November 18, 2019

Arthur F. Hultin

Chair, Truro Zoning Board of Appeals

PO Box 2030

Truro, MA 02666

Dear Mr. Hultin,

As a year-round Truro resident, I am writing to support the application for a Comprehensive Permit submitted by Community Housing Resource to provide 40 residential rentals at 22 Highland Rd.

The state of affordable housing on Cape Cod (and across the state) is bleak and it of vital importance that we seize this chance to make affordable rental a reality in Truro.

I am a board member of Highland Affordable Housing and I understand fully how hard it is to find year round accommodations. I am also a mediator in the Summary Process court and see first hand the struggle Cape Cod residents are faced with the possibility of losing their homes.

I want to speak out for those who would benefit from the opportunity this project would provide for so many who are otherwise being driven from Truro or the Cape because of the paucity of affordable rentals. We need to ensure the diversity of our town and to provide for ALL to live here.

Thank you for your attention.

Sincerely,

Jane J. Lea

Jane Lea

PO Box 1045

North Truro, MA 02652

Jeffrey Ribeiro

From: Russell Braun <arjaybraun@gmail.com>
Sent: Thursday, November 21, 2019 2:38 PM
To: Jeffrey Ribeiro
Subject: cloverleaf proposal

Jeff
Please have the ZBA read this email into the public record. I had opportunity to review the application for the proposed affordable housing at the cloverleaf and have the following thoughts-

Unlike the prior project completed in Truro by Community Housing the proposed site is exposed, viewable by everyone traveling Rte 6 through Truro. As such the proposal's visual impacts are crucial. My main concern is building height and the developer seeks forgiveness from the 30' maximum from mean grade for multiple buildings. This may or may not create a visual impact from the road but, based on the drawings submitted, there is no way of visually evaluating this.

Please understand that in my past professional life I designed and developed affordable housing and am enthusiastic supporter. Locally, I have been involved with Ted Malone's projects as a building official and I know that his organization does a fine job. We're lucky to have him.

The town, in its enthusiasm and push to provide more affordable housing for its citizens must be careful to make sure that this project fits the environment and nature of the setting and character of the town. As such we need to examine the proposal with a critical eye. I encourage the ZBA to make sure that it fully understands how this proposal fits in the setting. Based on the application package, I do not see how this is possible. The project team should have, at its disposal, adequate modeling and imaging tools to enable the ZBA and citizenry to view 3D images of this proposal not only from within the project site but also from Highland Rd., Rte 6 and other critical viewshed locations. If the proponent has not done so (prior commitment prevents me from attending today's meeting to see the full presentation) I am suggesting via this email that the ZBA request this.

This is a big deal for the town so its important that we ensure it gets done right. Architectural building elevations, which flatten a view of a building thus distorting what it really looks like, do not tell a true story and certainly dont give the public a picture of what it might be seeing on a daily basis.

Take a deep breath. Do it right. I have some other technical questions but nothing major. Thank you for your consideration

Russ Braun
510 Shore Rd.

November 17, 2019

Arthur F. Hultin

Chair, Truro Zoning Board of Appeals

PO Box 2030

Truro, MA 02666

Dear Mr. Hultin,

I am a year-round resident of Truro and I fully support the application for a Comprehensive Plan submitted by Community Housing Resource, Inc to create 40 affordable residential units at the Clover Leaf Project.

As you are well aware, the need for affordable housing in our town has reached a critical stage. We need to act on this opportunity to provide for all of our community.

I am speaking for those who cannot speak up and who would benefit from this project. The current cost of housing is so burdensome and good people are leaving far too often. We need Truro to thrive as a fully inclusive community.

Thank you for your time and efforts.

Sincerely,

Jennifer L. Shannon

Jennifer L. Shannon

PO Box 1045

North Truro, MA 02652

The Commonwealth of Massachusetts
MASSACHUSETTS SENATE

SENATOR JULIAN CYR
ASSISTANT MAJORITY WHIP
Cape and Islands District

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Chair
JOINT COMMITTEE ON MENTAL HEALTH, SUBSTANCE USE AND RECOVERY

Vice Chair
JOINT COMMITTEE ON HOUSING

JOINT COMMITTEE ON HEALTH CARE FINANCING
JOINT COMMITTEE ON PUBLIC HEALTH
JOINT COMMITTEE ON EDUCATION
JOINT COMMITTEE ON MUNICIPALITIES AND REGIONAL GOVERNMENT

November 21, 2019

Truro Zoning Board of Appeals
24 Town Hall Road
P.O. Box 2030
Truro, MA 02666

Dear Board members,

I regret that I am unable to join you in person due to obligations in Boston, but I wanted to take this opportunity to write in strong support of the Cloverleaf Community Housing project.

Attainable housing is one of the steepest challenges we face in Truro and across the Outer Cape. The vibrancy of our year-round community is eroding because residents cannot afford to live in our town. This poses difficulties for our workforce and sustainability of our schools. Older adults who are hoping to age in place and younger folks who hope to return to their community are being priced out of the neighborhoods they grew up in.

This project provides significant promise to meaningfully address the housing crisis by providing 40 units of affordable rental housing. That is 40 homes - 70 bedrooms - where families in our community can solidify their roots and build a modest life.

This project has been made possible by significant investment from the Commonwealth, first through the attainment of the Cloverleaf site from the Massachusetts Department of Transportation at a cost of $14,000. On Tuesday, the Commonwealth announced a $1.2

million MassWorks grant which will be used to provide municipal water to the site, allowing for many more units than if the project had to rely on well water. These efforts by residents of Truro and folks on the state level provide for a tremendous opportunity to benefit our small town.

I urge your favorable and swift consideration of this project.

Respectfully,

Julian Cyr

SUBMITTAL BY PAUL KIERNAN AT 12/5/2019 PUBLIC HEARING

Mr. Chairman and members of the Truro Zoning Board of Appeals,

Truro was given a 3.91-acre parcel by the state, commonly known as the "Cloverleaf", exclusively for affordable housing.

Truro issued a RFP (Request for Proposal) for a housing development plan ", estimated at between 30 and 40 units, ...". The guidelines suggested "...small but multi-unit structures." It also mentioned "Outdoor common areas are encouraged; gathering areas, playground areas and garden areas."

There was no mention within the RFP of the possible impact of such a concentrated development on Truro's aquifer.

There were 2 responses. The Town chose Mr. Malone, the developer of "Sally's Way".

The "Sally's Way" development resides on the same 10.66 acre parcel as the Truro Library and the Truro Community Center. In consists of 8 buildings, 16 units and 31 bedrooms.

The 10.66-acre parcel is in excess of 464,000 sq. ft. (464,349.6 sq. ft.) and appears to able to accommodate the 31 "Sally's Way" bedrooms using Truro's current Board of Health regulations and appears to meet State DEP Title V requirements.

This proposed development is very different.

The Cloverleaf parcel has 3.91-acres or slightly more than 170,000 sq. ft.

Under Truro's current Board of Health regulations this parcel could legally accommodate 17 bedrooms, not the proposed 70 bedrooms.

The 40B application you are reviewing asks this board to waive Truro's Board of Health Regulations.

The applicant on page 13, within paragraph 5, states "...The alternative of denitrification Alternative / Innovative septic technology would be an **excessive cost** that would burden the housing development budget as an upfront cost as well as ongoing annual system maintenance / monitoring expenses. Also, since the intent of bylaw is in part to protect private wells, it is noteworthy that the mapped ground water flow indicates an eastward flow from the proposed Title 5 Septic System leach field away from wells on the abutting properties."

The applicant has since corrected the ground water flow direction from eastward under the National Seashore toward the ocean to westward directly toward the North Truro Limited Business District and the private homes beyond.

I recently learned from a knowledgeable, long-standing member of Truro's Board of Health that when the State designed the run-off flow from Route 6, it unfortunately chose to direct the highway runoff in the same direction.

The safety of our water supply cannot be ignored. Nor should a developer's cost factor into our decision on whether or not to approve the current plan if in the end we sacrifice our water supply.

A 70-bedroom development anywhere else within the Truro Residential District would require 700,000 square feet (in excess of 16 acres) and the leaching fields would be evenly distributed throughout the 700,000 sq. ft. area.

Most likely these bedrooms developed elsewhere would only see daily use for less than 6 months a year.

In the plan before you, the effluent from the proposed 40 units, 70 bedrooms, and other sources of waste water (washers, toilets, showers, tubs, sinks, etc.) are all proposed to enter the aquifer via two 75' by 75' side by side leach fields. This concentration of effluent is problematic.

These proposed units will most likely be used on a year-round basis for the next 99 years.

This proposal will create a new source of highly concentrated pollutants where currently there is none.

The main problem is nitrogen, nitrogen as nitrate, and phosphorous in the water.

Maximum Contaminant Level (MCL) for nitrate measured as nitrogen in drinking water is **10 milligrams per liter** (parts per million) as established by the Federal Environmental Protection Agency. In addition, EPA has set an MCL for nitrite in drinking water at 1 milligram per liter (mg/L).

The Cape Cod Commission Staff comments, dated Dec. 3, 2019, state on page 3, paragraph 4, that:

"The project is proposed to be served by a Title V compliant septic system. Based on a preliminary calculation performed by the Commissions Staff, the project's site-wide nitrogen loading concentration (based on a proposed design flow for 70 bedrooms and 7700 gallons per day) would be nearly **19 mg/L**."

Because of the current elevated levels of nitrogen in the North Truro Limited Business District and surrounding properties' wells, how can we avoid making things worse?

I believe everyone can agree that affordable housing is a real problem in Truro. But it is not the only problem.

Recently the Town of Eastham and the Town of Wellfleet's center have spent millions trying to fix their drinking water problems.

If not for Truro, where would Provincetown get their water?

Safe drinking water needs every protection.

Here experts can give you and the Town the necessary facts to make the right decisions.

Alternative / Innovative septic technology may help, a downward redesign of the project or a combination of these and/or other ideas as yet unexplored may be the answer.

GOOD LUCK.

Paul Kiernan

Submittal by Debra Best-Parker at Public Hearing December 5, 2019

To:
Zoning Board of Appeals
Town of Truro

Thank you for allowing me to provide my comments to this board regarding the Cloverleaf Development Project being reviewed now.

I understand and fully support the need for additional affordable housing in our town but this plan is unacceptable as is. In this day and age, I find it hard to believe that we did not select a builder undertaking a project of this magnitude that uses "green" technologies. The plans have designs to accommodate solar panels on the roofs but do not have them. If the developer does not put them on, who will?

I implore you not to grant many, if not most, of the waivers being requested for this project; number one being the septic system proposed. If the septic system isn't adequate and too expensive to implement correctly so as to not pollute our town's drinking water, the project should be curtailed. Build a smaller number of units, 16 instead of the 70 bedrooms, which the acreage allows according to our town regulations, for good reason. Claiming another property as part of the land for the project is a shell game and unacceptable. The waivers for 3 stories, building heights, and inadequate easements, need not have been brought to you at all, since the builder knew our regulations and chose to ignore them in the presented plan.

I assume the water being brought to the site is from the Provincetown Water Department. If that is the case, the grounds cannot have automated irrigation systems running between June and October, based on the watering regulations of that department. Is this regulation also to be ignored? I am not allowed to violate this regulation for my landscaping. If they follow the rules, what is the plan for maintaining the new plantings in the proposed landscaping plan?

I feel powerless to influence this runaway train and I feel this board is being put in an untenable position of having to either accept this project at the expense of our environment or be accused of being against affordable housing. The builder should not have designed something that required all these waivers and instead, work within the framework of our town regulations

Please rule to insist that the design be changed to protect the water supply and follow our town regulations, which have been formulated and adopted to protect our town, both ecologically and aesthetically.

Respectfully,
Debra Best-Parker
2 Waterview Heights Road
N. Truro, MA 02652

Submitted by Jim Slater at 12/5/2019 Public Hearing

Data by Technical Bulletin

Welcome | Placetypes | Water | Ocean | Cultural Heritage | Wetlands & Wildlife | Open Space | Coastal Resiliency | Transportation | Community Design | Economy | Energy | Waste Management | Capital Facilities and Infrastructure | Housing

Water Resources

Goal:

To maintain a sustainable supply of high quality untreated drinking water and protect, preserve, or restore the ecological integrity of Cape Cod's fresh and marine surface water resources.

Objectives:

- ☐ Objective WR1 – Protect and preserve **groundwater** quality
- ☐ Objective WR2 – Protect, preserve, and restore **fresh water** resources
- ☐ Objective WR3 – Protect, preserve, and restore **marine water** resources
- ☐ Objective WR4 – Manage and treat **stormwater** to protect and preserve water quality
- ☐ Objective WR5 – Manage groundwater withdrawals and discharges to maintain hydrologic balance and protect surface and groundwater resources

Resource Areas: (If a data layer is not loading, please try zooming in)

Wellhead Protection Areas

Potential Water Supply Areas

Marine Watersheds

Subembayments

Subwatersheds

Fresh Water Recharge Areas

Impaired Areas

Plumes

Hazardous materials sites

Hazardous Materials (c21e)

Hazardous Materials (AUL)

Water Infrastructure

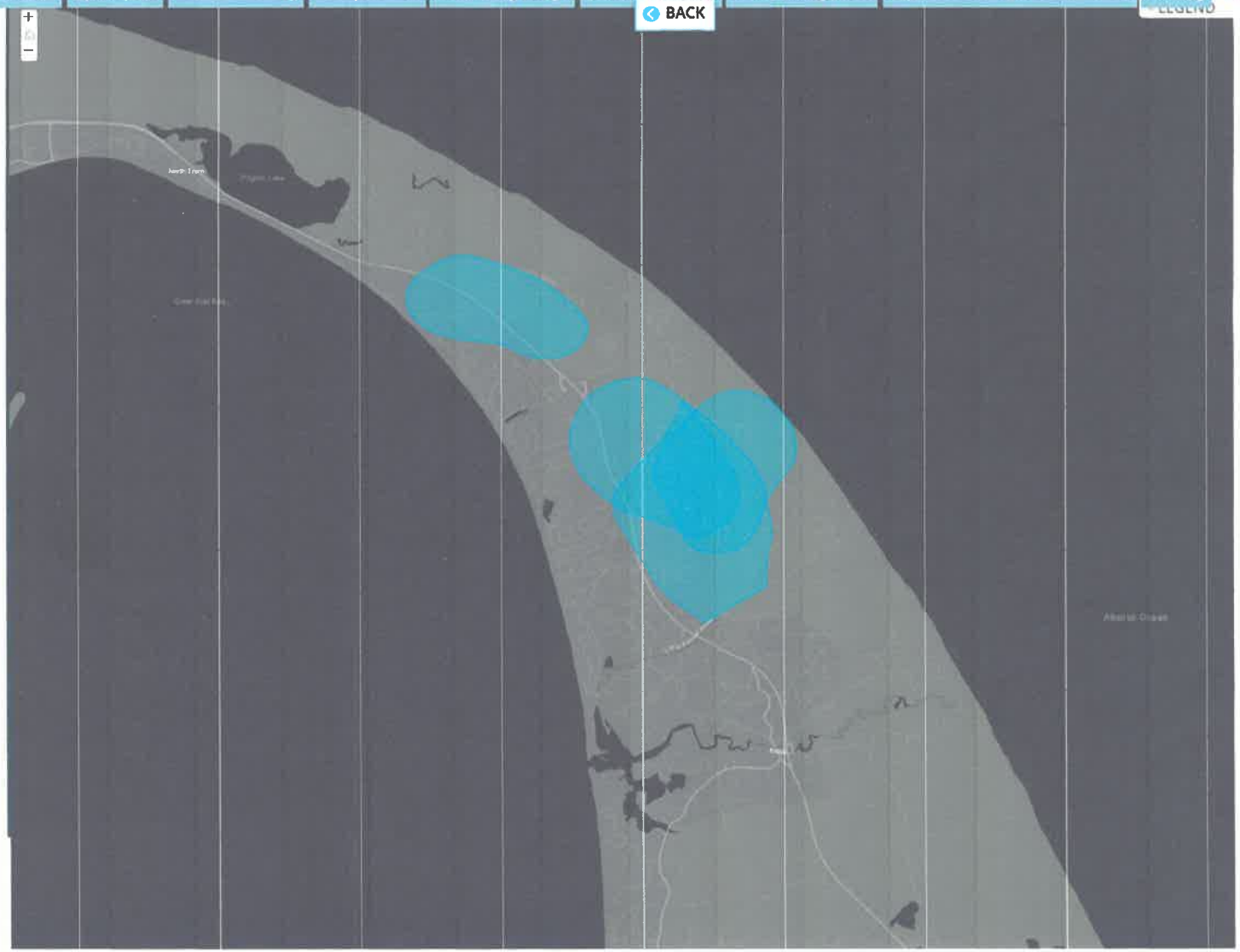
Public Water Supplies

Sewered Areas

Groundwater Discharge

Ground Water Lenses

Sagamore



SUBMITTED BY PETER HERRIDGE AT 12/5/2019 HEARING (SINGLE PAGE SUBMITTED)

Fwd: Re: Cloverleaf project

Kevin Kuechler <kevinkuechler27@gmail.com>
Tue 12/3/2019 10:18 PM
To: jackriemer@hotmail.com <jackriemer@hotmail.com>

---------- Forwarded message ---------
From: **Kevin Kuechler** < kkuechler@comcast.net>
Date: Tue, Dec 3, 2019 at 5:17 PM
Subject: Fwd: Re: Cloverleaf project
To: <kevinkuechler27@gmail.com>

---------- Original Message ----------
From: Kevin Kuechler <kkuechler@comcast.net>
To: jack riemer <jackriemer@hotmail.com>
Date: December 3, 2019 at 3:19 PM
Subject: Re: Cloverleaf project

Hi Jack,

i understand that the Cloverleaf project's developer is seeking relief from Truro's wastewater regulations. I am vehemently opposed to this.

Both Truro and Provincetown rely on Truros ground water for all of their drinking water. Furthermore, it appears that the ground water at the Cloverleaf flows toward the Bay via Pond Village. The ground water quality in Pond Village is worse than the average ground water quality in Truro, likely due to shallow ground water and older housing stock. For this reason, I would recommend stricter waste water treatment standards at the Cloverleaf rather than what is being proposed.

Relaxing waste water treatment standards at the Cloverleaf would be like allowing these homes to be built with coal-burning boilers for heat and hot water. This would set a truly bad precedent.

Kevin Kuechler

> On December 3, 2019 at 1:46 PM jack riemer <jackriemer@hotmail.com> wrote:
>
> Hi Kevin ,

SUBMITTAL BY PETER HERRIDGE AT 12/5/2019 HEARING

Nitrate Sampling Program Update

Water Resources Oversight Committee
June 1, 2010

Purpose: Establish baseline nitrate data for the entire town. Results will be used to identify areas where additional ground water protection may be warranted. This is the third year of sampling. A bottle has been sent to nearly all residences over the past three years. 1181 (51%) bottles were returned for analysis.

Approach: Test nitrates in ground water through a voluntary program whereby one-third of the town is chosen for sampling each year. We modeled our program on the one that is already established in Eastham. Nitrates were chosen since they indicate areas where septic systems may be operating ineffectively. Elevated nitrates can also be an indicator of other types of contamination as well.

Results:

- The average concentration of nitrates across the town is 1.11 mg/L.
- The range of values is from 0 to 11 mg/L.
- 45 out of 1181 samples (3.8%) came back at 5 mg/L (local standard) or higher.
- 2 out of 1181 samples (0.2%) came back at 10 mg/L (EPA standard) or higher.

Data:

North of Pond Village (Maps 2 -35):

- 123 samples
- Average concentration = 1.23 mg/L
- 2.4% of samples at 5 mg/L or higher

Pond Village/Shearwater (Maps 36, 38, and 39):

- 169 samples
- Average concentration = 1.5 mg/L
- 5.9% of samples at 5 mg/L or higher

South of Shearwater to North of Pamet River (Maps 37, 40, 41, 42, 43, 44)

- 318 samples
- Average concentration = 1.34 mg/L
- 5% of samples at 5 mg/L or higher

Pamet River Valley (Maps 45-51):

- 391 samples
- Average concentration = 0.99 mg/L
- 3.6% of samples at 5 mg/L or higher

Nitrate Sampling Program Update (continued)

June 1, 2010

South Truro (Maps 52-64):

- 179 samples
- Average concentration = 0.51 mg/L
- 1.1% of samples at 5 mg/L or higher

Interpretation:

- Nitrate concentrations are fairly low from the Pamet River Valley south to the Town line.
- Nitrate concentrations from Pond Village to north of the Pamet River Valley are somewhat higher, though still low compared to existing standards.

Next Steps:

- Re-test the 45 locations with nitrates at 5 mg/L or higher.
- Finalize database so that additional mapping and analysis can be performed
- Develop new notification card that provides suggestions to homeowners for ways to reduce local nitrate concentrations.
- Work closely with BOS, PLOG, BOH, and others to strengthen ground water protections in Truro.

Pg 2 of 2

December 12, 2019

To the Truro Zoning Board of Appeals:

In support of Clover Leaf

My name is Katherine Black and I live at 36 Corn Hill Road. I retired to Truro five years ago, after more than a lifetime as a summer resident. I say more than because my mother spent the summers here including the summer she was pregnant with me. My great grandmother used to come to Truro. My grandfather bought property near Corn Hill, my father subdivided it and passed it on. So I am one of the lucky ones who gets to live here in this Cape Cod paradise.

When I was able to move here year round, the first thing I became aware of was the housing crisis. I joined Highland Affordable Housing and learned that not only is there little or no housing for those who work in Wellfleet, Truro and Provincetown, people are being forced to leave the Outer Cape in droves when they lose their housing. Whether their rental has been sold or they need to downsize, there is basically nothing to move to. So yes, we need work force housing, but we also need senior housing and family housing. I love Truro as much as anyone, and I want to protect it as much as anyone, but I hate to see 75% of homes empty all winter as the average age of year-rounders creeps ever higher. I have done what I can as an individual by building a habitable studio apartment that I rent to a Truro resident who had lost her housing. I have also participated in home sharing, renting rooms in my house. But I am just one person. The Clover Leaf project can make a real impact on the crisis!

I have faith in our Town and State regulations to protect our environment. I trust our Town employees to oversee those regulations. I am dismayed by the lack of trust I observe in those who are fighting this project.

Why isn't there an attitude of "How can we make this work?" instead of "How can we put a stop to it?"

Sincerely,

Kat Black

Katherine Black

P.O. Box 1065
Truro, MA 02666
bunkerblackat@gmail.com

Submitted by Peter Herridge Dec 5, 2019

SUBMITTAL BY PETER HERRIDGE AT 12/5/2019 HEARING

International Journal of *Environmental Research and Public Health*

Review

Drinking Water Nitrate and Human Health: An Updated Review

Mary H. Ward [1,*], Rena R. Jones [1], Jean D. Brender [2], Theo M. de Kok [3], Peter J. Weyer [4], Bernard T. Nolan [5], Cristina M. Villanueva [6,7,8,9] and Simone G. van Breda [3]

1 Occupational and Environmental Epidemiology Branch, Division of Cancer Epidemiology and Genetics, National Cancer Institute, 9609 Medical Center Dr. Room 6E138, Rockville, MD 20850, USA; rena.jones@nih.gov
2 Department of Epidemiology and Biostatistics, Texas A&M University, School of Public Health, College Station, TX 77843, USA; jdbrender@sph.tamhsc.edu
3 Department of Toxicogenomics, GROW-school for Oncology and Developmental Biology, Maastricht University Medical Center, P.O Box 616, 6200 MD Maastricht, The Netherlands; t.dekok@maastrichtuniversity.nl (T.M.d.K.); s.vanbreda@maastrichtuniversity.nl (S.G.v.B.)
4 The Center for Health Effects of Environmental Contamination, The University of Iowa, 455 Van Allen Hall, Iowa City, IA 52242, USA; peter-weyer@uiowa.edu
5 U.S. Geological Survey, Water Mission Area, National Water Quality Program, 12201 Sunrise Valley Drive, Reston, VA 20192, USA; btnolan@usgs.gov
6 ISGlobal, 08003 Barcelona, Spain; cvillanueva@isiglobal.org
7 IMIM (Hospital del Mar Medical Research Institute), 08003 Barcelona, Spain
8 Department of Experimental and Health Sciences, Universitat Pompeu Fabra (UPF), 08003 Barcelona, Spain
9 CIBER Epidemiología y Salud Pública (CIBERESP), 28029 Madrid, Spain
* Correspondence: wardm@mail.nih.gov

Received: 17 May 2018; Accepted: 14 July 2018; Published: 23 July 2018

Abstract: Nitrate levels in our water resources have increased in many areas of the world largely due to applications of inorganic fertilizer and animal manure in agricultural areas. The regulatory limit for nitrate in public drinking water supplies was set to protect against infant methemoglobinemia, but other health effects were not considered. Risk of specific cancers and birth defects may be increased when nitrate is ingested under conditions that increase formation of *N*-nitroso compounds. We previously reviewed epidemiologic studies before 2005 of nitrate intake from drinking water and cancer, adverse reproductive outcomes and other health effects. Since that review, more than 30 epidemiologic studies have evaluated drinking water nitrate and these outcomes. The most common endpoints studied were colorectal cancer, bladder, and breast cancer (three studies each), and thyroid disease (four studies). Considering all studies, the strongest evidence for a relationship between drinking water nitrate ingestion and adverse health outcomes (besides methemoglobinemia) is for colorectal cancer, thyroid disease, and neural tube defects. Many studies observed increased risk with ingestion of water nitrate levels that were below regulatory limits. Future studies of these and other health outcomes should include improved exposure assessment and accurate characterization of individual factors that affect endogenous nitrosation.

Keywords: drinking water; nitrate; cancer; adverse reproductive outcomes; methemoglobinemia; thyroid disease; endogenous nitrosation; *N*-nitroso compounds

1. Introduction

Since the mid-1920s, humans have doubled the natural rate at which nitrogen is deposited onto land through the production and application of nitrogen fertilizers (inorganic and manure),

Submittal at 12/19/2019 Public Hearing.

Cloverleaf Comments

Susan Howe
12 Houser Way

-Although I'm the chair of the Truro Commission on Disabilities and the president of the Truro Historical Society, am speaking as a private citizen here tonight. I'm speaking because I love Truro,not because I have a vested interest in any aspect of the project.
-I was here last week. I looked around and saw people with a vested financial interest in Affordable housing, people with an emotional interest like those serving on the Housing Authority, People who are more interested in issues like climate change, water resources, recreation, open space....
-I think we need to consider the interests of everyone—the REAL needs of our citizens—the townspeople of Truro.
-the Cloverleaf property was given to Truro by the state to build Affordable housing, but does it have to have so many units? Can't we consider a smaller project be built?
-This is not about age or socioeconomic status. It is about all of us working together to improve our town.
-For the record, I am pro Affordable (Subsidized) housing. I am also pro housing that is affordable. (Most of us understand that these are two separate needs.) I am also pro environmental protection, pro preservation of our natural resources, and pro honoring our history and preserving as much of that history as we can. I am pro housing that is as accessible as possible since the vast majority of us are aging and getting less physically able. This is called "Universal Design". Many of us want to "age in place" and need an environment that we can navigate as we get older. When people present the opinion that this is the reason to build subsidized housing to house the "work force" they are not understanding that anyone making a livable wage makes TOO MUCH money to qualify. Ask the carpenters, plumbers, electricians, nurses, allied health professionals, landscapers, farmers, etc.
-People who were upset about Peter Herridge speaking his mind should be at least equally upset about the Affordable Housing train that is speeding along without consideration of the long term effects.
-What I love about Truro is that we are a town of individuals and that means we have differing opinions on some issues. Personally, I would like to see a development built that will not tax our precious environment AND would meet the needs of Truro citizens who need housing that is affordable to them (there are many more of us who have limited incomes but make too much money to qualify for a subsidy than those who do qualify. This needs to be reflected in whatever we build. To me, the ideal development would be as "green" as possible (ie solar

panels, safe sewerage and water protection systems)and be universally accessible. It could be a showplace for the rest of Cape Cod, the state of MA and the nation.
-Sadly, I think this is only a pipe dream. Our town seems as divided as our nation.
-So, Where is the art of compromise? Communication? Collaboration? This doesn't have to be contentious. There don't have to be winners and losers.
-Won't you-the Zoning Board of Appeals-protect the rural nature of Truro while approving development that will be good for all of us—not just those who have a vested interest?
-The late Maureen Burgess was very concerned about these issues and it is my honor to present them to you here tonight, not in her name but in my own. Still I have no doubt that Maureen is in agreement with many of my remarks.
-Thank you.

Dear Members of the ZBA,

I am writing in support of the Cloverleaf Project. I have spent every summer here since 1985 and moved here when I retired a little over a year ago. Truro is really a special place. I believe that in order to maintain the character of Truro; it is important that there be year-around housing available to people with different incomes. I believe the economic and community health of Truro depends on such diversity.

The provision of mixed income year-around rental housing is especially important to people in my age group. There are life-long residents who need to "downsize". They have recently been widowed, require more accessible housing or are just unable to maintain a large home. Yet they want to live out their life in Truro. Where do they find housing that meets their needs as well as their income?

More and more of us are going to face medical issues as we age. We are going to need more health care professionals such as a visiting nurse or physical therapist, to help us with those issues. Already it takes months to get assigned a primary care provider at Outer Cape Health. I was recently told it would take weeks to be assigned a nurse as my primary care provider and I would not have my first appointment for months. The cost of housing is a major reason, we cannot get a young doctor and her family to move to the Outer Cape.

I have never wanted to live in housing restricted to people over 55. Yet that is what Truro will become without housing that is affordable to young people at the beginning of their careers.

I am living in a year-round rental very close to the Cloverleaf Project and am looking forward to those new neighbors. They will be a welcomed addition to this community.

Thank you for your consideration of my position,

Betty Gallo

From:	Elizabeth Bradfield
To:	Jeffrey Ribeiro
Cc:	Lisa Sette
Subject:	Re: Cloverleaf- comments for the ZBA
Date:	Tuesday, December 17, 2019 5:31:53 PM

Dear Jeffrey Ribeiro,

I am an abutter to the Cloverleaf development (6 Professional Hts. Rd. #2) and a year-round resident of North Truro. I work locally for the Dolphin Fleet, lead classes for Wellfleet Audubon, and teach at the Fine Arts Work Center on occasion. My spouse, Lisa Sette, works at the Center for Coastal Studies. This is to say, we're one of the many non-retired folks here scrambling to make a living. I have attended the site walk-through and ZBA meetings regarding the Cloverleaf development, as has my spouse, Lisa Sette. I was shy about speaking at the 12/12/19 meeting, which I attended, but I am hoping our comments can be heard here, via email.

Overall, as someone who feels incredibly lucky to have found a place to live/own year-round on the Cape in North Truro after renting/caretaking on the outer cape for 10+ years, we are a firm proponent of year-round affordable housing. HOWEVER, as an abutter and a resident of North Truro, we are concerned about the Cloverleaf development's size and some of its other details.

We know this project was begun before we acquired the property behind the Truro Elementary School, but it seems we have time/space to really think about development in North Truro in a holistic way now. 20 Units in Cloverfield and, say, 30, in the elementary school property seems like a wiser plan for our resources, particularly when those living in Cloverfield will be seriously impacted by the noise of the highway (living nearby, I can attest to its pervasive impact). 40 units is a LOT to be squeezed into that small parcel.

Our first and primary concern is the septic runoff. We have a well on Professional Hts, and it seems that the majority of effluent from the Cloverleaf development would flow our way. Even with the additional proposed "scrubber" technology, we are concerned about this septic addition to our neighborhood (I've heard concerns about the regulation of the campground to the east, at Adventure Bound, at other meetings and do not believe that has been addressed). If there were a power failure or a huge storm (the development only has plans for a 50-year storm runoff event, and that itself seems inadequate), what would be the efficacy of the septic field? Nitrogen is one concern, but equally concerning are the pharmacudicals of 40 units.

Our second concern is traffic/concentration/planning. 40 units is quite a bit to fit into the small space of the Cloverfield development. Highland Road is busy - is the town ready/willing to provide a sidewalk on the north/west side of the road to the bus stop/corner? Can we put in a four way stop at the 6A/Highland Road intersection (already a problem)?

Light pollution has been addressed, but not completely--coyotes, turkeys and foxes utilize wildlife corridors in our area, and that is part of its charm and draw. We want to make sure that any public lights are low-level and not bright white. There are plenty of studies stating that lower public lighting is just as safe -- and often safer -- than really bright developments. See Dark Sky Initiative for supporting studies: https://www.darksky.org/light-pollution/

Also, I have seen the utility poles that are along the side of road that is set to have a driveway for Cloverfield--a member of the Truro fire department expressed concern that Truro does not have a ladder truck. Should Truro acquire a ladder truck the electrical in its current form is not adequate -- at least to my untrained eye -- to a ladder truck's clearance rates. While that IS a concern of the electric utilities, it points to another practical barrier to be overcome and paid for by the development.

Furthermore, we are concerned about sound-- particularly during construction. Sound pollution could be mitigated by setting a 8 am - 5 pm, Monday - Friday bylaw, but we do not have such an ordinance in place -- from what I understand --at present. Whatever time of year this contstruction is to occur, acknowledging that there are nearby residents who are concerned is of vital importance. This is not just a seasonal concern, but that of year-round residents who work hard to keep their residences and need the respite of a quiet weekend.

Again, overall, we are very much in favor of innovative plans for affordable housing and wise development in Truro. But, particularly since we have a chance now to step back and consider a town-wide plan that integrates Cloverleaf and the property behind the Elementary School, I think we need to take that time. We know a lot of people have put in a lot of hard work here, and we truly appreciate that.

Thank you for registering my concerns. My partner and I wish we could be at the 12/19 meeting, but I have other obligations that will keep me away. If there is a more appropriate place and space for me to express my thoughts, please do let me know. We hope to be able to attend the January meeting.

Sincerely,
Elizabeth Bradfield and Lisa Sette

On Tue, Dec 17, 2019 at 11:13 AM Elizabeth Bradfield <lizbradfield@gmail.com> wrote:

> Dear Jeffrey Ribeiro,
>
> I am an abutter to the Cloverleaf development (6 Professional Hts. Rd. #2) and a year-round resident of North Truro. I work locally for the Dolphin Fleet, lead classes for Wellfleet Audubon, and teach at the Fine Arts Work Center on occasion. My spouse, Lisa Sette, works at the Center for Coastal Studies. This is to say, we're one of the many non-retired folks here scrambling to make a living. I have attended the site walk-through and ZBA meetings regarding the Cloverleaf development, as has my spouse, Lisa Sette. I was shy about speaking at the 12/12/19 meeting, which I attended, but I am hoping our comments can be heard here, via email.

Overall, as someone who feels incredibly lucky to have found a place to live/own year-round on the Cape in North Truro after renting/caretaking on the outer cape for 10+ years, we are a firm proponent of year-round affordable housing. HOWEVER, as an abutter and a resident of North Truro, we are concerned about the Cloverleaf development's size and some of its other details.

We know this project was begun before we acquired the property behind the Truro Elementary School, but it seems we have time/space to really think about development in North Truro in a holistic way now. 20 Units in Cloverfield and, say, 30, in the elementary school property seems like a wiser plan for our resources, particularly when those living in Cloverfield will be seriously impacted by the noise of the highway (living nearby, I can attest to its pervasive impact). 40 units is a LOT to be squeezed into that small parcel.

Our first and primary concern is the septic runoff. We have a well on Professional Hts, and it seems that the majority of effluent from the Cloverleaf development would flow our way. Even with the additional proposed "scrubber" technology, we are concerned about this septic addition to our neighborhood (I've heard concerns about the regulation of the campground to the east, at Adventure Bound, at other meetings and do not believe that has been addressed). If there were a power failure or a huge storm (the development only has plans for a 50-year storm runoff event, and that itself seems inadequate), what would be the efficacy of the septic field? Nitrogen is one concern, but equally concerning are the pharmacudicals of 40 units.

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understand --at present. Whatever time of year this contstruction is to occur, acknowledging that there are nearby residents who are concerned is of vital importance. This is not just a seasonal concern, but that of year-round residents who work hard to keep their residences and need the respite of a quiet weekend.

Again, overall, we are very much in favor of innovative plans for affordable housing and wise development in Truro. But, particularly since we have a chance now to step back and consider a town-wide plan that integrates Cloverleaf and the property behind the Elementary School, I think we need to take that time. We know a lot of people have put in a lot of hard work here, and we truly appreciate that.

Thank you for registering my concerns. My partner and I wish we could be at the 12/19 meeting, but I have other obligations that will keep me away. If there is a more appropriate place and space for me to express my thoughts, please do let me know. We hope to be able to attend the January meeting.

Sincerely,
Elizabeth Bradfield and Lisa Sette

--
Elizabeth Bradfield
www.ebradfield.com
www.broadsidedpress.org
PO Box 24, Provincetown MA 02657

--
Elizabeth Bradfield
www.ebradfield.com
www.broadsidedpress.org
PO Box 24, Provincetown MA 02657

December 15, 2019

Arthur F. Hultin Jr.
Chair, Truro Zoning Board of Appeals
24 Town Hall Road
P.O. Box 2030
Truro, MA 02666

Dear Mr. Hultin,

As property owners in Truro (81 Slough Pond Rd.) we are writing to support the application for a Comprehensive Permit submitted by Community Housing Resource to create affordable 40 residential units at 22 Highland Road in Truro.

The Outer Cape has struggled for decades with a shortage of affordable housing and this single development will bring a significant body of housing units within reach of the permanent residents who have been crowded out of the market by rising real estate values.

We know many working families and citizens who are committed to the area but struggle to make ends meet. Truro's leaders need to find ways to make decent housing within reach for residents beyond those who inherit homes or have the wealth to afford property there. We are grateful that the state of Massachusetts and advocates for working people on the Cape have developed this plan. Please help keep Truro and the Cape a place where everyone has a chance to live and thrive and contribute to our community.

Thank you for your attention to this matter.

Sincerely,

Emily Bingham and Stephen Reilly

Jeffrey Ribeiro

From: sally brotman <sallybrotman@comcast.net>
Sent: Thursday, December 19, 2019 1:56 PM
To: Sally Brotman
Subject: Fact Check on Nitrates
Attachments: Herrige1.pdf; Herridge2.pdf

I hope you have time to read the article and handout attached below before the ZBA hearing at 5:30 pm at Town Hall this evening.
sally

An article in the Provincetown Independent today (reference below) was written in response to the unsigned hand-out that was hand-delivered to a home in North Truro (attached).

The Provincetown Independent investigation of the facts casts doubt on the claims made by the opposition to the Cloverleaf project.

FACT CHECK

Claims on Nitrates by Town Official Don't Hold Water

A second look at Cloverleaf project opponents' arguments

BY EDWARD MILLER · DEC 19, 2019

https://provincetownindependent.org/news/2019/12/19/statements-on-nitrates-dont-hold-water/

THE CLOVERLEAF COMPLEX FACT SHEET
VITAL INFORMATION ABOUT IMPACT ON WATER QUALITY

The clock is ticking to approve plans to build the "Cloverleaf" affordable housing project – a privately-owned complex - at the intersection of Highland Rd and Route 6 on town-owned land.

The plan puts 70 bedrooms in 11 buildings on less than 4 acres of land using a traditional septic system.

- **Housing that is affordable for Truro folks is much needed.**
- **But this density of use requires an advanced nitrate-reducing septic system.**
- **The developer is seeking a waiver from the Truro Board of Health regulations, in order to avoid installing and operating this necessary septic system.**

Truro's wastewater regulations require that the sewage and waste water output for a **complex of this size be spread out evenly over 16 acres of land**, not over less than 4 acres. The purpose of Truro's waste water regulations is to **prevent the pollution of our shared aquifer - and our town's and our own only source of drinking water.**

The developer **does not want to properly treat this sewage and wastewater to reduce toxic nitrate levels because it would cost HIM too much money!**

But the cost to area residents, the environment and our shared aquifer is much greater, now and for generations to come.

What's The Problem?

In a word: ***NITRATES***. The State's EPA sets maximum levels at **10 mg/L and Truro sets a safe level at 5 mg/L.** The Cape Cod Commission estimates the nitrogen loading from the Cloverleaf complex will be **19 mg/l – almost twice the EPA's limit and four times Truro's accepted safe level.**

So What?

Nitrate levels above 10 mg/L are a serious matter - Even 5mg/l - Truro's standard – is deeply concerning. But 10mg/l can be lethal to newborns and infants and will almost certainly increase rates of cancer, neural tube birth defects and thyroid disease in the entire affected region.

Who Will Be Affected?

The ground water beneath the Cloverleaf parcel flows toward the west and southwest of the Cloverleaf site, including Pond Village, already designated an **area of critical interest** by Weston & Sampson.** This designation was based on elevated nitrate levels detected in **Pond Village, among the highest in Truro.**

Everyone in Truro could be affected with potentially higher tax rates and lower property values, along with possible litigation.

(over, please) →

** See their Integrated Water Resources Management Plan, Phase I for Truro (October, 2014), posted on Truro's website. (Please see figure 4.3) https://www.truro-ma.gov/water-resources-oversight-committee/pages/truro-phase-i-wrmp

We already know that:

- The town's nitrate tests from 2010 showed the average nitrate level measured in wells in **Pond Village and parts of Shearwater were 40% higher than the town-wide average.**
- Sampling of homes in Pond Village in 2013 and 2016 indicated that at least 12 homes now have drinking water with **nitrate levels approaching Truro's limit of 5mg/L. Some homes have drinking water with nitrates above 9 mg/L.**
- If not properly treated, waste water produced at the Cloverleaf with 19 mg/l of nitrate could plausibly cause nitrate levels in Pond Village and other nearby areas **to soon exceed 10 mg/L and to rise steadily thereafter,** given that nitrate levels are already elevated.
- **Water has no property boundaries** - and flows where it will.

What Can WE Do?

As a "40B" project, the Truro Zoning Board of Appeals - with input from the Board of Health and Conservation Commission – will decide whether to grant the developer's permit.

We can demand that the developer put safety, health and water quality of the Cloverleaf residents and the surrounding area above profits and install the necessary septic system to reduce nitrate discharge to safe levels - or else face denial of the permit to develop the Cloverleaf project until this is done.

What Can YOU Do?

Let YOUR voice be heard. **Call on the ZBA to permit affordable housing *without putting* residents, neighbors and the environment in harm's way.** *Here are ways to do that while* public discussion is still open.

- **ATTEND the coming ZBA MEETINGS on the CLOVERLEAF Project**
  Dec 19 | Town Hall | 5:30 PM - check the Town website for other dates and times
  https://www.truro-ma.gov/zoning-board-of-appeals

- **CALL OR WRITE ZBA Members to express your views ASAP.**
  Arthur Hultin **John Thornley** **John Dundas**
  Fred Todd **Chris Lucy** **Darrell Shedd** **Heidi Townsend**

- **CALL OR WRITE SELECT BOARD Members to express your views and ask for your** comments to be read into the record.

Jan Worthington jworthington@truro-ma.gov
Kristen Reed kreed@truro-ma.gov
Bob Weinstein rweinstein@truro-ma.gov
Sue Areson sareson@truro-ma.gov

- ***Inform Neighbors And Friends*** - tell everyone you know that our water quality is at risk.

- ***Support environmentally responsible affordable housing!***

Thank You.

FACT CHECK

Claims on Nitrates by Town Official Don't Hold Water

A second look at Cloverleaf project opponents' arguments

BY EDWARD MILLER · DEC 19, 2019

TRURO — Some statements made at the Dec. 5 zoning board of appeals hearing related to groundwater nitrogen levels expected to be produced by the proposed "Cloverleaf" housing development did not pass a fact check conducted by the ***Independent*** this week.

The planning board's Peter Herridge, an opponent of the plan to build 40 units of affordable housing on Highland Road in North Truro, failed to provide evidence on water standards and the risk of nitrate poisoning to back up his statements. And local officials refuted some of his claims about recent nitrogen measurements and potential future pollution.

Herridge told the ***Independent*** that he is part of a group that has created a "fact sheet" about the development and added that his group is prepared to sue the town to stop the Cloverleaf housing from being built.

That document, circulating in Truro on Tuesday, warns that, if the housing is built with the wastewater waiver the developer has requested, it will create conditions "lethal to newborns and infants" and will "almost certainly" increase rates of cancer, birth defects, and thyroid disease.

The U.S. standard for nitrate in drinking water limits it to 10 mg per liter. Herridge told the ZBA that the safety standard in Germany is 4.4 mg per liter. "By Germany's standard, people in these hot spots in Pond Village are already drinking toxic water," Herridge said.

Journal of Environmental Research and Public Health by Mary H. Ward et al. But that article states that the standard in Germany and throughout the European Union is actually slightly higher than in the U.S., 11.3 mg per liter.

Moreover, the Ward article casts doubt on Herridge's predictions about the lethal consequences of nitrate in groundwater, concluding that "the number of well-designed studies of individual health outcomes is still too few to draw firm conclusions about risk from drinking water nitrate ingestion."

In a Dec. 15 email, Herridge wrote, "I am a physician and a pharmaceutical patent attorney and I know my biochemistry and this is a serious threat to our water aquifer and the developer could easily properly treat his sewage on site or reduce the number of bedrooms to what the land can tolerate and that is all we want."

The developer has proposed that, by including undevelopable National Seashore land surrounding the site in wastewater calculations, the project will meet the town's standards. The ZBA has asked Malone to hire a consultant to study that possibility.

Herridge told the ***Independent*** that the Cloverleaf area has the worst nitrogen contamination in Truro. "In 2010, six percent of the homes had levels over 5 mg.," he said. "The town did not check them more recently, but I have, and the levels are increasing. Some homes in Pond Village are over 10 mg and have gotten warning notices from the board of health."

But Truro Health Agent Emily Beebe said, "I'm not aware of any tests over 10 mg in that neighborhood. There are some readings that are over 3, and a couple over 5. The average in Pond Village of all the samples we could find is less than 2."

Tracey Rose, chair of the Truro Board of Health, said, "To the best of my knowledge, no letters have gone to Pond Village residents nor has anyone's well tested at that high level."

In an interview, Herridge also had harsh words for Ted Malone, the Cloverleaf developer.

"The developer is making money off poor people," Herridge said. "The reason he didn't want to treat his sewage is that it costs too much. He's trying to con the town into giving him the most lucrative deal they can."

Herridge's flyer states that "the developers are refusing to install and operate" an advanced nitrate-reducing septic system.

Jay Coburn of the Community Development Partnership disputed this, saying that town officials were still weighing whether an advanced system should be required, and that Malone was exploring the cost of putting in a small wastewater treatment plant at Cloverleaf, should one be needed.

Coburn defended Malone's track record as a developer. Malone, he said, "is one of the most highly respected small-scale developers in Massachusetts." Coburn also noted that the state

Kathleen Henry, president of the board of Highland Affordable Housing, also defended Malone.

"There's no question, Ted Malone is in business," Henry said. "He's not running a charity. But he is a person of the highest ideals, with a deep commitment to affordable housing and to giving people not just the basics but housing that's beautiful as well as efficient and clean and safe."

Referring to Malone's development near the Truro Library, Henry said, "You only have to visit Sally's Way to see the kind of work that he does."

FILED UNDER: ENVIRONMENT, FEATURED, LAND USE, NEWS, TOWNS, TRURO
TAGGED WITH: CLOVERLEAF

About Edward Miller
Ed Miller is the editor of the Provincetown Independent.

Jeffrey Ribeiro

From: K.S. O'Brien <ksmytheobrien@gmail.com>
Sent: Monday, December 23, 2019 11:11 AM
To: Jeffrey Ribeiro
Cc: Jan Worthington; Robert Weinstein; Susan Areson; Kristen Reed
Subject: Cloverleaf project

Dear Zoning Board of Appeals:

Please confirm receipt of this email correspondence and read it into the record.

As year round Truro residents we are opposed to the Cloverleaf complex as it is currently proposed. Some of our environmental, health and other concerns are described below.

For purely financial gain, the developer is trying to skirt regulations that assure the safety of water quality of not only our town but of Provincetown as well. It the ZBA/town approves the waste water waivers as requested, it is acting in a highly irresponsible manner and complicit in endangering the health of our citizens and those from surrounding towns. If the ZBA approves the project as currently presented, Truro opens itself up to innumerable extremely costly lawsuits.

If the building height variances are approved, this project will inalterably change the aesthetics of the town for all future generations and contribute to destroying the rural character of the town. The lot will no doubt be clear cut to accommodate this monstrosity which will add to the carbon load, not to mention displacing innumerable wildlife.

It is our understanding that an individual (who has been pushing hard for this project to progress) who is active in our town government will ultimately receive financial gain from the involvement in and management of this development, if constructed. This is a travesty and huge conflict of interest and could open the town up for further costly and embarrassing lawsuits. As the old saying goes, follow the money. This basically stinks to high heaven of a suspect process with back door dealings and we are horrified that it is being crammed down on the citizens of Truro.

The ZBA and the town need to do the right thing. If this is to be built at all, it must be significantly scaled back to conform with current regulations. We the citizens will hold you accountable. We urge you to act responsibly and uphold the guidelines of current regulations - which are there to protect our health and that of future generations - and deny the developer's request as presented.

Sincerely,
Kate and Dennis O'Brien
24 Bayberry Road
Truro MA

From: **Ralph Freidin** rfreidinmd@gmail.com
Subject: Cloverleaf Affordable Housing
Date: January 6, 2020 at 10:33 AM
To: jworthington@truro-ma.gov, kreed@truro-ma.gov, rweinstein@truro-ma.gov, sareson@truro-ma.gov

Dear Select Board Members

Although I am not a permanent resident of Truro, I am a part-time resident, concerned citizen and member of the Truro Part-time Resident Taxpayers' Association.

I am troubled by the environmental impact of the nitrate levels produced by the Cloverleaf Affordable Project as it is currently proposed. Clearly affordable housing is a concern for Truro, as it is for entire outer cape. However, the need for housing must be balanced by its environmental impact. It is the Zoning Board's responsibility to the full and part-time Truro residents to assure that both objectives are achieved fairly and safely. Until the developer can guarantee that nitrate production from the Cloverleaf project will be, and remain, within the limits established by Massachusetts EPA, the project should no go forward.

Would you please read my comments at the January 16 ZBA meeting that I am unable to attend.

Thank You,

Ralph B Freidin]
51 Castle Road, Truro

From: Alison Ellis elliali@gmail.com
Subject: Impact on water quality of the Cloverleaf project in No. Truro
Date: January 6, 2020 at 9:47 AM
To: jworthington@truro-ma.gov

Dear Ms. Worthington:
I am writing regarding the "Cloverleaf" affordable housing project. Although I am supportive of affordable housing, the number of structures proposed on the limited acreage, the impact this will have on traffic congestion and pedestrian traffic on Highland Road and the intersections of Routes 6 and 6A in downtown North Truro, is of great concern. I am also greatly concerned that Truro's wastewater regulations may be set aside, that the developer has requested a waiver from the regulations. My condo is in the center of No. Truro, on Highland Road. We already have to filter our water and use bottled water because the tap water gives us diarrhea.

I request that the Select Board do the necessary to encourage the Zoning Board of Appeal to deny the permit to develop the Cloverleaf project until and unless the developer chooses a method certain to keep water safe. My preference is that the developer reduce the number of units to conform to the natural leaching capacity of a 4-acre site. This request is due to both concerns about increased congestion, especially in the summer months, and concerns about further deterioration of the water quality.

I ask that my views and this request be read into the record. Thank you for your kind attention.

Alison Ellis

Trustee

4 Highland Road Condo Trust

From: dottyelms@comcast.net
Subject: Housing Project
Date: Jan 6, 2020 at 7:55:55 AM
To: Jan Worthington jworthington@truro-ma.gov

Dear Ms. Worthington,
I am writing to express my opposition to the 4acre housing project at Highland Rd. and Rte. 6.
Please account for my opposition at ZBA meeting.
Sincerely,
Dorothy and Michael Elms
122 Shore Rd.
North Truro, Ma.

Sent from Windows Mail

Do No Harm

As full time residents in Pond Village we beg of you to **Do No Harm** as you vote on the building permit for the Cloverleaf Project .

You are the Guardians appointed to protect our fragile ecosystem in the midst of progress.

Affordable housing is crucial to Truro but do not try to solve one problem by creating another.

Please Please with this project and many others going forward, **protecting our water supply needs to be first priority.**

Your decisions effect us ALL.

Sincerely,

Joe and Sharon Buteau
14 Pond Village Ave
North Truro

Jeffrey Ribeiro

From: Margaret Stewart <mgstewart1@msn.com>
Sent: Sunday, January 12, 2020 9:26 PM
To: Jeffrey Ribeiro
Subject: Cloverleaf Housing

I am writing to express my concern over the proposed affordable housing after attending the ZBA meetings on Dec 12 and 19. As a year round resident of North Truro, I worry about the impact this large complex will have on our already fragile water quality. Since the board has decided to hire an independent specialist to evaluate the situation, I will wait until their report is finalized to make judgement on the effect on nitrate levels.
I assume this report will be made public on the Truro website so that we may all be equally informed on this issue.

My next concern is how out of scale this project will be with the rest of Truro which is evidenced by the number of waivers that is required. While the ostensible reason for this housing is to assure that families and low income folks will have a place in our town, we are literally shoe horning them into a location at a density not only double that allowed by town law but also in no way representative of the lifestyle that attracts
people to live here. If we are sincere in wanting to diversify our citizenry I suggest we look at moving this housing
to the Walsh property where the space would allow for a true Truro environment.

I am also concerned with the cavalier attitude I sensed at the meetings about the abutters and other residents of
Highland Road which is now a quiet country lane. I saw no compassion for what those people are going to go through during the construction not to mention a permanent change to their neighborhood. I was surprised that the idea of bringing town water to them at the same time it is brought in for Cloverleaf was dismissed out of hand. I was thinking that we needed to relocate them for the duration of the construction at town cost.

I understand the board members are doing their best to evaluate this complicated project but it seems to me there is too much at stake for the North Truro community to make this decision in such a compressed timeframe.
If there was ever a time to measure twice and cut once this is it...the unintended consequences are frightening.

Sincerely,
Margaret Stewart

Sent from Mail for Windows 10

Jeffrey Ribeiro

From: Ruymann <ruymann@aol.com>
Sent: Monday, January 13, 2020 4:09 PM
To: Jeffrey Ribeiro
Cc: fwruymann@gmail.com; ruymann@aol.com; Jan Worthington; Robert Weinstein; Susan Areson
Subject: Cloverleaf project

Dear Members of the Truro Zoning Board of Appeals,

As residents of the Pond Village neighborhood, we have become aware of the debate surrounding the Septic Issues related to the Cloverleaf Project and wish to express our concern and to add some information for your consideration as you proceed with deliberations. Please distribute this letter to the members of the Truro ZBA prior to the January 16 meeting at Town Hal - 5:30 .

Let me preface this information by briefly sketching for you my 30+ year career as a GI physician. I took a position as a GI hospitalist at Cape Cod Hospital in Nov. 2017 so Karen and I could move full-time into our North Truro home. Prior to this I served for 10 years as Chief of GI for Mt. Auburn Hospital, and previous to that - as Chief of GI for Harvard Vanguard Medical Associates. One of my immediate concerns upon taking up my new position at CCH was the huge increase in pancreatic cancers I was diagnosing on residents of the Cape as opposed to my Cambridge and Boston populations. There seems to be no data to explain this phenomena, and as you may know pancreatic cancer is usually deadly.

Elevated Nitrate levels are known to cause cancers in animals, and studies suggest that they may be carcinogenic to humans as well. Much of the initial research is found in the WHO study of 2011 on Nitrites and Nitrates in drinking water. https://www.who.int/water_sanitation_health/dwq/chemicals/nitratenitrite2ndadd.pdf

In order to better understand the information I was receiving on the Cloverleaf Project, I researched and learned that "Nitrates cannot be removed from home drinking water with filters such as Brita, Refrigerator filters, PUR, or any other carbon-based filters. There are two accepted methods of nitrate filtering: reverse osmosis and ion-exchange technology, both of which incur significant cost to the homeowner." Private wells can have a high nitrate levels and reverse osmosis systems to reduce nitrate levels can be up to 70% effective.

Nitrates also react directly with hemoglobin in human blood to produce met-hemoglobin which destroys the ability of blood cells to transport oxygen. This condition is especially serious in babies under three months of age, causing a condition known as Methemoglobinemia or "blue baby" disease.

