

Spatial characteristics of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ reveal shrub-induced successional process in a coastal wetland

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ABSTRACT

Woody plant encroachment into grasslands is a worldwide phenomenon. Still, there is a research gap in quantifying the ecological processes of coastal wetland ecosystems. Here, we combined soil stable isotope technology with geostatistical methods to quantify the spatial characteristics of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ in a coastal wetland experiencing native shrub *Tamarix chinensis* encroachment in the Yellow River Delta (YRD), China, and to clarify the possible mechanisms by which shrub-induced successional processes determined the spatial distribution of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$. The results showed that soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ significantly decreased from bare land to grass and to shrub at the vegetation type level. Pearson correlation showed that soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were positively related to soil salinity, but negatively related to plant variables (grass cover, shrub crown width and above-ground biomass). Scaling method indicated that soil $\delta^{13}\text{C}$ variation occurred at two different scales, 40 m and 150 m, representing influences of shrub *T. chinensis* and environmental heterogeneity, respectively. Soil $\delta^{15}\text{N}$ variation was observed to occur at the 75 m scale, suggesting the combination effect of *T. chinensis* and *Suaeda glauca*-dominated grass species. Furthermore, other soil and plant variables also exhibited two-scale characteristics similar to soil $\delta^{13}\text{C}$. Therefore, the vegetation succession processes of coastal wetlands experiencing shrub encroachment could be understood well by combining soil stable isotope with GIS spatial tools. The spatial characteristics of soil stable isotopes help us establish strategies for protecting and managing coastal wetlands.

1. Introduction

Woody plant encroachment into grass-dominated ecosystems in arid and semi-arid regions is a worldwide phenomenon during the past 100 years (Van Auken, 2000, 2009). Recently, research has indicated that the global shrub encroachment in terrestrial grasslands is broadly coincident with the expansion and increasing density of woody trees and shrubs in coastal wetlands (Saintilan and Rogers, 2015). For example, a consistent expansion and increasing density of mangrove forest over the past century has been observed in southern Texas and Florida of the U.S. (Bianchi et al., 2013; Smith and Dukes, 2013; Zhai et al., 2019), and in Mexico (Lopez-Medellin et al., 2011) and Peru (Saintilan et al., 2014). In northern China, some tidal wetlands dominated by grass species had experienced the expansion of a native species, *Tamarix chinensis*, (a C₃-shrub) in recent decades due to human activities such as seawall reclamation (Qin et al., 2015).

Shrub encroachment has dramatically altered the structure and function of coastal wetland ecosystems (Kelleway et al., 2017), which

would also strongly affect soil moisture and microclimate (Thompson et al., 2017), spatial heterogeneity of vegetation and habitats (Bird et al., 2017; Shirley and Battaglia, 2006), and species diversity of insects and birds (Schirmel and Fartmann, 2014).

The carbon sequestration rate per unit area of wetland ecosystems is approximately 10 times greater than that of terrestrial forest ecosystems (McLeod et al., 2011). Therefore, any plant-induced shifts in coastal wetlands could potentially influence the carbon cycle process and the climate changes via the impacts on soil properties and dynamics, both regionally and globally.

It is necessary and important to investigate in detail the ecological implications of shrub encroachment in coastal wetlands, especially the relationship between shrub-induced successional process and the evolution of soil C and N pools. In addition, shrub encroachment can not only alter size of soil pools of C and N and but also increase the spatial heterogeneity of those pools. These driving factors depend on spatial scales. Therefore it is hard to quantify their spatial characteristics by traditional sampling investigations undetecting the spatial variance

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information (Liu et al., 2010). However, scaling method can provide a new solution to quantifying the spatial relationships from one scale to another one, because one of the important functions of scaling is to clarify the domains of scale and consequently explain the relationships in a given domain (Schneider, 2001).

The natural abundance of stable C and N isotopes in soils can be widely used to detect historical changes of C and N pools and their transformation rates in natural ecosystems (Lynch et al., 2006). Changes in stable isotopic composition of soil organic matter in arid and sub-arid regions experiencing woody shrub encroachment have been investigated widely (Bai et al., 2008; Boutton et al., 1998; Liu et al., 2010; Zhou et al., 2018), which showed that depending on their input and output, soil C and N pools could have positive, negative or insignificant effects on their stable isotopes (Bai et al., 2009, 2013). However, there is a

knowledge gap in our current understanding of what impacts the stable C and N isotopes in coastal wetlands, in view of the small amount of research on this field, especially regarding spatial variation of these isotopes.

We have therefore in this work investigated the spatial characteristics of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ in a coastal wetland after shrub-induced disturbances, combining stable isotope technology with geostatistics methods. The objectives were: 1) to quantify the spatial patterns of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ at vegetation type and transect levels and 2) to clarify the mechanism of shrub-induced succession determining the spatial patterns of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$.

