<u>Supporting Information</u>

Kaushal, S.S., P.M. Groffman, L.E. Band, E. Elliott, C.A. Shields, and C. Kendall. Tracking nonpoint nitrogen pollution in human-impacted watersheds.

Site Description

The Gwynns Falls is a 17,150 ha watershed that traverses a land use gradient from the urban core of Baltimore City, through older urban residential (1900-1950) areas, older suburban (1950-1980) zones in the middle reaches, and finally through suburbanizing areas and a rural/suburban fringe in the headwaters in Baltimore County (Doheny et al. 1997, Groffman et al. 2004, Shields et al. 2008, Kaushal et al. 2008).

Site	Land Use	Total Drainage Area (ha)	Reach Drainage Area (ha)	Land Use (Forest/Resident/Ag)			% Lawn	% ISC*
Longitudinal Gwynns Falls Reaches								
GFGL	Suburban	81	81	10	37	0	25.4	27.5
GFGB	Suburban	1,066	985	13	65	9	30.1	28.0
GFVN	Suburb/Urban	8,348	7,282	25	47	9	27.6	28.6
GFCP	Urban	16,278	7,930	19	50	5	24.8	34.9
Small Watersheds								
POBR	Forest	NA	32.3	100	0	0	0	0
BARN	Forest/Suburb	NA	381	63	34	0	16.0	3.7
DRKR	Suburb/Urban	NA	1,414	9	42	3	19.7	52.4
MCDN	Agricultural	NA	7.8	0	0	100	0	0

Table S1. Characteristics of Baltimore LTER site watersheds including longitudinal stations along the main channel of the Gwynns Falls and forested reference, agricultural, suburban and urban small watersheds (Groffman et al. 2004, Kaushal et al. 2008). *% ISC denotes watershed impervious surface cover and values are updated compared to previous older values presented by Groffman et al. 2004 and Kaushal et al. 2008.

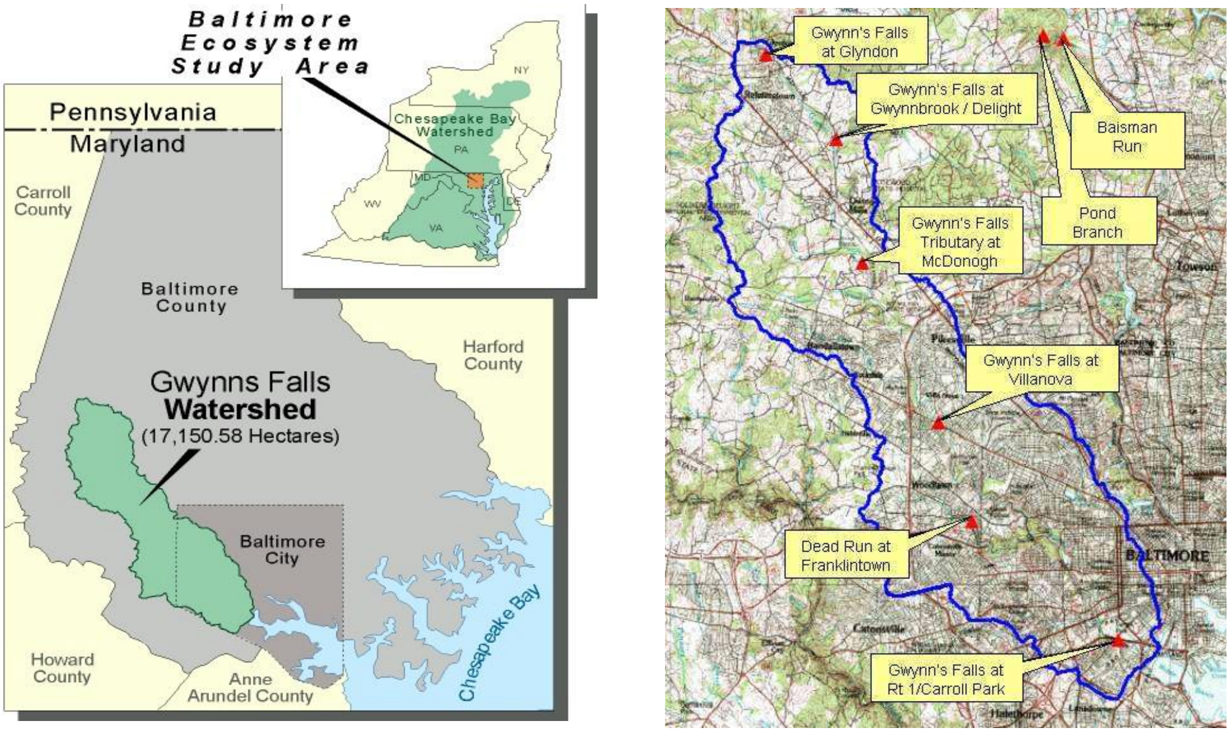
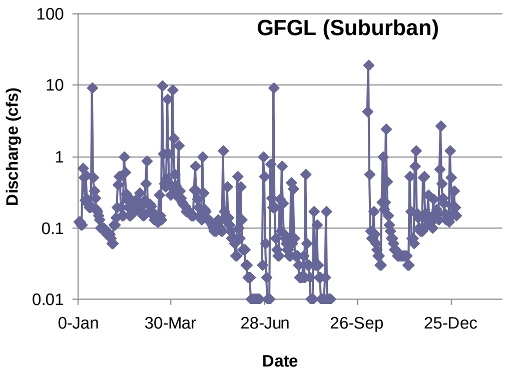


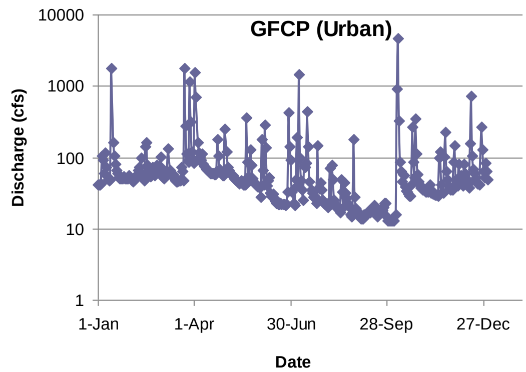
Figure S1. Baltimore LTER study sites, including small watersheds (Glyndon (suburban), McDonogh (agricultural), Pond Branch (forested), Baisman Run (suburban), and Dead Run (urban), and the longitudinal sites along the Gwynns Falls (Glyndon, Delight/Gwynnbrook, Villa Nova, and Carroll Park) (Kaushal et al. 2008).