More recent research is alarming. As reported in *HealthDay News* in June 2019 (WebMD), nitrates are responsible for nearly 12,600 cases of cancer per year in the US. Another study further refined this information: Eighty percent of nitrate cancers were categorized as colorectal, ovarian, thyroid, kidney, and bladder. Since 1962 the federal standard has stood at 10 mg/l, but increased risk of cancer has been noted at 1/10 of that level OR just 1mg/l. (See June 2019 study by Environmental Working Group https://www.ewg.org/release/ewg-nitrate-pollution-us-tap-water-could-cause-12500-cancer-cases-each-year)

Elevated Nitrite levels (one fewer O2 atom than a nitrate) are easily converted biologically to nitrates in human digestive processes and have been suggested to increase the risk of gliomas, thyroid, and gastric cancers. Gliomas comprise about 30% of all brain and central nervous system tumors and 80% of all malignant brain tumors. (https://www.abta.org/wp-content/uploads/2018/03/glioblastoma-brochure.pdf) A very dear friend here in Truro died this past last year from glioblastoma.

We live on Pilgrim Pond and have seen the surface of the Pond covered with green slime during portions of the past three summers. We have learned that nitrates have the same effect on aquatic plant growth as phosphates and thus the same negative effect on water quality. Plant and algae growth are stimulated by increased nitrate and nitrite levels, but if algae growth is too robust, oxygen levels in the water are reduced and fish die. Nitrites produce a serious illness (brown blood disease) in fish (https://water-research.net/index.php/nitrate) .

The above data is merely a brief compilation of the research available, but is very concerning. We urge the ZBA and the Town of Truro to protect the future health of residents of the Highland Avenue, Pond Village, and Shearwater Neighborhoods by working with the developer of the Cloverleaf Project to insure the installation of a robust septic system that protects all of us from these health risks. We ask that these above comments be read into the record.

Sincerely,
Frederick W. Ruymann
Karen M. Ruymann
2 Bayview Drive
North Truro

Jeffrey Ribeiro

From: G Little <easthillfarm@hotmail.com>
Sent: Wednesday, January 15, 2020 8:10 AM
To: Jeffrey Ribeiro
Subject: Cloverleaf Complex Water Quality concerns - Please forward to members of the Truro Zoning Board of Appeals for us

We, Elizabeth McLarney and Gordon Little, have a home at 25 Priest Road in North Truro. We are extremely concerned about the water quality issues being raised with the Cloverleaf Housing Project. While we strongly agree that affordable housing for Truro residents needs to be a priority for our town, **we do not agree that the developer should receive a waiver to avoid installing and operating the necessary septic system requirements for this parcel to insure nitrate discharge to safe levels.**

We ask that you support requiring the developer to install and operate an appropriate septic system, or deny the requested waiver insuring that Nitrate levels from this project do NOT put safety, health and water quality of Cloverleaf residents and the residents of the surrounding area at risk.

Sincerely yours,
Gordon B. Little and Elizabeth A. McLarney
residents 25 Priest Road

From: dickluciem DickLucieM@nycap.rr.com
Subject: Clover leaf Project
Date: Jan 16, 2020 at 1:52:03 PM
To: Jan Worthington jworthington@truro-ma.gov

Dear Ms. Worthington -

This note is late, as the Truro ZBA meeting is tonight, but I HAD to write.

I am a civil engineer and I know that a septic system which will remove nitrates from household sewage for 70 BRs needs careful design and careful construction inspection. It cannot be done with a standard design. Approval of this project should not be done without the above.

Please take this into account and notify the ZBA of my comments.

Thank you,

Richard Murray
576 Shore Road
North Truro
110 Clarks Chapel Road
Nassau, NY 12123-2612
518-810-3514
dickluciem@nycap.rr.com

Jeffrey Ribeiro

From: Mary Ann Larkin <maryannlarkin2@yahoo.com>
Sent: Thursday, January 16, 2020 10:44 AM
To: Jeffrey Ribeiro; Town Planner
Subject: Cloverleaf project from Mary Ann Larkin

Dear Zoning Board Members:

First of all, thank you for your service. You are true exemplars of democracy.

I've been reading about the Cloverleaf project in the papers and in e-mails people send me and I've come to the conclusion that more information is needed.

We need at least three educated opinions about what will happen with the additional housing. I saw that this board asked Mr. Malone to hire a consultant to weigh in on the issue. That's a good start. However, we need more than the builder's consultant's opinion. As you all know, the Association for the Preservation of Cape Cod has just completed a major study on the Cape's water. They would know of an expert who could evaluate this problem. Also, we have major universities such as MIT and Harvard who would have experts. Such experts will involve fees but with the purity of water at stake, we need to have experts. Perhaps some generous donor will step forward to pay these fees?

Finally, I have read that some processing system installed at the Cloverleaf could solve this problem. That also needs to be examined by experts. What if the system is put in and does not solve the problem?

I'm not sure why all these issues did not come up earlier, but people are busy and circumstances change, so now is the time to go forward to establish expert answers. So, I think it would be best to postpone any decision until we have more definitive answers to these questions. No one wants a third of the town to have a problem with water or even to be frightened that they are going to.

I feel sure that these issues can be explored and solutions found so that this wonderful housing project can go forward with the full support of everyone in Truro.

Thanking you again for your service,

Mary Ann Larkin
12 Pond Road
N. Truro MA 02652
202 832-3978

ps Please read this aloud at the meeting if that fits your format!!

March 12, 2020

Dear Truro Zoning Board of Appeals,

Thank you for your time and attention regarding this important matter. Please read this into the record.

As we know, the applicant/developer of the Cloverleaf project is seeking a waiver from the town's Health Regulations. These Health Regulations are designed to protect public health and drinking water supplies from sewage contamination. They limit the amount of wastewater discharge to 440 gallons/acre (this is also a Massachusetts Title 5 regulation). According to the town's consultant Horsley Witten Group this means that the Cloverleaf parcel can accept up to 1900 gallons/day. The Applicant proposes 7871 gallons/day. This is more than four times the acceptable rate.

To further assess the water quality impacts on nearby drinking water wells, Horsley Witten Group conducted a nitrate loading analysis. The State's EPA sets maximum levels of nitrates in groundwater at 10 mg/L and Truro sets a safe level at 5 mg/L. It has been established that high nitrate levels in ground water are deeply concerning and can cause serious health problems. Based on the consultant's review, in my opinion there are three main issues:

1) According to Horsley Witten Group's calculations the proposed system does not meet the 10 mg/liter nitrate drinking water standard at the property limit.
2) The proposed upgrade to a FAST system (rated at 20 mg/liter) does not meet the water quality standard.
3) Horsley Witten Group's letter suggests that alternative septic systems such as a Nitrex system "could potentially" (emphasis on the uncertainty here) have better results. However the Nitrex system is currently only provisionally approved by the DEP at a concentration of 25 mg/L, similar to the FAST system (see link: https://www.mass.gov/doc/nitrex-by-lombardo-associates-inc-provisional-use-approval/download). It could potentially gain General Approval at a lower concentration at some point in the future, but currently this does not exist. The only way to reliably meet the 10 mg/liter standard is to construct a conventional wastewater treatment plant permitted through MADEP's Groundwater Discharge Permit Program (314 CMR 5.00).

It is my understanding that future hearings will be held to review the proposed response to the peer review by the applicant. I, and I'm sure all concerned citizens, very much appreciate the high level of scrutiny and due diligence that the ZBA hopefully will continue to undertake to mitigate the potential negative impact on our ground water caused by this project. As far as I can determine, this negative outcome can be mitigated by one of two things occurring: (1) the project is significantly scaled down to be within the town's stated guidelines which are there to protect the citizens of Truro and the environment; or (2) the developer pays for the installation and operation of a Groundwater Discharge Program (under no circumstances should this costly system be shouldered by the taxpayers).

Thanks again.
Sincerely,
Kate O'Brien
24 Bayberry Road, Truro year-round resident

Submitted by Peter Herridge at 3/12/2020 Hearing

Journal Article submission to the ZBA

This packet contains five recent journal articles from well respected, refereed journals on medicine and environmental research. These articles are being submitted to be made of record in the Public Hearing held by the Zoning Board of Appeals (ZBA) of Truro, Massachusetts concerning the decision to grant a waste water waiver to the proposed Cloverleaf housing project held on March 12, 2020. It is the intention of this submission to demonstrate to the ZBA the fact that nitrate pollution in drinking water poses very serious risks of adverse medical consequences to people who drink this water on both an acute and prolonged basis. These articles also clearly show that even levels of nitrates that are below the maximum nitrate level presently allowed in the US, i.e. 10 mg of nitrogen as nitrate per liter of water, can cause a wide variety of serious medical consequences including cancer, birth defects, spontaneous abortion and thyroid disease.

The contents are:

1) *Exposure-based assessment and economic valuation of adverse birth outcomes and cancer risk due to nitrate in United States drinking water*. Author: Temkin A
2) *Nitrate toxicity and drinking water standards*. Author: Kross BC
3) *Thyroid cancer induction: Nitrates as independent risk factors or risk modulators after radiation exposure, with a focus on the Chernobyl accident*. Author: Drozd VA
4) *Drinking water nitrate and human health: an updated review.* Author: Ward MH
5) *Prenatal nitrate intake from drinking water and selected birth defects in offspring of the participants in the National Birth Defects Prevention Study*. Author: Brender JD

This packet is submitted to the Truro ZBA on March 12, 2020 by Peter Herridge MD, JD.

See end of packet for full journal articles.

Jeffrey Ribeiro

From: c.e.steinman <c.e.steinman@comcast.net>
Sent: Sunday, June 14, 2020 4:04 PM
To: Jeffrey Ribeiro; Art Hultin
Cc: Rae Ann Palmer
Subject: Comments for the ZBA Public Hearing Regarding the Cloverleaf Project
Attachments: Cloverleaf_Option_6-11-2020.pdf; Original cloverleaf_Site_Plan.pdf

To the Zoning Board of Appeals;:

I request that the following comments be read into the public record regarding the proposed Cloverleaf Project.

I had previously been in support of Building 21 with its fifteen apartments and commons spaces serving as a congregate housing component of the proposed Cloverleaf Project. Having had a 25-year career in the interior design for senior housing, including retirement communities, assisted living, nursing and Alzheimer care projects, I thought the proposal for Building 21 would offer Truro additional housing opportunities for our aging population. However, the devastating impact of the Coronavirus on seniors in congregate housing is a game changer.

With the uncertain future of the COVID-19 pandemic, Building 21 as currently designed might not be manageable, and importantly, not marketable either. Individual-entry apartments as elsewhere on the site are a safer option.

Attached is a concept sketch for replacing the large congregate building with three town-houses, similar in design to units 2, 4 and 6. Taking advantage of the site's topography, there could be six ground-floor apartments entered on the front and back of the town-house buildings that will have at-grade access. Those apartments could be designed for accessibility, similar to unit 2A. This would reduce the number of units in Building 21 from 15 to 9 in the town-houses, resulting in an overall reduction of 6 units. The total units will decrease from 40 to 34. At the same time, the redesign could increase the number of accessible units from 4 to 7.

This proposal is not trying to solve all the water quality issues presented in the peer engineering review, which may require appropriate on-site septic treatment and possibly an overall reduction in the number of units. However, the proposed design change with its reduction in the number of units could be a step in the right direction to help resolve the water quality issue while also making the project more beneficial to our community.

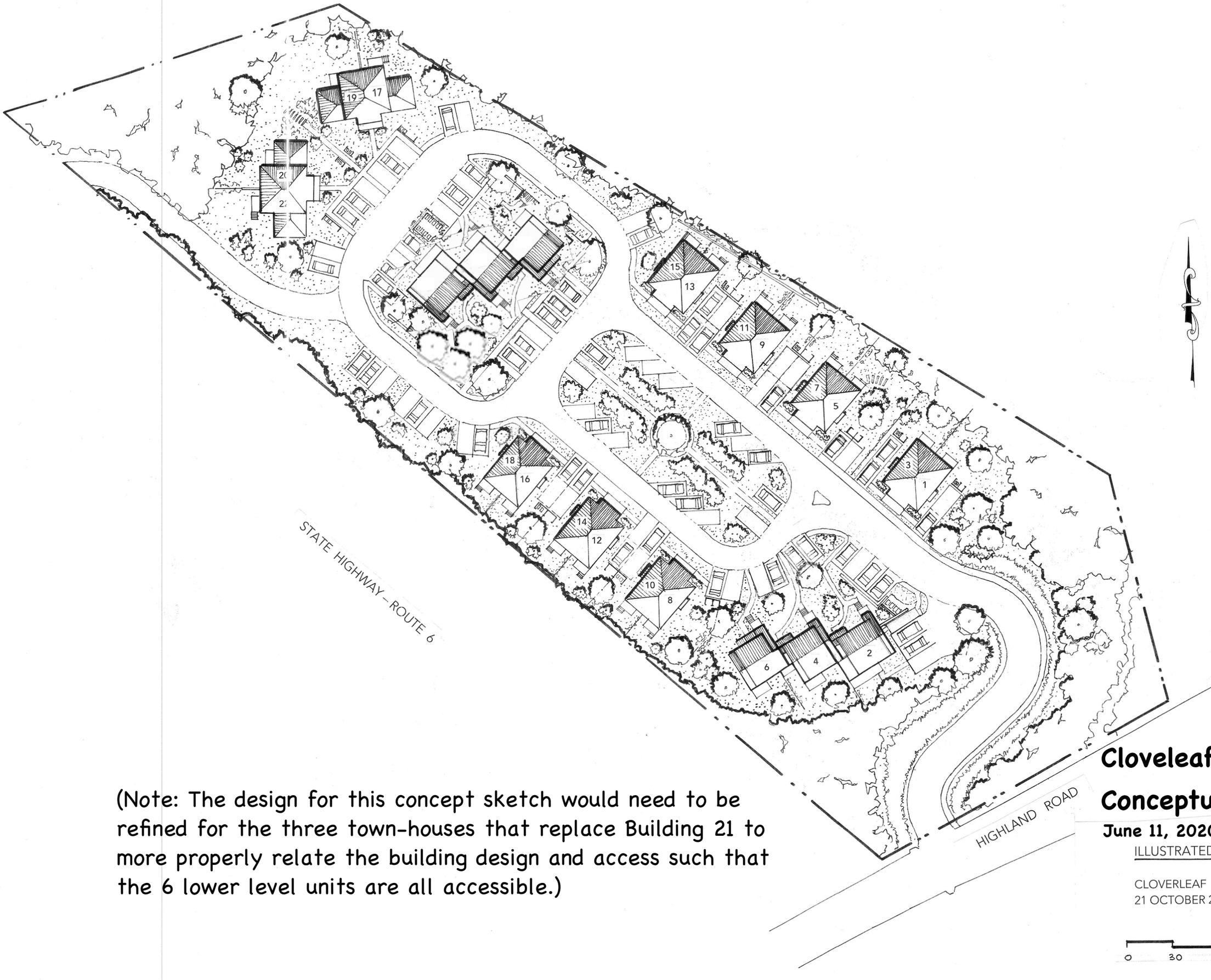
Thank you for the opportunity to comment and for your consideration,

Chuck Steinman
c.e.steinman@comcast.net
cell 617-974-1613

PO Box 781/Shore Road
North Truro, MA 02652

Cloverleaf Option Conceptual Site Plan, Replacing Building 21 with 3 Town-House Buildings:

Cloverleaf Original Illusrative Site Plan Showing Building 21:



Cloveleaf Option
Conceptual Site Plan

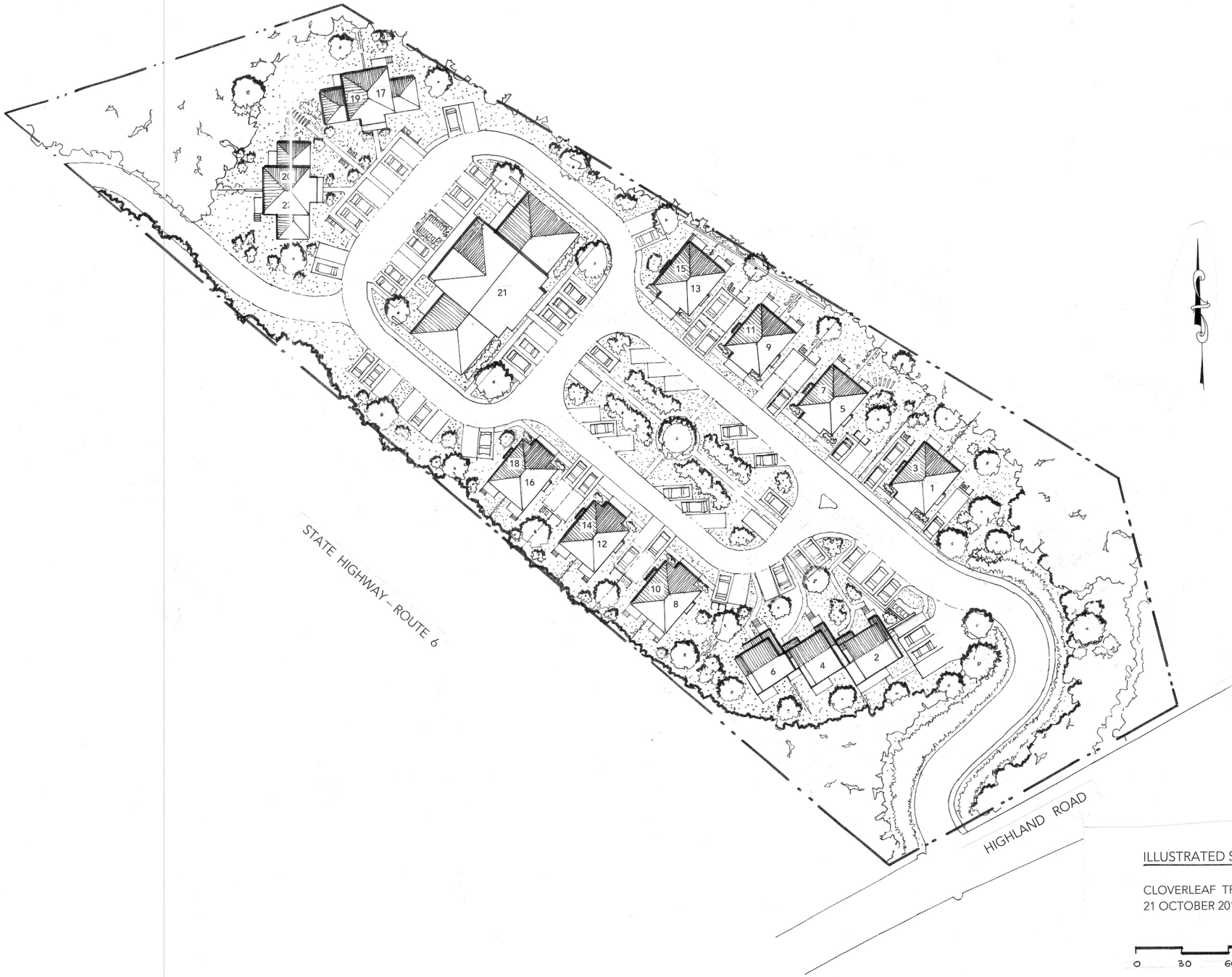
June 11, 2020

ILLUSTRATED SITE PLAN

CLOVERLEAF TRURO RENTAL HOUSING
21 OCTOBER 2019

0 30 60 90

(Note: The design for this concept sketch would need to be refined for the three town-houses that replace Building 21 to more properly relate the building design and access such that the 6 lower level units are all accessible.)



ILLUSTRATED SITE PLAN

CLOVERLEAF TRURO RENTAL HOUSING
21 OCTOBER 2019

0 30 60 90

Jeffrey Ribeiro

From: Steve Sollog
Sent: Thursday, June 25, 2020 12:58 PM
To: Jeffrey Ribeiro
Cc: Steve Sollog
Subject: To be read into ZBA meeting 6/25/20 5:30pm

In light of all the sacrifices and change due to the Covid crisis, why is this project not shelved until normal town participation can be resumed? There is going to be great financial upheaval the extent of which is not known at this time. Employment opportunities have evaporated, budget restructuring will be necessary. The unknowns should dictate a slowing down of this project.
Thank You
Steven Sollog

Jeffrey Ribeiro

From: jack riemer <jackriemer@hotmail.com>
Sent: Thursday, June 25, 2020 1:15 PM
To: Jeffrey Ribeiro
Subject: Truro ZBA June 25 2020

Jeffery,
Please accept the following to be read into the record during Public Comment at to-nights hearing:

1 Will the Cape Cod Commission review the revised plan?
2 What was the original cost per unit and what will be the cost per unit with the revised plan?
3 The Bio Microbics system at Noquochoke Village in Westport MA is on a 30 acre site , the Cloverleaf Development is less than 4 acres , why can't we have an example which is more representative of what we have in Truro?

Thank you,

Jack Riemer

Jeffrey Ribeiro

From: Peter Herridge <pherridge51@gmail.com>
Sent: Thursday, June 25, 2020 1:59 PM
To: Jeffrey Ribeiro
Subject: Question to be read at ZBA hearing

Dear Jeff,
I would like to have a question read at tonight's ZBA meeting and it is this; "will the engineering company that did the Peer Review be reviewing this new proposal and publicly reporting their finding and if so when"

Thank you.
On a different note I have to tell you that the PB and I will both really miss you and the excellent work you have done for us. You are the best planner we have ever had and the most professional. Our loss is the Cape Cod Commissions gain. I wish you all the best in you career which I have no doubt whatsoever will continue to be a great success.
Take care of yourself,
Peter

Jeffrey Ribeiro

From: Andrea Aldana <andrea@capecdp.org>
Sent: Thursday, June 25, 2020 2:31 PM
To: Jeffrey Ribeiro
Subject: Public Comment submission for 6/25 Cloverleaf Hearing
Attachments: Public Comment_Cloverleaf_6.25.2020.pdf

Importance: High

Hello Jeffrey,
As you may know, unfounded concerns about housing density and the spread of the coronavirus have been emerging across the state (and country) and may come up at tonight's hearing. I would like to submit the attached comments which debunk those concerns. Please distribute the document to the ZBA.

Thank you,

Andrea Aldana
Director of Housing Advocacy
Community Development Partnership
Capecdp.org **I** (508) 240-7873 x10 I andrea@capecdp.org

Submitted by:
Andrea Aldana
Director of Housing Advocacy
Community Development Partnership
Andrea@capecdp.org

Myth: affordable, dense housing will increase the spread of Covid-19

- Historically, many campaigns against density have been justified as a matter of health and sanitation. In fact, the same arguments started turning up in the Boston metropolitan area earlier in the pandemic and it appears that these inaccurate ideas have made their way to the Outer Cape.
- For the uninformed, the pandemic can become a rallying cry to maintain our sprawling suburban style neighborhoods that are designed to foster exclusion rather than inclusion while gobbling up open space and using land inefficiently.
- Yet this pandemic is reminding us that we need communities where teachers, child- and elder-care workers, nurses, doctors, janitors, construction workers, and baristas all share in the prosperity and the comfort of an affordable home.
- Let's not forget, we have a housing crisis that existed before the pandemic and the same crushing housing-cost burdens will exist after the pandemic. In fact, housing providers in the region are concerned that the pandemic will worsen the housing crisis due to loss of employment and increased expenses.
- Plus, our untenable and unjust rent burdens are disproportionately felt by service and essential workers whom we depend on, now more than ever.
- To blame density for the devastation caused by the coronavirus is to ignore all of the factors that really determine how badly communities suffer in crises such as the present one.
- In fact, no correlation exists between population density and rates of COVID-19 infection, according to recent studies examining the disease in China and Chicago.
- Crowding within homes is, indeed, bad for public health. Policies and economic inequity have forced many low-income people to live in crowded households. But density and crowding are different things. Crowding is what happens when, because of a lack of enough housing, families and roommates are forced into tight quarters designed for a smaller number of inhabitants. That crowding can increase the spread of a contagion.
- In contrast, density allows people to live in uncrowded homes under safer, more sanitary conditions.

- For example, while the nation's densest city, New York, experienced a severe COVID-19 outbreak—more than 195,000 confirmed cases and more than 19,000 deaths to date—one must simply look at our nation's second-densest city, San Francisco, to see a very different picture. San Francisco has so far experienced far fewer infections, with more than 2,000 confirmed cases and 36 deaths. Though New York City has approximately 10 times as many people as San Francisco, the former has suffered more than 500 times the deaths. One key reason is that the Bay Area responded to the pandemic earlier and more decisively than New York did, imposing social-distancing measures before major cities on the East Coast.
- Furthermore, the world's best coronavirus fighters so far are all in Asia where cities are denser than almost any place in North America: Hong Kong, Singapore, Seoul, and Taipei.
- An analysis by New York University's Furman Center found no relationship between the coronavirus and overall population density within New York City, with neighborhoods in Manhattan, the city's densest borough, having some of the lowest infection rates. However, the study did find that the virus is more prevalent in areas of New York where more people are crowding into homes.
- This contagion is not about whether you live in a densely populated area or a less densely populated area; it's about whether you have a good public health response to a pandemic, and Hong Kong and Singapore had a fantastic response," said California Sen. Scott Wiener in a *Politico* article.
- According to a recent study in the Journal of the American Planning Association (Journal of the American Planning Association, 2020, Vol. 0, No. 0):
    - "Density is not linked to rates of COVID-19 infection, after controlling for metropolitan area population, socioeconomics, and health care infrastructure in U.S. counties."
    - "COVID-19 death rates are lower in denser counties and higher in less dense counties, at a high level of statistical significance. This is likely due to better access to health care facilities and easier management of social distancing interventions such as sheltering in place."
    - "The fact that density is unrelated to confirmed virus infection rates and inversely related to confirmed virus death rates is important, unexpected, and profound."
    - "The role of planners and local governments in addressing pandemic outbreaks is crucial, but not through advocating for the low density and suburban types of development. Rather, planners and local governments play a key role in adopting measures tailored to their community for more effective implementation of social distancing measures and to mitigate the adverse impacts on businesses, households, and citizens."

Jeffrey Ribeiro

From: Eric Parker <bondeen1@comcast.net>
Sent: Thursday, June 25, 2020 3:21 PM
To: Jeffrey Ribeiro
Subject: Cloverleaf Hearing Timeline Question

Dear Mr. Ribeiro,
If you would, please ask this question or answer it at today's Cloverleaf public hearing, on my behalf.

Today's hearing is a resumption of public hearings on the Cloverleaf Project that had been placed on hold a few months back, due to the problems created for conducting public hearings, introduced by the COVID-19 pandemic.
Can you inform me and the town voters as to the effect on the 40B timeline originally stated and where the hearing schedule is currently at in that timeline? Has the Governor, in his declarations regarding impacts and changes to governing, specified anything regarding 40B timelines? I
Please let us know, if the timeframe is now more open-ended or, are there specific dates where hearings must end for the ZBA to render their decision(s) for this project.
Thank you.
Regards,
Eric Parker
2 Waterview Heights Road
North Truro, MA 02652

6/25/2020

To the Truro Zoning Board of Appeals,
J. Ribiero, Town Planner

I would like to comment on the current ZBA Agenda which is to decide whether to grant a Comprehensive Permit for the Community Housing Resource , Inc. Cloverleaf Project.

Truro currently has many environmental issues to assess - some more urgent than others. Because the proposed development is very large requiring massive and potentially harmful changes to the area I strongly suggest that the Truro town committees created to oversee important issues affecting Truro's environment , health and safety should be part of this permitting process. The three committees charged with this responsibility and who have the relevant expertise are the Energy Committee, the Climate Action Committee and the Water Resources Oversight Committee.
It would appear that choosing to ignore the professional input of these committees could lead to a decision with an outcome that is not as good as it should be. The ZBA deserves the benefits of this readily available expertise.

Sincerely,

Lucy Clark
7 Benson Road,
Truro, Ma 02666

June 25, 2020

Dear Members of the ZBA,

I wrote to the members of this committee in December, expressing my support for the Cloverleaf Project. At that time, I spoke about how important this project was to Truro residents like myself who are older. It is especially important to those Truro residents who are looking to "downsize".

Over the last months, many people who are older have "sheltered in place" in their homes in Truro. Those people often lived alone and were solely responsible for the upkeep of their homes during this crisis. They, as so many others, may have had their income affected by COVID19. I believe that more residents who are older are going to be looking to "downsize". This population's need for affordable housing is especially urgent now.

The apartment building that is part of the design of the Cloverleaf is the most likely building to address the needs of people who are older. The layout of that building allows more people with mobility problems to reside in the Cloverleaf complex. After this crisis has passed, the common areas that are part of the design of the apartment building will be a gift to the residents, especially older people living alone.

There had been some early indications that the density of housing might be a factor that contributes to the likelihood of people contracting COVID 19. But as research was done on the factors affecting transmission of the virus, there was no evidence that the density of the housing increased the risk of contracting the virus. There is research that the greater density of residents within a housing unit can have an impact on those residents' health. We also know that people living in nursing homes were at a greater risk of contracting the virus. Obviously, that is not the type of density that exists in the Cloverleaf apartment building. These apartments must meet stringent state standards for the number of people who can occupy each apartment. If when this housing becomes available, Covid19 continues to be a health crisis or we face a similar crisis in the future, the Cloverleaf apartments will provide safe and manageable place for people to "shelter in place".

This pandemic has highlighted the bravery and dedication of our healthcare providers. The need for more healthcare workers on the lower Cape was made very clear during this crisis. Also, more of us are going to face medical issues as we age. We are going to need more health care professionals such as visiting nurses or

physical therapists, etc. to help us with those issues. Yet many of those workers are going to need housing that is affordable to live here. The Cloverleaf Project is important to meeting those needs.

I am fortunate to live in a year-round rental very close to the Cloverleaf Project and am looking forward to those new neighbors.

Thank you for your consideration,

Betty Gallo

2 Pond Rd. Unit 2
North Truro

Jeffrey Ribeiro

From: Joanne Hollander <joanne.hollander@icloud.com>
Sent: Thursday, June 25, 2020 5:14 PM
To: Jeffrey Ribeiro
Subject: The Cloverleaf Project

Dear Mr. Ribeiro,

As a Truro resident living at 13 Tom's Hill Path, and a food product developer for several decades, my concern for water safety in Truro is paramount. We all share the Pamet aquifer as a source of our sole source of drinking water, which is already stressed from crumbling septic systems and the threat of salinity contamination from climate change rising tides.

What assurance do we have for the safety of our water with the proposed septic system for the Cloverleaf project? With so many people due to reside on 4 acres of land, how can we be assured that our water will not be contaminated further than it already is with carcinogenic chemicals such as PFOS and PFOA and MTBE and Glyphosate, not to mention nitrogen contamination which will likely occur with so many people depositing wasted in the septic system on such a small piece of land. Who has studied this on behalf of the Town of Truro?

What assurance do we have that this system will not fail, contaminating our precious drinking water? Has this project and septic system been reviewed by the Board of Heath, the Climate Action Committee, the Water Resources Oversight Committee and the Energy Committee?

How do we know and how can we trust that this project is in the best health interest for Truro residents?

Sincerely interested and concerned for all of our well-being,

Joanne Hollander

Jeffrey Ribeiro

From: Brian Boyle <beboyle@aol.com>
Sent: Thursday, June 25, 2020 5:15 PM
To: Jeffrey Ribeiro
Cc: mark@markfarber.com; rehigginssteele@gmail.com; jasn54@gmail.com; rtschwebel@gmail.com
Subject: COMMENTARY FROM TRURO ENERGY COMMITTEE for CLOVERLEAF HEARING

TO: Truro Zoning Board of Appeals, Truro Select Board, Town Planner
SUBJECT: COMMENTARY FROM TRURO ENERGY COMMITTEE for CLOVERLEAF HEARING

As some of you know, the Energy Committee has been actively engaged in the Cloverleaf project since late last year. We have had several meetings with those involved in the project and have several open requests for additional information.

Unfortunately, when the Covid-19 epidemic hit our state, our committee was essentially shut down, unable to meet, even by digital means. It has been just a few days now that we have had that ability, and our first meeting will occur next week. At that time we plan to initiate drafting a memo for your consideration.

That memo will summarize our work to date on increasing the sustainability of the project, and at the same time trying to make it even more affordable. We firmly believe that sustainability and affordability are not mutually exclusive.

However, to date we have not seen plans that give us any confidence that sustainability goals will be met. Moreover, if they are not, and the state mandates further increases in sustainability, as we believe they will, it could entail costly retrofits to the project. That would adversely impact the affordability of the few units designated as affordable.

It would be a shame if Truro went to all this trouble and ended up with a project that was neither sustainable nor truly affordable. And for that reason, the Energy Committee has been diligently pursuing this project, and willing and able to share our expertise, and questions still remaining.

Brian E. Boyle
Truro Energy Committee Chair

Jeffrey Ribeiro

From: Jim Nash <jasn54@gmail.com>
Sent: Thursday, June 25, 2020 6:22 PM
To: Jeffrey Ribeiro
Subject: ZBA Meeting Question Regarding Energy Efficient and Carbon Reducing Design Characteristics

TO: Truro ZBA, Truro Select Board, Town Planner
SUBJECT: Question Regarding Energy Efficient and Carbon Reducing Design Characteristics

My name is James Nash. I am a year-round Truro resident and own a home at 1 Captain Williams Way.

I would like the Cloverleaf Developer to please describe the timeline and their anticipated process to engage with qualified professionals and Town Committees to ensure optimal design characteristics and compliances and that meet and/or exceed jurisdictional energy efficiency goals.

I ask this question because I am aware of numerous opportunities for the Developer to work with local and State agencies that would provide services and grants to assist the Developer to incorporate cost effective and attainable fast-developing energy efficient design characteristics.

As an example, Massachusetts has set ambitious goals for carbon reduction and offers generous assistance to developers by providing a $6,275.00 grant per unit to defray costs for a tighter building envelope. This would result in immediate carbon reductions and allow for a 7 year payback to the developer of any additional costs to the project.

Eversource also works cooperatively with Developers to provide underground infrastructure to allow for EV charging stations. Inclusion of roof to ground-level internal conduits allowing the units to be at least solar-ready, if not solar-energy equipped, would also be desirable.

Thank you,
James Nash
508-274-4095

Jeffrey Ribeiro

From: c.e.steinman <c.e.steinman@comcast.net>
Sent: Friday, June 26, 2020 9:48 PM
To: Jeffrey Ribeiro; Art Hultin
Subject: Additional Comments for the Cloverleaf Housing Proposal
Attachments: Coronavirus Crisis Threatens Push for Denser Housing - The New York Times.pdf

Zoning Board of Appeals:

I understand that my concerns about the impact of COVID-19 on the design of Building 21 were challenged by a caller during the Public Hearing.

In support of my previous submittal, I would like to submit for the public record an article entitled ***Coronavirus Crisis Threatens Push for Denser Housing,*** by Kevin Williams, New York Times, May 5, 2020, as updated May 6, 2020..

Please see the sections in the attached copy of the article that are annotated with a red star in the margin. These sections are quoted as follows:

> "The whole discussion about housing will change. A lot of the bills and laws the Legislature have been discussing will be looked at in a different lens," he [Isaiah Madison, a board member of Livable California, a nonprofit group] said.
>
> "I wouldn't make any big development decisions right now," said Dr. Jackson, a former officer in the Epidemic Intelligence Service at the Centers for Disease Control and Prevention.
>
> The economic fallout is likely to last five years or more, he added, and people may be wearing masks for several years. Developers will have to factor the pandemic, and other crises, into their plans.
>
> "You have to plan out 100 years for building residences and creating buildings that are resilient and confront a multitude of hazards: terrorism, earthquakes, fires, climate change, energy shortages," Dr. Jackson said.
>
> The desire for denser developments might diminish, he [Mr. Youngentob, a Maryland developer] said, and his company may switch its focus to townhomes.
>
> "The forced interaction of sharing doors and elevators has caused some anxiety," Mr. Youngentob said. "Townhomes, where you come in and out of your door, and you know you are the only one touching your door handle, provide some comfort."

Thank you for your consideration,

Chuck Steinman
c.e.steinman@comcast.net
cell 617-974-1613

PO Box 781/Shore Road
North Truro, MA 02652

Zoning Board
Cloverleaf Complex
PO Box 2030
Truro, MA 02666

June 26, 2020

To: Zoning Board Members
Re: Cloverleaf Bus Stop

I am writing in reference to a suggestion made at the virtual meeting on 6/25/20 concerning the addition of a bus stop to be located in the area at the entrance to the Cloverleaf Complex at 22 Highland Rd. A bus stop already exists 150 yards from this site on the same side of the street, on the other side of the bridge. This stop contains a shelter and an off-street driveway where the bus comes in off of Highland and customers wait and enter the bus away from the main road. A sidewalk from the Cloverleaf Complex could be put in and attached to the sidewalk which is already in existence under the bridge and extended the remaining short distance to this area.

The area requested for the new bus stop is not part of the original land acquisition allocated to this development. As mentioned by Mr. Ribeiro, it belongs to the county and the town would have to acquire it for this purpose. The land in question already contains a large Verizon relay box. It doesn't have enough remaining area to provide safety issues such as protection from rear-end collisions, a shelter from the weather, or a safe area for the loading and unloading of passengers as exists at the current site.

I am also concerned about the proximity of the new bus stop to the on and off ramps for Route 6. A bus stop in this area would cause congestion and confusion with cars entering and exiting the ramps along with cars proceeding straight up and down Highland Road in addition to cars coming in and out of the complex. Having a bus stopped in this area would interrupt the flow of traffic presenting a safety hazard.

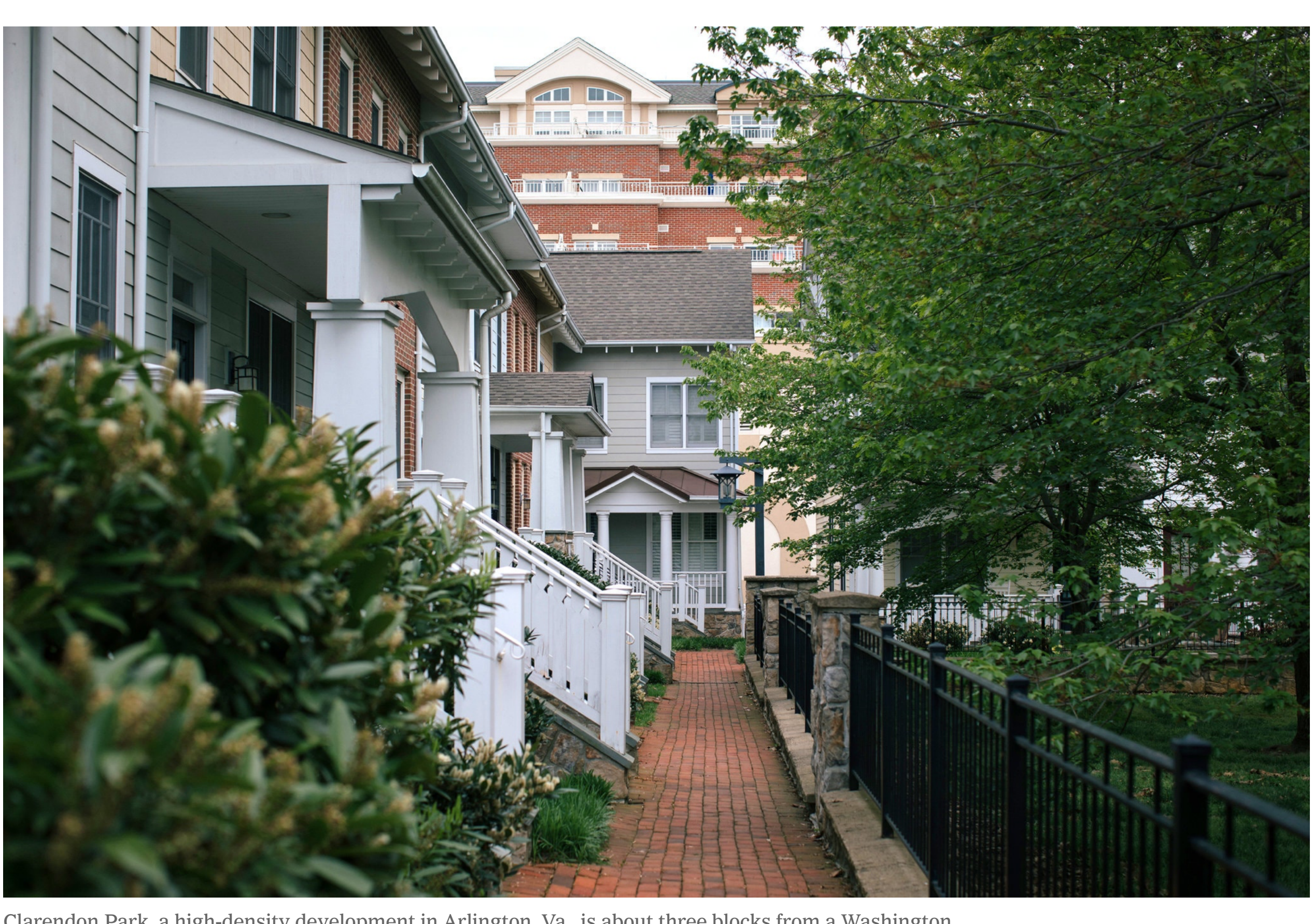
Further site review and a formal summer traffic study should be done before any further actions are taken.
Respectfully,

Lauren Anderson
30 Highland Rd
North Truro, MA 02652

SQUARE FEET

Coronavirus Crisis Threatens Push for Denser Housing

Transit-oriented developments were seen as a solution to severe housing shortages, but experts say developers need to rethink the design for a post-pandemic world.



Clarendon Park, a high-density development in Arlington, Va., is about three blocks from a Washington Metro station. Alyssa Schukar for The New York Times

By Kevin Williams

Published May 5, 2020 Updated May 6, 2020

SILVER SPRING, Md. — Katie and Timothy Carney were searching for a larger home to accommodate their growing family. But equally important to them was finding a place within easy distance of Washington's Metro rail line. They finally pounced when they saw a 2,500-square-foot colonial-style house in a mixed-use development less than a mile from the nearest rail stop in Silver Spring.

"He can leave his office downtown and be home in 50 minutes," Mrs. Carney said of her husband, who works as a journalist at The Washington Examiner. He can walk to the transit stop in less than 15 minutes.

That's music to the ears of planners and housing advocates trying to address the housing crisis ravaging cities like San Francisco and Seattle. But some developers worry that the coronavirus pandemic will stop the momentum as social distancing and telecommuting become the norm.

Transportation and denser housing have been the two focal points of urban residential development for the last decade, as cities try to combat a severe shortage of affordable housing. In areas where car commute times continue to climb, and freeways are at capacity, building denser communities along transit lines is seen as a panacea.

These projects, known as live-leave developments or more formally as transit-oriented developments, can be no-frills projects that focus on housing and getting people in and out fast. Or they can be more centered on amenities, meant to attract not only residents but commercial developers who find the density attractive for restaurants, coffee shops and boutiques. As an added benefit, developers can usually forgo expensive surface parking lots on prime real estate, as most residents use public transportation.

In California, legislation has been proposed to change zoning restrictions to make building transit-oriented developments near light rail lines easier. The bill was narrowly voted down in January because of concerns that it would strip local communities of too much control.

But the need for transit-oriented developments will not go away, even with the pandemic, said the bill's sponsor, Senator Scott Wiener, a Democrat who represents San Francisco.

"We need to take a deep breath and do the things we know will put an end to the pandemic," Mr. Wiener said, referring to the need for testing and a vaccine. But after the pandemic ends, California will still have a staggering homelessness problem of more 100,000 people that can be addressed only by building more housing, he said.

Bob Youngentob, chief executive of EYA, which builds transit-oriented developments in the Washington area, says the demand for that kind of housing could be tempered as Americans adapt to telecommuting. Alyssa Schukar for The New York Times

Transit-oriented development also carries economic weight. Developments at six stops along the Gold Line in Pasadena, Calif., have attracted 3,500 housing units and created 250,000 square feet of retail space, along with 603,000 square feet of office space, 421,000 square feet of hotel space and 306,000 square feet for other commercial construction, according to a 2016 study by Beacon Economics, an independent research firm in Los Angeles.

Those numbers are comparable to what has been seen in other markets, said Adam J. Fowler, research director of Beacon Economics. He agreed that the pandemic would not change the need for transit-oriented development.

But Isaiah Madison, a board member of Livable California, a nonprofit group that promotes local control, thinks the pandemic will force legislation on transit-oriented development in new directions.

"The whole discussion about housing will change. A lot of the bills and laws the Legislature have been discussing will be looked at in a different lens," he said.

Most experts say that the demand for transit-oriented development will still exist in some form after the crisis, but that the pandemic will leave a legacy.

Developers should take heed of the long-term effects of the pandemic, said Dr. Richard J. Jackson, professor emeritus in the department of environmental health sciences at the Fielding School of Public Health at the University of California, Los Angeles, who has studied transit and development.

"I wouldn't make any big development decisions right now," said Dr. Jackson, a former officer in the Epidemic Intelligence Service at the Centers for Disease Control and Prevention.

The economic fallout is likely to last five years or more, he added, and people may be wearing masks for several years. Developers will have to factor the pandemic, and other crises, into their plans.

"You have to plan out 100 years for building residences and creating buildings that are resilient and confront a multitude of hazards: terrorism, earthquakes, fires, climate change, energy shortages," Dr. Jackson said.

In the same way that better sanitation came after the Spanish flu 100 years ago, post-pandemic innovations for commercial developers will emerge, said Jennifer D. Roberts, an assistant professor of kinesiology at the University of Maryland School of Public Health who has studied human health as it relates to commuter rail proximity.

"This is an opportunity to think in new ways, but people will still want to live close to transit," she said.

The challenge for developers will be marrying density with safety, which will now require an interdisciplinary approach, Dr. Jackson said.

The era of a single architect designing buildings is over, he said, and transit-oriented development will need to bring in the best minds from design, health and transit to come up with living spaces that are conducive to community but also the well-being of residents.

The Coronavirus Outbreak >

Frequently Asked Questions and Advice

Updated June 16, 2020

- **I've heard about a treatment called dexamethasone. Does it work?**
  The steroid, dexamethasone, is the first treatment shown to reduce mortality in severely ill patients, according to scientists in Britain. The drug appears to reduce inflammation caused by the immune system, protecting the tissues. In the study,

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Developers are already starting to consider new design plans.

Transit-oriented developments "will be much, much more focused on public health," said John W. Hempelmann, a lawyer in Seattle and a former chairman of the Transit-Oriented Development Council of the Urban Land Institute.

Expect more open spaces, broader sidewalks, slimmer roads and promenades in the future, Mr. Hempelmann said. Consideration for social distancing, along with more robust preparation, will mitigate the effects of any future pandemics, he added.

Seattle has several transit-oriented developments under construction, including a 24,240-square-foot project in the University District, which will feature a mixed-use high-rise with more than 200 apartments a few blocks from a planned light rail stop.

"No one knew what T.O.D. meant 15 years ago," Mr. Hempelmann said. "Now, it is an extraordinarily popular concept."

EYA is building the Townhomes at Reston Station, a development about three blocks from a Washington Metro station in Reston, Va. Alyssa Schukar for The New York Times

But the excitement around transit-oriented development could be tempered as Americans adapt to telecommuting, decreasing the need for some to live close to mass transit, said Bob Youngentob, chief executive of EYA, a developer in Maryland that builds transit-oriented developments in the Washington area.

The desire for denser developments might diminish, he said, and his company may switch its focus to townhomes.

"The forced interaction of sharing doors and elevators has caused some anxiety," Mr. Youngentob said. "Townhomes, where you come in and out of your door, and you know you are the only one touching your door handle, provide some comfort."

Despite worries over the pandemic, Mrs. Carney, the Silver Spring resident, said the pandemic had not made her regret her family's decision to move near Washington's rail line.

"The hope is that this is not permanent, and this is why it is still worth it," she said. "If it were permanent, it might change my thinking."

To the Select Board and the Zoning Board of Appeals

STATEMENT ON CLOVERLEAF HOUSING DEVELOPMENT

To be a sustainable community, we need to have rental housing that will enable people to live here who cannot afford to buy, build, or rent a home at Truro's high market prices. The Cloverleaf property offers an opportunity to address this need. At first, most of us saw great promise in the proposal to develop the property. But much has changed. New concerns have emerged that cry out for consideration. Many in Truro believe we should reconsider the scope and some of the shortcomings of Cloverleaf Housing, but have been reluctant to speak out thus far because people who raise questions or concerns are immediately labeled "against affordable housing." This accusation is unfair, untrue, and very intimidating. It needs to stop.

At this point, the town should not be deprived of the opportunity and the time needed to consider the new realities that confront us all: health concerns that we never had before, a climate emergency requiring measures to reduce our carbon emissions to net-zero as quickly as possible, and new environmental threats from changes in climate we are already too late to avoid. We need to take our heads out of the sand that surrounds us and start taking the actions required to survive.

The pandemic—which we can't assume will be our last—has put everything into a new perspective. We know now that infectious diseases like Covid-19 disproportionately infect people living in densely populated housing. Doesn't this fact make it incumbent on us to reconsider whether it is responsible from a public health standpoint to house close to 100 people including at-risk workers and elders in close proximity on 4 acres of land?

Several Departments and Town Committees charged with matters directly bearing on the Cloverleaf project should be made part of the planning process, if they haven't already participated. They surely will have beneficial suggestions. In addition to the Board of Health, the Housing Authority, the Conservation Commission, the Planning Board, our Police and Fire Departments, and the ZBA, the Energy Committee, the Climate Action Committee, the Water Resources Oversight Committee, and the Local Comprehensive Plan Committee should all be involved.

Most Committees have not been meeting recently. The Water Resources Oversight Committee, which should be playing a key role in evaluating and advising on the Cloverleaf waste water problem, has not met since January. The Climate Action Committee, the Energy Committee and the Local Comprehensive Plan Committee are only now beginning to schedule meetings. Each of them may have important insights to bring to the table.

Waste water remains a matter of major concern, but there are other things that warrant attention. As currently planned, the Cloverleaf development will use fossil fuel and not have solar panels, heat pumps, or mini-splits. This will increase Truro's carbon footprint at a time when we must be rapidly reducing it, to do our part in avoiding catastrophic global climate change. Massachusetts has committed to ambitious reductions in carbon emissions. This requires that new buildings be built to be carbon-neutral. The technologies needed pay for themselves in a few years, actually saving money in the long run, as the Energy Committee can explain. If Cloverleaf isn't built to be carbon-free, Truro taxpayers will soon have to pay for costly retrofitting.

The Climate Action Committee is studying septic system corrosion from salinization caused by sea level rise. This is not merely a future concern: The Cape Cod Commission predicts flooding to occur at least annually in

Truro, starting this very year, which may increase salinization of our water table. The Climate Action Committee, the Water Resources Oversight Committee, the Board of Health, and local builders may be able to suggest alternative technologies to septic systems, such as composting toilets and biogas digesters. We need to hear from them.

In addition to all the concerns raised above, there exist many financial questions--are there investors who may profit from the Cloverleaf development, what measures will protect the tenants from unaffordable rent increases, what cost projections are there for future operation, maintenance and management , and what financial burden might the taxpayers have to carry? The land was given to the town, yet the townspeople have been provided no answers to these questions.

Cloverleaf is by far the biggest construction project Truro has ever contemplated and there are issues that have not been adequately addressed. Too few town committees and members of the public have been in on the planning of Cloverleaf. it was not as open a process as it might have been, and many unforeseen consequences have not been aired or discussed by the community. Now that so many questions have been raised, we need to slow things down and broaden public involvement. We must do better with the land given us by the State for affordable housing. Including more perspectives will result in a better plan. As one signer said, "We can work together to have it all: affordable housing, clean water, and carbon reduction."

JUNE 29, 2020

From the following 75* year-round and part-time residents

Marian Averback
Joanne Barkan
Elaine Beillin
Cheryl Best
Debbie Best-Parker
Sam Boleyn
Stephen M. Briscoe
Bonnie Brown
Peter Burgess (In memory of
Maureen Burgess)
Deb Citrin
Ann Courtney
Richard Courtney
Lisa Di Stefano
Tom Engelhardt
Nathalie Ferrier
Pamela Fichtner
Ron Fichtner
Michael O. Finkelstein
Jon Friedman
Nancy Garrity
Bill Golden
Claudia Goldstein
Mark Hammer
Kenneth Hawkey
Peter Herridge
Robert Higgins-Steele
Joanne Hollander
Daniel Holt
Joan Holt

Michael Holt
Robert Holt
Susan Howe
Dan Katz
Judyth Katz
Hank Keenan
Frank Korahais
Paul Krueger
Mary Ann Larkin
Roberta Lema
Helga Lupien
Sarah Lutz
Tracy Maclin
Regan McCarthy
Joan Moriarty
Dennis O'Brien
Kate O'Brien
Katherine Oehling
Alice C. Parcell
Eric Parker
Jack Riemer
Lorraine Rosenbaum
Martin Rosenbaum
James Rudd
Jane Rudd
Barbara Sass
Denise Seager
Jon Seager
Jon Slater
Andrew Smith
Caroline Smith
Snow, John
Bonnie Sollog

Steve Sollog
Janet St. Onge
Peter Sullivan
Jim Summers
Marie Danielle Tanguay
Karen Tosh
John van Rens
Laurie Veninger
Peter Weiler
Gwendolyn Willard
Anthony Wolff
Pamela Wolff
Steve Wynne

*There were others who expressed agreement with the Statement but didn't want their names made public.

Jeffrey Ribeiro

From: c.e.steinman <c.e.steinman@comcast.net>
Sent: Wednesday, July 1, 2020 12:27 PM
To: Jeffrey Ribeiro
Cc: Art Hultin; Fred Todd; John Thornley; John R. Dundas
Subject: Additional Comments for the Public Record Regarding the Design of the Proposed Cloverleaf Proje

Please read into the Public Record the following comments that are intended to make clear the points of my previously submitted comments:

The COVID pandemic has significant new implications for the health and safety for all Truro's residents, and in particular for future Cloverleaf residents. Many institutions that are cautiously grappling with reopening are extremely concerned about their liability for the spread of Covid-19. Not only are they limiting occupancy levels and creating safer environments, they fear potential lawsuits for not meeting strict State regulations for providing protective measures, or new standards for cleaning and sanitation. The potential liability of the Town and the developer must be taken into account.

The pre-pandemic design of the Building #21 congregate apartment is no longer an appropriate solution for Truro or its seniors. Given the high percentage of deaths in congregate retirement communities and nursing facilities, redesign of the apartment building must be taken seriously. In my comments in the June 25 packet and as further supported by the following excerpts from New York Times (*Coronavirus Crisis Threatens Push for Denser Housing,* by Kevin Williams, May 6, 2020), it is recommended that Building #21 be replaced with a 9 or 12 townhouse units similar to those previously proposed for the site, of which 6 or 8 could be accessible ground-floor units entered from the front and back. Pertinent to the recommended Cloverleaf redesign are the following excerpts from the NYT article:

> "**The whole discussion about housing will change**. A lot of the bills and laws the Legislature have been discussing will be **looked at in a different lens**," [Isaiah Madison, a board member of Livable California, a nonprofit group] said.
>
> "I wouldn't make any big development decisions right now," said Dr. Jackson, a former officer in the Epidemic Intelligence Service at the Centers for Disease Control and Prevention. "The economic fallout is likely to last five years or more," he added, "and people may be wearing masks for several years. **Developers will have to factor the pandemic, and other crises, into their plans**."
>
> "The desire for denser developments might diminish," [Mr. Youngentob, a Maryland developer] said, and his company may switch its focus to townhomes. "**The forced interaction of sharing doors and elevators has caused some anxiety**," Mr. Youngentob said, "**Townhomes, where you come in and out of your door, and you know you are the only one touching your door handle, provide some comfort**." (Emphasis added.)

Responsibility for the apartment building's daily cleaning and maintenance of commons areas, shared laundry rooms, elevators, stairwells, etc. will be an unanticipated burden and expense. As noted above, people are now fearful of occupying such spaces and are likely to prefer the privacy associated with duplexes or townhouses. The Town's responsibility for protecting its residents' health and safety has taken on a whole new dimension. Now is the time for the ZBA to press "**PAUSE**" to allow enough time for these concerns to be properly evaluated.

