UREA IN THE TRIBUTARIES OF THE CHESAPEAKE AND COASTAL BAYS OF MARYLAND

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Abstract. Concentrations of dissolved urea were monitored in several Chesapeake Bay tributaries from 1998 to 2002. Urea is a commonly used agricultural fertilizer and is also a breakdown product of poultry manure, which is used as an additional source of fertilizer throughout the watershed. Two trends were apparent. First, in several of the tributaries, seasonal peaks in ambient urea concentration coincided with the periods of the year (early spring and mid summer) when agricultural applications are most common. Second, highest annual mean concentrations (up to 2.6 μg atom N L^{-1}), as well as highest individual measurements (up to 24 μg atom N L^{-1}), were found for those tributaries with the most intensive agricultural and poultry operations. Peak urea concentrations were significantly higher than those which could be attained from *in situ* sources and regeneration. These elevated concentrations are of concern because this form of nitrogen has been shown to be a preferred form of nitrogenous nutrient for many phytoplankton, including some dinoflagellates which form harmful algal blooms. These results demonstrate that urea from land based sources can contribute to anthropogenic eutrophication.

Keywords: Chesapeake Bay, eutrophication, manure, nitrogen fertilizer, urea

1. Introduction

The nitrogenous nutrient urea has long been recognized to contribute substantially to marine and estuarine phytoplankton nutrition (e.g. McCarthy and Kamykowski, 1972; Carpenter *et al.*, 1972; Webb and Haas, 1976; Kristiansen, 1983; Glibert *et al.*, 1991; Tamminen and Irmisch, 1996). The availability of this form of nutrient, in terms of both total concentration and relative contribution to total nitrogen availability, is important for the understanding of the dynamics of phytoplankton. Although nitrate (NO_3^-) and ammonium (NH_4^+) are the most recognized forms of nitrogen suitable for phytoplankton growth, the ability of phytoplankton to utilize and to grow on specific organic compounds is well known (Antia *et al.*, 1991; Berman and Bronk, 2003). Of particular concern for enriched coastal areas is the recent observation that for some systems the organic fraction of nitrogen, including urea, may contribute to the triggering of harmful algal blooms (Paerl, 1988; Berg *et al.*, 1997; Glibert *et al.*, 2001; Anderson *et al.*, 2002).

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Urea is known to be produced *in situ* from zooplankton excretion (Corner and Newell, 1967; Mayzaud, 1973; Bidigare, 1983; Miller and Glibert, 1998), fish excretion (Wright *et al*., 1995; Chadwick and Wright, 1999; Walsh *et al*., 2000), bacterial regeneration (Mitamura and Saijo, 1980; Cho and Azam, 1995; Cho *et al*., 1996), and from release from sediments (Lomstein *et al*., 1989; Lund and Blackburn, 1989; Therkildsen and Lomstein, 1994). Urea is deposited from rain and atmospheric aerosols (Timperley *et al*., 1985; Cornell *et al*., 1998). Urea is also receiving increasing attention as an allochthonous form of nutrient. It is now the most commonly applied nitrogen fertilizer in many parts of the world (Smil, 2001; Glibert *et al*., unpublished). Urea represented more than 40% of global fertilizer sales a decade ago (Constant and Sheldrick, 1992), and its use has been increasing since. It is the preferred fertilizer for corn and soybean both in the Midwest and on the eastern seaboard of the U.S. (e.g. Overdahl *et al*., 1991). However, little is known about its transport and fate in the watershed.

The Chesapeake Bay is the largest estuary in the U.S. and has been undergoing significant eutrophication over the past several decades (e.g. Boynton *et al*., 1982, 1995; Fisher *et al*., 1988, 1992; Glibert *et al*., 1995). The Chesapeake Bay drains a large watershed with significant area of agricultural lands. The major agricultural source of nutrients to the mainstem of the Bay is the Susquehanna River at the head of the Bay. Agriculture is also intensive on the Delmarva Peninsula, the tri-state land area east of the Chesapeake Bay, where corn and soybeans are the primary crops. The Delmarva Peninsula also has some of the most intensive poultry production in the U.S., and the manure waste is used as fertilizer. As uric acid is the nitrogenous excretory product of poultry and its breakdown product is urea, manure is thus another important source of urea to both agricultural fields and runoff. The Delmarva Peninsula, in fact, has been identified by the U.S. Department of Agriculture as one of the top sub-watersheds in terms of vulnerability to manure leaching (USDA, 1997).