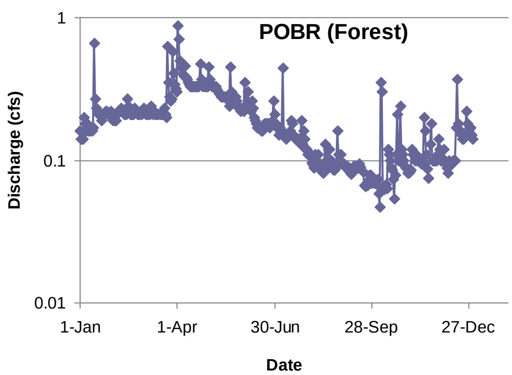
Methods

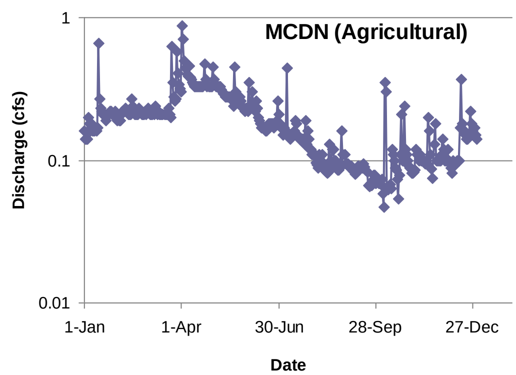
Discharge and Stream Chemistry

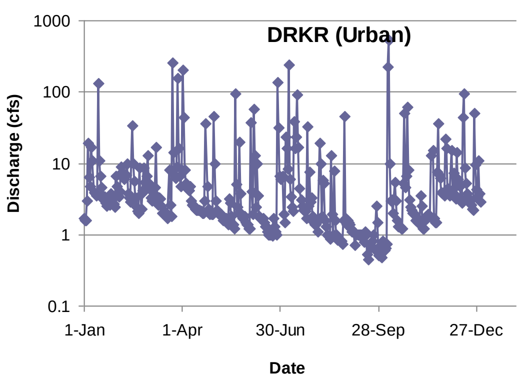
Stream discharge was continuously monitored at these sites by the U.S. Geological Survey (USGS). Stream samples were collected weekly regardless of flow conditions (no bias towards storm versus baseflow), filtered in the field, and returned to the laboratory for analysis of NO_3^- concentrations by ion chromatography or total N (dissolved and particulate) following persulfate digestion as described previously (Groffman et al. 2004, Kaushal et al. 2008a, Shields et al. 2008). Water samples are analyzed for dissolved organic carbon (DOC) using a Shimadzu TOC/TDN analyzer. Samples were collected from the middle of the stream in 125mL bottles and stored in a cooler during transport between sample sites.

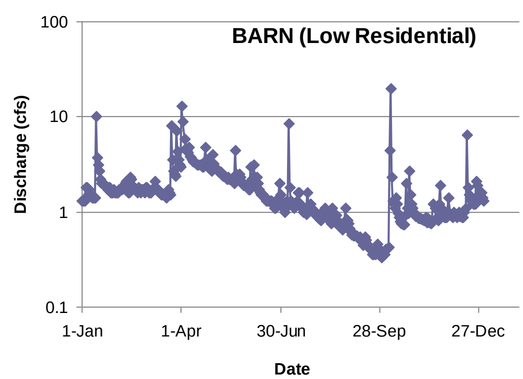


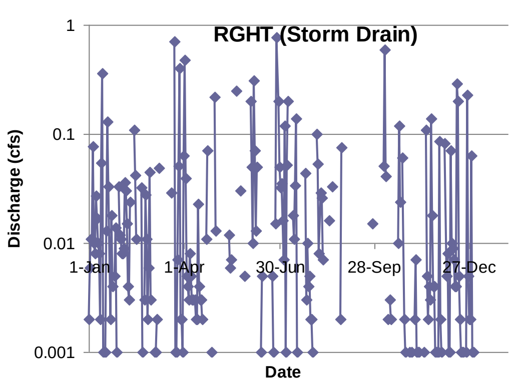












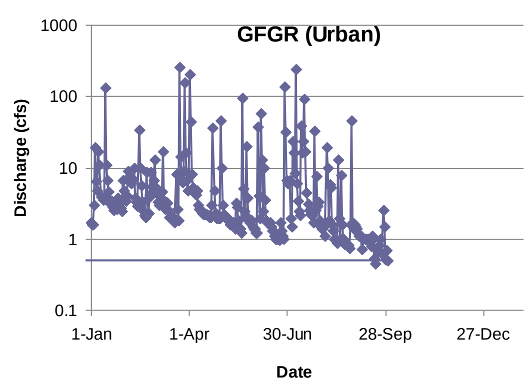


Figure S2. Hydrographs of streams showing available mean daily streamflow data during 2005.

Watershed N Exports and Mass Balance Calculations

Methods for estimating watershed N exports and mass balance calculations can be found in previous work (Groffman et al. 2004, Kaushal et al. 2008a, Shields et al. 2008). Nitrate-N and total N loads for the study watersheds were estimated using the Fluxmaster program developed by the USGS (Schwarz et al. 2006). The routine weekly sampling at these sites could have missed storm events, and we acknowledge that some bias may result if the discharge-concentration relations show a discontinuity above certain flow thresholds (Kaushal et al. 2008a, Shields et al. 2008). Error bars in Figure 1 indicate standard errors and uncertainty in exports estimated from the USGS Fluxmaster program (Schwarz et al. 2006).