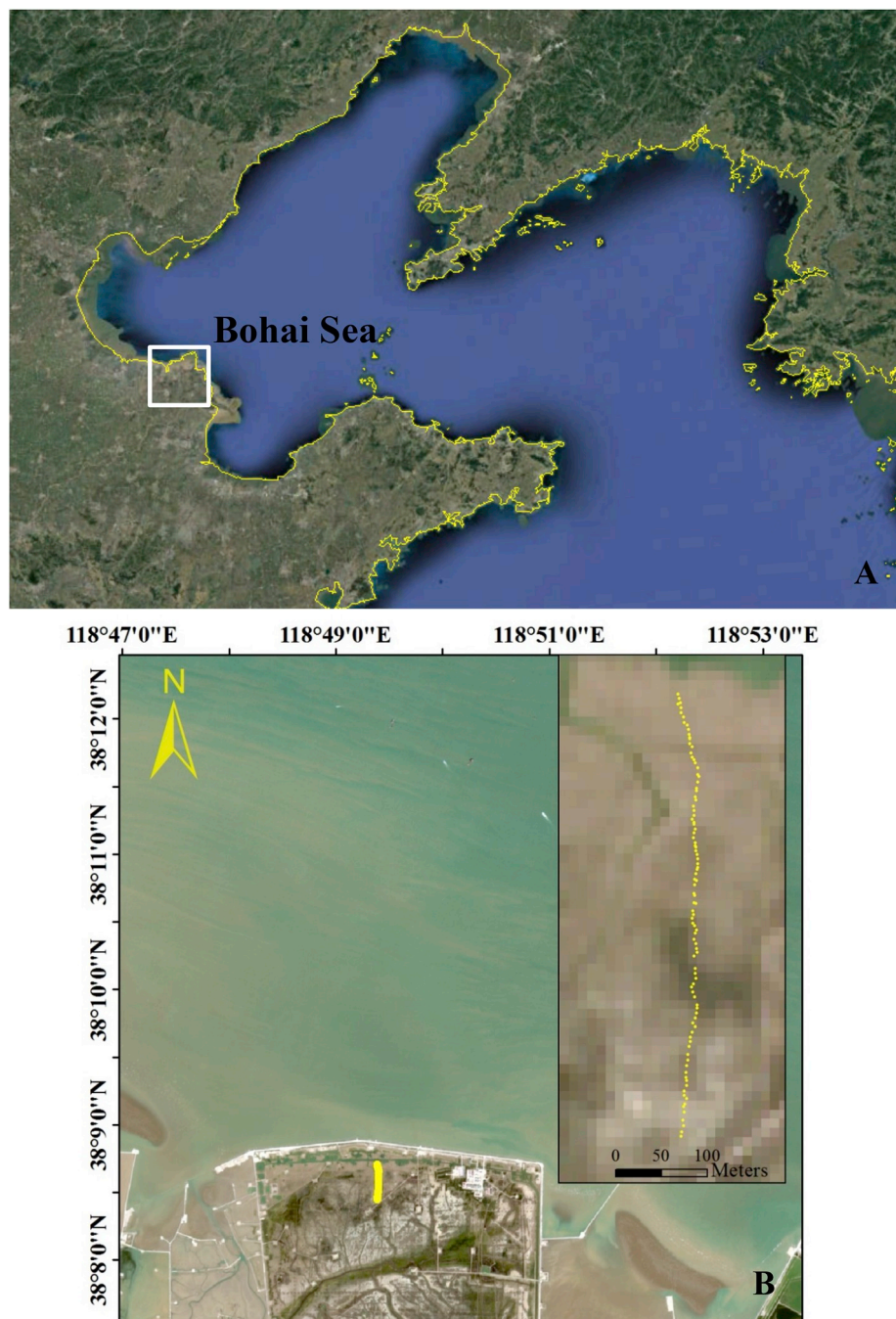


Fig. 1. Location of the Yellow River Delta (A) and the sampling transect (B)

2. Study area and methods

2.1. Study area

This research was conducted at the Yellow River Delta Natural Reserve (YRDNR), which is located on the southwest coast of the Bohai Sea (Fig. 1). The annual mean temperature, rainfall and evaporation are 12.1 °C, 551.6 mm and 1962 mm, respectively. The elevation is low, and most of the area is < 10 m above sea level. Tidal fluctuation is mixed semidiurnal, with successive tides of unequal size (He et al., 2009). The main soil type is sandy loam saline-alkali soil (coastal solonchak) (Yu et al., 2016).

In this region, land cover has changed dramatically over the past 30 years, and an approximately 658.6-km seawall has been constructed along the coastline to protect oil fields and agricultural activities from storm tides and erosion (Fu et al., 2011). Shrub species began to encroach on the tidal flats as a consequence of the land cover changes and seawall construction (Qin et al., 2015).

2.2. Sampling method

One transect (500 m × 4 m) from sea to land was established in 2013 at the northern YRDNR (Fig. 1), and sampling sites were designated at 5-m intervals using a global positioning system device (GPSMAP 62sc; Garmin, China). At each site, two soil cores (15 cm depth and 2 cm diameter, Liu et al., 2010; Bai et al., 2008) were collected: one for analyzing soil salinity, soil organic carbon (SOC), total nitrogen (TN), and C and N stable isotopes ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$), and another for measuring root biomass. Previous researches showed that mean residence time of SOC at depth of 0–15 cm ranged from about 30 to 50 years, which was able to capture the change of soil $\delta^{13}\text{C}$; while SOC at depth >15 cm has mean residence time >150 years (Boutton et al., 1998). Thus, soil sampling depth was limited to 0–15 cm, where soil $\delta^{13}\text{C}$ reflected the grassland condition that once dominated the area.

