Nitrate in drinking water and human health

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Agriculture at Risk: A Report to the Nation (1988) included a brief discussion of nitrate in the environment as a risk factor for human health. Specifically, the contribution from nitrogen-containing fertilizers to high levels of nitrate in food and drinking water was identified as an environmental health concern. Methemoglobinemia (blue-baby syndrome), various cancers and birth defects were listed as possibly being associated to exposure to elevated nitrate levels in drinking water. The authors of the report suggested a number of control strategies to limit the amount of nitrate entering groundwater supplies. Prevention of groundwater contamination at the source was cited as "the most effective and least costly control strategy available today." [1] However, little progress has been made since that time towards minimizing nitrate inputs into the environment. The increasing contamination of municipal wells and private well water (and surface water supplies) by nitrate, primarily from the widespread use of commercial fertilizers as well as human and animal waste, has been documented in many areas of the U.S. In Iowa, long term use of nitrogen fertilizers in both rural and urban areas has resulted in 30-40% of finished public water supplies with nitrate-nitrogen (NO_3-N) concentrations in excess of 5 mg/L, or parts per million (ppm). [2] The current U.S. Environmental Protection Agency maximum contaminant level (MCL) for public drinking water supplies is 10 ppm nitrate. (Nitrate in this paper refers to NO_3-N).

Exposure to nitrate *per se* is not of particular interest with respect to human health. However, nitrate can be reduced endogenously (within the human body) to nitrite through bacterial and other reactions; nitrite can be further reduced to N-nitroso compounds (NOCs). Infant (< 6 months of age) exposure to nitrite has been linked to development of methemoglobinemia. NOCs are some of the strongest known carcinogens, [3] can act systemically, [4] and have been found to induce cancer in a variety of organs in more than 40 animal species including higher primates. [5] While some vegetables (lettuce, spinach, celery, greens, etc.), contaminated drinking water, cigarette smoking, and certain medications all contribute to daily nitrate intake in the U.S. population, [6] drinking water can account for a substantial portion of that intake.

This paper will present what is known about the human health risks from long-term exposure to low levels (ppm) of nitrate in drinking water. An overview of the extent of nitrate contamination of water supplies will focus on Iowa. A brief discussion of how nitrate works in the human body will set the stage for a detailed description of what is known about health effects from exposure to nitrate in drinking water, focusing on both non-cancer and cancer outcomes. Finally, future needs with respect to research and education will be presented.

Occurrence of nitrate in Iowa water supplies

Iowa is a predominantly agricultural state, producing mainly corn, soybeans and hogs. In the 1990s, an average of 11-12 million acres were annually planted to corn. Table 1 shows the annual use of nitrogen fertilizer on corn in Iowa from 1991-1996; Iowa ranked second in the corn-belt behind Illinois in total tonnage nitrogen used during that time.

Table 1
Use of nitrogen fertilizers on corn in Iowa: 1991 –1996

Year	1,000 acres of corn	% planted w/ nitrogen	Lbs. Nitrogen / acre	Total nitrogen million lbs.
1991	12,500	98	120	1,469
1992	13,200	96	118	1,497
1993	12,000	98	114	1,335
1994	13,000	98	121	1,548
1995	11,700	97	120	1,365
1996	12,700	98	132	1,632

Source: USDA Agricultural Statistics Program

Nitrate in the environment comes from both rural and urban sources. Soil mineralization and atmospheric deposition of nitrogen are major contributors of nitrate to the environment. While urban use of nitrogen on lawns and gardens can contribute significant amounts, agricultural inputs are more important sources. [7] Results of a US Geological Survey (USGS) study of nitrogen contribution to the Gulf of Mexico are presented in Table 2. It has been estimated that 20% of the nitrate in the Mississippi River comes from Iowa. [8] Nutrients that are not fixed in the soil or used by plants can leach into shallow groundwater or run off into surface water during rainfall events. Other sources of nitrate include animal waste from livestock operations and urban areas, and human waste from septic systems and municipal wastewater treatment plants.

