Quantification of individual phosphorus forms in surface sediments of the Southern Caspian Sea - Iranian Coast: A sequential extraction procedure

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Received: December 2014 Accepted:Febrary 2016

Abstract

Seventy Two sediments from four stations (Anzali, Tonekabon, Noshahr and Amirabad), located in Caspian Sea-Iranian coast were examined on the basis of P-fractionation in autumn and winter of 2013-2014. Several forms of the phosphorus in sediment were separated and extracted according to sequential extraction method. Then the extracted phosphorus in each fraction was determined by UV–Vis spectrophotometry. In addition, Total concentrations of Ca, Fe and Al in sediment fractions were also measured by Atomic absorption spectroscopy. In autumn the rank order of P-fractions was: Ca-P>Residual-P>BD-P>Al-P>loosely-P and in winter it was: Ca-P>BD-P>Residual-P>Al-P>loosely adsorbed-P. In both season the loosely adsorbed phosphorus (NH4Cl–P) represented < 1% of the sedimentary inorganic phosphorus, while the reductant phosphorus (BD–P) ranged from 2.7 to 4%. The calcium bound phosphorus (Ca-P or HCl–P) showed considerable contribution (89-91%) to the sedimentary inorganic P-loads. The metal oxide bound phosphorus (Al-P or NaOH–P) was 1.2-1.5% and Residual-P was 2.3-4.5%. Concentration of BD- P increased with increasing depth in all stations and the concentration of other fractions increased in most stations. Sampling seasons had significant effect on variance of most P-fractions and other sediment features. TP and BD-P were positively correlated with Fe_{tot} (*p*<0.05).

Keywords: Phosphorus fractionation, Surface sediment, Caspian Sea, Iranian Coast

[DOR: 20.1001.1.15622916.2016.15.2.6.2]

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Introduction

Sediments play a fundamental role in determining concentration, distribution and final fate of several pollutants acting as a principal transport vehicle and the site of accumulation or release (Søndergaard *et al*., 1996; Kleeberg *et al*., 1997). Sediments can act as a sink and a possible source of metals for the environment. Some heavy metals are regarded as serious pollutants of an aquatic ecosystem because of their environmental persistence, toxicity and ability to be incorporated into food chains (Förstner and Wittman, 1983). Phosphorus has been recognized as the most critical nutrient limiting productivity in most lakes. The association of phosphate with iron, aluminum and calcium, and the adsorptive properties of carbonates and clays are of special interest (Jensen *et al*., 1992). The aim of this study was to investigate the different P-forms present in the sediments of four stations, located in southern Caspian Sea, Iran. For this purpose, a sequential extraction (Psenner *et al*., 1984) scheme was employed providing five P-fractions: the loosely adsorbed P, the iron bound P, the aluminum bound P and the calcium bound P and residual P. Phosphorus fractions were evaluated in relation to the concentrations of the most important P-binding elements Ca, Fe and Al, as well as to other sediment features.

Materials and methods

Site description

The Caspian Sea is the largest inland water body in the world, with a surface area of about 380,000 km^2 (the northern area 25%, middle 36% and southern area 39%) and volume of approximately 78,000 km^3. About 130 rivers of various sizes drain into the sea with an annual freshwater inflow of about 300 km^3 (Dumont, 1998). Sampling stations are illustrated in Table 1 and Fig. 1.

Sediment samples and procedure

The top 5 cm of the surface sediment samples from 3 depths of 5, 10 and 20 m were collected from four different sites of Caspian Sea in autumn and winter 2013-2014. Samples were collected with a stainless steel grab sampler, placed in nylon bags, closed tightly and then kept in a refrigerator (4ºC) during shipping. Samples were stored at -20º C until further treatment. In order to characterize various P-species in the sediment, a sequential extraction procedure proposed by Psenner *et al*. (1984) was used (Fig. 2). The concentration of phosphorus was determined using UV–Vis Cecil1020 spectrophotometer technique. All samples were analyzed at maximum wavelength of 885 nm according to the molybdate blue method (Riley and Murphy, 1962).

Table 1: Sampling stations in the southern of Caspian Sea- Iranian coast (2013-2014)

Station's name	symbols		5 m	10m	20m
Anzali	A5 A10 A20	Latitude	49° 29`	49° 29`	49° 29`
		Longitude	37° 29`	37° 29`	37° 30`
Tonekabon	T5 T10 T20	Latitude	50° 54`	50° 54`	50° 55`
		Longitude	36° 49`	36° 49`	36° 50`
Noshahr	N5 N10 N20	Latitude	51° 30	51° 30	51° 30
		Longitude	36° 40`	36° 41`	36° 41`
Amirabad	AA5 AA10 AA20	Latitude	53° 18`	53° 17`	53° 16`
		Longitude	36° 52`	36° 53`	36° 56`

Figure 1: Map of the Caspian Sea sampling stations in the southern of Caspian Sea-Iranian coast (2013-2014).