In this region, only shrub (*T. chinensis*) and grass species were noted because no tree species were found. For shrub species, the numbers and crown widths within a 2-m wide belt centered on this transect were recorded; for grass species, the percentage cover within a 0.5 m × 0.5 m plot at each site was recorded. Additionally, above-ground biomass including green grass biomass and litter mass was collected at each site, and then the dry weight was measured after oven-drying at 60 °C for at least 72 h (constant weight). In our area, the dominated grass are two *C₃* grass species, *Suaeda glauca* and *Phragmites communis*.

In sum, this transect was divided into three vegetation types: bare land (without plants), 55/100 sites; grass (without shrub), 24/100 sites and shrub (with shrub), 21/100 sites.

2.3. Lab analysis

Roots were obtained by washing the soil cores, and then the dry weight was measured after oven dried to a constant weight at 60 °C at least 72 h. Soil samples were air-dried and crushed, and then were passed through a 2-mm sieve after removing roots and gravel.

Soil electrical conductivity (EC, as an indicator of soil salinity) was measured using a conductivity meter (TP320; Mitong, Beijing, China). SOC and TN were measured using an elemental analyzer (vario MACRO cube; Elementar, Langensfeld, Germany). $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were measured using a stable isotope mass spectrometer (MAT253, Thermo Fisher, USA). C and N stable isotopes ratios ($^{13}\text{C}/^{12}\text{C}$ or $^{15}\text{N}/^{14}\text{N}$) were calculated using δ notation:

$$\delta = [(R_{\text{SAMPLE}} - R_{\text{STD}}) / R_{\text{STD}}] \times 10^3 \quad (1)$$

Where R_{SAMPLE} and R_{STD} is the $^{13}\text{C}/^{12}\text{C}$ ratio (or $^{15}\text{N}/^{14}\text{N}$ ratio for nitrogen isotopes) of the sample and the VPDB standard, respectively (Coplen, 1995). Precision of duplicate measures was 0.1‰ for $\delta^{13}\text{C}$ and

0.2‰ $\delta^{15}\text{N}$, respectively.

2.4. Statistical methods

One-way analysis of covariance (ANCOVA) was used to compare differences in soil (SOC, TN, C:N ratio, $\delta^{13}\text{C}$, $\delta^{15}\text{N}$ and salinity) and plant variables (total grass cover, shrub crown width, above ground biomass and root biomass) among the three vegetation types. First, the effect of distance (from sea to land) as covariance was excluded. *Post hoc* comparisons of those variables in different vegetation types were also conducted after ANCOVA, using SAS version 9.2 (SAS Institute, Cary, NC, USA). P value (<0.05) was used to indicate significant differences.

Mantel and cross-Mantel tests were used to test the spatial autocorrelation and spatial cross-correlations between soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ with other variables respectively using PASSaGE version 2 (Rosenberg and Anderson, 2011).

If Mantel test indicated that soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were spatially autocorrelated, the significance levels of these correlation coefficients will be assessed with a modified-t test, which has been to adjust the freedom degrees using the procedure of Clifford, Richardson and Hemon (CRH) (Clifford et al., 1989). Mantel tests worked by evaluating the relationship between two matrices. Cross-Mantel test indicated the spatial correlation between two variables.

Quadrat variance methods, which stems from the application of hierarchical analysis of variance to the data from grids or strips of continuous quadrats (Szmyt, 2014), were used to assess the spatial scales of those variables. Through calculating the variance of differences among blocks of different sizes, the patterns of variance can be determined and used to detect the scale characteristics. There are many similar quadrat methods (Liu et al., 2010), and we here chose three-term local quadrat variance (3TLQV) because of its lower sensitivity to trends in data and peak drift (Hanewinkel, 2004). 3TLQV was processed using GIS software (PASSaGE version 2) (Rosenberg and Anderson, 2011), in which the variance is calculated as follows:

$$V_3(b) = \sum_{i=1}^{n+1-3b} \left(\sum_{j=i}^{i+b-1} X_j - 2 \sum_{j=i+b}^{i+2b-1} X_j + \sum_{j=i+2b}^{i+3b-1} X_j \right)^2 / 8b(n+1-3b) \quad (2)$$

where X_1, X_2, \dots, X_n are counts of points, n is the number of quadrats, j is the number of blocks and b is block size (Dale et al., 2002).

