

revealed that the indoor wall surfaces were not having residual effect of the sprayed insecticide. It can be explained primarily to the quality of spray and smearing of houses with white-wash on the occasion of festivals. The high fever rate with low SPR warrants further investigation to find out the cause of fever other than malaria.

The study shows that the efforts of local malaria control operations are thwarted by the attitude of local population by not adhering to complete treatment, smearing of houses after indoor residual spray and not realizing the importance of IRS (as they do not allow spray in all rooms). Socio-cultural aspects of tribal inhabitants responsible for maintaining high degree of malaria have been well documented⁷⁻⁹ from intense malarious regions of Assam, Orissa and Madhya Pradesh. Inaccessibility due to difficult terrain and non-compliance to treatment by inhabitants are the prime reasons resulting in outbreaks of malaria in tribal areas¹⁰⁻¹³.

There is need to impart health education to tribal communities by audiovisuals (i) showing the benefits of adhering to radical treatment to avoid deaths, (ii) to educate them not to smear the sprayed surfaces and ensure complete coverage of houses and rooms, (iii) promotion of insecticide-treated bed nets in Ashram schools, and (iv) introduction of blister packs for specific age groups of fever/malaria cases for more effective radical treatment. The finding of *P. malariae* necessitates the importance of careful blood slide examination to know the actual prevalence of parasite species requiring specific treatment strategy. There is also need to monitor drug resistance in *P. falciparum* to chloroquine in different areas and to ensure whether two rounds of IRS are sufficient to exert vector control in an area with transmission window open for around eight months.

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Nitrous oxide fluxes in a tropical shallow urban pond under influencing factors

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Fluxes of nitrous oxide (N₂O) were measured *in situ* monthly for two years from a tropical shallow urban pond, receiving influx of agricultural run-off from the surrounding water shade and domestic sewage, located in Ujjain city, Madhya Pradesh. Results revealed that the shallow pond is a continuous source of N₂O with relatively low efflux value (0.00 to 0.51 mg m⁻² d⁻¹) and maximum emission generally occurred during the hot seasons (77% of annual emission), with annual mean of 1.0 kg N₂O ha⁻¹ yr⁻¹. Several influencing factors like pH, temperature, total nitrogen, inorganic nitrogen stock and organic carbon were studied concomitantly with N₂O gas flux measurement. N₂O emissions were positively and significantly correlated with sediment total nitrogen and surface water temperature. The study concludes that the shallow tropical water body is a source of N₂O flux, but does not have significant efflux compared to the terrestrial habitats.

AQUATIC ecosystems are considered to be significant sources of nitrous oxide (N₂O), a stable trace gas contributing to the

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anticipated global warming and also acting as a reactant in the destruction of stratospheric ozone following photolytic oxidation to nitric oxide (NO)¹. N₂O is 296 times more effective as an absorber of infrared radiation than CO₂ on a 100-year timescale and its radiative forcing is 6% of the total from all of the long-lived and globally mixed greenhouse gases². Its atmospheric concentration^{2,3} has increased by 46 ppbv (16%) since 1750 and there is a net increase in atmospheric N₂O from 275 ppbv in 1900 to 314 ppbv in 1998 to a projected concentration of 340–350 in 2020. Our knowledge about processes and factors contributing to N₂O production and emission has improved due to research during the last several years^{4–9}, but uncertainties about the contribution to atmospheric N₂O from individual sources still exist. Wetlands and ponds are important retainers of nitrogen and anthropogenic nitrogen inputs via drainage water from fertilized agriculture field and domestic sewage are known to increase microbial nitrogen transformation in aquatic environment¹⁰, resulting in the increase in N₂O fluxes and ratio of N₂O:N₂ from wetland sediment. Thus, it seems that eutrophication might enhance N₂O emission to the atmosphere^{11,12}. According to Samuelsson and Klemetsson¹³, published data on N₂O efflux from freshwater sediments are sparse. There is an urgent need for information contributing to the assessment of the relative importance of wetlands ponds and lakes in N₂O emissions to the atmosphere.

