The skewed N : P stoichiometry resulting from changing atmospheric deposition chemistry drives the pattern of ecological nutrient limitation in the Ganges

Despite the fact that the human alteration of nitrogen (N) cycle is in part linked with other major biogeochemical cycles, particularly that of carbon (C)[1] and phosphorus (P)[2], most of the geosphere–biosphere models do not explicitly consider the changing state of C/N and N/P coupling and the associated shifts in ecosystem functioning[3]. Such coupling occurs with specific elemental stoichiometries and regulates the status of ecosystem functioning (e.g. C : N : P in autotrophic growth)[2] and climate change drivers[4]. On a global scale, atmospheric deposition (AD) has become the dominant vector of N and P inputs[3,5]. However, most of the information on AD-induced changes in N and P dynamics is from the temperate world; the projected trends in tropical regions are a cause of concern[6,7]. In this long-term study (March 2007 to February 2013) conducted at seven sites (Figure 1) along a 35 km stretch of the Ganga River at Varanasi (25°18′N lat. and 83°1′E long.), we show that the increasingly high input of AD-nutrients has shifted the N : P stoichiometry and, by implication, the pattern of ecological nutrient limitation in the Ganga River.

Site selection was based on sub-catchment heterogeneity and the magnitude of atmospheric loading. Site 1 was in a relatively natural state and all other sites were human-disturbed. AD samples, collected using bulk samplers, were analysed for NO_3^-, NH_4^+ and PO_4^{3-} spectrophotometrically. Surface run-off and mid-stream river water samples were analysed for dissolved organic nitrogen (DON), dissolved inorganic nitrogen (DIN), dissolved reactive phosphorus (DRP) and dissolved organic carbon (DOC) following standard methods[8]. Chlorophyll *a* biomass was measured using acetone extraction procedure and gross primary productivity (GPP) following light and dark bottle method[8]. Significant effects of site and time series were tested using analysis of variance (ANOVA).

Our results showed that although there was over 1.4–2.2 fold increase in AD-NO_3^-, NH_4^+ and PO_4^{3-}, the N : P stoichiometric ratio of AD declined over time (Figure 2). AD was highest at site 7 and lowest at site 1. The run-off concentrations and N : P stoichiometry showed trends similar to AD. DIN appeared the dominant vector of hydrologic N-export from polluted sub-catchments accounting for 55–70% of total N being added to the river through run-off. Concentration of DIN and DRP as well as the N : P stoichiometric ratio in the river increased over time. Concentration of DOC in run-off and in the river increased consistently over time; the river C : N ratio, however, showed an opposite trend. Concentration of river DOC showed significant seasonality. Chlorophyll *a* biomass and GPP showed marked synchrony with AD and river nutrients (Figure 2).

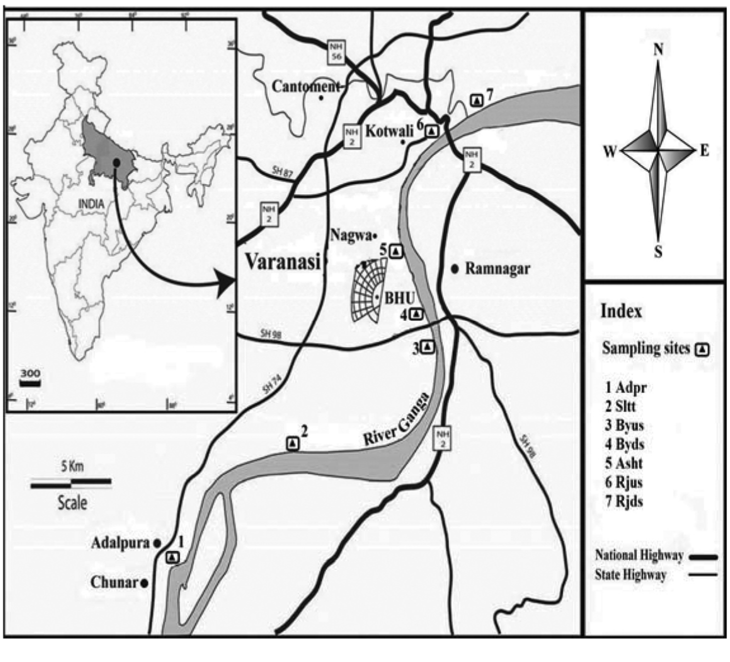

Figure 1. Location map showing sampling sites at the Ganga River. 1, Adalpura (Adpr); 2, Shultankeshwar ghat (Sltt); 3, Bypass upstream (Byus); 4, Bypass downstream (Byds); 5, Assighat (Asht); 6, Rajghat upstream (Rjus); 7, Rajghat downstream (Rjds).