Inputs of N from atmospheric deposition were taken from the U.S. Environmental Protection Agency's Clean Air Status and Trends Network (CASTNET) site at Beltsville, MD, approximately 50 km south of the Gwynns Falls watershed. Both wet and dry N deposition are measured at this site, although there may be uncertainty for deposition for the Gwynns Falls due to distance and difference in local emissions. Fertilizer input to lawns in the Glyndon watershed (14.4 kg N/ha/y over the whole watershed area) was calculated from measurements of lawn area and a detailed survey of residential lawn-care practices in the Glyndon watershed conducted in 2002 (Law et al. 2004). Fertilizer inputs to the agricultural watershed were from Maryland Cooperative Extension Service recommended application rates for maize production (120 kg N/ha/y) and estimates of N fixation by soybeans (30 kg N ha/y) for a mean annual input of 60 kg N ha/y (Groffman et al. 2004, Kaushal et al. 2008a).

Leaking sewers were not included in the "input" term (denominator) in calculating watershed N retention. Baisman Run is the only watershed with septic systems and a watershed mass balance was not estimated in this watershed. It is difficult to quantify the impacts of leaks, but they may impact the retention calculation. This potential impact on watershed N budgets is discussed in detail in Groffman et al. (2004). Groffman et al. (2004) noted that "sewage leaks increase the apparent yield and decrease the apparent retention of N from the watershed. At the same time, infiltration of stream flow into sanitary sewers is a well-documented problem in urban areas and may remove a significant amount of both streamflow and N from our output computations." We also did not take into account N inputs from pet waste, which may occur in the suburban watershed. Although these areas of uncertainty exist, the major components of the mass balance input model are well-constrained at the Baltimore LTER site via data from fertilizer inputs from field surveys, nearby NADP data for atmospheric deposition, and routine monitoring of outputs across baseflow and storms (Groffman et al. 2004, Kaushal et al. 2008).

Isotopic Sampling

The isotopic composition of soil water underneath fertilized lawns and atmospheric deposition was measured by analyzing samples (n = 10) of soil solution collected by zero tension lysimeters installed at 50 cm depth in long-term lawn study plots on the campus of the University of Maryland Baltimore County (Groffman et al. 2009) and atmospheric deposition (n = 6) from the National Atmospheric Deposition Program (NADP/NTN) collection site in Carroll County Maryland (Elliott et al. 2007). Lysimeter samples were collected from 2 grass lawn plots with 2 lysimeters each from December 2003 to March 2004 and atmospheric deposition samples were pooled bi-monthly during 2000. The procedures for compositing and analyzing atmospheric samples can be found in (Elliott et al. 2007).

Stream samples analyzed for isotopes were distributed among the sites as follows: BARN (exurban) (n = 20), DRKR (urban) (n = 15), DRKR (urban storm samples) (n = 47), GFCP (urban and mixed land use) (n = 16), GFGL (suburban) (n = 15), MCDN (agriculture) (n = 16), POBR (forest) (n = 18), RGHT (storm drain) (n = 7), GFGR (urban tributary to Gwynns Falls, contaminated with sewage, then repaired) (n = 14).

Results

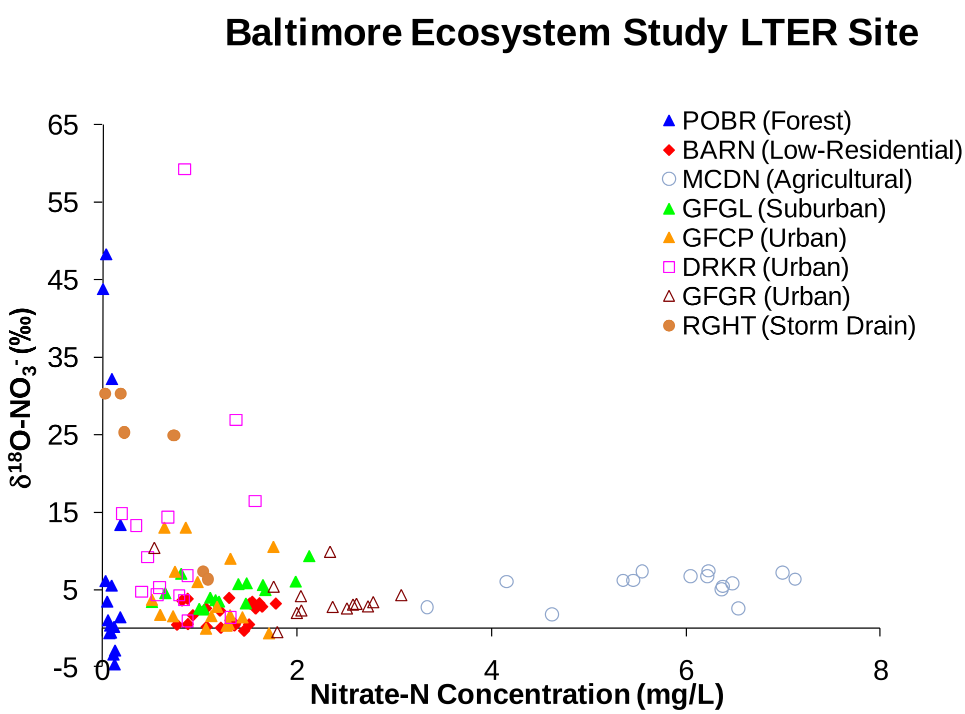


Figure S3. There were peaks in δ^{18}O values at low nitrate concentrations in the forest stream and in DRKR Dead Run (Urban). The forested watershed (POBR) also generally showed low $\delta^{15}N\text{-}NO_3^-$ in the range previously reported for soil N and fertilizer/rain, but high δ^{18}O values indicate significant atmospheric nitrate contributions to select samples.