There have been intensive efforts to quantify the nutrient loading to the Chesapeake Bay (e.g. Boynton *et al*., 1995; Boesch *et al*., 2000; Boynton and Kemp, 2000), and significant efforts to reduce the nutrient loading to the Bay are underway (Boesch, 2002). Such efforts include the signing of multi-state agreements, such as the Chesapeake Bay Agreement, to reduce loads and monitor water quality (http://www.chesapeakebay.net/agreement.htm). To date, however, most of the efforts to quantify both the rates of nutrient loading and the ambient concentrations in the Bay are based on inorganic nutrients.

Here we present a five-year study of urea availability in several tributaries of Chesapeake Bay. This study builds on a previous study which focused on the long-term trends in concentration and bioavailability of urea along the longitudinal axis of Chesapeake Bay (Lomas *et al*., 2002). To our knowledge, this is the only watershed for which such a time series of urea concentration is available.

2. Methods

Samples were collected over a five-year period, from 1998 to 2002, at up to 27 stations per year (Table I; Figure 1). Samples were collected by the Maryland Department of Natural Resources as part of their intensive monitoring for algal blooms and associated environmental parameters. Due to reductions in funding, the numbers of stations monitored decreased during the last two years of this study. Sample locations were concentrated along the eastern shore of the Chesapeake Bay,

TABLE I

Summary of numbers of stations sampled and frequency

Year	Tributary systems	Number of stations sampled	Number of samples collected
1998	Transquaking and Chicamacomico Rivers	5	18
	Manokin and Kings Creek	6	23
	Pocomoke River	5	18
	Coastal Bays	7	37
	Tangier and Pocomoke Sounds	2	5
1999	Transquaking and Chicamacomico Rivers	5	65
	Manokin and Kings Creek	6	42
	Pocomoke River	5	69
	Coastal Bays	6	42
	Tangier and Pocomoke Sounds	2	26
2000	Transquaking and Chicamacomico Rivers	6	63
	Manokin and Kings Creek	6	41
	Pocomoke River	5	63
	Coastal Bays	6	35
	Tangier and Pocomoke Sounds	2	19
	Middle River	3	36
2001	Transquaking and Chicamacomico Rivers	5	68
	Manokin and Kings Creek	6	41
	Pocomoke River	5	70
	Coastal Bays	6	51
	Tangier and Pocomoke Sounds	2	22
	Middle River	3	21
2002	Transquaking and Chicamacomico Rivers	4	29
	Manokin and Kings Creek	5	28
	Pocomoke River	2	16
	Coastal Bays	4	23
	Tangier and Pocomoke Sounds	2	5
	Middle River	3	21
Total			997

