

*Research Article*

# Characteristics of seawater intrusion in Soc Trang province, Vietnam

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Received: 18 October 2023; Accepted: 22 November 2023; Published: 25 March 2024

**Abstract:** Seawater intrusion poses many severe issues in many coastal areas around the world, including the Mekong Delta, Vietnam. For sustainable water management in such regions requires, it is crucial to understand the characteristics of salinization. This study aims to investigate the characteristics of the salinization phenomenon in water systems and its controlling factors based on hydrogeochemical analysis, seawater-freshwater mixing model and End Members Mixing Analysis. The results revealed that seawater intrusion negatively affects strongly on surface water quality with different magnitudes, depending on seasonal variation and tidal regime. It was estimated that surface water mixed with approximately 29.5% and 4.12% of seawater in the dry season and rainy season, respectively. However, it was found that the salinity in groundwater showed less seasonal variation and was distributed heterogeneously following the depth of screened wells and distance to the sea. The mixing ratio between seawater and groundwater varied widely, ranging from less than 10% in deep groundwater up to a maximum of 38% in shallow groundwater. Surface water salinization in the study area is controlled by upstream discharge, tidal regime, and operation of sluice gates. Meanwhile, groundwater salinization might be governed by geological characteristics, original recharge sources, human intervention, and natural variation, especially excessive groundwater exploitation in the Mekong Delta. Therefore, integrated sustainable groundwater and surface water use and management strategies in the Mekong Delta are needed in the context of human-nature intervention.

**Keywords:** Seawater intrusion; Seawater-Freshwater Mixing; Soc Trang; Vietnam.

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## 1. Introduction

Seawater intrusion (SWI) is the movement of salt sources into water system caused by natural processes and/or human activities, especially groundwater pumping. It results in high salinity concentration in water resources threatening to sustainable water use and management in many coastal regions around the world [1]. In particular, high salinity concentration in water system not only strikes directly to crops and ecosystems [2, 3] but also potentially threatens to human health [4–6]. It also causes land degradation via interconnection between salt water and soil layers, resulting in the difficulty to recovering agricultural lands in affected areas [7]. Therefore, understanding characteristics of salinization is key to preventing undesirable effects on agricultural activities and freshwater supply systems in many coastal regions in the world. Over the last several decades, SWI has been reported in different magnitudes from regional to global scales. For example, the earliest report to salinization in all coastal aquifers has been conducted by [8], and afterward, this issue has been intensively

*J. Hydro-Meteorol.* **2024**, *18*, 1-11; doi:10.36335/VNJHM.2024(18).1-11 <http://vnjhm.vn/>

reported by [9, 10]. Salinization has been occurring in the United States since 1960s posing many challenges for water management. Likewise, many coastal regions in Asia have experienced salinization. For instance, coastal areas of China have been facing with salinization due to a rapid expanding saline-affected areas since 2000s [11]. A great efforts have been made by the Chinese government to mitigate and prevent effects of water salinization processes, however, it has still been a severe issue as an increase of groundwater extraction along the coast [12]. Similarly, seawater intrusion is a serious water problem in coastal regions of India [13, 14]. In Africa, groundwater salinization was also found in many coastal regions [15] such as in the Nile Delta [16], in coastal areas of Libya [17], Morocco [18, 19], and Tunisia [20–22]. In addition, seawater intrusion has been well documented in many coastal regions in Europe [1]. In recently, water salinization is predicted to increase risks of water shortage and threat to national and global food security under impacts of climate change and sea level rise. Although numerous studies related to SWI have been intensively done for several decades [23], most of previous studies targeted to separated water resource, either surface water system or aquifer system. Naturally, surface water and groundwater are generally interconnected [24], therefore investigation of seawater intrusion into both water resources is crucial to sustainable water management along the coast.

