



Optimized digestion methods: organic phosphorus sequential extraction, total phosphorus, and nitrogen simultaneous determination in sediments

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Received: 4 October 2017 / Accepted: 16 February 2018 / Published online: 27 February 2018
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Abstract

Purpose The organic phosphorus (OP) that is found in sediments influences the water quality of the overlying water. Accurate methods for OP extraction are important to identify phosphorus fractions in the sediments. We optimized the following: (1) the pH range of solutions after digestion; (2) the amounts of TP, H₂SO₄, and K₂S₂O₈ in the digestion; and (3) the digestion efficiency between acid K₂S₂O₈ and alkaline K₂S₂O₈ in total phosphorus (TP) and total nitrogen (TN) measurements. Moreover, we explained the related experimental phenomena and the results with an in-depth mechanism.

Materials and methods Sediments samples were taken from three locations (Yellow Sea, Jiehe River, and Jialai River, China). Each sample was analyzed to optimize sequential OP extraction based on the Ivanoff method.

Results and discussion We focused on the digestion step and digestion efficiency between acidic K₂S₂O₈ and alkaline K₂S₂O₈, total phosphorus (TP), and total nitrogen (TN) measurements. The results indicate that the optimal pH range of solutions after digestion is from 3 to 5.5. The TP digestion efficiency in combination with H₂SO₄ + K₂S₂O₈ (acid) is, on average, 8% higher than that of NaOH + K₂S₂O₈ (alkaline). However, the results for TN suggest the opposite. Optimal amounts of TP, H₂SO₄, and K₂S₂O₈ for digestion were also clearly determined.

Conclusions The pH of solutions after digestion affects the color development necessary for effective phosphorus (P) determination. The TP and TN could be determined simultaneously using the alkaline K₂S₂O₈ digestion system under the experimental conditions. Finally, the optimal orthogonal combinations and mass ratios of P, K₂S₂O₈, and H₂SO₄, respectively, were determined to be $\sim 1.5 \times 10^{-5}:1:8$ (NaHCO₃, 16 h), $\sim 5.6 \times 10^{-5}:1:3$ (HCl, 3 h), $\sim 1.6 \times 10^{-5}:1:5$ (NaOH, 16 h), and $\sim 1.4 \times 10^{-5}:1:8$ (NaOH, 16 h, pH = 0.2). We improved the accuracy and precision of the Ivanoff method using an optimized sequential extraction method.

Keywords Organic phosphorus · Sequential extraction method · Simultaneous determination · Total phosphorus · Total nitrogen

Responsible editor: Shiming Ding

Electronic supplementary material The online version of this article (<https://doi.org/10.1007/s11368-018-1959-6>) contains supplementary material, which is available to authorized users.

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1 Introduction

Phosphorus (P) is an important and limiting nutrient for primary production in aquatic environments (Kaiserli et al. 2002; Ahlgren et al. 2005; Fowdar et al. 2017). However, excessive P from industrial and agricultural emissions has already caused some serious environmental pollution problems (Smi 2000). Phosphorus pollution can trigger eutrophication, biodiversity loss, and oxygen depletion in water (Chowdhury et al. 2017). According to figures from the US EPA, it costs communities approximately US\$2.2 billion every year to remedy drinking water and recreational water and to protect endangered species that are threatened by algal blooms (Dodds et al. 2009).

Sediments that adsorb large amounts of P are thought to be an important source of P for overlying water (Monbet et al. 2007; Hu et al. 2011). The P in sediments can be divided into inorganic P (IP) and organic P (OP), of which OP accounts for 12–42% of the total P (TP) (Ding et al. 2010). Studies have shown that considerable quantities of released P are from OP found in sediments (Suzumura and Kamatani 1995; Slomp et al. 2002). Under certain conditions, the decomposition of OP in sediments becomes an important source of soluble reactive P (SRP) in water (Bostrom et al. 1989). Similar to the sequential extraction of IP (Hieltjes and Lijklema 1980), OP can also be divided into many fractions (Zhu et al. 2013). However, the fractions and distributions of OP in sediments are poorly understood because there are currently no effective fractionation and characterization approaches (Zhang et al. 2008). Bowman and Cole (1978) developed a chemical fractionation method for OP in soil, and they divided OP into four distinct fractions using a sequential extraction method: labile OP, moderately labile OP, moderately resistant OP, and highly resistant OP. However, there are some problems associated with this method, such as the incomplete removal of high concentrations of phytate in most soils and sediments (Oluyedun et al. 1991; Golterman et al. 1998; Turner et al. 2002). Their fractionation method has been further improved by Tiessen et al. (1983), Sharpley and Smith (1985), and Ivanoff et al. (1998). For example, Ivanoff et al. (1998) developed a sequential extraction method for OP in soils. The OP recovery was improved by using an additional step to determine residual OP and biomass OP. Currently, there are no unanimously accepted fractionation procedures for analyzing OP in sediments (Li et al. 2013), and the sequential extraction procedure is still being improved and modified to estimate the fractionation of P in soil or sediment (Waterlot 2018). Although it is a classical method, there are still some problems with the Ivanoff method. For example, the proportions of OP, H_2SO_4 , and persulfate in the digestion are uncertain. In addition, the concentration of P during extraction and digestion are determined without pH adjustments. Furthermore, a pH range for the effective measurement of TP by the ascorbic acid-molybdenum blue method is not obtained (Murphy and Riley 1962; Tiessen et al. 1983). The previous problems could affect the accuracy of OP determination. Therefore, it is essential to optimize the fractionation method of OP in sediments. Additionally, during the determination process, OP maybe converted to IP under the $\text{K}_2\text{S}_2\text{O}_8$ digestion condition. There is a similar scenario for TN measurement where different forms of N change to nitrate nitrogen (NO_3^- -N) when TN is affected by $\text{K}_2\text{S}_2\text{O}_8$. Therefore, the feasibility of determining TN using a TP digestion system is worth studying. There have been studies of the simultaneous determination of TP and TN using either an alkaline or acid peroxydisulfate, but those authors did not give an in-depth description of the mechanisms or phenomena (Worsfold et al. 2005).