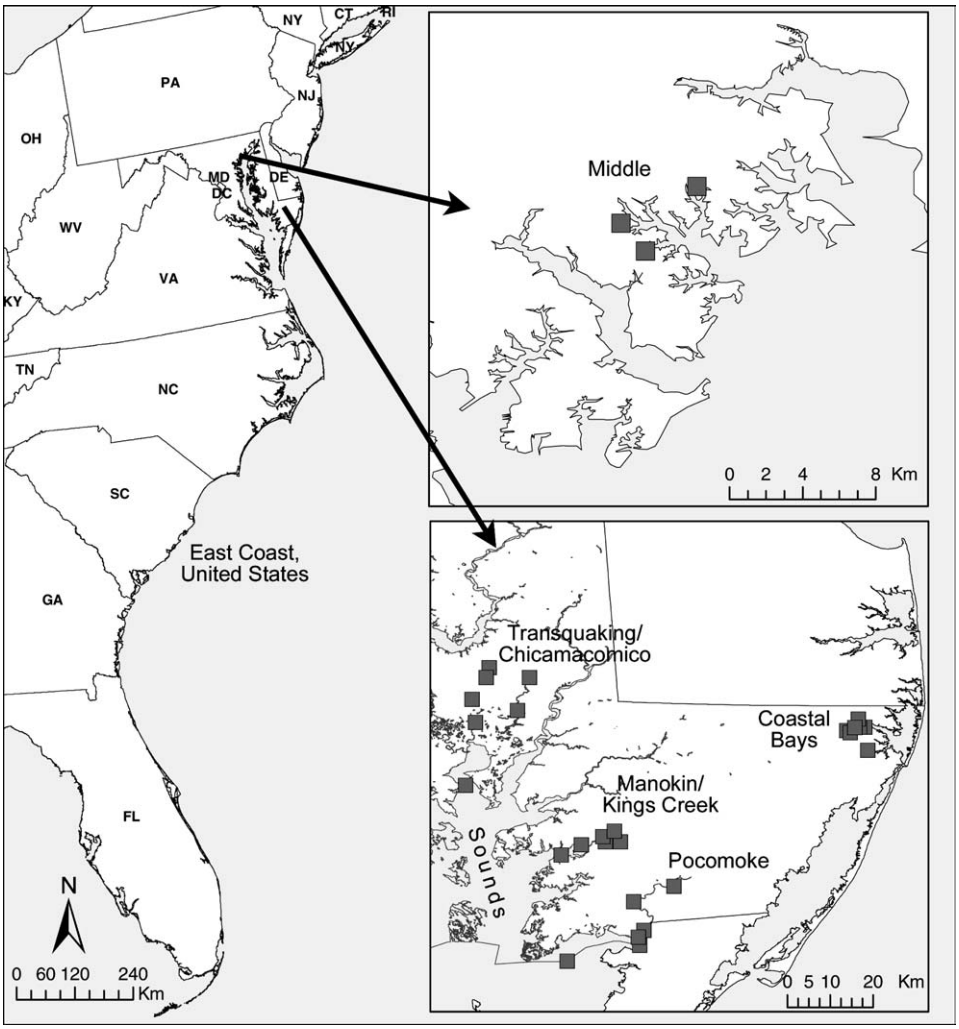

Figure 1. Station map showing the location of Chesapeake Bay, the Coastal Bays and the individual tributary systems that were sampled.

but some samples were also collected from the northwestern shore (Figure 1). In addition, some samples were collected from the Coastal Bays of Maryland. Sample collection each year was initiated in April and terminated in October; thus, no winter values are available. Samples were collected at a minimum on a monthly basis, and for some stations and years, on a weekly basis.

All sampling was from surface or just below surface. Samples were collected in acid-cleaned polyethylene or polycarbonate bottles, kept on ice until return to the laboratory, where they were immediately filtered through precombusted GF/F filters and frozen. All samples were processed at the Horn Point Analytical Services

Laboratory using the urease method of McCarthy (1970) for consistency with the Lomas *et al*. (2002) study. All samples were analyzed in triplicate. Variability in triplicate analyses was 7.8%. Samples were also analyzed for other nutrient and biological parameters; here, the NH_4^+ concentrations only are shown for comparison. Concentrations of NH_4^+ were analyzed according to Parsons *et al*. (1984).

Data on riverine flow were obtained from the U.S. Geological Survey monitoring gauges (http://md.usgs.gov/monthly/bay1.html). Although it is recognized that rainfall, and consequently flow, can be locally variable, only data from the gauging station on the Pocomoke River (Figure 1) were used in this analysis.

3. Results

In order to present spatial and temporal trends in urea distributions, data were grouped by tributary (except where noted), by month, and by year.

Variability within each station of each tributary system was large. This was exemplified through an examination of a single station from each of two years, from each of the tributary systems examined (one year only for Middle River). The within-stations trends for 1999 were quite different from those of 2000, but both years showed high variability. For Middle River, Kings Creek, the Pocomoke River, and the Pocomoke Sound, concentrations fluctuated from near 0 to $\sim 2.5 \mu$g at N L^{-1} from sampling point to sampling point (Figure 2). Larger fluctuations were observed for the Chicamacomico and Coastal Bays (Figure 2); at these stations, concentrations exceeding 5 μg at N L^{-1} were noted.

Mean seasonal variation across all years was examined by month. All samples from a given month within each tributary system from all years were combined into the monthly average. Although within a given year there may be some bias in that not all tributaries were sampled on the same day, when combined with other years, this source of variability was minimal. In several of the tributary systems, distinct and statistically significant peaks can be discerned (Figure 3). In the Manokin/Kings Creek systems, such peaks were found to occur in May and July, whereas these peaks were offset by one month in the Transquaking/Chicamacomico systems. In Middle River, mean concentrations revealed a significant pulse of urea in April, while in the Coastal Bays, a large pulse was noted in the mean data for June. Similarly, in the Pocomoke River, a substantially deeper river than the Manokin/Kings Creek or Transquaking/Chicamacomico systems, the mean concentration was highest in June (Figure 3).

