Assessment of the Sources of Nitrate in the Changjiang River, China Using a Nitrogen and Oxygen Isotopic Approach

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The Changjiang River is the largest freshwater river in China. Here, the sources and variability in nitrate of the Changjiang River are assessed for the first time using dual isotopic approach. Water samples were collected once in August 2006 from the main channel of the Changjiang and its major tributaries. The concentrations and isotopic composition of nitrate were then analyzed for the waters in the Changjiang River. The $\delta^{15}N$ and $\delta^{18}O$ of NO_3^- ranges from 7.3‰ to 12.9‰ and 2.4‰ to 11.2‰ in the Changjiang River waters, respectively. The ranges of isotopic compositions of nitrate suggested that nitrification (including "modified fertilizer") and urban sewage effluent are the major sources of nitrate in the Changjiang River. The high $\delta^{18}O$-NO_3^- values were observed in the water of the upper reaches, indicated that the current drought might be one important reason for shifting of isotopes in the special sampling period. In addition, there was a strong positive relationship between $\delta^{15}N$-NO_3^- and $\delta^{18}O$-NO_3^-, which indicated that denitrification added to the enrichment of heavy isotopes of nitrate.

Introduction

River systems play a very important role in nutrient transportation and transformation in the Earth's surface environments. Nitrate levels have generally increased in natural rivers over the last century (*1*, *2*). Increasing nutrient inputs into estuaries and coastal oceans have been linked to eutrophication and seasonal hypoxia. In addition, according to the Bulletin of Marine Environmental Quality of China (*3*), approximately 30−80 red tide events were recorded each year from 2000 to 2005 in the East China Sea. Zhu et al. (*4*) reported that the characteristics of phytoplankton during red tide events were related to nutrient loading into the Changjiang River Estuary. Additionally, high levels of nitrogen pose a serious threat to drinking water supplies due to the promotion of eutrophication in lakes and reservoirs.

Identification of the sources and the variability of nitrate is an important step in improving the management practices associated with maintaining water quality in rivers. Stable isotopic compositions of nitrate can provide useful information that can be used to track the source of nitrate in rivers due to the distinct isotopic characteristics of the main sources of nitrate, such as rain, chemical fertilizers, and nitrate derived from nitrification (*5*, *6*). Furthermore, dual isotopic models can be useful to identify denitrification in aquatic ecosystems because during this process ^{15}N and ^{18}O are preferentially enriched in residual nitrate (*7*, *8*). Recent studies have shown that N and O isotopes of nitrate were good indictors to identify the sources and transformations of nitrate in small and middle catchments (*6*, *9*–*13*). Meanwhile, there are a few studies using dual isotopic approach to investigate the nitrate behavior in large river basins, such as Mississippi River (*14*, *15*), the Illinois River (*16*) and five German rivers (*17*). These studies have shown the various degree of success of dual isotopic technique to understand the nitrate sources in rivers.

The Changjiang River is the largest river with a big freshwater resource in China, playing an important role in socio-economic development of China. The concentration of dissolved nitrate in the Changjiang River has been well documented (*2*, *18*, *19*), but to date no isotopic studies have been conducted to identify the sources of nitrate in the river. In this study, the chemical parameters and isotopic compositions of dissolved nitrate in the water were analyzed in the Changjiang River Basin. This information was then used to evaluate the spatial variations in nitrate as well as the sources of nitrate and its possible transformation in the river.

Experimental Section

Characteristics of the Changjiang Drainage Basin. The Changjiang River is the longest river in China and the third longest river in the world. The river has a basin area of 18 $\times$ 10^5 km^2 with a mainstream river length of over 6300 km. Changjiang originates in the Tibetan Plateau at an elevation higher than 5000 m. The upper, middle, and lower reaches of the river are geographically divided by Yichang in Hubei Province and Hukou in Jiangxi Province. The major tributaries of the Changjiang River are the Yalongjiang, Minjiang, Jialingjiang, and Wujiang rivers in the upper reach, and the Hanjiang River, Dongting Lake, and Poyang Lake River System in the middle reach. Taihu Lake is the biggest lake and there is no large tributary in the lower reach (Figure 1).