Respectfully submitted,

Chuck Steinman

PO Box 781/Shore Road, North Truro

Zoning Board
Cloverleaf Complex
PO Box 2030
Truro, MA 02666

June 26, 2020

To: Zoning Board Members
Re: Cloverleaf Bus Stop

I am writing in reference to a suggestion made at the virtual meeting on 6/25/20 concerning the addition of a bus stop to be located in the area at the entrance to the Cloverleaf Complex at 22 Highland Rd. A bus stop already exists 150 yards from this site on the same side of the street, on the other side of the bridge. This stop contains a shelter and an off-street driveway where the bus comes in off of Highland and customers wait and enter the bus away from the main road. A sidewalk from the Cloverleaf Complex could be put in and attached to the sidewalk which is already in existence under the bridge and extended the remaining short distance to this area.

The area requested for the new bus stop is not part of the original land acquisition allocated to this development. As mentioned by Mr. Ribeiro, it belongs to the county and the town would have to acquire it for this purpose. The land in question already contains a large Verizon relay box. It doesn't have enough remaining area to provide safety issues such as protection from rear-end collisions, a shelter from the weather, or a safe area for the loading and unloading of passengers as exists at the current site.

I am also concerned about the proximity of the new bus stop to the on and off ramps for Route 6. A bus stop in this area would cause congestion and confusion with cars entering and exiting the ramps along with cars proceeding straight up and down Highland Road in addition to cars coming in and out of the complex. Having a bus stopped in this area would interrupt the flow of traffic presenting a safety hazard.

Further site review and a formal summer traffic study should be done before any further actions are taken.
Respectfully,

Lauren Anderson
30 Highland Rd
North Truro, MA 02652

The Cloverleaf Proposal

(It was my intention to read this statement into the record of the Truro Zoning Board of Appeals during their deliberations of Mr-12-20 on the "Cloverleaf Project". Given that COVID-19 requires us all to maintain a "social distance" for the duration, which has already postponed several ZBA hearings so far... I offer this here and now.)

It is my hope that each of you on *our* Truro Zoning Board of Appeals cares enough about *our* small rural town of Truro to examine this project carefully enough to realize how *really bad* this proposal is and why it just doesn't belong here.

My name is Stephen Williams. And 35 years ago I was employed (for 16 years) by the Town as Truro's Building Commissioner, Agent to the Board of Health, and Zoning Enforcement Officer. Each of those positions involved the enforcement of specific codes. First, there was the Massachusetts Building Code (which is ***not*** a textbook for quality construction so much as a table of *minimum standards).* Then there was 105 CMR: "The minimum standards for human habitation." And then there was 310 CMR: "Title V", or the minimum standards regarding the proper disposal of "sanitary waste". And finally, Truro's Zoning Bylaw with it's minimum standards for lot area, frontage and the maximum height of buildings etc. All of these codes have one thing in common: They are all agreed upon and accepted as ***minimum standards*** **in Truro.** In effect, if you cannot, *or in this case deliberately choose not to,* meet these **minimum standards**, you are failing our most basic societal norms!

In my opinion, this so-called "Cloverleaf Proposal" is a travesty! **It fails to meet so many minimum standards** that I hardly know where to begin. And, the fact that the proponents are asking that you give them **so many waivers** (see *their* "Exhibit T") from all the various **minimum standards** (as noted above) proves that better than anything I can say here tonight.

In the first place this project involves **an unnecessarily and *extremely* dense cluster of "apartments"**, under the Chapter 40-B allowance for creating "affordable housing." But if you look at it closely, what you'll really see is what amounts to an entire subdivision compressed and forced onto less than 4 acres of land. **Such density might be appropriate in more urban areas where three-story apartment buildings are the norm** but in

Truro it is a **transgressive imposition** which is wholly out of keeping with the rural character of this community. What the developers are hoping to do here, is to build **a cul-de-sac with 40 "units" (in 12 duplexes and one 17 room "dormitory")** ***with a total of no less than 70 bedrooms...*** **all on only 3.9 acres of land!** But if you stop to consider this more critically, what you'll see is a very aggressive effort to force a ***disproportionally large*** **'privately run'** municipal housing project (with no on-site manager?) into too small a space... thinly disguised as an "affordable housing" project.

Some of you may remember Harold Harris. Mr. Harris owned 65± acres of land just west of "Noon's Pit" which, if you took away all the 'unbuildable wetland area' around the pond, left about 40± acres that was subsequently developed by Geiger-Phillips into a sub-division called Shearwater... perhaps the best laid out subdivision in Truro. To show you how dense the "Cloverleaf Project" actually is imagine taking that entire subdivision of 40± homes and compressing it down to make it fit onto just 3.9 acres of land. Well, that's what the proponents of Cloverleaf are trying to do. What they are seeking to do here, ***although they will vehemently deny it,*** is to create a small urban enclave, **a very dense** ***mini-ghetto*** **of public housing** (their recipe for an instantaneous slum? "Just add way too many people, boil, and stir briskly...") while they try to distract this community by waving the holy flag of "affordable housing" at us as though that alone would justify this kind of urban density. ***There is no need or reason that "public housing" or "affordable housing" should be this concentrated... especially when there are better and larger alternative sites immediately available... such as the Walsh Property south of the school.***

Their problem is that to do this, the developers need the ZBA approval for a slew of **waivers to the various minimum standards that still apply**... even after invoking Chapter 40-B. And no matter how much hype & bombast they assail you with, ***it is my considered opinion that they do not qualify for any of the waivers they're seeking.*** Not one!

I think it is important to note here and now that ***the proponents of this project are not philanthropists!*** Their only interest in Truro, and why they're promoting this project so aggressively, is for the profits they can make off this development... which are sure to be considerable! So, when

they come before you, holding up an empty bowl like Oliver Twist and begging, "Please Sir, can I have some more?" remember they're not asking you for an extra serving of porridge! What they ***ARE*** asking for, is that ***YOU*,** **on the ZBA,** give them another serving of more, and greater, windfall profits. **And,** ***without any demonstrable public benefit, each and every*** **minimum** ***design*** **standard** ***waiver you give them is just another cashier's check they can put in their pocket as they the drive out of town.***

Before I go any further with this, I think it is important to interject a bit of history here. When the Town and the State began discussions about a transfer of the Cloverleaf site to the Town it was initially premised on the need to relocate the Truro Highway Department. And, for that purpose, the Cloverleaf site was, **and still is**, perfect! It's certainly big enough and it doesn't require **ANY** waivers of minimum standards... and, easy access to all of the roadways around it is demonstrable. The Highway Department **WOULD NOT REQUIRE THE 1.2 MILION DOLLAR WATER SUPPLY** now envisioned for the "Cloverleaf project", nor would the septic outflow be near as dangerously toxic as **the proposed 7871 gallons per day from 70 bedrooms**. But somewhere along the way in those discussions, the zealots who champion "affordable housing" ('uber alles'), came in waving the flag of their holy crusade and seem to have successfully interposed their project on this site. **And what a dreadfully inappropriate fit it is!**

Given that the applicants have apparently chosen NOT to provide any full-size or scaled-drawing, and that their on-line plans are impossible to read, let alone scale, just one look at "Exhibit T" (*THEIR assessment* of what setback violations are necessary) should confirm that **at least seven of the proposed buildings appear to intrude into our *minimum standard* of 25' setbacks from all property lines.** If this is so they'll need "sideline variances". Variances of this nature, de novo or otherwise, require that they ***shall*** meet three (3) criteria. Lot shape; Soil conditions and Topography; and "Hardship"... financial or otherwise. There is no case to be made here by Lot Shape. There is no case to be made here for Soil Conditions & Topography. And, in my opinion, their only case for Financial Hardship is one they've deliberately created and brought upon themselves! Designing from scratch they've had more than ample

opportunity to meet our Town-wide *minimum setbacks requirements* ***and the fact that they've chosen not to do so only illustrates that*** **the high density level of their proposal is well beyond the capacity of this site and is driven more by a quest for private profit than any stated "public good".** A much more modest proposal in keeping with the rural nature of this Town is clearly preferable. And if more affordable housing units have to wait another year or two, well... remember that the Truro Highway Department has been waiting for more than 40 years!

Next and **again, because the applicants have chosen NOT to submit any scaled-drawings,** it is not possible to determine any actual elevations of the 13 buildings to be constructed at "Cloverleaf". But "Exhibit T" (see also page 12 of their proposal) confirms multiple variances will be needed for the "height of building" & "number of stories" regulations in our Zoning Bylaw. Not only are they *demanding* to be allowed to construct 3-story buildings, a violation of the Zoning Bylaws maximum 2-story rule, but **they also want to exceed the "maximum building height" allowance of 30' *by as much as 11½ feet!*.** Designing from scratch, they've had more than ample opportunity to make these 13 buildings meet all of the *minimum standards* of our zoning bylaw and not stand out as being so blatantly special or so *"different"* from the rest of this Town. **The fact that they have chosen not to do so once again demonstrates a presumptuous sense of entitlement on their part... rather than making any effort to design their project so that it fits into the minimum standards of this still rural Town. Who are these people anyway? And what makes them think they are so special that they deserve ANY variances from the *minimum standards* of this town which has gotten along quite well without them? *More people? Really?* If they really knew *anything* about Truro they'd know that what we *really* need here, even more than 'affordable housing', is: *MORE "AFFORDABLE" JOBS!***

And while renting out the "Cloverleaf" will likely be "sold out" in only a couple of minutes, it will most likely ***be occupied predominantly with people from other nearby towns*** because I doubt that we, as a community, actually need **as many as 70 rooms *ALL AT ONCE*!** And what's the point of bringing many more people into Truro if there's not enough work here

now? The **'inventory percentage'** of "affordable housing units" needed in Truro is actually meaningless if ***the people who already live here, and who need it most,*** are not the first and primary beneficiaries of the concept! A better approach for Truro would be incremental... say 5 or 6 units a year.

Their "parking plan" is also *deeply flawed*: I count only 69 spaces but with 70 bedrooms, **each capable of a 2-person occupancy**, even 140 spaces may prove to be inadequate. Double-depth spaces, as shown for several of the duplex units, isn't just bad planning... it's a prescription for further chaos and discord among renters! What if a resident wanted to invite friends over for a visit. Where can *they* park? What if someone gave a party? With a road width of 16'±, will fuel trucks be able to pass cars parked in the road? How about Fire and Rescue vehicles? Snowplows?

I should also note that I've seen no mention of any on-site supervisor, like the 'building superintendent' in most urban apartment buildings! **Our motels, cottage colonies and Condo Units *are all required* to have on-site managers.** And, given the obvious potential for conflict between various renters jammed so close together, not to mention the 17 room ***"dormitory"*** which, without some manner of on-site adult supervision, could easily become as notoriously unmanageable as a college dorm on 'spring-break', EXACTLY WHO IS GOING TO BE RESPONSIBLE for keeping order in "The Cloverleaf"? The Truro Police? Surveillance cameras like the six we have at the dump? Should we anticipate "facial recognition" technology?

The "Cloverleaf" at 3.9 acres is really not an appropriate location for "affordable housing" ***under any conditions,*** but applying the minimum standards of Title V, (yet another table of ***minimum standards*** to wit: 10,000 sf. of lot area per bedroom) the site can only accommodate **a maximum of 17 bedrooms!** At that level the project would actually meet all the other minimum standards noted above, ***it would also NOT require the 1.2 million dollar expense of public water or any septic variances*** and it *would* fit in with the rural character of this Town. **The proposed 70 bedrooms properly requires 700,000 sf. of lot area... that's 16 acres of land!** So obviously, this project will require a slew of waivers from the **minimum standards** of Title V as well. And when any project needs so many waivers from so many different **minimum standards** we should

conclude that this project does not, cannot, and never will meet the **minimum standards**, or the rural character, of this community we love and share called Truro.

There is also a jurisdictional aspect here that is most troubling: The Zoning Board of Appeals is set up to review zoning issues! Chapter 40-B is a zoning matter but there are no zoning issues involved when it comes to Title V. Given the legal precept of "Federal Sovereignty" wherein towns must defer to states which must defer to the federal government (or, "the sovereign"), **it seems entirely unlikely that our ZBA can legally waive ANY State minimum standards for Title V. (See page 3 of the Chapter 40-B handbook). But once again, one must ask, on what basis do these applicants feel that their cause is SO deserving that they should merit any such consideration? I would further posit that the Truro Board of Health may waive 'local' septic regulations *IN EXCESS* of State law (If *they* could find any *'public health'* justification!) but they must also defer to the State DEP if any waivers to State Law are being requested!**

I have read that the proponents of this project seem to think they should be given GPD credit for much of the 15.6 acres of State and Park **vacant adjacent land** so they can meet their Title V (GPD) requirements of lot-area per-bedroom per-day. This is so pathetically self-serving I don't know whether to laugh or cry. In 1963 my father bought some land from Donald Schlesinger on North Pamet Road. His house lot sat atop an esker *abutting the National Seashore.* Should he be eligible to claim **"that vacant NPS adjacent land"** for septic purposes should his kids want to build a 20 room "affordable apartment" complex there? How about 10 rooms? **310 CMR 15.00: (Title V) is NOT AMBIGUOUS when it refers to site-specific area requirements for the ON-SITE disposal of "sanitary waste".**

And in none of the reports available on-line have I seen any mention of the other lots adjacent to the Cloverleaf site whose health and safety will likely be threatened by **the dangerous and disproportionately high volume of 7871 gallons of septic waste being created there *EVERYDAY!* On only 3.9 acres of land?** Olin Sparks owned one of those lots and the protective "zone of contribution" of his well includes an arc which falls within the perimeter of the empty Cloverleaf site. ***The discharge of 2,872,915 gallons***

per year of septic waste JUST outside the drawdown cone of a SFR well on an adjacent lot? Come on now. **That's just criminal malfeasance!**

No one disputes that we need affordable housing in Truro. But that need (*devoid of its hysterical hype*) should not blind us to the fact that this particular project is needlessly and unjustifiably too dense, and too much at odds with the rural character of Truro to fit into this community. **With only 17 bedroom units there's no problem**... although the location is still bad and inappropriate. **The problem is that these proponents are trying to jam 20 pounds of sugar into a 5 pound bag AND TO CONSTRUCT AN ADDITONAL 53 MORE BEDROOMS THAT JUST DON'T FIT ON ANY 3.9 acre lot!** What should happen, *in my opinion,* is that this site should to be given back to the Town for the purpose of re-locating the Highway Department soon or next year and that some section of land in the Walsh Property should be designated for **lower-density** "affordable" housing. **It's late into this project now... people have invested time and money and *THEY'RE NOT GOING TO BE HAPPY* if the ZBA stands it's ground to defend Truro from the depredation of *these* so-called "developers".** But just because it's a Chapter 40-B development doesn't mean it has to be *so dense* and such an obviously unappealing place to live, with as many as 140 people having to live on top of each other on less that 4 acres of land! **We're not a city!** At the Walsh Property the density of inhabitants to lot area can be spread out more to reflect the rural character of the Town and create affordable housing where people can actually live *(with their children!)* without falling over their neighbors, or being assailed by the constant air-pollution and 24/7 din of traffic noise from the adjacent highway, or being constantly irritated by their next door neighbor's choice of music, or high volume. And tell me, how do you comfortably share or enjoy the leftover outside space of a 3.9 acre lot with 139 other people on a hot summer afternoon? In short, you can't! In short:

~~ This proposal is esthetically offensive and morally indefensible ~~

And just because low-income working people aren't rich enough to buy a house in Shearwater is no reason for them to have to suffer the added indignity of being herded together like animals so tightly into the *"concentration-camp"* like densities of THIS 'duplex disaster' aka the

"Cloverleaf" proposal. All that's missing are a few guard towers, a barb-wire perimeter fence and a road sign that says "Arbeit Mach Frei". And with as many as 140± people on 3.9 acres? That's 35 people per acre! Really? OR PERHAPS THAT'S THE POINT! *YES*, you can live in our Town and *YES* you can work for us. But just because we let you out on a daily 'work-release' don't ever assume more than that. Because if this were really the "workers paradise" its proponents purport it to be, then maybe they should demonstrate the truth of their claim by volunteering to actually live there *for just one year!* Do you believe that *any of them* would find it so desirable living year-round packed into such a suffocating "sardine-can subdivision" with as many as 139 other people, assailed by the incessant traffic noise (24-7) of Route 6 not to mention *the constant monoxide exhaust stench of traffic wafting in from a major highway only a few yards away?* What a slum! What a sad and dreary and unhealthy setting for raising children in the country!

If this is really the best we can do for minimum-wage workers, we should be ashamed! I mean, would any of you, the members of our ZBA, actually WANT to live in "Cloverleaf? Would you want to raise *your* family there?

In closing I'd like to note that **if** the proponents of this proposal claim that such high density (while perhaps not essential to this project) is still essentially necessary if they're going to be able to finance it and make a profit.... (**and if I were sitting as a member on this ZBA hearing, I would suggest:** *"That maybe they're just not the right developers for this project! That maybe they should just take their drawings and go home."*) I'm sure there are other developers who would gladly compete to build 'affordable housing' in Truro *in a more humane way*, with a code-conforming, no waivers needed, **17 bedrooms** ± ... and could make a decent living do so!

~~ As presented, this proposal is NOT in the public interest! ~~

~~~~~ ~~~~~ ~~~~~ ~~~~~ ~~~~~ ~~~~~ ~~~

Stephen Williams – P.O. Box 1111 – Truro – Mass – 02666 - #508-349-3358
~~~~~

July 5, 1995

CONCENTRATION VERSES DISPERSAL

a theoretical concept regarding the historic and future siting of municipal buildings & functions in Truro

We all know that Truro is still a rural community.

It is that quality which makes this Town so special to its inhabitants and to its visitors. Although the preservation of that rural quality should be a priority for all of us it is not a foregone conclusion.

In the first place Truro has only recently surpassed the mid-point of its projected potential for growth. With 2100 homes here now there remains enough undeveloped land which, if its use were to be maximized, could be divided so as to allow for another 1900 home-sites. Given a 36 year average of 35 new homes a year we can anticipate a total "build out" of 4000 homes around the year 2050. Increasing the minimum lot size to 1 acre and, through the Planning Board, urging developers to minimize subdivisions in exchange for lesser developmental standards (read: "fewer and larger lots but with lower developmental costs with roughly equal profit") would help a lot.

In the second place over the last 25 years, Truro has been "urged" to adopt an artifically lowered tax-rate... largely at the expense of its own infrastructure. The consequence of this "do nothing" policy preference means that in the decade of the 1990's Truro has had to: 1. rebuild its school (4 million plus _X_ 20 year debt service); 2. construct a new Police/Fire building (2 million plus _X_ 20 year debt service). And, out of necessity, Truro still faces more public works projects: 3. a new and enlarged central library (? million plus debt service); 3. a community center/Council On Aging (? million plus debt service); 4. a new or equally expensive rehab of Town Hall (? million plus debt service); 5. "capping" the landfill and moving the Highway Department down to the transfer station (? million plus debt service) ; not to mention the repaving of most of its roads (80,000 per mile).

When the "Lema Motel site" was bought through a friendly purchase for a new Police/Fire/Rescue building, Truro was "given" (remember that it was the developer who approached the Town first) a once-in-a-lifetime opportunity. This location is geographically central in Truro and provides a fast acceleration down-hill run for heavy fire trucks headed in either direction. For the Police & Fire Departments this location is perfect.

But given the size of this central location (6.7 acres) many other factions and/or proponents of particular projects (library, Town Hall, Community Center/Council on Aging) have begun to

envision this site as appropriate for their uses as well.

So, it is the purpose of this exposition to request that everyone in Truro consider the longterm effects of such "concentration" (most especially at this location on a two-lane major thoroughfare) and to offer, as an alternative, a valid argument for maintaining our historical "dispersal" of Town buildings and functions.

Truro, once again, is still a rural community. Inherent to that quality is the dispersal of private homes and public functions: the Town Hall is separate from the Police/Fire which is separate from the libraries which are separate from the dump which is separate from the school etc. This dispersal of functions means that all of these separate entities can operate simoultaneously without people tripping over one another regardless of which function they are engaging. Because of this dispersal, traffic jams are avoided because the congestion of people-plus-vehicles is broken up and not concentrated in any one place at one time.

There are some people in Truro who feel that all, or at least most, town functions should now be concentrated exclusively at the Lema Motel site. Their argument, as I understand it, is that such centralization would be "convenient" and that the geographic centrality of this location has merit or "fairness" because no one would have to travel too far from their particular end of Town.

Although this argument has an apprearance of logic it avoids one of the most important issues of proper siting which is: what will be the longterm effects of such centralization and are the side effects of such concentration sufficient to warrant another approach... such as deliberate dispersal?

Consider for a moment the number of cars which might be traveling in and out of the Lema site if all Town functions are concentrated there. Police/Fire/Rescue/library/community center/ Council on Aging/Town Hall (which includes: Selectmen meetings/ tax-treasurer-clerk/assessing department/ accounting department/ building department/licensing department, plus all committees: Finance Committee/Board of Health/Conservation Commmittee/Planning Board/Zoning Board of Appeals/Water Study Committee/Building Needs Committee/Beach Commission/Personnel Board/Harbor Commission/ Recycling Committee/Affordable Housing Committee/Recreation Committee/Library Trustees/Cemetary Commission/Golf Course Advisory Committee/Board of Fire Engineers/Historical Commission, etc.

This is an incomplete list but even so that's a lot of separate functions, a lot of people, a lot of cars, and a huge parking lot! Time-phasing public meetings is impractical if it's possible at all but whether cars arrive and depart in waves or in a random manner, one thing remains predictable: a heavy concentration of cars in ANY location in Truro is bound to cause traffic problems and to concentrate them all in only one location would promote more "urban" traffic conditions or gridlock. The

parallel concept of establishing "centralized parking" and running mini-bus shuttles to our beaches is similarly ill advised. "Why" one might ask, "do we live here?"

Getting into the Lema site from the north is a given... you just slow down (slowing traffic behind you in the process) and turn right. And getting out isn't bad either IF you're going south... just wait for a break in the traffic and you're back on the road. But getting in from the south across heavy oncoming traffic is likely to completely stop northbound traffic as you wait for an opening... and remember there are two lanes converging into one on-coming lane just 600 feet ahead of you. Getting out of the Lema site going north is going to be a major problem... especially if both north and southbound traffic is heavy since one needs a simoultaneous break from both directions to get across the highway. Imagine crossing the highway at night or worse, a rainy night. My instinct would be to go south to the school and turn around. And don't forget that in 55 years the number of Truro residents will have increased by another 1900 homes, nearly double what it is now.

Consider how an uninterrupted traffic flow may allow as many as 4400 cars to pass a given spot in an hour at 50 miles per hour. But when that flow is slowed by vehicles stopping to let other vehicles cross the road or, in the worst case scenario, by a traffic light, that number is dramatically reduced as traffic slows down to a bumper-to-bumper traffic jam. If a downhill run is good start for heavy fire apparatus it is also conversely a bad start for traffic trying to accelerate again on an uphill run from both directions. Envision the South Main Street entrance into Wellfleet (by the Mobil Station) where, on-the-flat, a single traffic light can and does back up traffic as far south as the Norseman in Eastham. And then consider whether centralization or dispersal is best suited to keeping Truro a "rural" community. A single "trip-light" at the Lema site would destroy the traffic flow through central Truro and could back up traffic to Pilgrim lake on the north end and past the Wellfleet border to the south. You might only want to get home, or to Perry's Market, or to the Post Office, or across the road, but the effects could be an unmitigated disaster that we, its engineers, would have to live with and endure.

An alternative to such concentration which would not impose this condition would be maintaining the historic dispersal of Town functions into different areas of Town, each of which this writer concedes may have its own particular logistical problems which over time are likely to increase... but which, if concentration is avoided, will at least not be cumulative. The school, recently rebuilt and expanded, is an established point. The Police/Fire building, recently constructed, is also an established point. So, what's left? The library, the community center, the Council on Aging, and the Town Hall with all its various boards and committees and departments.

Imagine how the dispersal of these functions might look. The

Library, for example, could be situated on the recently acquired SAADA LAND (it's west of Rt 6, east of 6A, north of Highland Road and south of Standish Way). This 13.67 acre location (5 acres of which was given to the "Affordable Housing Committee" at the last Town Meeting but which could still be recovered) provides the possibility for several access points onto Route 6 (4-lane) as well as a northern access from Standish Way. Additionally, the Town might negotiate with Ms. Bloespflug (the abutting owner of Babe's Bakery on Route 6A (aka: Shore Road) for a road width access (30') to 6A. This would mean that people who want to use the new consolidated library might arrive and depart by three independent multiple network road systems without major inconvenience or disruption of traffic flow.

There has been talk of moving the Highway Department to the landfill site after the landfill is "capped". This move makes practical sense. The Highway Department would no longer need to shuttle back and forth between Town Hall and the dump and the storage of their heavy equipment would be out of sight. As it is now, their heavy equipment rumbles through Truro Center daily and abutting neighbors have complained about the appearance of so much equipment being stored there.

The Town Hall, in my opinion, is part of this Towns' history as well as its architectural heritage. Restoration may not be the cheapest solution but it is the only solution which acknowledges that this building (for all its internal space limitation) is still a cultural landmark for this Town which must be preserved. Once upon a time there were three steeples in Truro... at least until the South Truro Church burned down. It is unthinkable that anyone should seriously consider tearing down this structurally sound (albeit cosmetically uncared for) building in favor of a "modern" building which looks like it was imported from New Jersey. **OUR** classic New England Town Hall can be rehabilitated and, if necessary, an annex of similar architectural style constructed nearby. After the Highway Department moves there will be no shortage of space on Town Hall Hill.

Left out of this scenario so far are the community center and the Council On Aging. These functions are equally important but properly siting them really should wait until we see what traffic conditions are actually like at the new Library. If the conditions there are inviting, both might go to that site. If the Library site is too congested they might go beside the Town Hall. If additional sites are needed or desireable we should move to obtain them.

Residents of Aldrich Road and South Highland Road should be especially sensitive to the fact that an accident in front of the Lema site would necessitate detouring traffic around it via those roads. Residents in the rest of Truro should try to imagine the resulting crawl of traffic which they may have to endure if accidents outside the Lema site result in detours or ultimately a traffic light. It usually takes a fatality or two before the State

is willing to discuss traffic lights on open highways.

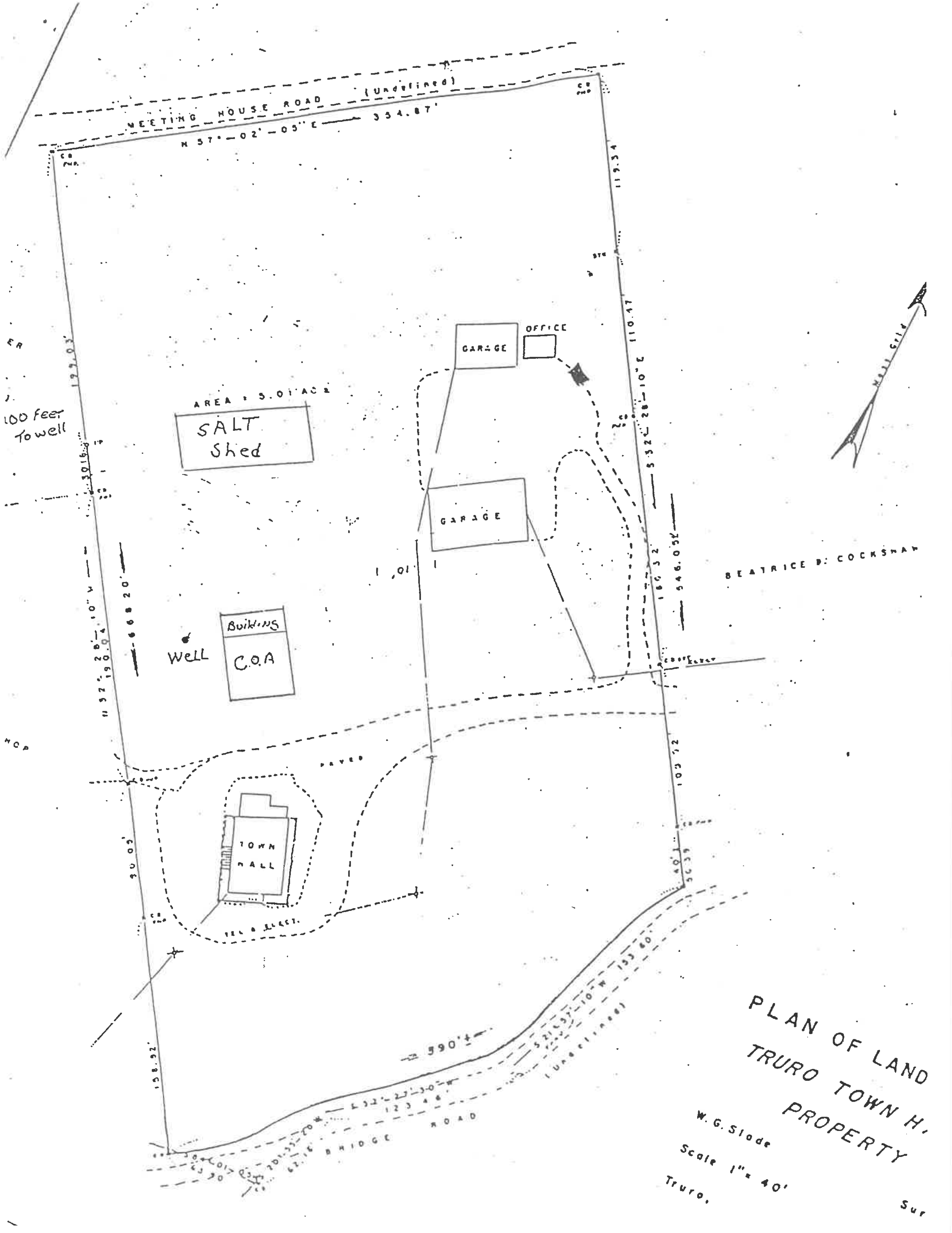
I offer these observations only to provoke the public thought and discussion that is necessary to resolve such issues which are community-shared. With all the best intentions, those who advocate "concentration of all Town Buildings at the Lema site" may unwittingly create a situation which is at once unpleasant, impractical, dangerous, and financially uncorrectable in the future. If we are offered the "carrot of convenience" of a central location which is also situated on a major thoroughfare where south bound traffic is converging and slowing into one lane from two and where northbound traffic has been already congested by a previous 15 miles of single lane travel, we should also think about whether this site is going to be accessible at all... especially, during the summer. Because, if traffic makes access and egress from this site awkward, or dangerous or simply so infuriating that people opt to avoid going there we are, in effect, stabbing ourselves in the proverbial foot.

The siting of Town buildings is very important if they are to serve this community and work as well as we all hope they will. Burdening new and improved facilities with predictable and inevitable access and egress traffic problems is a fast prescription for longterm failure which will injure this community more than not having the buildings at all. Why, for instance, build a new library at all if for half the year getting to it is so difficult or frightening that the people who love Cobb or Pilgrim libraries simply stop going to the library.

If one accepts that maintaining the historic and practical "dispersal" of Town buildings and functions is more harmonious with the rural character of Truro than the urban practice of "concentrating" everything in one place then the availability of space would seem to suggest the following sequence: FIRST; building a new and consolidated library, SECOND: capping the landfill and then moving the Town Highway Department, THIRD: rebuilding Town Hall with an annex or addition if necessary; FOURTH: determining where the community center and Council On Aging should best be situated. As to this community's obligation to provide "Affordable Housing", the least disruptive location, ironically enough, would be at the Lema Site where the minimal traffic effect of a few more residences would not be so disruptive.

In my opinion, the citizens of Truro should resist the tempting urban theories about growth and concentration. If we let ourselves get swept away by such imagry all of us may end up by collectively destroying the spacious beauty of this Town which keeps us here or has brought us here. It is an old economic axiom that "maintenance is not growth". But if we work together we can both grow and maintain the beauty of Truro's historical rural character. The two are not mutually exclusive unless we choose to maintain nothing of our past and to grow only in urban ways.

Stephen Williams



MEETING HOUSE ROAD (Undefined)
N 57°-02'-05" E 354.87'
GARAGE
OFFICE
AREA = 5.01 AC±
SALT Shed
100 Feet To well
GARAGE
BEATRICE D. COCKSHAW
S 32°-28'-10" E
546.05'
N 32°-28'-10" W
668.20'
Buildings
C.O.A
Well
PAVED
TOWN HALL
TEL & ELECT.
590'±
BRIDGE ROAD
PLAN OF LAND
TRURO TOWN H.
PROPERTY
W. G. Slade
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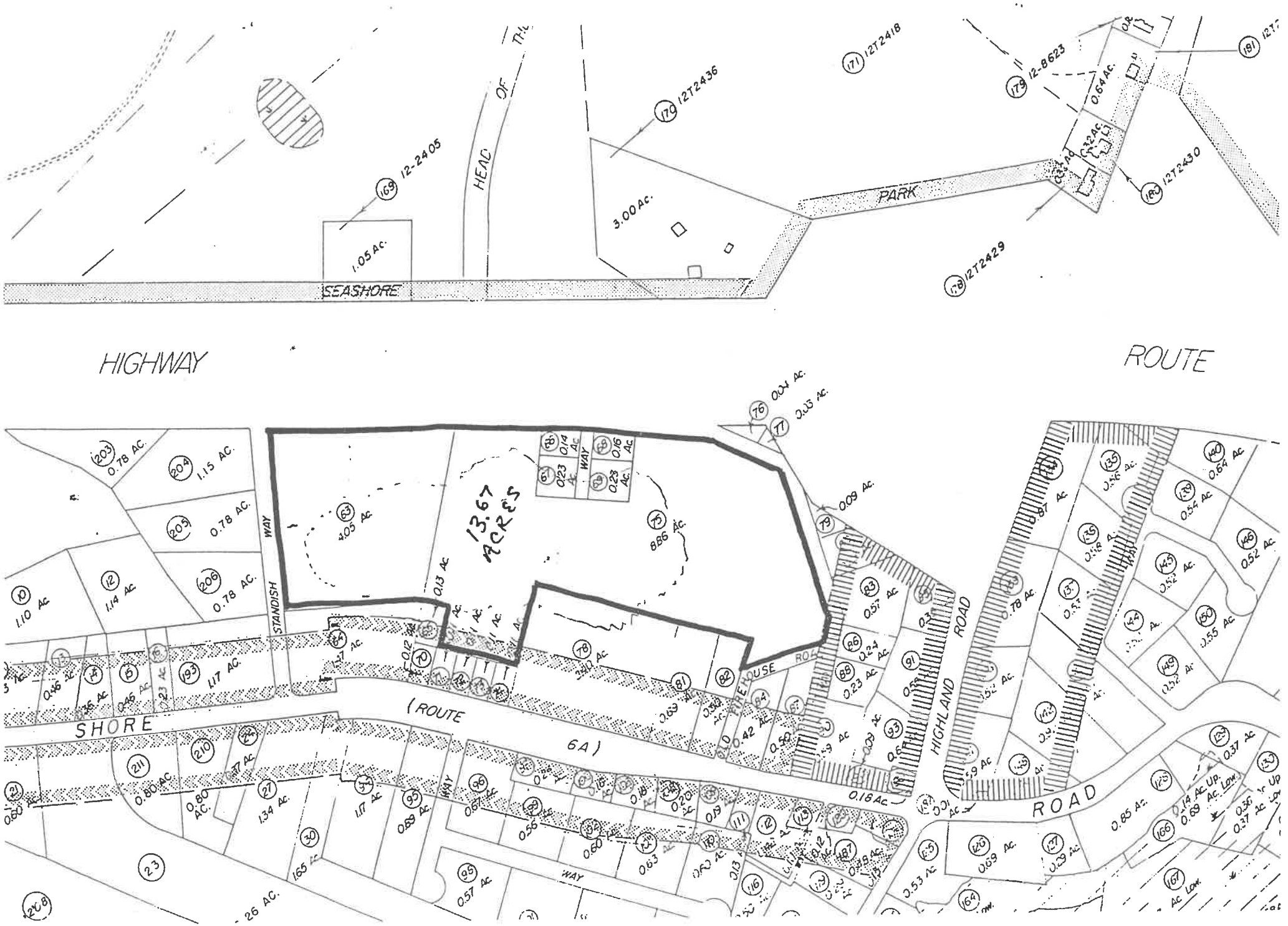
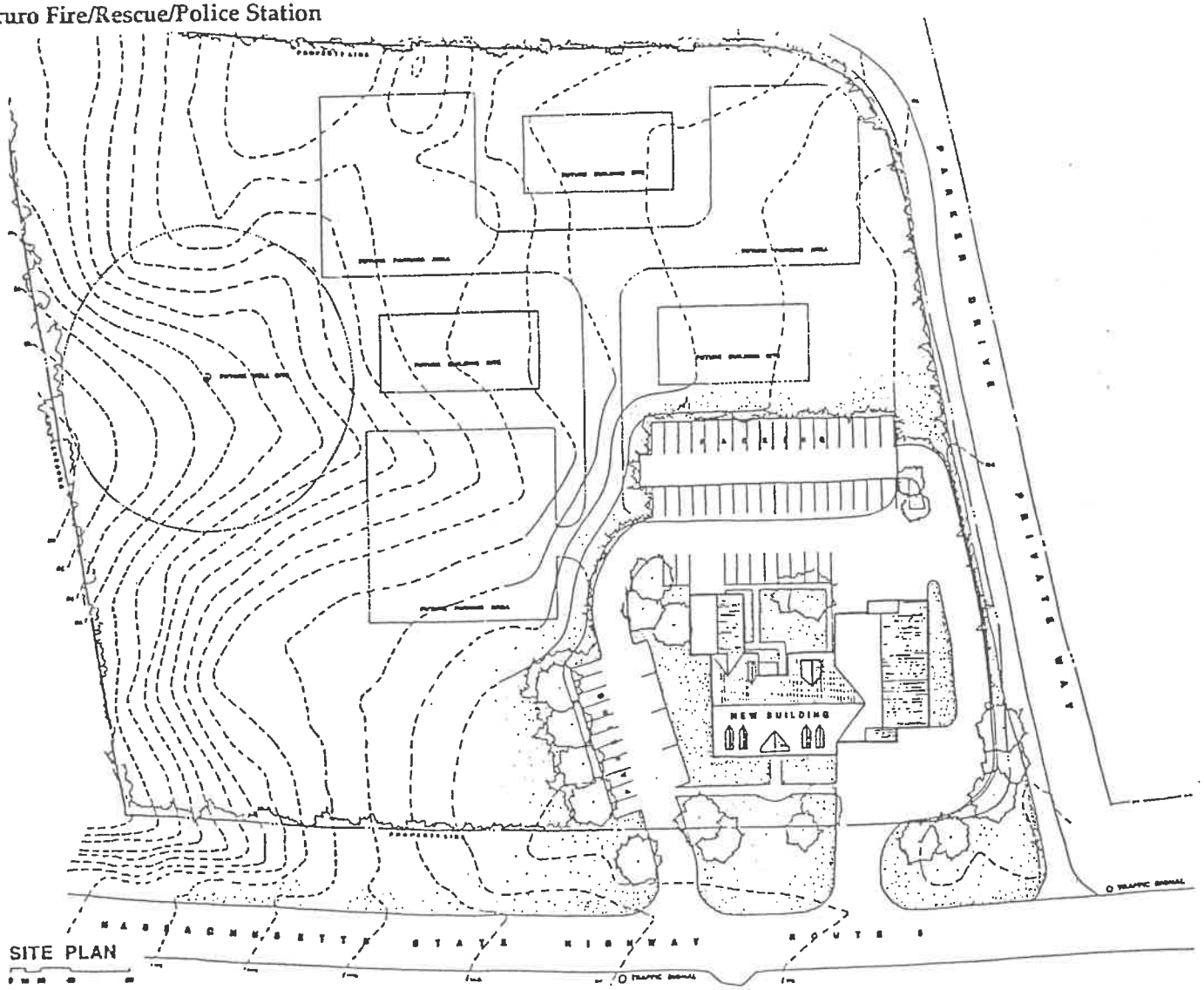

HIGHWAY
ROUTE
SEASHORE
HEAD OF THE
PARK
1.05 AC.
3.00 AC.
12T2436
12T2418
12-8623
12-2405
12T2430
12T2429
0.64 AC.
13.67 ACRES
STANDISH WAY
SHORE
(ROUTE 6A)
FIREHOUSE ROAD
HIGHLAND ROAD
ROAD
WAY
4.05 AC.
8.86 AC.
0.78 AC.
1.15 AC.
1.10 AC.
1.14 AC.
1.17 AC.
0.80 AC.
1.34 AC.
0.69 AC.
0.57 AC.
0.59 AC.
0.85 AC.
26 AC.

Truro Fire/Rescue/Police Station



SITE PLAN

July 8, 2020

To Truro Town Planner and Truro ZBA,

We are staunch supporters of affordable housing and welcome our new neighbors to be, but as abutters to the proposed clover leaf project we also have a range of concerns we would like addressed.

It is a sea change to go from the privacy and open space that we currently enjoy to neighboring a 3.9 acre development that, once occupied, would house 2-7% of the town's entire population.

Our concerns range from water quality to privacy to safety and property value.

Our water is pristine, and we're happy to supply current water tests. In the July 8th meeting of the Truro Board of Health, Mark Nelson of Horsley Whitten, spoke candidly of nitrogen contamination to wells usually being caused by a neighboring septic. Unless we receive a guarantee that this project will not affect our water quality, we believe that the lease holder should be required to pay for annual water tests to monitor our water quality. If our water quality diminishes, and we need a new well, we believe the lease holder should pay for whatever costs we would incur, including costs for permitting and surveying.

Our parcel, unique among the residential abutters, falls in the seashore district, which permits our pursuit of agriculture. Our property is home to many agricultural realities that could be hazardous and/or could fall under the heading of "attractive nuisance". We have bee hives and chickens and roosters to name a few, which would be very tempting to children and dogs and others who don't read signs.

We believe the developer should be required to install and maintain what we agree to as adequate fencing and privacy screening along our 227 foot shared property line. Considering the upheaval that shifting 10,000 cubic yards of earth will cause, we feel this fencing and privacy screening should to be in place before construction and need assurance that it will be kept up once the project is complete and occupied.

Initially we were told that the area adjacent to our property would be left as a natural buffer, but have since heard it offered up for many uses, from playgrounds, to the site of a well for watering plantings, to additional parking. This area is also slated on the site

plan as "reserved for future" which means that in case of a failure of their septic system, a perforated pipe trenched into this area would handle 4,285 gallons of discharge per day. Consequently, we would like the buffer zone clearly defined, and want assurances that the project won't continue to creep towards our parcel through the defined reserve area and buffer.

There is a phrase we see on the “Drainage and Subcatchment areas” map, called “Limit of Work”. With the exception of the installation of the fencing and privacy screening, we would like this line be pushed back from our property line on their north west boundary marker- much as the “limit of work” is from the other residential abutter to the southeast of the clover leaf parcel. Essentially, we don't want any work done up against our property line, and we want the originally proposed buffer zone to remain in place.

Building 21 is enormous. A comparable residential building does not exist in our town. When justifying this building, it was compared with the Grace Gouveia building, which is a significantly smaller residential building in another town (Provincetown), and the main building at the Truro Vineyards, which shares the same height, but is otherwise smaller and not residential. To give you another view on this building please see our attachment. This is a crude rendering that we created after the balloon test of December 21. You can see how it towers over us as abutters.There are claims that building 21 will not rise above the tree line but once the 10,000 cubic yards of ground are moved to create the topography for this development, the existing tree line will be gone. With the slope of the site, there is no way to create a privacy screen between these apartments and our property. As you can see, this means the two upper floor of this building on the north side, 8 apartments, or 32 windows in all, directly overlook us, no matter where we go on our property, and they will be always in our line of sight, too. A residential building of this magnitude does not fit into our community's rural character, and we believe it should be revisited and modified.

We believe that creating a development that houses between 2-7% of the town's population on a single 3.9 acre parcel risks unnecessary crowding, overuse of the site, and should be carefully considered. As a town we recently purchased the Walsh property, which is nearly 70 acres. During deliberations about purchasing that large parcel, we agreed as a town to explore using a portion of that land for affordable housing. This means that the clover leaf parcel needn't shoulder the burden of our entire affordable housing deficit. Reining in the scope of building 21 would reduce the threat to our single source aquifer, create more unified support of the project, and still create a significant amount of housing.

Thank you for your consideration,

Naomi Czekaj-Robbins and Christopher Czekaj

423 Route 6

Truro, MA 02666

To: The Select Board and the Zoning Board of Appeals

We support moving forward with the Cloverleaf development. We reject the claims of the Cloverleaf opposition letter from Joan Holt and others.

1. **THEY SAY:** Density will create a Public Health – Covid Problem

FACT: Each apartment is a separate living unit. Guests and service people will be required to follow State mandated health regulations.

2. **THEY SAY:** Town Committees have yet to weigh in

FACT: The Housing Authority and Select Board have provided ongoing opportunity for town boards and the public to weigh in. The Select Board unanimously approved the project and selected a developer in a well-attended public meeting. The following boards received the Cloverleaf application on November 14th: Select Board, Planning Board, Conservation Commission, Board of Health, Housing Authority, Open Space Committee, Energy Committee, Water Resources Oversight Committee. The ZBA is conducting televised meetings and is responsive to public comment. The Commission on Disabilities provided input on accessibility. The Energy Committee provided input and an independent report on sustainable design was conducted. Installation of solar panels will depend on the availability of grants and rebates to offset the cost at the time of construction.

3. **THEY SAY:** Waste water remains a major concern

FACT: In response to public concerns, an independent peer review of the wastewater and septic handling has been conducted a de-nitrification system was recommended and the developer will install this. Nitrogen outflow will be less than 10 ppm in accordance with State public health regulations.

4. **THEY SAY:** There may be septic system corrosion from sea level rise

FACT: The septic system will be installed 47 feet above sea level.

5. **THEY SAY:** The Cloverleaf is a private for-profit development...designed to provide maximum profit not maximum affordable housing:

FACT: Rents for the 31 affordable units are set based on area median income. They are regulated, and the only way they go up is if the area median income rises. There are 8 market rate rental units; these rents help subsidize the affordable units and help make the project financially feasible**.**

6. **THEY SAY:** If Cloverleaf isn't built to be carbon-free, Truro taxpayers will soon have to pay for costly retrofitting.

FACT: There are no 40b State statute requirements whatsoever on carbon free development. Furthermore, this would be an unfair hurdle to single out an affordable housing project to meet this aspiration of the Truro Climate Action Committee, would delay and likely make the project economically unfeasible.

7. **THEY SAY:** We need to slow things down

WE SAY: We need to keep moving forward to address our housing crisis.

Respectfully yours,

Sally Brotman
Carl Brotman
Susan Todd
Kathleen Henry
Kim Marrkand
Adrian Cyr
Annette Cyr
Jane Lea
Jen Shannon
Linnet Hultin
Betty Gallo
Mara Glatzel
Tim Dickey
Lisa Wisotzky
Mark Wisotzky
Will Hilldreth
Linda Hassett
Bob Hassett
Anne Wyckoff
Gregory Campora
Christopher Bellonci
Edouard Fontenot
Laura Hebert
Nola Glatzel
Jules Glatzel
Wendelin Glatzel
Kait Blem
Kolby Blem
Ellery Althaus
Julian Cyr
Kevin Grunwald
Shawn Grunwald
Richard Fishman
Jay Vivian
Barbara Wood
Paul Wisotzky
Jay Coburn
Raphael Richter
Maureen Cronin
Peter Graham
Claire Adams
Dante Rosselli
Richard Wood
Marne Hodgin
Richard Hersh
Judith Myers
Mary Abt
Kathleen Morris
Nicholas Brown
Lucy Brown
Garry Wyckoff
Ryan Cooke
Curtis Hartman
Nick Norman
Mark Kundeman
Keith Althus
Susan Baker
Robert M Paanessiti
Cindy Kleine
Andre Gregory
Rita Burke
Glenn Pasanen
Emily Bingham
Stephen Reily
John F Guerra
Ave Gaffney
Shar Prilo
Mark Adams
Mary Reinhardt
Roy Barnhart
Rita Burke
Any Kandall
Gullermo Chang
Annmarie Barrett Chang
Christen Roberts
Eric Jansen
Dilys Staaterman
Peter Staaterman
Kathleen Collins
Jeffrey Love
Deborah Landa
Sheldon Schwartz
Susan Kurtzman
Henry Janowsky
Andrew Rahorcsak

Submitted by Peter Herridge Dec 5, 2019

Submittal by Peter Herridge at 12/5/2019 Hearing

International Journal of *Environmental Research and Public Health*

MDPI

Review

Drinking Water Nitrate and Human Health: An Updated Review

Mary H. Ward [1,*], Rena R. Jones [1], Jean D. Brender [2], Theo M. de Kok [3], Peter J. Weyer [4], Bernard T. Nolan [5], Cristina M. Villanueva [6,7,8,9] and Simone G. van Breda [3]

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Abstract: Nitrate levels in our water resources have increased in many areas of the world largely due to applications of inorganic fertilizer and animal manure in agricultural areas. The regulatory limit for nitrate in public drinking water supplies was set to protect against infant methemoglobinemia, but other health effects were not considered. Risk of specific cancers and birth defects may be increased when nitrate is ingested under conditions that increase formation of *N*-nitroso compounds. We previously reviewed epidemiologic studies before 2005 of nitrate intake from drinking water and cancer, adverse reproductive outcomes and other health effects. Since that review, more than 30 epidemiologic studies have evaluated drinking water nitrate and these outcomes. The most common endpoints studied were colorectal cancer, bladder, and breast cancer (three studies each), and thyroid disease (four studies). Considering all studies, the strongest evidence for a relationship between drinking water nitrate ingestion and adverse health outcomes (besides methemoglobinemia) is for colorectal cancer, thyroid disease, and neural tube defects. Many studies observed increased risk with ingestion of water nitrate levels that were below regulatory limits. Future studies of these and other health outcomes should include improved exposure assessment and accurate characterization of individual factors that affect endogenous nitrosation.

Keywords: drinking water; nitrate; cancer; adverse reproductive outcomes; methemoglobinemia; thyroid disease; endogenous nitrosation; *N*-nitroso compounds

1. Introduction

Since the mid-1920s, humans have doubled the natural rate at which nitrogen is deposited onto land through the production and application of nitrogen fertilizers (inorganic and manure),

the combustion of fossil fuels, and replacement of natural vegetation with nitrogen-fixing crops such as soybeans [1,2]. The major anthropogenic source of nitrogen in the environment is nitrogen fertilizer, the application of which increased exponentially after the development of the Haber–Bosch process in the 1920s. Most synthetic fertilizer applications to agricultural land occurred after 1980 [3]. Since approximately half of all applied nitrogen drains from agricultural fields to contaminate surface and groundwater, nitrate concentrations in our water resources have also increased [1].

The maximum contaminant level (MCL) for nitrate in public drinking water supplies in the United States (U.S.) is 10 mg/L as nitrate-nitrogen (NO_3-N). This concentration is approximately equivalent to the World Health Organization (WHO) guideline of 50 mg/L as NO_3 or 11.3 mg/L NO_3-N (multiply NO_3 mg/L by 0.2258). The MCL was set to protect against infant methemoglobinemia; however other health effects including cancer and adverse reproductive outcomes were not considered [4]. Through endogenous nitrosation, nitrate is a precursor in the formation of *N*-nitroso compounds (NOC); most NOC are carcinogens and teratogens. Thus, exposure to NOC formed after ingestion of nitrate from drinking water and dietary sources may result in cancer, birth defects, or other adverse health effects. Nitrate is found in many foods, with the highest levels occurring in some green leafy and root vegetables [5,6]. Average daily intakes from food are in the range of 30–130 mg/day as NO_3 (7–29 mg/day NO_3-N) [5]. Because NOC formation is inhibited by ascorbic acid, polyphenols, and other compounds present at high levels in most vegetables, dietary nitrate intake may not result in substantial endogenous NOC formation [5,7].

Studies of health effects related to nitrate exposure from drinking water were previously reviewed through early 2004 [8]. Further, an International Agency for Research on Cancer (IARC) Working Group reviewed human, animal, and mechanistic studies of cancer through mid-2006 and concluded that ingested nitrate and nitrite, under conditions that result in endogenous nitrosation, are probably carcinogenic [5]. Here, our objective is to provide updated information on human exposure and to review mechanistic and health effects studies since 2004. We summarize how the additional studies contribute to the overall evidence for health effects and we discuss what future research may be most informative.

2. Drinking Water Nitrate Exposures in the United States and Europe

Approximately 45 million people in the U.S. (about 14% of the population) had self-supplied water at their residence in 2010 [9]. Almost all (98%) were private wells, which are not regulated by the U.S. Environmental Protection Agency (EPA). The rest of the population was served by public water supplies, which use groundwater, surface water, or both. The U.S. Geological Survey's National Water Quality Assessment (USGS-NAWQA) Project [10] sampled principal groundwater aquifers used as U.S. public and private drinking water supplies in 1988–2015. Nitrate levels in groundwater under agricultural land were about three times the national background level of 1 mg/L NO_3-N (Figure 1) [11]. The mixed land use category mostly had nitrate concentrations below background levels reflecting levels in deeper private and public water supply wells. Based on the NAWQA study, it was estimated that 2% of public-supply wells and 6% of private wells exceeded the MCL; whereas, in agricultural areas, 21% of private wells exceeded the MCL [10]. The USGS-NAWQA study also revealed significant decadal-scale changes in groundwater nitrate concentrations among wells sampled first in 1988–2000 and again in 2001–2010 for agricultural, urban, and mixed land uses [12]. More sampling networks had increases in median nitrate concentration than had decreases.

A study of U.S. public water supplies (PWS) using data from EPA's Safe Drinking Water Information System estimated that the percentage of PWS violating the MCL increased from 0.28 to 0.42% during 1994–2009; most increases were for small to medium PWS (<10,000 population served) using groundwater [13]. As a result of increasing nitrate levels, some PWS have incurred expensive upgrades to their treatment systems to comply with the regulatory level [14–16].

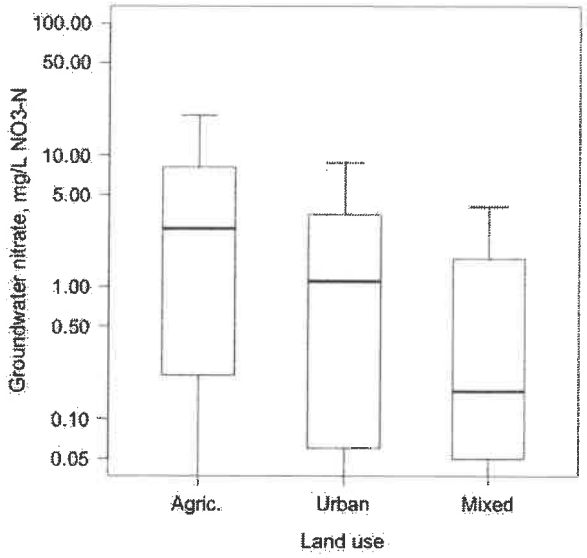

Figure 1. Boxplots of nitrate concentrations in shallow groundwater beneath agricultural and urban land uses, and at depths of private and public drinking water supplies beneath mixed land use. The number of sampled wells were 1573 (agricultural land), 1054 (urban), and 3417 (mixed). The agricultural and urban wells were sampled to assess land use effects, whereas the mixed category wells were sampled at depths of private and public supplies. Median depths of wells in the agricultural, urban, and mixed categories were 34, 32, and 200 feet, respectively. The height of the upper bar is 1.5 times the length of the box, and the lower bound was truncated at the nitrate detection limit of 0.05 mg/L NO_3-N.

In Europe, the Nitrates Directive was set in 1991 [17,18] to reduce or prevent nitrate pollution from agriculture. Areas most affected by nitrate pollution are designated as 'nitrate vulnerable zones' and are subject to mandatory Codes of Good Agricultural Practice [18]. The results of compliance with this directive have been reflected in the time trends of nitrate in some countries. For example, nitrate levels in groundwater in Denmark increased in 1950–1980 and decreased since the 1990s [19]. Average nitrate levels in groundwater in most other European countries have been stable at around 17.5 mg/L NO_3 (4 mg/L NO_3-N) across Europe over a 20-year period (1992–2012), with some differences between countries both in trends and concentrations. Average concentrations are lowest in Finland (around 1 mg/L NO_3 in 1992–2012) and highest in Malta (58.1 mg/L in 2000–2012) [20]. Average annual nitrate concentrations at river monitoring stations in Europe showed a steady decline from 2.7 NO_3-N in 1992 to 2.1 mg/L in 2012 [20], with the lowest average levels in Norway (0.2 mg/L NO_3-N in 2012) and highest in Greece (6.6 mg/L NO_3-N in 2012).