For all years examined, the highest individual values measured ranged from 2.5 μg at N L^{-1} in 2000 to >24 μg at N L^{-1} in 2002 (Figure 4). The extreme high values represented pulsed additions to the watershed, as they were neither sustained in time (from sampling interval to sampling interval) or in space. The stations with the highest measured concentrations were consistently located in the Transquaking/Chicamacomico systems, and the Coastal Bays. In fact, stations

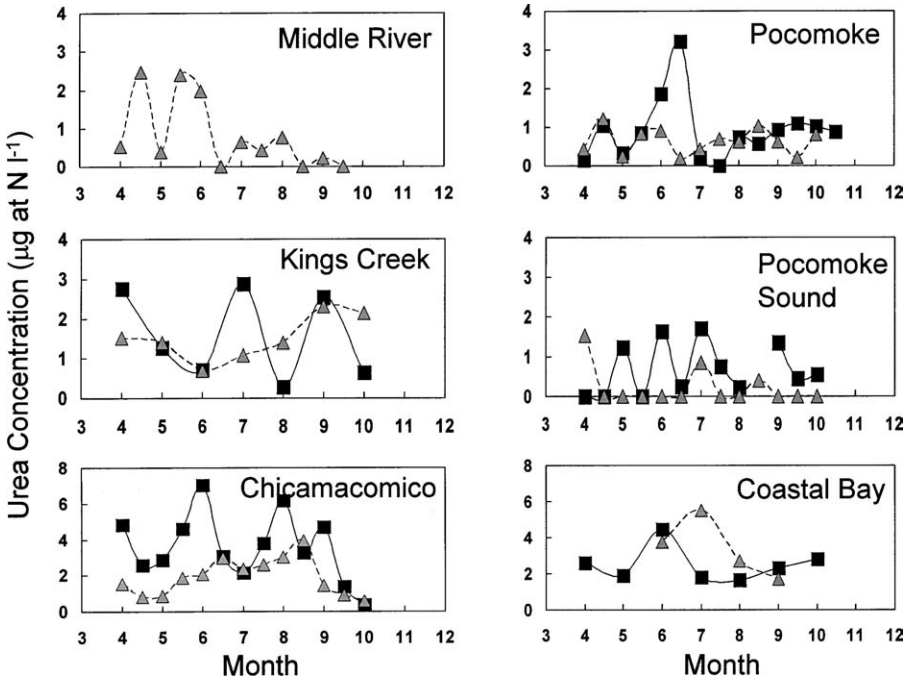

Figure 2. Concentrations of urea for one representative station for each of the tributary systems monitored. Data from two years of sampling are shown: 1999 (squares) and 2000 (triangles). The tributary and individual stations are indicated in the respective panels. Note that data from 1999 for Middle river are not available.

from the Transquaking/Chicamacomico systems had 56% of the five highest recorded values for each year, while those from the Coastal Bays had 20% of the highest recorded values.

The distribution downstream in urea concentrations was also highly variable, as exemplified by the change in concentration of the three stations in the Transquaking River during 1999 and 2000 (Figure 5). Whereas in 1999, urea concentrations were found to decrease by as much as an order of magnitude from station to station, in 2000 concentrations did not vary to nearly the same extent in space.

Mean concentrations of NH_4^+ generally exceeded those of urea, but for several tributaries through much of the summer months concentrations of urea and NH_4^+ were similar, as shown for 1999 (Figure 6). The timing of the maximum concentrations of NH_4^+ differed from those of urea, with a significant pulse of NH_4^+ noted in May in the Manokin/Kings Creek systems, and well into the autumn for the Transquaking/Chicamacomico systems.

Intuitively, flow regime should be related to ambient concentration, in that it is rainfall that determines flow and runoff. However, other factors, such as timing of fertilizer applications, soil type, and moisture content complicate this relationship. Thus, when urea concentrations for 1999 were examined as a function of flow, there