The objectives of this study are to (1) investigate and optimize the pH range of the digestion solution for TP measurement by the phosphor-molybdate colorimetric method; (2) identify the digestion efficiency of TP and TN using the two digestion systems $\text{H}_2\text{SO}_4 + \text{K}_2\text{S}_2\text{O}_8$ and $\text{NaOH} + \text{K}_2\text{S}_2\text{O}_8$; (3) investigate the feasibility of determining TP and TN using the same digestion system; and (4) investigate the relationship between the TP digestion efficiency and the concentration of P, as well as dosages of H_2SO_4 and $\text{K}_2\text{S}_2\text{O}_8$. Moreover, the aims of this study are to explain the related experimental phenomena and results with an in-depth mechanism and to improve the accuracy and precision of the classical Ivanoff OP sequential extraction method by using an optimized method.

2 Materials and methods

2.1 Study site and sampling

The Jiehe River and Jiaolai River are located in the northwest of the Shandong Peninsula, and they flow northwest into the Laizhou Bay of the Bohai Sea, China (Fig. 1). Both rivers have been heavily contaminated. Pollution in the Jiehe River is mainly caused by local mining and smelting activities (Bi and Zhao 2017). Pollution in the Jiaolai River is mainly caused by upstream industrial effluent (Cai et al. 2015; Li et al. 2016). Sediment samples were collected using a Van Veen grab sampler (Fig. 1). Composite surface sediment samples were stored in sealed plastic bags in an adiabatic box and then frozen at $-24\text{ }^\circ\text{C}$ until the analysis was performed. The samples were freeze dried and sieved (100 mesh) prior to the analysis.

2.2 Measurements

The TP was measured by calcination in a muffle furnace ($550\text{ }^\circ\text{C}$) and was then extracted using a solution of 1 mol L^{-1} HCl. Total IP (TIP) was measured by extraction with 1 mol L^{-1} HCl (16 h) without calcination. The difference between TP and TIP was total OP (TOP) (Aspila et al. 1976). The three solid-phase fractions of OP determination (readily labile OP, moderately labile OP, and nonlabile OP) in the sediment samples were based on the Ivanoff method (Ivanoff et al. 1998).

1. Readily labile OP: A total of 1.0-g sediment samples that had been sieved through a 100-mesh sieve was initially extracted with 50 mL of 0.5 mol L^{-1} NaHCO_3 at pH 8.5 for 16 h and was then centrifuged at 4000 g for 15 min. After the centrifugation, the extract was collected to determine both IP and TP. The difference between TP and IP was readily labile OP.