Seawater intrusion has become the widespread environmental problem in Vietnam, threatening to water resources and ecosystem in coastal regions, especially in the Red River Delta and the Vietnamese Mekong Delta [25]. In natural conditions without human intervention, saline boundary in the Vietnamese Mekong Delta (VMD) extends to approximately 30-40 km inland [26]. However, the salinity boundary in recent years has been extremely expanding to inland due to changes of upstream discharge to the VMD [27]. For example, extreme drought and salinization have been occurred in the dry season 2016 caused huge losses of agricultural productions and resulted in severe freshwater shortage in many provinces of the delta including Tien Giang, Ben Tre, Tra Vinh, Soc Trang, Bac Lieu, Ca Mau, and Kien Giang. The salinity boundary has extended to 60-90 km inland [28]. As a result, large areas of agricultural lands, especially paddy fields and shrimp farming were destroyed, causing huge economic losses [29]. Water salinization also puts a high pressure on freshwater supply system due to an increase in salinity concentration in both surface water and groundwater [30]. Therefore, previous studies have been conducted to understand the impacts of seawater intrusion in the Mekong Delta based on different scenarios. For instance, most of researcher used hydraulic model to simulate the salt concentration based under affected by water practices in the upstream of the Mekong River Basin and socio-economic development in the Mekong Delta [26–28] as well as climate change [36–38]. However, the understanding of how much percentage of seawater contributing to surface water and groundwater along the coastal is still limited. Hence, this study aims to investigate characteristics of water salinization for sustainable water resources management in coastal area of the VMD. The main objectives are followed: (i) characterizing the tempo-spatial variation of salinity concentration in both surface and groundwater; (ii) examining the main factors controlling seawater intrusion in water system of the study area.

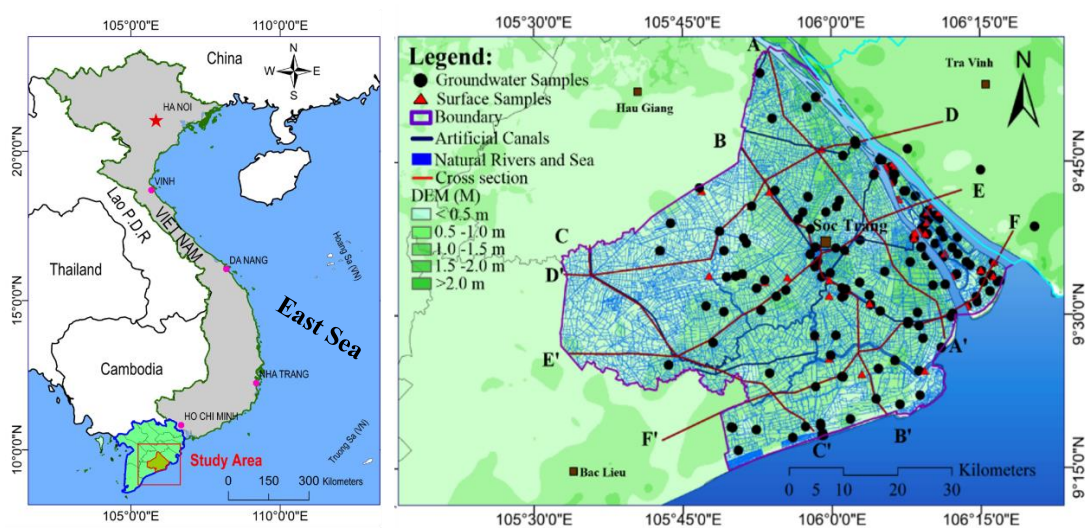
## **2. Materials and Methods**

### *2.1. Description of study site*

The research is situated in Soc Trang province, within the Mekong Delta of Vietnam, covering an approximate area of 2,300 km<sup>2</sup> with an average population of 1.32 million people (General Statistics Office of Vietnam 2016). The climate in this region is characterized by two distinct seasons - the rainy season and the dry season. The rainy season, influenced by the Southwest Monsoon, occurs from May to October annually and contributes more than 85% of the total annual rainfall of 1,875 mm. In contrast, the dry season, dominated by the

Northeast Monsoon, lasts from November to April, with a minimal contribution of just 15% to the annual rainfall. Despite significant seasonal variations in rainfall, other climate parameters such as temperature, humidity, and evaporation exhibit only minor differences. For instance, the mean monthly air temperature remains quite consistent between the dry and rainy seasons, hovering around 26.7°C, while evaporation averages around 34 mm. Humidity is approximately 85% in the dry season and 87% in the rainy season, with occasional exceptionally high daily air temperatures exceeding 36°C, particularly in the warmest months of April and May.

The hydrological system in the study area is highly intricate due to the interconnectedness of rivers, the sea, and an extensive canal network in relation to upstream discharge, tide regime, and local hydraulic construction operations (Figure 1). The region is traversed by two major rivers, Hau and My Thanh, which converge with the canal system and directly connect to the East Sea (Figure 2). Upstream discharge and interactions between river dynamics and marine influences are the primary factors driving seawater intrusion into the coastal river and canal system [31–34]. The impacts of salinization have expanded recently due to human-induced activities in the Mekong River Basin [35, 36]. Changes in upstream discharge and human activities can strongly influence both the hydrological regime and water quality, posing threats such as seawater intrusion, surface water contamination, and potential risks to groundwater resources in the area [37–39].