POM Isotope Analysis

Organic C and N isotopes in streams varied across land use with percentage impervious surface coverage in the watershed (Figure S5). There were elevated levels of dissolved organic carbon (DOC) in the storm drain relative to other streams at the BES LTER site (Figure S4). In addition, the δ^{13}C-POM in the storm drain was elevated compared to other watersheds and δ^{13}C-POM increased with increasing percentage impervious surface coverage (Figure S5) and is in the range expected for terrestrial carbon sources and C4 plants. δ^{15}N-POM was most variable in watersheds with the lowest impervious surface cover; isotopic variability increased with increasing percentage impervious surface coverage (Figure S5).

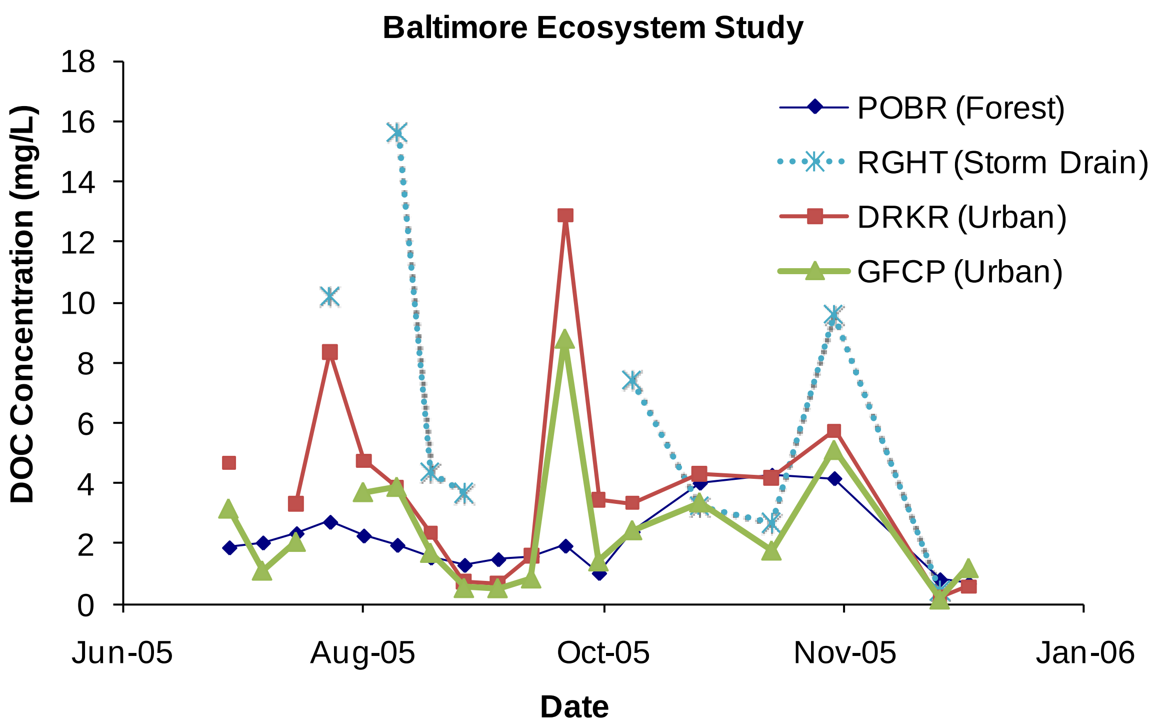


Figure S4. Examples of elevated concentrations of DOC in the storm drain relative to other streams.