One of the main characteristics of natural wetlands is the permanent or temporary water saturation¹⁴, and dominated by aquatic macrophytes which play an important role in the transport of oxygen in the wetlands and are able to oxidize the rhizosphere in the deep water and sediment¹⁵. The available oxygen is consumed by aerobic microorganisms near aerobic microsites where ammonium-oxidizing and nitrate-oxidizing bacteria nitrify available NH₄⁺ to NO₂⁻ and NO₃⁻ respectively. Nitrifying bacteria are widespread in the wetlands and play an important role in the turnover of nitrogen and pathway of coupled nitrification–denitrification; and denitrification depends on the rate at which NO₃⁻ is produced by nitrification¹⁶. The NO₃⁻ thus formed then diffuses into the anaerobic zone, where it is reduced to N₂O and N₂ by denitrifiers¹⁷. Under waterlogged conditions, the anaerobic sediment/soil has the potential for reducing N₂O to N₂ and the major product of denitrification¹⁸ under such strictly anaerobic soils is N₂ rather than N₂O. The coupled nitrification–denitrification, therefore, constitutes an important control of N₂O fluxes from the wetland sediment/soil, thus resulting as a source or sink of nitrous oxide. N₂O formation and consumption in wetlands are microbiological processes that are controlled by many factors. The present investigation aims to: (i) quantify N₂O fluxes periodically in hot and cool seasons under nutrient loading from the surrounding watershed in urban shallow pond, and (ii) monitoring diagnostic physico-chemical factors influencing N₂O flux in the water column and sediment interface.

For the study of N₂O flux measurement in the aquatic ecosystem, we considered the shallow, eutrophicated Solah

Sagar pond, which is a natural and perennial water body situated on Ujjain–Agar road, 5 km from Ujjain city (23°11'N lat and 75°43'E long and 491.7 m amsl), Madhya Pradesh, Central India. The north and west banks of the pond are surrounded by agricultural crop fields and the eastern side by a residential colony. The pond is shallow at the margins and the depth gradually increases towards the central area, attaining a maximum depth of approximately 3.5 m. The surface area of the pond is about 65,918 m² during the rainy season when it is completely filled. It harbours almost natural water, except receiving domestic urban wastewater through the earthen channel as point source from the adjoining urban habitation and agricultural drained water from the surrounding agricultural fields during the rainy season as non-point sources. Basically the pond has been utilized on a lease system for the cultivation of water-chestnut (*Trapa bispinosa*, family Trapaceae) and fish farming by the local fishermen (Bhoi)¹⁹. The entire luxuriant growth of water-chestnut sown during late March to mid-April every year gradually dies and sinks into the water, through decomposition after the manual harvesting of commercial nuts during October–November. After chestnut harvesting and before its sowing in the next season, the water body under study becomes dominated by water hyacinth (*Eichhornia crassipes*) cover. Every year the dense growth of water hyacinth is manually removed as noxious weed from the wetland for chestnut cultivation. The wetland is kept idle for 1–2 months between weed (*E. crassipes*) removal and crop (*Trapa bispinosa*) sowing. During this period an intensive growth of *Ceratophyllum demersum* (submerged plant) and *Lemna minor* (free-floating plant) occurred.

The climate of the area is typically monsoonal with hot summer (May–June) and cold winter (January–February). The year can be divided into three distinct seasons such as summer (March to mid-June), rainy (mid-June to mid-October) and winter (November to February). The mean monthly maximum temperature varied from 21.0 (January) to 36.5°C (May) and the mean minimum temperature varied from 13.0 (January) to 29.5°C (May). Average rainfall amounted to 1015 mm during the study period. Most of the precipitation occurs in July and August, although the rainy season extends to mid-October. About 80–90% of the total annual precipitation is received during this period.

Gas samplings were carried out using closed chamber technique as standardized by National Physical Laboratory (NPL), New Delhi²⁰. For the collection of gas samples from the sediment–water interface, a circular motorcycle rubber tube filled with air was attached to the bottom rim of a transparent perspex chamber made from 6 mm plexiglass sheet to make it float on the surface of water. The chamber cover is fitted with a sampling port and a port to which a plastic pressing pump is attached for uniform mixing of air inside the chamber. The sampling port is a hole plugged with a rubber septum, through which a 50 ml glass syringe was inserted with hypodermic needle to collect gas samples from

the chamber. Gas samples were transferred to pre-evacuated, sealed glass vials of 100 ml volume at 0, 10, 20 and 30 min and ambient air sample was also collected near the chamber to compare with 0 min flux at the time of calculation. Flux measurements were made in the late morning at 10 am and afternoon by 3 pm on each sampling day. The temperature inside the perspex chamber was recorded at the time of sample collection (0, 10, 20, 30 min) using a thermometer (–10 to 100°C range, Co Immersion Zeel, England) fixed on the inside wall of the chamber for the calculation of box volume at STP. Water temperature was also measured with a thermometer adjacent to the chamber.