**Figure 1.** Map of sampling location and the river-canal system in Soc Trang Province, Vietnam.

Similarly, the hydrogeology of the region is intricate, comprising several multi-layered formations. From a management perspective, the aquifer system can be categorized into seven subdivision aquifers: Holocene (qh), Upper Pleistocene (qp3), Upper-Middle Pleistocene (qp23), Lower Pleistocene (qp1), Middle Pliocene (n22), Lower Pliocene (n21), and Upper Miocene (n13) aquifer layers. Generally, each hydrogeological unit consists of upper and lower parts with distinct characteristics. The former comprises low-permeability silt, clay, or silt clay with very low water yield, while the latter is relatively permeable, consisting of fine to coarse sand, gravel, and pebbles [40]. In this study, we group groundwater in seven aquifers into four classes following SGW (aquifer qh); DGW1 (aquifer qp3); DGW2 (qp23); DGW3 (qp1); DGW4 (n22, n21 and n13).

## 2.2. Methodologies

### 2.2.1. Hydrogeochemical data analysis

Between 2013 and 2018, a comprehensive set of geochemical analyses was conducted on a total of 278 groundwater samples and 79 surface water samples collected from the study

area. These water samples were meticulously gathered in 100 ml plastic bottles and meticulously prepared for both chemical and stable isotopes analysis. Additionally, field measurements were undertaken to acquire essential physical parameters – namely, temperature (T°C), pH, dissolved oxygen (DO), and electrical conductivity (EC) – utilizing portable instruments from HANNA.

### 2.2.2. Hydro-geochemical Evaluation Methods

Fresh seawater mixing analysis.

It was presumed that Cl<sup>-</sup> serves as a conservative tracer primarily originating from seawater. Consequently, the fraction of seawater ( $f_{sea}$ ) in the water sample can be determined based on chloride concentrations in both seawater and freshwater, as outlined in [41]:

$$f_{sea} = \frac{m_{Cl^-,sample} - m_{Cl^-,fresh}}{m_{Cl^-,sea} - m_{Cl^-,fresh}} \quad (2)$$

where  $m_{Cl^-,sample}$ ,  $m_{Cl^-,fresh}$  and  $m_{Cl^-,sea}$  represent the concentrations of Cl<sup>-</sup> in the water sample, freshwater, and seawater, respectively, all expressed in mmol/L. In coastal delta regions, fresh groundwater primarily contains Ca<sup>2+</sup> and HCO<sub>3</sub><sup>-</sup>, resulting from mineral dissolution processes. Consequently, the presence of all other ions in water samples may be attributed to seawater admixture. Under such circumstances,  $m_{i,fresh} = 0$  for all components except Ca<sup>2+</sup> and HCO<sub>3</sub><sup>-</sup> [1], and the seawater fraction ratio in Equation 2 is modified accordingly:

$$f_{sea} = \frac{m_{Cl^-,sample}}{m_{Cl^-,sea}} = \frac{m_{Cl^-,sample}}{566} \quad (3)$$

The concentration of an ion *i* ( $m_i$ ) in the blended fresh seawater was determined by applying the mass fraction of seawater ( $f_{sea}$ ), as outlined in Equation 2, as follows:

$$m_{i,mix} = f_{sea} \times m_{i,sea} + (1 - f_{sea})m_{i,fresh} \quad (4)$$

where  $m_{i,mix}$  represents the concentration of ion *i* in mmol/L in the anticipated mixed fresh-seawater. Subscripts mix, sea, and fresh denote the conservative mixture, seawater end member, and freshwater end member, respectively. The alteration in concentration,  $m_{i,reaction}$  ( $\Delta_{mi}$ ), due to reactions without mixing is then expressed as:

$$\Delta_{mi} = m_{i,sample} - m_{i,mix} \quad (5)$$

where  $m_{i,sample}$  is the measured concentration in the sample, expressed in mmol/L. Deviation in the chemical composition from a fresh-seawater mixture signifies chemical reactions resulting from seawater intrusion. A positive value indicates that the ion has been introduced to the water sample, such as through desorption from the exchange complex, while a negative value may suggest absorption processes.