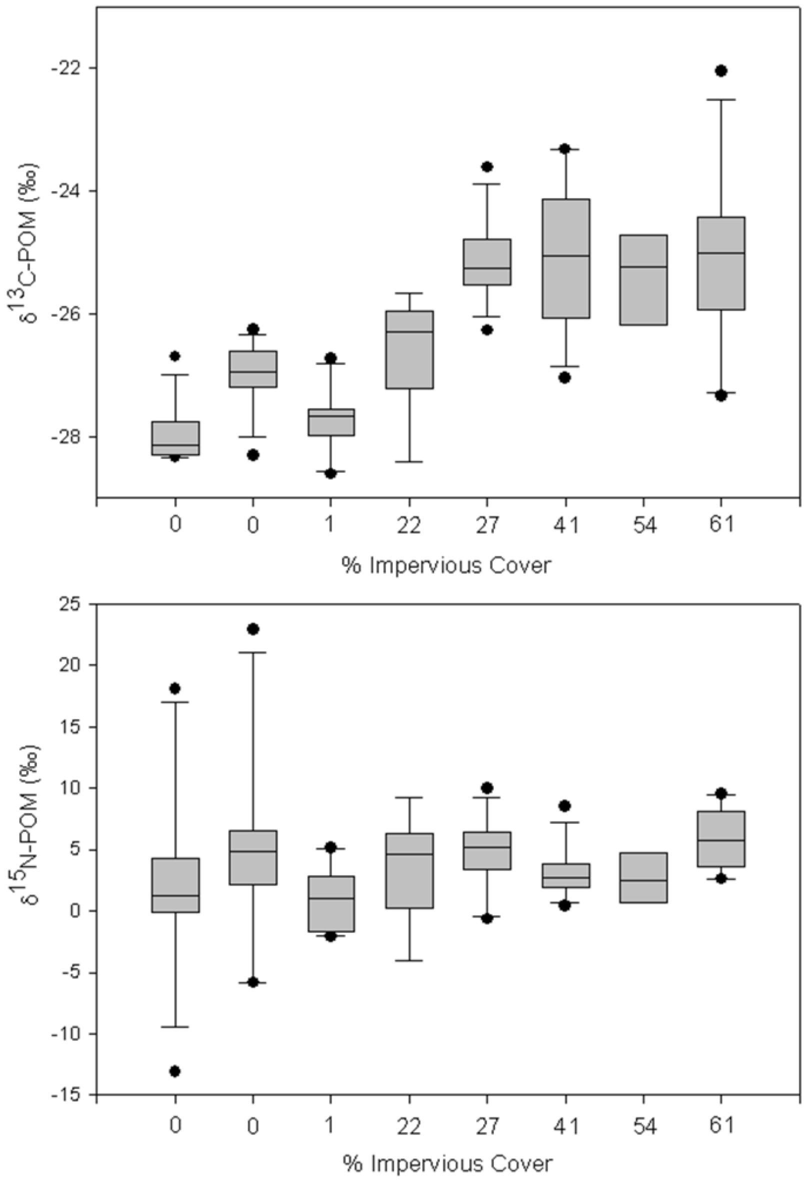


Figure S5. Patterns in δ^{13}C-POM and δ^{15}N-POM in streams across a gradient in watershed impervious surface coverage. Impervious surface cover based on previous estimates from Groffman et al. (2004) and Kaushal et al. (2008). Box-whisker plots describe the normal distribution of data (minimum value, lower quartile (Q1), median (Q2), upper quartile (Q3), and maximum value). *The storm drain (RGHT) drains an urban watershed with approximately 54% impervious cover.

Relationships between Isotopic Composition and Runoff

The isotopic composition of nitrate showed varying relationships with runoff across land use (Figure S6). In particular, $\delta^{15}N\text{-}NO_3^-$ and $\delta^{18}O\text{-}NO_3$ in the agricultural catchment (MCDN) showed significant decreasing linear relationships with increasing runoff ($p < 0.05$) (Figure S6). Nitrogen and oxygen isotope signatures showed significant contrasting patterns with runoff at the most downstream urban site in the Gwynns Falls (GFCP) ($p < 0.05$); $\delta^{15}N\text{-}NO_3^-$ showed a significant linear decrease with increasing runoff ($p < 0.05$) whereas $\delta^{18}O\text{-}NO_3$ showed a significant linear increase with increasing runoff ($p < 0.05$) (Figure S6).

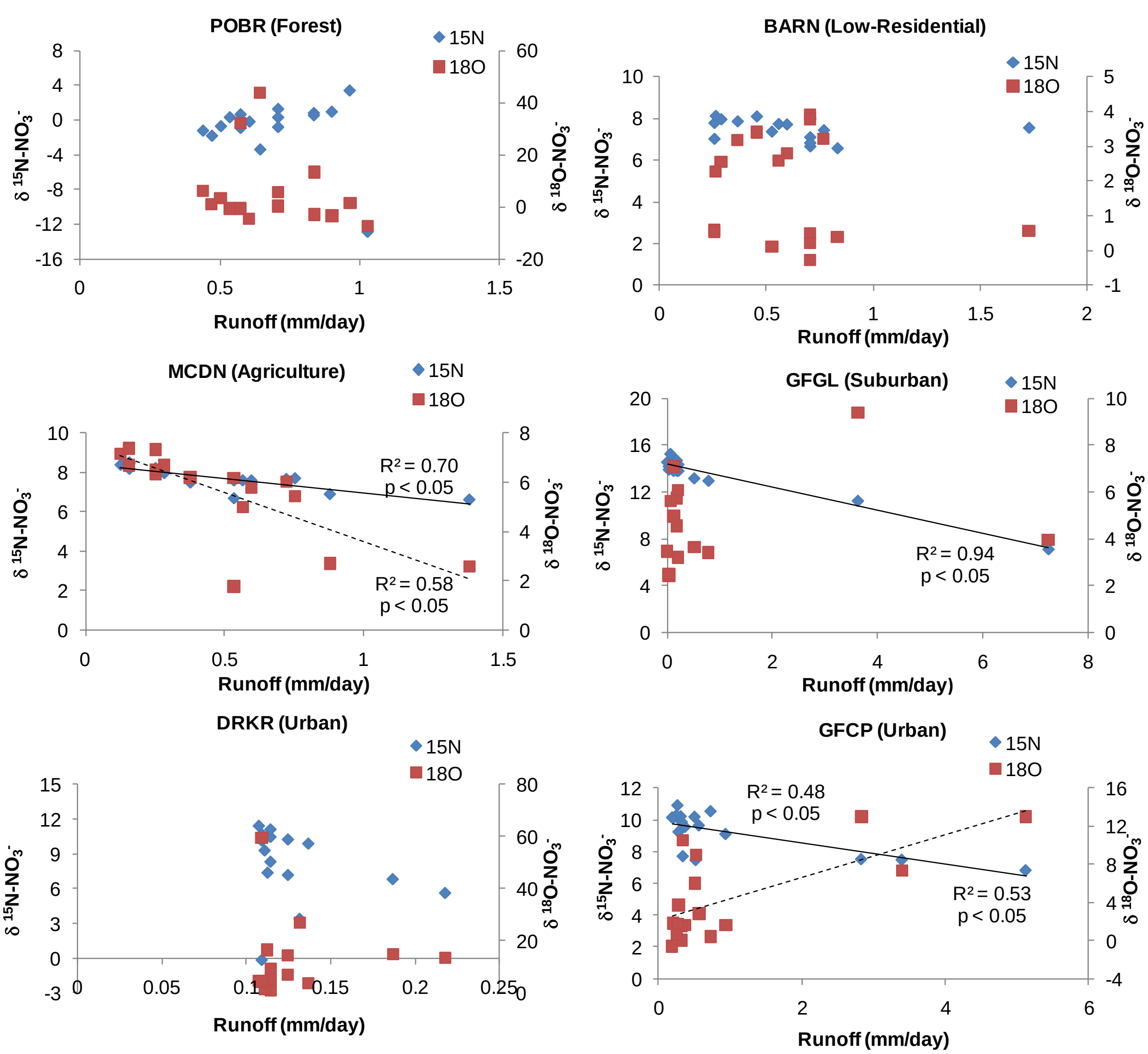


Figure S6. Relationships between $\delta^{15}N\text{-}NO_3^-$ and $\delta^{18}O\text{-}NO_3^-$ of nitrate in streams across varying runoff conditions.