3. Results

3.1. Spatial changes of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ at vegetation type level

ANCOVA results indicated that soil characteristics such as $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, TN, SOC, as well as soil salinity (all $P < 0.05$) were significantly different among vegetation types. In bare land, average values of soil $\delta^{15}\text{N}$ (4.70‰), soil $\delta^{13}\text{C}$ (−23.12‰), C:N ratio (27.68) and salinity (4.34 mS/cm) were the highest, whereas average values of soil TN (0.05%) and SOC (1.32%) were the lowest. Contrarily, the lowest value of soil $\delta^{13}\text{C}$ (−23.83‰) and $\delta^{15}\text{N}$ (3.12‰) and the highest values of soil SOC (1.44%) and TN (0.07%) were observed in shrub (Table 1).

In addition, shrub has total grass cover and above ground biomass (73.6% and 81.29 g m^{−2}) than higher than grass (66.66% and 70.77 g m^{−2}) and bare land (0% and 0 g m^{−2}). However, root biomass of shrub and grass types were not significantly different (Table 1).

3.2. Spatial changes of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ along the sea-land transect and their relation to other variables

Soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ decreased from −20.59‰ to −26.01‰ and from 6.11‰ to 0.85‰, respectively, along the sea-land gradient (Fig. 2). Mantel test showed that soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were spatially autocorrelated ($r = 0.52$ and 0.22 , respectively, $P < 0.0001$). Cross-Mantel correlation indicated that soil $\delta^{13}\text{C}$ was spatially correlated with $\delta^{15}\text{N}$ ($r = 0.24$). Our

Table 1

Mean and standard error (SE) of variables with ANCOVA results in bareland, grass and shrub types in the Yellow River Delta.

	Bare land		Grass		Shrub		ANCOVA
	mean	SE	mean	SE	mean	SE	P
Soil $\delta^{13}\text{C}$ (‰)	-23.12 ^a	0.093	-23.73 ^b	0.14	-23.83 ^c	0.18	<0.001
Soil $\delta^{15}\text{N}$ (‰)	4.70 ^a	0.12	3.59 ^b	0.17	3.13 ^c	0.22	<0.001
Soil SOC (%)	1.32 ^a	0.02	1.39 ^b	0.03	1.44 ^c	0.03	<0.001
Soil TN (%)	0.05 ^a	0.00	0.06 ^b	0.00	0.07 ^c	0.00	<0.001
Soil C:N ratio	27.68 ^a	0.54	23.66 ^b	0.80	23.68 ^b	1.00	= 0.002
Soil salinity(mS)	4.34 ^a	0.16	3.05 ^b	0.29	2.22 ^c	0.23	<0.001
Elevation(m)	-2.02 ^a	3.78	-0.94 ^a	3.03	-1.15 ^a	3.62	0.2930
Total grass cover (%)	0.00 ^a	0.00	66.66 ^b	39.3	73.6 ^c	29.84	<0.001
Above ground biomass(g m ⁻²)	0.00 ^a	0.00	70.77 ^b	44.26	81.29 ^c	44.23	<0.001
Root biomass(g m ⁻²)	0.05 ^a	0.12	0.27 ^b	0.30	0.23 ^b	0.30	<0.001

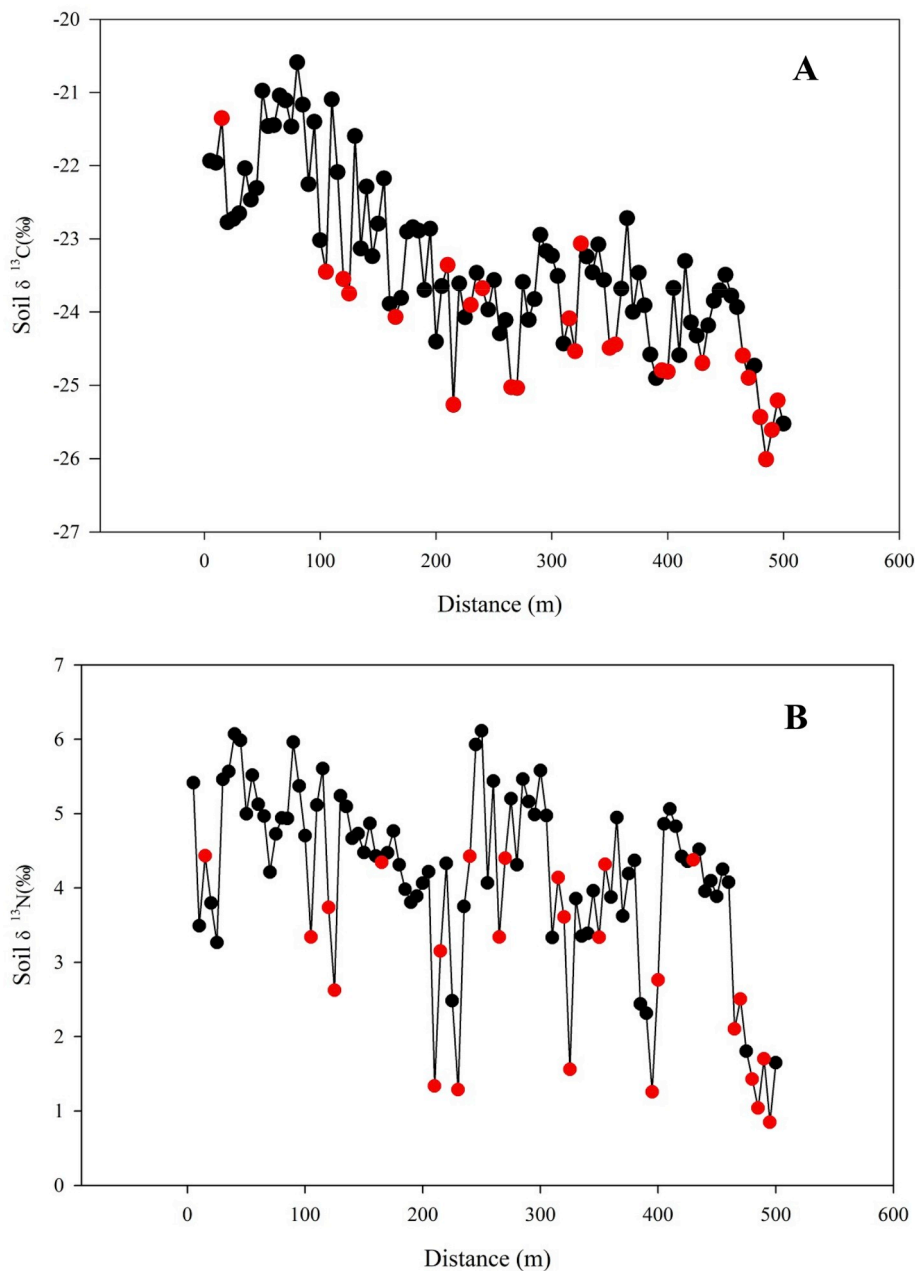
**Fig. 2.** Spatial distributions of soil $\delta^{13}\text{C}$ (A) and $\delta^{15}\text{N}$ (B) with shrubs (red points) and without shrubs (black points)