The major portion of the Changjiang Basin is located in the humid subtropical zone and has a typical East Asian monsoon climate. The Changjiang River runoff primarily originates from precipitation. In a normal year, the upper, middle, and lower reaches account for 46.7%, 47.3%, and 6.3% of the total runoff, respectively (*19*). The basic characteristics of major tributaries in Changjiang River are listed in Table 1. The major tributaries listed in Table 1 contributed about 40% of water discharge for the Changjiang River. Approximately 25% of the cultivated land in China is located within the basin, 2.41 $\times$ 10^7 ha of which is composed of upland and paddy fields (*20*). The cultivated land cover ratio and population density for major tributaries of Changjiang River are calculated by a county-level statistical database of China (http://www.data.ac.cn/), and are listed in Table 1.

Sampling and Analytical Methods. Water samples were collected from the Changjiang main channel and the largest major tributaries (Yalongjiang, Minjiang, Jialingjiang, Wujiang, Hanshui and Dongting Lake, Poyang Lake) during August 2006 (Figure 1). In addition, samples were collected from urban wastewater in Yibin (YBWS) and from a small urban

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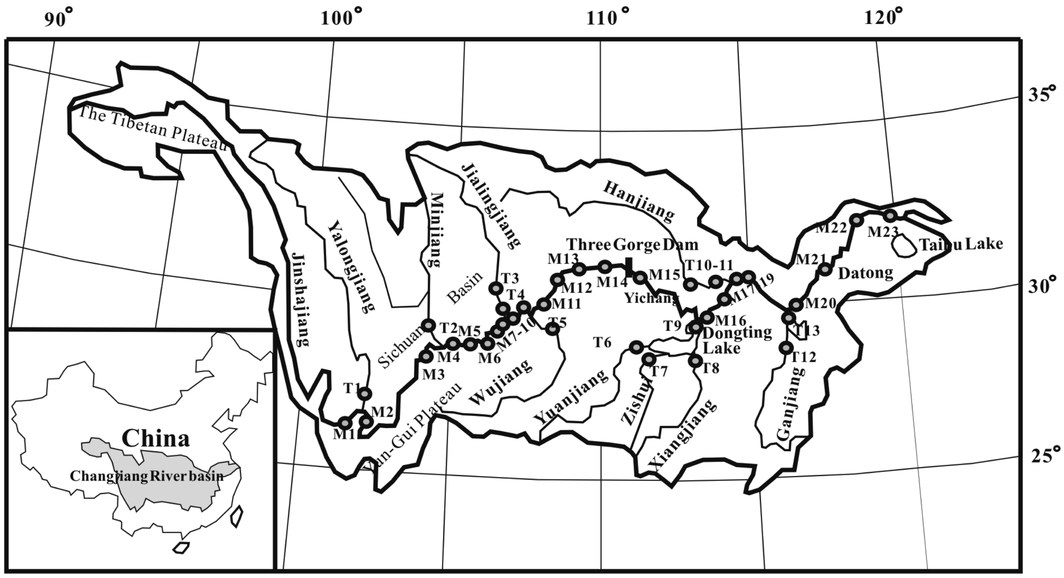

FIGURE 1. Changjiang River basin and locations of sampling sites.

TABLE 1. Characteristics of Major Tributaries in Changjiang River

	area[a] (km^2)	discharge[a] (m^3/s)	cultivated land ratio[b] (%)	population density[b] ($person/km^2$)
Yalongjiang (T1)	128444	1914	1.6	19
Minjiang (T2)	133000	2850	8.5	139
Jialingjiang (T3)	160747	2120	14.0	239
Wujiang (T5)	87920	1650	15.2	258
Yuanjiang (T6)	89163	2170	12.1	201
Zishui (T7)	28142	759	17.2	390
Xiangjiang (T8)	94660	2370	15.8	318
Hanjiang(T10)	159000	1710	17.7	236
Ganjiang (T12)	83500	2130	14.9	276

[a] Data are from http://www.cjw.com.cn/ (Changjiang Water Resources Commission). [b] Calculated from a county-level statistical database of China (http://www.data.ac.cn/).

river in Wuhan (WHWS) that had been contaminated by industrial and domestic wastewater (*21*). A portion of each water sample was filtered through 0.45-μm cellulose-acetate filter paper into polyethylene bottles with airtight caps and preserved with HCl to prevent biological activity.