Discussion

Denitrification in Agricultural and Low-Residential Catchments

As mentioned in the text, there was a marginally significant linear relationship between $\delta^{15}N\text{-}NO_3^-$ and nitrate concentrations ($p = 0.05$) suggesting that although denitrification was occurring, it may not havebeen sufficient to reduce a substantial proportion of the elevated nitrate concentrations in the stream (Figure S7).

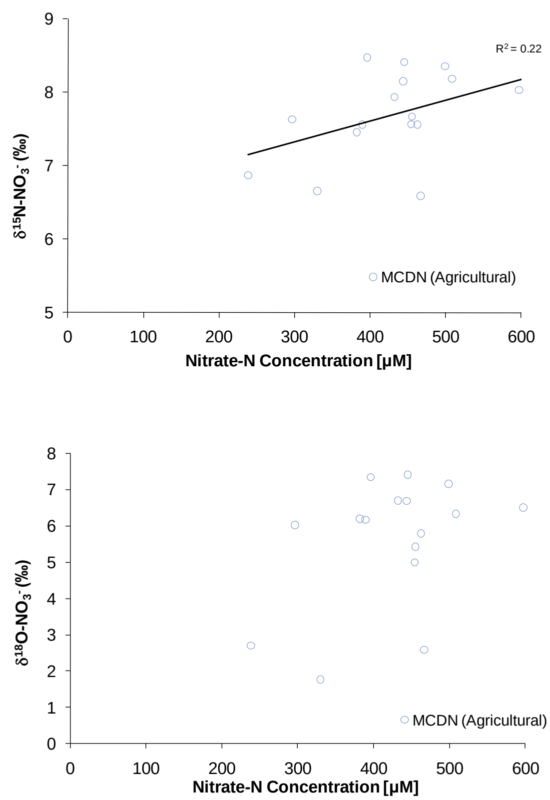


Figure S7. Relationships between $\delta^{15}N\text{-}NO_3^-$ and $\delta^{18}O\text{-}NO_3$ and nitrate concentrations in MCDN.

Importance of Wastewater vs. Lawn Fertilizer in Catchments

A mass balance analysis provides further information on the complexity of determining the vulnerability of various nonpoint sources to watershed N export. There are approximately 170 septic systems and 370 people located within the BARN watershed (Law et al. 2004, Wu and Band, *unpublished data*). Assuming that individuals release a mean of 4.8 kg N/yr per capita (Valiela et al. 1997), human waste contributes a total input of 4.7 kg N/ha/yr in the BARN watershed. This does not take into account further losses of septic N due to denitrification, volatilization of ammonia, or adsorption of ammonium within septic systems, which can be substantial (Valiela et al. 1997). Thus, an upper estimate of N inputs derived from human waste (without taking into account any losses in septic systems) is still only 50% of estimated watershed N inputs from lawn fertilization (9.5 kgN/ha/yr) (Law et al. 2004). However, septic system NO_3^- may be more vulnerable to watershed N export to the stream than fertilizer NO_3^- because it enters the environment in highly concentrated form and below the biologically active zone of the soil and can enter ground water (Gold et al. 1990, Kaushal et al. 2006). In contrast, fertilizer is added to surface soils with growing plants, and has been shown to be highly retained in Baltimore lawns (Raciti et al. 2008). Likely septic systems contribute NO_3^- to BARN and other suburban streams and should be addressed in efforts to reduce N exports from these watersheds.

The potential importance of wastewater is consistent with information from other chemical tracers of sewage inputs at this site including fluoride, which increases along the Gwynns Falls as it traverses from suburban areas to progressively urban areas (Figure S8).