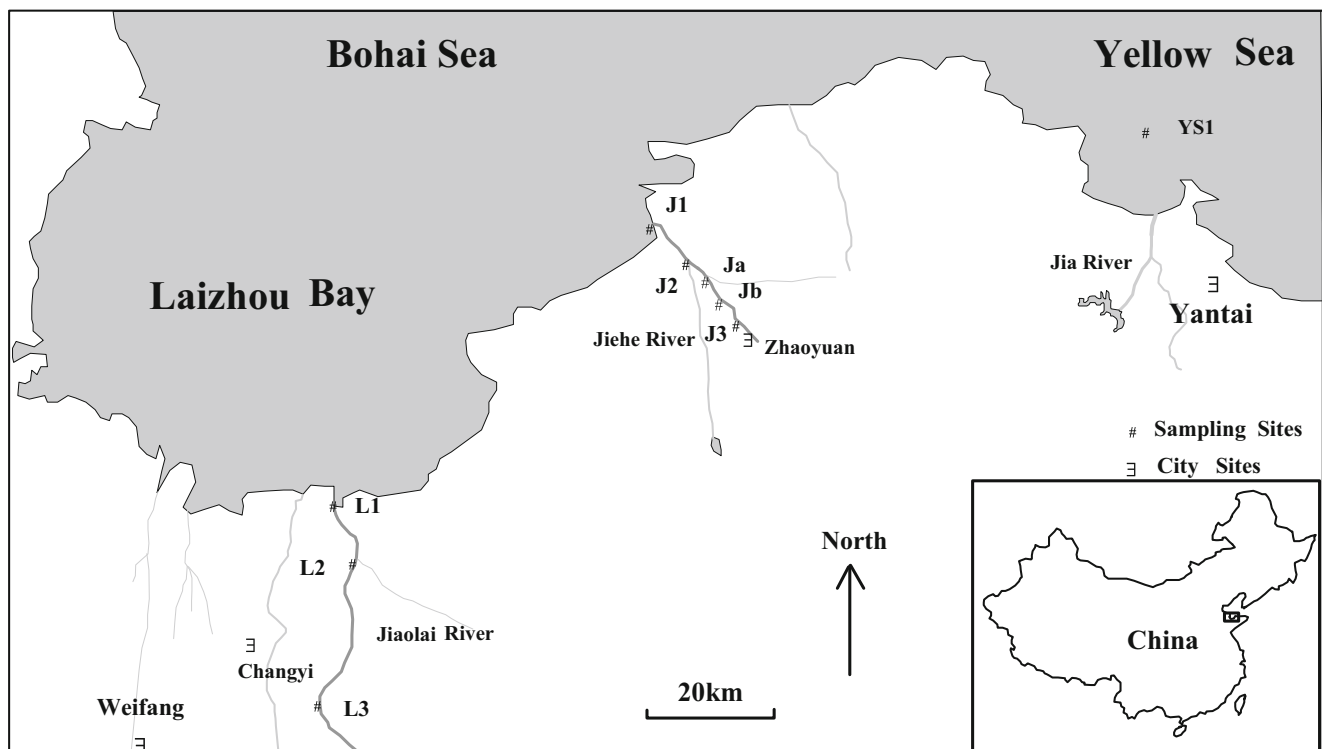


Fig. 1 The location of the sampling sites. One is located in the north Yellow Sea (YS1), three of them are in the Jiaolai River (L1, L2, and L3), and five of them are in the Jiehe River (J1, J2, J3, Ja, and Jb)

- Moderately labile OP: The residue from the NaHCO_3 extraction was extracted with 50 mL of $1.0 \text{ mol L}^{-1} \text{HCl}$ for 3 h. The residue from the HCl extraction was rinsed with deionized water and then extracted using 50 mL of $0.5 \text{ mol L}^{-1} \text{NaOH}$ for 16 h. The NaOH extract was acidified to pH 0.2 with concentrated HCl to separate the nonlabile fraction (humic acid-P) from the moderately labile fraction (fulvic acid-P). All extracts were centrifuged and analyzed for TP and IP. The HCl-OP and fulvic acid-OP were moderately labile OP.
- Nonlabile-OP: The rinsed residue from the NaOH extraction was ashed at $550 \text{ }^\circ\text{C}$ for 1 h and then extracted with $1 \text{ mol L}^{-1} \text{H}_2\text{SO}_4$ for 24 h. After centrifugation, the extract was analyzed for TP. The sum of H_2SO_4 -TP and humic acid-OP was nonlabile-OP.

Total extracted OP is the sum of the three solid-phase fractions of OP described previously. The P concentration in all extracts was determined according to the molybdenum blue method (Murphy and Riley 1962). Total organic carbon (TOC) and TN in the sediment samples were analyzed using a total organic carbon analyzer (TOC-VCPH, Shimadzu, Japan). The TN in all solutions was measured using a spectrophotometric method where all forms of nitrogen were oxidized to NO_3^- -N, and the final concentration of NO_3^- -N was used as TN (Chinese EPA 2002). The concentrations of urea (organic N) in all samples were

measured using a spectrophotometry method that is based on the reaction of diacetylmonoxime and urea in an acidic solution (Zhao et al. 2011). The nitrite determination method was based on the reactions of sulfonamides with nitrite under acidic conditions, followed by coupling with *N*-(1-naphthyl) ethylenediamine dihydrochloride (NED) (Pandurangappa and Venkataramanappa 2007). Sediment grain sizes were measured using a laser particle size analyzer (Malvern Mastersizer 2000F, Malvern, UK). Triplicates were analyzed for all samples, and the results are expressed as their average. The relative standard deviation (RSD) of the measurements is $\leq 5\%$.

2.3 The pH gradient experiments of digestion solutions

The eight parallel solutions containing OP ($P < 30 \text{ } \mu\text{g}$) derived from sediment extraction were added to eight 50-mL colorimetric tubes. The eight parallel solutions were digested for TP determination (Bowman 1989). The parallel digestion solutions were spiked with $2.5 \text{ mol L}^{-1} \text{NaOH}$ or $2.5 \text{ mol L}^{-1} \text{H}_2\text{SO}_4$ to adjust pH to approximately 1, 2, 3, 5, 7, 9, 11, and 13 after digestion. The previous mixtures were diluted by adding Milli-Q water to the 50-mL comparison tubes for P determination. The reported results are the average values of three parallel experiments.