The concentrations of Cl^- and NO_3^- were determined by ionic chromatography Dionex 90 with a precision of 5%. Deuterium and oxygen isotopes of water were analyzed in a Micromass IsoPrime mass spectrometer coupled to an automated line based on the equilibration between H-water and H_2 gas with a Pt catalyst, and between O-water and CO_2 gas. δD and $\delta^{18}O\text{-}H_2O$ have a precision of 3‰ and 0.2‰, respectively.

The methods used to isolate nitrate and subsequently convert it to $AgNO_3$ were adapted from Silva et al. (*22*). Nitrate was first collected on the ion-exchange columns using an anion resin, after which 2 M HCl was used to elute the nitrate from the resins. The SO_4^{2-} and PO_4^{3-} were removed by precipitation with excess $BaCl_2$. The DOC was removed by activated carbon. The sample was then passed through a cation exchange resin. An excess of Ag_2O was then used to remove the Cl and neutralize the solution to achieve a pH of about 6, after which the $AgNO_3$ solution was freeze-dried for isotopic analysis.

The ratios of isotopic N and O were determined using elemental analysis-isotope ratio mass spectrometry (EA-IRMS), and reported in the δ notation relative to N_2(air) and the Standard Mean Ocean Water (SMOW) in permil (‰). For nitrogen isotopic analysis, $AgNO_3$ was transferred into tin capsules and combusted in the combustion reactor, which was held at 1030 °C and contained CrO as well as silvered cobaltous/cobaltic oxide. N_2 was produced in a reduction reactor filled with copper held at 650 °C and sent to the mass spectrometer. For $\delta^{18}O\text{-}NO_3^-$ analysis, $AgNO_3$ was enclosed in silver capsules and pyrolyzed with the elemental analyzer at 1250 °C. The CO and N_2 produced by the pyrolysis were separated by GC column packed with a 5-Å molecular sieve. Isotopic standards N-1(IAEA), N-3(IAEA), and working standards were converted to gas for calibration and to check on reproducibility. The isotope results are given in δ units defined as:

$$\delta(‰) = ((R_{sample}/R_{standard}) - 1) \times 1000 \qquad (1)$$

where R represents the $^{15}N/^{14}N$ or $^{18}O/^{16}O$ ratios, respectively. The value of $\delta^{18}O$-N3(IAEA) was reported from +22.7‰ to +25.6‰ (*22–24*). Here, the $\delta^{18}O$-N3(IAEA) was assigned as +22.7‰ for isotopic calibration of $\delta^{18}O\text{-}NO_3$ (*23*). The analytical precision of the $\delta^{15}N$ and $\delta^{18}O$ values was generally better than 0.2‰ and 0.5‰, respectively.

Results and Discussion

Water Chemistry and Isotope Hydrology. The NO_3^- concentrations and isotopic compositions of the samples are listed in Table 2. In the natural waters, the concentration of NO_3^- was found to range from 4 to 94.7 μmol/L with a mean value of 60.4 μmol/L. Forest, bushes, shrubs, and grasslands cover major portions of the Jinshajiang (M1) and Yalongjiang (T1) rivers. The population densities of upper Jinshajiang (M1) and Yalongjiang rivers are general lower than 10 persons/km^2 (http://www.cjw.com.cn/). The contents of NO_3^- were found to be lower than 10 μmol/L in samples