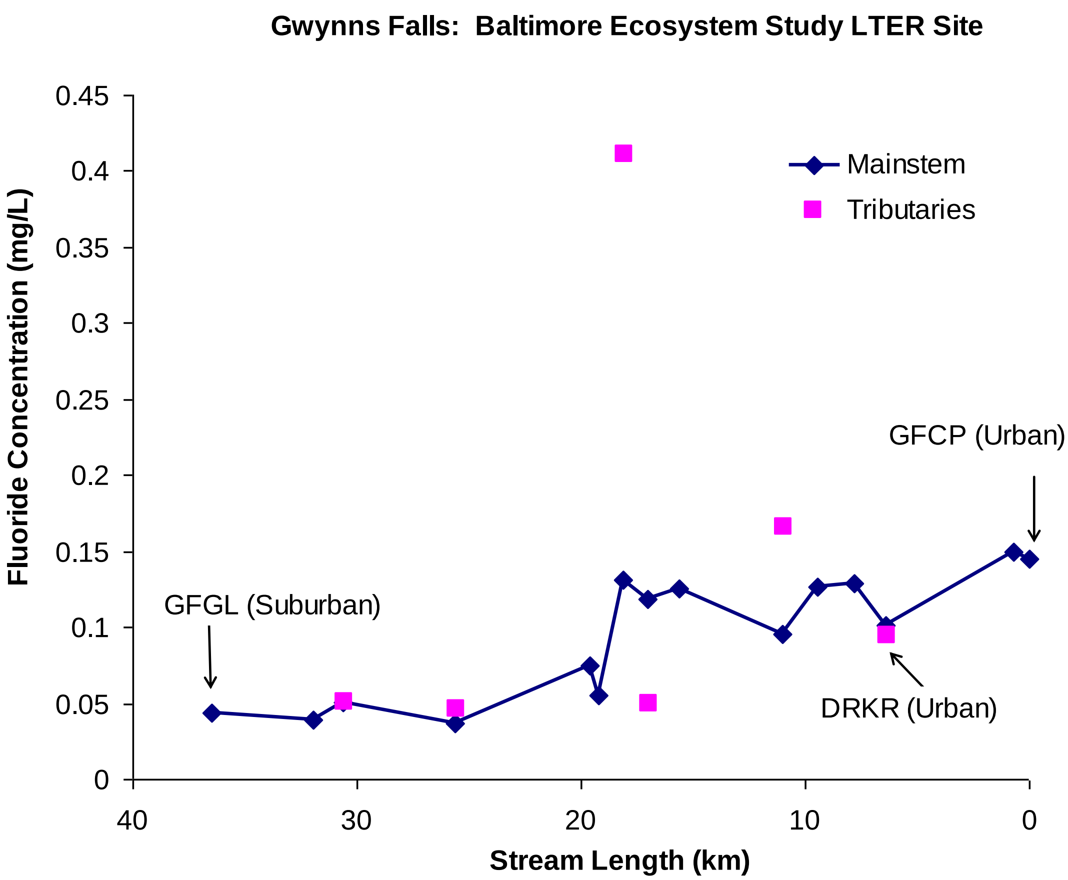


Figure S8. Fluoride concentrations indicate leaks in potable water and/or sewage along the Gwynns Falls (Kaushal and Belt In Preparation).

Nitrogen Transformations in an Urban Watershed with Storm Drain

Our results indicate that δ^{13}C-POM and δ^{15}N-POM in streams shifted with percentage impervious surface coverage in the watershed. Interestingly, our results were similar to Ulseth and Hershey (2005) which showed a pattern with δ^{13}C-POM across urbanization. We observed the opposite pattern as Ulseth and Hershey (2005), however, and this may have been due to their focus on the impacts of wastewater treatment plants on carbon subsidies in streams. There were no wastewater treatment plants in our study watersheds and our results suggest the importance of carbon subsidies from the landscape to streams. Increases in δ^{13}C-POM suggests changes in organic carbon sources from C3 plants to C4 plants from extensive residential lawns in Baltimore, Maryland. Another more likely hypothesis is that the shift in signatures may reflect terrestrial organic carbon sources that are less biologically degraded and more labile in urbanized watersheds. Work has shown that δ^{13}C-POM and δ^{15}N-POM can vary across watershed land use in rivers due to changes in terrestrial contributions and algal production (Kendall et al. 2001). Previous work has also suggested that the δ^{13}C-POM decreases with respect to the original value as a result of microbial alteration and selective preservation of ^{13}C depleted organic compounds (Lehmann et al. 2002). There can be substantial biological degradation of particulate organic carbon in forest streams, but physical degradation and fragmentation due to storms can be particularly important in urban streams (Gessner et al. 1999, Paul et al. 2006). Thus, there may be increased quantity and lability of carbon delivered to urban streams compared to forest streams, and this may also influence N transformations along hydrologic flowpaths in urban watersheds (Sivirichi et al. 2011). A shift in organic carbon sources, inputs, and quality in response to urbanization may influence N transformations in storm drains, and this warrants further study in urban drainage networks.

Changes in N Sources during Storms and Management Implications

Estimates from the mixing model showed that both wastewater and atmospheric deposition were important sources of nitrate-N to streams, but varied across runoff, nitrate-N concentration, and daily nitrate-N loads (Figure S9-S10).