The collected gas samples were brought to the laboratory and analysed for N₂O fluxes at NPL, using a SRI 8610 C gas chromatograph (SRI Instruments, USA) equipped with a 63 Ni Electron Capture Detector (ECD) and Porapack Q column. The operating temperature of the detector, injector and oven was 350, 150 and 80°C, respectively, with nitrogen as a carrier gas at the flow rate of 30 ml/min. The gas chromatograph was calibrated each day before starting N₂O flux measurement, using 211 and 1242 ppb N₂O as primary standards.

N₂O flux was calculated using the formula:

$$F = \frac{\Delta x}{10^6} \times BV(\text{STP}) \times \frac{44 \times 10^3}{22400} \times \frac{1}{A} \times \frac{60}{t}$$

where F is the efflux of nitrous oxide in $\mu\text{g m}^{-2} \text{h}^{-1}$, Δx the change in concentration of nitrous oxide in ppbv from time '0' to 't' min, BV(STP) the Box volume at standard temperature and pressure in cm^3 , and A the area within the chamber in m^2 .

After performing gas sampling, the bottom sediment of the pond was sampled by the Ekman's grab sampler (suitable for soft sediment) at every sampling date after taking water sample near the gas sampling point. The amount of $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$ was obtained from extracts of field sediments with 2 M KCl (1:5 w/v) and analysis was done by Bremner's distillation method, total nitrogen (TKN) by microkjeldahl method, organic carbon (C) in oven-dried sediment sample by dichromate oxidation and titration with ferrous ammonium sulphate²¹. Microbial biomass nitrogen contents were determined by the chloroform-fumigation-extraction method²², sediment and water pH using a pH meter equipped with glass electrode (1:2.5 soil: water ratio, w/v), and water surface temperature by mercury thermometer (Co Immersion Zeel, England).

The periodic N₂O flux in the eutrophicated wetland ecosystem (Solah Sagar pond) during the two years ranged from 0.00 to 21.24 $\mu\text{g m}^{-2} \text{h}^{-1}$ (Figure 1a). The fluxes were high during summer compared to rainy and winter season (Figure 1b). Water temperature in the surface 5 cm water column ranged from 17 to 31°C, while average temperature was almost same during summer and rainy season, and 9°C lower during winter (Table 1). The $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$ ranged from 0 to 4.55 and 1 to 3.27 mg l^{-1} respectively, in

the water column of the pond. Total nitrogen in the pond sediment ranged from 0.27 to 0.6% during the study. No marked seasonality was observed in sediment inorganic N-pool, but $\text{NH}_4^+\text{-N}$ was 8 to 11 times greater than $\text{NO}_3^-\text{-N}$ (Table 1). Organic carbon and C:N ratio in the pond sediment was higher during winter than summer and rainy season (Table 1), pH varied from 7.0 to 8.9 and organic carbon from 1.5 to 4.02%. The overall trend indicates that N₂O fluxes were positively correlated with water temperature ($r = 0.56$, $P < 0.004$, $n = 24$; Figure 2a) and total nitrogen of the sediment ($r = 0.47$, $P < 0.019$, $n = 24$; Figure 2b). N₂O emissions from the wetland sediment did not correlate with an increase in the ammonium-N and nitrate-N concentration of the overlying water or bed sediment.

N₂O efflux recorded during the two years of study indicated that the shallow pond is a continuous source of N₂O with relatively lower value of N₂O efflux (Figure 1a) compared to terrestrial ecosystems^{23,24}. However, during January 1998, N₂O flux value was recorded as zero. As we know, soils can act as source and sink of nitrous oxide and N₂O fluxes in somewhat identical habitat, e.g. submerged rice field, indicate that not only zero flux but several negative fluxes (less than zero) were also observed²⁵. Also, under waterlogged conditions, the anaerobic soils/sediments have greater potential for reducing N₂O to N₂. The major product of denitrification¹⁸ under strictly anaerobic soils is N₂ rather than N₂O and eutrophic lakes^{26,27} may act as sinks for atmospheric N₂O. Low annual rate of N₂O emission from the wetlands may be because of inhibited nitrogen mineralization and nitrification rate due to lack of oxygen in the sediment layer under continuous submergence²⁸, as well as less diffusion from sediment to bed space under pressure of standing water column and dissolution of N₂O in standing water²⁹.