TABLE 2. Ions and Isotopic Compositions in the Changjiang River Waters, China

sample	river, location	Cl^{a} (μmol/L)	NO_3^{a} (μmol/L)	$\delta^{18}O_{H2O}$ ‰	δD ‰	$\delta^{15}N_{NO3}$ ‰	$\delta^{18}O_{NO3}$ ‰
M1	Jinshajiang, Panzhihua	1134.9	6.2	−13.8	−110.8	nd	nd
T1	Yalongjiang, Panzhihua	30.5	5.3	−14.7	−111.4	nd	nd
M2	Jinshajiang, Panzhihua	557.0	4.0	−14.4	−107.2	nd	nd
M3	Jinshajiang, Yibin	440.2	23.8	−13.0	−104.0	12.4	10.8
T2	Minjiang, Yibin	146.0	88.5	−9.9	−81.5	9.7	9.2
M4	Jinshajiang, Yibin	307.3	52.3	−11.8	−90.8	12.5	9.9
M5	Jinshajiang, Luzhou	347.9	49.9	−11.9	−88.8	12.1	9.9
M6	Jinshajiang, Luzhou	376.8	75.8	−11.4	−84.4	12.4	9.1
T3	Jialingjiang,Chongqing	223.2	63.7	−8.8	−66.1	10.9	7.5
M7	Changjiang, Chongqing	299.3	42.9	−12.2	−87.8	12.1	10.4
T4	Jialingjiang, Chongqing	237.9	64.7	−8.9	−66.7	11.4	11.2
M8	Changjiang, Chongqing	314.6	83.0	−11.1	−80.3	10.4	10.1
M9	Changjiang, Chongqing	331.6	71.3	−11.6	nd	12.3	11.2
M10	Changjiang, Chongqing	325.1	51.7	−11.6	−86.2	9.5	9.9
T5	Wujiang, Fuling	157.7	94.7	−7.1	−50.4	11.1	7.8
M11	Changjiang, Fuling	329.5	53.6	−11.9	−88.4	12.9	10.9
M12	Changjiang, Wanzhou	378.2	41.8	−12.2	−90.5	10.9	6.1
M13	Three Gorges Dam Reservoir	312.6	76.6	nd	nd	8.9	4.7
M14	Three Gorges Dam Reservoir	380.3	50.7	−12.0	nd	9.1	6.8
M15	Changjiang, Yichang	425.3	63.2	−11.4	−82.5	9.6	4.4
T6	Yuanjiang, Taoyuan	89.0	41.9	−6.5	−45.2	11.2	7.1
T7	Zishui, Yiyang	115.7	56.0	−6.4	−47.7	9.9	7.4
T8	Xiangjiang, Changsha	183.2	80.5	−6.6	−55.1	9.0	6.3
T9	Dongting Lake	182.0	59.8	−6.4	−47.3	7.7	3.8
M16	Changjiang, Yueyang	265.8	60.1	−7.5	−58.4	7.9	3.5
M17	Changjiang, Wuhan	272.0	73.7	−9.2	−68.9	7.3	3.6
T10	Hanjiang, Wuhan	229.0	45.2	−7.7	−55.0	10.0	3.6
T11	Hanjiang, Wuhan	216.2	40.9	−7.7	−57.6	9.5	4.0
M18	Changjiang, Wuhan	257.0	58.7	−8.5	−62.5	9.8	2.7
M19	Changjiang, Wuhan	283.1	74.6	−9.3	−70.5	8.4	4.2
T12	Ganjiang, Nanchang	216.7	56.1	−6.2	−48.6	9.9	7.6
T13	Poyang Lake	182.0	38.8	−6.0	−45.6	12.3	8.3
M20	Changjiang, Pengze	258.4	66.6	−8.3	−60.0	8.3	2.4
M21	Changjiang, Datong	277.3	65.3	−8.3	−61.3	9.6	3.2
M22	Changjiang, Nanjing	286.6	63.9	−8.2	−62.5	9.8	3.4
M23	Changjiang, Zhangjiagang	302.2	66.7	−8.3	−64.4	8.3	2.6
YBWS	Waste water, Yibin	1124.7	1477.7	−10.1	−77.2	8.9	5.6
WHWS	Contaminated water, Wuhan	603.8	43.0	nd	nd	9.3	3.0

[a] Data are from Chetelat et al. (*21*). nd: not determined.