Bio-available phosphorus that is the sum of loosely adsorbed-P, BD-P and Al-P was also determined. The preparation of Ca, Fe and Al was done according to Standard method MOOPAM, (2005) and were analyzed by atomic absorption spectroscopy using GFAAS techniques (Thermo M5). Total organic matter (TOM) in sediments was analyzed as the loss of ignition at 500ºC for 2 h (Jensen and Anderson, 1992). Main characteristics of the studied sediments are shown in Table 2.

Table 2: Main characteristics of the studied sediments, Caspian Sea- Iranian coast (2013-2014)

Mean (±SE)	Autumn	Winter
Fe_{tot}(mg/g.dw)	24.5(±0.7)	23.5(±0.8)
Al_{tot}(mg/g.dw)	19.9(±1.5)	18.2(±0.9)
Ca_{tot}(mg/g.dw)	62.1(±3.5)	83.2(±3.1)
TOM(%)	2.9(±0.4)	2.1(±0.2)
Eh	-82.8(±0.3)	-64.5(±0.3)
pH	8.2(±0.0)	8.1(±0.0)

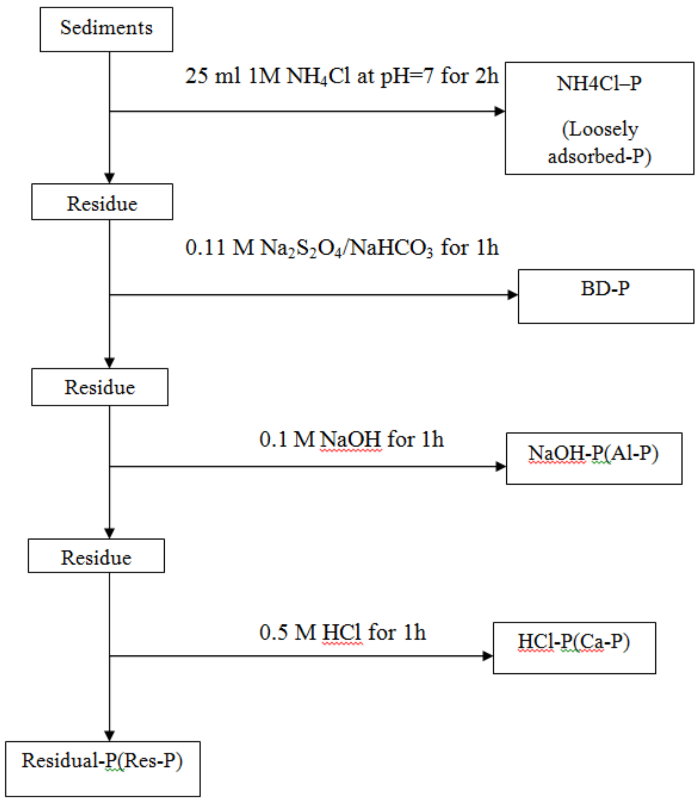


Figure 2: Sequential extraction method followed in the present study (Psenner *et al*., 1984).

Data treatment and testing for appropriateness

The results according to the Shapiro-Wilk test showed that the data transferred did not reject hypothesis of normality. To examine the validity and suitability of these data for the PCA, two widely used statistical tests, namely Kaiser Meyer Olkin (KMO) test which measures the sampling adequacy and Bartlett's tests, were performed (Ghiyasvand, 2008). The initial set of components generated by PCA is not readily interpretable therefore it is usually transformed by Varimax rotation. In this study, KMO coefficient was equal to 0.60. All data obtained from the two sampling periods were used for statistical analyses using SPSS 22.0 software.