Table 2
Estimated nitrogen contribution to the Gulf of Mexico from specific sources

Source of nitrogen	% of nitrate flux from the source	% of total nitrogen flux from source
Fertilizer and mineralized soil nitrogen	58 +/- 9	50 +/- 9
Animal manure	16 +/- 9	15 +/- 10
Other, including atmospheric deposition, groundwater, soil erosion, and urban runoff	16 +/- 6	24 +/- 6
Municipal and industrial point sources	9 +/- 2	11 +/- 2

Source: Goolsby et al., 1999.

A 1996-98 study by the USGS National Water Quality Assessment (NAWQA) Program documented high levels of nitrate in several eastern Iowa river basins, finding that these streams ranked in the upper 25th percentile nationally for median nitrate concentrations. [9] Although lower than surface water levels, nitrate concentrations in alluvial aquifers in agricultural areas in eastern Iowa were greater than groundwater sampled elsewhere within the corn belt. [9] The NAWQA study found that the type of land use also affects shallow groundwater quality. In Iowa, nitrate was found in 94% of samples taken from shallow alluvial aquifers in agricultural areas compared to 77% of samples taken in urban areas, with a median nitrate level of 5.1 ppm in agricultural areas compared to a median of 1.8 ppm in urban areas. [9] Additionally, 39% of samples taken from shallow alluvial aquifers in agricultural areas exceeded the MCL of 10 ppm, while none of the samples taken in urban areas exceeded the MCL. [9]

Information on public water supplies from the Iowa Safe Drinking Water Act database (1988-95) shows that approximately 30-40% of public water supplies had water samples with nitrate levels >5 ppm. [2] Earlier historical data on Iowa municipal water supplies document an increase in nitrate levels since the mid-1950s. A study of almost 400 Iowa community water supplies shows an increase in nitrate levels in drinking water supplies from 1955-64, which peaks in the 1980s and then drops slightly through the 1990s (Table 3). [10]

Table 3
Nitrate in community drinking water supplies, Iowa Women's Health Study

NO_3-N concentration	1955-64[a]	1976-82[b]	1983-88[b]	1989-2000[b]
Median (ppm)	0.40	1.11	5.11	3.82
Mean (ppm)	1.31	5.94	5.76	5.26

Source: CHEEC
a: raw (source water) and finished water
b: finished water only

In 1988-89, the Iowa Statewide Rural Well Water Survey collected water samples from about 685 private rural wells across the state. Based on the results of that survey, it was estimated that 18.3% of private rural drinking water wells were contaminated with nitrate in excess of 10 ppm. [11] These data show that residents of Iowa (and potentially, residents of other corn belt states) have experienced long term exposure to nitrate in drinking water supplies, both from municipal water systems as well as private wells.

Nitrate within the human body

Nitrate is considered to be of low toxicity, but nitrite and NOCs are biologically active in mammalian systems. As reduction (nitrosation) of nitrate produces nitrite and ultimately NOCs, it is important to consider the various exogenous sources of nitrate exposure to the human body. Dietary nitrate, mainly in vegetables and processed meats, constitutes a major source of ingested nitrate. Nitrate levels in vegetables vary depending on the type of vegetable and how it is grown and stored. Concentrations in fresh vegetables are fairly low, generally in the range of 1-2 mg/kg and rarely over 10/mg/kg. [12] It has been estimated that vegetables can provide up to 85% of the average daily human dietary intake of nitrate. [13] Other dietary sources include cured meats, which contain considerable amounts of nitrate and nitrite from the curing process and from added preservatives.

While nitrate from food is a main source for human exposure, nitrate from drinking water also plays an important role. Elevated nitrate levels in drinking water may result in higher body burdens of nitrate in people consuming that water. For example, compared to persons who are not exposed to nitrate in drinking water, persons consuming water with 10 ppm nitrate have about twice the total nitrate intake. At 20 ppm nitrate the total intake is threefold and above 20 ppm nitrate drinking water is estimated to contribute over 80% of dietary nitrate intake. [14] Nitrate in drinking water is positively associated with urine nitrate levels as well as excretion of nitrosoproline, a biomarker of endogenous nitrosation. [14, 15] Other dietary variables and lifestyle factors can impact nitrate in the body. Cigarette smoking accelerates nitrosation reactions, while intake of vitamin C or E within 1-2 hours of consumption of a nitrate source inhibits these reactions. [16]