2.4 The TP digestion by systems of H₂SO₄ + K₂S₂O₈ and NaOH+ K₂S₂O₈

To compare the digestion efficiency of TP using H₂SO₄ + K₂S₂O₈ and NaOH+ K₂S₂O₈, 12 parallel solutions containing OP (Sect. 2.3) of accurately measured aliquots ($P < 30 \mu\text{g}$) were added into 12 colorimetric tubes (50 mL). The tubes were divided into three series: (1) tubes A–E were spiked with different amounts of 2.5 mol L⁻¹ NaOH (0.5, 1, 2, 4, and 6 mL) and 3 mL of 5% K₂S₂O₈ (wt%); (2) tube F was not spiked with 5% K₂S₂O₈ (wt%), H₂SO₄, or NaOH addition and tube G was spiked with 3 mL of 5% K₂S₂O₈ (wt%) without the H₂SO₄ or NaOH addition. Group G is the standard method published by the Chinese EPA (2002); (3) tubes H–L were spiked with different amounts of 2.5 mol L⁻¹ H₂SO₄ (0.5, 1, 2, 4, and 6 mL) and 3 mL of 5% K₂S₂O₈ (wt%). Milli-Q water was added to these 12 tubes until they were filled to the 25-mL tick mark for digestion (Halstead et al. 1999). The resulting concentration of TP was used to represent digestion efficiency. After digestion, the pH values of the 12 parallel digestion solutions were adjusted to the effective colorimetric pH range by adding 1 or 6 mol L⁻¹ NaOH and 1 or 6 mol L⁻¹ H₂SO₄, and then the tubes were diluted by Milli-Q water to the 50-mL tick mark for the determination of P.

2.5 TN digestion by H₂SO₄ + K₂S₂O₈ and NaOH+ K₂S₂O₈

Generally, the digestion system of NaOH+ K₂S₂O₈ is used for the determination of TN in water because the digestion efficiency can be promoted by the alkalinity associated with K₂S₂O₈ (Halstead et al. 1999). To investigate whether the digestion system of H₂SO₄+ K₂S₂O₈ can be simultaneously applied to the digestion and determination of TN and TP, an experiment that compared the digestion efficiency of TN was performed.

The standard artificial solution containing 5 mg L⁻¹ organic nitrogen, 5 mg L⁻¹ NH₃-N, and 5 mg L⁻¹ NO_x-N was from analytically pure urea (CO(NH₂)₂), NH₄Cl, and KNO₃. In natural water, urea is a common type of organic nitrogen and a significant part of the nitrogen from agricultural fertilizer and animal metabolism sources. It is also common in the laboratory. Consequently, urea was selected to represent organic nitrogen. Then, the standard N solutions (2 mL) were added into 12 colorimetric tubes (25 mL). The tubes were divided into three series: (1) tubes 1–5 were spiked with 5 mL of different concentrations (0.025, 0.125, 0.25, 0.375, and 0.5 mol L⁻¹) of NaOH solutions containing 4% K₂S₂O₈ (wt%), (2) tube 6 was a blank group that did not contain H₂SO₄ or NaOH but contained 4% K₂S₂O₈ (wt%), tube 7 was spiked with 4% alkaline K₂S₂O₈ (wt%) and was analyzed with a standard oxidation digestion-UV spectrophotometry method for TN determination as published by the Chinese EPA (Zhou et al. 2012), and (3) tubes 8–12 were spiked with 5 mL of different concentrations

(0.025, 0.125, 0.25, 0.375, and 0.5 mol L⁻¹) of H₂SO₄ solutions that contained 4% K₂S₂O₈ (wt%). Milli-Q water was added to the 12 tubes until the 10-mL tick mark for TN digestion (Halstead et al. 1999). Then, 1 mL of HCl (1 + 9) was added and diluted by Milli-Q water to the 25-mL tick mark for TN determination after digestion.

2.6 Orthogonal experiment of TP digestion efficiency

To investigate the optimal conditions of TP digestion (where the TP value was regarded as an index), an orthogonal experiment of three factors at four levels was designed using the SPSS 19.0 software (SPSS, IBM Statistics 19.0 edition, USA) (Table 1).

3 Results and discussion

3.1 Sediment characteristics

The chemical components and fractions of P of the sediment samples are shown in Table 2. The concentrations of TP in the sediments range from 297 to 2797 mg kg⁻¹. The concentration of IP dominates the P fraction, ranging from 271 to 1197 mg kg⁻¹ (60–91.2% of the TP). Additionally, OP content ranges from 26 to 380 mg kg⁻¹, accounting for 8.8–40% of the TP. Concentrations of TOC and TN in sediments range from 0.11 to 2.3 and 0.04 to 0.36 (wt%), respectively.

The Jiaolai River is a freshwater deficient river. Its water volume is mainly dependent on the influx from marine environments. As a consequence, marine silt and sand have been deposited at the sediment-water interface in the river. Therefore, sediments that are close to the estuary are dominated by larger particles in the Jiaolai River than the Jiehe River. However, the Jiehe River is a normal freshwater river that flows into the sea. There is more clay in Jiehe River sediments (Fig. 2). These findings suggest that the rivers have different grain size distributions, dependent on local geology. Additionally, their grain size distributions could influence distribution of pollutants (Jafarabadi et al. 2017). As pollutants accumulate more easily in small particles, a higher content of fine particulate matter in the sediment leads to a higher

Table 1 Design of orthogonal experiment

Levels	A P extracting solution (mL) ^a	B H ₂ SO ₄ (mL)	C K ₂ S ₂ O ₈ (mL)
1	1	1	1
2	2	2	2
3	3	3	3
4	5	5	5

^a The moderately labile OP extracting solution (1 mol L⁻¹ HCl, 3 h) was 1, 2, 3, and 4