Levels in finished public drinking water have been published only for a few European countries. Trends of nitrate in drinking water supplies from 1976 to 2012 in Denmark showed a decline in public supplies but not in private wells [21]. In Spain, median concentrations were 3.5 mg/L NO_3 (range: 0.4–66.8) in 108 municipalities in 2012 [22], and 4.2 mg/L (range: <1–29) in 11 provinces in 2010 [23]. Levels in other countries included a median of 0.18 mg/L (range: <0.02–7.9) in Iceland in 2001–2012 [24], a mean of 16.1 mg/L (range: 0.05–296 mg/L) in Sicily, Italy in 2004–2005 [25] and a range from undetected to 63.3 mg/L in Deux-Sèvres, France in in 2005–2009 [26].

Nitrate levels in bottled water have been measured in a few areas of the EU and the U.S. and have been found to be below the MCL. In Sicily, the mean level was 15.2 mg/L NO_3(range: 1.2–31.8 mg/L) in 16 brands [25] and in Spain, the median level was 5.2 mg/L NO_3 (range: <1.0–29.0 mg/L) in 9 brands [23]. In the U.S., a survey of bottle water sold in 42 Iowa and 32 Texas communities found

varying but generally low nitrate levels. Nitrate concentrations ranged from below the limit of detection (0.1 mg/L NO_3-N) to 4.9 mg/L NO_3-N for U.S. domestic spring water purchased in Texas.

There are few published studies of nitrate concentrations in drinking water outside the U.S. and Europe. Nitrate concentrations in groundwater were reported for Morocco, Niger, Nigeria, Senegal, India-Pakistan, Japan, Lebanon, Philippines and Turkey with maximum levels in Senegal (median 42.9 mg/L NO_3-N) [5]. In India, nitrate in drinking water supplies is particularly high in rural areas, where average levels have been reported to be 45.7 mg/L NO_3 [27,28] and 66.6 mg/L NO_3 [28]; maximum levels in drinking water exceeded 100 mg/L NO_3 in several regions [27,29]. Extremely high levels of nitrate have been reported in The Gaza Strip, where nitrate reached concentrations of 500 mg/L NO_3 in some areas, and more than 50% of public-supply wells had nitrate concentrations above 45 mg/L NO_3 [30].

3. Exposure Assessment in Epidemiologic Studies

With the implementation of the Safe Drinking Water Act in 1974, more than 40 years of monitoring data for public water supplies in the U.S. provide a framework of measurements to support exposure assessments. Historical data for Europe are more limited, but a quadrennial nitrate reporting requirement was implemented as part of the EU Nitrates Directive [17,18]. In the U.S., the frequency of sampling for nitrate in community water systems is stipulated by their sources (ground versus surface waters) and whether concentrations are below the MCL, and historically, by the size of the population served and vulnerability to nitrate contamination. Therefore, the exposure assessment for study participants who report using a public drinking water source may be based on a variable number of measurements, raising concerns about exposure misclassification. In a study of bladder cancer risk in Iowa, associations were stronger in sensitivity analyses based on more comprehensive measurement data [31]. Other studies have restricted analyses to subgroups with more complete or recent measurements [32–35], with implications for study power and possible selection biases. Sampling frequency also limits the extent to which temporal variation in exposure can be represented within a study population, such as the monthly or trimester-based estimates of exposure most relevant for etiologic investigations of adverse reproductive outcomes. In Denmark, limited seasonal variation in nitrate monitoring data suggested these data would sufficiently capture temporal variation for long-term exposure estimates [36]. Studies have often combined regulatory measurements with questionnaire and ancillary data to better characterize individual variation in nitrate exposure, such as to capture changes in water supply characteristics over time or a participant's duration at a drinking water source [31,33,37,38]. Most case-control studies of drinking water nitrate and cancer obtained lifetime residence and drinking water source histories, whereas cohort studies typically have collected only the current water source. Many studies lacked information about study participants' water consumption, which may be an important determinant of exposure to drinking water contaminants [39].

Due to sparse measurement data, exposures for individuals served by private wells are more difficult to estimate than exposures for those on public water supplies. However, advances in geographic-based modeling efforts that incorporate available measurements, nitrogen inputs, aquifer characteristics, and other data hold promise for this purpose. These models include predictor variables describing land use, nitrogen inputs (fertilizer applications, animal feeding operations), soils, geology, climate, management practices, and other factors at the scale of interest. Nolan and Hitt [40] and Messier et al. [41] used nonlinear regression models with terms representing nitrogen inputs at the land surface, transport in soils and groundwater, and nitrate removal by processes such as denitrification, to predict groundwater nitrate concentration at the national scale and for North Carolina, respectively. Predictor variables in the models included N fertilizer and manure, agricultural or forested land use, soils, and, in Nolan and Hitt [40], water-use practices and major geology. Nolan and Hitt [40] reported a training R^2 values of 0.77 for a model of groundwater used mainly for private supplies and Messier, Kane, Bolich and Serre [41] reported a cross-validation testing R^2 value of 0.33 for a point-level

private well model. These and earlier regression approaches for groundwater nitrate [42–46] relied on predictor variables describing surficial soils and activities at the land surface, because conditions at depth in the aquifer typically are unknown. Redox conditions in the aquifer and the time since water entered the subsurface (i.e., groundwater age) are two of the most important factors affecting groundwater nitrate, but redox constituents typically are not analyzed, and age is difficult to measure. Even if a well has sufficient data to estimate these conditions, the data must be available for all wells in order to predict water quality in unsampled areas. In most of the above studies, well depth was used as a proxy for age and redox and set to average private or public-supply well depth for prediction.

Recent advances in groundwater nitrate exposure modeling have involved machine-learning methods such as random forest (RF) and boosted regression trees (BRT), along with improved characterization of aquifer conditions at the depth of the well screen (the perforated portion of the well where groundwater intake occurs). Tree-based models do not require data transformation, can fit nonlinear relations, and automatically incorporate interactions among predictors [47]. Wheeler et al. [48] used RF to estimate private well nitrate levels in Iowa. In addition to land use and soil variables, predictor variables included aquifer characteristics at the depth of the well screen, such as total thickness of fine-grained glacial deposits above the well screen, average and minimum thicknesses of glacial deposits near sampled wells, and horizontal and vertical hydraulic conductivities near the wells. Well depth, landscape features, nitrogen sources, and aquifer characteristics ranked highly in the final model, which explained 77% and 38% of the variation in training and hold-out nitrate data, respectively.

Ransom et al. [49] used BRT to predict nitrate concentration at the depths of private and public-supply wells for the Central Valley, California. The model used as input estimates of groundwater age at the depth of the well screen (from MODFLOW/MODPATH models) and depth-related reducing conditions in the groundwater. These estimates were generated by separate models and were available throughout the aquifer. Other MODFLOW-based predictor variables comprised depth to groundwater, and vertical water fluxes and the percent coarse material in the uppermost part of the aquifer where groundwater flow was simulated by MODFLOW. Redox variables were top-ranked in the final BRT model, which also included land use-based N leaching flux, precipitation, soil characteristics, and the MODFLOW-based variables described above. The final model retained 25 of an initial 145 predictor variables considered, had training and hold-out R^2 values of 0.83 and 0.44 respectively, and was used to produce a 3D visualization of nitrate in the aquifer. These studies show that modeling advances and improved characterization of aquifer conditions at depth are increasing our ability to predict nitrate exposure from drinking water supplied by private wells.

4. Nitrate Intake and Endogenous Formation of *N*-Nitroso Compounds

Drinking water nitrate is readily absorbed in the upper gastrointestinal tract and distributed in the human body. When it reaches the salivary glands, it is actively transported from blood into saliva and levels may be up to 20 times higher than in the plasma [50–53]. In the oral cavity 6–7% of the total nitrate can be reduced to nitrite, predominantly by nitrate-reducing bacteria [52,54,55]. The secreted nitrate as well as the nitrite generated in the oral cavity re-enter the gastrointestinal tract when swallowed.

Under acidic conditions in the stomach, nitrite can be protonated to nitrous acid (HNO_2), and subsequently yield dinitrogen trioxide (N_2O_3), nitric oxide (NO), and nitrogen dioxide (NO_2). Since the discovery of endogenous NO formation, it has become clear that NO is involved in a wide range of NO-mediated physiological effects. These comprise the regulation of blood pressure and blood flow by mediating vasodilation [56–58], the maintenance of blood vessel tonus [59], the inhibition of platelet adhesion and aggregation [60,61], modulation of mitochondrial function [62] and several other processes [63–66].

On the other hand, various nitrate and nitrite derived metabolites such as nitrous acid (HNO_2) are powerful nitrosating agents and known to drive the formation of NOC, which are

suggested to be the causal agents in many of the nitrate-associated adverse health outcomes. NOC comprise *N*-nitrosamines and *N*-nitrosamides, and may be formed when nitrosating agents encounter *N*-nitrosatable amino acids, which are also from dietary origin. The nitrosation process depends on the reaction mechanisms involved, on the concentration of the compounds involved, the pH of the reaction environment, and further modifying factors, including the presence of catalysts or inhibitors of *N*-nitrosation [66–69].

Endogenous nitrosation can also be inhibited, for instance by dietary compounds like vitamin C, which has the capacity to reduce HNO_2 to NO; and alpha-tocopherol or polyphenols, which can reduce nitrite to NO [54,70–72]. Inhibitory effects on nitrosation have also been described for dietary flavonoids such as quercetin, ferulic and caffeic acid, betel nut extracts, garlic, coffee, and green tea polyphenols [73,74]. Earlier studies showed that the intake of 250 mg or 1 g ascorbic acid per day substantially inhibited *N*-nitrosodimethylamine (NDMA) excretion in 25 women consuming a fish meal rich in amines (nitrosatable precursors) for seven days, in combination with drinking water containing nitrate at the acceptable daily intake (ADI) [75]. In addition, strawberries, garlic juice, and kale juice were shown to inhibit NDMA excretion in humans [76]. The effect of these fruits and vegetables is unlikely to be due solely to ascorbic acid. Using the *N*-nitrosoproline (NPRO) test, Helser et al. [77] found that ascorbic acid only inhibited nitrosamine formation by 24% compared with 41–63% following ingestion of juices (100 mL) made of green pepper, pineapple, strawberry or carrot containing an equal total amount of ascorbic acid.

The protective potential of such dietary inhibitors depends not only on the reaction rates of *N*-nitrosatable precursors and nitrosation inhibitors, but also on their biokinetics, since an effective inhibitor needs to follow gastrointestinal circulation kinetics similar to nitrate [78]. It has been argued that consumption of some vegetables with high nitrate content, can at least partially inhibit the formation of NOC [79–81]. This might apply for green leafy vegetables such as spinach and rocket salad, celery or kale [77] as well as other vegetables rich in both nitrate and natural nitrosation inhibitors. Preliminary data show that daily consumption of one bottle of beetroot juice containing 400 mg nitrate (the minimal amount advised for athletes to increase their sports performances) for one day and seven days by 29 young individuals results in an increased urinary excretion of apparent total nitroso compounds (ATNC), an effect that can only be partially inhibited by vitamin C supplements (1 g per day) [82].

Also, the amount of nitrosatable precursors is a key factor in the formation of NOC. Dietary intakes of red and processed meat are of particular importance [83–87] as increased consumption of red meat (600 vs. 60 g/day), but not white meat, was found to cause a three-fold increase in fecal NOC levels [85]. It was demonstrated that heme iron stimulated endogenous nitrosation [84], thereby providing a possible explanation for the differences in colon cancer risk between red and white meat consumption [88]. The link between meat consumption and colon cancer risk is even stronger for nitrite-preserved processed meat than for fresh meat leading an IARC review to conclude that processed meat is carcinogenic to humans [89].

In a human feeding study [90], the replacement of nitrite in processed meat products by natural antioxidants and the impact of drinking water nitrate ingestion is being evaluated in relation to fecal excretion of NOC, accounting for intakes of meat and dietary vitamin C. A pilot study demonstrated that fecal excretion of ATNC increased after participants switched from ingesting drinking water with low nitrate levels to drinking water with nitrate levels at the acceptable daily intake level of 3.7 mg/kg. The 20 volunteers were assigned to a group consuming either 3.75 g/kg body weight (maximum 300 g per day) red processed meat or fresh (unprocessed) white meat. Comparison of the two dietary groups showed that the most pronounced effect of drinking water nitrate was observed in the red processed meat group. No inhibitory effect of vitamin C intake on ATNC levels in feces was found (unpublished results).

5. Methemoglobinemia

The physiologic processes that can lead to methemoglobinemia in infants under six months of age have been described in detail previously [8,91]. Ingested nitrate is reduced to nitrite by bacteria in the mouth and in the infant stomach, which is less acidic than adults. Nitrite binds to hemoglobin to form methemoglobin, which interferes with the oxygen carrying capacity of the blood. Methemoglobinemia is a life-threatening condition that occurs when methemoglobin levels exceed about 10% [8,91]. Risk factors for infant methemoglobinemia include formula made with water containing high nitrate levels, foods and medications that have high nitrate levels [91,92], and enteric infections [93]. Methemoglobinemia related to high nitrate levels in drinking water used to make infant formula was first reported in 1945 [94]. The U.S. EPA limit of 10 mg/L NO_3-N was set as about one-half the level at which there were no observed cases [95]. The most recent U.S. cases related to nitrate in drinking water were reported by Knobeloch and colleagues in the late 1990s in Wisconsin [96] and were not described in our prior review. Nitrate concentrations in the private wells were about two-times the MCL and bacterial contamination was not a factor. They also summarize another U.S. case in 1999 related to nitrate contamination of a private well and six infant deaths attributed to methemoglobinemia in the U.S. between 1979–1999 only one of which was reported in the literature [96,97]. High incidence of infant methemoglobinemia in eastern Europe has also been described previously [98,99]. A 2002 WHO report on water and health [100] noted that there were 41 cases in Hungary annually, 2913 cases in Romania from 1985–1996 and 46 cases in Albania in 1996.

Results of several epidemiologic studies conducted before 2005 that examined the relationship between nitrate in drinking water and levels of methemoglobin or methemoglobinemia in infants have been described previously [8]. Briefly, nitrate levels >10 mg/L NO_3-N were usually associated with increased methemoglobin levels but clinical methemoglobinemia was not always present. Since our last review, a cross-sectional study conducted in Gaza found elevated methemoglobin levels in infants on supplemental feeding with formula made from well water in an area with the highest mean nitrate concentration of 195 mg/L NO_3 (range: 18–440) compared to an area with lower nitrate concentration (mean: 119 mg/L NO_3; range 18–244) [101]. A cross-sectional study in Morocco found a 22% increased risk of methemoglobinemia in infants in an area with drinking water nitrate >50 mg/L (>11 as NO_3-N) compared to infants in an area with nitrate levels <50 mg/L nitrate [102]. A retrospective cohort study in Iowa of persons (aged 1–60 years) consuming private well water with nitrate levels <10 mg/L NO_3-N found a positive relationship between methemoglobin levels in the blood and the amount of nitrate ingestion [103]. Among pregnant women in rural Minnesota with drinking water supplies that were mostly ≤3 mg/L NO_3-N, there was no relationship between water nitrate intake and women's methemoglobin levels around 36 weeks' gestation [104].

6. Adverse Pregnancy Outcomes

Maternal drinking water nitrate intake during pregnancy has been investigated as a risk factor for a range of pregnancy outcomes, including spontaneous abortion, fetal deaths, prematurity, intrauterine growth retardation, low birth weight, congenital malformations, and neonatal deaths. The relation between drinking water nitrate and congenital malformations in offspring has been the most extensively studied, most likely because of the availability of birth defect surveillance systems around the world.

Our earlier review focused on studies of drinking water nitrate and adverse pregnancy outcomes published before 2005 [8]. In that review, we cited several studies on the relation between maternal exposure to drinking water nitrate and spontaneous abortion including a cluster investigation that suggested a positive association [105] and a case-control study that found no association [106]. These studies were published over 20 years ago. In the present review, we were unable to identify any recently published studies on this outcome. In Table 1, we describe the findings of studies published since 2004 on the relation between drinking water nitrate and prematurity, low birthweight, and congenital malformations. We report results for nitrate in the units (mg/L NO_3 or NO_3-N) that

were reported in the publications. In a historic cohort study conducted in the Deux-Sèvres district (France), Migeot et al. [26] linked maternal addresses from birth records to community water system measurements of nitrate, atrazine, and other pesticides. Exposure to the second tertile of nitrate (14–27 mg/L NO_3) without detectable atrazine metabolites was associated with small-for-gestational age births (Odds Ratio (OR) 1.74, 95% CI 1.1, 2.8), but without a monotonic increase in risk with exposures. There was no association with nitrate among those with atrazine detected in their drinking water supplies. Within the same cohort, Albouy-Llaty and colleagues did not observe any association between higher water nitrate concentrations (with or without the presence of atrazine) and preterm birth [107].

Stayner and colleagues also investigated the relation between atrazine and nitrate in drinking water and rates of low birth weight and preterm birth in 46 counties in four Midwestern U.S. states that were required by EPA to measure nitrate and atrazine monthly due to prior atrazine MCL violations [108]. The investigators developed county-level population-weighted metrics of average monthly nitrate concentrations in public drinking water supplies. When analyses were restricted to counties with less than 20% private well usage (to reduce misclassification due to unknown nitrate levels), average nitrate concentrations during the pregnancy were associated with increased rates of very low birth weight (<1.5 kg Rate Ratio $(RR)_{per\ 1\ ppm}$ = 1.17, 95% CI 1.08, 1.25) and very preterm births (<32 weeks $RR_{per\ 1\ ppm}$ = 1.08, 95% CI 1.02, 1.15) but not with low birth weight or preterm birth overall.

In record-based prevalence study in Perth Australia, Joyce et al. mapped births to their water distribution zone and noted positive associations between increasing tertiles of nitrate levels and prevalence of term premature rupture of membranes (PROM) adjusted for smoking and socioeconomic status [109]. Nitrate concentrations were low; the upper tertile cut point was 0.350 mg/L and the maximum concentration was 1.80 mg/L NO_3-N. Preterm PROM was not associated with nitrate concentrations.

Among studies of drinking water nitrate and congenital malformations, few before 2005 included birth defects other than central nervous system defects [8]. More recently, Mattix et al. [110] noted higher rates of abdominal wall defects (AWD) in Indiana compared to U.S. rates for specific years during the period 1990–2002. They observed a positive correlation between monthly AWD rates and monthly atrazine concentrations in surface waters but no correlation with nitrate levels. Water quality data were obtained from the USGS-NAWQA project that monitors agricultural chemicals in streams and shallow groundwater that are mostly not used as drinking water sources. A case-control study of gastroschisis (one of the two major types of AWD), in Washington State [111] also used USGS-NAWQA measurements of nitrate and pesticides in surface water and determined the distance between maternal residences (zip code centroids) and the closest monitoring site with concentrations above the MCL for nitrate, nitrite, and atrazine. Gastrochisis was not associated with maternal proximity to surface water above the MCL for nitrate (>10 mg/L NO_3-N) or nitrite (>1 mg/L NO_2-N) but there was a positive relationship with proximity to sites with atrazine concentrations above the MCL. In a USA-wide study, Winchester et al. [112] linked the USGS-NAWQA monthly surface water nitrate and pesticide concentrations computed for the month of the last menstrual period with monthly rates of 22 types of birth defects in 1996–2002. Rates of birth defects among women who were estimated to have conceived during April through July were higher than rates among women conceiving in other months. In multivariable models that included nitrate, atrazine, and other pesticides, atrazine (but not nitrate or other pesticides) was associated with several types of anomalies. Nitrate was associated with birth defects in the category of "other congenital anomalies" (OR 1.18, 95% CI 1.14, 1.21); the authors did not specify what defects were included in this category. None of these three studies included local or regional data to support the assumption that surface water nitrate and pesticide concentrations correlated with drinking water exposures to these contaminants.

Using a more refined exposure assessment than the aforementioned studies, Holtby et al. [113] conducted a case-control study of congenital anomalies in an agricultural county in Nova Scotia,

Canada. They linked maternal addresses at delivery to municipal water supply median nitrate concentrations and used kriging of monthly measurements from a network of 140 private wells to estimate drinking water nitrate concentrations in private wells. They observed no associations between drinking water nitrate and all birth defects combined for conceptions during 1987–1997. However, the prevalence of all birth defects occurring during 1998–2006 was associated with drinking water nitrate concentrations of 1–5.56 mg/L NO_3-N (OR 2.44, 95% CI 1.05, 5.66) and ≥5.56 mg/L (OR 2.25, 95% CI 0.92, 5.52).

None of the studies of congenital anomalies accounted for maternal consumption of bottled water or the quantity of water consumed during the first trimester, the most critical period of organ/structural morphogenesis. Attempting to overcome some of these limitations, Brender, Weyer, and colleagues [38,114] conducted a population-based, case-control study in the states of Iowa and Texas where they: (1) linked maternal addresses during the first trimester to public water utilities and respective nitrate measurements; (2) estimated nitrate intake from bottled water based on a survey of products consumed and measurement of nitrate in the major products; (3) predicted drinking water nitrate from private wells through modeling (Texas only); and (4) estimated daily nitrate ingestion from women's drinking water sources and daily consumption of water. The study populations were participants of the U.S. National Birth Defects Prevention Study [115]. Compared to the lowest tertile of nitrate ingestion from drinking water (<0.91 mg/day NO_3), mothers of babies with spina bifida were twice as likely (95% CI 1.3, 3.2) to ingest ≥5 mg/day NO_3 from drinking water than control mothers. Mothers of babies with limb deficiencies, cleft palate, and cleft lip were, respectively, 1.8 (95% CI 1.1, 3.1), 1.9 (95% CI 1.2, 3.1), and 1.8 (95% CI 1.1, 3.1) times more likely to ingest ≥5.4 mg/day of water NO_3 than controls. Women were also classified by their nitrosatable drug exposure during the first trimester [116] and by their daily nitrate and nitrite intake based on a food frequency questionnaire [117]. Higher ingestion of drinking water nitrate did not strengthen associations between maternal nitrosatable drug exposure and birth defects in offspring [38]. However, a pattern was observed of stronger associations between nitrosatable drug exposure and selected birth defects for women in the upper two tertiles of total nitrite ingestion that included contributions from drinking water nitrate and dietary intakes of nitrate and nitrite compared to women in the lowest tertile. Higher intake of food nitrate/nitrite was found to also modify the associations of nitrosatable drug exposure and birth defects in this study [118,119] as well as in an earlier study of neural tube defects conducted in south Texas [120]. Multiplicative interactions were observed between higher food nitrate/nitrite and nitrosatable drug exposures for conotruncal heart, limb deficiency, and oral cleft defects [118].

In summary, five out of six studies, conducted since the 1980s of drinking water nitrate and central nervous system defects, found positive associations between higher drinking water nitrate exposure during pregnancy and neural tube defects or central nervous system defects combined [38,120–123]. The sixth study, which did not find a relationship, did not include measures of association, but compared average drinking water nitrate concentrations between mothers with and without neural tube defect-affected births, which were comparable [124].

Table 1. Studies of drinking water nitrate [a] and adverse pregnancy outcomes published January 2005–March 2018.

First Author, Year, Country	Study Design Regional Description	Years of Outcome Ascertainment	Exposure Description	Pregnancy Outcome	Summary of Findings
Albouy-Llaty, 2016 France [107]	Historic cohort study Deux-Sèvres	2005–2010	Measurements of atrazine metabolites and NO_3 in community water systems (263 municipalities) were linked to birth addresses	Preterm birth.	No association for >26.99 mg/L vs. <14.13 mg/L NO_3 in community water systems with or without atrazine detections, adjusted for neighborhood deprivation
Brender, 2013 Weyer, 2014 USA [38]	Population-based case-control study Iowa and Texas	1997–2005	Maternal addresses during the first trimester linked to public water utility nitrate measurements; nitrate intake from bottled water estimated with survey and laboratory testing; nitrate from private wells predicted through modeling; nitrate ingestion (NO_3) estimated from reported water consumption	Congenital heart defects Limb deficiencies Neural tube defects Oral cleft defects	≥5 vs. <0.91 mg/day NO_3 from drinking water spina bifida OR = 2.0 (95% CI: 1.3, 3.2) ≥5.42 vs. <1.0 mg/day NO_3 from water: limb deficiencies OR = 1.8 (CI: 1.1, 3.1); cleft palate OR = 1.9 (CI: 1.2, 3.1) cleft lip OR = 1.8 (CI: 1.1, 3.1)
Holtby, 2014 Canada [113]	Population-based case-control study Kings County, Nova Scotia	1988–2006	Maternal addresses at delivery linked to municipal water supply median nitrate (NO_3-N) concentrations; nitrate in rural private wells estimated from historic sampling and kriging	Congenital malformations combined into one group	Conceptions in 1987–1997: no association with nitrate concentrations Conceptions in 1998–2006: 1–5.56 mg/L NO_3-N (vs. <1 mg/L) OR = 2.44 (CI: 1.05, 5.66); ≥5.56 mg/L OR = 2.25 (CI: 0.92, 5.52)
Joyce, 2008 Australia [109]	Record-based prevalence study Perth	2002–2004	Linked birth residences to 24 water distribution zones; computed average NO_3-N mg/L from historical measurements; independent sampling conducted for 6 zones as part of exposure validation; also evaluated trihalomethanes (THM)	Premature rupture of membranes at term (PROM) (37 weeks' gestation or later)	ORs for tertiles (vs. <0.125 mg/L NO_3-N): 0.125–0.350 mg/L OR = 1.23 (CI: 1.03, 1.52); >0.350 mg/L OR = 1.47 (CI: 1.20, 1.79) No association with THM levels
Mattix, 2007 USA [110]	Ecologic study Indiana	1990–2002	Monthly abdominal wall defect rates linked to monthly surface water nitrate and atrazine concentrations (USGS-NAWQA monitoring data [b])	Abdominal wall birth defects	No correlation observed between nitrate levels in surface water and monthly abdominal wall defects Positive correlation with atrazine levels

Table 1. *Cont.*

First Author, Year, Country	Study Design Regional Description	Years of Outcome Ascertainment	Exposure Description	Pregnancy Outcome	Summary of Findings
Migeot, 2013 France [26]	Historic cohort study Deux-Sèvres	2005–2009	Measurements of atrazine metabolites and NO_3 in community water systems (263 municipalities) were linked to birth addresses	Small-for-gestational age (SGA) births	ORs for tertiles (vs. <14.13 mg/L NO_3) in community water systems with no atrazine detections: 14–27 mg/L OR = 1.74 (CI: 1.10, 2.75); >27 mg/L OR = OR 1.51 (CI: 0.96, 2.4); no association with nitrate when atrazine was detected
Stayner, 2017 USA [108]	Ecologic study 46 counties in Indiana, Iowa, Missouri, and Ohio	2004–2008	Counties had one or more water utility in EPA's atrazine monitoring program; excluded counties with >20% of population on private wells and >300,000 population. Computed county-specific monthly weighted averages of NO_3-N in finished drinking water; exposure metric was average 9 months prior to birth	Preterm birth Low birth weight	Average nitrate not associated with low birth weight and preterm birth Very low birth weight: RR for 1 ppm increase in NO_3-N = 1.17 (CI: 1.08, 1.25); Very preterm birth RR for 1 ppm increase = 1.08 (CI: 1.02, 1.15)
Waller, 2010 USA [111]	Population-based case-control study Washington State	1987–2006	Calculated distance between maternal residence and closest stream monitoring site with concentrations >MCL for NO_3-N, NO_2-N, or atrazine in surface water (USGS-NAWQA data [b])	Gastroschisis	Gastroschisis was not associated with maternal residential proximity to surface water with elevated nitrate (>10 mg/L) or nitrite (>1 mg/L)
Winchester, 2009 USA [112]	Ecologic study USA-wide	1996–2002	Rates of combined and specific birth defects (computed by month of last menstrual period) linked to monthly surface water nitrate concentrations (USGS-NAWQA data [b]); also evaluated atrazine and other pesticides (combined)	Birth defects categorized into 22 groups	Birth defect category "other congenital anomalies": OR for continuous log nitrate = 1.15 (CI: 1.12, 1.18); adjusted for atrazine and other pesticides: OR = 1.18, CI: 1.14, 1.21); No association with other birth defects

Abbreviations: CI, 95% CI confidence interval; OR, odds ratio; RR, rate ratio; USGS-NAWQA, U. S. Geological Survey National Water Quality Assessment; [a] nitrate units are specified as reported in publications. NO_3 can be converted to NO_3-N by multiplying by 0.2258; [b] USGS-NAWQA data for 186 streams in 51 hydrological study areas; streams were not drinking water sources.

7. Cancer

Most early epidemiologic studies of cancer were ecologic studies of stomach cancer mortality that used exposure estimates concurrent with the time of death. Results were mixed, with some studies showing positive associations, many showing no association, and a few showing inverse associations. The results of ecologic studies through 1995 were reviewed by Cantor [125]. Our previous review included ecologic studies of the brain, esophagus, stomach, kidney, ovary, and non-Hodgkin lymphoma (NHL) published between 1999 and 2003 that were largely null [8]. We did not include ecologic studies or mortality case-control studies in this review due to the limitations of these study designs, especially their inability to assess individual-level exposure and dietary factors that influence the endogenous formation of NOC.

Since our review of drinking water nitrate and health in 2005 [8], eight case-control studies and eight analyses in three cohorts have evaluated historical nitrate levels in PWS in relation to several cancers. Nitrate levels were largely below 10 mg/L NO_3-N. Most of these studies evaluated potential confounders and factors affecting nitrosation. Table 2 shows the study designs and results of studies published from 2005 through 2018, including findings from periodic follow-ups of a cohort study of postmenopausal women in Iowa (USA) [31,37,126–129]. In the first analysis of drinking water nitrate in the Iowa cohort with follow-up through 1998, Weyer and colleagues [130] reported that ovarian and bladder cancers were positively associated with the long-term average PWS nitrate levels prior to enrollment (highest quartile average 1955–1988: >2.46 mg/L NO_3-N). They observed inverse associations for uterine and rectal cancer, but no associations with cancers of the breast, colon, rectum, pancreas, kidney, lung, melanoma, non-Hodgkin lymphoma (NHL), or leukemia. Analyses of PWS nitrate concentrations and cancers of the thyroid, breast, ovary, bladder, and kidney were published after additional follow-up of the cohort. The exposure assessment was improved by: (a) the computation of average nitrate levels and years of exposure at or above 5 mg/L NO_3-N, based on time in residence (vs. one long-term PWS average nitrate estimate used by Weyer and colleagues); and (b) by estimation of total trihalomethanes (TTHM) and dietary nitrite intake.

Thyroid cancer was evaluated for the first time after follow-up of the cohort through 2004. A total of 40 cases were identified [37]. Among women with >10 years on PWS with levels exceeding 5 mg/L NO_3-N for five years or more, thyroid cancer risk was 2.6 times higher than that of women whose supplies never exceeded 5 mg/L. With follow-up through 2010, the risk of ovarian cancer remained increased among women in the highest quartile of average nitrate in PWS [129]. Ovarian cancer risk among private well users was also elevated compared to the lowest PWS nitrate quartile. Associations were stronger when vitamin C intake was below median levels with a significant interaction for users of private wells. Overall, breast cancer risk was not associated with water nitrate levels with follow-up through 2008 [128]. Among women with folate intake $\geq$ 400 µg/day, risk was increased for those in the highest average nitrate quintile (Hazard Ratio (HR) = 1.40; 95% CI: = 1.05–1.87) and among private well users (HR = 1.38; 95% CI: = 1.05–1.82), compared to those with the lowest average nitrate quintile. There was no association with nitrate exposure among women with lower folate intake. With follow-up through 2010, there were 130 bladder cancer cases among women who had used PWS >10 years. Risk remained elevated among women with the highest average nitrate levels and was 1.6 times higher among women whose drinking water concentration exceeded 5 mg/L NO_3-N for at least four years [31]. Risk estimates were not changed by adjustment for TTHM, which are suspected bladder cancer risk factors. Smoking, but not vitamin C intake, modified the association with nitrate in water; increased risk was apparent only in current smokers (p-interaction <0.03). With follow-up through 2010, there were 125 kidney cancer cases among women using PWS; risk was increased among those in the 95th percentile of average nitrate (>5.0 mg/L NO_3-N) compared with the lowest quartile (HR = 2.2, 95% CI: 1.2–4.2) [127]. There was no positive trend with the average nitrate level and no increased risk for women using private wells, compared to those with low average nitrate in their public supply. An investigation of pancreatic cancer in the same population (follow-up through 2011)

found no association with average water nitrate levels in public supplies and no association among women on private wells [126].

In contrast to the positive findings for bladder cancer among the cohort of Iowa women, a cohort study of men and women aged 55–69 in the Netherlands with lower nitrate levels in PWS found no association between water nitrate ingestion (median in top quintile = 2.4 mg/day NO_3-N) and bladder cancer risk [131]. Dietary intake of vitamins C and E and history of cigarette smoking did not modify the association. A hospital-based case-control study of bladder cancer in multiple areas of Spain [33] assessed lifetime water sources and usual intake of tap water. Nitrate levels in PWS were low, with almost all average levels below 2 mg/L NO_3-N. Risk of bladder cancer was not associated with the nitrate level in drinking water or with estimated nitrate ingestion from drinking water, and there was no evidence of interaction with factors affecting endogenous nitrosation.

Several case-control studies conducted in the Midwestern U.S. obtained lifetime histories of drinking water sources and estimated exposure for PWS users. In contrast to findings of an increased risk of NHL associated with nitrate levels in Nebraska PWS in an earlier study [132], there was no association with similar concentrations in public water sources in a case-control study of NHL in Iowa [35]. A study of renal cell carcinoma in Iowa [34] found no association with the level of nitrate in PWS, including the number of years that levels exceeded 5 or 10 mg/L NO_3-N. However, higher nitrate levels in PWS increased risk among subgroups who reported above the median intake of red meat intake or below the median intake of vitamin C (p-interaction <0.05). A small case-control study of adenocarcinoma of the stomach and esophagus among men and women in Nebraska [133] estimated nitrate levels among long-term users of PWS and found no association between average nitrate levels and risk.

A case-control study of colorectal cancer among rural women in Wisconsin estimated nitrate levels in private wells using spatial interpolation of nitrate concentrations from a 1994 water quality survey and found increased risk of proximal colon cancer among women estimated to have nitrate levels >10 mg/L NO_3-N compared to levels < 0.5 mg/L. Risk of distal colon cancer and rectal cancer were not associated with nitrate levels [134]. Water nitrate ingestion from public supplies, bottled water, and private wells and springs over the adult lifetime was estimated in analyses that pooled case-control studies of colorectal cancer in Spain and Italy [135]. Risk of colorectal cancer was increased among those with >2.3 mg/day NO_3-N (vs. <1.1 mg/day). There were no interactions with red meat, vitamins C and E, and fiber except for a borderline interaction (p-interaction = 0.07) for rectum cancer with fiber intake. A small hospital-based case-control study in Indonesia found that drinking water nitrate levels above the WHO standard (>11.3 mg/L as NO_3-N) was associated with colorectal cancer [136]. A national registry-based cohort study in Denmark [32] evaluated average nitrate concentrations in PWS and private wells in relation to colorectal cancer incidence among those whose 35th birthday occurred during 1978–2011. The average nitrate level was computed over residential water supplies from age 20 to 35. Increased risks for colon and rectum cancer were observed in association with average nitrate levels ≥9.25 mg/L NO_3 (≥2.1 as NO_3-N) and ≥3.87 mg/L NO_3 (>0.87 as NO_3-N), respectively, with a significant positive trend. Because the study did not interview individuals, it could not evaluate individual-level risk factors that might influence endogenous nitrosation.

A case-control study of breast cancer in Cape Cod, Massachusetts (US) [137] estimated nitrate concentrations in PWS over approximately 20 years as an historical proxy for wastewater contamination and potential exposure to endocrine disruption compounds. Average exposures >1.2 mg/L NO_3-N (vs. <0.3 mg/L) were not associated with risk. A hospital-based case-control study in Spain found no association between water nitrate ingestion and pre- and post-menopausal breast cancers [138].

Table 2. Case-control and cohort studies of drinking water nitrate and cancer (January 2004–March 2018) by cancer site.

First Author (Year) Country	Study Design, Years Regional Description	Exposure Description	Cancer Sites Included	Summary of Drinking-Water Findings [a,b]	Evaluation of Effect Modification [c]
Zeegers, 2006 Netherlands [131]	Cohort Incidence, 1986–1995 204 municipal registries across the Netherlands	1986 nitrate level in 364 pumping stations, exposure data available for 871 cases, 4359 members of the subcohort	Bladder	Highest vs. lowest quintile intake from water (≥1.7 mg/day NO_3-N [median 2.4 mg/day] vs. <0.20) RR = 1.11 (CI: 0.87–1.41; *p*-trend = 0.14)	No interaction with vitamin C, E, smoking
Espejo-Herrera, 2015 Spain [33]	Hospital-based multi-center case-control Incidence, 1998–2001 Asturias, Alicante, Barcelona, Vallès-Bages, Tenerife provinces	Nitrate levels in PWS (1979–2010) and bottled water (measurements of brands with highest consumption based on a Spanish survey); analyses limited to those with ≥70% of residential history with nitrate estimate (531 cases, 556 controls)	Bladder	Highest vs. lowest quartile average level (age 18-interview) (≥2.26 vs. 1.13 mg/L NO_3-N) OR = 1.04 (CI: 0.60–1.81) Years >2.15 mg/L NO_3-N (75th percentile) (>20 vs. 0 years) OR = 1.41 (CI: 0.89–2.24)	No interaction with vitamin C, E, red meat, processed meat, average THM level
Jones, 2016 USA [31]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2010 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence with nitrate and trihalomethane estimates (20,945 women; 170 bladder cases); no measurements for private wells Adjusted for total trihalomethanes (TTHM)	Bladder	Highest vs. lowest quartile PWS average (≥2.98 vs. <0.47 mg/L NO_3-N) HR = 1.47 (CI: 0.91–2.38; *p*-trend = 0.11) Years >5 mg/L (≥4 years vs. 0) HR = 1.61 (CI: 1.05–2.47; *p*-trend = 0.03) Private well users (vs. <0.47 mg/L NO_3-N on PWS) HR = 1.53 (CI: 0.93–2.54)	Interaction with smoking (*p*-interaction = 0.03); HR = 3.67 (CI: 1.43–9.38) among current smokers/≥2.98 mg/L vs. non-smokers/<0.47 mg/L NO_3-N); No interaction with vitamin C, TTHM levels
Mueller, 2004 USA, Canada, France, Italy, Spain [139]	Pooled case-control studies Incidence among children <15 years (USA <20 years) 7 regions of 5 countries	Water source during pregnancy and first year of child's life (836 cases, 1485 controls); nitrate test strip measurements of nitrate and nitrite for pregnancy home (except Italy) (283 cases, 537 controls; excluding bottled water users: 207 cases, 400 controls)	Brain, childhood	Private well use versus PWS associated with increased risk in 2 regions and decreased risk in one; No association with nitrate levels in water supplies Astrocytomas (excludes bottled water users): ≥1.5 vs. <0.3 mg/L NO_2-N OR = 5.7 (CI: 1.2–27.2)	Not described
Brody, 2006 USA [137]	Case-control Incidence, 1988–1995 Cape Cod, Massachusetts	Nitrate levels in public water supplies (PWS) since 1972 was used as an indicator of wastewater contamination and potential mammary carcinogens and endocrine disrupting compounds; excluded women on private wells	Breast	Average ≥1.2 mg/L NO_3-N vs. <0.3 OR = 1.8, (CI: 0.6–5.0); summed annual NO_2-N ≥ 10 vs. 1–< 10 mg/L OR = 0.9, CI: 0.6–1.5); number of years >1 mg/L NO_3-N ≥8 vs. 0 years OR = 0.9 (CI: 0.5–1.5)	Not described

Table 2. *Cont.*

First Author (Year) Country	Study Design, Years Regional Description	Exposure Description	Cancer Sites Included	Summary of Drinking-Water Findings [a,b]	Evaluation of Effect Modification [c]
Inoue-Choi, 2012 USA [128]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2008 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence (20,147 women; 1751 breast cases); no measurements for private wells	Breast	Highest vs. lowest quintile PWS average (≥3.8 vs. ≤0.32 mg/L NO_3-N) HR = 1.14 (CI: 0.95–1.36; *p*-trend = 0.11); Private well (vs. ≤ 0.32 mg/L NO_3-N) HR = 1.14 (CI: 0.97–1.34); Private well (vs. ≤0.32 mg/L NO_3-N on PWS) HR = 1.38 (CI: 1.05–1.82); No association among those with low folate <400 µg/day	Interaction with folate for PWS (*p*-interaction = 0.06). Folate >400 µg/d: (>3.8 vs. ≤0.32 mg/L NO_3-N) HR = 1.40 (CI: 1.05–1.87; *p*-trend = 0.04)
Espejo-Herrera, 2016 Spain [138]	Hospital-based multi-center case-control Incidence, 2008–2013 Spain (8 provinces)	Nitrate levels in PWS (2004–2010), bottled water measurements and private wells and springs (2013 measurements in 21 municipalities in León, Spain, the area with highest non-PWS use) Analyses include women with ≥70% of period from age 18 to 2 years before interview (1245 cases, 1520 controls)	Breast	Water nitrate intake based on average nitrate levels (age 18 to 2 years prior to interview) and water intake (L/day). Post-menopausal women: >2.0 vs. 0.5 mg/day NO_3-N OR = 1.32 (0.93–1.86); Premenopausal women: >1.4 vs. 0.4 mg/day NO_3-N OR = 1.14 (0.67–1.94)	No interaction with red meat, processed meat, vitamin C, E, smoking for pre- and post-menopausal women
McElroy, 2008 USA [134]	Population-based case-control, women Incidence, 1990–1992 and 1999–2001 Wisconsin	Limited to women in rural areas with no public water system (475 cases, 1447 controls); nitrate levels at residence (presumed to be private wells) estimated by kriging using data from a 1994 representative sample of 289 private wells	Colorectal	All colon cancers: Private wells ≥10.0 mg/L NO_3-N vs. <0.5 OR = 1.52 (CI: 0.95–2.44); Proximal colon cancer: OR = 2.91 (CI: 1.52–5.56)	Not described
Espejo-Herrera, 2016 Spain, Italy [135]	Multi-center case-control study Incidence, 2008–2013 Spain (9 provinces) and population-based controls; Italy (two provinces) and hospital-based controls	Nitrate levels in PWS (2004–2010) for 349 water supply zones, bottled water (measured brands with highest consumption), and private wells and springs (measurements in 2013 in 21 municipalities in León, Spain, the area with highest non-PWS use) Analyses include those with nitrate estimates for ≥70% of period 30 years before interview (1869 cases, 3530 controls)	Colorectal	Water nitrate intake based on average nitrate levels (estimated 30 to 2 years prior to interview) and water intake (L/day) Highest vs. lowest exposure quintiles (≥2.3 vs. <1.1 mg /day NO_3-N) OR = 1.49 (CI:1.24–1.78); Colon OR = 1.52 (CI: 1.24–1.86), Rectum OR = 1.62 (CI: 1.23–2.14)	Interaction with fiber for rectum (*p*-interaction = 0.07); >20 g/day fiber + >1.0 mg/L NO_3-N vs. <20 g/day + ≤1.0 mg/L HR = 0.72 (CI: 0.52–1.00). No interaction with red meat, vitamin C, E

Table 2. *Cont.*

First Author (Year) Country	Study Design, Years Regional Description	Exposure Description	Cancer Sites Included	Summary of Drinking-Water Findings [a,b]	Evaluation of Effect Modification [c]
Fathmawati, 2017 Indonesia [136]	Hospital-based case-control Incidence, 2014–2016 Indonesia (3 provinces)	Nitrate levels in well water collected during the raining season (Feb-March 2016) and classified based on >11.3 or ≤11.3 mg/L as NO_3-N and duration of exposure >10 and ≤10 years Analyses included participants who reported drinking well water (75 cases, 75 controls)	Colorectal	Water nitrate > WHO standard vs. below (> 11.3 vs. ≤11.3 mg/L NO_3-N) OR = 2.82 (CI: 1.08–7.40); > 10 years: 4.31 (CI: 11.32–14.10); ≤10 years: 1.41 (CI: 0.14–13.68)	Not described
Schullehner, 2018 Denmark [32]	Population-based record-linkage cohort of men and women ages 35 and older, 1978–2011 Denmark	Nitrate levels in PWS and private wells among 1,742,321 who met exposure assessment criteria (5944 colorectal cancer cases, including 3700 with colon and 2308 with rectal cancer)	Colorectal	Annual average nitrate exposure between ages 20–35 among those who lived ≥75% of study period at homes with a water sample within 1 year (61% of Danish population). Highest vs. lowest exposure quintile (≥2.1 vs. 0.16 mg/L NO_3-N); Colorectal: HR = 1.16 (CI: 1.08–1.25); colon: 1.15 (CI: 1.05–1.26); rectum: 1.17 (CI: 1.04–1.32)	No information on dietary intakes or smoking
Ward, 2007 USA [34]	Population-based case control Incidence, 1986–1989 Iowa	Nitrate levels in PWS among those with nitrate estimates for ≥70% of person-years ≥1960 (201 cases, 1244 controls)	Kidney (renal cell carcinomas)	Highest vs. lowest quartile PWS average (≥2.8 mg/L NO_3-N vs. <0.62) OR = 0.89 (CI 0.57–1.39); Years >5mg/L NO_3-N 11+ vs. 0 OR = 1.03 (CI: 0.66–1.60)	Interaction with red meat intake (p-interaction = 0.01); OR = 1.91 (CI 1.04–3.51) among 11+ years >5 mg/L NO_3-N and red meat >1.2 servings/day. Interaction with vitamin C showed similar pattern (p-interaction = 0.13)
Jones, 2017 USA [127]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2010 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence. PWS measurements for nitrate and TTHM; no measurements for private wells (20,945 women; 163 kidney cases)	Kidney	Nitrate and TTHM metrics computed for duration at water source (11+ years) 95th percentile vs. lowest quartile PWS average (≥5.00 vs. <0.47 mg/L NO_3-N) HR = 2.23 (CI: 1.19–4.17; p-trend = 0.35) Years >5 mg/L (≥4 years vs. 0) HR = 1.54 (CI: 0.97–2.44; p-trend = 0.09) Private well users (vs. <0.47 mg/L NO_3-N in PWS) HR = 0.96 (CI: 0.59–1.58)	No interaction with smoking, vitamin C
Ward, 2006 USA [35]	Population-based case-control Incidence, 1998–2000 Iowa	Nitrate levels in PWS among those with nitrate estimates for ≥70% of person-years ≥1960 (181 case, 142 controls); nitrate measurements for private well users at time of interviews (1998–2000; 54 cases, 44 controls)	Non-Hodgkin lymphoma	Private wells: >5.0 mg/L NO_3-N vs. ND OR = 0.8 (CI 0.2–2.5) PWS average: ≥2.9 mg/L NO_3-N vs. <0.63 OR = 1.2 (CI 0.6–2.2) Years ≥5mg/L NO3-N: 10+ vs. 0 OR = 1.4 (CI: 0.7–2.9)	No interaction with vitamin C, smoking

Table 2. *Cont.*

First Author (Year) Country	Study Design, Years Regional Description	Exposure Description	Cancer Sites Included	Summary of Drinking-Water Findings [a,b]	Evaluation of Effect Modification [c]
Inoue-Choi, 2015 USA [129]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2010 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence; PWS measurements for nitrate and TTHM; no measurements for private wells (17,216 women; 190 ovarian cases)	Ovary	Nitrate and TTHM metrics computed for reported duration at water source (11+ years) Highest vs. lowest quartile PWS average (≥2.98 mg/L vs. <0.47 mg/L NO_3-N) HR = 2.03 (CI = 1.22–3.38; *p*-trend = 0.003) Years >5 mg/L (≥4 years vs. 0) HR = 1.52 (CI: 1.00–2.31; *p*-trend = 0.05) Private well users (vs. <0.47 mg/L NO_3-N in PWS) HR = 1.53 (CI: 0.93–2.54)	No interaction with vitamin C, red meat intake, smoking for PWS nitrate Interaction with private well use and vitamin C intake (*p*-interaction = 0.01)
Quist, 2018 USA [126]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2011 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence; nitrate and TTHM estimates for PWS (20,945 women; 189 pancreas cases); no measurements for private wells Adjusted for TTHM (1955–1988), measured levels in 1980s, prior year levels estimated by expert)	Pancreas	Nitrate and TTHM metrics computed for reported duration at water source (11+ years) 95th percentile vs. lowest quartile PWS average (≥5.69 vs. <0.47 mg/L NO_3-N) HR = 1.16 (CI: 0.51–2.64; *p*-trend = 0.97) Years >5 mg/L (≥4 years vs. 0) HR = 0.90 (CI: 0.55–1.48; *p*-trend = 0.62) Private well users (vs. <0.47 mg/L NO_3-N) HR = 0.92 (CI: 0.55–1.52)	No interaction with smoking, vitamin C
Ward, 2008 USA [133]	Population-based case control Incidence, 1988–1993 Nebraska	Controls from prior study of lymphohematopoetic cases and controls interviewed in 1992–1994; Proxy interviews for 80%, 76%, 61% of stomach, esophagus, controls, respectively. Nitrate levels (1965–1985) in PWS for ≥70% of person-years (79 distal stomach, 84, esophagus, 321 controls); Private well users sampling at interview (15 stomach, 22 esophagus, 44 controls)	Stomach and esophagus (adenocarcinomas)	Highest vs. lowest quartile PWS average (>4.32 vs. <2.45 mg/L NO_3-N): stomach OR = 1.2 (CI 0.5–2.7); esophagus OR = 1.3 (CI: 0.6–3.1); Years >10 mg/L NO_3-N (9+ vs. 0): stomach OR = 1.1 (CI: 0.5–2.3); esophagus OR = 1.2 (CI: 0.6–2.7) Private well users (>4.5 mg/L NO_3-N vs. <0.5) stomach OR = 5.1 (CI: 0.5–52; 4 cases, 13 controls); esophagus OR = 0.5 (CI: 0.1–2.9; 8 cases; 13 controls)	No interaction with vitamin C, processed meat, or red meat for either cancer
Ward, 2010 USA [37]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2004 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence (21,977 women; 40 thyroid cases); no measurements for private wells	Thyroid	Highest vs. lowest quartile PWS average (>2.46 vs. <0.36 mg/L NO_3-N) HR = 2.18 (CI: 0.83–5.76; *p*-trend = 0.02) Years >5 mg/L (≥5 years vs. 0) HR = 2.59 (CI: 1.09–6.19; *p*-trend = 0.04); Private well (vs. <0.36 mg/L NO_3-N on PWS) HR = 1.13 (CI: 0.83–3.66) Dietary nitrate intake quartiles positively associated with risk (*p*-trend = 0.05)	No interaction with smoking, vitamin C, body mass index, education, residence location (farm/rural vs. urban)

ND = not detected; PWS = public water supplies; [a] nitrate or nitrite levels presented in the publications as mg/L of the ion were converted to mg/L as NO_3-N or NO_2-N; [b] Odds ratios (OR) for case-control studies, incidence rate ratios (RR) and hazard ratios (HR) for cohort studies, and 95% confidence intervals (CI); [c] Factors evaluated are noted. Interaction refers to reported $p \leq 0.10$ from test of heterogeneity.

Animal studies demonstrate that in utero exposure to nitrosamides can cause brain tumors in the exposed offspring. Water nitrate and nitrite intake during pregnancy was estimated in a multi-center case-control study of childhood brain tumors in five countries based on the maternal residential water source [139]. Results for the California and Washington State sites were reported in our previous review [8,140]. Nitrate/nitrite levels in water supplies were measured using a nitrate test strip method in four countries including these U.S. sites; most of these measurements occurred many years after the pregnancy. Measured nitrate concentrations were not associated with risk of childhood brain tumors. However, higher nitrite levels (>1.5 mg/L NO_2-N) in the drinking water were associated with increased risk of astrocytomas.

8. Thyroid Disease

Animal studies demonstrate that ingestion of nitrate at high doses can competitively inhibit iodine uptake and induce hypertrophy of the thyroid gland [141]. An early study of women in the Netherlands consuming water with nitrate levels at or above the MCL, found increased prevalence of thyroid hypertrophy [142]. Since the last review, five studies have evaluated nitrate ingestion from drinking water (the Iowa cohort study also assessed diet) and prevalence of thyroid disease. A study of school-age children in Slovakia found increased prevalence of subclinical hypothyroidism among children in an area with high nitrate levels (51–274 mg/L NO_3) in water supplies compared with children ingesting water with nitrate ≤50 mg/L (11 mg/L NO_3-N). In Bulgarian villages with high nitrate levels (75 mg/L NO_3) and low nitrate levels (8 mg/L), clinical examinations of the thyroids of pregnant women and school children revealed an approximately four- and three-fold increased prevalence of goiter, respectively, in the high nitrate village [143,144]. The iodine status of the populations in both studies was adequate. Self-reported hypothyroidism and hyperthyroidism among a cohort of post-menopausal women in Iowa was not associated with average nitrate concentrations in PWS [37]. However, dietary nitrate, the predominant source of intake, was associated with increased prevalence of hypothyroidism but not hyperthyroidism. Modeled estimates of nitrate concentrations in private wells among a cohort of Old Order Amish in Pennsylvania (USA) were associated with increased prevalence of subclinical hypothyroidism as determined by thyroid stimulating hormone measurements, among women but not men [145].

9. Other Health Effects

Associations between nitrate in drinking water and other non-cancer health effects, including type 1 childhood diabetes (T1D), blood pressure, and acute respiratory tract infections in children were previously reviewed [8]. Since 2004, a small number of studies have contributed additional mixed evidence for these associations. Animal studies indicate that NOC may play a role in the pathology of T1D through damage to pancreatic beta cells [146]. A registry-based study in Finland [147] found a positive trend in T1D incidence with levels of nitrate in drinking water. In contrast, an ecological analysis in Italy showed an inverse correlation with water nitrate levels and T1D rates [148]. A small T1D case-control study in Canada with 57 cases showed no association between T1D and estimated intake of nitrate from drinking water (highest quartile >2.7 mg/day NO_3-N) [149]. Concentrations of nitrate in drinking water (median ~2.1 mg/L NO_3-N) were not associated with progression to T1D in a German nested case-control study of islet autoantibody-positive children, who may be at increased risk of the disease [150].

In a prospective, population-based cohort study in Wisconsin (USA), increased incidence of early and late age-related macular degeneration was positively associated with higher nitrate levels (≥5 mg/L vs. <5 mg/L NO_3-N) in rural private drinking water supplies [151]. The authors suggested several possible mechanisms, including methemoglobin-induced lipid peroxidation in the retina.

Potential benefits of nitrate ingestion include lowering of blood pressure due to production of nitric oxide in the acidic stomach and subsequent vasodilation, antithrombotic, and immunoregulatory effects [152]. Experimental studies in animals and controlled feeding studies in humans have

demonstrated mixed evidence of these effects and on other cardiovascular endpoints such as vascular hypertrophy, heart failure, and myocardial infarction (e.g., [152–154]). Ingested nitrite from diet has also been associated with increased blood flow in certain parts of the brain [155]. Epidemiologic studies of these effects are limited to estimation of dietary exposures or biomarkers that integrate exposures from nitrate from diet and drinking water. Recent findings in the Framingham Offspring Study suggested that plasma nitrate was associated with increased overall risk of death that attenuated when adjusted for glomerular function (HR: 1.16, 95% CI: 1.0–1.35) but no association was observed for incident cardiovascular disease [156]. No epidemiologic studies have specifically evaluated nitrate ingested from drinking water in relation to these outcomes. Another potential beneficial effect of nitrate is protection against bacterial infections via its reduction to nitrite by enteric bacteria. In an experimental inflammatory bowel disease mouse model, nitrite in drinking water was associated with both preventive and therapeutic effects [157]. However, there is limited epidemiologic evidence for a reduced risk of gastrointestinal disease in populations with high drinking water nitrate intake. One small, cross-sectional study in Iran found no association between nitrate levels in public water supplies with mean levels of ~5.6 mg/L NO_3-N and gastrointestinal disease [158].