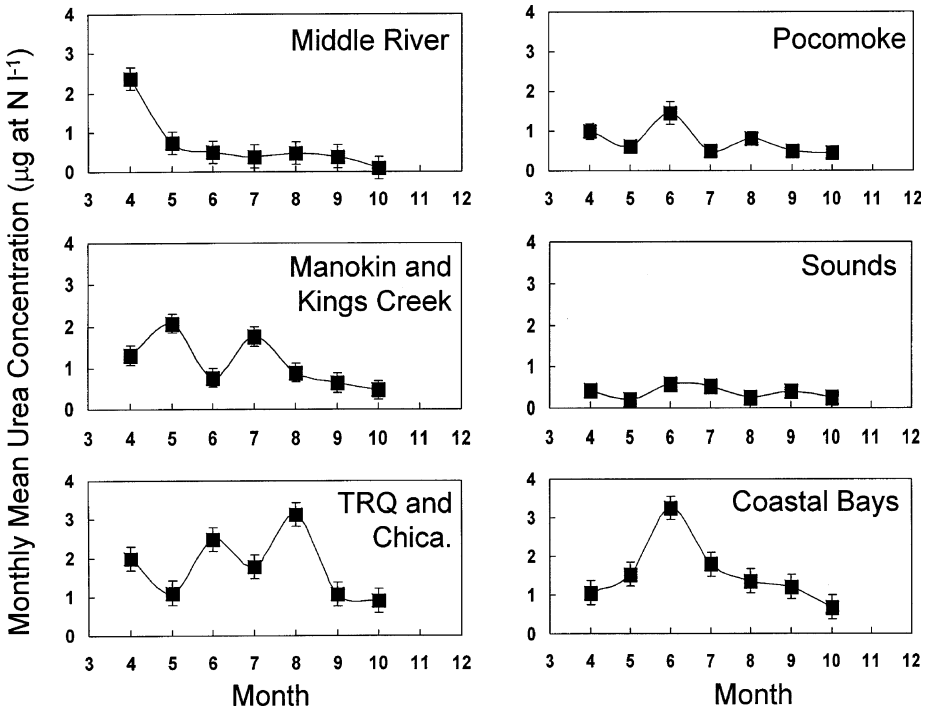

Figure 3. Monthly mean concentrations of urea for all stations monitored for the tributary system indicated on the respective panels. Data shown are the average of all years of sampling. Error bars are ±1 S.E. "TRQ" and "Chica." refers to the Transquaking and Chicamicomico Rivers.

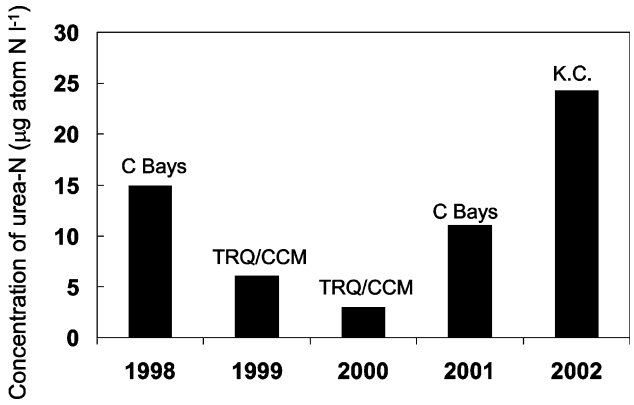

Figure 4. Maximum recorded concentrations of urea for each of the years indicated. The tributary system for which the maximum was recorded is indicated above the bar: "C Bays" refers to Coastal Bays, "TRQ/CCM" to the Transquaking and Chicamacomico Rivers, and "K.C." refers to Kings Creek.

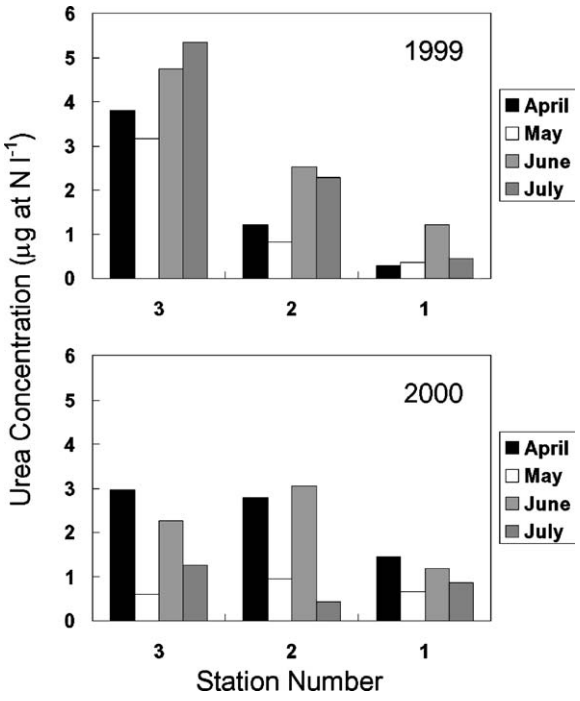

Figure 5. Concentration of urea in three stations of the Transquaking river for each of the months indicated for 1999 (upper panel) and 2000 (lower panel).