Table 2 General chemical characteristics of the studied sediments

Parameters	YS1	L1	L2	L3	J1	J2	J3	Ja	Jb
TP (mg kg ⁻¹)	533	409	297	695	539	715	677	1197	2797
IP (mg kg ⁻¹)	441	361	271	445	429	630	488	1007	2417
OP (mg kg ⁻¹) ^b	92 (17.3)	48 (11.7)	26 (8.8)	250 (40)	110 (20.4)	85 (11.9)	189 (27.9)	190 (15.9)	380 (13.6)
TOC (%)	0.217	0.140	0.110	1.975	0.933	0.289	0.524	0.656	2.302
TN (%)	0.063	0.049	0.044	0.314	0.209	0.073	0.085	0.111	0.363

^b Values in the parentheses are the percentage of TP in sediments (%)

likelihood that more pollutants (such as TP, IP, OP, TN, and TOC) will be in a river (Fig. 2).

In the present study, the characteristics, origins, and pollution degrees of these sediment samples are different, which is a better representation in order to verify the reliability of the optimized OP sequential extraction method.

3.2 The optimal pH range of a solution after digestion for TP determination

The relationship between TP concentrations and solution pH is shown in Fig. 3. Triplicates were analyzed, and the average values are expressed in the results. Figure 3 shows that measured TP concentrations are low (close to zero) when the pH is lower than 2 or higher than 12. Generally, the redox activity of ascorbic acid depends on pH, so molybdenum phosphates could be reduced by ascorbic acid under strongly acidic conditions (Snehalatha et al. 1997; Mowry and Ogren 1999). Additionally, ascorbic acid is more likely to be oxidized under alkaline conditions than acidic conditions (Hou et al. 2017). Therefore, the ascorbic acid has possibly been oxidized because the pH is higher than 12. In addition, TP is still not strictly consistent with the digestion solution pH values (2–12). As shown in Fig. 3, the experiment reveals that the

optimal pH range for TP measurement is 3–5.5 (weak acid conditions). The low concentration of TP at high pH (6.5–12) levels may be due to ascorbic acid being more likely to be oxidized under alkaline conditions.

3.3 The digestion efficiency of H₂SO₄ + K₂S₂O₈ and NaOH+ K₂S₂O₈

Figure 4 shows that the TP digestion efficiency with H₂SO₄ + K₂S₂O₈ is relatively higher than with NaOH+ K₂S₂O₈. For group F, the background concentration of IP is 70.8 mg kg⁻¹ in the sediment. For groups A–C, the TP digestion efficiency is not promoted with amounts of 2.5 mol L⁻¹ NaOH lower than 2 mL (3 < pH < 5) compared to group G. However, the TP digestion efficiency increases as the amount of NaOH increases (groups D and E). This finding is because the thermal decomposition of persulfate was promoted by OH⁻ (H⁺ can be neutralized by OH⁻), which can enhance O₂^{·-} with strong oxidizing properties (Halstead et al. 1999).

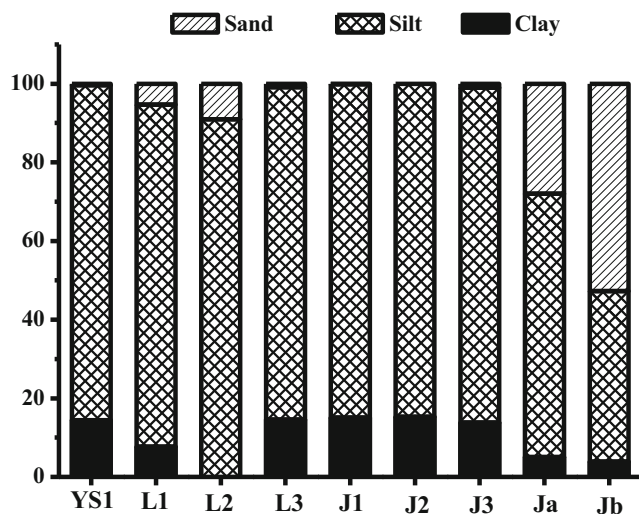
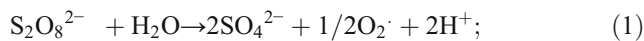


Fig. 2 Grain size distributions of the nine representative sediment samples

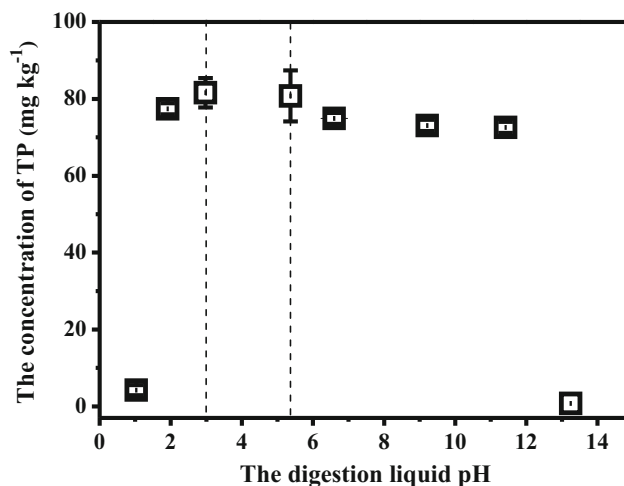


Fig. 3 The influence of different digestion liquids pH on the determination of TP