10. Discussion

Since our last review of studies through 2004 [8], more than 30 epidemiologic studies have evaluated drinking water nitrate and risk of cancer, adverse reproductive outcomes, or thyroid disease. However, the number of studies of any one outcome was not large and there are still too few studies to allow firm conclusions about risk. The most common endpoints studied were colorectal cancer, bladder, and breast cancer (three studies each) and thyroid disease (four studies). Considering all studies to date, the strongest evidence for a relationship between drinking water nitrate ingestion and adverse health outcomes (besides methemoglobinemia) is for colorectal cancer, thyroid disease, and neural tube defects. Four of the five published studies of colorectal cancer found evidence of an increased risk of colorectal cancer or colon cancer associated with water nitrate levels that were mostly below the respective regulatory limits [32,134,135,159]. In one of the four positive studies [159], increased risk was only observed in subgroups likely to have increased nitrosation. Four of the five studies of thyroid disease found evidence for an increased prevalence of subclinical hypothyroidism with higher ingestion of drinking water nitrate among children, pregnant women, or women only [37,144,145,160]. Positive associations with drinking water nitrate were observed at nitrate concentrations close to or above the MCL. The fifth study, a cohort of post-menopausal women in Iowa, had lower drinking water nitrate exposure but observed a positive association with dietary nitrate [37]. To date, five of six studies of neural tube defects showed increased risk with exposure to drinking water nitrate below the MCL. Thus, the evidence continues to accumulate that higher nitrate intake during the pregnancy is a risk factor for this group of birth defects.

All but one of the 17 cancer studies conducted since 2004 were in the U.S. or Europe, the majority of which were investigations of nitrate in regulated public drinking water. Thyroid cancer was studied for the first time [37] with a positive finding that should be evaluated in future studies. Bladder cancer, a site for which other drinking water contaminants (arsenic, disinfection by-products [DBPs]) are established or suspected risk factors, was not associated with drinking water nitrate in three of the four studies. Most of the cancer studies since 2004 evaluated effect modification by factors known to influence endogenous nitrosation, although few observed evidence for these effects. Several studies of adverse reproductive outcomes since 2004 have indicated a positive association between maternal prenatal exposure to nitrate concentrations below the MCL and low birth weight and small for gestational age births. However, most studies did not account for co-exposure to other water contaminants, nor did they adjust for potential risk factors. The relation between drinking water nitrate and spontaneous abortion continues to be understudied. Few cases of methemoglobinemia, the health concern that lead to the regulation of nitrate in public water supplies, have been reported in the U.S. since the 1990s. However, as described by Knobeloch et al. [96], cases may be underreported

and only a small proportion of cases are thoroughly investigated and described in the literature. Based on published reports, [100] areas of the world of particular concern include several eastern European countries, Gaza, and Morocco, where high nitrate concentrations in water supplies have been linked to high levels of methemoglobin in children. Therefore, continued surveillance and education of physicians and parents will be important. Biological plausibility exists for relationships between nitrate ingestion from drinking water and a few other health outcomes including diabetes and beneficial effects on the cardiovascular system, but there have been only a limited number of epidemiologic studies.

Assessment of drinking water nitrate exposures in future studies should be improved by obtaining drinking water sources at home and at work, estimating the amount of water consumed from each source, and collecting information on water filtration systems that may impact exposure. These efforts are important for reducing misclassification of exposure. Since our last review, an additional decade of PWS monitoring data are available in the U.S. and European countries, which has allowed assessment of exposure over a substantial proportion of participants' lifetimes in recent studies. Future studies should estimate exposure to multiple water contaminants as has been done in recent cancer studies [31,33,127,129]. For instance, nitrate and atrazine frequently occur together in drinking water in agricultural areas [161] and animal studies have found this mixture to be teratogenic [162]. Regulatory monitoring data for pesticides in PWS has been available for over 20 years in the U.S.; therefore, it is now feasible to evaluate co-exposure to these contaminants. Additionally, water supplies in agricultural areas that rely on alluvial aquifers or surface water often have elevated levels of both DBPs and nitrate. Under this exposure scenario, there is the possibility of formation of the nitrogenated DBPs including the carcinogenic NDMA, especially if chloramination treatment is used for disinfection [163,164]. Studies of health effects in countries outside the U.S. and Europe are also needed.

A comprehensive assessment of nitrate and nitrite from drinking water and dietary sources as well as estimation of intakes of antioxidants and other inhibitors of endogenous nitrosation including dietary polyphenols and flavonoids is needed in future studies. Heme iron from red meat, which increases fecal NOC in human feeding studies, should also be assessed as a potential effect modifier of risk from nitrate ingestion. More research is needed on the potential interaction of nitrate ingestion and nitrosatable drugs (those with secondary and tertiary amines or amides). Evidence from several studies of birth defects [38,118–120] implicates nitrosatable drug intake during pregnancy as a risk factor for specific congenital anomalies especially in combination with nitrate. Drugs with nitrosatable groups include many over-the-counter and prescription drugs. Future studies with electronic medical records and record-linkage studies in countries like Denmark with national pharmacy data may provide opportunities for evaluation of these exposures.

Populations with the highest exposure to nitrate from their drinking water are those living in agricultural regions, especially those drinking water from shallow wells near nitrogen sources (e.g., crop fields, animal feeding operations). Estimating exposure for private well users is important because it allows assessment of risk over a greater range of nitrate exposures compared to studies focusing solely on populations using PWS. Future health studies should focus on these populations, many of which may have been exposed to elevated nitrate in drinking water from early childhood into adulthood. A major challenge in conducting studies in these regions is the high prevalence of private well use with limited nitrate measurement data for exposure assessment. Recent efforts to model nitrate concentrations in private wells have shown that it is feasible to develop predictive models where sufficient measurement data are available [41,48,49]. However, predictive models from one area are not likely to be directly translatable to other geographic regions with different aquifers, soils, and nitrogen inputs.

Controlled human feeding studies have demonstrated that endogenous nitrosation occurs after ingestion of drinking water with nitrate concentrations above the MCL of 10 mg/L NO_3-N (~44 mg/L as NO_3). However, the extent of NOC formation after ingestion of drinking water with nitrate

concentrations below the MCL has not been well characterized. Increased risks of specific cancers and central nervous system birth defects in study populations consuming nitrate below the MCL is indirect evidence that nitrate ingestion at these levels may be a risk factor under some conditions. However, confounding by other exposures or risk factors can be difficult to rule out in many studies. Controlled human studies to evaluate endogenous nitrosation at levels below the MCL are needed to understand interindividual variability and factors that affect endogenous nitrosation at drinking water nitrate levels below the MCL.

A key step in the endogenous formation of NOC is the reduction of nitrate, which has been transported from the bloodstream into the saliva, to nitrite by the nitrate-reducing bacteria that are located primarily in the crypts on the back of the tongue [165–167]. Tools for measuring bacterial DNA and characterizing the oral microbiome are now available and are currently being incorporated into epidemiologic studies [168,169]. Buccal cell samples that have been collected in epidemiologic studies can be used to characterize the oral microbiome and to determine the relative abundance of the nitrate-reducing bacteria. Studies are needed to characterize the stability of the nitrate-reducing capacity of the oral microbiome over time and to determine factors that may modify this capacity such as diet, oral hygiene, and periodontal disease. Interindividual variability in the oral nitrate-reducing bacteria may play an important role in modifying endogenous NOC formation. The quantification of an individual's nitrate-reducing bacteria in future epidemiologic studies is likely to improve our ability to classify participants by their intrinsic capacity for endogenous nitrosation.

In addition to characterizing the oral microbiome, future epidemiologic studies should incorporate biomarkers of NOC (e.g., urinary or fecal NOC), markers of genetic damage, and determine genetic variability in NOC metabolism. As many NOC require α-hydroxylation by CYP2E1 for bioactivation and for formation of DNA adducts, it is important to investigate the influence of polymorphisms in the gene encoding for this enzyme. Studies are also needed among populations with medical conditions that increase nitrosation such as patients with inflammatory bowel disease and periodontal disease [8]. Because NOC exposures induce characteristic gene expression profiles [170,171], further studies linking drinking water intake to NOC excretion and gene expression responses are relevant to our understanding of health risks associated with drinking water nitrate. The field of 'Exposome-research' [172,173] generates large numbers of genomics profiles in human population studies for which dietary exposures and biobank materials are also available. These studies provide opportunities to measure urinary levels of nitrate and NOC that could be associated with molecular markers of exposure and disease risk.

Nitrate concentrations in global water supplies are likely to increase in the future due to population growth, increases in nitrogen fertilizer use, and increasing intensity and concentration of animal agriculture. Even with increased inputs, mitigation of nitrate concentrations in water resources is possible through local, national, and global efforts. Examples of the latter are the International Nitrogen Initiative [174] and the EU Nitrates Directive [17,18], which aim to quantify human effects on the nitrogen cycle and to validate and promote methods for sustainable nitrogen management. Evidence for the effectiveness of these efforts, which include the identification of vulnerable areas, establishment of codes of good agricultural practices, and national monitoring and reporting are indicated by decreasing trends in groundwater nitrate concentrations in some European countries after the implementation of the EU Nitrates Directive [19]. However, the effect of this initiative was variable across the EU. In the U.S., nitrogen applications to crop fields are not regulated and efforts to reduce nitrogen runoff are voluntary. Although strategies such as appropriate timing of fertilizer applications, diversified crop rotations, planting of cover crops, and reduced tillage can be effective [175], concentrations in U.S. ground and surface water have continued to increase in most areas [10]. Climate change is expected to affect nitrogen in aquatic ecosystems and groundwater through alterations of the hydrological cycle [176]. Climatic factors that affect nitrate in groundwater include the amount, intensity, and timing of precipitation. Increasing rainfall intensity, especially in

the winter and spring, can lead to increases in nitrogen runoff from agricultural fields and leaching to groundwater.

11. Conclusions

In summary, most adverse health effects related to drinking water nitrate are likely due to a combination of high nitrate ingestion and factors that increase endogenous nitrosation. Some of the recent studies of cancer and some birth defects have been able to identify subgroups of the population likely to have greater potential for endogenous nitrosation. However, direct methods of assessing these individuals are needed. New methods for quantifying the nitrate-reducing bacteria in the oral microbiome and characterizing genetic variation in NOC metabolism hold promise for identifying high risk groups in epidemiologic studies.

To date, the number of well-designed studies of individual health outcomes is still too few to draw firm conclusions about risk from drinking water nitrate ingestion. Additional studies that incorporate improved exposure assessment for populations on PWS, measured or predicted exposure for private well users, quantification of nitrate-reducing bacteria, and estimates of dietary and other factors affecting nitrosation are needed. Studies of colorectal cancer, thyroid disease, and central nervous system birth defects, which show the most consistent associations with water nitrate ingestion, will be particularly useful for clarifying these risks. Future studies of other health effects with more limited evidence of increased risk are also needed including cancers of the thyroid, ovary, and kidney, and the adverse reproductive outcomes of spontaneous abortion, preterm birth, and small for gestational age births.

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Submitted by Peter Herridge at 3/12/2020 Hearing

Journal Article submission to the ZBA

This packet contains five recent journal articles from well respected, refereed journals on medicine and environmental research. These articles are being submitted to be made of record in the Public Hearing held by the Zoning Board of Appeals (ZBA) of Truro, Massachusetts concerning the decision to grant a waste water waiver to the proposed Cloverleaf housing project held on March 12, 2020. It is the intention of this submission to demonstrate to the ZBA the fact that nitrate pollution in drinking water poses very serious risks of adverse medical consequences to people who drink this water on both an acute and prolonged basis. These articles also clearly show that even levels of nitrates that are below the maximum nitrate level presently allowed in the US, i.e. 10 mg of nitrogen as nitrate per liter of water, can cause a wide variety of serious medical consequences including cancer, birth defects, spontaneous abortion and thyroid disease.

The contents are:

1) *Exposure-based assessment and economic valuation of adverse birth outcomes and cancer risk due to nitrate in United States drinking water*. Author: Temkin A
2) *Nitrate toxicity and drinking water standards*. Author: Kross BC
3) *Thyroid cancer induction: Nitrates as independent risk factors or risk modulators after radiation exposure, with a focus on the Chernobyl accident*. Author: Drozd VA
4) *Drinking water nitrate and human health: an updated review.* Author: Ward MH
5) *Prenatal nitrate intake from drinking water and selected birth defects in offspring of the participants in the National Birth Defects Prevention Study*. Author: Brender JD

This packet is submitted to the Truro ZBA on March 12, 2020 by Peter Herridge MD, JD.

Prenatal Nitrate Intake from Drinking Water and Selected Birth Defects in Offspring of Participants in the National Birth Defects Prevention Study

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Background: Previous studies of prenatal exposure to drinking-water nitrate and birth defects in offspring have not accounted for water consumption patterns or potential interaction with nitrosatable drugs.

Objectives: We examined the relation between prenatal exposure to drinking-water nitrate and selected birth defects, accounting for maternal water consumption patterns and nitrosatable drug exposure.

Methods: With data from the National Birth Defects Prevention Study, we linked addresses of 3,300 case mothers and 1,121 control mothers from the Iowa and Texas sites to public water supplies and respective nitrate measurements. We assigned nitrate levels for bottled water from collection of representative samples and standard laboratory testing. Daily nitrate consumption was estimated from self-reported water consumption at home and work.

Results: With the lowest tertile of nitrate intake around conception as the referent group, mothers of babies with spina bifida were 2.0 times more likely (95% CI: 1.3, 3.2) to ingest ≥ 5 mg nitrate daily from drinking water (vs. < 0.91 mg) than control mothers. During 1 month preconception through the first trimester, mothers of limb deficiency, cleft palate, and cleft lip cases were, respectively, 1.8 (95% CI: 1.1, 3.1), 1.9 (95% CI: 1.2, 3.1), and 1.8 (95% CI: 1.1, 3.1) times more likely than control mothers to ingest ≥ 5.42 mg of nitrate daily (vs. < 1.0 mg). Higher water nitrate intake did not increase associations between prenatal nitrosatable drug use and birth defects.

Conclusions: Higher water nitrate intake was associated with several birth defects in offspring, but did not strengthen associations between nitrosatable drugs and birth defects.

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Introduction

Nitrate is one of the most widespread chemical contaminants in aquifers around the world (Spalding and Exner 1993). Results from several epidemiologic studies have suggested an association between prenatal exposure to nitrates in drinking water and birth defects in offspring, including neural tube defects (NTDs) (Brender et al. 2004; Croen et al. 2001; Dorsch et al. 1984), central nervous system defects overall (Arbuckle et al. 1988), oral cleft defects (Dorsch et al. 1984), musculoskeletal defects (Dorsch et al. 1984), and congenital heart defects (Cedergren et al. 2002). In these studies, exposure was assigned on the basis of nitrate levels detected in drinking-water sources without further estimating individual consumption of nitrate from such sources. It is noteworthy that previous associations observed between birth defects and nitrates in drinking water were often observed at levels below the current allowable maximum contaminant level for nitrate (10 mg/L as nitrate-nitrogen or 45 mg/L as total nitrate) set by the U.S. Environmental Protection Agency (National Primary Drinking Water Regulations 2010).

Once ingested and absorbed, approximately 25% of nitrate is secreted in saliva (Mensinga et al. 2003), where about 20% is converted to nitrite by bacteria in the mouth (Spiegelhalder et al. 1976). This endogenously formed nitrite, along with nitrite from dietary and drinking-water sources, can react with nitrosatable compounds such as amine- and amide-containing drugs to form *N*-nitroso compounds in the stomach (Gillatt et al. 1985). *N*-Nitroso compounds have been found to be teratogens in animal models (Nagao et al. 1991; Platzek et al. 1983). These compounds are formed to a greater extent in the presence of a nitrosatable compound if nitrite concentration is high (Choi 1985); and when combined with higher nitrite, nitrosatable compounds have been reported to be more strongly associated with exencephaly and skeletal malformations in mice (Teramoto et al. 1980) and with NTDs (Brender et al. 2004, 2011b) and other types of birth defects in humans (Brender et al. 2012). In a small case–control study of Mexican-American women, nitrosatable drug exposure was more strongly associated with NTDs in offspring of women whose drinking-water nitrate measured ≥ 3.5 mg/L than among births to women

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with lower measured nitrate in their drinking water (Brender et al. 2004).

The objectives of our study were to *a*) examine the relation between prenatal exposure to drinking-water nitrate and birth defects in offspring (selected from defect groups previously associated with higher nitrate in drinking water), accounting for maternal water consumption patterns; and *b*) investigate whether higher daily exposure to drinking-water nitrate or total nitrite that included contributions from diet and drinking water strengthened associations between prenatal exposure to nitrosatable drugs and selected birth defects in offspring.

Methods

Study population and design. To address the study objectives, we used data from the Iowa and Texas sites of the National Birth Defects Prevention Study (NBDPS), an ongoing population-based case–control study of birth defects in the United States (includes sites in 10 states) that began in 1997 (Yoon et al. 2001). The Iowa and Texas sites identify deliveries with major birth defects from live births, stillbirths, and elective terminations as part of their population-based birth defect surveillance. In the NBDPS, case classification is standardized, and clinical information on potentially eligible births is evaluated by a clinical geneticist at each study site and also independently reviewed by one or more other clinical geneticists. For the present study, women with estimated dates of delivery from 1 October 1997 through 31 December 2005 who had deliveries with an NTD, oral cleft, limb deficiency, or congenital heart defect were included. Control infants (live births without any major congenital malformations and whose mothers resided in the study area at delivery) were randomly selected from live birth certificates in Iowa and from hospital delivery records in Texas (proportional to the number of births in each hospital in the geographic regions of study). These comparison infants served as controls for all case groups. The institutional review boards (IRBs) at each NBDPS site and the Centers for Disease Control and Prevention approved the NBDPS study protocol, and the IRBs at the University of Iowa, Texas A&M University, and Texas Department of State Health Services also approved the present project.

Data collection. After providing informed consent, case and control mothers were interviewed in English or Spanish by female interviewers using a computer-assisted telephone interview (Yoon et al. 2001). Mothers were questioned about their use of prescription and over-the-counter medications during the index pregnancy, vitamin supplements taken, diet, beverage consumption, work characteristics, and water use. Residential histories were collected for the period 3 months before conception through pregnancy, including the month/year that the mother started and stopped living in each location. A water module was added to the NBDPS interview in 1999, and questions about personal water use were asked of all mothers beginning in 2000, including sources (private well, unfiltered tap, filtered tap, bottled, other); presence and type of filtration; quantity of water drank at home and at work or school on an average day; and any changes including month/year of change in source or quantity of drinking water consumed. Only women who completed the water module were included in the water nitrate analyses, and their estimated dates of delivery ranged from 1998 through 2005.

Assessment of nitrate in municipal tap water. After maternal residential addresses were geocoded, we used an approach developed by the Water Subcommittee of the NBDPS Environmental/Occupational Work Group to link geocoded addresses to municipal water supplies. This included *a*) linking geocoded maternal addresses to public water utilities that had digitized boundary maps available; *b*) if utility boundary maps were not available, linking maternal addresses to water utilities using census place names (census place city boundaries were identified through linkage of municipal water system names to census place names); and *c*) contacting water utilities to confirm whether they provided water for maternal addresses that could not be matched using the first two approaches.

Under the federal Safe Drinking Water Act (SDWA 1974), public water supplies using groundwater are required to sample annually for nitrate, and surface water utilities are initially required to sample quarterly, then annually. In Iowa, SDWA and other public water supply data are maintained by the Center for Health Effects of Environmental Contamination at the University of Iowa (Iowa City, IA, USA). In Texas, routine monitoring data for drinking-water nitrate were obtained from the Texas Commission on Environmental Quality (Austin, TX, USA); public water suppliers are required by Texas law to report water monitoring results to this state agency.

Water samples taken during the actual dates of residence during 1 month before conception (B1) through the end of the third month of pregnancy (P3) were given the highest priority for inclusion and averaged if more than one sample result was available. If sample results for this period (B1P3) were unavailable, results were selected, in order of priority, as *a*) any results of samples up to 12 months before the start of B1 through 12 months after the end of P3, or *b*) results of samples taken closest to the earliest date of B1 and results closest to the last day of P3. Using the same approach, we also obtained water nitrate estimates for 1 month before through 1 month postconception (B1P1) for analyses involving NTDs to better reflect the critical exposure window for these defects.

Assessment of nitrate in bottled water. Analyses of maternal responses to water use indicated that 341 Iowa and 1,069 Texas mothers (with deliveries having the specified birth defects in this project or control births) reported using bottled water exclusively near the beginning of pregnancy, and a large number of participants in both states reported drinking bottled water in addition to tap water. To estimate exposure to nitrate in bottled water, we conducted a bottled water survey in Iowa and Texas from January through May 2010 in which representative samples of bottled water were collected in major metropolitan and municipal areas that women resided in or nearby. In addition, dispensed waters sold by the gallon were obtained in Iowa stores and in Texas stores, water mills, and kiosks. All samples were tested for nitrate at the State Hygienic Laboratory at The University of Iowa with U.S. Environmental Protection Agency Method 300.0 (Pfaff 1993). Median values were assigned for each city based on multiple bottled water samples collected and respective test results. These median levels were assigned to residents of that city; for cities where bottled water was not collected, the median level of the closest city where water was collected was used.

Estimation of nitrate in private well water. Residential addresses of Texas mothers reporting drinking water from private wells were linked to the relevant aquifers. Nearly one-half of the reported private wells were located in the Ogallala Aquifer, with the other reported wells mainly located in five additional major Texas aquifers, including the Edwards-Trinity, Trinity, Carrizo-Wilcox, Gulf Coast, and Hueco-Mesilla Bolson aquifers. We modeled groundwater flow and nitrate transport in these major aquifers and estimated the temporal dynamics of nitrate level at private well locations during the index pregnancies. The modeling effort for individual wells (based on the hydrogeology and the spatial scale of the aquifers) was done separately using two different models: *a*) MODFLOW-MT3DMS (McDonald and Harbaugh 1988; Zheng and Wang 1988) and *b*) HYDRUS-PHRREQC (HP1) model (Jacques and Šimůnek 2005). The wells in the Ogallala Aquifer were modeled using the MODFLOW-MT3DMS because this aquifer encompassed nearly one-half of the private well users, and spanned a large area, which required large scale modeling. Wells in other aquifers were modeled using the HP1 model because the private well users in these aquifers were either localized (e.g., Hueco-Mesilla

Bolson, Trinity) or located on a scattered aquifer such as the Seymour Aquifer. The Seymour Aquifer is known as a scattered aquifer because it is in separate areas of erosional remnants of the Seymour Formation of Pleistocene age in parts of 20 Texas counties. Each model was run for 4–9 years depending on the case or control dates of B1P3 and was validated using available historical sampling data from wells in the respective areas. Daily nitrate concentrations obtained from the models were averaged for the respective exposure windows of each Texas mother who reported drinking private well water.

Estimation of daily intake of nitrate from drinking water. Nitrate levels in drinking water varied considerably by source. Median levels for bottled water, public water supplies, and private wells (estimated through modeling) were respectively 0.33, 5.0, and 17.6 mg/L as nitrate. For mothers living in more than one residence during the two exposure windows of interest, average nitrate levels from reported drinking-water sources at each residence were obtained and weighted by number of months lived at each address. We developed a program for estimating daily intake of nitrate from drinking water during the exposure windows, using STATA® (Release 11; StataCorp, College Station, TX) that took into account the reported sources of drinking water with respective nitrate concentrations and quantity consumed at home and work, use of water filters and type, consumption of tea and coffee, and any reported changes in water consumption or source during 1 month preconception through the first trimester. We developed two environmental exposure metrics including daily intake of nitrate from drinking water (milligrams) during B1P1 that was used in all analyses of NTDs, and water nitrate intake during B1P3 for analyses of heart, limb, and oral cleft defects. Nitrate intake from drinking-water sources was categorized into tertiles for each exposure period based on the control mothers' distributions. We were able to estimate daily intake of nitrate from these sources for 87% of case mothers and 88% of control mothers who completed the water module of the NBDPS interview. Reasons for nonlinkage included nitrate in drinking water of private well users not estimated (9% of the Iowa cases/controls) and insufficient/missing addresses or an address outside the United States during the exposure windows of interest.

Classification of nitrosatable drugs. In the NBDPS interview, mothers were questioned about prescription and nonprescription drugs used (including start and stop dates) for specific illnesses and disorders and were also prompted for specific products. Methods used to classify drugs with respect to nitrosatability have been described in detail in previous publications (Brender et al. 2011a, 2011b). Briefly, the active ingredients of reported medications used were identified, cross-referenced against previously compiled lists of nitrosatable medicinal compounds (Brambilla and Martelli 2007; McKean-Cowdin et al. 2003), and categorized based on the presence of amine (secondary or tertiary) and amide functional groups in their molecular structures. We focused on exposure to any nitrosatable drugs during the month before and after conception in relation to NTDs and during the first trimester for the other birth defects. Approximately 24% of the control mothers in the NBDPS took one or more nitrosatable drugs during the first trimester (Brender et al. 2011a). The most commonly taken nitrosatable drugs included certain types of antiemetic medications, decongestants, antihistamines, and anti-infectives that contained secondary amines, tertiary amines, or amides as part of their molecular structures.

Estimation of total nitrite exposure. To estimate daily intake of nitrate and nitrite from dietary sources, we used a combination of sources, including *a*) the 58-item food frequency questionnaire (FFQ) that elicited information about dietary intake during the year before pregnancy and that was adapted from the short Willett FFQ (Willett et al. 1985), and *b*) additional detailed questions about consumption of breakfast cereals from 3 months before to the end of pregnancy. Procedures were described in detail in a previous publication (Griesenbeck et al. 2009b); briefly, *a*) weighted means for nitrates and nitrites (milligrams/100 g) were calculated for each food item based on the relevant literature; *b*) the respective means were multiplied by the serving size (grams) assigned to each food; *c*) nitrates and nitrites in each serving size were multiplied by the number of servings by month; and *d*) nitrates and nitrites across all food items were summed and then divided by 30 to obtain daily intake of dietary nitrate and nitrite (milligrams). Using the formula suggested by Choi (1985), we estimated total nitrite exposure from food and water as the sum of dietary nitrite intake and 5% of estimated nitrate intake from diet and water sources. Total nitrite intake was further categorized into tertiles based on the control mothers' distributions. In this population, median contributions of food and drinking-water nitrate to daily intake of nitrate were 94% and 6%, respectively. Approximately 97% and 3% of total nitrite exposure was from food and drinking water, respectively.

Statistical analysis. To account for correlation of nitrate intake by geographic location, mixed-effects (random-effects) models for logistic regression were used with mothers nested within cities of residence (nearest city, if rural address) (Goldstein 2010). Mothers in the lowest tertile of nitrate intake from drinking water during B1P1 for analyses of NTDs and B1P3 for the other birth defects served as the referent categories. For limb deficiencies, oral cleft defects, and congenital heart defects, we restricted analyses to isolated birth defects. Covariables were selected *a priori* and based on the literature, and only those cases and controls for which complete data on all pertinent covariables in each analysis were included. For NTDs, covariables included maternal race/ethnicity, education, study site, and any folic acid supplementation during B1P1. In addition to maternal race/ethnicity, education, and study site, covariables for analyses of oral clefts also included maternal age, any smoking 1 month before conception through the first trimester, and folic acid supplementation during the first trimester. Covariables for analyses of limb deficiencies included maternal race/ethnicity, education, age, study site, and multivitamin supplementation during the first trimester. For heart defects, maternal race/ethnicity, education, smoking, study site, and multivitamin supplementation during the first trimester were incorporated into the logistic models. The associations between tertile of prenatal nitrate intake from drinking water and birth defects in offspring were assessed for linear trend by treating the three levels of nitrate intake as a continuous variable in the logistic model and testing the significance of linearity with the z-test in STATA® (equivalent to the Wald chi-square test).

As part of a sensitivity analysis, we repeated the above analyses for the subset of participants who reported drinking only municipal tap water during the period around conception and the first trimester. We also examined the association between measured nitrate (milligrams per liter) in municipal water and selected birth defects for which we used the cut points reported by Croen et al. (2001) and Dorsch et al. (1984) (< 5 mg/L, 5–15 mg/L, and > 15 mg/L).

Nitrosatable drug exposure (any vs. none) during B1P1 and the first trimester was stratified by tertiles of nitrate intake from drinking water and by total nitrite from food and water sources. In analyses involving total nitrite, we excluded women with daily caloric intakes of < 500 or > 5,000 kcal, and also adjusted the odds ratios (ORs) for total energy intake (kilocalories per day). We tested for departure from additivity (biologic interaction) in these associations using a statistical program developed by Andersson et al. (2005) that was adapted for STATA®. This program calculated the relative excess risk due to interaction (RERI) and attributable proportion due to interaction (AP) (and their respective 95% CIs). Departures from additive effects were considered present if the confidence intervals of either measure excluded zero. To

assess multiplicative interaction, the product terms of any nitrosatable drug use with water nitrate and total nitrite intake were included in the logistic models, and multiplicative interaction was considered present if the p-value associated with the interaction term was < 0.05.

Results

Maternal interviews for offspring with estimated dates of delivery from 1997 through 2005 numbered 317 with NTDs, 177 with limb deficiencies, 654 with oral cleft defects, 2,011 with congenital heart defects, and 1,551 unaffected live births. Maternal participation rates for births with NTDs, limb deficiencies, oral clefts, congenital heart defects, and controls were, respectively, 66%, 72%, 74%, 62%, and 64%. Median time from estimated date of delivery to maternal interview ranged from 9 months for control mothers to 13 months for women with NTD-affected pregnancies. Table 1 shows the characteristics of the case and control mothers. Among participants who completed the water module questions, the proportions of control mothers and mothers of babies with heart defects were similar with respect to usual home sources of drinking water. In contrast, mothers of babies with NTDs, limb deficiencies, and oral clefts were more likely than control mothers to report drinking municipal tap water.

Table 1. Selected characteristics of Iowa and Texas case mothers and control mothers in the National Birth Defects Prevention Study, 1997–2005 [*n* (%)].

Characteristic	Controls (*n* = 1,551)	Cases			
		NTDs (*n* = 317)	Limb deficiencies (*n* = 177)	Oral cleft defects (*n* = 654)	Heart defects (*n* = 2,011)
Race/ethnicity					
Non-Hispanic white	901 (58.2)	165 (52.2)	93 (52.5)	393 (60.2)	1,033 (51.5)
Non-Hispanic black	27 (1.7)	9 (2.9)	5 (2.8)	12 (1.8)	60 (3.0)
Hispanic	555 (35.9)	132 (41.8)	67 (37.9)	218 (33.4)	833 (41.5)
Asian/Pacific Islander	21 (1.4)	1 (0.3)	2 (1.1)	12 (1.8)	19 (0.9)
All others	44 (2.8)	9 (2.8)	10 (5.7)	18 (2.8)	62 (3.1)
Missing	3	1	0	1	4
Education (years)					
< 12	286 (18.8)	64 (20.3)	27 (15.6)	138 (21.3)	408 (20.6)
12	443 (29.2)	87 (27.6)	57 (33.0)	192 (29.7)	574 (29.0)
13–15	436 (28.7)	105 (33.3)	57 (32.9)	186 (28.7)	606 (30.6)
> 15	353 (23.3)	59 (18.7)	32 (18.5)	131 (20.2)	390 (19.7)
Missing	33	2	4	7	33
Age at delivery (years)					
< 18	95 (6.1)	11 (3.5)	7 (4.0)	29 (4.4)	98 (4.9)
18–19	130 (8.4)	29 (9.1)	19 (10.7)	61 (9.3)	159 (7.9)
20–24	380 (24.5)	79 (24.9)	48 (27.1)	208 (31.8)	535 (26.6)
25–29	453 (29.2)	100 (31.5)	55 (31.1)	170 (26.0)	551 (27.4)
30–34	344 (22.2)	68 (21.5)	35 (19.8)	114 (17.4)	446 (22.2)
> 34	149 (9.6)	30 (9.5)	13 (7.3)	72 (11.0)	222 (11.0)
Study center					
Iowa	759 (48.9)	146 (46.1)	80 (45.2)	306 (46.8)	769 (38.2)
Texas	792 (51.1)	171 (53.9)	97 (54.8)	348 (53.2)	1,242 (61.8)
Smoking[a]					
No	1,199 (78.7)	259 (82.2)	132 (76.3)	471 (72.6)	1,548 (78.1)
Yes	324 (21.3)	56 (17.8)	41 (23.7)	178 (27.4)	433 (21.9)
Missing/out of range	28	2	4	5	30
Nitrosatable drug exposure[b]					
No	1,166 (77.6)	216 (70.8)	120 (71.9)	482 (76.4)	1,475 (76.2)
Yes	336 (22.4)	89 (29.2)	47 (28.1)	149 (23.6)	460 (23.8)
Total daily nitrite intake[c]					
≤ 4.78 mg/day	726 (66.1)	145 (62.5)	72 (55.8)	334 (68.2)	1,004 (63.5)
> 4.78 mg/day	372 (33.9)	87 (37.5)	57 (44.2)	156 (31.8)	578 (36.5)
Multivitamin use[d]					
No	206 (13.6)	33 (10.6)	22 (12.9)	100 (15.7)	304 (15.5)
Yes	1,308 (86.4)	277 (89.4)	148 (87.1)	537 (84.3)	1,658 (84.5)
Missing	37	7	7	17	49
Usual home source of drinking water[e]					
Tap water, municipal	738 (58.3)	173 (64.3)	96 (64.0)	354 (61.7)	1,011 (56.3)
Tap water, private well	72 (5.7)	19 (7.1)	14 (9.3)	42 (7.3)	99 (5.5)
Bottled water exclusively	455 (36.0)	77 (28.6)	40 (26.7)	178 (31.0)	685 (38.2)
Not available[f]	286	48	27	80	216

[a]Any smoking between date of conception and end of first trimester. [b]Exposure during the first trimester of pregnancy. [c]Total daily nitrite intake = 5% (drinking water nitrate + dietary nitrate) + dietary nitrite. [d]Use during the first trimester of pregnancy. [e]Reported primary drinking water source at the beginning of pregnancy. [f]Water module questions were added in 1999.

Numbers of births with complete information for maternal daily nitrate intake from water sources and other covariables were 227, 94, 415, 1,046, and 1,105, respectively, for all NTDs, isolated limb deficiencies, oral cleft defects, congenital heart defects, and controls. Adjusting for maternal race/ethnicity, education, study site, and folic acid supplementation, maternal nitrate intake of ≥ 5 mg per day from drinking water was associated with NTD-affected pregnancies [adjusted odds ratio (aOR) 1.43; 95% CI: 1.01, 2.04], although this association appeared to be specific to spina bifida (Table 2). Mothers of babies with spina bifida were 1.4 times more likely (95% CI: 0.86, 2.32) than control mothers to ingest between 0.91 and 4.9 mg nitrate per day and 2 times more likely (95% CI: 1.27, 3.22) to ingest ≥ 5 mg nitrate from drinking water around conception (p for trend = 0.003). During B1P3, mothers of babies with isolated limb deficiencies, cleft palate, and cleft lip without cleft palate were, respectively, 1.8 (95% CI: 1.05, 3.08), 1.9 (95% CI: 1.17, 3.09), and 1.8 times (95% CI: 1.08, 3.07) more likely than control mothers to ingest > 5.41 mg per day of nitrate from drinking water. We noted significant linear trends ($p < 0.05$) in the associations between maternal water nitrate and these defects in offspring (Table 2). In contrast, we saw minimal or no associations between maternal nitrate intake from drinking water and congenital heart defects in offspring. Restriction of analyses to women who reported drinking only tap water from municipal water supplies did not materially change the aORs associated with the highest tertile of water intake for spina bifida (aOR = 1.93; 95% CI: 0.99, 3.76), cleft lip without cleft palate (aOR = 1.96; 95% CI: 0.88, 4.36), or cleft palate (aOR = 1.55; 95% CI: 0.78, 3.10), but the aOR for any limb deficiency increased to 3.19 (95% CI: 1.09, 9.35) (see Supplemental Material, Table S1). A significant linear trend was observed for only cleft lip in relation to measured nitrate in drinking water among offspring of women who reported drinking municipal water (see Supplemental Material, Table S2). An aOR of 2.31 (95% CI: 1.20, 4.47) was noted for this defect among offspring of women who consumed water with nitrate levels > 15 mg/L relative to women who drank water with nitrate levels < 5 mg/L.

No specific patterns of stronger associations between nitrosatable drug exposure (any versus none) and birth defects among women with higher daily intake of nitrate from drinking water were evident when aORs were stratified according to tertile of daily nitrate intake from drinking water (see Supplemental Material, Table S3). For several birth defect groups, the strongest associations with

nitrosatable drug exposure were estimated for women in the lowest tertiles of estimated nitrate intake from drinking water [e.g., aORs = 2.54 (95% CI: 1.20, 5.37) and 2.89 (95% CI: 1.15, 7.25) for NTDs and cleft palate, respectively]. The CIs for the RERI and AP included 0, indicating no significant departures from additivity, and the *p*-values for the interaction terms for water and nitrosatable drug exposure were > 0.05, indicating no significant departures from multiplicative effects.

On the other hand, when estimated nitrate from drinking water and diet were combined with dietary nitrite intake to estimate total nitrite exposure from these sources, the strongest associations between nitrosatable drug exposure and several birth defects were observed among women with the highest estimated total nitrite exposure (the lower two tertiles of intake combined because of similarity of ORs) (see Supplemental Material, Table S4). Associations between nitrosatable drug exposure and birth defects were stronger in the highest tertile of total nitrite (vs. the lower two tertiles combined) for NTDs (aOR = 1.76; 95% CI: 0.90, 3.43 vs. aOR = 1.41; 95% CI: 0.87, 2.29), cleft lip without cleft palate (aOR = 2.01; 95% CI: 0.90, 4.48 vs. aOR = 0.80; 95% CI: 0.42, 1.52), cleft palate (aOR = 2.51; 95% CI: 1.24, 5.06 vs. aOR = 0.95; 95% CI: 0.55, 1.64), limb deficiencies (aOR = 1.64; 95% CI: 0.80, 3.35 vs. aOR = 1.00; 95% CI: 0.53, 1.89), atrioventricular septal defects (aOR = 5.10; 95% CI: 1.40, 18.6 vs. aOR = 1.93; 95% CI: 0.76, 4.87), and single ventricle (aOR = 3.25; 95% CI: 1.13, 9.31 vs. aOR = 0.74; 95% CI: 0.27, 2.02). Significant departures from additivity were noted for the joint estimated effects of total nitrite intake and nitrosatable drug exposures for cleft lip, cleft palate, limb deficiencies, and single ventricle; multiplicative interaction was also present in this association with cleft palate (see Supplemental Material, Table S4).

Discussion

Results from this large population-based case–control study suggest that prenatal nitrate intake from drinking water is associated with NTDs, oral cleft defects, and limb deficiencies in offspring. Previous publications that have reported significant associations between drinking-water nitrates and birth defects hypothesized that nitrate might act as a teratogen through its contribution to the endogenous formation of *N*-nitroso compounds (Croen et al. 2001; Dorsch et al. 1984). In the present study, however, higher daily intake of nitrate from drinking water did not strengthen associations between nitrosatable drugs and the various birth defects examined. On the other hand, associations between nitrosatable drugs and birth defects were stronger among women in the highest tertile of estimated total nitrite intake, a measure based on intake of dietary nitrite and nitrate from diet and drinking water. In this study, nitrate levels in the drinking water tended to be low, with a median contribution of nitrate per day from this source of 6% in the study population. In a recent review, the World Health Organization (2011) noted that the contribution of drinking water to nitrate intake is usually < 14%.

Previous studies have assigned exposure based on measured nitrate in drinking water instead of estimating daily ingestion. For

Table 2. Maternal daily nitrate intake from drinking water and selected birth defects in offspring.

Birth defect	Daily nitrate intake from water (mg/day)[a]	Cases [*n* (%)]	Controls [*n* (%)]	Unadjusted OR (95% CI)[b]	Adjusted OR (95% CI)[b]	*p*-Value for linear trend
Any NTD[c]	< 0.91	67 (29.5)	367 (33.3)	1.00	1.00	0.038
	0.91–4.9	65 (28.6)	360 (32.7)	0.99 (0.68, 1.43)	1.00 (0.68, 1.45)	
	≥ 5.0	95 (41.9)	374 (34.0)	1.39 (0.99, 1.96)	1.43 (1.01, 2.04)	
Spina bifida[c]	< 0.91	30 (22.4)	367 (33.3)	1.00	1.00	0.003
	0.91–4.9	42 (31.3)	360 (32.7)	1.43 (0.87, 2.33)	1.41 (0.86, 2.32)	
	≥ 5.0	62 (46.3)	374 (34.0)	2.03 (1.28, 3.21)	2.02 (1.27, 3.22)	
Anencephaly[c]	< 0.91	31 (43.7)	367 (33.3)	1.00	1.00	0.348
	0.91–4.9	17 (23.9)	360 (32.7)	0.56 (0.30, 1.03)	0.58 (0.32, 1.08)	
	≥ 5.0	23 (32.4)	374 (34.0)	0.73 (0.42, 1.27)	0.78 (0.44, 1.37)	
Any limb deficiency[d,e]	< 1.0	23 (24.5)	370 (33.5)	1.00	1.00	0.028
	1.0–5.41	29 (30.9)	367 (33.2)	1.27 (0.72, 2.24)	1.17 (0.66, 2.07)	
	≥ 5.42	42 (44.7)	368 (33.3)	1.84 (1.08, 3.11)	1.79 (1.05, 3.08)	
Any oral cleft defect[e,f]	< 1.0	122 (29.4)	370 (33.5)	1.00	1.00	0.007
	1.0–5.41	120 (28.9)	366 (33.2)	0.99 (0.74, 1.33)	0.98 (0.73, 1.32)	
	≥ 5.42	173 (41.7)	367 (33.3)	1.43 (1.09, 1.88)	1.45 (1.10, 1.92)	
Cleft lip without cleft palate[e,f]	< 1.0	24 (24.0)	370 (33.5)	1.00	1.00	0.019
	1.0–5.41	29 (29.0)	366 (33.2)	1.22 (0.70, 2.14)	1.13 (0.64, 1.99)	
	≥ 5.42	47 (47.0)	367 (33.3)	1.97 (1.18, 3.30)	1.82 (1.08, 3.07)	
Cleft palate[e,f]	< 1.0	29 (25.2)	370 (33.5)	1.00	1.00	0.007
	1.0–5.41	32 (27.8)	366 (33.2)	1.12 (0.66, 1.88)	1.12 (0.66, 1.90)	
	≥ 5.42	54 (47.0)	367 (33.3)	1.88 (1.17, 3.01)	1.90 (1.17, 3.09)	
Conotruncal heart defects[e,g]	< 1.0	58 (35.4)	370 (33.5)	1.00	1.00	0.403
	1.0–5.41	41 (25.0)	367 (33.2)	0.71 (0.47, 1.09)	0.72 (0.47, 1.11)	
	≥ 5.42	65 (39.6)	368 (33.3)	1.13 (0.77, 1.65)	1.18 (0.80, 1.74)	
Right ventricular outflow tract obstruction heart defects[e,g]	< 1.0	36 (30.0)	370 (33.5)	1.00	1.00	0.083
	1.0–5.41	31 (25.8)	367 (33.2)	0.87 (0.53, 1.43)	0.89 (0.54, 1.48)	
	≥ 5.42	53 (44.2)	368 (33.3)	1.48 (0.95, 2.32)	1.47 (0.93, 2.33)	
Left ventricular outflow tract obstruction heart defects[e,g]	< 1.0	44 (28.2)	370 (33.5)	1.00	1.00	0.522
	1.0–5.41	58 (37.2)	367 (33.2)	1.33 (0.88, 2.02)	1.31 (0.86, 2.00)	
	≥ 5.42	54 (34.6)	368 (33.3)	1.23 (0.81, 1.88)	1.16 (0.75, 1.78)	
Septal heart defects[e,g]	< 1.0	203 (35.8)	370 (33.5)	1.00	1.00	0.853
	1.0–5.41	210 (37.0)	367 (33.2)	1.04 (0.82, 1.33)	0.92 (0.69, 1.22)	
	≥ 5.42	154 (27.2)	368 (33.3)	0.76 (0.59, 0.98)	0.98 (0.71, 1.34)	

[a]For NTDs, water nitrate intake 1 month preconception to 1 month postconception was estimated. For limb, oral cleft, and congenital heart defects, water nitrate intake 1 month preconception through the first trimester was estimated. [b]Crude and adjusted ORs include only cases and controls with complete information for covariates. [c]Adjusted for maternal race/ethnicity, education, study center, and folic acid supplementation. [d]Adjusted for maternal race/ethnicity, education, age, multivitamin supplementation, and study center. [e]Isolated defect. [f]Adjusted for maternal race/ethnicity, education, age, folic acid supplementation, smoking, and study center. [g]Adjusted for maternal race/ethnicity, education, multivitamin supplementation, smoking, and study center.

women who drank water from groundwater sources, measured levels of total nitrate as low as 5–15 mg/L have been significantly associated with birth defects (Dorsch et al. 1984) including anencephaly (Croen et al. 2001). Although we noted significant ORs in the relation between measured nitrate levels at ≥ 5 mg/L and several birth defects, we saw a significant linear trend only for cleft lip without cleft palate in our study population. Other studies have reported elevated, but not statistically significant, ORs for central nervous system defects (Arbuckle et al. 1988) and NTDs (Brender et al. 2004) for measured nitrate levels respectively at 26 mg/L (relative to 0.1 mg/L) and ≥ 3.5 mg/L (relative to < 3.5 mg/L). Positive associations were restricted to groundwater drinkers in several of these studies, and the authors suggested that other agents correlated with nitrate in groundwater might be responsible for the associations noted (Croen et al. 2001; Dorsch et al. 1984).

In contrast to findings from a study of nitrosatable drugs and NTDs in Mexican Americans (Brender et al. 2004), in the present study, higher intake of nitrate from drinking water did not strengthen the association between nitrosatable drug use and NTDs, nor was this pattern noted for the other birth defects examined. In two earlier studies (Brender et al. 2011b, 2012) of NBDPS, which included participants from all 10 sites, associations between prenatal nitrosatable drug exposure and several birth defects, including NTDs, cleft palate, conotruncal heart defects, atrioventricular septal defects, and single ventricle defects were stronger among women with the highest estimated intake of nitrite from dietary sources than in women with lower estimated dietary intakes. Similarly in the present study, associations between nitrosatable drug use and several of the same defects were stronger with higher estimated total nitrite intake, which included intake from drinking-water as well as dietary sources. Water nitrate contributed, on average (median), approximately 3% of total daily nitrite in the present study population. Therefore, water nitrate might be associated with birth defects for reasons other than its contribution to the endogenous formation of *N*-nitroso compounds. Nitrate has been found to occur with other contaminants in drinking water, especially in conjunction with pesticides, arsenic and other trace metals, and water disinfection by-products (Toccalino et al. 2012).

In the present study, we focused on nitrate contamination in drinking-water sources without examining the presence of other water contaminants. Another study limitation was the potential for measurement errors in nitrate content of drinking-water sources and daily consumption of water nitrate. Estimates of nitrate in sources from public water systems were based on data from routine monitoring in which we linked addresses to the most time-relevant sample results available. Our approach for assigning nitrate levels to municipal drinking-water sources was not validated, although we developed and followed a detailed set of standard operating procedures for such assignment (Griesenbeck et al. 2009a). The high percentage of bottled water users presented a challenge in exposure assessment because participants were not specifically questioned about types of bottled water consumed. Therefore, nitrate content from this source was estimated from nitrate measured in bottled water samples from neighborhood grocery store surveys. However, associations noted between nitrate intake from drinking-water nitrate and birth defects changed very little when the analysis was restricted to women who reported drinking tap water from municipal water supplies only. We estimated nitrate content in private wells through complex models that took into account local conditions; however, this modeling effort was restricted to private well users in Texas. Although it is possible that some participants might have not accurately recalled the types and amounts of water that they consumed during early pregnancy, Shimokura et al. (1998) found good agreement (Pearson's $r = 0.78$) between a questionnaire on past use and a 3-day water diary for drinking-water intake in a sample of pregnant women. Given that all exposure assessments in this study of drinking-water nitrate were completed with the study teams blinded to case–control status, misclassification of daily nitrate intake from drinking water would most likely be nondifferential and have led to an underestimation of the true ORs. Measurement error might have also occurred with the estimation of dietary intake of nitrate and nitrite, and this limitation is discussed in detail in previous publications (Brender et al. 2011a, 2012) along with the potential for bias in participant recall of drugs taken during early pregnancy.

Conclusion

In this large, population-based case–control study, women who had babies with NTDs, limb deficiencies, and oral cleft defects were significantly more likely than control mothers to ingest ≥ 5 mg of nitrate per day from drinking water. However, study findings suggest that endogenous formation of *N*-nitroso compounds might not be the underlying mechanism for potential teratogenesis with this water contaminant, because higher intake of nitrate from drinking water did not strengthen associations between prenatal nitrosatable drug exposure and birth defects in offspring. Given that nitrate contamination occurs in conjunction with other water contaminants, future studies of birth defects might focus on prenatal exposure to mixtures of contaminants in drinking water.

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Review

Drinking Water Nitrate and Human Health: An Updated Review

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Abstract: Nitrate levels in our water resources have increased in many areas of the world largely due to applications of inorganic fertilizer and animal manure in agricultural areas. The regulatory limit for nitrate in public drinking water supplies was set to protect against infant methemoglobinemia, but other health effects were not considered. Risk of specific cancers and birth defects may be increased when nitrate is ingested under conditions that increase formation of *N*-nitroso compounds. We previously reviewed epidemiologic studies before 2005 of nitrate intake from drinking water and cancer, adverse reproductive outcomes and other health effects. Since that review, more than 30 epidemiologic studies have evaluated drinking water nitrate and these outcomes. The most common endpoints studied were colorectal cancer, bladder, and breast cancer (three studies each), and thyroid disease (four studies). Considering all studies, the strongest evidence for a relationship between drinking water nitrate ingestion and adverse health outcomes (besides methemoglobinemia) is for colorectal cancer, thyroid disease, and neural tube defects. Many studies observed increased risk with ingestion of water nitrate levels that were below regulatory limits. Future studies of these and other health outcomes should include improved exposure assessment and accurate characterization of individual factors that affect endogenous nitrosation.

Keywords: drinking water; nitrate; cancer; adverse reproductive outcomes; methemoglobinemia; thyroid disease; endogenous nitrosation; *N*-nitroso compounds

1. Introduction

Since the mid-1920s, humans have doubled the natural rate at which nitrogen is deposited onto land through the production and application of nitrogen fertilizers (inorganic and manure),

the combustion of fossil fuels, and replacement of natural vegetation with nitrogen-fixing crops such as soybeans [1,2]. The major anthropogenic source of nitrogen in the environment is nitrogen fertilizer, the application of which increased exponentially after the development of the Haber–Bosch process in the 1920s. Most synthetic fertilizer applications to agricultural land occurred after 1980 [3]. Since approximately half of all applied nitrogen drains from agricultural fields to contaminate surface and groundwater, nitrate concentrations in our water resources have also increased [1].

The maximum contaminant level (MCL) for nitrate in public drinking water supplies in the United States (U.S.) is 10 mg/L as nitrate-nitrogen (NO_3-N). This concentration is approximately equivalent to the World Health Organization (WHO) guideline of 50 mg/L as NO_3 or 11.3 mg/L NO_3-N (multiply NO_3 mg/L by 0.2258). The MCL was set to protect against infant methemoglobinemia; however other health effects including cancer and adverse reproductive outcomes were not considered [4]. Through endogenous nitrosation, nitrate is a precursor in the formation of *N*-nitroso compounds (NOC); most NOC are carcinogens and teratogens. Thus, exposure to NOC formed after ingestion of nitrate from drinking water and dietary sources may result in cancer, birth defects, or other adverse health effects. Nitrate is found in many foods, with the highest levels occurring in some green leafy and root vegetables [5,6]. Average daily intakes from food are in the range of 30–130 mg/day as NO_3 (7–29 mg/day NO_3-N) [5]. Because NOC formation is inhibited by ascorbic acid, polyphenols, and other compounds present at high levels in most vegetables, dietary nitrate intake may not result in substantial endogenous NOC formation [5,7].

Studies of health effects related to nitrate exposure from drinking water were previously reviewed through early 2004 [8]. Further, an International Agency for Research on Cancer (IARC) Working Group reviewed human, animal, and mechanistic studies of cancer through mid-2006 and concluded that ingested nitrate and nitrite, under conditions that result in endogenous nitrosation, are probably carcinogenic [5]. Here, our objective is to provide updated information on human exposure and to review mechanistic and health effects studies since 2004. We summarize how the additional studies contribute to the overall evidence for health effects and we discuss what future research may be most informative.

2. Drinking Water Nitrate Exposures in the United States and Europe

Approximately 45 million people in the U.S. (about 14% of the population) had self-supplied water at their residence in 2010 [9]. Almost all (98%) were private wells, which are not regulated by the U.S. Environmental Protection Agency (EPA). The rest of the population was served by public water supplies, which use groundwater, surface water, or both. The U.S. Geological Survey's National Water Quality Assessment (USGS-NAWQA) Project [10] sampled principal groundwater aquifers used as U.S. public and private drinking water supplies in 1988–2015. Nitrate levels in groundwater under agricultural land were about three times the national background level of 1 mg/L NO_3-N (Figure 1) [11]. The mixed land use category mostly had nitrate concentrations below background levels reflecting levels in deeper private and public water supply wells. Based on the NAWQA study, it was estimated that 2% of public-supply wells and 6% of private wells exceeded the MCL; whereas, in agricultural areas, 21% of private wells exceeded the MCL [10]. The USGS-NAWQA study also revealed significant decadal-scale changes in groundwater nitrate concentrations among wells sampled first in 1988–2000 and again in 2001–2010 for agricultural, urban, and mixed land uses [12]. More sampling networks had increases in median nitrate concentration than had decreases.

A study of U.S. public water supplies (PWS) using data from EPA's Safe Drinking Water Information System estimated that the percentage of PWS violating the MCL increased from 0.28 to 0.42% during 1994–2009; most increases were for small to medium PWS (<10,000 population served) using groundwater [13]. As a result of increasing nitrate levels, some PWS have incurred expensive upgrades to their treatment systems to comply with the regulatory level [14–16].

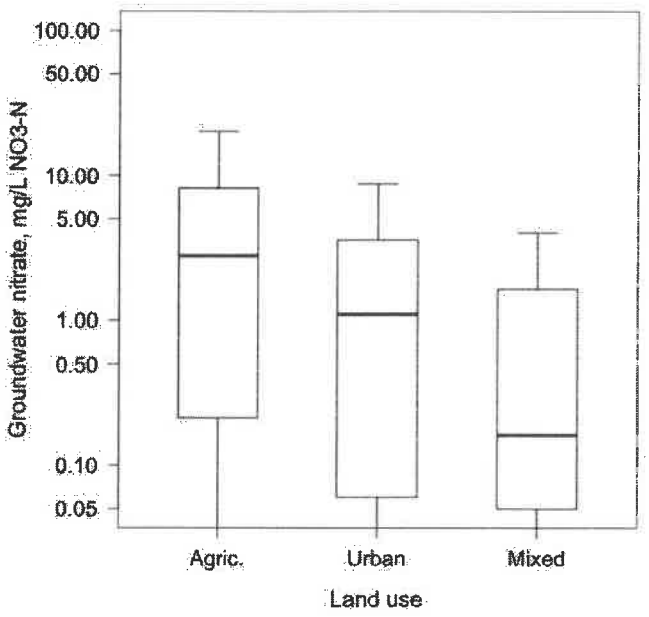

Figure 1. Boxplots of nitrate concentrations in shallow groundwater beneath agricultural and urban land uses, and at depths of private and public drinking water supplies beneath mixed land use. The number of sampled wells were 1573 (agricultural land), 1054 (urban), and 3417 (mixed). The agricultural and urban wells were sampled to assess land use effects, whereas the mixed category wells were sampled at depths of private and public supplies. Median depths of wells in the agricultural, urban, and mixed categories were 34, 32, and 200 feet, respectively. The height of the upper bar is 1.5 times the length of the box, and the lower bound was truncated at the nitrate detection limit of 0.05 mg/L NO_3-N.