In addition to urban-industrial activities, which intensify as one travels from site 1 to 7, the city side of the river, along with urban-industrial release, annually receives massive emission from burning of over 36,000 dead bodies using over 25,000 tonnes of dry wood in the process of cremation. We compare N : P stoichiometry of AD to run-off in order to assess the potential effect of AD on run-off nutrient flushing. Despite rising trends in AD input over time, a declining trend in N : P stoichiometry indicates relatively more P loading from biomass burning along the study gradient. This has relevance switching over P-limitation of phytoplankton production to N-limitation in the long-run. The site-wise N : P stoichiometry of AD and run-off, however, did not exactly follow a correspondence. For instance, site 1 with highest AD-N : P showed lowest N : P ratio for run-off. With a few exceptions, N : P ratio was relatively higher at the agricultural site (site 3), indicating the influence of local controls such as agriculture[9].

Both the concentrations of DIN and DRP as well as the elemental ratios may have a suite of biological effects, and the anthropogenic drivers could profoundly affect these parameters. For instance, the AD-nutrients enhance phytoplankton production[9] and, by implication, the turnover of DOC. The pulsed increase in

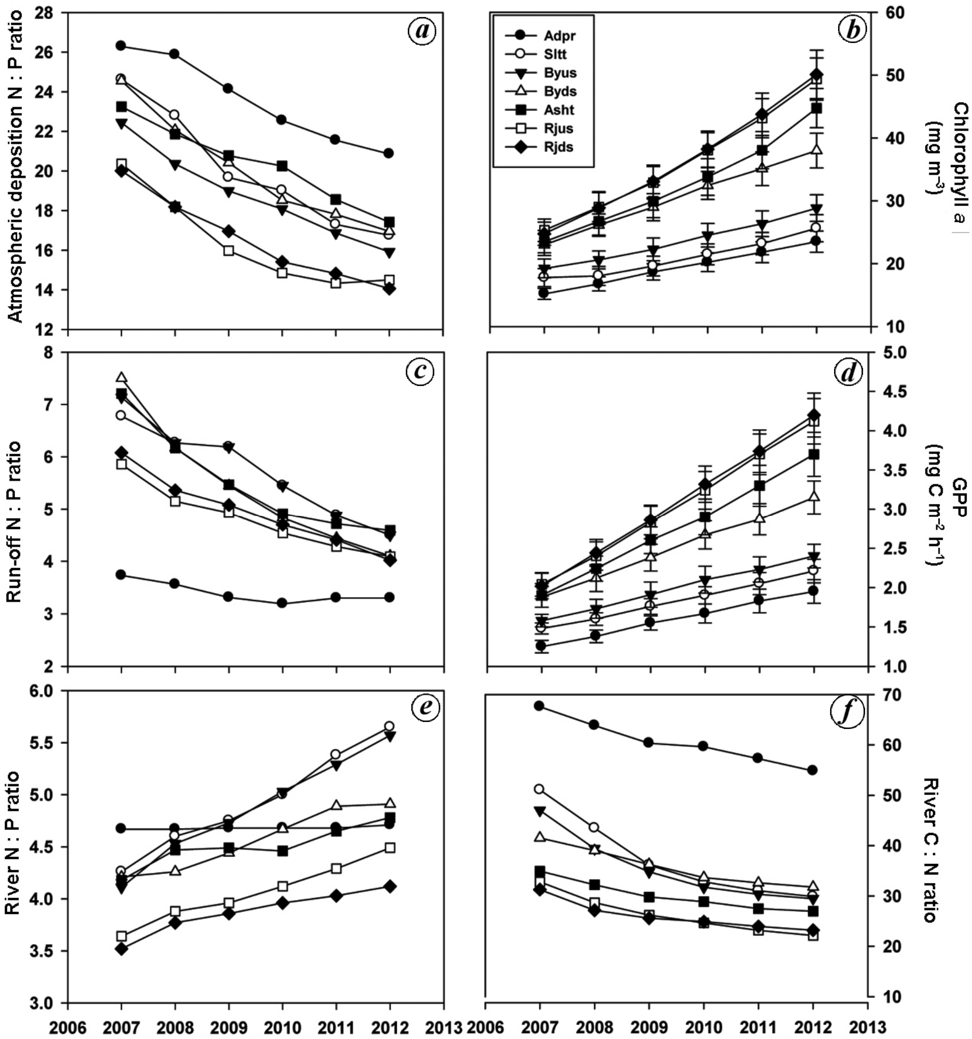

Figure 2. Year-wise trends in N : P stoichiometry of atmospheric deposition (***a***), runoff (***c***) and river water (***e***); and chlorophyll *a* biomass (***b***), gross primary productivity (***d***) and C : N ratio (***f***) at seven study sites of Ganga River. For chlorophyll *a* biomass and primary productivity, the values are mean (n = 36) ± 1 SE.