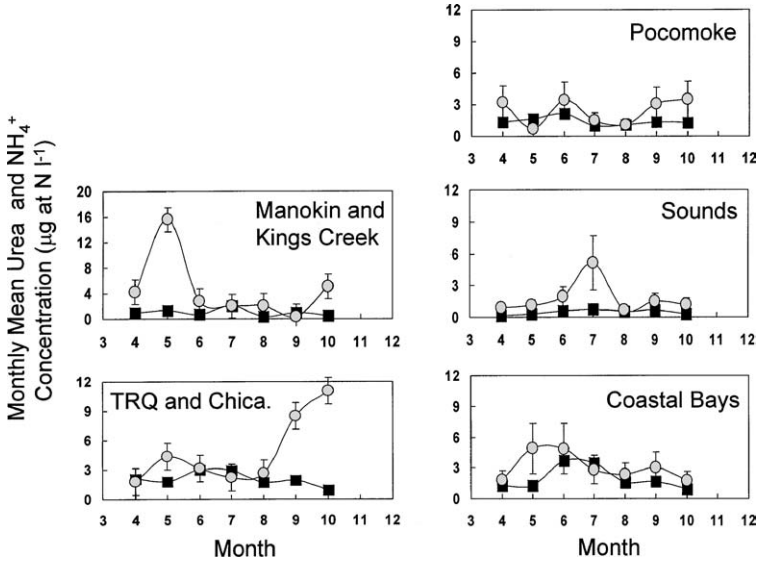

Figure 6. Monthly mean concentrations of urea (squares) and NH_4^+ (circles) for all stations monitored with the tributary system indicated. Data shown are for 1999 only. No data from that year are available for Middle river. Note the change of scale for the Manokin and Kings Creek data. Error bars are ±1 S.E.

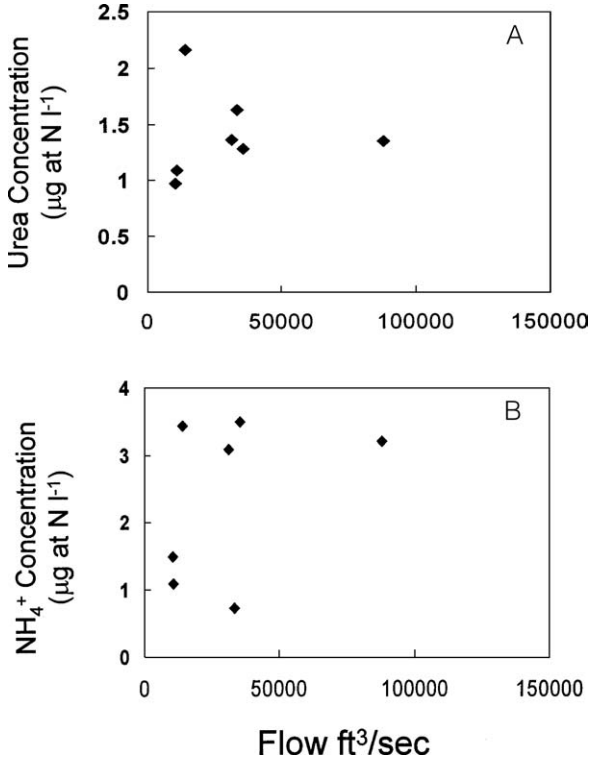

Figure 7. Concentrations of urea (panel A) and NH_4^+ (panel B) as a function of riverine flow measured at a gauging site on the Pocomoke river for 1999. There was no significant relationship for either nitrogen nutrient.

was no consistent relationship (Figure 7). Similarly, using 1999 as an example, there was also no relationship with NH_4^+ concentrations (Figure 7). While NH_4^+ is a breakdown product of urea, it is also a mineral fertilizer and thus subject to runoff as well.

When examined by year, there was substantial variability within each tributary system (Figure 8), although differences were not significant. The time scale of this analysis, five years, is too short to discern any long-term trend, as this period of study encompassed years that were both above and below average in terms of rainfall and runoff. Although a declining trend seems to be the case for the Coastal Bays from 1998 to 2002, this trend may have been influenced by the smaller number of samples collected in 2002; thus, the pulses of high urea concentration observed in previous years may have been missed.

Mean concentrations for all stations and years combined were above 0.5 μg at N L^{-1}, and for the Transquaking/Chicamacomico systems and the Coastal Bays were above 1.5 μg at N L^{-1} (Figure 9). Although the variability around each of these means was large, the mean concentrations for the Transquaking/Chicamacomico and the Coastal Bays were significantly different from those of Middle River,