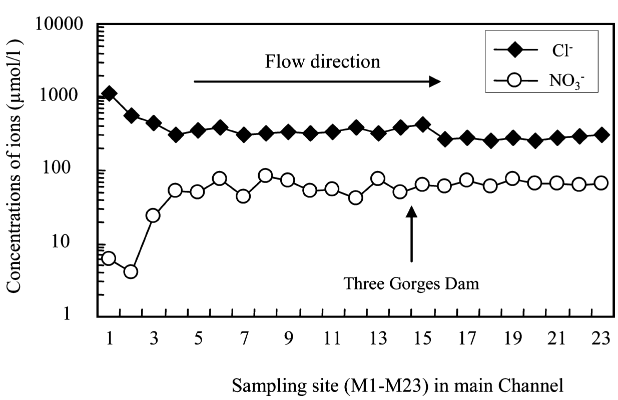

FIGURE 2. Variations of NO_3^- and Cl^- in the Changjiang main channel.

collected upstream (M1−2), which should have been due to the limited human activities. The NO_3^- concentration increased quickly from M1 to M4 (Figure 2) when the Changjiang River flowed into Sichuan basin, which is a major agricultural field of Southwest China. There were somewhat higher levels of nitrate (>60 μmol/L) in the major tributaries, including the Minjiang, Jialingjiang, Wujiang, and Xiangjiang, which indicates that these tributaries contribute a large amount of nitrate to the main channel of the Changjiang River.

Chloride is a good indicator of sewage impacts and dilution because it is not subject to physical, chemical, and biological processes (*25*). High Cl^- contents in first station mainly derived from evaporite dissolution (*21*). The changes in Cl^- levels from upstream to downstream suggest that dilution might play a major role in the variation of Cl^-. The NO_3^- contents obviously increased from upstream to downstream, which was opposite to the Cl^- contents of waters in the main channel of the Changjiang (Figure 2). Nitrogen from groundwater/soil−water discharge to the river and its tributaries would result in greater nitrate concentrations and lower chloride concentrations, which might be responsible for the opposing trends of NO_3^- and Cl^- from upstream to downstream.

The hydrological gauging station of Datong in the lower reaches of Changjiang is located at the upstream limit of the tidally influenced river segment. Nutrient fluxes from the Changjiang River to the East Sea can be estimated based on the concentrations of nutrients and discharge of water at Datong Station. Liu et al. (*18*) demonstrated that the NO_3^- level was very low upstream and remained fairly stable at 70−98 μmol/L in April−May, 1997. The NO_3^- concentrations ranged from 70.7 to 97.1 μmol/L from 1998 to 1999 at Datong Station (*2*). In the present study, the nitrate content measured at Datong Station was 66.7 μmol/L, which is similar to the data collected at the end of the last century.

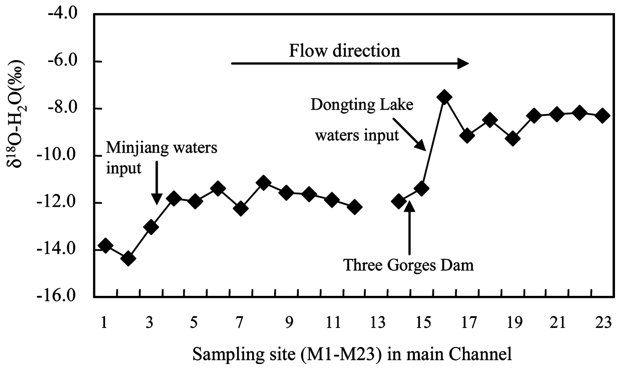

FIGURE 3. Variations of oxygen isotopic compositions of waters in the Changjiang main channel.

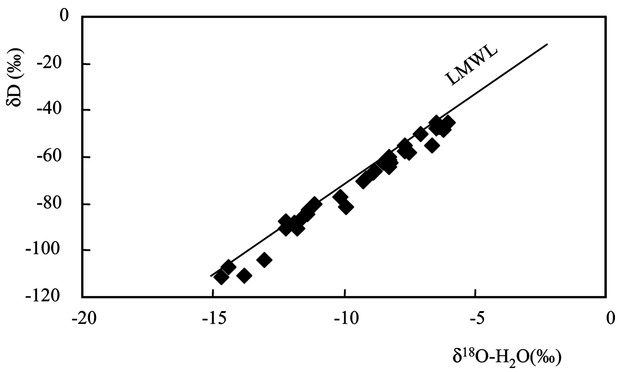

FIGURE 4. Isotopic compositions (δ^{18}O, δD) of waters in Changjiang River basin together with the Local Meteoric Water Line (IAEA/WMO, 2006).