The summer season has been marked with temperature increase (ambient temperature 38–42°C, water column temperature 24–31°C) and highest value of sediment microbial biomass nitrogen (Table 1). This indicates enhanced level of microbial activity, which increased the decomposition of organic matter and enhanced the process of nitrification and denitrification. Lowering of the water table during summer has been found to increase nitrogen mineralization in wetlands³⁰, and water table reduction increases N₂O fluxes^{31–33}. The emergent Trapa plants being rooted initially during March–April, can help in developing an oxic zone in the sediments near the rhizosphere, because oxygen is transported by aerenchymatous parts of plants and it may help maintain a small nitrifying population even under waterlogged condition³⁴. Nitrification is possible in the shallow oxic zones at the soil–water interface and around the root of plants. These oxic zones are of great importance for N₂O emission from vegetated wetland sediments for two reasons. First, these are the sites where N₂O is potentially produced during nitrification. The N₂O thus produced may then rapidly be lost to the atmosphere via the gas vascular system of the plant³⁵. Second, these are sites where

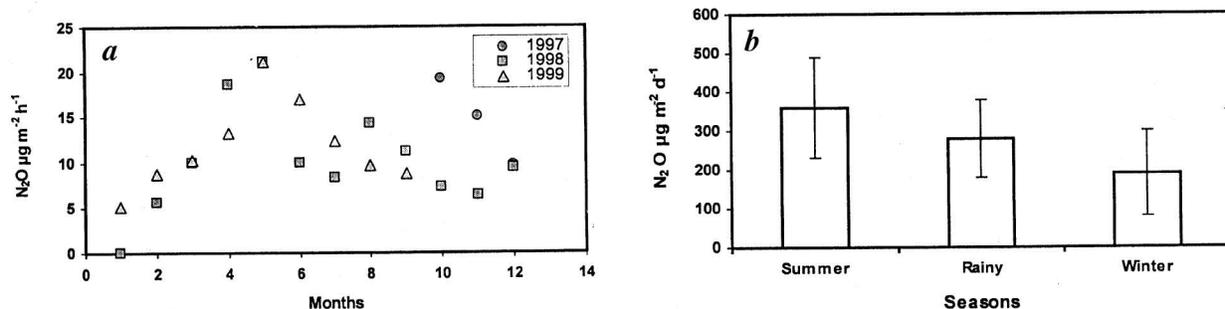


Figure 1. Monthly (a) and seasonal (b) variation in nitrous oxide efflux monitored during two years of study (1997–99).

Table 1. Seasonal variation in physico-chemical parameters of water column and sediment in the shallow pond (1997–1999)

Parameter	Hot season		Cool season
	Summer	Rainy	Winter
Sediment			
pH	7.79	7.45	7.24
NH ₄ ⁺ -N (mg kg ⁻¹ ODS)	24.1 (5.69)	21.38 (4.24)	20.77 (2.59)
NO ₃ ⁻ -N (mg kg ⁻¹ ODS)	2.81 (0.56)	1.89 (0.42)	2.75 (0.87)
Total-N (TKN) (%)	0.44 (0.08)	0.44 (0.10)	0.42 (0.11)
Organic carbon (%)	2.61 (0.75)	2.30 (0.61)	3.00 (0.58)
C : N ratio	6.11 (2.18)	5.48 (1.74)	7.63 (2.74)
Microbial biomass-N (mg kg ⁻¹ ODS)	98.0 (6.2)	60.5 (7.31)	39.0 (4.65)
Pond water			
Water temperature (°C)	27.0 (2.2)	26.7 (3.0)	18.7 (1.6)
NH ₄ ⁺ -N (mg l ⁻¹)	2.81 (1.54)	2.59 (1.23)	2.80 (0.89)
NO ₃ ⁻ -N (mg l ⁻¹)	1.90 (0.71)	1.45 (0.37)	1.77 (0.68)

ODS, Oven dry soil, TKN, Total Kjeldahl nitrogen; Values in parentheses are standard deviation.

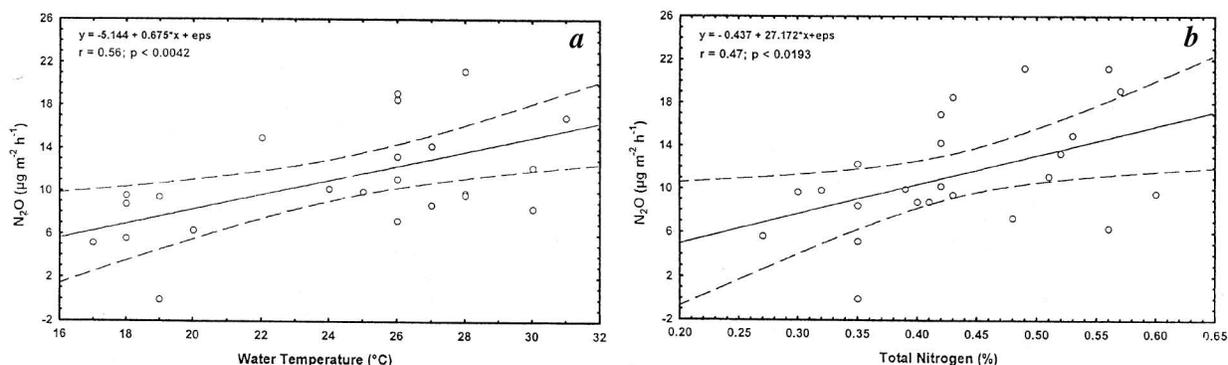


Figure 2. Relationship between N₂O flux and (a) pond water temperature and (b) sediment total nitrogen. Dotted curves show 95% confidence interval for the regression.