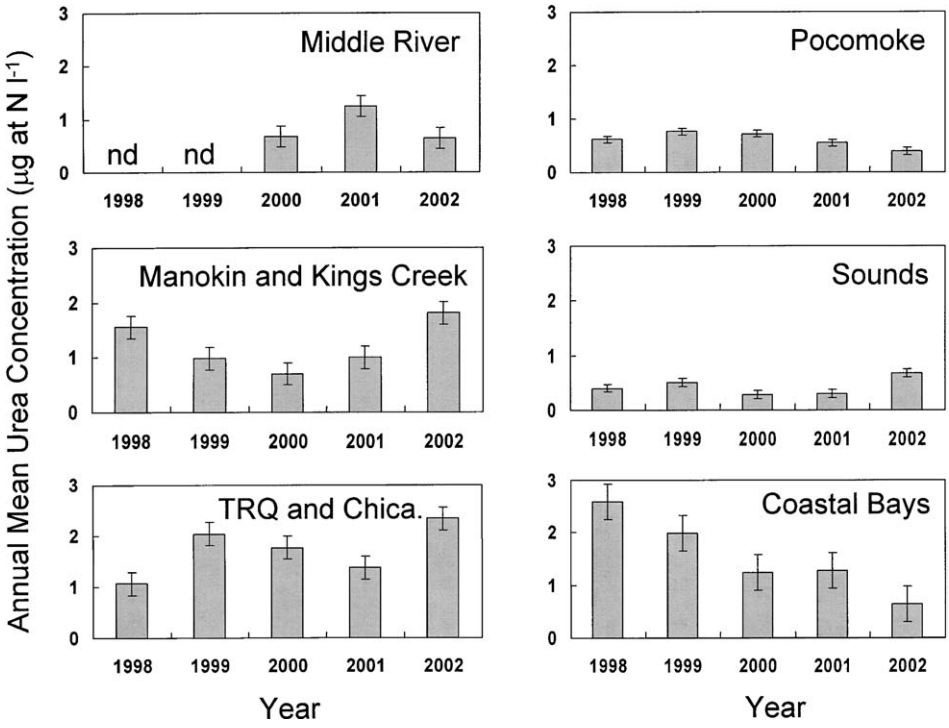

Figure 8. Annual mean concentrations of urea for all stations for each of the tributary systems indicated by year of sampling. Error bars are ±1 S.E.

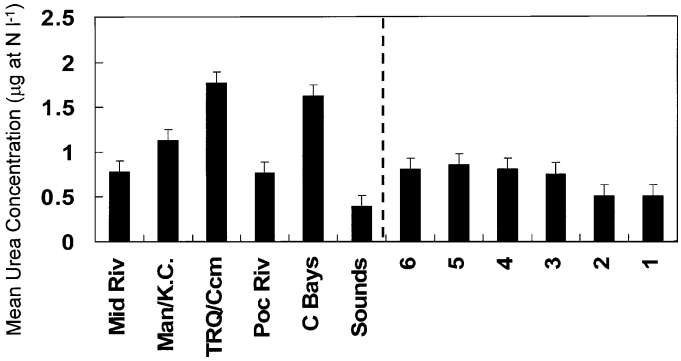

Figure 9. Mean concentrations of all values from all years for the tributary systems monitored in comparison to values previously published by Lomas *et al*. (2002) for six sections along the longitudinal axis of the Chesapeake Bay. The six segments are identified in Figure 1. Error bars are ±1 S.E. "Mid Riv" refers to Middle River, "Man/K.C." refers to Manokin and Kings Creek, "TRQ/CCM" refers to Transquaking and Chicamacomico Rivers, "Poc Riv" refers to Pocomoke River, and "C Bays" refers to Coastal bays.

Pocomoke River, and the combined values of Pocomoke Sound and Tangier Sound, and from the mainstem of Chesapeake Bay, based on data previously presented for six sections of the Bay (Lomas *et al*., 2002).

4. Discussion

Organic nitrogen has been receiving increasing attention in recent years, as it is now well recognized to have multiple sources, and to be an important contributor to nitrogen cycling and to the nitrogen nutrition of both bacteria and phytoplankton (reviewed by Antia *et al*., 1991; Berman and Bronk, 2003). Little appreciation has been given to the increasing anthropogenic nature of organic compounds, including urea. This study demonstrated that not only is urea a significant form of nitrogen entering the tributaries of Chesapeake Bay, but it is also highly variable.

The use of urea as an agricultural nutrient has been increasing, both in Maryland's tributaries and globally (e.g. Constant and Sheldrick, 1992; Smil, 2001). Worldwide use of urea as a nitrogen fertilizer and feed additive has increased more than 100-fold in the past four decades, with a doubling in just the past decade alone (Glibert *et al*., unpublished). As an agricultural fertilizer, it is often preferred over inorganic nitrogenous nutrients because of its stability in storage, its ease of application, and its retention within soils (Stehouwer and Johnson, 1990; Shoji *et al*., 2001). In coated form, urea becomes a slow-release fertilizer and this is one of the most popular forms for applications to lawns, golf courses, and parks, as well as many crops (Overdahl *et al*., 1991).