The δ^{18}O values of water ranged from −14.7‰ to −6.0‰ throughout the basin and increased from upstream to downstream in the Changjiang main channel (Figure 3). There were two abrupt increases between M2 and M4, M15 and M16. The former was due to waters from other small tributaries and Minjiang river of Southwest China. The latter was due to waters from Dongting Lake (T9) with a big discharge. The δ^{18}O values of water in the main channel of the Changjiang below Yichang were similar to the values of water from the tributaries in the middle and lower reaches, which suggests that they were the major sources of water to the Changjiang main channel below Yichang during the sampling period. As indicated by Figure 4, the compositions of δD and δ^{18}O generally fall on the local meteoric water line (LMWL) calculated using data from the Global Network of Isotopes in Precipitation (GNIP) from stations in China (*26*), which indicates that modern precipitation was the major water source for the Changjiang River and there was no strong evaporation taking place along the river during sampling period.

δ^{15}N and δ^{18}O of nitrate in Changjiang River Water. The N and O isotopic composition of nitrate in samples collected from the Changjiang River are listed in Table 2. The δ^{15}N of NO_3^- ranged from 7.3‰ to 12.9‰ in the Changjiang River with a mean value of 10.2‰. There was a narrow range of δ^{15}N-NO_3^- in the water of the tributaries, with most samples having δ^{15}N-NO_3^- values between 9‰ and 12‰. The NO_3^- in river water may be derived from rain, chemical fertilizer, nitrification of soil organic nitrogen, and sewage effluents. It has been reported that nitrogen species from rain have values lower than +0‰ in the Sichuan Basin of the Changjiang Catchment (*27*). Chemical fertilizers from Jiangshu, Guiyang, and Sichuan provinces in the Changjiang Basin have been shown to have δ^{15}N values of −1.0 ± 1.4‰, 0 ± 1.4‰, and +1.0 ± 2.6‰, respectively (*25, 27, 28*), which is in agreement with the data reported in the literature (*6, 29*). The δ^{15}N values of particulate organic nitrogen (PON) in the Changjiang have been found to vary from 2.8‰ to 6.0‰ (*30*). The δ^{15}N values of nitrate derived from livestock waste and sewage generally range from +8‰ to +25‰ (*6, 31*). The nitrogen isotopes of nitrate might be affected by ammonia volatilization and biological assimilation. In the present study, the N isotopic compositions of nitrate in urban wastewater (YBWS, WHWS) were 8.9‰ and 9.3‰, respectively, which is in good agreement with the range of δ^{15}N-NO_3^- values reported in these studies (*6, 29*).

The δ^{18}O of NO_3^- was found to range from 2.4‰ to 11.2‰ in the Changjiang River with a mean value of 6.8‰. The O isotopic compositions of nitrate in urban wastewater (YBWS, WHWS) were 5.6‰ and 3.0‰, respectively. The pattern of δ^{18}O of NO_3^- is more useful than that of δ^{15}N for identifying nitrate from wet deposition and microbial nitrification as well as chemical fertilizer because there is a large variability in the δ^{18}O-NO_3^- values among these sources (*5, 6*). The δ^{18}O-NO_3^- values of precipitation vary greatly from +25‰ to +75‰ due to complex atmospheric processes. The isotopic composition of oxygen in chemical NO_3^- fertilizers is +23.5‰, which is similar to that of atmospheric oxygen (*7*). There are several factors that lead to shifting of the δ^{18}O-NO_3^- values by nitrification, such as water evaporation, diffusion of oxygen in the soil, discriminating ratio of oxygen from the air and water in different environments, and rapid microbial mineralization-immobilization turnover processes in the soil zone (*6, 32, 33*). δ^{18}O-NO_3^- values would have a wide range for nitrate derived from nitrification. Based on the pattern of δ^{18}O-NO_3^- observed in this study, nitrate derived from nitrification of nitrogen-containing organic materials and sewage effluent should be the major source of nitrate in the Changjiang River. In theory, the δ^{18}O values of nitrate produced by microbial nitrification could be calculated because approximately one-third of the oxygen in NO_3^- should be derived from oxygen in the air and have a value of +23.5‰, while two-thirds should be derived from water oxygen (*34*). Therefore, nitrate derived from microbial nitrification should have δ^{18}O values between 0‰ and +6‰ in the Changjiang River. In this study, nitrate measured in the Changjiang River has δ^{18}O values somewhat higher than would be expected for nitrification alone.