Table 2Cross-Mantel correlation results for soil $\delta^{13}\text{C}$, $\delta^{15}\text{N}$ and other soil and plant variables in the Yellow River Delta.

	Soil SOC	Soil TN	Soil C:N ratio	Soil Salinity	Elevation	Total grass cover	Shrub crown width	Above ground biomass	Root biomass
Soil $\delta^{13}\text{C}$	-0.008	0.031	-0.032	0.126 ^a	0.054	0.1472 ^a	0.215 ^a	0.089	0.029
Soil $\delta^{15}\text{N}$	0.028	-0.009	-0.009	0.422 ^a	0.067	0.4374 ^a	0.396 ^a	0.2507 ^a	0.080

^a significant level > 0.0001, the same as following.

results also indicated strongly spatial correlations between soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ with soil salinity ($r = 0.13$ and 0.42), total grass cover ($r = 0.15$ and 0.44) and shrub crown width ($r = 0.22$ and 0.40). In addition, soil $\delta^{15}\text{N}$ was spatially related with above ground biomass ($r = 0.26$) (Table 2).

Pearson correlation (modified t -test) also showed soil $\delta^{13}\text{C}$ was closely related with $\delta^{15}\text{N}$ ($r = 0.59$). Further, soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were significantly and positively correlated with soil salinity ($r = 0.40$ and 0.67 , respectively). Soil $\delta^{13}\text{C}$ were significantly and negatively correlated with shrub crown width ($r = -0.47$), followed by total grass cover ($r = -0.41$), above ground biomass ($r = -0.33$), while soil $\delta^{15}\text{N}$ was significantly and negatively correlated with total grass cover ($r = -0.67$), followed by shrub crown width ($r = -0.58$), above ground biomass ($r = -0.54$) and root biomass ($r = -0.3$) (Table 3).

3.3. Spatial scaling of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ patterns

Three-term local quadrat variance (TLQV) results revealed soil $\delta^{13}\text{C}$ variation at two spatial scales (40 m and 150 m, respectively) and soil $\delta^{15}\text{N}$ variation at one spatial scale of 75 m (Fig. 3). In addition, a smaller scale of variation (30–40 m) for soil SOC, TN, C:N ratio, salinity and larger scale of variation (130–150 m) for grass cover, shrub crown width, above ground biomass, and root biomass were also detected as soil $\delta^{13}\text{C}$ variation (Table 4).

4. Discussion

Because C_3 plants have $\delta^{13}\text{C}$ values ranging from -32‰ to -22‰ , while C_4 plants have $\delta^{13}\text{C}$ values ranging from -17‰ to -9‰ (Bai et al., 2008), we could identify the sources of SOC in the YRD wetlands experiencing shrub encroachment by analysing their stable isotope compositions. Our results agreed with previous research, which showed that soil $\delta^{13}\text{C}$ values ranged from -26‰ to -22‰ (Fig. 2) (Li et al., 2016), indicating that natural wetlands in this region possessed mainly C_3 plant species (e. g., *S. glauca*, *P. communis* and *T. chinensis*).