In Europe, the Nitrates Directive was set in 1991 [17,18] to reduce or prevent nitrate pollution from agriculture. Areas most affected by nitrate pollution are designated as 'nitrate vulnerable zones' and are subject to mandatory Codes of Good Agricultural Practice [18]. The results of compliance with this directive have been reflected in the time trends of nitrate in some countries. For example, nitrate levels in groundwater in Denmark increased in 1950–1980 and decreased since the 1990s [19]. Average nitrate levels in groundwater in most other European countries have been stable at around 17.5 mg/L NO_3 (4 mg/L NO_3-N) across Europe over a 20-year period (1992–2012), with some differences between countries both in trends and concentrations. Average concentrations are lowest in Finland (around 1 mg/L NO_3 in 1992–2012) and highest in Malta (58.1 mg/L in 2000–2012) [20]. Average annual nitrate concentrations at river monitoring stations in Europe showed a steady decline from 2.7 NO_3-N in 1992 to 2.1 mg/L in 2012 [20], with the lowest average levels in Norway (0.2 mg/L NO_3-N in 2012) and highest in Greece (6.6 mg/L NO_3-N in 2012).

Levels in finished public drinking water have been published only for a few European countries. Trends of nitrate in drinking water supplies from 1976 to 2012 in Denmark showed a decline in public supplies but not in private wells [21]. In Spain, median concentrations were 3.5 mg/L NO_3 (range: 0.4−66.8) in 108 municipalities in 2012 [22], and 4.2 mg/L (range: <1−29) in 11 provinces in 2010 [23]. Levels in other countries included a median of 0.18 mg/L (range: <0.02−7.9) in Iceland in 2001−2012 [24], a mean of 16.1 mg/L (range: 0.05−296 mg/L) in Sicily, Italy in 2004−2005 [25] and a range from undetected to 63.3 mg/L in Deux-Sèvres, France in in 2005−2009 [26].

Nitrate levels in bottled water have been measured in a few areas of the EU and the U.S. and have been found to be below the MCL. In Sicily, the mean level was 15.2 mg/L NO_3(range: 1.2−31.8 mg/L) in 16 brands [25] and in Spain, the median level was 5.2 mg/L NO_3 (range: <1.0−29.0 mg/L) in 9 brands [23]. In the U.S., a survey of bottle water sold in 42 Iowa and 32 Texas communities found

varying but generally low nitrate levels. Nitrate concentrations ranged from below the limit of detection (0.1 mg/L NO_3-N) to 4.9 mg/L NO_3-N for U.S. domestic spring water purchased in Texas.

There are few published studies of nitrate concentrations in drinking water outside the U.S. and Europe. Nitrate concentrations in groundwater were reported for Morocco, Niger, Nigeria, Senegal, India-Pakistan, Japan, Lebanon, Philippines and Turkey with maximum levels in Senegal (median 42.9 mg/L NO_3-N) [5]. In India, nitrate in drinking water supplies is particularly high in rural areas, where average levels have been reported to be 45.7 mg/L NO_3 [27,28] and 66.6 mg/L NO_3 [28]; maximum levels in drinking water exceeded 100 mg/L NO_3 in several regions [27,29]. Extremely high levels of nitrate have been reported in The Gaza Strip, where nitrate reached concentrations of 500 mg/L NO_3 in some areas, and more than 50% of public-supply wells had nitrate concentrations above 45 mg/L NO_3 [30].

3. Exposure Assessment in Epidemiologic Studies

With the implementation of the Safe Drinking Water Act in 1974, more than 40 years of monitoring data for public water supplies in the U.S. provide a framework of measurements to support exposure assessments. Historical data for Europe are more limited, but a quadrennial nitrate reporting requirement was implemented as part of the EU Nitrates Directive [17,18]. In the U.S., the frequency of sampling for nitrate in community water systems is stipulated by their sources (ground versus surface waters) and whether concentrations are below the MCL, and historically, by the size of the population served and vulnerability to nitrate contamination. Therefore, the exposure assessment for study participants who report using a public drinking water source may be based on a variable number of measurements, raising concerns about exposure misclassification. In a study of bladder cancer risk in Iowa, associations were stronger in sensitivity analyses based on more comprehensive measurement data [31]. Other studies have restricted analyses to subgroups with more complete or recent measurements [32–35], with implications for study power and possible selection biases. Sampling frequency also limits the extent to which temporal variation in exposure can be represented within a study population, such as the monthly or trimester-based estimates of exposure most relevant for etiologic investigations of adverse reproductive outcomes. In Denmark, limited seasonal variation in nitrate monitoring data suggested these data would sufficiently capture temporal variation for long-term exposure estimates [36]. Studies have often combined regulatory measurements with questionnaire and ancillary data to better characterize individual variation in nitrate exposure, such as to capture changes in water supply characteristics over time or a participant's duration at a drinking water source [31,33,37,38]. Most case-control studies of drinking water nitrate and cancer obtained lifetime residence and drinking water source histories, whereas cohort studies typically have collected only the current water source. Many studies lacked information about study participants' water consumption, which may be an important determinant of exposure to drinking water contaminants [39].

Due to sparse measurement data, exposures for individuals served by private wells are more difficult to estimate than exposures for those on public water supplies. However, advances in geographic-based modeling efforts that incorporate available measurements, nitrogen inputs, aquifer characteristics, and other data hold promise for this purpose. These models include predictor variables describing land use, nitrogen inputs (fertilizer applications, animal feeding operations), soils, geology, climate, management practices, and other factors at the scale of interest. Nolan and Hitt [40] and Messier et al. [41] used nonlinear regression models with terms representing nitrogen inputs at the land surface, transport in soils and groundwater, and nitrate removal by processes such as denitrification, to predict groundwater nitrate concentration at the national scale and for North Carolina, respectively. Predictor variables in the models included N fertilizer and manure, agricultural or forested land use, soils, and, in Nolan and Hitt [40], water-use practices and major geology. Nolan and Hitt [40] reported a training R^2 values of 0.77 for a model of groundwater used mainly for private supplies and Messier, Kane, Bolich and Serre [41] reported a cross-validation testing R^2 value of 0.33 for a point-level

private well model. These and earlier regression approaches for groundwater nitrate [42–46] relied on predictor variables describing surficial soils and activities at the land surface, because conditions at depth in the aquifer typically are unknown. Redox conditions in the aquifer and the time since water entered the subsurface (i.e., groundwater age) are two of the most important factors affecting groundwater nitrate, but redox constituents typically are not analyzed, and age is difficult to measure. Even if a well has sufficient data to estimate these conditions, the data must be available for all wells in order to predict water quality in unsampled areas. In most of the above studies, well depth was used as a proxy for age and redox and set to average private or public-supply well depth for prediction.

Recent advances in groundwater nitrate exposure modeling have involved machine-learning methods such as random forest (RF) and boosted regression trees (BRT), along with improved characterization of aquifer conditions at the depth of the well screen (the perforated portion of the well where groundwater intake occurs). Tree-based models do not require data transformation, can fit nonlinear relations, and automatically incorporate interactions among predictors [47]. Wheeler et al. [48] used RF to estimate private well nitrate levels in Iowa. In addition to land use and soil variables, predictor variables included aquifer characteristics at the depth of the well screen, such as total thickness of fine-grained glacial deposits above the well screen, average and minimum thicknesses of glacial deposits near sampled wells, and horizontal and vertical hydraulic conductivities near the wells. Well depth, landscape features, nitrogen sources, and aquifer characteristics ranked highly in the final model, which explained 77% and 38% of the variation in training and hold-out nitrate data, respectively.

Ransom et al. [49] used BRT to predict nitrate concentration at the depths of private and public-supply wells for the Central Valley, California. The model used as input estimates of groundwater age at the depth of the well screen (from MODFLOW/MODPATH models) and depth-related reducing conditions in the groundwater. These estimates were generated by separate models and were available throughout the aquifer. Other MODFLOW-based predictor variables comprised depth to groundwater, and vertical water fluxes and the percent coarse material in the uppermost part of the aquifer where groundwater flow was simulated by MODFLOW. Redox variables were top-ranked in the final BRT model, which also included land use-based N leaching flux, precipitation, soil characteristics, and the MODFLOW-based variables described above. The final model retained 25 of an initial 145 predictor variables considered, had training and hold-out R^2 values of 0.83 and 0.44 respectively, and was used to produce a 3D visualization of nitrate in the aquifer. These studies show that modeling advances and improved characterization of aquifer conditions at depth are increasing our ability to predict nitrate exposure from drinking water supplied by private wells.

4. Nitrate Intake and Endogenous Formation of *N*-Nitroso Compounds

Drinking water nitrate is readily absorbed in the upper gastrointestinal tract and distributed in the human body. When it reaches the salivary glands, it is actively transported from blood into saliva and levels may be up to 20 times higher than in the plasma [50–53]. In the oral cavity 6–7% of the total nitrate can be reduced to nitrite, predominantly by nitrate-reducing bacteria [52,54,55]. The secreted nitrate as well as the nitrite generated in the oral cavity re-enter the gastrointestinal tract when swallowed.

Under acidic conditions in the stomach, nitrite can be protonated to nitrous acid (HNO_2), and subsequently yield dinitrogen trioxide (N_2O_3), nitric oxide (NO), and nitrogen dioxide (NO_2). Since the discovery of endogenous NO formation, it has become clear that NO is involved in a wide range of NO-mediated physiological effects. These comprise the regulation of blood pressure and blood flow by mediating vasodilation [56–58], the maintenance of blood vessel tonus [59], the inhibition of platelet adhesion and aggregation [60,61], modulation of mitochondrial function [62] and several other processes [63–66].

On the other hand, various nitrate and nitrite derived metabolites such as nitrous acid (HNO_2) are powerful nitrosating agents and known to drive the formation of NOC, which are

suggested to be the causal agents in many of the nitrate-associated adverse health outcomes. NOC comprise *N*-nitrosamines and *N*-nitrosamides, and may be formed when nitrosating agents encounter *N*-nitrosatable amino acids, which are also from dietary origin. The nitrosation process depends on the reaction mechanisms involved, on the concentration of the compounds involved, the pH of the reaction environment, and further modifying factors, including the presence of catalysts or inhibitors of *N*-nitrosation [66–69].

Endogenous nitrosation can also be inhibited, for instance by dietary compounds like vitamin C, which has the capacity to reduce HNO_2 to NO; and alpha-tocopherol or polyphenols, which can reduce nitrite to NO [54,70–72]. Inhibitory effects on nitrosation have also been described for dietary flavonoids such as quercetin, ferulic and caffeic acid, betel nut extracts, garlic, coffee, and green tea polyphenols [73,74]. Earlier studies showed that the intake of 250 mg or 1 g ascorbic acid per day substantially inhibited *N*-nitrosodimethylamine (NDMA) excretion in 25 women consuming a fish meal rich in amines (nitrosatable precursors) for seven days, in combination with drinking water containing nitrate at the acceptable daily intake (ADI) [75]. In addition, strawberries, garlic juice, and kale juice were shown to inhibit NDMA excretion in humans [76]. The effect of these fruits and vegetables is unlikely to be due solely to ascorbic acid. Using the *N*-nitrosoproline (NPRO) test, Helser et al. [77] found that ascorbic acid only inhibited nitrosamine formation by 24% compared with 41–63% following ingestion of juices (100 mL) made of green pepper, pineapple, strawberry or carrot containing an equal total amount of ascorbic acid.

The protective potential of such dietary inhibitors depends not only on the reaction rates of *N*-nitrosatable precursors and nitrosation inhibitors, but also on their biokinetics, since an effective inhibitor needs to follow gastrointestinal circulation kinetics similar to nitrate [78]. It has been argued that consumption of some vegetables with high nitrate content, can at least partially inhibit the formation of NOC [79–81]. This might apply for green leafy vegetables such as spinach and rocket salad, celery or kale [77] as well as other vegetables rich in both nitrate and natural nitrosation inhibitors. Preliminary data show that daily consumption of one bottle of beetroot juice containing 400 mg nitrate (the minimal amount advised for athletes to increase their sports performances) for one day and seven days by 29 young individuals results in an increased urinary excretion of apparent total nitroso compounds (ATNC), an effect that can only be partially inhibited by vitamin C supplements (1 g per day) [82].

Also, the amount of nitrosatable precursors is a key factor in the formation of NOC. Dietary intakes of red and processed meat are of particular importance [83–87] as increased consumption of red meat (600 vs. 60 g/day), but not white meat, was found to cause a three-fold increase in fecal NOC levels [85]. It was demonstrated that heme iron stimulated endogenous nitrosation [84], thereby providing a possible explanation for the differences in colon cancer risk between red and white meat consumption [88]. The link between meat consumption and colon cancer risk is even stronger for nitrite-preserved processed meat than for fresh meat leading an IARC review to conclude that processed meat is carcinogenic to humans [89].

In a human feeding study [90], the replacement of nitrite in processed meat products by natural antioxidants and the impact of drinking water nitrate ingestion is being evaluated in relation to fecal excretion of NOC, accounting for intakes of meat and dietary vitamin C. A pilot study demonstrated that fecal excretion of ATNC increased after participants switched from ingesting drinking water with low nitrate levels to drinking water with nitrate levels at the acceptable daily intake level of 3.7 mg/kg. The 20 volunteers were assigned to a group consuming either 3.75 g/kg body weight (maximum 300 g per day) red processed meat or fresh (unprocessed) white meat. Comparison of the two dietary groups showed that the most pronounced effect of drinking water nitrate was observed in the red processed meat group. No inhibitory effect of vitamin C intake on ATNC levels in feces was found (unpublished results).

5. Methemoglobinemia

The physiologic processes that can lead to methemoglobinemia in infants under six months of age have been described in detail previously [8,91]. Ingested nitrate is reduced to nitrite by bacteria in the mouth and in the infant stomach, which is less acidic than adults. Nitrite binds to hemoglobin to form methemoglobin, which interferes with the oxygen carrying capacity of the blood. Methemoglobinemia is a life-threatening condition that occurs when methemoglobin levels exceed about 10% [8,91]. Risk factors for infant methemoglobinemia include formula made with water containing high nitrate levels, foods and medications that have high nitrate levels [91,92], and enteric infections [93]. Methemoglobinemia related to high nitrate levels in drinking water used to make infant formula was first reported in 1945 [94]. The U.S. EPA limit of 10 mg/L NO_3-N was set as about one-half the level at which there were no observed cases [95]. The most recent U.S. cases related to nitrate in drinking water were reported by Knobeloch and colleagues in the late 1990s in Wisconsin [96] and were not described in our prior review. Nitrate concentrations in the private wells were about two-times the MCL and bacterial contamination was not a factor. They also summarize another U.S. case in 1999 related to nitrate contamination of a private well and six infant deaths attributed to methemoglobinemia in the U.S. between 1979–1999 only one of which was reported in the literature [96,97]. High incidence of infant methemoglobinemia in eastern Europe has also been described previously [98,99]. A 2002 WHO report on water and health [100] noted that there were 41 cases in Hungary annually, 2913 cases in Romania from 1985–1996 and 46 cases in Albania in 1996.

Results of several epidemiologic studies conducted before 2005 that examined the relationship between nitrate in drinking water and levels of methemoglobin or methemoglobinemia in infants have been described previously [8]. Briefly, nitrate levels >10 mg/L NO_3-N were usually associated with increased methemoglobin levels but clinical methemoglobinemia was not always present. Since our last review, a cross-sectional study conducted in Gaza found elevated methemoglobin levels in infants on supplemental feeding with formula made from well water in an area with the highest mean nitrate concentration of 195 mg/L NO_3 (range: 18–440) compared to an area with lower nitrate concentration (mean: 119 mg/L NO_3; range 18–244) [101]. A cross-sectional study in Morocco found a 22% increased risk of methemoglobinemia in infants in an area with drinking water nitrate >50 mg/L (>11 as NO_3-N) compared to infants in an area with nitrate levels <50 mg/L nitrate [102]. A retrospective cohort study in Iowa of persons (aged 1–60 years) consuming private well water with nitrate levels <10 mg/L NO_3-N found a positive relationship between methemoglobin levels in the blood and the amount of nitrate ingestion [103]. Among pregnant women in rural Minnesota with drinking water supplies that were mostly $\leq$3 mg/L NO_3-N, there was no relationship between water nitrate intake and women's methemoglobin levels around 36 weeks' gestation [104].

6. Adverse Pregnancy Outcomes

Maternal drinking water nitrate intake during pregnancy has been investigated as a risk factor for a range of pregnancy outcomes, including spontaneous abortion, fetal deaths, prematurity, intrauterine growth retardation, low birth weight, congenital malformations, and neonatal deaths. The relation between drinking water nitrate and congenital malformations in offspring has been the most extensively studied, most likely because of the availability of birth defect surveillance systems around the world.

Our earlier review focused on studies of drinking water nitrate and adverse pregnancy outcomes published before 2005 [8]. In that review, we cited several studies on the relation between maternal exposure to drinking water nitrate and spontaneous abortion including a cluster investigation that suggested a positive association [105] and a case-control study that found no association [106]. These studies were published over 20 years ago. In the present review, we were unable to identify any recently published studies on this outcome. In Table 1, we describe the findings of studies published since 2004 on the relation between drinking water nitrate and prematurity, low birthweight, and congenital malformations. We report results for nitrate in the units (mg/L NO_3 or NO_3-N) that

were reported in the publications. In a historic cohort study conducted in the Deux-Sèvres district (France), Migeot et al. [26] linked maternal addresses from birth records to community water system measurements of nitrate, atrazine, and other pesticides. Exposure to the second tertile of nitrate (14–27 mg/L NO_3) without detectable atrazine metabolites was associated with small-for-gestational age births (Odds Ratio (OR) 1.74, 95% CI 1.1, 2.8), but without a monotonic increase in risk with exposures. There was no association with nitrate among those with atrazine detected in their drinking water supplies. Within the same cohort, Albouy-Llaty and colleagues did not observe any association between higher water nitrate concentrations (with or without the presence of atrazine) and preterm birth [107].

Stayner and colleagues also investigated the relation between atrazine and nitrate in drinking water and rates of low birth weight and preterm birth in 46 counties in four Midwestern U.S. states that were required by EPA to measure nitrate and atrazine monthly due to prior atrazine MCL violations [108]. The investigators developed county-level population-weighted metrics of average monthly nitrate concentrations in public drinking water supplies. When analyses were restricted to counties with less than 20% private well usage (to reduce misclassification due to unknown nitrate levels), average nitrate concentrations during the pregnancy were associated with increased rates of very low birth weight (<1.5 kg Rate Ratio $(RR)_{per\ 1\ ppm}$ = 1.17, 95% CI 1.08, 1.25) and very preterm births (<32 weeks $RR_{per\ 1\ ppm}$ = 1.08, 95% CI 1.02, 1.15) but not with low birth weight or preterm birth overall.

In record-based prevalence study in Perth Australia, Joyce et al. mapped births to their water distribution zone and noted positive associations between increasing tertiles of nitrate levels and prevalence of term premature rupture of membranes (PROM) adjusted for smoking and socioeconomic status [109]. Nitrate concentrations were low; the upper tertile cut point was 0.350 mg/L and the maximum concentration was 1.80 mg/L NO_3-N. Preterm PROM was not associated with nitrate concentrations.

Among studies of drinking water nitrate and congenital malformations, few before 2005 included birth defects other than central nervous system defects [8]. More recently, Mattix et al. [110] noted higher rates of abdominal wall defects (AWD) in Indiana compared to U.S. rates for specific years during the period 1990–2002. They observed a positive correlation between monthly AWD rates and monthly atrazine concentrations in surface waters but no correlation with nitrate levels. Water quality data were obtained from the USGS-NAWQA project that monitors agricultural chemicals in streams and shallow groundwater that are mostly not used as drinking water sources. A case-control study of gastroschisis (one of the two major types of AWD), in Washington State [111] also used USGS-NAWQA measurements of nitrate and pesticides in surface water and determined the distance between maternal residences (zip code centroids) and the closest monitoring site with concentrations above the MCL for nitrate, nitrite, and atrazine. Gastrochisis was not associated with maternal proximity to surface water above the MCL for nitrate (>10 mg/L NO_3-N) or nitrite (>1 mg/L NO_2-N) but there was a positive relationship with proximity to sites with atrazine concentrations above the MCL. In a USA-wide study, Winchester et al. [112] linked the USGS-NAWQA monthly surface water nitrate and pesticide concentrations computed for the month of the last menstrual period with monthly rates of 22 types of birth defects in 1996–2002. Rates of birth defects among women who were estimated to have conceived during April through July were higher than rates among women conceiving in other months. In multivariable models that included nitrate, atrazine, and other pesticides, atrazine (but not nitrate or other pesticides) was associated with several types of anomalies. Nitrate was associated with birth defects in the category of "other congenital anomalies" (OR 1.18, 95% CI 1.14, 1.21); the authors did not specify what defects were included in this category. None of these three studies included local or regional data to support the assumption that surface water nitrate and pesticide concentrations correlated with drinking water exposures to these contaminants.

Using a more refined exposure assessment than the aforementioned studies, Holtby et al. [113] conducted a case-control study of congenital anomalies in an agricultural county in Nova Scotia,

Canada. They linked maternal addresses at delivery to municipal water supply median nitrate concentrations and used kriging of monthly measurements from a network of 140 private wells to estimate drinking water nitrate concentrations in private wells. They observed no associations between drinking water nitrate and all birth defects combined for conceptions during 1987–1997. However, the prevalence of all birth defects occurring during 1998–2006 was associated with drinking water nitrate concentrations of 1–5.56 mg/L NO_3-N (OR 2.44, 95% CI 1.05, 5.66) and ≥5.56 mg/L (OR 2.25, 95% CI 0.92, 5.52).

None of the studies of congenital anomalies accounted for maternal consumption of bottled water or the quantity of water consumed during the first trimester, the most critical period of organ/structural morphogenesis. Attempting to overcome some of these limitations, Brender, Weyer, and colleagues [38,114] conducted a population-based, case-control study in the states of Iowa and Texas where they: (1) linked maternal addresses during the first trimester to public water utilities and respective nitrate measurements; (2) estimated nitrate intake from bottled water based on a survey of products consumed and measurement of nitrate in the major products; (3) predicted drinking water nitrate from private wells through modeling (Texas only); and (4) estimated daily nitrate ingestion from women's drinking water sources and daily consumption of water. The study populations were participants of the U.S. National Birth Defects Prevention Study [115]. Compared to the lowest tertile of nitrate ingestion from drinking water (<0.91 mg/day NO_3), mothers of babies with spina bifida were twice as likely (95% CI 1.3, 3.2) to ingest ≥5 mg/day NO_3 from drinking water than control mothers. Mothers of babies with limb deficiencies, cleft palate, and cleft lip were, respectively, 1.8 (95% CI 1.1, 3.1), 1.9 (95% CI 1.2, 3.1), and 1.8 (95% CI 1.1, 3.1) times more likely to ingest ≥5.4 mg/day of water NO_3 than controls. Women were also classified by their nitrosatable drug exposure during the first trimester [116] and by their daily nitrate and nitrite intake based on a food frequency questionnaire [117]. Higher ingestion of drinking water nitrate did not strengthen associations between maternal nitrosatable drug exposure and birth defects in offspring [38]. However, a pattern was observed of stronger associations between nitrosatable drug exposure and selected birth defects for women in the upper two tertiles of total nitrite ingestion that included contributions from drinking water nitrate and dietary intakes of nitrate and nitrite compared to women in the lowest tertile. Higher intake of food nitrate/nitrite was found to also modify the associations of nitrosatable drug exposure and birth defects in this study [118,119] as well as in an earlier study of neural tube defects conducted in south Texas [120]. Multiplicative interactions were observed between higher food nitrate/nitrite and nitrosatable drug exposures for conotruncal heart, limb deficiency, and oral cleft defects [118].

In summary, five out of six studies, conducted since the 1980s of drinking water nitrate and central nervous system defects, found positive associations between higher drinking water nitrate exposure during pregnancy and neural tube defects or central nervous system defects combined [38,120–123]. The sixth study, which did not find a relationship, did not include measures of association, but compared average drinking water nitrate concentrations between mothers with and without neural tube defect-affected births, which were comparable [124].

Table 1. Studies of drinking water nitrate [a] and adverse pregnancy outcomes published January 2005–March 2018.

First Author, Year, Country	Study Design Regional Description	Years of Outcome Ascertainment	Exposure Description	Pregnancy Outcome	Summary of Findings
Albouy-Llaty, 2016 France [107]	Historic cohort study Deux-Sèvres	2005–2010	Measurements of atrazine metabolites and NO_3 in community water systems (263 municipalities) were linked to birth addresses	Preterm birth	No association for >26.99 mg/L vs. <14.13 mg/L NO_3 in community water systems with or without atrazine detections, adjusted for neighborhood deprivation
Brender, 2013 Weyer, 2014 USA [38]	Population-based case-control study Iowa and Texas	1997–2005	Maternal addresses during the first trimester linked to public water utility nitrate measurements; nitrate intake from bottled water estimated with survey and laboratory testing; nitrate from private wells predicted through modeling; nitrate ingestion (NO_3) estimated from reported water consumption	Congenital heart defects Limb deficiencies Neural tube defects Oral cleft defects	≥5 vs. <0.91 mg/day NO_3 from drinking water spina bifida OR = 2.0 (95% CI: 1.3, 3.2) ≥5.42 vs. <1.0 mg/day NO_3 from water: limb deficiencies OR = 1.8 (CI: 1.1, 3.1); cleft palate OR = 1.9 (CI: 1.2, 3.1) cleft lip OR = 1.8 (CI: 1.1, 3.1)
Holtby, 2014 Canada [113]	Population-based case-control study Kings County, Nova Scotia	1988–2006	Maternal addresses at delivery linked to municipal water supply median nitrate (NO_3-N) concentrations; nitrate in rural private wells estimated from historic sampling and kriging	Congenital malformations combined into one group	Conceptions in 1987–1997: no association with nitrate concentrations Conceptions in 1998–2006: 1–5.56 mg/L NO_3-N (vs. <1 mg/L) OR = 2.44 (CI: 1.05, 5.66); ≥5.56 mg/L OR = 2.25 (CI: 0.92, 5.52)
Joyce, 2008 Australia [109]	Record-based prevalence study Perth	2002–2004	Linked birth residences to 24 water distribution zones; computed average NO_3-N mg/L from historical measurements; independent sampling conducted for 6 zones as part of exposure validation; also evaluated trihalomethanes (THM)	Premature rupture of membranes at term (PROM) (37 weeks' gestation or later)	ORs for tertiles (vs. <0.125 mg/L NO_3-N): 0.125–0.350 mg/L OR = 1.23 (CI: 1.03, 1.52); >0.350 mg/L OR = 1.47 (CI: 1.20, 1.79) No association with THM levels
Mattix, 2007 USA [110]	Ecologic study Indiana	1990–2002	Monthly abdominal wall defect rates linked to monthly surface water nitrate and atrazine concentrations (USGS-NAWQA monitoring data [b])	Abdominal wall birth defects	No correlation observed between nitrate levels in surface water and monthly abdominal wall defects Positive correlation with atrazine levels

Table 1. *Cont.*

First Author, Year, Country	Study Design Regional Description	Years of Outcome Ascertainment	Exposure Description	Pregnancy Outcome	Summary of Findings
Migeot, 2013 France [26]	Historic cohort study Deux-Sèvres	2005–2009	Measurements of atrazine metabolites and NO_3 in community water systems (263 municipalities) were linked to birth addresses	Small-for-gestational age (SGA) births	ORs for tertiles (vs. <14.13 mg/L NO_3) in community water systems with no atrazine detections: 14–27 mg/L OR = 1.74 (CI: 1.10, 2.75); >27 mg/L OR = OR 1.51 (CI: 0.96, 2.4); no association with nitrate when atrazine was detected
Stayner, 2017 USA [108]	Ecologic study 46 counties in Indiana, Iowa, Missouri, and Ohio	2004–2008	Counties had one or more water utility in EPA's atrazine monitoring program; excluded counties with >20% of population on private wells and >300,000 population. Computed county-specific monthly weighted averages of NO_3-N in finished drinking water; exposure metric was average 9 months prior to birth	Preterm birth Low birth weight	Average nitrate not associated with low birth weight and preterm birth Very low birth weight: RR for 1 ppm increase in NO_3-N = 1.17 (CI: 1.08, 1.25); Very preterm birth RR for 1 ppm increase = 1.08 (CI: 1.02, 1.15)
Waller, 2010 USA [111]	Population-based case-control study Washington State	1987–2006	Calculated distance between maternal residence and closest stream monitoring site with concentrations >MCL for NO_3-N, NO_2-N, or atrazine in surface water (USGS-NAWQA data [b])	Gastroschisis	Gastroschisis was not associated with maternal residential proximity to surface water with elevated nitrate (>10 mg/L) or nitrite (>1 mg/L)
Winchester, 2009 USA [112]	Ecologic study USA-wide	1996–2002	Rates of combined and specific birth defects (computed by month of last menstrual period) linked to monthly surface water nitrate concentrations (USGS-NAWQA data [b]); also evaluated atrazine and other pesticides (combined)	Birth defects categorized into 22 groups	Birth defect category "other congenital anomalies": OR for continuous log nitrate = 1.15 (CI: 1.12, 1.18); adjusted for atrazine and other pesticides: OR = 1.18, CI: 1.14, 1.21); No association with other birth defects

Abbreviations: CI, 95% CI confidence interval; OR, odds ratio; RR, rate ratio; USGS-NAWQA, U. S. Geological Survey National Water Quality Assessment; [a] nitrate units are specified as reported in publications. NO_3 can be converted to NO_3-N by multiplying by 0.2258; [b] USGS-NAWQA data for 186 streams in 51 hydrological study areas; streams were not drinking water sources.

7. Cancer

Most early epidemiologic studies of cancer were ecologic studies of stomach cancer mortality that used exposure estimates concurrent with the time of death. Results were mixed, with some studies showing positive associations, many showing no association, and a few showing inverse associations. The results of ecologic studies through 1995 were reviewed by Cantor [125]. Our previous review included ecologic studies of the brain, esophagus, stomach, kidney, ovary, and non-Hodgkin lymphoma (NHL) published between 1999 and 2003 that were largely null [8]. We did not include ecologic studies or mortality case-control studies in this review due to the limitations of these study designs, especially their inability to assess individual-level exposure and dietary factors that influence the endogenous formation of NOC.

Since our review of drinking water nitrate and health in 2005 [8], eight case-control studies and eight analyses in three cohorts have evaluated historical nitrate levels in PWS in relation to several cancers. Nitrate levels were largely below 10 mg/L NO_3-N. Most of these studies evaluated potential confounders and factors affecting nitrosation. Table 2 shows the study designs and results of studies published from 2005 through 2018, including findings from periodic follow-ups of a cohort study of postmenopausal women in Iowa (USA) [31,37,126–129]. In the first analysis of drinking water nitrate in the Iowa cohort with follow-up through 1998, Weyer and colleagues [130] reported that ovarian and bladder cancers were positively associated with the long-term average PWS nitrate levels prior to enrollment (highest quartile average 1955–1988: >2.46 mg/L NO_3-N). They observed inverse associations for uterine and rectal cancer, but no associations with cancers of the breast, colon, rectum, pancreas, kidney, lung, melanoma, non-Hodgkin lymphoma (NHL), or leukemia. Analyses of PWS nitrate concentrations and cancers of the thyroid, breast, ovary, bladder, and kidney were published after additional follow-up of the cohort. The exposure assessment was improved by: (a) the computation of average nitrate levels and years of exposure at or above 5 mg/L NO_3-N, based on time in residence (vs. one long-term PWS average nitrate estimate used by Weyer and colleagues); and (b) by estimation of total trihalomethanes (TTHM) and dietary nitrite intake.

Thyroid cancer was evaluated for the first time after follow-up of the cohort through 2004. A total of 40 cases were identified [37]. Among women with >10 years on PWS with levels exceeding 5 mg/L NO_3-N for five years or more, thyroid cancer risk was 2.6 times higher than that of women whose supplies never exceeded 5 mg/L. With follow-up through 2010, the risk of ovarian cancer remained increased among women in the highest quartile of average nitrate in PWS [129]. Ovarian cancer risk among private well users was also elevated compared to the lowest PWS nitrate quartile. Associations were stronger when vitamin C intake was below median levels with a significant interaction for users of private wells. Overall, breast cancer risk was not associated with water nitrate levels with follow-up through 2008 [128]. Among women with folate intake $\geq$ 400 µg/day, risk was increased for those in the highest average nitrate quintile (Hazard Ratio (HR) = 1.40; 95% CI: = 1.05–1.87) and among private well users (HR = 1.38; 95% CI: = 1.05–1.82), compared to those with the lowest average nitrate quintile. There was no association with nitrate exposure among women with lower folate intake. With follow-up through 2010, there were 130 bladder cancer cases among women who had used PWS >10 years. Risk remained elevated among women with the highest average nitrate levels and was 1.6 times higher among women whose drinking water concentration exceeded 5 mg/L NO_3-N for at least four years [31]. Risk estimates were not changed by adjustment for TTHM, which are suspected bladder cancer risk factors. Smoking, but not vitamin C intake, modified the association with nitrate in water; increased risk was apparent only in current smokers (p-interaction <0.03). With follow-up through 2010, there were 125 kidney cancer cases among women using PWS; risk was increased among those in the 95th percentile of average nitrate (>5.0 mg/L NO_3-N) compared with the lowest quartile (HR = 2.2, 95% CI: 1.2–4.2) [127]. There was no positive trend with the average nitrate level and no increased risk for women using private wells, compared to those with low average nitrate in their public supply. An investigation of pancreatic cancer in the same population (follow-up through 2011)

found no association with average water nitrate levels in public supplies and no association among women on private wells [126].

In contrast to the positive findings for bladder cancer among the cohort of Iowa women, a cohort study of men and women aged 55–69 in the Netherlands with lower nitrate levels in PWS found no association between water nitrate ingestion (median in top quintile = 2.4 mg/day NO_3-N) and bladder cancer risk [131]. Dietary intake of vitamins C and E and history of cigarette smoking did not modify the association. A hospital-based case-control study of bladder cancer in multiple areas of Spain [33] assessed lifetime water sources and usual intake of tap water. Nitrate levels in PWS were low, with almost all average levels below 2 mg/L NO_3-N. Risk of bladder cancer was not associated with the nitrate level in drinking water or with estimated nitrate ingestion from drinking water, and there was no evidence of interaction with factors affecting endogenous nitrosation.

Several case-control studies conducted in the Midwestern U.S. obtained lifetime histories of drinking water sources and estimated exposure for PWS users. In contrast to findings of an increased risk of NHL associated with nitrate levels in Nebraska PWS in an earlier study [132], there was no association with similar concentrations in public water sources in a case-control study of NHL in Iowa [35]. A study of renal cell carcinoma in Iowa [34] found no association with the level of nitrate in PWS, including the number of years that levels exceeded 5 or 10 mg/L NO_3-N. However, higher nitrate levels in PWS increased risk among subgroups who reported above the median intake of red meat intake or below the median intake of vitamin C (p-interaction <0.05). A small case-control study of adenocarcinoma of the stomach and esophagus among men and women in Nebraska [133] estimated nitrate levels among long-term users of PWS and found no association between average nitrate levels and risk.

A case-control study of colorectal cancer among rural women in Wisconsin estimated nitrate levels in private wells using spatial interpolation of nitrate concentrations from a 1994 water quality survey and found increased risk of proximal colon cancer among women estimated to have nitrate levels >10 mg/L NO_3-N compared to levels < 0.5 mg/L. Risk of distal colon cancer and rectal cancer were not associated with nitrate levels [134]. Water nitrate ingestion from public supplies, bottled water, and private wells and springs over the adult lifetime was estimated in analyses that pooled case-control studies of colorectal cancer in Spain and Italy [135]. Risk of colorectal cancer was increased among those with >2.3 mg/day NO_3-N (vs. <1.1 mg/day). There were no interactions with red meat, vitamins C and E, and fiber except for a borderline interaction (p-interaction = 0.07) for rectum cancer with fiber intake. A small hospital-based case-control study in Indonesia found that drinking water nitrate levels above the WHO standard (>11.3 mg/L as NO_3-N) was associated with colorectal cancer [136]. A national registry-based cohort study in Denmark [32] evaluated average nitrate concentrations in PWS and private wells in relation to colorectal cancer incidence among those whose 35th birthday occurred during 1978–2011. The average nitrate level was computed over residential water supplies from age 20 to 35. Increased risks for colon and rectum cancer were observed in association with average nitrate levels $\geq$9.25 mg/L NO_3 ($\geq$2.1 as NO_3-N) and $\geq$3.87 mg/L NO_3 (>0.87 as NO_3-N), respectively, with a significant positive trend. Because the study did not interview individuals, it could not evaluate individual-level risk factors that might influence endogenous nitrosation.

A case-control study of breast cancer in Cape Cod, Massachusetts (US) [137] estimated nitrate concentrations in PWS over approximately 20 years as an historical proxy for wastewater contamination and potential exposure to endocrine disruption compounds. Average exposures >1.2 mg/L NO_3-N (vs. <0.3 mg/L) were not associated with risk. A hospital-based case-control study in Spain found no association between water nitrate ingestion and pre- and post-menopausal breast cancers [138].

Table 2. Case-control and cohort studies of drinking water nitrate and cancer (January 2004–March 2018) by cancer site.

First Author (Year) Country	Study Design, Years Regional Description	Exposure Description	Cancer Sites Included	Summary of Drinking-Water Findings [a,b]	Evaluation of Effect Modification [c]
Zeegers, 2006 Netherlands [131]	Cohort Incidence, 1986–1995 204 municipal registries across the Netherlands	1986 nitrate level in 364 pumping stations, exposure data available for 871 cases, 4359 members of the subcohort	Bladder	Highest vs. lowest quintile intake from water (≥1.7 mg/day NO_3-N [median 2.4 mg/day] vs. <0.20) RR = 1.11 (CI: 0.87–1.41; *p*-trend = 0.14)	No interaction with vitamin C, E, smoking
Espejo-Herrera, 2015 Spain [33]	Hospital-based multi-center case-control Incidence, 1998–2001 Asturias, Alicante, Barcelona, Vallès-Bages, Tenerife provinces	Nitrate levels in PWS (1979–2010) and bottled water (measurements of brands with highest consumption based on a Spanish survey); analyses limited to those with ≥70% of residential history with nitrate estimate (531 cases, 556 controls)	Bladder	Highest vs. lowest quartile average level (age 18-interview) (≥2.26 vs. 1.13 mg/L NO_3-N) OR = 1.04 (CI: 0.60–1.81) Years >2.15 mg/L NO_3-N (75th percentile) (>20 vs. 0 years) OR = 1.41 (CI: 0.89–2.24)	No interaction with vitamin C, E, red meat, processed meat, average THM level
Jones, 2016 USA [31]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2010 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence with nitrate and trihalomethane estimates (20,945 women; 170 bladder cases); no measurements for private wells Adjusted for total trihalomethanes (TTHM)	Bladder	Highest vs. lowest quartile PWS average (≥2.98 vs. <0.47 mg/L NO_3-N) HR = 1.47 (CI: 0.91–2.38; *p*-trend = 0.11) Years >5 mg/L (≥4 years vs. 0) HR = 1.61 (CI: 1.05–2.47; *p*-trend = 0.03) Private well users (vs. <0.47 mg/L NO_3-N on PWS) HR = 1.53 (CI: 0.93–2.54)	Interaction with smoking (*p*-interaction = 0.03); HR = 3.67 (CI: 1.43–9.38) among current smokers/≥2.98 mg/L vs. non-smokers/<0.47 mg/L NO_3-N); No interaction with vitamin C, TTHM levels
Mueller, 2004 USA, Canada, France, Italy, Spain [139]	Pooled case-control studies Incidence among children <15 years (USA <20 years) 7 regions of 5 countries	Water source during pregnancy and first year of child's life (836 cases, 1485 controls); nitrate test strip measurements of nitrate and nitrite for pregnancy home (except Italy) (283 cases, 537 controls; excluding bottled water users: 207 cases, 400 controls)	Brain, childhood	Private well use versus PWS associated with increased risk in 2 regions and decreased risk in one; No association with nitrate levels in water supplies Astrocytomas (excludes bottled water users): ≥1.5 vs. <0.3 mg/L NO_2-N OR = 5.7 (CI: 1.2–27.2)	Not described
Brody, 2006 USA [137]	Case-control Incidence, 1988–1995 Cape Cod, Massachusetts	Nitrate levels in public water supplies (PWS) since 1972 was used as an indicator of wastewater contamination and potential mammary carcinogens and endocrine disrupting compounds; excluded women on private wells	Breast	Average ≥1.2 mg/L NO_3-N vs. <0.3 OR = 1.8, (CI: 0.6–5.0); summed annual NO_3-N ≥ 10 vs. 1–< 10 mg/L OR = 0.9, CI: 0.6–1.5); number of years >1 mg/L NO_3-N ≥8 vs. 0 years OR = 0.9 (CI: 0.5–1.5)	Not described

Table 2. *Cont.*

First Author (Year) Country	Study Design, Years Regional Description	Exposure Description	Cancer Sites Included	Summary of Drinking-Water Findings [a,b]	Evaluation of Effect Modification [c]
Inoue-Choi, 2012 USA [128]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2008 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence (20,147 women; 1751 breast cases); no measurements for private wells	Breast	Highest vs. lowest quintile PWS average (≥3.8 vs. ≤0.32 mg/L NO_3-N) HR = 1.14 (CI: 0.95–1.36; *p*-trend = 0.11); Private well (vs. ≤ 0.32 mg/L NO_3-N) HR = 1.14 (CI: 0.97–1.34); Private well (vs. ≤0.32 mg/L NO_3-N on PWS) HR = 1.38 (CI: 1.05–1.82); No association among those with low folate <400 µg/day	Interaction with folate for PWS (*p*-interaction = 0.06). Folate ≥400 µg/d: (≥3.8 vs. ≤0.32 mg/L NO_3-N) HR = 1.40 (CI: 1.05–1.87; *p*-trend = 0.04)
Espejo-Herrera, 2016 Spain [138]	Hospital-based multi-center case-control Incidence, 2008–2013 Spain (8 provinces)	Nitrate levels in PWS (2004–2010), bottled water measurements and private wells and springs (2013 measurements in 21 municipalities in León, Spain, the area with highest non-PWS use) Analyses include women with ≥70% of period from age 18 to 2 years before interview (1245 cases, 1520 controls)	Breast	Water nitrate intake based on average nitrate levels (age 18 to 2 years prior to interview) and water intake (L/day). Post-menopausal women: >2.0 vs. 0.5 mg/day NO_3-N OR = 1.32 (0.93–1.86); Premenopausal women: >1.4 vs. 0.4 mg/day NO_3-N OR = 1.14 (0.67–1.94)	No interaction with red meat, processed meat, vitamin C, E, smoking for pre- and post-menopausal women
McElroy, 2008 USA [134]	Population-based case-control, women Incidence, 1990–1992 and 1999–2001 Wisconsin	Limited to women in rural areas with no public water system (475 cases, 1447 controls); nitrate levels at residence (presumed to be private wells) estimated by kriging using data from a 1994 representative sample of 289 private wells	Colorectal	All colon cancers: Private wells ≥10.0 mg/L NO_3-N vs. <0.5 OR = 1.52 (CI: 0.95–2.44); Proximal colon cancer: OR = 2.91 (CI: 1.52–5.56)	Not described
Espejo-Herrera, 2016 Spain, Italy [135]	Multi-center case-control study Incidence, 2008–2013 Spain (9 provinces) and population-based controls; Italy (two provinces) and hospital-based controls	Nitrate levels in PWS (2004–2010) for 349 water supply zones, bottled water (measured brands with highest consumption), and private wells and springs (measurements in 2013 in 21 municipalities in León, Spain, the area with highest non-PWS use) Analyses include those with nitrate estimates for ≥70% of period 30 years before interview (1869 cases, 3530 controls)	Colorectal	Water nitrate intake based on average nitrate levels (estimated 30 to 2 years prior to interview) and water intake (L/day) Highest vs. lowest exposure quintiles (≥2.3 vs. <1.1 mg /day NO_3-N) OR = 1.49 (CI:1.24–1.78); Colon OR = 1.52 (CI: 1.24–1.86), Rectum OR = 1.62 (CI: 1.23–2.14)	Interaction with fiber for rectum (*p*-interaction = 0.07); >20 g/day fiber + >1.0 mg/L NO_3-N vs. <20 g/day + ≤1.0 mg/L HR = 0.72 (CI: 0.52–1.00). No interaction with red meat, vitamin C, E

Table 2. *Cont.*

First Author (Year) Country	Study Design, Years Regional Description	Exposure Description	Cancer Sites Included	Summary of Drinking-Water Findings [a,b]	Evaluation of Effect Modification [c]
Fathmawati, 2017 Indonesia [136]	Hospital-based case-control Incidence, 2014–2016 Indonesia (3 provinces)	Nitrate levels in well water collected during the raining season (Feb-March 2016) and classified based on >11.3 or ≤11.3 mg/L as NO_3-N and duration of exposure >10 and ≤10 years Analyses included participants who reported drinking well water (75 cases, 75 controls)	Colorectal	Water nitrate > WHO standard vs. below (> 11.3 vs. ≤11.3 mg/L NO_3-N) OR = 2.82 (CI: 1.08–7.40); > 10 years: 4.31 (CI: 11.32–14.10); ≤10 years: 1.41 (CI: 0.14–13.68)	Not described
Schullehner, 2018 Denmark [32]	Population-based record-linkage cohort of men and women ages 35 and older, 1978–2011 Denmark	Nitrate levels in PWS and private wells among 1,742,321 who met exposure assessment criteria (5944 colorectal cancer cases, including 3700 with colon and 2308 with rectal cancer)	Colorectal	Annual average nitrate exposure between ages 20–35 among those who lived ≥75% of study period at homes with a water sample within 1 year (61% of Danish population). Highest vs. lowest exposure quintile (≥2.1 vs. 0.16 mg/L NO_3-N); Colorectal: HR = 1.16 (CI: 1.08–1.25); colon: 1.15 (CI: 1.05–1.26); rectum: 1.17 (CI: 1.04–1.32)	No information on dietary intakes or smoking
Ward, 2007 USA [34]	Population-based case control Incidence, 1986–1989 Iowa	Nitrate levels in PWS among those with nitrate estimates for ≥70% of person-years ≥1960 (201 cases, 1244 controls)	Kidney (renal cell carcinomas)	Highest vs. lowest quartile PWS average (≥2.8 mg/L NO_3-N vs. <0.62) OR = 0.89 (CI 0.57–1.39); Years >5mg/L NO_3-N 11+ vs. 0 OR = 1.03 (CI: 0.66–1.60)	Interaction with red meat intake (p-interaction = 0.01); OR = 1.91 (CI 1.04–3.51) among 11+ years >5 mg/L NO_3-N and red meat ≥1.2 servings/day. Interaction with vitamin C showed similar pattern (p-interaction = 0.13)
Jones, 2017 USA [127]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2010 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence. PWS measurements for nitrate and TTHM; no measurements for private wells (20,945 women; 163 kidney cases)	Kidney	Nitrate and TTHM metrics computed for duration at water source (11+ years) 95th percentile vs. lowest quartile PWS average (≥5.00 vs. <0.47 mg/L NO_3-N) HR = 2.23 (CI: 1.19–4.17; p-trend = 0.35) Years >5 mg/L (≥4 years vs. 0) HR = 1.54 (CI: 0.97–2.44; p-trend = 0.09) Private well users (vs. <0.47 mg/L NO_3-N in PWS) HR = 0.96 (CI: 0.59–1.58)	No interaction with smoking, vitamin C
Ward, 2006 USA [35]	Population-based case-control Incidence, 1998–2000 Iowa	Nitrate levels in PWS among those with nitrate estimates for ≥70% of person-years ≥1960 (181 case, 142 controls); nitrate measurements for private well users at time of interviews (1998–2000; 54 cases, 44 controls)	Non-Hodgkin lymphoma	Private wells: >5.0 mg/L NO_3-N vs. ND OR = 0.8 (CI 0.2–2.5) PWS average: ≥2.9 mg/L NO_3-N vs. <0.63 OR = 1.2 (CI 0.6–2.2) Years ≥5mg/L NO3-N: 10+ vs. 0 OR = 1.4 (CI: 0.7–2.9)	No interaction with vitamin C, smoking

Table 2. *Cont.*

First Author (Year) Country	Study Design, Years Regional Description	Exposure Description	Cancer Sites Included	Summary of Drinking-Water Findings [a,b]	Evaluation of Effect Modification [c]
Inoue-Choi, 2015 USA [129]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2010 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence; PWS measurements for nitrate and TTHM; no measurements for private wells (17,216 women; 190 ovarian cases)	Ovary	Nitrate and TTHM metrics computed for reported duration at water source (11+ years) Highest vs. lowest quartile PWS average (≥2.98 mg/L vs. <0.47 mg/L NO_3-N) HR = 2.03 (CI = 1.22–3.38; *p*-trend = 0.003) Years >5 mg/L (≥4 years vs. 0) HR = 1.52 (CI: 1.00–2.31; *p*-trend = 0.05) Private well users (vs. <0.47 mg/L NO_3-N in PWS) HR = 1.53 (CI: 0.93–2.54)	No interaction with vitamin C, red meat intake, smoking for PWS nitrate Interaction with private well use and vitamin C intake (*p*-interaction = 0.01)
Quist, 2018 USA [126]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2011 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence; nitrate and TTHM estimates for PWS (20,945 women; 189 pancreas cases); no measurements for private wells Adjusted for TTHM (1955–1988), measured levels in 1980s, prior year levels estimated by expert)	Pancreas	Nitrate and TTHM metrics computed for reported duration at water source (11+ years) 95th percentile vs. lowest quartile PWS average (≥5.69 vs. <0.47 mg/L NO_3-N) HR = 1.16 (CI: 0.51–2.64; *p*-trend = 0.97) Years >5 mg/L (≥4 years vs. 0) HR = 0.90 (CI: 0.55–1.48; *p*-trend = 0.62) Private well users (vs. <0.47 mg/L NO_3-N) HR = 0.92 (CI: 0.55–1.52)	No interaction with smoking, vitamin C
Ward, 2008 USA [133]	Population-based case control Incidence, 1988–1993 Nebraska	Controls from prior study of lymphohematopoetic cases and controls interviewed in 1992–1994; Proxy interviews for 80%, 76%, 61% of stomach, esophagus, controls, respectively. Nitrate levels (1965–1985) in PWS for ≥70% of person-years (79 distal stomach, 84, esophagus, 321 controls); Private well users sampling at interview (15 stomach, 22 esophagus, 44 controls)	Stomach and esophagus (adenocarcinomas)	Highest vs. lowest quartile PWS average (>4.32 vs. <2.45 mg/L NO_3-N): stomach OR = 1.2 (CI 0.5–2.7); esophagus OR = 1.3 (CI: 0.6–3.1); Years >10 mg/L NO_3-N (9+ vs. 0): stomach OR = 1.1 (CI: 0.5–2.3); esophagus OR = 1.2 (CI: 0.6–2.7) Private well users (>4.5 mg/L NO_3-N vs. <0.5) stomach OR = 5.1 (CI: 0.5–52; 4 cases, 13 controls); esophagus OR = 0.5 (CI: 0.1–2.9; 8 cases; 13 controls)	No interaction with vitamin C, processed meat, or red meat for either cancer
Ward, 2010 USA [37]	Population-based cohort of postmenopausal women ages 55–69 Incidence, 1986–2004 Iowa	Nitrate levels in PWS (1955–1988) and private well use among women >10 years at enrollment residence (21,977 women; 40 thyroid cases); no measurements for private wells	Thyroid	Highest vs. lowest quartile PWS average (>2.46 vs. <0.36 mg/L NO_3-N) HR = 2.18 (CI: 0.83–5.76; *p*-trend = 0.02) Years >5 mg/L (≥5 years vs. 0) HR = 2.59 (CI: 1.09–6.19; *p*-trend = 0.04); Private well (vs. <0.36 mg/L NO_3-N on PWS) HR = 1.13 (CI: 0.83–3.66) Dietary nitrate intake quartiles positively associated with risk (*p*-trend = 0.05)	No interaction with smoking, vitamin C, body mass index, education, residence location (farm/rural vs. urban)

ND = not detected; PWS = public water supplies; [a] nitrate or nitrite levels presented in the publications as mg/L of the ion were converted to mg/L as NO_3-N or NO_2-N; [b] Odds ratios (OR) for case-control studies, incidence rate ratios (RR) and hazard ratios (HR) for cohort studies, and 95% confidence intervals (CI); [c] Factors evaluated are noted. Interaction refers to reported $p \leq 0.10$ from test of heterogeneity.

Animal studies demonstrate that in utero exposure to nitrosamides can cause brain tumors in the exposed offspring. Water nitrate and nitrite intake during pregnancy was estimated in a multi-center case-control study of childhood brain tumors in five countries based on the maternal residential water source [139]. Results for the California and Washington State sites were reported in our previous review [8,140]. Nitrate/nitrite levels in water supplies were measured using a nitrate test strip method in four countries including these U.S. sites; most of these measurements occurred many years after the pregnancy. Measured nitrate concentrations were not associated with risk of childhood brain tumors. However, higher nitrite levels (>1.5 mg/L NO_2-N) in the drinking water were associated with increased risk of astrocytomas.

8. Thyroid Disease

Animal studies demonstrate that ingestion of nitrate at high doses can competitively inhibit iodine uptake and induce hypertrophy of the thyroid gland [141]. An early study of women in the Netherlands consuming water with nitrate levels at or above the MCL, found increased prevalence of thyroid hypertrophy [142]. Since the last review, five studies have evaluated nitrate ingestion from drinking water (the Iowa cohort study also assessed diet) and prevalence of thyroid disease. A study of school-age children in Slovakia found increased prevalence of subclinical hypothyroidism among children in an area with high nitrate levels (51–274 mg/L NO_3) in water supplies compared with children ingesting water with nitrate ≤50 mg/L (11 mg/L NO_3-N). In Bulgarian villages with high nitrate levels (75 mg/L NO_3) and low nitrate levels (8 mg/L), clinical examinations of the thyroids of pregnant women and school children revealed an approximately four- and three-fold increased prevalence of goiter, respectively, in the high nitrate village [143,144]. The iodine status of the populations in both studies was adequate. Self-reported hypothyroidism and hyperthyroidism among a cohort of post-menopausal women in Iowa was not associated with average nitrate concentrations in PWS [37]. However, dietary nitrate, the predominant source of intake, was associated with increased prevalence of hypothyroidism but not hyperthyroidism. Modeled estimates of nitrate concentrations in private wells among a cohort of Old Order Amish in Pennsylvania (USA) were associated with increased prevalence of subclinical hypothyroidism as determined by thyroid stimulating hormone measurements, among women but not men [145].

9. Other Health Effects

Associations between nitrate in drinking water and other non-cancer health effects, including type 1 childhood diabetes (T1D), blood pressure, and acute respiratory tract infections in children were previously reviewed [8]. Since 2004, a small number of studies have contributed additional mixed evidence for these associations. Animal studies indicate that NOC may play a role in the pathology of T1D through damage to pancreatic beta cells [146]. A registry-based study in Finland [147] found a positive trend in T1D incidence with levels of nitrate in drinking water. In contrast, an ecological analysis in Italy showed an inverse correlation with water nitrate levels and T1D rates [148]. A small T1D case-control study in Canada with 57 cases showed no association between T1D and estimated intake of nitrate from drinking water (highest quartile >2.7 mg/day NO_3-N) [149]. Concentrations of nitrate in drinking water (median ~2.1 mg/L NO_3-N) were not associated with progression to T1D in a German nested case-control study of islet autoantibody-positive children, who may be at increased risk of the disease [150].

In a prospective, population-based cohort study in Wisconsin (USA), increased incidence of early and late age-related macular degeneration was positively associated with higher nitrate levels (≥5 mg/L vs. <5 mg/L NO_3-N) in rural private drinking water supplies [151]. The authors suggested several possible mechanisms, including methemoglobin-induced lipid peroxidation in the retina.