Nitrate Sources and Impacting Factors As Determined by a Dual Isotopic Approach. There are these sources contributing to the NO_3^- budget in the Changjiang River: precipitation, chemical fertilizer, nitrification of nitrogen-containing organic materials, and urban sewage effluent. The contribution of nitrate from precipitation is probably negligible for the Changjiang River waters due to the low δ^{15}N-NO_3^- values and high δ^{18}O-NO_3^- values (*5, 6*). In China, urea and ammonium salt are the majority of synthetic nitrogen fertilizer. KNO_3 usually is used for vegetables and fruit tree in some places. About 7.3 Tg N/a from synthetic fertilizers were input in Changjiang Basin (*20*). However, chemical nitrate fertilizer was likely not directly input into the rivers at the time of sampling because intensive fertilizing activities are not conducted in August and it has low δ^{15}N-NO_3^- values and high δ^{18}O-NO_3^- values. The isotopic composition of nitrate from chemical fertilizer would be modified by a biological process during the flow of water into rivers from the soil and unsaturated zone (*33*). In this study, the NO_3^- contents in the urban wastewater samples (YBWS, WHWS) were 1477.7 and 43 μmol/L, respectively. The contents of ammonium in the YBWS and WHWS were 0.4 and 3.3 mg N/L. It has been reported that there were 2.08 × 10^{10} tons of industrial wastewater and 9.75 × 10^9 tons of domestic sewage input into Changjiang River. About 80% of these wastewaters were input in the middle and lower reaches of the Changjiang Basin in 2006 (*35*). The wastewater with high nitrogen load would be one important source of nitrate

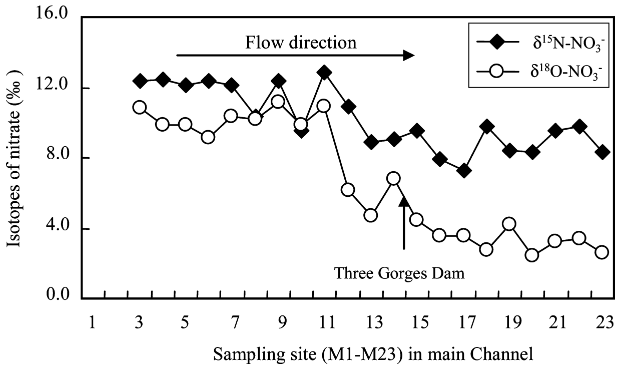

FIGURE 5. Variations of isotopic compositions for nitrate in the Changjiang main channel.

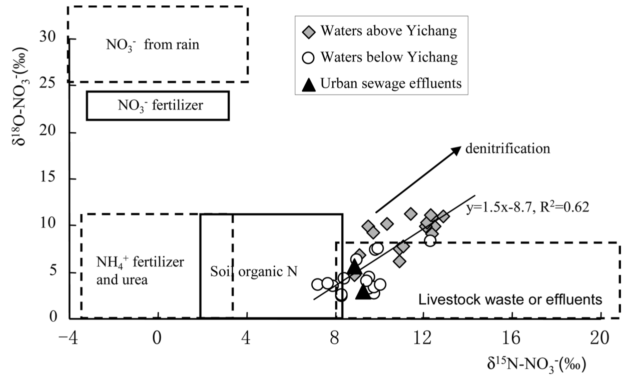

FIGURE 6. Relationship between δ^{15}N and δ^{18}O of nitrate in the Changjiang River waters. The isotopic composition of various sources in the diagram (modified after refs *6, 8*, and *34*).

for the Changjiang River waters. Thus, nitrification of nitrogen-containing organic matter (including "modified fertilizer") and urban sewage effluent are likely the major sources of nitrate in the Changjiang River.