Mammalian metabolic processes can form nitrate endogenously. It has been estimated that about 1 mg/kg body weight/day is formed in humans; that amount may be substantially increased by the presence of bacterial infection. [13] The process of nitrosation is a complex series of chemical reactions which ultimately results in the production of NOCs. In essence, nitrate is a precursor to nitrite, which forms via bacterial reduction in the saliva, stomach, large intestine or infected urinary bladder. Nitrite then reacts with nitrosable substrates (amines, amides and amino acids) to produce NOCs. Ingested nitrate is readily absorbed from the small intestine and rapidly distributed through the body. [17, 18] Blood nitrate is selectively transported and secreted in the saliva by an active transport system. [19] An estimated 25% of ingested nitrate is secreted in the saliva. [20] In addition, almost 65-70% of orally ingested nitrate is excreted in the urine. [18] Thus, about 5% of total dietary nitrate is available for reduction to nitrite in the saliva.

Mammalian nitrate reductase activity can account for half the reduction of nitrate to nitrite, the other half being affected by microorganism enzyme action. NOCs may then be synthesized in the stomach from reingested nitrite and secondary amines also present in food. The presence of certain iatrogenic bacteria enhances nitrosation in the gut. [21] The fate of ingested nitrate in the body is not totally understood. Gangolli, in an excellent overview of the toxicology of nitrate, nitrite and NOCs, states that “The toxicokinetics and metabolic disposition of nitrate, nitrite and NOC are closely inter-related and have a pivotal role in influencing the bioavailability and toxicity of nitrite and NOC derived from ingested nitrate.” [13]

Non-cancer health effects from exposure to nitrate in drinking water

The development of methemoglobinema in infants is perhaps the most well documented health outcome resulting from acute nitrite toxicity, which may result from reduction of ingested nitrate to nitrite. The EPA’s drinking water MCL of 10 ppm nitrate was established due to concerns about methemoglobinemia. The process involves nitrite mediating the oxidation of the heme ion in hemoglobin, which causes a change from the ferrous to the ferric state. The resulting methemoglobin cannot combine reversibly with oxygen or carbon dioxide, potentially resulting in anemic hypoxia. This inability of the blood to effectively transport oxygen and carbon dioxide can result in acute distress to the system, and in severe cases can cause a bluish-tinge in the skin color, hence the term “blue baby”. The World Health Organization cited numerous cases of nitrite intoxication following ingestion of well water containing high levels of nitrate, almost 98% of which were associated with nitrate levels in the range of 44-88 ppm. [22] Very few cases of methemoglobinemia are reported in the U.S. Common complicating factors in determining the role of nitrate in the development of methemoglobinemia include the prevalence of bacterial contamination of a water supply and/or co-existing bacterial infection in the infant.

An ongoing debate centers on the importance of nitrate in drinking water as a risk factor for methemoglobinemia. [23, 24, 25]

Other non-cancer health outcomes associated with exposure to nitrate in drinking water have been reported. A German study found an increased incidence of hyperthyroidism (goiter) in persons drinking water from shallow wells with high nitrate levels compared to persons drinking water from deeper wells with low nitrate levels. [26] A Colorado study reported an increased risk of developing insulin dependent diabetes in residents of counties whose water supplies had nitrate levels between 0.77–8.2 ppm, compared to counties with water nitrate levels < 0.77 ppm. [27]

Maternal transfer of nitrate, nitrite and NOCs is suggested by a number of studies on reproductive outcomes tentatively linked to high nitrate levels in water supplies. An Australian study reported an increased risk for central nervous system malformations in infants whose mothers consumed drinking water from private wells with nitrate levels at 26 ppm. [28] Another Australian study reported nitrate levels in drinking water between 5-15 ppm and >15 ppm associated with 3 and 4 times the risk of delivering malformed children compared to an area where nitrate concentration in the water supply was <5 ppm. [29] A California study found an increased risk for neural tube defects (anencephaly) in babies of women who consumed drinking water with nitrate levels >10 ppm during pregnancy. [30] However, the association was only seen in women who consumed groundwater only (not mixed water supplies with similar nitrate concentrations); the authors concluded that nitrate may not be the exposure of interest.