Potential benefits of nitrate ingestion include lowering of blood pressure due to production of nitric oxide in the acidic stomach and subsequent vasodilation, antithrombotic, and immunoregulatory effects [152]. Experimental studies in animals and controlled feeding studies in humans have

demonstrated mixed evidence of these effects and on other cardiovascular endpoints such as vascular hypertrophy, heart failure, and myocardial infarction (e.g., [152–154]). Ingested nitrite from diet has also been associated with increased blood flow in certain parts of the brain [155]. Epidemiologic studies of these effects are limited to estimation of dietary exposures or biomarkers that integrate exposures from nitrate from diet and drinking water. Recent findings in the Framingham Offspring Study suggested that plasma nitrate was associated with increased overall risk of death that attenuated when adjusted for glomerular function (HR: 1.16, 95% CI: 1.0–1.35) but no association was observed for incident cardiovascular disease [156]. No epidemiologic studies have specifically evaluated nitrate ingested from drinking water in relation to these outcomes. Another potential beneficial effect of nitrate is protection against bacterial infections via its reduction to nitrite by enteric bacteria. In an experimental inflammatory bowel disease mouse model, nitrite in drinking water was associated with both preventive and therapeutic effects [157]. However, there is limited epidemiologic evidence for a reduced risk of gastrointestinal disease in populations with high drinking water nitrate intake. One small, cross-sectional study in Iran found no association between nitrate levels in public water supplies with mean levels of ~5.6 mg/L NO_3-N and gastrointestinal disease [158].

10. Discussion

Since our last review of studies through 2004 [8], more than 30 epidemiologic studies have evaluated drinking water nitrate and risk of cancer, adverse reproductive outcomes, or thyroid disease. However, the number of studies of any one outcome was not large and there are still too few studies to allow firm conclusions about risk. The most common endpoints studied were colorectal cancer, bladder, and breast cancer (three studies each) and thyroid disease (four studies). Considering all studies to date, the strongest evidence for a relationship between drinking water nitrate ingestion and adverse health outcomes (besides methemoglobinemia) is for colorectal cancer, thyroid disease, and neural tube defects. Four of the five published studies of colorectal cancer found evidence of an increased risk of colorectal cancer or colon cancer associated with water nitrate levels that were mostly below the respective regulatory limits [32,134,135,159]. In one of the four positive studies [159], increased risk was only observed in subgroups likely to have increased nitrosation. Four of the five studies of thyroid disease found evidence for an increased prevalence of subclinical hypothyroidism with higher ingestion of drinking water nitrate among children, pregnant women, or women only [37,144,145,160]. Positive associations with drinking water nitrate were observed at nitrate concentrations close to or above the MCL. The fifth study, a cohort of post-menopausal women in Iowa, had lower drinking water nitrate exposure but observed a positive association with dietary nitrate [37]. To date, five of six studies of neural tube defects showed increased risk with exposure to drinking water nitrate below the MCL. Thus, the evidence continues to accumulate that higher nitrate intake during the pregnancy is a risk factor for this group of birth defects.

All but one of the 17 cancer studies conducted since 2004 were in the U.S. or Europe, the majority of which were investigations of nitrate in regulated public drinking water. Thyroid cancer was studied for the first time [37] with a positive finding that should be evaluated in future studies. Bladder cancer, a site for which other drinking water contaminants (arsenic, disinfection by-products [DBPs]) are established or suspected risk factors, was not associated with drinking water nitrate in three of the four studies. Most of the cancer studies since 2004 evaluated effect modification by factors known to influence endogenous nitrosation, although few observed evidence for these effects. Several studies of adverse reproductive outcomes since 2004 have indicated a positive association between maternal prenatal exposure to nitrate concentrations below the MCL and low birth weight and small for gestational age births. However, most studies did not account for co-exposure to other water contaminants, nor did they adjust for potential risk factors. The relation between drinking water nitrate and spontaneous abortion continues to be understudied. Few cases of methemoglobinemia, the health concern that lead to the regulation of nitrate in public water supplies, have been reported in the U.S. since the 1990s. However, as described by Knobeloch et al. [96], cases may be underreported

and only a small proportion of cases are thoroughly investigated and described in the literature. Based on published reports, [100] areas of the world of particular concern include several eastern European countries, Gaza, and Morocco, where high nitrate concentrations in water supplies have been linked to high levels of methemoglobin in children. Therefore, continued surveillance and education of physicians and parents will be important. Biological plausibility exists for relationships between nitrate ingestion from drinking water and a few other health outcomes including diabetes and beneficial effects on the cardiovascular system, but there have been only a limited number of epidemiologic studies.

Assessment of drinking water nitrate exposures in future studies should be improved by obtaining drinking water sources at home and at work, estimating the amount of water consumed from each source, and collecting information on water filtration systems that may impact exposure. These efforts are important for reducing misclassification of exposure. Since our last review, an additional decade of PWS monitoring data are available in the U.S. and European countries, which has allowed assessment of exposure over a substantial proportion of participants' lifetimes in recent studies. Future studies should estimate exposure to multiple water contaminants as has been done in recent cancer studies [31,33,127,129]. For instance, nitrate and atrazine frequently occur together in drinking water in agricultural areas [161] and animal studies have found this mixture to be teratogenic [162]. Regulatory monitoring data for pesticides in PWS has been available for over 20 years in the U.S.; therefore, it is now feasible to evaluate co-exposure to these contaminants. Additionally, water supplies in agricultural areas that rely on alluvial aquifers or surface water often have elevated levels of both DBPs and nitrate. Under this exposure scenario, there is the possibility of formation of the nitrogenated DBPs including the carcinogenic NDMA, especially if chloramination treatment is used for disinfection [163,164]. Studies of health effects in countries outside the U.S. and Europe are also needed.

A comprehensive assessment of nitrate and nitrite from drinking water and dietary sources as well as estimation of intakes of antioxidants and other inhibitors of endogenous nitrosation including dietary polyphenols and flavonoids is needed in future studies. Heme iron from red meat, which increases fecal NOC in human feeding studies, should also be assessed as a potential effect modifier of risk from nitrate ingestion. More research is needed on the potential interaction of nitrate ingestion and nitrosatable drugs (those with secondary and tertiary amines or amides). Evidence from several studies of birth defects [38,118–120] implicates nitrosatable drug intake during pregnancy as a risk factor for specific congenital anomalies especially in combination with nitrate. Drugs with nitrosatable groups include many over-the-counter and prescription drugs. Future studies with electronic medical records and record-linkage studies in countries like Denmark with national pharmacy data may provide opportunities for evaluation of these exposures.

Populations with the highest exposure to nitrate from their drinking water are those living in agricultural regions, especially those drinking water from shallow wells near nitrogen sources (e.g., crop fields, animal feeding operations). Estimating exposure for private well users is important because it allows assessment of risk over a greater range of nitrate exposures compared to studies focusing solely on populations using PWS. Future health studies should focus on these populations, many of which may have been exposed to elevated nitrate in drinking water from early childhood into adulthood. A major challenge in conducting studies in these regions is the high prevalence of private well use with limited nitrate measurement data for exposure assessment. Recent efforts to model nitrate concentrations in private wells have shown that it is feasible to develop predictive models where sufficient measurement data are available [41,48,49]. However, predictive models from one area are not likely to be directly translatable to other geographic regions with different aquifers, soils, and nitrogen inputs.

Controlled human feeding studies have demonstrated that endogenous nitrosation occurs after ingestion of drinking water with nitrate concentrations above the MCL of 10 mg/L NO_3-N (~44 mg/L as NO_3). However, the extent of NOC formation after ingestion of drinking water with nitrate

concentrations below the MCL has not been well characterized. Increased risks of specific cancers and central nervous system birth defects in study populations consuming nitrate below the MCL is indirect evidence that nitrate ingestion at these levels may be a risk factor under some conditions. However, confounding by other exposures or risk factors can be difficult to rule out in many studies. Controlled human studies to evaluate endogenous nitrosation at levels below the MCL are needed to understand interindividual variability and factors that affect endogenous nitrosation at drinking water nitrate levels below the MCL.

A key step in the endogenous formation of NOC is the reduction of nitrate, which has been transported from the bloodstream into the saliva, to nitrite by the nitrate-reducing bacteria that are located primarily in the crypts on the back of the tongue [165–167]. Tools for measuring bacterial DNA and characterizing the oral microbiome are now available and are currently being incorporated into epidemiologic studies [168,169]. Buccal cell samples that have been collected in epidemiologic studies can be used to characterize the oral microbiome and to determine the relative abundance of the nitrate-reducing bacteria. Studies are needed to characterize the stability of the nitrate-reducing capacity of the oral microbiome over time and to determine factors that may modify this capacity such as diet, oral hygiene, and periodontal disease. Interindividual variability in the oral nitrate-reducing bacteria may play an important role in modifying endogenous NOC formation. The quantification of an individual's nitrate-reducing bacteria in future epidemiologic studies is likely to improve our ability to classify participants by their intrinsic capacity for endogenous nitrosation.

In addition to characterizing the oral microbiome, future epidemiologic studies should incorporate biomarkers of NOC (e.g., urinary or fecal NOC), markers of genetic damage, and determine genetic variability in NOC metabolism. As many NOC require α-hydroxylation by CYP2E1 for bioactivation and for formation of DNA adducts, it is important to investigate the influence of polymorphisms in the gene encoding for this enzyme. Studies are also needed among populations with medical conditions that increase nitrosation such as patients with inflammatory bowel disease and periodontal disease [8]. Because NOC exposures induce characteristic gene expression profiles [170,171], further studies linking drinking water intake to NOC excretion and gene expression responses are relevant to our understanding of health risks associated with drinking water nitrate. The field of 'Exposome-research' [172,173] generates large numbers of genomics profiles in human population studies for which dietary exposures and biobank materials are also available. These studies provide opportunities to measure urinary levels of nitrate and NOC that could be associated with molecular markers of exposure and disease risk.

Nitrate concentrations in global water supplies are likely to increase in the future due to population growth, increases in nitrogen fertilizer use, and increasing intensity and concentration of animal agriculture. Even with increased inputs, mitigation of nitrate concentrations in water resources is possible through local, national, and global efforts. Examples of the latter are the International Nitrogen Initiative [174] and the EU Nitrates Directive [17,18], which aim to quantify human effects on the nitrogen cycle and to validate and promote methods for sustainable nitrogen management. Evidence for the effectiveness of these efforts, which include the identification of vulnerable areas, establishment of codes of good agricultural practices, and national monitoring and reporting are indicated by decreasing trends in groundwater nitrate concentrations in some European countries after the implementation of the EU Nitrates Directive [19]. However, the effect of this initiative was variable across the EU. In the U.S., nitrogen applications to crop fields are not regulated and efforts to reduce nitrogen runoff are voluntary. Although strategies such as appropriate timing of fertilizer applications, diversified crop rotations, planting of cover crops, and reduced tillage can be effective [175], concentrations in U.S. ground and surface water have continued to increase in most areas [10]. Climate change is expected to affect nitrogen in aquatic ecosystems and groundwater through alterations of the hydrological cycle [176]. Climatic factors that affect nitrate in groundwater include the amount, intensity, and timing of precipitation. Increasing rainfall intensity, especially in

the winter and spring, can lead to increases in nitrogen runoff from agricultural fields and leaching to groundwater.

11. Conclusions

In summary, most adverse health effects related to drinking water nitrate are likely due to a combination of high nitrate ingestion and factors that increase endogenous nitrosation. Some of the recent studies of cancer and some birth defects have been able to identify subgroups of the population likely to have greater potential for endogenous nitrosation. However, direct methods of assessing these individuals are needed. New methods for quantifying the nitrate-reducing bacteria in the oral microbiome and characterizing genetic variation in NOC metabolism hold promise for identifying high risk groups in epidemiologic studies.

To date, the number of well-designed studies of individual health outcomes is still too few to draw firm conclusions about risk from drinking water nitrate ingestion. Additional studies that incorporate improved exposure assessment for populations on PWS, measured or predicted exposure for private well users, quantification of nitrate-reducing bacteria, and estimates of dietary and other factors affecting nitrosation are needed. Studies of colorectal cancer, thyroid disease, and central nervous system birth defects, which show the most consistent associations with water nitrate ingestion, will be particularly useful for clarifying these risks. Future studies of other health effects with more limited evidence of increased risk are also needed including cancers of the thyroid, ovary, and kidney, and the adverse reproductive outcomes of spontaneous abortion, preterm birth, and small for gestational age births.

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Thyroid Cancer Induction: Nitrates as Independent Risk Factors or Risk Modulators after Radiation Exposure, with a Focus on the Chernobyl Accident

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Thyroid cancer · Nitrate pollution · Ionizing radiation · Chernobyl · Salivary glands · Risk stratification · Belarus

Abstract
In recent decades, differentiated thyroid cancer (DTC) incidence has been increasing worldwide. The important contributions to this phenomenon of "overdiagnosis" driven by wider use of improved ultrasound systems are amply documented, notwithstanding the "real" carcinogenic effects of ionizing radiation, e.g., from the Chernobyl accident or health care interventions. Less well understood is the role of nitrates – as environmental pollutants, in diet, and in medication – in thyroid carcinogenesis. Increasing exposure to nitrates is associated with rising incidence of esophageal, stomach, bladder, and colon cancers. Recent data suggest that in agricultural areas with higher mean nitrate levels in groundwater, DTC risk is also elevated. Our work in Belarus after Chernobyl has shown that children in districts with high nitrate concentrations in drinking water had significantly higher thyroid cancer incidence after irradiation than did their counterparts in areas with lower nitrate concentrations. Notwithstanding thyroid shielding, increasing use of computed tomography and dental X-rays heightens radiation exposure of the salivary glands in the general population, especially in children and adolescents. When nitrate intake is increased, salivary gland irradiation may potentially result in carcinogenic elevations in plasma nitric oxide concentrations. In conclusion, excess nitrate intake seems to be an independent risk factor for DTC. Additionally, we hypothesize from our data that high nitrate levels modulate the carcinogenic effect of radiation on the thyroid. Cohort studies, case-control studies, or both, are needed to quantify the effects of nitrates on DTC risk in the presence or absence of radiation exposure, e.g., that associated with diagnostic or therapeutic health care interventions.

Introduction

During the last 30 years, the incidence of differentiated thyroid cancer (DTC) has steadily increased worldwide, most markedly in France, Italy, the Republic of Korea, Australia, and the USA [1–8]. In the USA, DTC incidence

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is rising more rapidly than that of any other malignancy except liver cancer [1], with the annual percent change (both genders) increasing from 2.4% in 1980–1997 to 6.6% in 1997–2009 [2, 8]. In Europe, the increase in the last 2–3 decades has ranged from 5.3% (Switzerland) to 155.6% (France).

In Belarus and Ukraine, DTC incidence has also substantially increased in the past 25 years; there is strong evidence that this increase was mainly due to radiation exposure of children and adolescents after the Chernobyl accident [9–11]. Nonetheless, an appreciable proportion of thyroid cancers diagnosed in young people in these countries may be related to screening or other confounders [10, 12].

The main factors contributing to the worldwide increase in the incidence of DTC continue to be debated. Today, it is generally accepted that the widespread use of ultrasound, introduced in the 1980s to diagnose structural thyroid diseases, has led to earlier, more frequent detection of this neoplasm. Better diagnostics were estimated to account for 60% or more of DTC diagnoses in 2003–2007 in women under the age of 80 years in France, Italy, the USA, Australia, and the Republic of Korea, and 30% or more in other very high-income countries [13]. Consistent with these estimates, small thyroid cancers that are best discovered using new technologies (ultrasound and fine-needle aspiration biopsy) have shown a sharply increased incidence [14]. However, improved medical surveillance and "overdiagnosis" do not completely explain the rise in rates of papillary thyroid carcinoma (PTC), since a significant increase also has been observed for larger tumors (>10 mm) [15–18].

DTC and Radiation Exposure

Also well accepted as an explanation for greater frequency of DTC diagnoses is radiation exposure. External radiotherapy in childhood for cancer, tinea capitis, or an enlarged thymus or tonsils has been long known to be associated with an elevated risk of DTC [19–21]. Additionally, DTC was the first solid tumor to be found in excess among atomic bomb survivors in Japan [22]. An updated pooled analysis of 12 studies [23] identified a consistent risk model across the full range of external radiation doses to the thyroid, with relative risk (RR) increasing approximately supralinearly through 2–4 Gy, and then leveling and declining above approximately 30 Gy, although RRs remained elevated. Radiogenic effects occurred for both PTC and nonpapillary thyroid tumors. For doses >0.10 Gy, RRs increased significantly with dose ($p < 0.01$), with no significant departure from linearity. The excess relative risk (ERR) estimate per Gy was significant within 10 years of radiation exposure at 2.76 (95% CI: 0.94–4.98), and remained elevated 50 years and more after exposure [23].

In several other studies, dental radiography was associated with an increased risk of thyroid cancer [24, 25] and parotid gland tumors [26]. One case-control study [24] found a significant association between self-reported dental X-ray exposure, particularly multiple exposures, and DTC risk (odds ratio [OR]: 2.1, 95% CI: 1.4–3.1, $p < 0.001$) with a dose-response pattern ($p < 0.0001$ for trend). American Dental Association recommendations stress the need to shield the thyroid during dental X-ray examination [27].

Pediatric DTC rates in Belarus began to increase as early as 4 years after the Chernobyl accident [28, 29]. Ecological studies of DTC incidence in Belarus and Ukraine following Chernobyl estimated a linear ERR per Gy of 18.9 and excess absolute risk per Gy of 2.7 [9]. Cohort studies with measurement-based individual thyroid dose estimates reported ERRs per Gy of 5.3 and 2.2 for DTC in Ukraine and Belarus [11, 30–35], respectively.

Nitrates as Pollutants, in Diet, and in Medication

Beyond diagnostic activity and radiation exposure, additional factors may contribute to increased DTC incidence, and require further investigation. In particular, nutritional exposure to chemical pollutants such as nitrates in drinking water, specifically during intrauterine life and early childhood, might affect thyroid cell propensity to mutagenesis. In general, there are five primary sources of exposure to nitrate and its metabolite nitrite: environmental/atmospheric exposure to nitric oxide (NO) and nitrogen, dietary exposure to nitrate and nitrite in food and in drinking water, and endogenous production of NO and swallowing of nitrate-rich saliva [36, 37].

The largest proportion of reactive nitrogen, i.e., NO, nitrogen dioxide, nitric acid, nitrous oxide, nitrite, nitrate, ammonia, nitrogen oxides, and organic compounds such as urea, amines, proteins, and nucleic acids, in the environment comes from agriculture in the form of fertilizers and animal waste [38, 39]. The past 60 years have witnessed an exponential increase in the use of nitrogen-rich manure and reactive nitrogen as fertilizers [40]. Although they boost agricultural productivity, nitrogen-

rich fertilizers let nitrates seep through the soil into both groundwater and surface water. There, these substances can accumulate for years until the concentration is adverse to human health. Because of water pollution, high amounts of nitrate might be present in fruits and vegetables, specifically those grown in greenhouses. Additionally, high nitrate levels may be found in cured and processed meats due to the addition of these chemicals as preservatives or color enhancers. Medications, including antidiarrheals, diuretics, vasodilators, and the cytotoxic chemotherapy agent nitrosourea, also contribute to nitrate exposure in humans [39, 41].

In Belarus, between 1960 and 1990, mean use of nitrogen fertilizers increased more than 20-fold, from 4 to 92 kg/hectare, while the average nitrate concentration in groundwater rose almost 40-fold, from 1.1 to 41.6 mg/L [42]. Groundwater from open wells is the main source of drinking water in rural Belarus. According to the Belarusian Ministry of Health, about 1% of pipeline water samples have nitrate concentration exceeding the World Health Organization (WHO)-recommended maximum contaminant level of 45 mg/L [43]. In contrast, about 40% of water samples from open wells exceed that maximum contaminant level. In Brest and Gomel Oblasts, the proportion of such samples reaches 40–60%, while in Mogilev Oblast, it is about 20% [43].

Physiology and Pathophysiology of Nitrates

In the past 30 years, the roles of NO in physiology and pathophysiology have been extensively studied. Nitrate is metabolized by the nitrate-nitrite-NO pathway. As a gas (in the pure state and under standard temperature and pressure conditions) with an unshared electron, NO participates in various biological processes. In the body, under normal oxygen pressure, NO is produced by NO synthetase from L-arginine. In hypoxia, nitrite is reduced by a variety of reductases, including deoxyhemoglobin, to produce NO. Further reduction/oxidation of NO can lead to metabolite production (nitrogen dioxide, nitrate) [44].

Nitrate and NO are known to affect the iodine metabolism of the thyroid. Nitrate is a competitive inhibitor of the sodium-iodine symporter and prevents iodide uptake by the gland. Thyroid hormone synthesis is thereby compromised, leading to thyrotropin elevation. The resultant chronic thyroid stimulation can lead to proliferative changes, including hypertrophy and hyperplasia as well as neoplasia [45–47].

There are other mechanisms by which ingested nitrate may produce detrimental effects on health. One is through formation of methemoglobin, which inhibits the oxygen-carrying capacity of blood; another is through endogenous formation of N-nitroso compounds that may act as carcinogens [41, 46]. Nitrosamine synthesis depends on temperature and pH, and may be stimulated by low-level gamma radiation [48–50].

The salivary glands play a very important role in the metabolism of nitrate and the nitrate-nitrite-NO pathway [36, 37]. Dietary nitrate is rapidly completely absorbed in the upper gastrointestinal tract. Sixty percent of ingested nitrate is excreted in the urine within 48 h [36, 37]. However, approximately 25% of circulating nitrate is taken up by the salivary glands and secreted into the mouth in saliva. Salivary nitrate concentrations are 10- to 20-fold above blood levels, and may reach several millimolars. Oral facultative anaerobic bacteria, residing mainly in the tongue's crypts, then reduce nitrate to nitrite and NO via nitrate-reducing enzymes. This relatively effective process results in nitrite levels that are 1,000-fold higher in saliva than in plasma.

Nitrates and Radiotherapy

Therapeutic irradiation increases NO levels in salivary gland tissue. NO produced in irradiated tissues mediates cellular regulation through posttranslational modification of a number of proteins [44]. Evidence exists for the role of NO as an intrinsic radiosensitizer [51]. On the other hand, administration of an NO synthesis inhibitor ameliorated the dysfunction of irradiated salivary glands, indicating that NO helps mediate the dry mouth symptoms occurring after irradiation [52].

Radiation-induced bystander effects may be modulated by NO [53–56]. NO synthase activation and NO overproduction after exposure to ionizing radiation not only affect bystander cells with activated NO synthase, but also can stimulate specific cell-signaling mechanisms. These NO-dependent effects include the promotion of genomic instability and the accumulation of DNA reduplication errors in bystander cells, without the direct DNA damage seen in irradiated cells. Hydrophobic properties of NO, permitting the diffusion of the substance through the cytoplasm and plasma membranes, allow this signaling molecule to easily spread from irradiated cells to bystander cells without the involvement of gap-junctional intercellular communication [56].

Nitrate Carcinogenicity: Relation to DTC

The first report of negative health effects of nitrate, namely, methemoglobin formation, was in 1945, after observation of cyanosis in infants in Iowa, USA [57]. Long-term exposure to nitrate and nitrite has been evaluated in relation to multiple tumor types; positive associations were reported for cancers of the esophagus, stomach, bladder, and colon [41, 50].

NO as a carcinogen heavily depends on concentration in a nonlinear manner: the specific activity of this analyte at very low levels blocks tumor growth, while moderate concentrations promote tumor angiogenesis and cell survival via lymphocyte suppression [44]. High NO levels may induce chromosomal breaks directly, or indirectly by inhibiting DNA repair activities [58]. NO can cause irreversible injury to several fundamental cancer control genes. The substance plus superoxide rapidly react to form peroxynitrite, which can cause oxidative damage to DNA. NO can also block DNA synthesis through inhibition of ribonucleotide reductase, the rate-limiting enzyme in DNA production [58–60]. Additionally, NO can directly inhibit enzymes in the mitochondrial electron transport chain or act indirectly by interfering with DNA repair mechanisms, leaving the cell susceptible to other DNA-damaging agents [59]. NO has been shown to have a role in stimulating vascular endothelial growth factor-D (VEGF-D) expression in vitro [61]. The formation of the NO biomarker, nitrotyrosine, was also correlated with VEGF-D expression in human PTC. In that setting, NO may induce lymph node metastasis via VEGF-D stimulation. In vitro, NO has both genotoxic and metastasis-promoting properties. Increased NO generation in cancer cells may contribute to tumor hemangiogenesis or lymphangiogenesis by upregulating VEGF-D [61]. The effects of NO are mediated in part by its metabolites, such as peroxynitrite. Data suggest that NO stimulates CXC chemokine receptor 4 (CXCR4) expression in vitro [62]. Nitrotyrosine formation was also correlated with CXCR4 expression and lymph node metastasis in human PTC [62].

Regarding DTC, Ward et al. [63] found an increased risk of this neoplasm in agricultural areas with higher mean nitrate levels in public water supplies and with longer-term consumption of water with nitrate-N concentrations exceeding 5 mg/L (subjects with ≤5 years' consumption at levels of >5 mg/L, RR: 2.6, 95% CI: 1.1–6.2). Increased dietary nitrate intake was associated with a heightened risk of DTC (RR: 2.9, 95% CI: 1.0–8.1, $p = 0.046$) and with the prevalence of hypothyroidism (OR: 1.2, 95% CI: 1.1–1.4), but not hyperthyroidism [63].

With regard to thyroid radiation dose, de Vathaire et al. [64] investigated potential modifiers of the thyroid dose response to radiation therapy in survivors of pediatric solid tumors other than thyroid cancer. The risk of DTC as a second primary malignancy increased with a thyroid dose of up to 10 Gy, then leveled off for higher doses. The excess RR per Gy of radiation to the thyroid was 4.7 (95% CI: 1.7–22.6). Patients also receiving nitrosourea chemotherapy had a 6.6-fold (95% CI: 2.5–15.7-fold) higher risk than those who did not.

Exposure to Nitrates and Radiation after Chernobyl

Recently published data suggest a synergistic influence of nitrates in drinking water and the thyroid radiation dose on the incidence of childhood DTC in Belarus after the Chernobyl accident [12]. The highest mean thyroid dose (320 mGy) and the highest incidence of pediatric thyroid cancer in 1986–2005 (11 per 100,000 patient-years) was found in the most contaminated area, Gomel Oblast [12]. However, there was a notable exception to the general dose-incidence rate pattern, i.e., substantial difference in rates of pediatric thyroid cancer in Mogilev versus Brest Oblasts (1.50 vs. 5.51 per 100,000 patient-years). Whereas the estimated thyroid doses from iodine-131 were comparable in the two regions (65 vs. 51 mGy), nitrate contamination of drinking water significantly differed (mean levels in open well water, 40 vs. 185 mg/L). Radiation dose was significantly associated with thyroid cancer incidence ($p = 0.029$), but the effect of radiation significantly varied according to the nitrate concentration in drinking water ($p = 0.004$). A plausible interpretation of these observations is that the radiation effect on thyroid cells might be modified by patients' ingestion of nitrate from drinking water [12]. Comparison of maps respectively showing levels of groundwater pollution by nitrates (Fig. 1a) and Chernobyl-related radiation doses (Fig. 1b) also suggests that both factors may influence DTC risk.

Conclusions

Based on our own experience and on published data, we hypothesize that thyroid cancer may be induced by coincidence of several conditions: (1) excessive nitrate uptake via drinking water increases nitrite production,

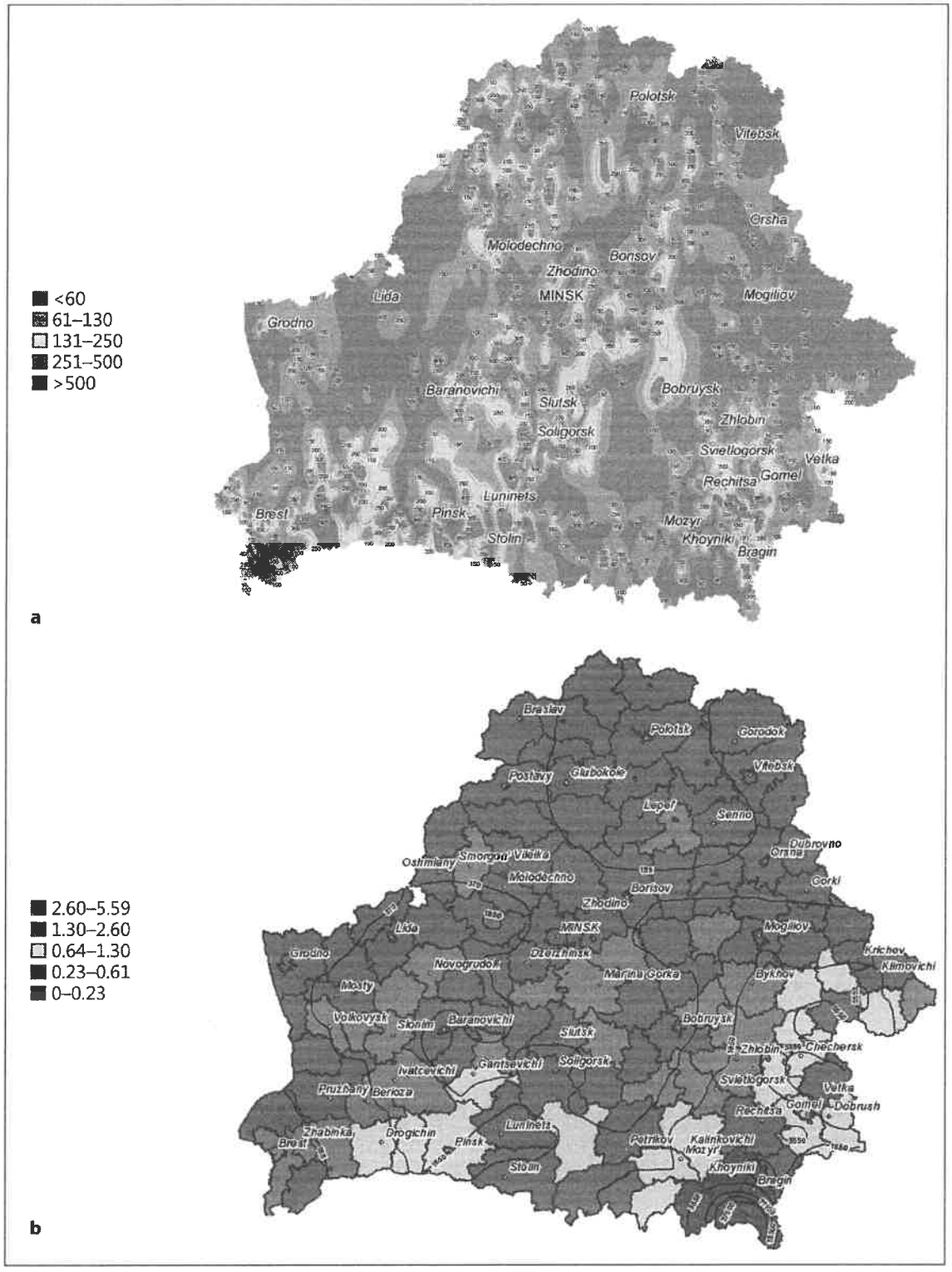

Fig. 1. Maps of Belarus depicting by district (oblast) the level of groundwater pollution with nitrate (mg/L) measured in open wells in 1988–1990 (**a**) and the prevalence (per 1,000) of pediatric thyroid cancer in 1986–2005 in the cohort ages 0–18 years at exposure to radioactive fallout from the Chernobyl accident (**b**). In **b**, areas exposed to such fallout are bounded with black lines, and radioactive contamination due to radioiodine in 1986 (in kBq/m^2) is shown in small numerals. Notably, areas of greatest pediatric thyroid cancer prevalence tend to coincide with areas characterized by both a high radiation exposure and high nitrate pollution of groundwater.

which leads to the development of hypoxia in the blood, especially in children, and to overproduction of NO, which is carcinogenic per se; (2) radiation exposure of the salivary glands, e.g., by dental X-ray examination, may also lead to increased plasma levels of NO; and (3) if one or both of these processes coincide with radiation exposure of the thyroid, the considerably increased NO concentrations in the body presumably enhance the carcinogenic effect of the radiation.

The role of radiation in thyroid carcinogenesis is well documented. The influence of other factors and confounders and their synergistic effects is less well understood. Studies of radiation-induced DTC in Belarus after the Chernobyl accident have shown that children living in areas with high nitrate concentration in drinking water have a significantly elevated thyroid cancer risk. A plausible interpretation is that the radiation effect might be modulated by ingested nitrates. Further cohort studies or case-control studies with individual exposure estimates are required to quantify the effect of nitrate on DTC risk in the context of growing use of medical radiation for diagnostic and therapeutic purposes. Such studies examining the increasing exposure to nitrates alone and in combination with ionizing radiation may provide a better understanding of the considerable increase in thyroid cancer incidence in many countries.

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The authors have no conflicts of interest to disclose.

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NITRATE TOXICITY AND DRINKING WATER STANDARDS – A REVIEW

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Abstract. The current US EPA maximum contaminant level (MCL) for public drinking water supplies and the health advisory level (HAL) for other private water supplies is 10 mg/L, expressed as NO_3-N. Unlike other drinking water standards, the nitrate standard has no safety factor, which typically is about a 10-fold safety factor to account for differences in human susceptibility. Guidance, action, or advisory levels for nitrate in drinking water are lower in several countries, including Germany (4.4 mg/L), South Africa (4.4 mg/L), and Denmark (5.6 mg/L). Clearly health and regulatory officials in other countries believe that the current WHO and USA drinking water standard for nitrate is not adequate to protect their most susceptible population from methemoglobinemia. In addition to acute methemoglobinemia, other potential health effects of nitrate exposure in drinking water include cancer, disruption of thyroid function, birth defects, and developmental disorders in children. Is the current drinking water standard in the US and World Health Organization for nitrate adequate? I think not! Clearly the most susceptible human population (infants under four months of age with existing diarrhea conditions) is not adequately protected from methemoglobinemia. In addition, recent studies suggest other possible linkages between nitrate in drinking water and adverse health consequences for adults. Particularly troublesome is the finding of a positive association between nitrate in drinking water (at levels below the USA drinking water standard) and bladder cancer and ovarian cancer in a large cohort of women in Iowa, USA. Given this framework, the regulatory authorities should establish a safety factor of two, which would reduce the current MCL and HAL for nitrate to 5.0 mg/L NO_3-N. This regulatory mandate would encourage a prudent public health strategy of limiting human nitrate exposure.
Key words: nitrate, drinking water, safety factor, methemoglobinemia

Rezumat. Nivelul maxim admis de contaminare a sistemelor publice de aprovizionare cu apă potabilă, stabilit de Agenţia de Protecţie a Mediului din SUA pentru nitraţi, ca şi nivelul maxim recomandat pentru alte sisteme particulare de apă este de 10 mg/l, exprimat în NO_3-N. Spre deosebire de alte standarde pentru apa de băut, în standardul american pentru nitrat nu s-a aplicat un **factor de siguranţă** (de regulă, cu valoarea 10) pentru a se lua în considerare diferenţele de susceptibilitate existente în populaţie. Nivelurile maxime acceptate pentru nitraţi în apa de băut sunt mai mici în unele ţări ca Germania (4,4 mg/l), Africa de Sud (4,4 mg/L) şi Danemarca (5,6 mg/L). Aceasta înseamnă că, în aceste ţări, nivelul maxim acceptat în SUA nu este considerat suficient de scăzut pentru a proteja de methemoglobinemie grupele cele mai susceptibile ale populaţiei. Pe lângă methemoglobinemie, ca efect acut, există şi alte patologii asociate expunerii la nitraţii din apă, cum sunt cancerul, unele disfuncţii tiroidiene, malformaţii congenitale şi tulburări de dezvoltare la copii. La întrebarea dacă limita admisă în SUA şi recomandată de OMS pentru nitraţi în apă, este corespunzătoare, răspunsul este negativ, fiindcă nu asigură o protecţie suficientă, astfel încât grupele cele mai susceptibile ale

populației (copii sub vârsta de 4 luni, cu factori favorizanți ai diareei) să nu facă methemoglobinemie. În plus, studii recente sugerează existența unor posibile asocieri între nitrații din apă și o serie de efecte adverse asupra sănătății adulților. Sunt relevante, în acest sens, rezultatele care indică o asociere pozitivă între nitrații din apă (în concentrații sub limita admisă în SUA) și cancere de vezică și ovariene, constatată într-un studiu de cohortă efectuat pe femei în statul Iowa, SUA. Având în vedere aceste aspecte, consider că se impune stabilirea unui factor de siguranță în valoare de 2, care ar reduce limita maximă admisă pentru nitrați la 5,0 mg/L NO_3-N. Această decizie de reglementare ar încuraja o strategie prudentă pentru limitarea expunerii populației la nitrați.
Cuvinte cheie: nitrați, apă de băut, factor de siguranță, methemoglobinemie

Health Effects

The current US EPA maximum contaminant level (MCL) for public drinking water supplies and the health advisory level (HAL) for other private water supplies is 10 mg/L, expressed as NO_3-N. These health-based standards and advisory levels are intended to prevent infant methemoglobinemia. Nitrate generally has a low human toxicity, but becomes a hazard when it is reduced to nitrite by bacterial action in the human gastrointestinal tract. Nitrite coverts oxygen-carrying hemoglobin to methemoglobin, which then cannot transfer oxygen. The resulting condition is methemoglobinemia, or the so-called blue baby disorder.

The most susceptible population to nitrate/nitrite toxicity is infants less than four months of age. Their high sensitivity is due to a combination of factors: higher gastric pH which allows greater bacterial activity in the stomach and subsequent enhanced conversion of ingested nitrate to nitrite, higher proportion of fetal hemoglobin which is more readily oxidized to methemoglobin than adult hemoglobin, and infant NADH - dependent methemoglobin reductase (the enzyme responsible for converting methemoglobin to normal hemoglobin) has about half the activity of the adult enzyme (1).

Several thousand cases of infant methemoglobinemia have been reported in the literature, with an overall case fatality rate of about 5-10 percent. Recent outbreaks have been reported in Central and Eastern Europe, especially in rural areas utilizing private well-water supplies (2,3). The most recent fatal case of methemoglobinemia in the United States occurred in South Dakota in 1986 (4).

The major source of nitrate intake for infants is from drinking water mixed with infant formula. Boiling of drinking water to kill bacteria (a common practice in rural areas) concentrates the nitrate that is present. Feeding practices that include early introduction of certain fruits and vegetables, which contain naturally high nitrate levels (beets, spinach, carrot or apple juice), can also enhance the risk of infant methemoglobinemia (5,6).

There is presently no evidence to support earlier hypotheses that breast-fed infants may develop

methemoglobinemia when their mothers consumed nitrate contained in well water. So far, available data from experimental animals and limited human studies indicate that neither nitrate nor nitrite accumulate or concentrate in the mammary gland or milk (7).

Methemoglobinemia is a syndrome of elevated methemoglobin level, high blood nitrite, and frequently is associated with acute diarrhea. A newly proposed hypothesis being advanced by the Center for Global Food Issues is that nitrates in water or food are not the cause of infant methemoglobinemia, but rather the infant's own body is the primary culprit as it responds to pathogens or an indigestible protein (8). In brief, this theory suggests that when an infant has acute diarrhea or other severe gastrointestinal disturbance, either from a bacterial infection in the gut or a protein intolerance (perhaps from cow's milk supplemental feedings) (9), the entire metabolism of the infant is altered in such a way as to cause methemoglobin formation, irrespective of the infant's nitrate intake from food or water. The proposed mechanism of methemoglobinemia in this case is the release of nitric oxide from the white blood cells responding to the inflammation in the gastrointestinal system. The nitric oxide is converted by the body into nitrate, then to nitrite, and finally to ammonia, which is cleared by the kidneys. This process is known as endogenous nitrate production, and has been described recently in the literature for about 45 infants studied in Israel (10). During such episodes of severe gastrointestinal disturbances, the nitric oxide is overproduced, resulting in an accumulation of nitrite in the body. The outcome has been called endogenous methemoglobinemia.

In addition to acute methemoglobinemia, other health effects including cancer (11,12), disruption of thyroid function (13), birth defects (14), developmental disorders in animals (15) and developmental disorders in children (16) are under current study with respect to their relationship to nitrate exposures in drinking water. Although the recent National Research Council report concluded that the current drinking water standards for nitrate were adequate to protect human health in the United States, this conclusion was hedged somewhat by this same subcommittee's recommendation that limiting infant exposure to nitrate would be a sensible public-health measure (17).

Basis for Drinking Water Standard

The current nitrate standard established in 1987 is based on a literature review of 278 cases of methemoglobinemia reported in the United States between 1945 and 1950. The study reported that none of these cases occurred when nitrate concentrations in drinking water were below 10 mg/L (18). Unlike other drinking water standards, the nitrate standard has **no safety factor**, which typically is about a 10-fold safety factor to account for differences in human susceptibility.

Other studies and case reports in the literature strongly suggest that a safety factor is needed. Studies conducted in Germany in 1964 indicated that about 4% of the 249 cases of methemoglobinemia occurred in infants consuming water containing less than 11 mg/L. of nitrate (19). Other case reports in the literature indicated that infants with severe diarrhea are also susceptible to methemoglobinemia following ingestion of drinking water containing less than 10 mg/L. of nitrate-N (20). A recent report of methemoglobinemia in Wisconsin involved an infant consuming formula mixed with private well water containing 9.9 mg/L of nitrate-N and up to 7.8 mg/L of copper (21). Guidance, action, or advisory levels for nitrate in drinking water are lower in several countries, including Germany (4.4 mg/L), South Africa (4.4 mg/L), and Denmark (5.6 mg/L). Clearly health and regulatory officials in other countries believe that the current drinking water standard for nitrate is not adequate to protect their most susceptible population from methemoglobinemia.

Extent of Nitrate Contamination

Another important issue related to nitrate toxicity and drinking water resources is the current extent of groundwater and surface water contamination in the United States. A national drinking water survey conducted by the US EPA indicated about 1.2% of community-water wells and about 2.4% of rural domestic wells have nitrate levels that exceed the health advisory level. It is estimated that about 1.5 million people, including about 22,500 infants, are served by rural domestic wells and that another 3 million people, including about 43,500 infants, are served by community water wells that exceed health advisory levels for nitrate (22).

An Environmental Working Group review of nearly 200,000 public water sampling records found that nearly 2-million people - including an estimated 15,000 infants under the age of four months - drank water from 2,016 water systems that were reported to the US EPA for violating the nitrate standard at least once between 1986 and 1995. All of these water systems were termed "significant noncompliers" by the US EPA and 60% were repeat violators. An additional 3.8 million people drink water from private wells that are contaminated above the 10 mg/L nitrate standard. In seven states - California, Pennsylvania, New York, Illinois, Wisconsin, Minnesota, and Iowa - more than 100,000 rural residents are exposed to nitrate above the federal standard via private drinking water (23).

In a statewide survey conducted in Iowa, about 18% of rural domestic wells had nitrate contamination above the 10 mg/L. limit (24). Similarly, an assessment of Safe Drinking Water Act database in Iowa indicates that from 1988 to 1995, the MCL was exceeded in 21% of the samples, and was greater than 5 mg/L in 43% of the samples. Some trends in the data were also noted. The median concentration of nitrate in finished water supplies

decreased from 1991 to 1995, which was also represented in a decline in the percentage of samples exceeding the MCL - 21% in 1991 to 4% in 1995 (25). However, in another study of groundwater sources in Iowa, the trend for nitrate contamination is not as clear. Since 1982 multiple samples of untreated groundwater used for Iowa municipal water supplies indicate no significant temporal trends in either the frequency of detection or median nitrate concentrations in these wells (26).

In the rural areas of America, nitrate contamination of drinking water supplies continues to be an important public health issue. In the event that future research proves a relationship between nitrate exposure to infants and subsequent adverse health effects such as cancer or developmental disorders, the population at risk to excess nitrate exposures will indeed be huge.

Naturally occurring groundwater resources without influence from anthropogenic pollution sources such as fertilizer, sewage sludge, and animal manure generally contain nitrate at levels below 3.0 mg/L (27). For example, the natural background concentration of nitrate in Iowa groundwater is typically less than 2.0 mg/L (28).

Sources of Nitrate in Drinking Water

Within the US, each year there are about 8-billion pounds more nitrogen available in farm fields than can be utilized by the crops (29). This excess nitrogen generally moves through the soil into groundwater, or is transported during rainfall events into surface waters. Some natural degradation (denitrification) also occurs. Other sources of nitrate such as sewage treatment plants, private septic systems, animal manure, legume crops, and atmospheric deposition can be important in specific, localized groundwater systems. In Iowa, the Department of Natural Resources estimates that about 55-60% of the nitrate environmental loading is from commercial fertilizer applications. Moreover, these area sources of nitrate contamination appear to be more significant than point sources or poor well construction. For example, in Iowa's statewide rural well water study, single source problems such as locating near or in animal feedlots accounted for only 3% of the total rural wells, and accounted for only about 1% of the wells exceeding the nitrate standard (1).

Policy Issues

In order to protect pristine groundwater resources and to recognize the uncertainty in current human health-based standards for nitrate toxicity, a non-degradation groundwater protection strategy for nitrate should be established for all areas where the existing groundwater quality is better than the current drinking water standard. In other words, industrial, municipal, and agricultural pollution sources should not be allowed to contaminate groundwater resources up to the current 10 mg/L level for nitrates. A

regulatory framework that accepts or encourages the so-called license to pollute concept is unwise, particularly if an allowable level of nitrate pollution is based on the flawed assumption that the current drinking water standard for nitrate is adequate to protect human health.

Is the current drinking water standard for nitrate adequate? I think not! Clearly the most susceptible human population (infants under four months of age with existing diarrhea conditions) is not adequately protected from methemoglobinemia. In addition, recent studies suggest other possible linkages between nitrate in drinking water and adverse health consequences for adults. Particularly troublesome is the recent finding of a positive association between nitrate in drinking water (at levels below the drinking water standard) and bladder cancer and ovarian cancer in a large cohort of women in Iowa (12). If further studies confirm strong associations between nitrate in drinking water and cancer, then a revised regulatory safety factor would be applied. However, in the meantime, a safety factor of at least two is needed to adequately protect the vulnerable, helpless infant population. Given this framework, the regulatory authority should implement a maximum contaminant level goal (MCLG) of 3 mg/L of NO_3-N in order to limit infant exposures to nitrate.

By mandating a safety factor of two, which would reduce the current MCL and HAL for nitrate to 5.0 mg/L NO_3-N, and by promulgating a MCLG of 3.0 mg/L of NO_3-N; the United States regulatory approach for nitrate in drinking water would become consistent with other European countries and would encourage the prudent public health strategy of limiting human nitrate exposure.

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Exposure-based assessment and economic valuation of adverse birth outcomes and cancer risk due to nitrate in United States drinking water.

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ABSTRACT

Background: Nitrate ingestion from drinking water has been associated with an increased risk of adverse birth outcomes as well as elevated risk of colorectal cancer and several other cancers. Yet, to date, no studies have attempted to quantify the health and economic impacts due to nitrate in drinking water in the United States.
Methods: This study presents a first-of-its-kind comprehensive assessment of nitrate exposure from drinking water for the entire United States population. This exposure assessment serves as the basis for our analysis of the annual nitrate-attributable disease cases in the United States and the associated economic losses due to medical costs and lost productivity. Additionally, through a meta-analysis of studies on drinking water nitrate and colorectal cancer, we examine the exposure-response relationship for nitrate and cancer risk.
Results: On the basis of national nitrate occurrence data and relative risk ratios reported in the epidemiology literature, we calculated that annually, 2939 cases of very low birth weight, 1725 cases of very preterm birth, and 41 cases of neural tube defects could be related to nitrate exposure from drinking water. For cancer risk, combining nitrate-specific risk estimates for colorectal, ovarian, thyroid, kidney, and bladder cancers results in a range of 2300 to 12,594 annual nitrate-attributable cancer cases (mean: 6537 estimated cases). For medical expenditures alone, this burden of cancer corresponds to an annual economic cost of 250 million to 1.5 billion U.S. dollars, together with a potential 1.3 to 6.5 billion dollar impact due to lost productivity. With the meta-analysis of eight studies of drinking water nitrate and colorectal cancer, we observed a statistically significant positive association for nitrate exposure and colorectal cancer risk and calculated a one-in-one million cancer risk level of 0.14 mg/L nitrate in drinking water.
Conclusion: Health and economic analyses presented here suggest that lowering exposure to nitrate in drinking water could bring economic benefits by alleviating the impacts of nitrate-associated diseases.

1. Introduction

A large body of epidemiological research has found an elevated risk of cancer, adverse birth outcomes and other health impacts associated with the presence of nitrate in drinking water (Ward et al., 2018). These effects are often observed at drinking water nitrate concentrations significantly lower than the levels associated with methemoglobinemia, or blue-baby syndrome, a life-threatening condition that can kill an infant through oxygen deprivation. The U.S. drinking water standard for nitrate of 10 mg/L nitrate (as nitrogen) was first set in 1962 in order to protect against methemoglobinemia. The Canadian legal limit for nitrate in drinking water is equivalent to the U.S. standard, and the European standard is comparable, allowing up to 50 mg/L of nitrate as nitrate (corresponding to 11.3 mg/L nitrate as nitrogen).

For decades, methemoglobinemia was considered to be the primary health concern due to nitrate ingestion from water. This viewpoint is reflected in recent regulatory risk assessments published by government agencies, for example Health Canada (2013) and California Office of Environmental Health Hazard Assessment (OEHHA 2018a). Yet, the epidemiological evidence linking nitrate in drinking water with human health harms raises questions about whether the nitrate limit of 10 mg/L protects the general population against adverse health outcomes.

Recent epidemiological studies with large study populations conducted in Spain and Italy (Espejo-Herrera et al., 2016) and in Denmark (Schullehner et al., 2018) reported statistically significant increases in colorectal cancer risk associated with nitrate in drinking water at levels of 0.7–2 mg/L. Amongst these studies, the highest risk was observed for men with high red meat intake and highest exposure to nitrate from

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List of abbreviations

DALY	Disability-Adjusted Life Year
HR	Hazard Ratio
OEHHA	California Office of Environmental Health Hazard Assessment
OR	Odds Ratio
RR	Relative Risk
VOLY	Value of a Life Year
USGS	United States Geological Survey
U.S. EPA	U.S. Environmental Protection Agency
YLD	Years Lost due to Disability
YLL	Years of Life Lost

drinking water (Espejo-Herrera et al., 2016). These European publications corroborate the results from an earlier study conducted in Iowa, a region of the United States with a history of elevated nitrate in drinking water, where elevated colorectal cancer risk was observed for drinking water nitrate levels above 5 mg/L, for individuals with above median meat consumption and below median Vitamin C intake (De Roos et al., 2003). Additionally, statistically significant increases in the risk of ovarian, thyroid, kidney and bladder cancers associated with exposure to nitrate have been reported in studies of an Iowa cohort of women 55–69 years old (Inoue-Choi et al., 2015; Jones et al., 2017; Jones et al., 2016; Ward et al., 2010).

Notably, not all epidemiological studies report elevated risk for colorectal cancer and nitrate exposure, and some publications report null findings. For example, studies of a female-only cohort in Iowa observed no association between drinking water nitrate and colorectal cancer risk alone or when risk factors such as red meat intake and antioxidant intake levels were also considered (Jones et al., 2019; Weyer et al., 2001).

Cancer development upon nitrate ingestion is a complex process likely mediated by the endogenous formation of N-nitroso compounds, which are potent mutagens and carcinogens. In the body, nitrate can be reduced to nitrite and further metabolized to yield nitrosating agents capable of reacting with dietary amines to form such compounds. The World Health Organization's International Agency for Research on Cancer classified ingested nitrate as probably carcinogenic to humans, specifically when nitrate is ingested under conditions that promote endogenous nitrosation (IARC, 2010). Dietary consumption of nitrate-preserved meats and red meat in general contributes to nitrosation and has been associated with greater cancer risk. In contrast, intake of nitrate in the presence of compounds that inhibit endogenous nitrosation, such as Vitamin C and E, may prevent or reduce the formation of N-nitroso compounds (Khatri et al., 2017). There is some evidence that endogenous formation of N-nitroso compounds occurs upon ingestion of nitrate from drinking water, as documented by the urinary excretion of N-nitroso compounds (Mirvish et al., 1992; van Maanen et al., 1996).

In addition to cancer risk, exposure to drinking water nitrate during pregnancy has been associated with an elevated risk of adverse birth outcomes such as neural tube birth defects or other birth defects (Brender et al., 2013). Mother's exposure during pregnancy has been also associated with small for gestational age at birth (Migeot et al., 2013) as well as very preterm birth and very low birth weight (Stayner et al., 2017a). Potential mechanism(s) underlying these reproductive and developmental effects remain to be elucidated. While N-nitroso compound formation following nitrate ingestion exhibits developmental toxicity in animal studies, epidemiological data suggest that a different mechanism of nitrate toxicity might be involved in adverse birth outcomes (Brender et al., 2013). Additional pathways of nitrate toxicity could include inhibition of iodine uptake into the thyroid and changes in the thyroid function (Cao et al., 2010; Horton et al., 2015; Tonacchera et al., 2004) as well as interference with steroidogenesis (Edwards et al., 2018; Hamlin et al., 2016; Poulsen et al., 2018).

In light of the epidemiological data suggesting potential health harms at current levels of nitrate in drinking water, a population-wide assessment of nitrate-attributable health and economic impacts for the United States is both timely and practical. The present study utilizes nitrate occurrence data for public water systems in all 50 U.S. states to estimate the annual number of nitrate-associated adverse pregnancy outcomes, cancer cases and associated economic costs for the U.S. population as a whole. Additionally, we carried out a meta-analysis of studies on nitrate and colorectal cancer and determined nitrate's carcinogenic potency, also called the cancer slope factor, using established risk assessment methodologies (U.S. EPA, 1992). Together, these data form a solid platform for developing risk-based health benchmarks and drinking water standards that would protect human health from nitrate-attributable adverse effects.

2. Methods

2.1. Exposure assessment for nitrate in community water systems in the United States

This study is based on a national-level dataset for nitrate occurrence in public water systems in the United States for 2010–2017. The dataset is posted in an open access database available at https://www.ewg.org/tapwater/, which, to our knowledge, represents the most comprehensive, freely searchable source of tap water contaminant occurrence data for the U.S. Within the database, and throughout this paper, all nitrate concentrations in drinking water are expressed for nitrate as nitrogen, which is the standard metric in the United States for reporting drinking water nitrate concentrations. For the purposes of exposure assessment in this analysis, we calculated the arithmetic mean for all nitrate test results available for each individual public water system for 2010–2017, and this calculated value was assigned as the exposure level for this system. Test results reported as "non-detects" were assigned a value of zero and included in the overall data array for the calculation of averages. This approach is conservative and exerts a downward effect of the overall exposure estimates because, at least in some states, the detection limit of nitrate for purposes of reporting is higher than what is achievable with the analytical capabilities of the most sensitive test methods.

Population statistics for community water systems were obtained from the U.S. EPA Envirofacts database (https://www3.epa.gov/enviro/facts/sdwis/search.html), and supplemented with data available from state drinking water programs. These population numbers represent an estimate, and the specific number of customers and residents served by an individual water system may differ. Analyzing the population statistics in our dataset we found that for 38 out of 50 states, the overall population data for residents served by community water systems were within 10% of what was expected based on the 2017 Census data. For 8 states in our dataset, the calculated population was within 20% of expected, while for remaining 4 states (Alaska, Alabama, Massachusetts, and Mississippi), the population calculated from the U.S. EPA Envirofacts data diverged by more than 20% from the population expected from the census data. Based on this analysis, we applied a state-specific population adjustment factor where needed, to bring our estimates for the total population served by community water systems in each state in concordance with the 2017 census data.

2.2. Exposure assessment for nitrate in private water wells in the United States

To assess nitrate exposure for private well users, we developed an

extrapolation model that incorporates nitrate testing data for groundwater-based community and non-community systems that serve up to 50 people. Non-community systems are defined by the U.S. EPA as "a public water system that regularly supplies water to at least 25 of the same people at least six months per year" or a system that "provides water in a place such as a gas station or campground where people do not remain for long periods of time". Over 95% of non-community water systems are groundwater systems (U.S. EPA, 2018b), and over 90% of very small community systems use groundwater (National Research Council, 1997).

Our approach incorporates information on the number of people who use private water wells in each state (Kenny et al., 2009; U.S. EPA, 2011). For this analysis, we treated the nitrate concentrations in the non-community water systems and the smallest community water systems as a proxy for nitrate levels in private wells. Private water wells are likely to have the same depth or be shallower compared to public water systems and would likely have same or worse nitrate concentration profiles as what is found in the very small community or non-community water systems. Thus, our modeling approach represents a conservative scenario with respect to private well users' exposure to nitrate.