[DOR: 20.1001.1.15622916.2016.15.2.6.2]

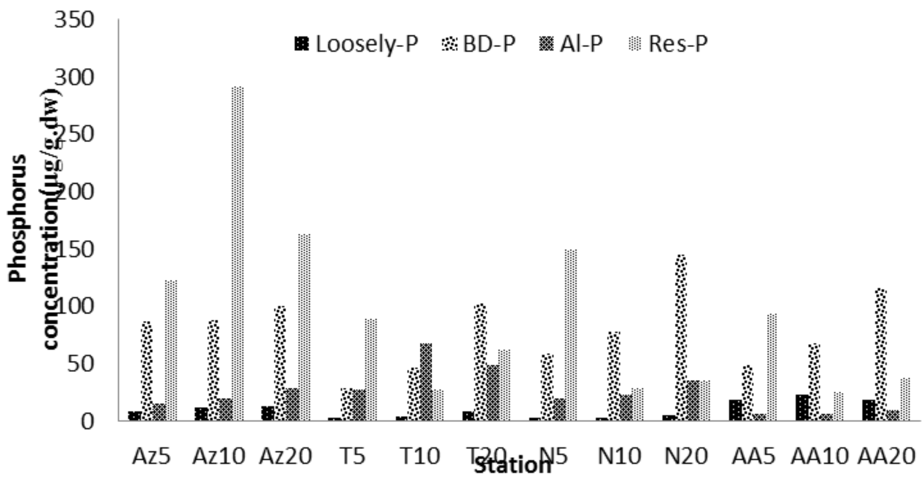
Results

The concentration of total P (TP) showed significant differences in seasons and depths ($p<0.05$). As shown in Table 3, the concentrations of TP were higher in autumn. The concentration of total inorganic P (TIP-sum of loosely adsorbed-P, BD-P, Al-P and Ca-P) showed significant differences in seasons ($p<0.05$). The concentrations of bioavailable-P (sum of loosely adsorbed-P, BD-P and Al-P) showed significant differences in seasons and stations ($p<0.05$). The concentration of this form of phosphorus is higher in the western and central part of the Caspian Sea (Figs. 3 and 4). BD-P showed significant differences in seasons, stations and depth. Al-P didn't show significant differences in seasons but in stations. Unlike Al-P, loosely adsorbed-P showed significant differences in different seasons. Results of the current study indicated that the most abundant form of phosphorus in both seasons was Ca-P. But, relative abundances of other forms of phosphorus in autumn follow the order: Res-P>BD-P>Al-P>loosely adsorbed-P and in winter: BD-P>Res-P>Al-P>loosely adsorbed-P. The loosely adsorbed-P represented <1% of the sedimentary inorganic phosphorus, while the BD/Al-P ranged from 4 to 5%. The Res-P ranged between 2.3 and 4.5% (Figs. 3 and 4). The Ca-P showed considerable contribution 91% (Figs. 5 and 6).

PCA was applied to the 9 variables collected during the two seasons (Table 4). The results of PCA showed that three main factors were enough to explain more than 72.5% of cumulated variance. PC1 and PC2 account for 39.1% and 16.7% of the total variance, respectively. PC1 explained TP, Ca/BD-P, Fe_{tot}, Al_{tot} and TOM while PC2 included loosely adsorbed/BD-P and Ca_{tot}. PC3 explained relatively lower variance (16.5%) with a high loading factor on loosely adsorbed and Al bound phosphorus.

Table 3: Mean (±SE) of different forms of phosphorus during different seasons and depths in the southern Caspian Sea surface sediments (2013-2014).

		Autumn			Winter		
		5m	**10m**	**20m**	**5m**	**10m**	**20m**
Loosely-P	Mean±SE	8.4(±3.6)	11.1(±4.8)	13.5(±3.0)	3.1(±1.0)	4.4(±1.4)	6.8(±1.3)
BD-P	Mean±SE	56.1(±12.0)	70.4(±8.9)	116.1(±10.3)	40.0(±5.6)	47.8(±2.4)	66.6(±6.4)
Al-P	Mean±SE	17.3(±4.5)	29.3(±13.3)	30.6(±8.0)	24.6(±3.8)	30.1(±3.6)	35.1(±10.4)
Ca-P	Mean±SE	17.84.4(±50.2)	1889.3(±74.3)	1926.1(±64.0)	1706.9(±88.0)	1759.2(±84.4)	1825.8(±102.2)
Res-P	Mean±SE	116.0(±13.0)	94.0(±65.7)	70.5(±30.8)	54.6(±19.8)	33.2(±8.1)	45.3(8.8)
TP	Mean±SE	2008.5(±24.0)	2080.1(±39.1)	2208.4(±51.2)	1828.1(±102.5)	1874.9(±91.0)	1979.9(±105.0)



Figurer 3: The variation of Loosely/BD/Al and Res-P between the stations in marine sediment Caspian Sea-Iranian coast, autumn (2013).