End members mixing analysis (EMMA)

In this study, the EMMA model was employed to compute the ratio of each water source contributing to the surface water and groundwater within the study area. EMMA, a widely used hydro-geochemical mixing model in hydrogeological studies, has been applied extensively to identify and quantify the contribution ratios of various water sources to both surface water and groundwater [42–45]. The model utilizes the dominant chemical compositions of potential end members within the watershed to determine the percentages of each original parent water contributing to the final mixture [46, 47]. The mixing model relies on four assumptions: (i) surface water/groundwater is a final mixture of different water sources, (ii) the mixing process follows a linear and hydrodynamic mechanism, (iii) the mixed compositions act as conservative tracers, and (iv) the source solutions exhibit dominant concentrations [44]. However, hydrological processes within the watershed can vary in time and space, emphasizing the importance of careful consideration in the selection of end members before applying the EMMA approach [44, 45].

In coastal estuarial regions, water sources typically originate from various contributors, including rainfall, surface water, seawater, and groundwater from different aquifers. To

simplify the complexity of contributors, it was assumed that surface water primarily derived from rainfall, upstream surface water, and seawater, while groundwater originated from rainfall and seawater. The estimation of the contribution ratio for each end member to surface water and groundwater is based on the following equations:

For surface water:

$$f_R + f_{UPW} + f_{SW} = 1 \tag{6}$$

$$f_R \times \delta_R + f_{UPW} \times \delta_{UPW} + f_{SW} \times \delta_{SW} = \delta_S \tag{7}$$

$$f_R \times Cl_R + f_{UPW} \times Cl_{UPW} + f_{SW} \times Cl_{SW} = Cl_S \tag{8}$$

For groundwater:

$$f_R + f_{SW} = 1 \tag{9}$$

$$f_R \times Cl_R + f_{SW} \times Cl_{SW} = Cl_G \tag{10}$$

Where *f* represents the contribution ratio of each end member to the final mixtures (*f<sub>R</sub>* – local ratio of rainfall, *f<sub>SW</sub>* – ratio of seawater, and *f<sub>UPW</sub>* – ratio of upstream freshwater). The notation  $\delta$  and Cl denote the stable isotope of oxygen-18 (‰) and chloride concentration (mg/L), respectively, considered as conservative tracers.

In this study, the selected end members include R for rainwater (with  $\delta_{18}O = -9.29\text{‰}$  and Cl = 0.99 mg/L), SW for seawater (with  $\delta_{18}O = -0.82\text{‰}$  and Cl = 16020.59 mg/L), both collected during a field survey in 2017, and UPW for average upstream fresh surface water (with  $\delta_{18}O = -7.20\text{‰}$  and Cl = 6.75 mg/L).

### 3. Results and discussion

#### 3.1. Tempo-spatial variation of salinity

The statistical analysis of hydro-geochemistry was presented in Table 1.

**Table 1.** Statistical summary of hydrochemical parameters of surface water and groundwater in the study area.

Parameters	pH	EC	TDS	Na	K	Ca	Mg	Cl	HCO <sub>3</sub>	SO <sub>4</sub>	NO <sub>3</sub>
<i>Surface water in dry season (N=38)</i>											
Min.	6.32	133.10	86.52	20.43	1.67	9.36	0.66	26.28	67.10	11.45	0.72
Averg.	7.49	9669.27	6285.02	1748.51	60.64	56.00	212.48	3015.84	93.14	838.12	67.14
Max.	8.04	24400.00	15860.00	4877.02	255.06	174.34	625.91	9354.58	186.65	2667.21	234.40
STD	0.44	6718.23	4366.85	1410.23	56.06	40.61	172.97	2462.84	23.55	705.66	81.56
<i>Surface water in rainy season (N=41)</i>											
Min.	6.57	13.48	8.76	1.84	1.34	0.68	0.05	0.99	7.32	0.31	0.34
Averg.	7.34	937.37	609.29	151.10	12.40	17.31	21.49	253.82	76.06	33.61	3.48
Max.	7.77	13320.00	8658.00	2632.92	98.98	111.39	334.46	4642.72	136.07	536.06	54.13
STD	0.27	2189.41	1423.12	429.23	20.68	16.87	53.70	758.53	19.22	86.41	8.98
<i>Groundwater in dry season (N=137)</i>											
Min.	6.27	116.70	75.86	16.88	0.52	1.44	1.28	2.85	14.03	1.83	0.27
Averg.	7.26	1307.59	849.94	178.27	22.06	35.41	33.08	189.62	318.30	76.03	5.65
Max.	8.68	11710.00	7611.50	2407.00	149.35	475.29	455.53	3705.75	779.23	474.30	59.48
STD	0.47	1380.76	897.50	260.64	21.64	43.57	42.66	421.99	156.80	81.96	9.29
<i>Groundwater in rainy season (N=140)</i>											
Min.	6.12	129.80	84.37	33.10	2.38	0.11	0.03	5.27	16.48	0.02	0.11
Averg.	7.32	2216.79	1440.91	459.95	15.79	65.91	69.21	741.18	329.98	178.24	15.59
Max.	8.55	21200.00	13780.00	8535.80	278.81	970.48	1290.06	16970.45	649.25	3239.46	264.18
STD	0.48	3327.06	2162.59	1164.28	30.14	151.53	157.18	2385.36	148.50	405.71	45.11