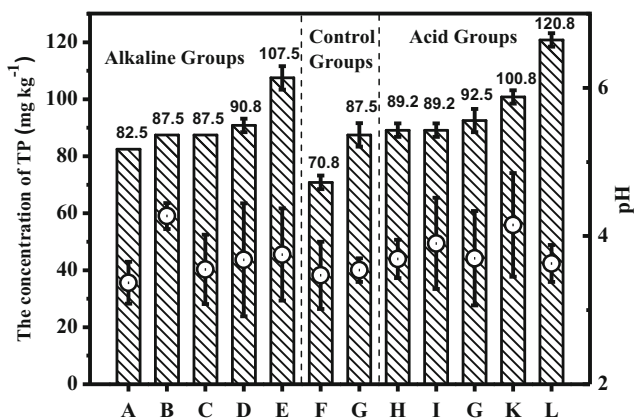
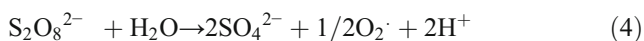
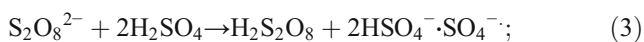


Fig. 4 The TP digestion efficiency was under the alkaline, the acid peroxydisulfate, and the standard method digestive conditions (left). All of the pH in the digestion of each group was adjusted to the optimal interval (3–5.5) prior to effective TP measurement (right)

For groups H–L, the OP digestion efficiency was promoted with 2.5 mol L⁻¹ H₂SO₄, and the efficiency of promotion was higher than the groups with added NaOH. This result was due to the following two reactions (Eqs. (3) and (4)) (Fuller et al. 2016)



Because SO₄⁻ and O₂[·] are strong oxidizing radicals, the OP digestion efficiency of H₂SO₄ + K₂S₂O₈ is higher than that of NaOH+ K₂S₂O₈. Additionally, the pH of the digestion solution in all groups (A–L) was adjusted to the optimal pH range after digestion and prior to effective TP measurement (3 < pH < 5.5) as shown in Fig. 4.

Based on these results, the digestion system of H₂SO₄+ K₂S₂O₈ is more effective than NaOH+ K₂S₂O₈ for determining TP in a sediment extraction. However, both the alkaline and the acid peroxydisulfate can be recommended for TP determination rather than the Chinese EPA standard method (group G).

For TN determination, the digestion efficiency of NaOH+ K₂S₂O₈ is significantly higher than that of H₂SO₄+ K₂S₂O₈ (Fig. 5a). For groups 1–5, the TN digestion efficiency is similar to group 7. This result suggests that the alkaline K₂S₂O₈ digestion method for TN is a stable and reliable. In addition, NH₄⁺ is more easily oxidized with increasing pH, so NH_{3(aq)} is more easily oxidized than NH₄⁺. If there is excessive addition of NaOH, NH₄⁺ could be hydrolyzed into NH₃·H₂O, which could be oxidized to N₂ by S₂O₈²⁻ (Ball et al. 1985) (Eq. (5)). Therefore, TN digestion efficiency began to decrease in group 5.

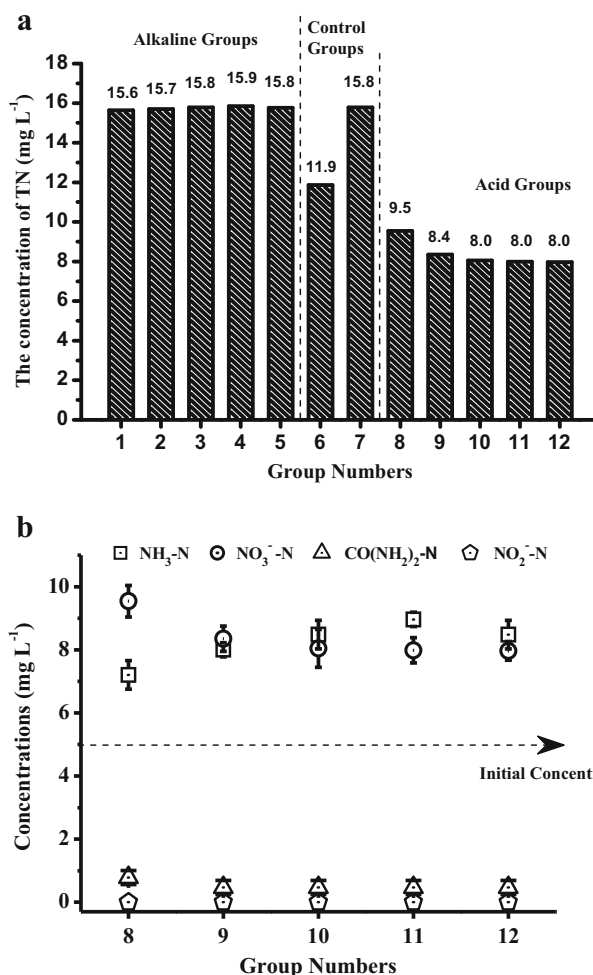


Fig. 5 The TN digestion efficiency was under the alkaline, acid peroxydisulfate, and the standard method digestive conditions (a). The concentrations of different forms of N in the acid group tubes after digestion (b). The dotted arrow represent the initial spiked three forms N (NO₃⁻-N, CO(NH₂)₂-N, and NH₃-N) concentrations in the standard artificial solution before digestion