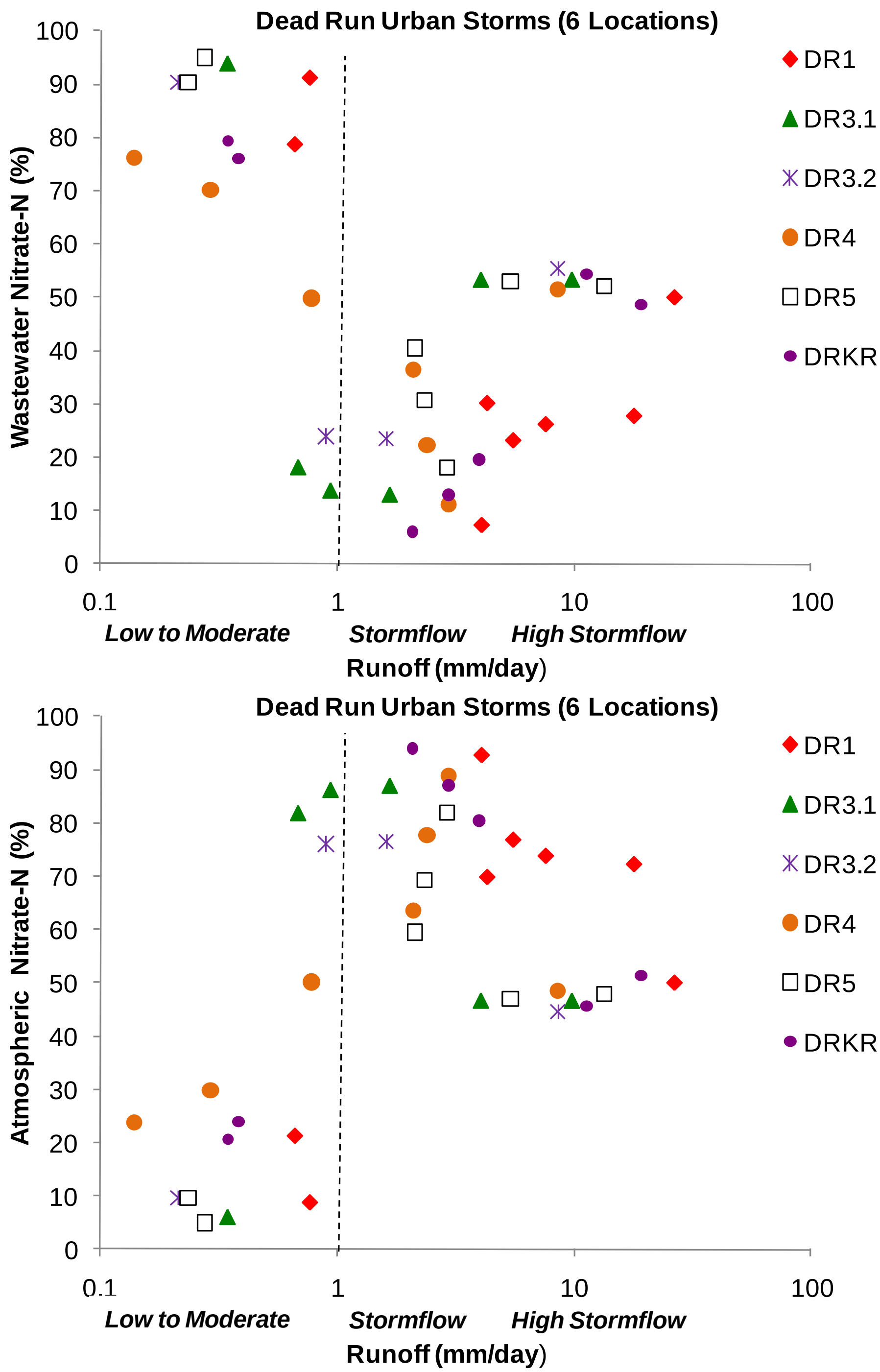
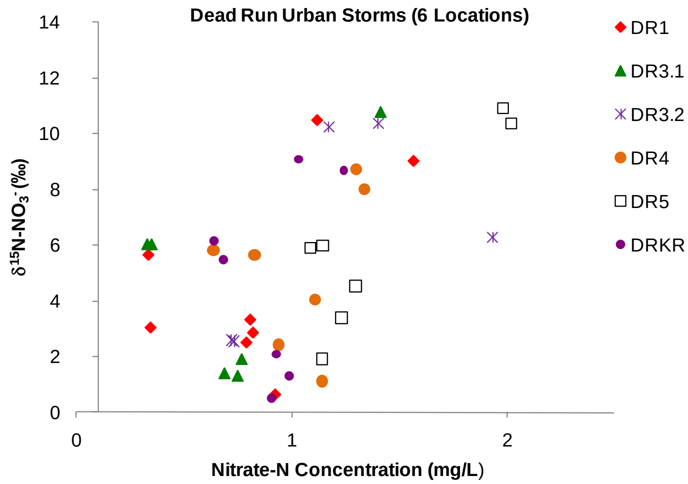
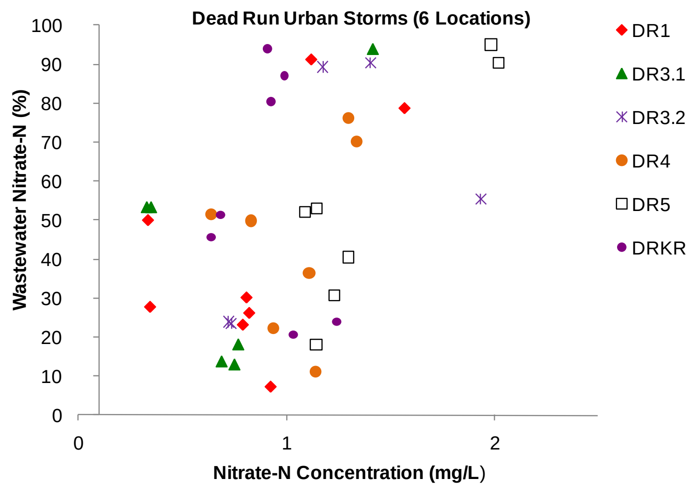


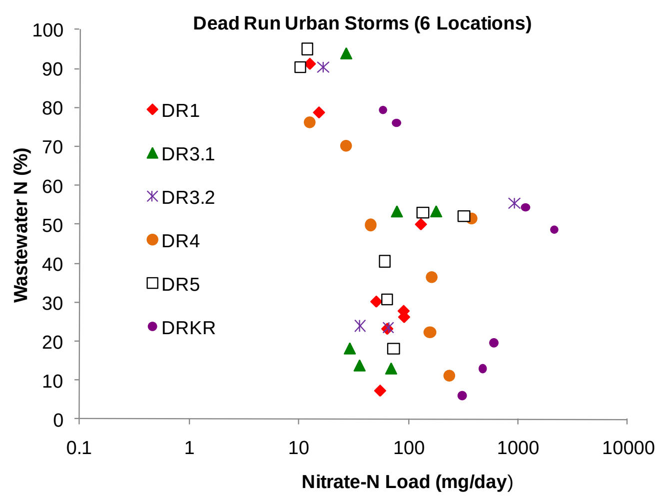
Figure S9. Source apportionment of wastewater vs. atmospheric nitrate-N using a simple 2 endmember mixing model in Dead Run subwatersheds across runoff conditions. Values plotted represent estimated proportions for individual samples, and ranges for subwatersheds are presented in Table 2 of the paper.



Dead Run Urban Storms (6 Locations)
$\delta^{15}N\text{-}NO_3^-$ (‰)
Nitrate-N Concentration (mg/L)
DR1
DR3.1
DR3.2
DR4
DR5
DRKR



Dead Run Urban Storms (6 Locations)
Wastewater Nitrate-N (%)
Nitrate-N Concentration (mg/L)
DR1
DR3.1
DR3.2
DR4
DR5
DRKR



Dead Run Urban Storms (6 Locations)
Wastewater N (%)
Nitrate-N Load (mg/day)
DR1
DR3.1
DR3.2
DR4
DR5
DRKR

Figure S10. Relationship between nitrate-N concentration and $\delta^{15}N\text{-}NO_3^-$. Source apportionment of wastewater vs. atmospheric nitrate-N in samples from Dead Run subwatersheds across nitrate-N concentrations and daily nitrate-N loads.

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