Note: EC is expressed in  $\mu\text{S/cm}$ , all chemical compositions were in mg/L

In general, hydrogeochemical data reveal distinctive characteristics between surface water and groundwater. The pH values for both surface water and groundwater ranged from 6.12 to 8.68, with an average and standard deviation of  $7.32 \pm 0.45$ , indicating primarily alkaline nature. Surface water salinity exhibited significant seasonal variation. For example, the electrical conductivity (EC) values of surface water varied widely between the dry season and the rainy season, with averages and standard deviations of  $9,669.27 \pm 6718.23 \mu\text{S}/\text{cm}$  and  $937.37 \pm 2,189.41 \mu\text{S}/\text{cm}$ , respectively. Similarly, total dissolved solids (TDS) values in surface water displayed substantial seasonal and spatial fluctuations (Fig. 2), ranging from 8.76 to 15,860 mg/l, with mean values and standard deviations of  $1,634.78 \pm 2,654.10 \text{ mg}/\text{l}$  compared to  $609.29 \pm 1423.12 \text{ mg}/\text{l}$  in the dry season (Table 1). Low to slightly saline surface water (TDS = 1,000 – 3,000 mg/l) was observed during the rainy season from the coast up to 60 km inland (Figure 2), possibly due to a high rate of freshwater discharge into the coastal estuary. This contrasts with the dry season, where a higher TDS concentration (moderately saline > 3,000 mg/l) is noted due to seawater intrusion.

On the other hand, groundwater exhibited less seasonal variation in hydro-geochemistry but displayed spatial heterogeneity in salinity based on distance to the sea and well depth. Elevated TDS concentrations (ranging from 3,000 to 14,000 mg/l) were observed not only in coastal groundwater wells with depths less than 100m but also in inland areas located more than 10 km from the coastline. In contrast, deep groundwater wells (depths exceeding 100m) showed relatively lower TDS concentrations. The heightened salinity in coastal groundwater may be attributed to the effects of seawater intrusion into the freshwater body through a complex mechanism, involving saline water infiltration, modern seawater intrusion, and aquifer leaks or upwelling. Furthermore, TDS concentrations in groundwater varied widely due to differences in the magnitudes of mineral dissolution and precipitation within heterogeneous aquifers.

### 3.2. Salinization and freshening processes

Based on a simple fresh-seawater mixing model, the deviation of  $\text{Na}^+$  (called  $\text{Na}_{\text{re-act}}$ ) for all water samples was estimated to investigate the possibility of salinization and freshening processes. Most of water samples experienced freshening process which has a decrease of the fraction of seawater ( $f_{\text{sea}}$ ) in corresponding to an increase of  $\text{Na}_{\text{re-act}}$ . More noticeably, some surface water samples presented higher fraction ratio ( $f_{\text{sea}} > 0.1$ ) and high positive  $\text{Na}_{\text{re-act}}$  value indicated that they might be strongly affected by freshening process as an increase upstream freshwater discharge into the VMD in rainy season. Meanwhile, several groundwater samples have significantly experienced freshening process because of fresh groundwater submarine flow to the sea. Conversely, groundwater and surface samples had high fraction ratio ( $f_{\text{sea}} > 0.1$ ) and more negative value of  $\text{Na}_{\text{re-act}}$  might experience salinization processes.