In groups 8–12, the TN digestion efficiency was suppressed with H₂SO₄, and the efficiency decreased as H₂SO₄ increased. Different forms of N were measured in the five acidic groups (Fig. 5b). The amounts of NH₃-N first increased and then decreased as the amounts of H₂SO₄ increased (from 7.2 to 8.9 and finally to 8.5 mg L⁻¹). However, the concentrations of urea-N (CO(NH₂)₂-N) in the acidic groups decreased as H₂SO₄ increased (from 0.78 to 0.47 mg L⁻¹), and nitrite-N (NO₂⁻-N) was not detected in the acidic groups.

The concentrations of NH₃-N, CO(NH₂)₂-N, and NO₂⁻-N were 5 mg L⁻¹ in the original standard artificial solutions. After the digestion, NH₃-N increased, but CO(NH₂)₂-N decreased to nearly 0 mg L⁻¹ in the acidic groups. Previous studies suggested that it is difficult to oxidize NH₃-N under acidic conditions (Nakamura et al. 2014). The primary decomposition products of urea are NH₃ and HNCO (Eq. (6)). However, there is no

Table 3 Disposition and result of orthogonal experimental design L16 (4^3)

Number	A			B			C		
	TP1 (mg kg ⁻¹)	TP2 (mg kg ⁻¹)	TP3 (mg kg ⁻¹)	TP4 (mg kg ⁻¹)	TP1 (mg kg ⁻¹)	TP2 (mg kg ⁻¹)	TP3 (mg kg ⁻¹)	TP4 (mg kg ⁻¹)	
1	2	3	5	3.63	14.73	3.93	2.43		
2	5 ^c	5	5	8.73	30.43	9.73	5.83		
3	1	2	5	1.93	8.23	2.03	1.53		
4	5 ^c	2	3	8.83	30.23	9.53	5.93		
5	2	2	2	3.63	15.73	4.03	2.73		
6	5 ^c	1	2	8.43	30.23	9.53	6.13		
7	3	3	3	5.53	22.83	6.03	3.83		
8	3	2	1	5.33	23.13	6.03	3.83		
9	1	1	1	1.93	8.13	2.13	1.43		
10	2	5	1	3.93	15.13	4.03	2.83		
11	1	3	2	1.93	8.13	2.43	1.43		
12	3	5	2	5.63	22.33	5.83	3.73		
13	2	1	3	3.53	15.13	4.23	2.83		
14	1	5	3	2.23	8.33	2.33	1.93		
15	5 ^c	3	1	8.83	30.03	10.03	6.33		
16	3	1	5	5.33	23.23	6.33	4.43		

^c The moderately labile OP extracting solution (1 M HCl, 3 h) was 4 mL

consensus as to whether HNCO is produced or not (Alzueta et al. 1998). Therefore, $\text{CO}(\text{NH}_2)_2\text{-N}$ was decomposed to $\text{NH}_3\text{-N}$ and “X-N,” thereby increasing the amount of $\text{NH}_3\text{-N}$ in the solution. Then, the X-N was oxidized to $\text{NO}_3^-\text{-N}$, increasing the content of $\text{NO}_3^-\text{-N}$. The original $\text{NH}_3\text{-N}$ in the acid tubes was not oxidized during the digestion. Finally, all forms of N added up to $\sim 15 \text{ mg L}^{-1}$ in each tube after digestion, observing the N conservation law. Hence, the mechanisms of digestion for $\text{NH}_3\text{-N}$, $\text{CO}(\text{NH}_2)_2\text{-N}$, and $\text{NO}_3^-\text{-N}$ are most likely the following reactions (Eqs. (7) and (8)):



Based on these results, the digestion system of $\text{H}_2\text{SO}_4 + \text{K}_2\text{S}_2\text{O}_8$ should not be selected for TN measurement. The alkaline $\text{K}_2\text{S}_2\text{O}_8$ method can be selected over the Chinese

EPA standard method (group G for TP and group 7 for TN) for simultaneous determination of TP and TN, under certain conditions (Worsfold et al. 2005). However, understanding what types of oxidation agents or conditions could convert $\text{NH}_3\text{-N}$ to $\text{NO}_3^-\text{-N}$ directly (no N_2 production) in strong acid conditions and understanding whether there are other digestion agents that could simultaneously oxidize all forms of P to IP (for TP determination) and all forms of N to $\text{NO}_3^-\text{-N}$ (for TN measurement) remains to be discovered.