Anecdotal reports of reproductive effects are also found in the literature. The CDC's Morbidity and Mortality Weekly Report described a case study in LaGrange County, Indiana, where three women experienced a total of six spontaneous abortions during a two year time period (1991-93). [31] These women resided in proximity to each other and consumed drinking water from private wells that contained high levels of nitrate (19-26 ppm). There were several other women residing in the area who gave birth to full-term babies; none of these women had high nitrate in their water supplies.

Finally, two studies reported that nitrate in drinking water may have genotoxic effects in persons consuming that water. A study of Greek children residing in areas with drinking water nitrate levels between 55.7-88 ppm found a significant increase in the mean number of chromatid/chromosome breaks in children who consumed water with nitrate levels >70.5 ppm. [32] Another study reported increased HPRT (hypoxanthine phosphoribosyltransferase gene) variant frequencies and endogenous formation of NOCs in persons who consumed well water with >25 ppm nitrate. [33]

Nitrate in drinking water and cancer risks

Public concern about possible links between nitrate in drinking water and adverse health effects has heightened in recent years, as coverage of nitrate contamination of ambient water and drinking water supplies has become a regular feature in the popular press. There is a considerable amount of experimental data suggesting a role for nitrate in the formation of carcinogenic NOCs. However, there is a lack of epidemiologic data addressing the possible association of nitrate in drinking water with cancer risk.

Most of the epidemiologic data are derived from ecologic studies, which have inherent weaknesses related to assigning aggregate exposures (generally at the community or regional level) to individuals. In addition, most previous ecologic studies had limited nitrate data, relied on cancer mortality (rather than incidence) data, and used residence at the time of death as a surrogate for lifetime residence. In addition, few studies took into account an induction (or latency) period. Finally, ecologic studies rarely account for other possible risk factors for the disease under study. In essence, these studies are weak from a number of standpoints.

In general, ecologic studies of nitrate in drinking water as a risk factor for cancer have reported mixed findings. Studies of stomach cancer have been mixed, [34] while elevated risks for cancer of the esophagus, nasopharynx, urinary bladder, and prostate have been reported. [34, 35] However, an ecologic comparison of cancer incidence rates in northern England found that gastric and esophageal cancer rates in an areas with high water nitrate levels were comparable to rates in areas with low water nitrate levels. [36] The same study reported an increased incidence of adult brain and central nervous system tumors in the high nitrate areas. An ecologic study in Nebraska found a positive association with non-Hodgkin lymphoma, [37] while a study from Denmark [38] found no association for lymphatic or hematopoietic (blood or bone marrow) cancers. Two ecologic studies reported no association between nitrate in drinking water and risk for ovarian cancer. [38, 39] A positive association between municipal drinking water nitrate levels and risk for uterine cancer was reported in ecologic studies in Canada [39] and Denmark. [38]

Of particular interest is bladder cancer. As mentioned previously, 70% of orally ingested nitrate is excreted in the urine [6] and endogenous nitrosation occurs in the bladder. [40] Nitrosation byproducts appear in the urine after oral ingestion of nitrate in drinking water [15] and NOCs are carcinogenic in the bladder in laboratory animal studies. [41] Studies conducted in Canada [39] and Spain [42] both found no association with nitrate levels in drinking water and bladder cancer incidence or mortality, respectively, whereas another study from Spain [43] found a positive association for bladder cancer incidence, although the estimates were based on small numbers. Follow-up studies of workers involved in the manufacture of nitrogen-based fertilizers have found no association with bladder cancer. [44, 45, 46]