We analyzed the state-level profiles of nitrate occurrence in 2010–2017 in non-community and community water systems serving less than 50 people, and determined the state-level percentage of those systems that provide water with average nitrate concentrations exceeding a defined nitrate concentration level. For the purposes of this analysis, the nitrate occurrence distribution in the above dataset was considered equivalent to the nitrate occurrence distribution in the private wells in the same state.

To validate this approach, we utilized data from the U.S. EPA analysis of state-specific U.S. Geological Survey data on the percentage of area groundwater contaminated with nitrate above 5 mg/L (U.S. EPA, 2011). We compared these EPA estimates with our modeled estimates of the percentage of private well users in each state relying on water with more than 5 mg/L nitrate (Supplementary Table 1). These two metrics are distinct yet related, as one reflects the area of groundwater impacted by nitrate, and the other reflects a possible number of private well users impacted. In a correlation analysis, for 31 states that constitute 91% of the overall U.S. population served by private wells, the median of the absolute difference between the two metrics approaches zero, indicating overall concordance between the two datasets.

2.3. Calculation of nitrate-attributable cases of disease

In order to calculate the nitrate-attributable cases of diseases or health conditions, namely cancer and adverse reproductive outcomes, we adapted, with modifications, a published methodology for calculation of nitrate-attributable colorectal cancer cases in Europe (van Grinsven et al., 2010). The calculations formula incorporates relative risk from epidemiological studies, size of the population exposed to nitrate concentration above a specific cut-off level, and the current annual incidence proportions of a specific disease or health condition, available from the Centers for Disease Control and Prevention (U.S. Cancer Statistics, 2017). We first calculated baseline incidence proportion, referred to as Inc_B in the following equation:

$$Inc_B = \text{Disease Cases} / ((PoP_E * R_E) + (Pop_U * R_U)$$

where.

Inc_B = baseline incidence proportion in the unexposed population.
Disease Cases = National disease incidence proportion * total U.S. population.
Pop_E = exposed population (estimated number of people from public water systems and private wells drinking water with nitrate above a specified concentration).
Pop_U = unexposed population (total population minus Pop_E).
R_E = relative risk of the exposed population (odds ratio for a disease or a health condition in exposed population from epidemiological literature).
R_U = relative risk of the unexposed population (value = 1).

We than calculated the nitrate attributable cases using the following equation:

$$\text{Nitrate Attributable Cases} = Pop_E * \Delta R * Inc_B$$

where,

$\Delta R = R_E - R_U$ or the increased risk in the exposed population

To identify relevant epidemiological literature for cancer risk estimates and nitrate exposure levels, we queried the Pubmed database with a search term "drinking water nitrate and cancer", or a combination of such terms. An assumption of this methodology is the causal link between exposure to nitrate in drinking water and cancer development, therefore only studies indicating positive findings were used in our analysis of nitrate-attributable cancer cases. Five different risk scenarios for colorectal cancer were selected, based on reported nitrate exposure and significant increases in odds ratios or hazard ratios in studies by De Roos et al. (2003), Espejo-Herrera et al. (2016), and Schullehner et al. (2018). These studies were chosen because they had strong study designs incorporating large sample sizes, improved exposure assessment and control of factors influencing endogenous nitrosation. Of the three studies, Schullehner et al. (2018) presents a nation-wide assessment of colorectal cancer risk in Denmark coupled with reliable individually linked exposure data. For the assessment of other types of cancer risk related to nitrate, we used a kidney cancer risk scenario from Ward et al. (2007) for a cohort that included both men and women and reported similar risk estimates as Jones et al. (2017). For bladder cancer risk (Jones et al., 2016), ovarian cancer risk (Inoue-Choi et al., 2015), and thyroid cancer risk (Ward et al., 2010), risk estimates come from a well-defined cohort of over 20 thousand women 55–69 years old in Iowa who were enrolled in 1986 in the National Cancer Institute's Iowa Women's Health Study (National Cancer Institute, 2018).

For all studies analyzed here, odds ratios were interpreted as relative risk values since cancer is a rare event (Cochrane Collaboration, 2011). Risk estimates were used for exposure groups that found a significant increased risk relative to the lowest exposure group. Concentration cut-off levels were determined as the lower limit of the exposure group indicating an increased risk and are expressed as mg/L nitrate-nitrogen. Three studies reported elevated cancer risk from nitrate in drinking water relative to meat consumption. De Roos et al. (2003) classified this study population as above median meat consumers, while Espejo-Herrera et al. (2016) and Ward et al. (2007) further specified high red meat consumption. For these scenarios, we used increased relative risk values for R_E. Increased relative risk values were calculated using the following equation:

RE = OR above median meat/red meat consumer + nitrate /
OR above median meat/red meat consumer (no nitrate)

This approach accounts for the slight increased risk of cancer associated with red meat or meat consumption and no nitrate exposure and was used in the van Grinsven study (2010) and confirmed through personal communication with the author.

In some scenarios, we incorporated a population adjustment factor whereby the exposed population was adjusted to accurately reflect the characteristics of the at-risk population from our selected studies. Above median meat/red meat consumers were considered 50% of the total U.S. population. For scenarios applicable to women 55–69 years of age, we defined this group as 9% of the total U.S. population according to the 2017 U.S. census report. Women 55–69 years of age with no history of bilateral oophorectomy were considered 7% of the total U.S. population given that approximately 20% of women in this age range in

the United States have had bilateral oophorectomy surgery (Howe 1984).

A similar approach was employed for calculating nitrate-attributable cases of adverse birth outcomes, whereby we assessed the estimated numbers for nitrate-related neural tube defects, incidence of very low birth weight and very preterm births. Three thousand pregnancies in the U.S. each year are affected by neural tube defects (Oakeshott et al., 2010). Anencephaly and spina bifida account for approximately 80% of all neural tube defects based on incidence reported by the Centers for Disease Control and Prevention, and spina bifida is twice as common as anencephaly. Attributable cases were calculated based on national incidence proportions. For neural tube defects and very low birth weight outcomes, data was obtained from the 2016 Centers for Disease Control and Prevention National Health Statistics. For very preterm birth, data was obtained from 2014 to 2015 March of Dimes Perinatal Data Center (2019).

2.4. Assessment of economic costs for nitrate-attributable adverse birth outcomes

For all economic analyses presented here, costs are expressed in 2014 U.S. dollars. As recommended by Dunn et al. (2018), medical costs were indexed using the Bureau of Economic Analysis' Personal Consumption Expenditures health price index, while indirect economic losses were updated using the general Personal Consumption Expenditures price index (U.S. Department of Labor Bureau of Labor and Statistics, 2017). Full analysis of the direct and indirect economic costs for all nitrate-related adverse birth outcomes is beyond the scope of this manuscript and deserves its own investigation. In our analysis we incorporated the costs of hospitalization for medical concerns for three outcomes studied here (neural tube defects, very pre-term birth and very low birth weight) reported in the research literature. Due to uncertainty about potential overlap between the occurrence and registration of low birth weight and preterm birth, we did not aggregate the total costs for these birth outcomes but presented them separately.

For the very low birth weight, lost economic productivity was estimated based on the loss of IQ points (indirect costs) according to recently published methodology (Malits et al., 2018). Following this approach, low birth weight was considered to incur a 4.98-point loss in IQ, as defined through a meta-analysis of the impact of low birth weight on intelligence in adolescence and early adulthood (Kormos et al., 2014). Very low birth weight is a more severe health outcome compared to low birth weight, and thus out approach of assigning this IQ loss value to very low birth weight cases is conservative. Following the U.S. EPA economic analysis, each IQ point loss was valued at $11,745 – $15,883 in 2014 dollars (U.S. EPA, 2015a). Overall indirect economic cost is calculated by multiplying the number of nitrate-attributable very low weight births by the 4.98 IQ point loss per case and the cost of each IQ point loss (Malits et al., 2018).

2.5. Assessment of direct medical costs due to nitrate-attributable cancer cases

For cancer-related medical costs, we obtained annualized mean net costs of care per patient published by the National Cancer Institute, based on research by Mariotto et al. (2011), converted to 2014 U.S. dollars. We estimated the total costs per cancer case with the following formula:

$$\text{Total cost per case} = \text{Initial cost} + \text{Continuing Costs each Year} + \text{Cost for the Last Year of Life}$$

For calculation of continuing costs per year, annual continuing cost was multiplied by the median years lived with disease (Supplementary Table S2), minus 2 years, which represent the first year when the diagnosis is made and the last year of life. For the last year of life, National Cancer Institute gives two cost estimates, one for death due to cancer and another due to death from causes other than cancer (Mariotto et al., 2011). Here we average these two estimates to obtain a single average cost for the last year of life for specific cancers. Supplementary Table S2 lists calculated cost of medical care per cancer case for colorectal, ovarian, kidney and bladder cancer. We did not carry out cost of medical care calculations for thyroid cancer because the National Cancer Institute study did not include this type of cancer (Mariotto et al., 2011).

2.6. Assessment of economic losses due to nitrate-attributable cancer cases

For the indirect economic loss assessment, we used the World Health Organization metric for Disability-Adjusted Life Years (DALY), together with the Value of Life Year (VOLY) approach where the Value of Life Year is derived from research literature (World Health Organization, 2018; Desaigues et al., 2011; van Grinsven et al., 2010). This calculation incorporates two variables measuring the impact of a disease, namely the years of life lost (YLL) and the number of years lost due to disability (YLD) and is calculated as follows:

YLD = Years lived with disease * Disease-specific disability weight.

YLL = Average life expectancy for the population - median age at death for the disease.

DALY = Number of nitrate-attributable cases * (YLL + YLD).

Indirect Economic Loss = Total DALY * Value of Life Year (VOLY).

All parameters used in these calculations are listed in Supplementary Table S2. Median ages at diagnosis and death for specific cancers were obtained from the website of the National Cancer Institute's Surveillance, Epidemiology, and End Results Program (SEER). Years lived with disease (YLD) were calculated as the difference in years between the median age at death and the median age at diagnosis for the disease, multiplied by the disability weight for a specific cancer. Here, we used cancer site-specific disability weights for the diagnosis and primary therapy phase of the cancer: colorectal cancer (0.43), ovarian cancer (0.43), thyroid cancer (0.27), kidney cancer (0.27), bladder cancer (0.27), as described in Soerjomaataram et al. (2012).

There is a broad range of estimates in the research literature for the Value of Life Year that usually fall within one to three times the per capita GDP of a given country (Marseille et al., 2015). Similar to a recently published study of economic loss due to diseases attributable to environmental exposure (Grandjean and Bellanger 2017), we used a Value of Life Year derived from a nine-country European assessment (Desaigues et al., 2011). The Value of Life Year estimate of 40,000 euro recommended by Desaigues et al. (2011) was converted to 2014 U.S. dollars using the 2010 euro to USD conversion rate and adjusting for inflation between 2010 and 2014, resulting in a value of $57,757.

2.7. Meta-analysis of studies of colorectal cancer and nitrate

The U.S. National Library of Medicine Pubmed database was queried to identify academic literature using the search term "drinking water nitrate and colorectal cancer", or a combination of such terms. To be included in the dose-response analysis, studies needed to be of case-control or cohort study design, with risk values for colon or colorectal cancer reported as odds ratio (OR), relative risk (RR) or hazard ratio (HR). Studies on rectal cancer only were excluded due to a less robust dataset for this cancer site. Since colorectal cancer is a rare event (prevalent in less than 10% of the study population), OR were treated as RR for simplicity (Cochrane Collaboration, 2011). Additionally, studies had to report at least two levels of nitrate exposure quantified in mg/L, or mg/day (with estimations of water consumption), or mmol/L. Lastly, same study cases and controls could not be present in more than one study.

Data extracted from each study (Table 4) included dose estimates for each exposure group and the corresponding OR, RR or HR from the

Table 1
Estimated annual nitrate-attributable cases of adverse birth outcomes.

Data Imported from Peer-reviewed Literature				Calculated Outcomes			
Analysis ID and birth Outcome	Study author and publication year	Nitrate cut-off level (mg/L)	Risk in exposed Population	Estimated nitrate exposed births [a]	Number of annual attributable cases due to nitrate exposure from community water systems [b]	Number of annual attributable cases due to nitrate exposure from private wells [b]	Percent of annual adverse birth outcomes due to drinking water nitrate [c]
1 – Neural Tube Defect	Brender et al., 2013	4.5	1.43	126,575	32	9	1.4%
2 – Very low birth weight	Stayner 2017b	1	1.17	1,108,703	2592	347	5.3%
3 – Very preterm birth	Stayner 2017b	1	1.08	1,108,703	1522	204	2.7%

[a] The number of at-risk births is the percentage of total 2016 births that is equivalent to the same percentage of people exposed to the nitrate cut-off level relative the total U.S. population.
[b] Attributable cases were calculated based on national incidence proportions. For neural tube defects and very low birth weight outcomes, data was obtained from the 2016 Centers for Disease Control and Prevent National Health Statistics. For very preterm birth, data was obtained from 2014 to 2015 statistics from the March of Dimes Perinatal Data Center (https://www.marchofdimes.org/peristats/Peristats.aspx).
[c] Nitrate attributable cancer cases divided by total cases for each birth outcome based on 2014–2016 incidence statistics. Neural tube defects: 3000 cases; Very low birth weight: 55,242 cases; Very preterm birth: 63,134 cases.

analysis accounting for the most covariables as well as the number of cases and controls or person years. Mean/median values were used when provided in the study or directly provided to us by the author (Schullehner, personal communication). When mean/median values were not available, midpoint values were calculated. For the highest exposure groups where there was no upper concentration limit, dose estimates were calculated as the value plus the width of the previous interval. All values used for exposure/dose estimates are expressed as mg/L nitrate-nitrogen. To achieve this, several conversions were required for studies based on populations in Europe and Asia. For European studies reporting drinking water values as mg/L nitrate, values were multiplied by 0.2259. In the case of Espejo-Herrera et al. (2016), values reported as mg/day nitrate-N were first divided by the average water consumption rate of cases (1.4 L/day) and then converted to nitrate. Chiu et al. (2010) reported values as mmol/L nitrate-N, which were converted into mg/L concentrations.

All statistical analyses were performed in STATA (version 14, STATA, 2018). Generalized least squares regression analysis was used to generate study-specific slopes representing the estimated increase in log risk ratio per mg/L increase in nitrate concentration and standard errors for these slopes. Study-specific slopes and their standard errors were then incorporated into meta-analysis using a random effects model to derive a weighted pooled slope estimate with 95% confidence intervals based on the DerSimonian and Laird method (1986). A random effects model was used instead of a fixed effects model in order to account for both within-study variation and inter-study variation.

Study heterogeneity was assessed using the I^2 test and the heterogeneity chi-squared test for significance, whereby a p-value less than 0.1 considered to be significant (Higgins et al., 2003). I^2 values from 0 to 40%, 30–60%, 50–90% and greater than 75% are interpreted to represent low, moderate, substantial and considerable study heterogeneity, respectively (Deeks et al., 2011). To identify the source of heterogeneity, we conducted additional analysis by grouping studies based on similar covariables following methodology from Camargo et al. (2011). We also conducted sensitivity analysis by omitting single studies from the pooled estimates and examining the I^2 values and p-values for the meta-analysis of the remaining seven studies.

2.8. Analysis of risk-based benchmark values for nitrate protective of human health

We derived a cancer-based drinking water guideline for nitrate following established methodologies for the calculation of drinking water concentrations corresponding to a particular cancer risk level (U.S. EPA, 1992). Here we follow the California Office of Environmental Health Hazard formula (OEHHA, 2004) whereby:

$$C = R / CSF * BR * WCA$$

where.

C = drinking water concentration corresponding to a specified cancer risk level.

R = cancer risk level; in this study we use one-in-one-million or 10^{-6} risk level.

CSF = cancer slope factor.

BR = background cancer rate.

WCA = water consumption adjustment factor between populations.

Here, cancer slope factor is the pooled slope estimate for colorectal cancer, as calculated by meta-analysis; and the background cancer rate was the average annual U.S.-wide incidence of colorectal from 2011 to 2015 published by the Centers for Disease Control and Prevention, which is 39.4 cases per 100,000 people. A water consumption adjustment factor was used to account for differences in the amount of water consumed by the different populations in the included studies since differences in water consumption could lead to differences in internal dose. The water consumption adjustment factor was calculated by taking the inverse of the combined average minimum and maximum

Table 2
Estimated annual nitrate-attributable cases of colorectal, ovarian, thyroid, kidney and bladder cancers.

Data Imported from Peer-reviewed Literature					Calculated Outcomes			
Analysis ID and cancer type	Study author and publication year	Nitrate-N cut-off (mg/L) [a]	Cancer risk in exposed population	Additional risk factors and population adjustment (% of total U.S. Population) [b]	At-risk population [c]	Number of annual attributable cases due to nitrate exposure from community water systems [d]	Number of annual attributable cases due to nitrate exposure from private wells [d]	% of Total annual cancer cases attributable to nitrate exposure, rounded [e]
A - Colorectal	Espejo-Herrera et al., 2016	1.7	1.49	None (100%)	59,144,818	9054	1325	8.2%
B - Colorectal	Espejo-Herrera et al., 2016	0.7	1.3	Above median red meat consumers (50%)	55,479,150	5447	729	4.9%
C - Colorectal	Schullehner et al., 2018	0.9	1.11	None (100%)	96,442,751	3529	478	3.2%
D - Colorectal	Schullehner et al., 2018	2	1.15	None (100%)	46,871,865	2310	374	2.1%
E - Colorectal	De Roos et al., 2003	5	1.8	Above median meat consumers (50%)	3,989,662	939	294	1.0%
F - Ovarian	Inoue-Choi et al., 2015	3	2.03	Women 55–69 years of age with no history of bilateral oophorectomy (7%)	1,935,539	486	94	3.2%
G - Ovarian	Inoue-Choi et al., 2015	5	1.6	Women 55–69 years of age with no history of bilateral oophorectomy (7%)	600,570	84	26	0.6%
H - Thyroid	Ward et al., 2010	2.5	2.18	Women 55–69 years of age (9%)	3,066,241	880	167	2.2%
I – Thyroid	Ward et al., 2010	5	2.59	Women 55–69 years of age (9%)	750,712	281	88	0.8%
J – Kidney	Ward et al., 2007	5	1.7	Above median red meat consumers (50%)	3,989,662	346	108	0.9%
K – Bladder	Jones et al., 2016	5	1.61	Women 55–69 years of age (9%)	750,712	102	32	0.2%

[a] Nitrate concentration cut-offs were rounded to no more than one decimal place.
[b] Population adjustment factors reflect the additional risk factors or population characteristics, such as above median meat or red meat consumption, age and gender, and medical history as defined in the original epidemiological studies.
[c] At-risk population represents the size of the specified sub-population that is exposed to a given nitrate concentration.
[d] Attributable cancer cases were calculated based on age-adjusted annual cancer incidence from 2011 to 2015 obtained from the Centers for Disease Control and Prevention U.S. Cancer Statistics. For ovarian, thyroid and bladder cancers attributable cases were calculated on the basis of annual incidence from 2011 to 2015 for females 50 + years of age.
[e] Nitrate-attributed cancer cases divided by total expected cancer cases, rounded to one decimal place.

Table 3
Disability-adjusted life years (DALYs) and economic costs associated with estimated annual nitrate-attributable cancer cases.

Analysis ID and cancer type	Total nitrate attributable cases for community water systems and private wells [a]	Total years of life with disease [b]	Total years lost due to disability [b]	Total years of life lost [b]	Total DALYs[b]	Estimated indirect Economic Loss due to Lost Productivity, in 2014 U.S. dollars (billions)[c]	Combined medical costs of cancer treatment in 2014 U.S. dollars (billions) [d]
A - Colorectal	10,379	62,275	26,778	58,123	84,901	$4.90	$1.33
B - Colorectal	6176	37,053	15,933	34,583	50,516	$2.92	$0.79
C - Colorectal	4007	24,041	10,338	22,439	32,776	$1.89	$0.51
D - Colorectal	2684	16,104	6925	15,031	21,956	$1.27	$0.34
E - Colorectal	1233	7396	3180	6903	10,083	$0.58	$0.16
F - Ovarian	580	4062	1747	6441	8188	$0.47	$0.11
G - Ovarian	110	773	332	1226	1558	$0.09	$0.02
H - Thyroid	1047	23,026	6217	8478	14,695	$0.85	N/A
I – Thyroid	369	8113	2191	2987	5178	$0.30	N/A
J – Kidney	454	3179	858	3451	4310	$0.25	$0.06
K – Bladder	134	938	253	281	535	$0.03	$0.01

[a] Values from Table 2, combining the estimated cancer cases for private well users and for community water systems.
[b] Values in these columns refer to total years of life with disease, years lost due to disability, years of life lost and DALYs for all cases attributed to nitrate in each analysis. Calculations for cancer specific disability-adjusted life years are listed in Supplementary Table S2.
[c] Economic Loss = VOLY * Total DALYs where VOLY = $57,757.
[d] Economic loss due to medical costs of cancer treatment calculated on the basis of annualized mean net costs of care per patient published by the National Cancer Institute. As listed in Supplementary Table 2, medical costs per case of colorectal cancer are $127,890; per case of ovarian cancer are $196,452; per case of kidney cancer are $128,921; per case of bladder cancer are $92,127. No medical costs for thyroid cancer were listed by the National Cancer Institute study (Mariotto et al., 2011), indicated as N/A for "Not Available".

reported water consumption values (L/day) for each study included in our meta-analysis (Supplementary Table S4). This combined average value was 2.13 L/day. Where specific values for water consumption could not be identified, a value of 2 L/day was assumed, as common practice for U.S. EPA drinking water standards (U.S. EPA, 2018a).

3. Results

3.1. Annual nitrate-attributable disease cases

A unique and powerful feature of this analysis is our ability to calculate exposure information for the portion of the U.S. population, by state, that likely ingest nitrate above specified concentrations in drinking water. As expected, the population exposed negatively correlates with nitrate levels in the water supply, where a greater number of people are exposed to lower levels of nitrate and vice versa, with the exception of those with non-detectable levels (Fig. 1). From 2010 to 2017, approximately 81 million people served by community water systems in the U.S. had a mean drinking water nitrate level of 1 mg/L and above, while 6 million people had a mean level of 5 mg/L or more nitrate in their drinking water (Fig. 1). Similar calculations were conducted for nitrate exposure levels for private well users, and nitrate-attributable cases of disease were analyzed separately for private well and community water system users (Tables 1 and 2).

To assess the health risks associated with short-term exposure to drinking water nitrate during pregnancy, we calculated the number of nitrate-attributable adverse pregnancy outcomes. Such adverse outcomes affect a relatively small percent of the overall pregnancies. Centers for Disease Control and Prevention statistics show that approximately 0.07% of births have neural tube defects, while 1.4–1.6% of births are associated with very low birth weight or very preterm deliveries. Based on risk estimates reported in epidemiological studies on drinking water nitrate exposure and pregnancy outcomes (Brender et al., 2013; Stayner 2017b), we calculated that annually 2939 very low birth weight births, 1725 very preterm births, and 41 births with neural tube defects could be attributable to nitrate exposure (Table 1). Nitrate-attributable cases of neural tube defects, very low birth weight and very preterm birth account for 1.4, 5.3 and 2.7 percent of total annual cases of these adverse reproductive outcomes in the U.S.

Combining the exposed population and cancer case estimates for community water systems and private well users yields an estimated range of annual national nitrate-attributable colorectal cancer cases between 1233 and 10,379 cases, corresponding to between 1 percent and 8 percent of all annual U.S. colorectal cancer cases (Table 2). The lowest number of nitrate-attributable cancer cases was derived from a scenario based on findings from De Roos et al. (2003) (Scenario E) while the highest number of nitrate-attributable cancer cases was derived from the Espejo-Herrera et al. (2016) general population scenario (Scenario A). Previous published literature has estimated the number of nitrate attributable colorectal cancer cases in the European Union as approximately 4 percent of the annual incidence (van Grinsven et al., 2010), which is comparable to the range determined in our study.

This analysis was repeated for ovarian, thyroid, kidney and bladder cancer yielding an additional 110–580 ovarian, 369–1047 thyroid, 454 kidney and 134 bladder cancer cases respectively (Table 2). These additional cases represent approximately 0.6–3 percent of the annual US ovarian cancer cases, 0.8 to 2 percent of the thyroid cancer cases, 0.9 percent of the kidney cancer cases and just 0.2 percent of the annual bladder cancer cases. Adding estimated ovarian, thyroid, kidney and bladder cancers to the total colorectal cancer cases results in a modest increase in the total estimate for annual nitrate-attributable cancer cases, ranging from 2300 to 12,594, where 54–82% of cases correspond to colorectal cancer.

3.2. Medical costs and lost productivity costs due to nitrate-attributable diseases

Here we followed the examples of other studies by separately considering the direct and indirect costs of illness (U.S. EPA, 2010). For an economic assessment of costs related to neural tube defects, we relied on the lifetime direct costs for spina bifida of $577,000 to 791,900 per case (2014 U.S. dollars), as published by the National Center on Birth Defects and Developmental Disabilities, a part of the U.S. Centers for Disease Control and Prevention (Grosse et al., 2016). For 41 annual nitrate-attributable cases of neural tube defects, this cost per case corresponds to an economic impact of $24–32 million.

For premature births, we applied a value of $51,600 (in 2005 dollars) as reported by the Institute of Medicine (2007), corresponding to $67,022 in 2014 dollars, which translates to a medical cost of 116 million dollars for the 1725 annual nitrate-attributable cases of very preterm birth. Notably, there might be potential overlap between very preterm births and very low birth weight cases and additional epidemiological research is needed to better define these relative risks of nitrate-associated adverse birth outcomes. Further, following recently

Table 4
Number of cases and controls, estimated dose and relative risk values extracted from studies included in the meta-analysis.

Study	Cases	Controls	Exposure groups (mg/L)	Estimated dose (mg/L)	Relative risk	95% Confidence Interval lower limit	95% Confidence Interval upper limit
Case-Control Studies							
De Roos et al., (2003) [a b]	172	566	≤1	0.5	1		
Table 2	116	380	>1 ≤ 3	2	1.02	0.8	1.3
	27	124	>3 ≤ 5	4	0.7	0.4	1.1
	61	174	>5	7	1.2	0.8	1.7
Espejo-Herrera et al.,	778	1899	≤0.81	0.40	1		
(2016) [a c d]	447	803	>0.81–1.61	1.21	1.7	0.98	1.38
Table 2	644	828	>1.61	2.42	1.49	1.24	1.78
Chiu et al., (2010) [e f]	1921	2052	<0.38	0.06	1		
Table 3	730	732	0.39–0.57	0.43	1.02	0.9	1.15
	1056	923	>0.60	0.99	1.16	1.04	1.3
Yang et al., (2007) [f]	775	746	≤0.22	0.00	1		
Table 2	758	749	0.23–0.45	0.38	0.98	0.84	1.14
	701	739	0.48–2.86	0.74	0.98	0.83	1.16
Fathmawati et al., (2017)	56	67	≤11.3	5.65	1		
[a c]	19	8	>11.3	22.59	2.82	1.075	7.395
Table 2							
McElroy et al., (2008) [a g]	147	549	<0.5	0.25	1		
	104	274	0.5–1.9	1.20	1.39	1.02	1.89
	137	361	2.0–5.9	3.95	1.32	0.99	1.76
	57	159	6.0–9.9	7.95	1.28	0.88	1.88
	33	86	≥10.0	13.90	1.57	0.97	2.52
	Cases	**Person-years**	**Exposure Groups (mg/L**	**Estimated Dose (mg/L)**	**Risk Ratio**	**95% Confidence Interval Lower Limit**	**95% Confidence Interval Upper Limit**
Cohort Studies							
Weyer et al., (2001) [h i]	58	48,438	<0.36	0.20	1.00		
	86	48,163	0.36–1.00	0.70	1.53	1.09	2.16
	92	47,821	1.01–2.46	1.91	1.54	1.08	2.19
	64	48,011	>2.46	5.59	0.98	0.66	1.46
Schullehner et al., (2018)	788	4,071,980	<0.29	0.16	1.00		
[c j]	517	3,917,230	0.29–0.53	0.42	1.08	0.96	1.21
	478	4,169,923	0.53–0.87	0.66	0.97	0.87	1.09
	777	5,146,393	0.87–2.09	1.24	1.09	0.98	1.2
	1140	5,520,772	≥2.09	3.63	1.14	1.04	1.24

[a] Dose estimated as calculated midpoint.
[b] OR for second exposure group was originally 1 but changed to 1.02 by log-transforming the upper and lower confidence limits and exponentiating the midpoint of the two log-transformed confidence limits.
[c] Nitrate values were multiplied by 0.2259 to convert Nitrate-NO_3 to Nitrate-N.
[d] Converted mg/day to mg/L by dividing by average water consumption of cases (1.4 L/day).
[e] Exposure values were originally measured in mmol/L and converted into mg/L (multiplied by 14.0067).
[f] Dose estimated as median reported in the study.
[g] Case and control numbers are an approximation based on total sample size and percentages reported for each exposure group.
[h] Dose estimated as mean reported in the study.
[i] 25,736 women in the at-risk cohort.
[j] Dose estimated as median based on data received through personal communication with the authors.

published methodology (Malits et al., 2018), we estimated indirect costs due to lost productivity caused by IQ loss associated with low birth weight to be 172 million to 232 million dollars, at $11,745 – $15,883 (2014 dollars) per IQ point loss, for 2939 annual nitrate-attributable very low birth weight cases. Other economic costs, such as parental lost work days are not accounted for in this analysis, and the overall costs of nitrate-attributable adverse birth outcomes are likely to be greater than what is estimated here.

For economic valuation of nitrate-attributable cancer cases, we first estimated hospitalization and medical treatment costs, which are the direct cost of medical resources to treat disease that can be ascertained from national health care cost statistics. Next, we estimated economic losses due to disability and premature death of patients with nitrate-attributable cancer, which represent harder to define indirect costs. Our analysis does not include society-level non-medical costs associated with the illness, such as the loss of work time and productivity as well as the loss of leisure time for family members of patients with the disease, due to difficulties in estimating such economic impacts.

Based on the National Cancer Institute data for the cost of treatment, we calculated that a range of 250 million to 1.5 billion dollars of medical costs in 2014 dollars could be due to the nitrate-attributable cancer cases (Table 3). For the indirect economic costs, we used the Disability-Adjusted Life Years methodology, combined with the Value of Life Year approach. For all cancers combined, nitrate-attributable loss of years of life due to disability and premature death corresponds to the estimated range of 21,663 to 112,628 annual nitrate-attributable DALYs (Table 3). Using a published estimate of $57,757 (in 2014 dollars) for the Value of Life Year (Desaigues et al., 2011), this translates to 1.3 billion to 6.5 billion dollars in annual indirect economic losses.

3.3. Meta-analysis of colorectal cancer studies

Based on the risk estimates reported in epidemiological studies and potential number of nitrate-attributable cases calculated here, we concluded that colorectal cancers pose the greatest risk linked to exposure to nitrate in drinking water relative to other cancer sites, and thus presents an area where a meta-analysis would be warranted to define the exposure-response relationship.

In total, nineteen studies were returned based on our search query in Pubmed, of which 12 were relevant to our study question and eight

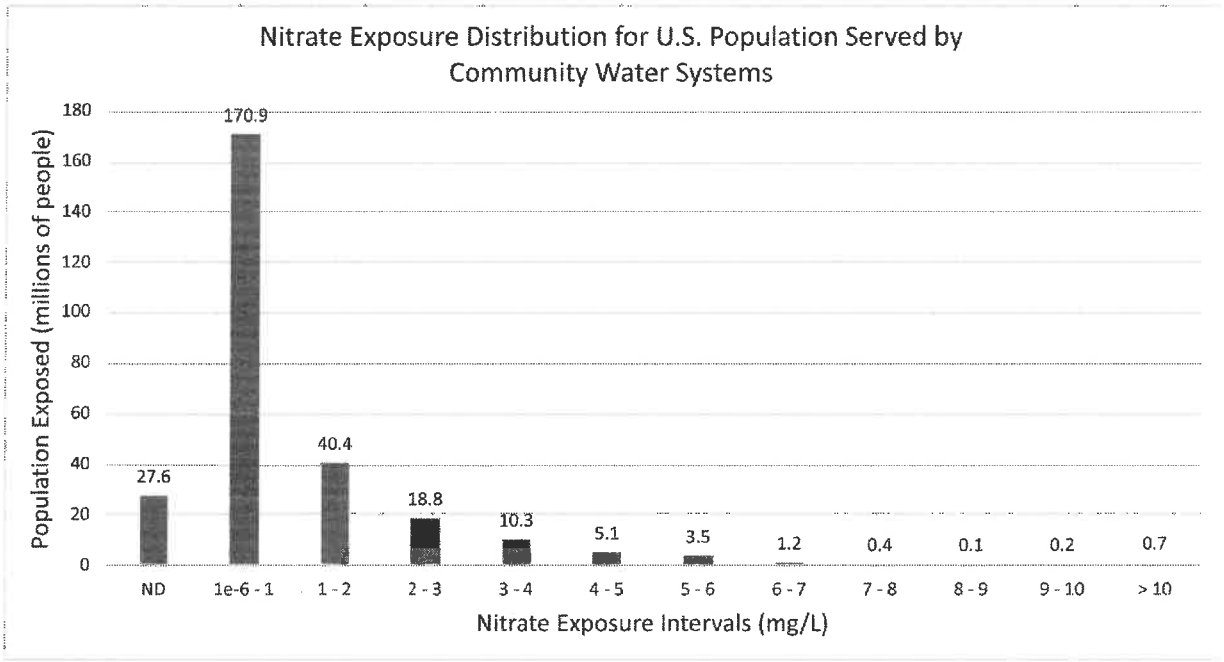

Fig. 1. U.S. population distribution for exposure to nitrate in drinking water at specific concentration ranges. Data from 2010 to 2017 for community water systems for all 50 states. The lower range of the nitrate exposure intervals represents the lowest average calculated. ND = non-detect. Source of data: Environmental Working Group Tap Water Database (https://www.ewg.org/tapwater/).

met our inclusion criteria (Supplementary Table 3). Of the studies not included, Morales-Suarez-Varela et al. (1995) and Gulis et al. (2002) were both ecological studies while Chang et al. (2010) included the same study population controls used in another publication already included in the meta-analysis (Chiu et al., 2010). Kuo et al. (2007) only assessed rectal cancer risks yet observed a significant increase in rectal cancer mortality for those exposed to a 0.72 mg/L median level of nitrate.

Findings of these studies were similar to others included in the meta-analysis, where the majority found positive associations between nitrate exposure in drinking water and colorectal cancer. Gulis et al. (2002) observed a positive trend for increased colorectal cancer in women exposed from low to high nitrate levels. Chang et al. (2010) found an increased risk of rectal cancer mortality at low concentrations of nitrate in drinking water (> 0.38 mg/L). Morales-Suarez-Varela et al. (1995) found no association between nitrate in drinking water and colon cancer mortality but did observe a statistically significant increase in risk of death from gastric cancer. Of the remaining eight studies, six were case-control studies resulting in a total of 8739 colorectal cancer cases and 12,219 controls, and two were cohort studies resulting in 4000 colorectal cancer cases over 1,758,862 person-years included in the meta-analysis.

Results of the generalized least squares regression analysis yielded positive study specific slopes for six studies, while negative study specific slopes were observed for the other two (Table 5). A study by Weyer et al. (2001) observed an increased risk in the second and third exposure groups, but a decreased risk in the highest exposure group, resulting in an overall negative slope. Overall, the dose response analysis of all studies (Fig. 2) yielded a statistically significant positive linear association between nitrate in drinking water and increased colorectal cancer risk, RR = 1.04 (95% CI 1.01-1.07) and a significant pooled linear slope estimate of 0.04 per mg/L increase (95% CI 0.009-0.072) (Table 5).

We observed substantial heterogeneity in our analysis (I^2 = 69.1%, p = 0.0002). Within the meta-analysis framework, heterogeneity can come from inconsistencies of study findings as well as study quality and study characteristics such as design and sample size (i.e. case control or cohort), geographic region (United States vs. Europe vs. Asia), and other variables explored in Table S5 and the literature (Camargo et al., 2011). Given the substantial amount of heterogeneity in the pooled estimate, an assessment was done to identify the source of heterogeneity among the studies by grouping studies based on certain

Table 5
Study specific dose-response slope estimates from general least squares regression and pooled slope estimate from meta-analysis of colorectal cancer risk and drinking water nitrate.

Study	Regression slope	Regression slope 95% Confidence Interval lower limit	Regression slope 95% Confidence Interval upper limit	Standard error
Case-Control Studies				
De Roos et al. (2003)	0.014	−0.034	0.062	0.025
Espejo-Herrera et al. (2016)	0.161	0.089	0.233	0.037
Chiu et al. (2010)	0.144	0.03	0.258	0.058
Yang et al. (2007)	−0.029	−0.256	0.198	0.116
Fathmawati et al. (2017)	0.046	0.003	0.09	0.022
McElroy et al. (2008)	0.026	−0.004	0.055	0.015
Cohort Studies				
Weyer et al. (2001)	−0.43	−0.108	0.021	0.033
Schullehner et al. (2018)	0.034	0.014	0.053	0.010
Pooled				
All studies	0.04	0.009	0.072	

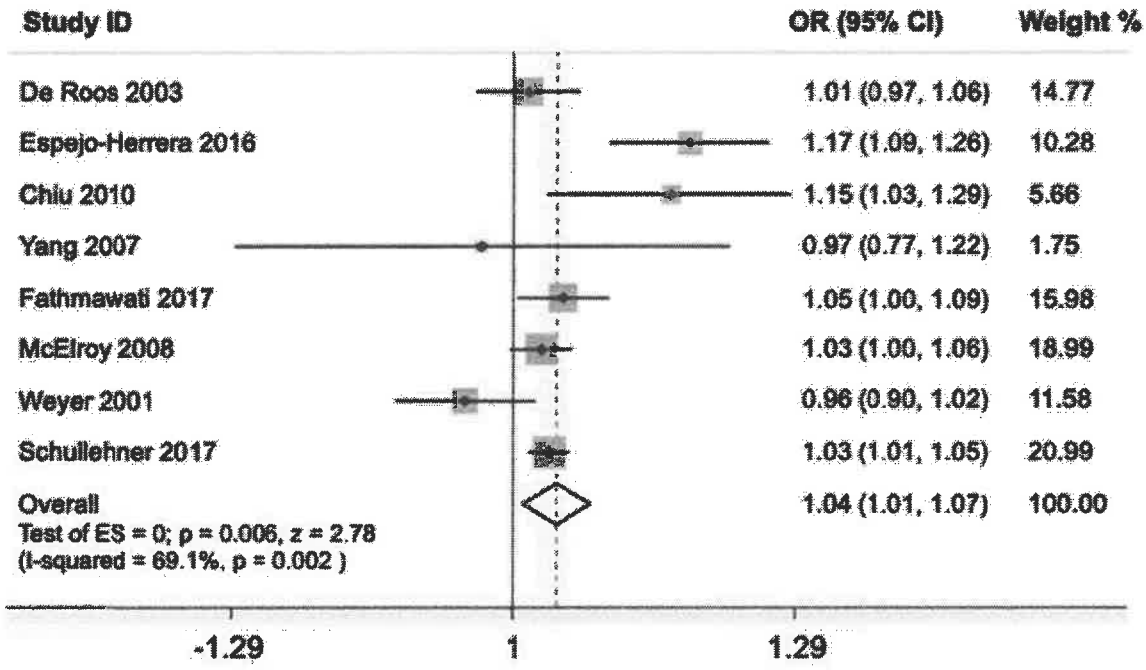

Fig. 2. Odds ratio (OR), 95% confidence intervals (95% CI), study weight within the overall meta-analysis and overall risk estimate based on studies of nitrate exposure from drinking water and colorectal cancer risk. ORs were obtained by exponentiating the study-specific slope estimates from generalized least squared regression to obtain log risk ratio estimates per mg/L increase in nitrate.

covariables as well as omitting single studies from the analysis (Supplementary Table S5 and S6).

Calculated pooled slopes from other study combinations based on covariables did not clearly identify a meaningful study covariable for which to attribute heterogeneity. There was some indication that for studies which did not account for dietary factors, a reduced slope estimate as well as reduced heterogeneity was observed. Additionally, these calculated slopes for analyses typically including more than two studies were within the 95% confidence intervals of the analysis including all eight selected studies (Supplementary Table S5).

After omitting single studies and rerunning the meta-analysis, one study in particular, Espejo-Herrera et al. (2016), was identified as the major source of statistical heterogeneity in the pooled analysis. Removing Espejo-Herrera from the pooled assessment reduced the heterogeneity (I^2 value) to 41.7%, which was no longer significant ($p = 0.113$). Given that Espejo-Herrera observed the greatest positive linear dose response for nitrate and colorectal cancer risk, the pooled slope estimate from the remaining seven studies was slightly reduced relative to the eight study meta-analysis, 0.027, yet remained statistically significant (Supplementary Table S6; Test of effect size = 0, $p = 0.019$). Espejo-Herrera used a strong study design that included exposure assessment from public water supplies, private wells and bottled water; accounted for factors that influence endogenous nitrosation; and pooled data from two European cohorts, increasing sample size. Given the high quality of this study, its inclusion is important to the calculation of the pooled slope. For the purposes of using this information to calculate a range of drinking water health benchmarks, it was determined that while removing Espejo-Herrera from the meta-analysis, statistically improves the heterogeneity, such an analysis would not accurately reflect the strength of evidence within the epidemiological literature and a more relevant analysis was not gained by omission of this study in an effort to reduce overall heterogeneity.

3.4. Risk-based drinking water benchmarks for nitrate

Based on the estimated nitrate-attributable colorectal cancer cases and colorectal cancer meta-analysis, we calculated an array of drinking water benchmarks corresponding to an annual one-in-one-million cancer risk (Table 6). First, we used our estimated nitrate-attributable cancer cases (Table 2), expressed as additional cases per million people at a given nitrate concentration to linearly extrapolate a concentration corresponding to one additional case of nitrate-attributable cancer per million people. This approach results in values that range from 0.04 to 1.3 mg/L. Similar to the nitrate attributable colorectal cancer cases, the lower range is derived from Espejo-Herrera et al. (2016) while the upper range is derived from De Roos et al. (2003). Second, using the cancer slope factor of 0.04 per mg/L increase in nitrate corresponding to pooled slope estimate from the meta-analysis, and following the equation outlined in section 2.8, results in a drinking water nitrate concentration of 0.14 mg/L (95% CI 0.08-0.63 mg/L) as the central estimate for annual one-in-one-million cancer risk level.

4. Discussion

Epidemiological data suggest that nitrate impacts on human health may occur at nitrate concentrations present in drinking water in the United States today. Among health impacts observed in epidemiological studies of nitrate in drinking water, colorectal cancer shows the strongest association, based on long-term studies with large numbers of study participants. National Cancer Institute statistics show that colorectal cancer is the fourth most prevalent cancer in the United States, with over 1.3 million people living with colorectal cancer in 2015 and 140,250 new cases estimated for 2018 (SEER, 2018). Recent trends suggest that both incidence and mortality due to colorectal cancer are decreasing slightly, with 2.4% and 2.6% decrease over the last decade, respectively (SEER, 2018). Yet, given the numbers of people affected by colorectal cancer, it remains imperative to continue research into risk factors for this disease and measures that can be taken to address them. Smoking, physical inactivity, high dietary intake of red meat and consumption of processed, nitrate-preserved meats are some of the known risk factors for colorectal cancer. Detection of additional risk factors and identification of measures to eliminate such risk would help decrease the health and economic impacts of colorectal cancer on society.

Through a combination of targeted study review and meta-analysis,

Table 6
Nitrate concentrations corresponding to one-in-one-million annual colorectal cancer risk derived from nitrate-attributable cancer case analysis and from meta-analysis.

Source of risk estimate	Nitrate cut-off concentration used in nitrate-attributable cancer case analysis (mg/L, Table 2)	Estimated nitrate- attributable cases per million at cut-off concentration [a]	Extrapolated concentration for annual one-in-one-million cancer risk [b]	Meta-analysis derived one-in-one-million cancer risk (95% Confidence Intervals) [c]
Nitrate-Attributable Cancer Case Analysis				
Espejo-Herrera et al., 2016 - A	1.7	30.70	0.06	
Espejo-Herrera et al., 2016 - B	0.7	18.51	0.04	
Schullehner et al., 2018 - C	0.9	11.94	0.08	
Schullehner et al., 2018 - D	2	7.71	0.26	
De Roos et al., 2003 - E	5	3.86	1.29	
Meta-Analysis				0.14 (0.08–0.63)

[a] Attributable cases per million was obtained by dividing the estimated number of nitrate-attributable cases by the total U.S. population from 2017 census estimates (325,719,178 people).
[b] Concentration corresponding to annual one-in-one-million cancer risk was obtained by dividing the nitrate concentration for a specified scenario (values in column 2) by the estimated number of attributable cases per million people in the U.S. population (values in column 3).
[c] Concentration corresponding to annual one-in-one million cancer risk obtained using the cancer slope factor derived from a meta-analysis of colorectal cancer risk and nitrate in drinking water and the equation listed in Methods section 2.8.

we developed a risk estimate of drinking water nitrate-attributable colorectal cancer in the United States. Our data suggest that exposure to nitrate in drinking water could account for 1–8% of total colorectal cancer cases, which translates into 1233–10,379 cancer cases annually. Of these cases, 12–24% are due to nitrate exposure for private well users, especially for people whose well water has 5 mg/L or more nitrate.

Given that our study focused on nitrate occurrence data in drinking water for 2010–2017 and that cancer is a disease with long latency, the findings presented in this study are most relevant for future cancer prevention efforts. Additionally, our analysis includes some uncertainty around the exact number and exposure information for people served by community water systems and private wells. However, we note that the impact of the population adjustment factor used here to account for this uncertainty is smaller than the variability observed in the range of reported risk estimates for nitrate-attributable diseases, and thus unlikely to influence significantly the disease case estimates presented here. Additional limitations in our estimation of nitrate-attributable disease cases and associated economic costs come from the assumption of causality necessary to perform such an analysis. Published studies have suggested that the lower limits of the health costs and exposure attributable cases may be zero (van Grinsven et al., 2010; U.S. EPA, 2005).

Exposure to drinking water contaminants has been described as a risk factor for other cancers such as exposure to drinking water disinfection byproducts and bladder cancer. In fact, using the risk estimate and slope calculated based on meta-analysis by Villanueva et al. (2003), the U.S. EPA (2005) estimated the annual number disinfection byproduct-attributable bladder cancer cases as 8899 (95% CI 4830-15,376). This estimated number of cancer cases is comparable to the number of nitrate-attributable colorectal cancer cases we present here. Of note, disinfection byproduct-attributable bladder cancer cases represent a greater percentage of the population-attributable fraction for this cancer site, approximately 16%, than nitrate-attributable colorectal cancer cases, 1–8%. This could be due to the steeper cancer slope factor for disinfection byproducts, 0.006 per μg/L increase compared to our calculated cancer slope factor for nitrate of 0.00004 per μg/L increase when expressed in the same units.

The latest research has produced strengthened epidemiological evidence for the risk of colorectal cancer at nitrate levels below the regulatory standard of 10 mg/L of nitrate as nitrogen. Even a small increase in risk, as suggested by our meta-analysis, can lead to large population-attributable risk and a large number of disease cases that could be avoided if these exposures were prevented (Rose, 2001). Additionally, our economic analysis suggests that this attributable risk to a large population also comes at large economic costs, initially felt by individuals and families as direct medical costs, and eventually translating into overall economic loss for the society because of loss of work time and productivity. The medical impacts for cancer treatment are particularly significant for the United States because patients in the U.S. may personally bear all or a large portion of these medical costs because of the lack of health insurance or limited coverage under existing insurance plans.

Current estimates for the annual prevalence cost of colorectal cancer in the United States is $14.1 billion (Yabroff et al., 2012). Based on our estimates that 1 to 8 percent of colorectal cancer cases could be attributed to nitrate exposure, expected medical costs would be $141 million to $1.1 billion, which is also reflected by the estimated medical costs presented in this study of $157 million to $1.3 billion. Given the increasingly aging population and the advancement of medical treatments, the annual costs of cancer are expected to grow 27 to 39 percent between 2010 and 2020 (Mariotto et al., 2011), highlighting the need for prevention strategies geared towards reducing the cancer burden.

For the calculation of indirect economic costs, we used a combination of the Disability-Adjusted Life Years approach together with the Value of Life Year (VOLY) valuation. Here we used a VOLY value of

$57,757 in 2014 US dollars, based on recent research literature (Desaigues et al., 2011; Grandjean and Bellanger, 2017; van Grinsven et al., 2010). It is possible that the VOLY value derived from these studies is underestimated. For example, an alternative VOLY value developed by the Institute for Clinical and Economic Review (2017) defined the value of one Quality-Adjusted Life Year between $100,000 and $150,000 with the median value of $125,000 was considered for use in this analysis. Additionally, in an assessment of economic loss due to cancer deaths in the United States, Yabroff et al. (2008) used a VOLY of $150,000. If either of these values were used, our calculated nitrate attributable economic losses would be up to approximately $12.8 billion (not adjusted for inflation). While such economic analyses produce only approximate estimates, the overall data presented form a solid foundation for the argument that existing levels of nitrate in U.S. drinking water may drive negative health and economic impacts on society and that lowering nitrate exposure from drinking water would protect public health.

For additional point of comparison, we note that U.S. EPA uses a different methodology for calculating the costs of environmental pollution, namely the "Value of Statistical Life" approach. A broad spread of estimates for the Value of Statistical Life ranging from $1 million to $10 million (2000 dollars) is reported in the literature (Viscusi and Aldy, 2003). In recent reports, U.S. EPA has recommended using a Value of Statistical Life of $7.9 million (in 2008 dollars) (U.S. EPA, 2010), while in a 2015 regulatory impact assessment, a value of $10 million was used (U.S. EPA, 2015b). There are scientific uncertainties around applying the Value of Statistical Life approach for the calculations of indirect economic loss due to cancer, since not every cancer case results in mortality. For the 2300 to 12,594 annual nitrate-attributable cancer cases calculated here, a Value of Statistical Life of $1 million translates into $2.3-$12.6 billion in indirect economic losses due to nitrate pollution of drinking water, while the Value of Statistical Life of $10 million would result in 10 times greater amount in indirect economic losses.

For the purposes of cost-benefit analysis, the estimates for the range of direct and indirect costs due to nitrate in drinking water can be compared with the costs of removing nitrate from drinking water. Based on the published methodology for estimating nitrate treatment costs per 1000 gallons of water treated (Jensen et al., 2012), a study published online by Environmental Working Group estimated that if all U.S. communities with drinking water nitrate concentrations at or above 5 mg/L, which lacked nitrate treatment as of 2014–2015, added ion exchange systems for nitrate removal, the total extra cost would range from about $102 million a year to almost $765 million a year (Weir Schechinger and Cox, 2018). If each of these communities without nitrate treatment opted for a reverse osmosis water treatment system instead, the added cost could be as high as $1.47 billion a year. These costs are particularly significant for small rural communities where water systems often lack funds for capital improvement. According to the same analysis, as much as $666 a year per person is added to the cost of providing drinking water in a very small community, while a reverse osmosis system could add as much as $2776 a year (Weir Schechinger and Cox, 2018).

Studies by the U.S. Geological Survey have pointed out a rising trend in nitrate concentrations in groundwater, particularly in the agricultural areas (Pennino et al., 2017; Rupert, 2008), and the number of nitrate-attributable disease may grow in future years. Every year, nitrogen-based fertilizer is spread in farming areas, and a significant portion of that nitrogen ends up as nitrate in surface water and ground water supplies that communities small and large depend on as a source of their drinking water. Nitrate contamination present in the ground water would likely stay there for years or decades, and the exposures identified in this study would likely continue or become more severe if nitrate removal technologies are not utilized.

Finally, our study has used two approaches to calculate a risk-based drinking water benchmark for nitrate. First, based on nitrate-attributable cancer cases from three studies, we calculated an array of cancer-based drinking water benchmarks for nitrate that range from 0.04 to 1.3 mg/L (Table 6). For a statistically valid central estimate of the one-in-one-million risk level, we used the cancer slop estimate for nitrate derived from a meta-analysis, to yield of value of 0.14 mg/L (95% CI: 0.08–0.63 mg/L). These benchmarks are based on annual background rates of colorectal cancer and therefore correspond to annual one-in-one-million cancer risk. Our heterogeneity analysis indicated that removing Espejo-Herrera from the pooled analysis would reduce the study heterogeneity. Using the pooled slope estimate and 95% confidence intervals from the seven study meta-analysis would still produce drinking water guideline values within this range.

In practice, regulatory agencies have considered a lifetime one-in-one-million risk (OEHHA 2018b) as the *de minimus* risk acceptable for general public exposure to cancer-causing chemicals. At lifetime risk level of one-in-one-million implies that not more than one person in a population of one million people drinking the water with the specified contaminant concentration daily for 70 years would be expected to develop cancer as a result of exposure to that chemical. Different government agencies use different risk frameworks and the choice of a specific risk level may depend on the specific policy context. For example, 10^{-6} risk level is used by the state of California for the development of public health goals for cancer-causing drinking water contaminants (OEHHA 2018b), while the state of Minnesota uses a 10^{-5} risk level for setting the water benchmarks for cancer-causing contaminants (Minnesota Administrative Rules Part 4717.7840).

Questions remain about the appropriate translation of the annual cancer risk benchmark into a lifetime benchmark. If the cancer risk were linear throughout the range of possible exposure concentrations and duration of exposures, then one could calculate the lifetime cancer risk benchmark by dividing the annual cancer risk benchmark by factor of 70, the length of life used in regulatory risk assessments or by using a lifetime background cancer rate, expressed as the number of cancer deaths divided by the number of total deaths. Future studies of the dose-response relationship for nitrate may help clarify whether such an approach can be used for deriving lifetime cancer risk benchmark for nitrate. As typical for epidemiological studies, data presented and analyzed here are suggestive but not conclusive for establishing causality and defining the dose-response function. To address this uncertainty, we present the calculations from the meta-analysis in the context of an array of estimates calculated based on relative risk reported by individual, high-quality epidemiological studies coupled with real nitrate exposure data that reinforces our confidence in the final assessment.

Another approach for derivation of drinking water benchmarks for nitrate can come from the consideration of non-cancer effects of nitrate exposure, specifically the effects on the developing fetus (Stayner, 2017b). These risks apply to nitrate exposure during pregnancy, which is a relatively short period of exposure and a window of greater vulnerability. Such epidemiological studies likely have greater reliability for the derivation of human-health protective water benchmarks, because they eliminate uncertainties due to interspecies extrapolation from laboratory animals to humans. On the other hand, uncertainty factors, sometimes also considered safety factors, may be appropriate for the assessment that involves LOAEL (Lowest Observed Adverse Effect Level) to NOAEL (No Observed Adverse Effect Level) extrapolation, where U.S. EPA-defined default uncertainty factor is 10 (U.S. EPA, 2002). Additionally, in some circumstances a children's health protection factor of 10 may also be warranted, to account for children's greater susceptibility to toxic chemicals (National Research Council, 1993). Applying a single uncertainty/safety factor of 10 to the two departure points for nitrate's developmental effects, 1 mg/L from Stayner et al. (2017a) and 4.5 mg/L from Brender et al. (2013), results in drinking water benchmarks of 0.1–0.45 mg/L, respectively. These values are consistent with health benchmarks developed on the basis of annual cancer risk due to nitrate.

Two key uncertainties remain, namely, the shape of the dose-response curve and the concentration of the nitrate in the water where no adverse effects would be observed. These can only be addressed by future toxicology and epidemiology studies. The topics of threshold effects and the shape of dose-response curve for environmental contaminants have been hotly debated in the risk assessment literature for decades (National Research Council 2009; Zeise et al., 1987). However, these uncertainties do not preclude the need to search for pragmatic solutions to water quality problems and nitrate pollution of water supplies that are faced by communities today.

Availability of data and materials

The U.S. nitrate occurrence dataset analyzed in this study is posted in an open access database available at https://www.ewg.org/tapwater/.

Declarations of interest

None.

Authors' contributions

All authors have made substantial contributions to conception and design of this research, analysis and interpretation of data, and manuscript preparation.

Submission declaration

This work is original, has not been previously published and is not under consideration for publication elsewhere.

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Ethics approval and consent to participate

Not applicable.

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Appendix A. Supplementary data

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