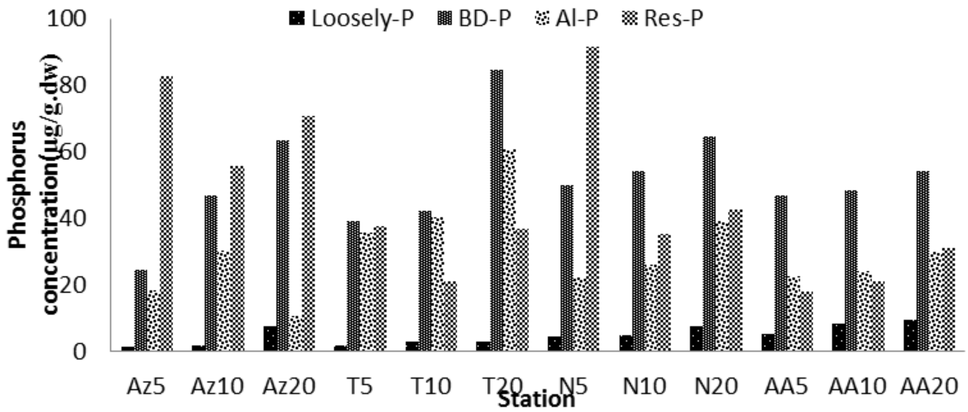


Figure 4: The variation of loosely adsorbed/BD/Al and Res-P between the stations in marine sediment Caspian Sea-Iranian coast, winter (2014).

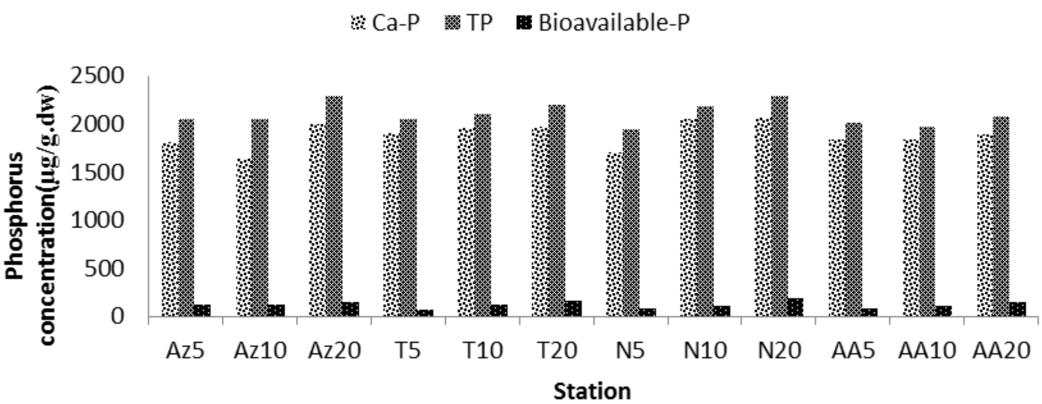


Figure 5: The variation of Ca/Bioavailable-P and TP between the stations in marine sediment Caspian Sea-Iranian coast, autumn (2013).

[DOR: 20.1001.1.15622916.2016.15.2.6.2]

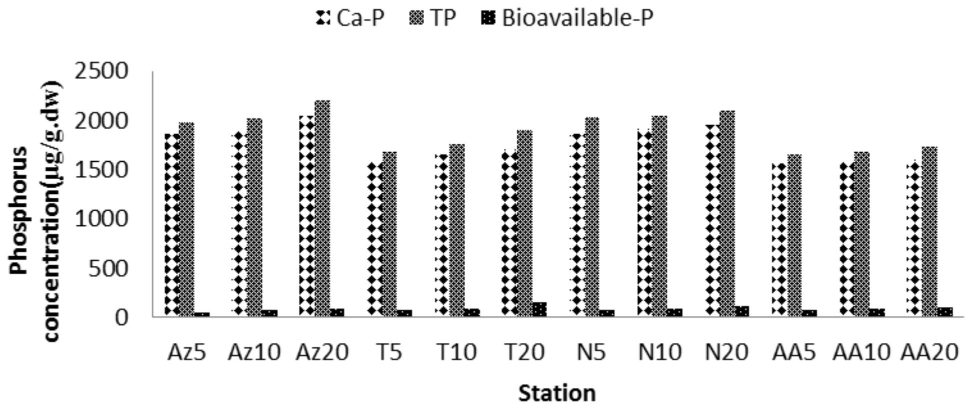


Figure 6: The variation of Ca/bioavailable-P and TP between the stations in marine sediment Caspian Sea-Iranian coast, winter (2014).