However, soil $\delta^{15}\text{N}$ ranged from 6.8‰ to 1‰ (Fig. 2), which was far lower than that in C_3 -dominated coastal marshes in San Francisco Bay estuary (1‰ – 17‰) (Cloern et al., 2002). The wetland ecosystems in the YRD, like many northern and temperate forests, are nitrogen-limited (Reay et al., 2008; Jia et al., 2018). N limitation might influence nitrogen availability of wetland plants and thus accumulation process of soil $\delta^{15}\text{N}$ (King et al., 2009; McLauchlan et al., 2010). More works about the effects of N limitation on N transformation and $\delta^{15}\text{N}$ changes in plant and soil ecosystems under different environmental conditions are needed.

4.1. Spatial patterns of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ at vegetation type level and alternative mechanisms

Vegetation shifts induced spatial variability in soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ at

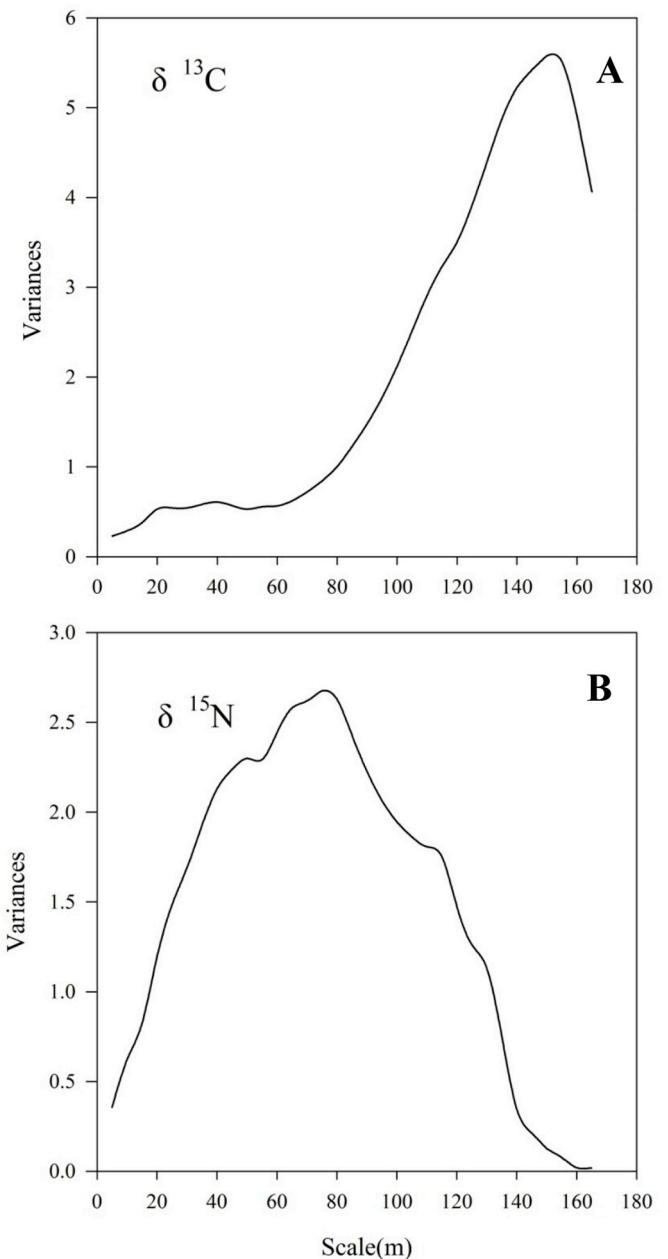


Fig. 3. Three term local quadrat variance method (3TLQV) results of soil $\delta^{13}\text{C}$ (A) and $\delta^{15}\text{N}$ (B) at the sea-land transect in the Yellow River Delta.

Table 3Pearson correlation results (modified t -test) for soil $\delta^{13}\text{C}$, $\delta^{15}\text{N}$ and other soil and plant variables in the Yellow River Delta.

	Soil SOC	Soil TN	Soil C:N ratio	Soil Salinity	Elevation	Total grass cover	Shrub crown width	Above ground biomass	Root biomass
Soil $\delta^{13}\text{C}$	-0.005	0.031	-0.037	0.4004**	-0.254	-0.4144**	-0.4718**	-0.3336**	-0.15
Soil $\delta^{15}\text{N}$	0.042	-0.009	-0.029	0.679**	-0.250	-0.6697**	-0.5836**	-0.536**	-0.380**

Table 4

Three term local quadrat variance method (3TLQV) results of soil and plant variables in the Yellow River Delta.

	Scale (m)
Soil $\delta^{13}\text{C}$	40,150
Soil $\delta^{15}\text{N}$	75
Soil SOC	40,130
Soil TN	30,130
Soil C:N ratio	30,125
Soil salinity	45, 130
Elevation	-
Total grass cover	40,135
Shrub crown width	25,140
Above ground biomass	35,145
Root biomass	40,130

–: no significant variation.

vegetation type level. Our results showed that soil $\delta^{13}\text{C}$ exhibited a spatial pattern similar to soil $\delta^{15}\text{N}$, in which they were decreased with shrub encroachment (from bare land, grass to shrub type) (Table 1). These findings are consistent with other studies reporting in both coastal and arid ecosystems, where the higher the coverage of C_3 -dominant vegetation became, the lower the soil $\delta^{13}\text{C}$ content that occurred (Biggs et al., 2002). The phenomenon (the lowest soil $\delta^{13}\text{C}$ in the shrubs) could be largely contributed to: (1) declines in $\delta^{13}\text{C}$ content of *T. chinensis* twigs because of the declines of soil salinity (Sun et al., 2016), and (2) changes in soil salinity can also increase turnover rates of soil organic matter and accumulation process of soil $\delta^{13}\text{C}$ (Telles et al., 2003). Since the dominant shrub and grass species are all C_3 -plants in our case, additional works should compare the influences of soil salinity on $\delta^{13}\text{C}$ content of C_3 shrub with C_3 grass.