3.4 The orthogonal experiment of OP digestion efficiency

Based on the results, the orthogonal experiment L16 (4^3) of OP was designed to investigate the impact of the amount of OP, H_2SO_4 , and $\text{K}_2\text{S}_2\text{O}_8$ on the OP digestion efficiency (Table 3). The values of TP1, TP2, TP3, and TP4 represent the P concentrations in four kinds of OP extracting solutions

Table 4 The result of orthogonal polar difference analysis

Number	TP1 (mg kg ⁻¹)			TP2 (mg kg ⁻¹)			TP3 (mg kg ⁻¹)			TP4 (mg kg ⁻¹)		
	A	B	C	A	B	C	A	B	C	A	B	C
K1	2.01	1.79	1.68	8.21	7.75	7.73	2.23	2.07	2.04	1.75	1.53	1.49
K2	1.84	1.64	1.83	7.59	7.84	7.75	2.03	1.99	2.07	1.49	1.48	1.45
K3	1.64	1.84	1.90	7.63	7.65	7.77	2.02	2.10	2.09	1.46	1.43	1.60
K4	1.74	1.95	1.82	7.56	7.74	7.74	1.94	2.06	2.01	1.34	1.59	1.49
R	0.37	0.31	0.22	0.65	0.19	0.04	0.29	0.11	0.08	0.41	0.16	0.15
Order	A > B > C			A > B > C			A > B > C			A > B > C		
Results	A1B4C3			A1B2C3			A1B3C3			A1B4C3		

Table 5 Concentrations and the recovery of different OP fractions in the sediments

Sediments	Labile OP	Moderately labile OP		Nonlabile OP		Total extracted OP (mg kg ⁻¹)	Total OP (mg kg ⁻¹)	Recovery (%)
	NaHCO ₃ -OP (mg kg ⁻¹)	HCl-OP (mg kg ⁻¹)	Fulvic acid-OP (mg kg ⁻¹)	Humic acid-OP (mg kg ⁻¹)	Residual-OP (mg kg ⁻¹)			
YS1	12.5	5	25.5	0.7	42.5	86.2	92	93.7
L1	15	5	8.7	2.5	17.5	48.7	48	101.5
L2	2.5	2.5	2.75	0	17.5	25.25	26	97.1
L3	35.8	40	55	46.5	74.2	251.5	250	100.6
J1	12.5	17.5	24.1	14.3	32.5	100.9	110	91.7
J2	12.5	5	22.2	9.9	37.5	87.1	85	102.5
J3	42.5	20	44.3	50.1	27.5	184.4	189	97.6
Ja	17.5	15	25	40	94.5	192	190	101.1
Jb	50	55	97.5	112.5	83.7	398.7	380	102.2

(0.5 mol L⁻¹ NaHCO₃, 16 h; 1 mol L⁻¹ HCl, 3 h; 0.5 mol L⁻¹ NaOH, 16 h; HCl added into 0.5 mol L⁻¹ NaOH extraction to adjust, pH = 0.2) after digestion.

According to the results of the orthogonal polar difference analysis, the order of influence of the factors is A > B > C (Table 4). Therefore, the amount of P is the major factor that influences the digestion efficiency, followed by H₂SO₄ and K₂S₂O₈. The optimal combinations of the extracting solution for P by NaHCO₃, HCl, NaOH, and NaOH (pH = 0.2) are A1B4C3, A1B2C3, A1B3C3, and A1B4C3, respectively. After calculating the optimal combinations, the mass ratios of P, K₂S₂O₈, and H₂SO₄, respectively, are ~ 1.5 × 10⁻⁵:1:8 (NaHCO₃, 16 h), ~ 5.6 × 10⁻⁵:1:3 (HCl, 3 h), ~ 1.6 × 10⁻⁵:1:5 (NaOH, 16 h), and ~ 1.4 × 10⁻⁵:1:8 (NaOH, 16 h, pH = 0.2). The digestion solution pH values were adjusted to the optimal pH range prior to analysis (pH 3–5.5).

Nine types of sediments from different sampling areas (Fig. 1) were analyzed to determine the OP concentrations under the established optimal conditions. The recovery (%) of OP was measured as the ratio of total extracted OP to TOP (Zhang et al. 2008). Table 5 shows that the recoveries were 92–103%. There is high recovery and reproducibility when using the optimized method.

4 Conclusions

A method of digesting OP in sediments using a sequential extraction based on the Ivanoff method was optimized in this study. The pH of solutions after digestion affects the color development necessary for effective P determination. The optimal pH for digestion ranges from 3 to 5.5. Additionally, the OP digestion efficiency using H₂SO₄ + K₂S₂O₈ is, on average, 8% higher than that of NaOH + K₂S₂O₈. The alkaline K₂S₂O₈ digestion method can be selected for simultaneous determination of TP and TN, under certain conditions. Finally, the optimal orthogonal

combinations and the mass ratios of P, K₂S₂O₈, and H₂SO₄, respectively, are ~ 1.5 × 10⁻⁵:1:8 (NaHCO₃, 16 h), ~ 5.6 × 10⁻⁵:1:3 (HCl, 3 h), ~ 1.6 × 10⁻⁵:1:5 (NaOH, 16 h), and ~ 1.4 × 10⁻⁵:1:8 (NaOH, 16 h, pH = 0.2). The accuracy and precision of the optimized Ivanoff method are improved in the present study.

Funding information This study was supported by the National Natural Science Foundation of China (Grant No. 41373100). Additional support was provided by the Science and Technology Program for Public Wellbeing of Shandong Province (Grant No. 2013kjhm060308) and the CAS Key Technology Talent Program.

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