Table 4: PCA results from correlation matrix at the different depths in the southern Caspian Sea surface sediment of the Iranian coast (2013-2014).

	Component		
	PCA1(39.1%)	**PCA2(16.7%)**	**PCA3(16.5%)**
TP	0.926		
Loosely-P	0.554-0.630		
BD-P	0.675	0.595	
Al-P	0.906		
Ca-P	0.770		
Fe_{tot}	0.804		
Al_{tot}	0. 519		
Ca_{tot}	0.890		
TOM	0.745		

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.

Discussion

Loosely adsorbed-P in the sediment can be easily released into water and taken up by phytoplankton. This fraction may contain pore water P, P released from $CaCO_3$-associated phosphorus or leached P from decaying cells of bacterial biomass in deposited phytodetrital aggregates (Gonsiorezyk *et al.*, 1998; Pettersson, 2001). As mentioned above this form of phosphorus had <1% of sedimentary total inorganic phosphorus. In current study, Al-P was associated to PC3. This factor suggests that Ca_{tot} may control the presence of loosely adsorbed-P and probably represents the autochthonous precipitation of P (Sallade and Sims, 1997; Gonsiorezyk *et al*., 1998).

BD–P represents the redox-sensitive P forms, mainly bound to Fe-hydroxides and Mn compounds (Kozerski and Kleeberg, 1998). This fraction is considered as potentially mobile pool of P and is algal available. As you can see in Table 2, redox potential is dominated

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in the sediments that we examined. Mean value of Eh in autumn is higher than in winter. In surface sediment BD–P can be used to determine the source of P and also to indicate the extent of environmental pollution although Fe_{tot} and Al_{tot} are heavy metals that influence the environment. PC1 showed that there was a positive correlation between TP,BD-P, Ca-P, Fe_{tot}, Al_{tot} and TOM. Results of the current study indicated that the most abundant forms of inorganic phosphorus were Ca-P and BD-P, so positive correlation between these form of phosphorus and TP is not unexpected. On the other hand, this component showed an important role of BD-P and Fe_{tot} (more important than aluminum) and organic matter in regulating mobile P-budget (Maine *et al*., 1996). It is also indicated that this P-form can be considered the most reactive one in this system.

The NaOH–P represents P bound to metal oxides, mainly of Al, which is exchangeable again with OH^- and inorganic P compounds soluble in bases (Kozerski and Kleeberg, 1998). Although, in the current study Al-P was associated to PC3, it contained high loading factor, variance and concentration, which had principal role to release. On the other hand, negative correlation between Al-P and loosely adsorbed-P represent the interrelationships among the algal available phosphorus fractions. Mineralization of organic matter could be one possible route of mobilized P from Al–P to the other P-fractions (Gonsiorezyk *et al*., 1998).

Res-P is the difference between TP (total phosphorus) – determined by digestion method and the P-sum in the fractions 1–4. Organic and refractory P compounds are included in this fraction. As mentioned above, the sampling month significantly affected variance of Res-P. In autumn, because of the existence of *Mnemiopsis leidyi* that secretes mucus with high organic matter, this kind of phosphorus increases.

The mean concentration of bioavailable-P was 116.8 (±10.6) and 87.3(±7.0) µg/g.dw in autumn and winter, respectively. In autumn, the high population of *M. leidyi* in the Caspian Sea decreased the grazers of phytoplankton (i.e. zooplankton) and indirectly increased the phytoplankton abundance (Shiganova *et al*., 2003). Meanwhile, a combination of suitable conditions has led to the bloom of *Nodularia spumigena* mainly in the western and central part of the Caspian Sea (Nasrollahzadeh *et al.,* 2011).

Generally, it can be concluded that the P from marine surface sediments mainly originated from inorganic P fraction (96.0%). The sampling month had a significant effect on the variance of TP and other sediment features in the study area. Most of the sedimentary inorganic P in the surface sediment mainly consisted of Ca-P, 91.0%, while the other forms only constituted a minor part.TP and BD-P showed a significant positive correlation with Fe_{tot} ($p<0.05$).

The rank order of the different P extracts in autumn was: Ca-P > Res-P> BD-P>Al-P>loosely adsorbed-P and in winter it was: Ca-P>BD-P>Res-P>Al-P>loosely adsorbed-P.

Acknowledgment

The authors sincerely thank the Director and the staff of Caspian Sea Ecology Research Center (CSERC), for providing facilities to carry out the work.

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