The observed lowest soil $\delta^{15}\text{N}$ in shrub patches could be explained as follows: 1) Shrub encroachment increased soil N pool because the return of litter in shrubs (81.29 g m^{-2}) were more than that in grasses (70.77 g m^{-2}) (Table 1). Bai et al. (2013) demonstrated that as the soil N pool increase, the soil $\delta^{15}\text{N}$ decreased. Zhang et al. (2019) showed that N_2O emissions in soils of grass communities were higher than that of woody *T. chinensis* communities in the YRD. Thus, compared with grassy areas, there is more N input to and less N output from the soil in shrub types in this delta, leading to lower soil $\delta^{15}\text{N}$. 2) Endophytic nitrogen-fixing bacteria found in the root systems of *Tamarix* family could increase

soil N input (Moseman et al., 2009). The biologically fixed $\delta^{15}\text{N}$ value is approximately 0‰ (atmospheric $\delta^{15}\text{N}$) (Houlton et al., 2006), far lower than soil $\delta^{15}\text{N}$. Therefore, this shrub could increase N input by biological fixation and subsequently reduce soil $\delta^{15}\text{N}$. However, no reports have been found concerning the biological fixation of this shrub species in the YRD.

4.2. Spatial patterns, scaling characteristics of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ at transect level and environmental mechanisms

On the transect, vegetation succession has strongly determined soil salinity as well as soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$. From sea to land, soil $\delta^{13}\text{C}$ had regularly decreased. Fig. 2 showed that soil $\delta^{13}\text{C}$ was low at a larger percentage of sites with shrub species (80%), whereas soil $\delta^{15}\text{N}$ was low at a smaller percentage of sites with shrub species (60%). Many researches have indicated that soil salinity is closely related to soil $\delta^{13}\text{C}$ due to positive relation with the plant $\delta^{13}\text{C}$ (such as Hussain and Al-Dakheel, 2018), while Doongar et al. found that plant $\delta^{15}\text{N}$ was negatively correlated with soil available P in a coastal ecosystem (Chaudhary et al., 2018). Therefore, it is important to understand the sensitivity of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ and their response to different soil environments.

Interestingly, scaling method (3TLVQ) detected significant differences between soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, where soil $\delta^{13}\text{C}$ was scaled at about 40 m and 150 m, whereas soil $\delta^{15}\text{N}$ was scaled at about 75 m (Fig. 3 and Table 4). Scale characteristics of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ can help us to deeply understand their spatial patterns and the driving mechanism.

We used a hypothesis model to explain the form of spatial pattern of soil C and N stable isotopes (Fig. 4). Soil $\delta^{13}\text{C}$ has two distinct scale characteristics which reflect the influences of shrub. For instance, 40 m was the appropriate average distance between the two centroid points of shrub *T. chinensis* in this region. This scale reflected the influence of individual shrub (Fig. 4-A). 150 m was about the distance between two large shrub clusters including different shrub patches and reflecting environmental heterogeneity (Fig. 4-A). Fig. 4-A had addressed the leading effects of shrub on soil $\delta^{13}\text{C}$.

However, soil $\delta^{15}\text{N}$ has only one scale characteristics which reflected combined effects of shrub and grass. 75 m was the approximate average distance between the two central points of shrub *T. chinensis* and grass *Suaeda glauca* patches. In the YRD, *S. glauca* is abundant under