ammonium is oxidized through nitrification to nitrate, which then can diffuse into the adjacent anoxic zones to be reduced in the sediment and thus to allow N₂O production³⁶. Arth *et al.*³⁷ show that nitrification is the prerequisite for N₂O production and denitrification was coupled to nitrification.

This is only possible in the oxidized soil surface and in the plant rhizosphere, where O₂ is available³⁸. The existence and activity of nitrifiers in the rhizosphere of aquatic plants have been reported^{39–41}. This indicates that the coupling of nitrification–denitrification is a common characteristic

near the plant root–sediment interface and is important in the control of the emissions and loss of N₂O and other gaseous N-compounds from the wetlands into the atmosphere^{37,42–44}. In contrast, during winter season lower temperature would result in a combination of decreased microbial activity (low microbial biomass nitrogen during winter, Table 1), increased N₂O solubility and slower gaseous diffusion in water⁴⁵.

Aquatic sediment is considered eutrophicated due to an increased load of nitrogen compounds and these increased nitrogen inputs are known to increase microbial processes like nitrification and denitrification in aquatic environments, resulting in increase in nitrous oxide production^{11,12}. In the present investigation total nitrogen content of wetland sediment ranged from 0.24 to 0.6% during the study period and positively correlated with N₂O fluxes ($r = 0.47$, $P < 0.05$, $n = 24$; Figure 2*b*). It has been reported that an increase in nutrient loading enhances both the efflux of nitrous oxide¹³ and the ratio of N₂O:N₂ released to the water from the sediment^{11,12,46}. According to Seitzinger and Kroeze⁴⁷, about 1% (range 0.2–2%) of the nitrogen input from fertilizers, atmospheric deposition, and sewage to watersheds is lost as N₂O from different water bodies, following nitrogen leaching and run-off from watersheds into aquatic systems. Besides nitrogen loading in the pond, water column temperature affects N₂O fluxes and a significant positive correlation ($r = 0.56$, $P < 0.005$, $n = 24$) between water temperature and N₂O emissions was observed (Figure 2*a*). Temperature mainly affects N₂O turnover by affecting the overall process rate of nitrification and/or denitrification. N₂O emission from denitrification seems to be enhanced at lower temperatures, whereas the potential nitrification and denitrification rates tend to increase with increasing temperature^{48–50}. However, the effect of temperature on N₂O production is complicated, since several biochemical processes with different time constants and different dependencies on other variables like oxygen penetration, nitrate and ammonium availability are involved in the overall process^{51,52}. The relationship between pH and N₂O emissions is not as clear-cut, although N₂O emission increases with decreasing pH under conditions where N₂O was emitted mainly via denitrification⁵³. The relationship between pH and N₂O emission is complex and generally difficult to quantify⁵⁴. It is generally accepted that optimal pH for denitrification is between 7 and 8 and higher soil pH leads to greater reduction^{55,56} of N₂O to N₂.

N₂O fluxes in the shallow pond are governed by nitrogen input in the sediment–water column under the cover of decomposing aquatic plants. The eutrophicated pond thus symbolizes by excessive vegetation and ramifying roots in the water column which provide micro-sites for nitrifying–denitrifying process to occur and regulate N₂O fluxes from the pond. Higher N₂O fluxes during the hot season was probably due to enhancement of nitrogen mineralization leading to N₂O fluxes and was reflected in increased level of ammonium and low level of nitrified nitrate-N in the

sediment. However, concentration of N₂O from aquatic sources is considerable as in the present study, but not the dominant source of atmospheric nitrous oxide⁵⁷, compared to terrestrial habitats^{23,24}. Shallow ponds (wetlands) may be the major sink of nitrogen as they can denitrify up to 90% of nitrogen which enters⁵⁸, leading to lower N₂O emissions as in the present study. Further studies need to be done to estimate the key influencing factors attenuating the fluxes of N₂O to control ozone depletion and global warming.

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