The use of poultry manure as an agricultural fertilizer is another source of urea to many areas, particularly mid-Atlantic farms. On the Delmarva Peninsula agricultural land represents about 35% of total land use. Intensive poultry operations in the region produce over 500,000,000 chickens and game hens annually from roughly 5500 farms (http://www.dpichicken.com). While acreage for corn production has decreased in the past decade, acreage for soybean, wheat, greenhouse and nursery production has increased. Our sampling regime was not designed to pinpoint specific sources of runoff, and indeed, this would be very difficult, as fertilizer and manure use varies from farm to farm, crop to crop, and year to year. Fertilizer usage per farm is also propriety information; nevertheless, of the systems monitored, concentrations were found to be highest in the Transquaking and Chicamacomico Rivers and the upper Coastal Bays. These sites were located in two of the top twenty counties in the U.S. for poultry production. These systems not only had maximum concentrations up to an order of magnitude higher than average concentrations, but the average concentrations for all stations and years combined were two-fold higher than most of the other tributaries. Furthermore, the results for these tributaries indicate peaks in spring and mid-summer, times that coincide with the annual application of urea or manure to crop lands. As these data have shown, urea is present in waters receiving runoff,

and the highest concentrations are found in those tributaries for which agriculture is intensive.

A trend of declining urea concentrations in waters of the mainstem of Chesapeake Bay was found during the decade from 1988 to 1998 in a long-term analysis of trends in urea availability covering the period from 1972 to 1998 (Lomas *et al*., 2002). This finding was particularly surprising given the change in fertilizer quality toward manures and liquid urea over this same period. One likely explanation is the very tight coupling between urea uptake and urea concentration that was found (Lomas *et al*., 2002). Furthermore, variations in rainfall on an annual basis may also be significant in this regard: from 1990 to 1994, Susquehanna River flow was lower than average, leading to a negative relationship between such flow and urea concentration, while 1996–1998 were wetter than average, leading to a positive relationship between flow and concentration (Lomas *et al*., 2002).

Other seasonal analyses have reported varying temporal patterns in urea. Several estuarine and coastal systems have been documented to have winter/spring peaks (Savidge and Huntley, 1977; Satoh *et al*., 1980; Kristiansen, 1983), while others have summer peaks (Berman, 1974; Webb and Haas, 1976; Turley, 1986). Such variability is not surprising when external sources, in additional to internal cycling processes, are considered.

The importance of these results is two-fold. First, this is the first long-term data set for this nitrogenous nutrient for these tributaries, and it demonstrates the potential contribution of this nutrient to eutrophication. Second, urea has been shown to be a preferred form of nitrogenous nutrient for numerous micro-flagellates, including dinoflagellates in estuarine systems (Glibert *et al*., 2001). In fact, urea has previously been shown to be correlated with the outbreaks of several harmful algae blooms within the Bay system. For example, Glibert and Terlizzi (1999) showed that for fish aquaculture ponds located near the Manokin River, concentrations of urea >1.5 μg at N L^{-1} were associated with harmful dinoflagellate blooms 75% of the time, whereas few such blooms were found to be associated with urea concentrations <1.5 μg at N L^{-1}. Furthermore, Glibert *et al*. (2001) and Lomas *et al*. (2001) found that elevated concentrations of urea preceded blooms of *Prorocentrum minimum* and *Aureococccus anophagefferens* in the Choptank River and Coastal Bays, respectively. Finally, the systems documented herein as having the highest maximum concentrations of urea on an annual basis, the Transquaking/Chicamacomico and Coastal Bays, have also been shown to be among those stations at which the harmful dinoflagellate *Pfiesteria piscicida* is most often found in either the sediment or water column (Glibert *et al*., in press). While *P. piscicida* is a heterotrophic dinoflagellate that primarily relies on grazing for its nutrition, it does have the capability for direct uptake of urea (Lewitus *et al*., 1999), and may also be stimulated indirectly by the stimulation of growth of other microorganisms (e.g. bacteria and cryptophytes) on which it feeds.

In summary, these data have shown that urea concentrations in the tributaries of Maryland's Chesapeake and Coastal Bays can be significant. While highly variable in space and time, the potential contribution of this nitrogenous nutrient to eutrophication cannot be ignored.

Acknowledgements

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