It could be observed that the δ^{15}N and δ^{18}O of NO_3^- levels decreased from upstream to downstream in the main channel of the Changjiang River (Figure 5). There is a distinct decreasing turnover point observed from M11 to M13, which might indicate that different sources or impacting factors are responsible for the shifting isotopic composition of nitrate. These waters are located in the Three Gorges reservoir area. The Three Gorges Dam was built by 2002. Algae/hydrophyte would easily grow in this large reservoir and produce a "new" organic matter. However, algae/biology generally prefers to uptake light isotopes of nitrate, which would lead to enrichment with heavy isotopes in residual nitrate (*14, 17*). Therefore, assimilation would not the major reason for the decreasing isotopic values of nitrate in the main channel of the Changjiang. The Three Gorges Reservoir submerged a big area, which would lead a large amount of "old" organic matter buried. The "new and old" labile organic matter would produce new nitrate due to remineralization, which might have modified the isotopes of nitrate in waters.

In 2006, drought occurred in most parts of China. Severe drought hit Chongqing and eastern part of Sichuan and the drought was regarded as moderate or relatively severe compared to the normal year, especially in August 2006 (*36*). Specifically, it was reported that the rainfall in 2006 was approximately 10−30% lower than during a normal year in most parts of the upper reach of the Changjiang and Hanjiang basins. Indeed, there were only 796 billion cubic meters of total water resources in the Changjiang River in 2006, which is 19% less than are present in a normal year (*35*). In August 2006, the mean discharges at Yichang, Hankou, and Datong stations on the Changjiang River were 9590, 19,900, and 27,600 m^3/s, respectively, which was lower than the historical monthly averages of 27,700, 39,800, and 45,200 m^3/s (http://www.hydroinfo.gov.cn/), respectively. In drought condition, water evaporation would lead to high values of δ^{18}O-NO_3^- during nitrification. Additionally, the sewage effluents would play more important role in contributing nitrate sources in rivers during the period with lower water discharge than normal. This is consistent with the heavier δ^{15}N and lighter δ^{18}O values of the nitrate observed in the present study, which suggested that urban sewage effluent was a significant source of nitrate for the Changjiang River.

No negative relationship between NO_3^- and δ^{15}N/ δ^{18}O-NO_3^- was observed (not shown). However, a strong positive relationship was found between δ^{15}N and δ^{18}O-NO_3^- (Figure 6), which indicates that denitrification may be responsible for the shifting δ^{15}N and δ^{18}O-NO_3^- values. The slope of the linear regression was 1.5, which is within the range of previously reported values 1.3−2.1 (*8, 37, 38*). Denitrification can reduce the nitrate load in water via transformation of dissolved nitrate to N_2 and N_2O as follows:

$$NO_3^- \rightarrow NO_2^- \rightarrow N_2O \rightarrow N_2$$

Denitrification might occur in the soil and unsaturated zone when organic carbon or FeS_2 take part in the reaction (*6, 8*). The riparian zone is an important area for denitrification to control the distribution of nitrate (*37*). There existed a large number of littoral zones in Changjiang basin due to the developed lake system and construction of the dam. These wet lands might be an important area for denitrification. Additionally, Pardo et al. (*11*) thought hydrological storage played an important role in control of the nitrate exported to streams. It is reported that the discharge of groundwater containing highly denitrified NO_3^- to the river and in-stream denitrification should be the important reason for heavier isotopic values of nitrate in the lower Illinois River (*15*). The interchange between the river waters and the hyporheic zone or lateral exchange with surficial groundwater at the shores might probably be strongly reduced, which can be expected to have significant influence on denitrification. In drought condition, groundwater and soil−water would be major water resources for the upper reach of Changjiang River. So in this study, the nitrate from groundwater storage pool and soil solution with denitrified NO_3^- might be another major reason for nitrate with high δ^{15}N and δ^{18}O values in Changjiang River waters, especially for the upper reach samples.

There were multiple sources of nitrate and impacting factors affecting the nitrate behavior in rivers in different seasons (*9, 16, 17*). It was difficult to understand the sources contribution and extent of nitrate transformation well and truly for the Changjiang River on the basis of only one sampling program. But this study could show that the nitrification of organic matter and sewage effluents mainly contributed nitrate to Changjiang River, and denitrification shifted the isotopes of nitrate in the special sampling period.

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