Few case-control studies have been conducted to evaluate nitrate in drinking water as a risk factor for cancer. The case-control study is a more powerful design than the ecologic study, as it accounts for individual exposures to factors (lifestyle, occupational, medical, dietary, etc.) that may impact the development of cancer. Cancer cases (and matched controls) are identified and surveyed for information on these variables, and are statistically compared for differences in risks for developing the disease. Case-control studies have reported mixed results for nitrate in drinking water and gastric cancer, [47, 48, 49] and null results for brain cancer. [50, 51] A population-based case-control study in Nebraska found long-term consumption (30 years) of community water with average levels of nitrate ≥4ppm (compared to <1.6 ppm) was positively associated with a two-fold risk for non-Hodgkin's lymphoma, and there was evidence of a dose response with increasing nitrate levels. [52]

At least two prospective cohort studies have investigated cancer and exposure to nitrate in drinking water. The prospective cohort is the most powerful of the epidemiologic study designs. In essence, the study group is defined at a specific point in time (prior to development of disease), surveyed for information on lifestyle, occupation, etc., and followed into the future to see what health problems they develop. A cohort study in the Netherlands found no association with gastric cancer incidence. [53] A cohort study of elderly women in Iowa reported an increased risk for bladder and ovarian cancer, an inverse association (protective effect) for rectal cancer and uterine cancer, and no association with several other cancers, including colon cancer and non-Hodgkin's lymphoma. [54] In this study, an almost three-fold risk was seen for bladder cancer in women who had consumed drinking water with nitrate concentrations >2.46 ppm for over 10 years, although the number of cases was quite small. As mentioned previously, biologic plausibility exists for exposure to exogenous nitrate and development of bladder cancer. The Iowa study did not corroborate the previous findings of an association with non-Hodgkin's lymphoma in Nebraska, [52] while the positive finding for ovarian cancer is in contrast to previous studies. [38, 39]

Future needs: research and education

Very few studies have looked at nitrate in drinking water and non-cancer health outcomes. The role of nitrate in drinking water in the development of methemoglobinemia in very young infants is under debate. Research proposals on evaluating the susceptibility of sensitive populations to acute effects of methemoglobin formation by other possible etiologic agents (nitrobenzene, dinitrotoluenes) in drinking water have been solicited by EPA. [55]

Studies investigating a possible association between exposure to nitrate in drinking water and the risk for cancer have reported mixed findings. Based on the research to date, it is very difficult to say with any certainty whether long term exposure to low levels of nitrate in drinking water is related to cancer development. Certainly, more research is needed in this area. Replication of studies such as the Nebraska study and the Iowa study is needed, to see if the positive finding for non-Hodgkin's lymphoma and bladder cancer are corroborated.

One limitation of previous cancer studies has been the use of an ecologic exposure variable for nitrate in drinking water. That is, the average nitrate level for a community drinking water supply for a specific time frame is assigned to every person who resides in that community during that time. This exposure classification scheme has been adopted mainly due to a lack of data on historical nitrate levels at the tap. Some studies have interpolated annual values for community water supply nitrate levels while others have used grosser averages, both methods using whatever data points are available. Regardless of the method employed, nitrate exposure assessment could stand improvement.

To that end, research is underway to evaluate seasonal nitrate variations in finished water for community drinking water supplies, as it appears from the historical data that finished drinking water nitrate levels can fluctuate depending on the time of year, in a similar fashion as raw water nitrate levels. In conjunction with that effort, comparison of raw water and finished water nitrate levels for municipal water supplies is in progress. Additionally, nitrate variation in distribution systems should be studied. Preliminary research has shown that ammonia levels may be elevated in distribution systems that use chloramines for disinfection. [56] Nitrate levels in such systems could fluctuate, as ammonia can be oxidized to nitrite and nitrate.

Data are generally scarce on private well water nitrate concentrations, so cancer studies are difficult in this population. In theory, this is a population could be at the greatest risk of exposure to high nitrate levels in water, as many private rural wells are shallow and thus more susceptible to contamination. Ongoing surveillance of water quality in private wells would be a good investment of resources in order to develop a baseline to track progress or decline in private rural drinking water quality.

With respect to education needs, both rural and urban residents must do a better job of protecting source water supplies, both at the local and watershed levels. Many state Extension programs are doing an excellent job of developing and disseminating information to the agricultural community on water quality issues. A similar effort may prove to be beneficial in urban areas.

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