chlorophyll *a* biomass and associated shift in DOC are important evidences that pelagic production is contributing to changes in river DOC. The overall trend in river DOC, however, could be the result of tightly coupled effect of sewage input, land use and AD-linked terrestrial flushing together with pelagic production[9–11]. We use C : N ratio and C : chlorophyll *a* ratio to address this issue. Meyers[12] referred C : N > 20 to land plant origin. In the present study C : N ratios were invariably high (> 22), indicating allochthonous influence. The contribution of land-derived DOC appeared highest at site 1 (C : N > 60), which declined consistently, although remained above 22, with increasing pelagic production downstream. An increasing trend in C : chlorophyll *a* ratio indicates rising contribution of autochthonous C to river DOC along AD gradient. Further, a declining trend in DOC : P ratio (from 173.6 in site 1 to 63.2 in site 7) through downstream sites is characteristic of low ecosystem respiration to production ratio[13].

To assess the potential effects of AD-nutrients on phytoplankton nutrient limitation, we compare the N : P stoichiometry of AD with phytoplankton uptake requirements. In tropical waters, where light and temperature regimes generally remain optimal, phytoplankton production is primarily nutrient-limited. Since we collected samples at 15–25 cm depth, we consider that phytoplankton production was not limited by light availability. Our results showed that the atmospheric input of N and P was lower than the stoichiometric requirements of phytoplankton. The stoichiometric mass balance computation indicated that complete utilization of AD-N would result in fixation of 463.34–2554.46 μmol C m^{-2} d^{-1}, while AD-P would be responsible for 61.10–1099.90 μmol C m^{-2} d^{-1}. These values represent 19.34–31.20% (for N) and 2.50–13.44% (for P) of average GPP observed in the present study. Baker *et al.*[5] observed that AD-N inputs support fixation of 120–1290 μmol C m^{-2} d^{-1} representing 0.7–7.6% of average depth integrated GPP in the tropical Atlantic Ocean. Izquierdo *et al.*[14] observed that AD-P accounted for 24–33% of AD-P induced annual production in the western

Mediterranean Sea during strong dust events. In the present study, despite a declining trend in the N : P stoichiometry of AD and run-off over time, N : P ratio in the river increased suggesting higher input of N relative to P (may be through sewage, etc.) during subsequent years. These changes indicate that the N-limitation of initial years/upstream sites may shift to P-limitation over time. A switchover from N to P-limitation would affect aquatic ecosystem functioning because P-limited algae constitute poor-quality food for consumers[2]. However, P augments phytoplankton growth in waters where N : P > 16 : 1 (average required cellular ratio). In the present study, despite a rising trend over time, the river N : P stoichiometric ratios were very low (< 6) and skewed in favour of P relative to N, indicating that P is unlikely the principal element limiting phytoplankton in the Ganges. This has serious concern because the producer diversity is reduced when resource supply ratios are skewed in favour of one particular nutrient relative to other[2]. Further, the feedback effect that enhances denitrification buffering against elevated N loading would enhance sediment P release causing a self-fertilizing effect. Thus, the management priorities reducing N sources alone will not work. Substantial and sustained intervention is also needed for reducing P sources. The study provides important cues on which the action plan for integrated river basin management can be keyed.

1. Gruber, N. and Galloway, J. N., *Nature*, 2008, **451**, 293–296.
2. Elser, J. J. *et al.*, *Science*, 2009, **326**, 835–837.
3. Galloway, J. N. *et al.*, *Science*, 2008, **320**, 889–892.
4. Langley, J. A. and Megonigal, J. P., *Nature*, 2010, **466**, 96–99.
5. Baker, A. R., Weston, K., Kelly, S. D., Voss, M., Streu, P. and Cape, J. N., *Deep Sea Res.*, 2007, **154**, 1704–1720.
6. Phoenix, G. K. *et al.*, *Global Change Biol.*, 2006, **12**, 470–476.
7. Pandey, J., Singh, A. V., Singh, A. and Singh, R., *Bull. Environ. Contam. Toxicol.*, 2013, **91**, 184–190.
8. APHA, *Standard Methods for the Examination of Water and Wastewater*, American Public Health Association, Washington DC, 1998.
9. Pandey, U. and Pandey, J., *Biogeochemistry*, 2013, **112**, 537–553.
10. Monteith, D. T. *et al.*, *Nature*, 2007, **450**, 537–540.
11. Chattopadhyay, S., Asa Rani, L. and Sangeetha, P. V., *Curr. Sci.*, 2005, **89**, 2163–2169.
12. Meyers, P. A., *Org. Geochem.*, 2003, **34**, 261–289.
13. Hanson, P. C., Pollard, A. I., Bade, D. L., Predick, K., Carpenter, S. R. and Foley, J. A., *Global Change Biol.*, 2004, **10**, 1285–1298.
14. Izquierdo, R., Benitez-Nelson, C. R., Masque, P., Castillo, S., Alastuey, A. and Avila, A., *Atmos. Environ.*, 2012, **49**, 361–370.

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