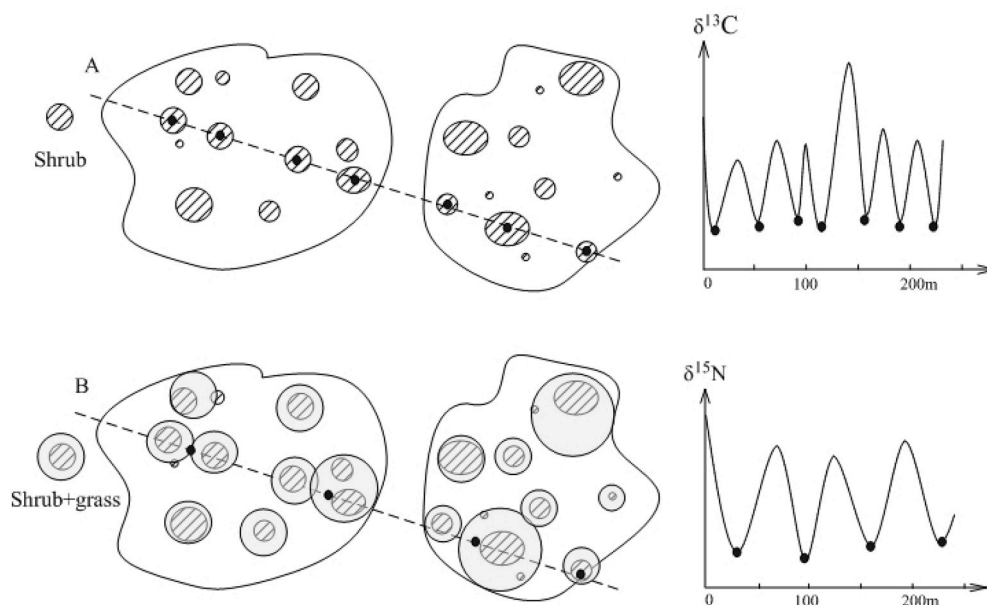


Fig. 4. Hypothesized spatial patterns and central point of shrub and grass patches and the expected spatial patterns of soil $\delta^{13}\text{C}$ (A) and $\delta^{15}\text{N}$ (B). Cross-lined circle: shrub patch; grey circle: grass patch; empty circle: shrub cluster; black dot: central point of shrub patch (A) and of shrub+grass patch (B).

T. chinensis canopies (He et al., 2012), probably because of the fertilizer island effects of *T. chinensis* (Liu et al., 2017), thereby making it very easy to form relatively homogenous patches. Therefore, the central point of individual shrub patches was shifted and subsequently the scale span was enlarged (Fig. 4-B). Here, grass might also play important roles in determining the patterns of soil $\delta^{15}\text{N}$. Research had indicated that *S. glauca* has higher ability to store nitrogen in its leaves and roots in an N-limited environment (Song et al., 2009), causing the corresponding changes in soil $\delta^{15}\text{N}$.

Moreover, the cross-Mantel and Pearson correlation analysis also evidenced that grass had influenced soil $\delta^{15}\text{N}$ more than shrubs (Tables 2 and 3), suggesting that the spatial pattern of soil $\delta^{15}\text{N}$ might contain information about $\delta^{15}\text{N}$ allocation in both grass and shrub.

4.3. Application to the protection and management of coastal wetland ecosystems

The relationship between ecological patterns and processes is usually scale-dependent (Levin, 1992). Our results showed that soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ had different spatial patterns at the vegetation type and transect levels after shrub encroachment, which suggested that biochemical processes should be given more spatial consideration in evaluation of coastal wetlands with shrub encroachment. In our study, spatial pattern of soil $\delta^{15}\text{N}$ remained constant, which can give more information about historical shrub patterns than soil $\delta^{13}\text{C}$, so, it can be considered as a good indicator for tracing the process of shrub expansion in this area in the following study.

Shrub encroachment in the YRD has lasted for decades (Qin et al., 2015), and the spatial information provided by soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ would be helpful not only for historical characteristics of soil and plant but also for future evolution of soil. For example, SOM fractions reflected the patterns of salinity stress in the past (van Groenigen and van Kessel, 2002), so soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ may also be used to reconstruct historic salinity patterns. To reconstruct plant-soil patterns and predict future evolution of landscapes under various disturbances (Bai et al., 2009), we need to combine stable isotope technology with remote sensing data and GIS spatial analysis tools.

5. Conclusions

The spatial patterns of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ at the vegetation type and transect levels have been strongly influenced by the expansion of native shrub *Tamarix chinensis* in the coastal wetland of the Yellow River Delta (YRD). Soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ decreased significantly from bare land to grass and to shrub patches, and from sea to land (with the distance to sea). Mantel test showed that both soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were spatially auto-correlated. Cross-Mantel correlation indicated that there were significantly spatial correlation between soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ with soil salinity, total grass cover and shrub crown width. Further Pearson correlation showed that soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were positively related to soil salinity, but negatively related to plant variables (grass cover, shrub crown width and above ground biomass). Scaling method showed significant differences in the spatial characteristics of soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, where soil $\delta^{13}\text{C}$ variation occurred at two different scales (40 m and 150 m), whereas soil $\delta^{15}\text{N}$ variation occurred at the 75 m scale. In particular, shrubs exhibited greater negative effects on soil $\delta^{13}\text{C}$ than on soil $\delta^{15}\text{N}$. To a certain degree, the spatial pattern of soil $\delta^{15}\text{N}$ can represented the historical patterns of shrub species in the YRD. The vegetation successional processes of coastal wetlands experiencing shrub encroachment could be rebuilt and their future trends could be also forecast, by combining soil stable isotopes with GIS spatial tools.

Declaration of competing interest

This manuscript is original, is suitable to the requirements of Estuarine, Coastal and Shelf Science, and it has not been previously

submitted to any other journal. All authors have agreed the works.

CRediT authorship contribution statement

Shiwei Zhou: Supervision, Writing - review & editing. **Jingjing Wu:** Investigation. **Xiaoli Bi:** Writing - original draft.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.ecss.2020.106621>.

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