### 3.3. Mixing ratios between fresh and seawater

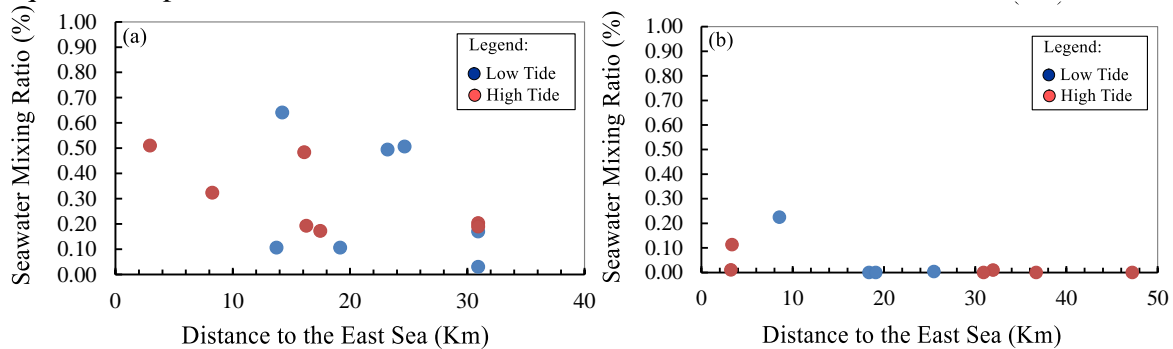
Mixing fresh and seawater strongly effects on chemical characteristics of coastal water resources. The magnitude of mixing between freshwater and seawater depends on local rainfall-runoff process, upstream discharge, and tidal regime. In the study area, surface water showed a seasonal fresh seawater mixing trend. In dry season, for instance, it was found that 29.5% of seawater mixing with freshwater surface water in the study area compared with just only 4.12% in rainy season (Table 2). The mixing ratios also showed wide spatial-temporal variation, depending on the distance to the East Sea and tidal regime. In the high tidal altitude period, the mixing ratios ranged from 50% in the coast to 20% in the inland, approximately 30 km to the sea while in the low tidal peak these ratios were lower than 10% and without mixing, respectively (Figure 2).

The hydraulic connection between river-canal system and the East Sea plays important role in mixing between surface water and seawater. The surface water had high mixing ratios and high chloride concentration along approximately 10 km from Hau River and 30 km to the East Sea. Although some surface water samples were located along the coast, they had very low TDS concentration. It is perhaps due to the operation of sluice gate to prevent seawater intrusion into the canal system along the coast.

**Table 2.** Ratios (in percentage) of each water sources contribution in surface water samples.

Water Sources	Water sources	Min	Average	Max	STD
Surface Water in Dry Season	Rainwater	0.00	0.08	0.66	0.18
	Upstream Water	0.14	0.62	0.94	0.23
	Seawater	0.03	0.30	0.64	0.19
Surface Water in Rainy Season	Rainwater	0.00	0.28	0.80	0.33
	Upstream Water	0.20	0.68	0.99	0.30
	Seawater	0.00	0.04	0.23	0.06
SGW	Rainwater	0.18	0.83	1.00	0.23
	Seawater	0.00	0.17	0.82	0.23
DGW1	Rainwater	0.79	0.99	1.00	0.03
	Seawater	0.00	0.01	0.21	0.03
DGW2	Rainwater	0.84	0.99	1.00	0.02
	Seawater	0.00	0.01	0.16	0.02
DGW3	Rainwater	0.62	0.67	0.72	0.05
	Seawater	0.28	0.33	0.38	0.05
DGW4	Rainwater	0.95	0.98	1.00	0.01
	Seawater	0.00	0.02	0.05	0.01

The mixing processes of freshwater and seawater in coastal water system might be explained via four steps based on chloride and stable isotopes signatures combined with hydrological system. At the beginning, upstream surface water discharges into the study area entering internal river-canal system and discharges directly into the sea in the low peak of tide. After that, a part of upstream water escapes out of river mouth while the remaining one will return into local canal system in the high tidal peak. The returned water then mixes with evaporated water in the large river system as well as local water from semi-isolated canals, aquaculture ponds and wastewater from residential areas.



**Figure 2.** The seawater-mixing ratio with fresh water following distance from the sea and tidal regime in the dry season (a) in the rainy season (b).

Eventually, the mixed water runs into the river mouth and enters the sea afterward before returning to the next circulation. These flow processes establish water circulation in coastal-

estuarial area resulting in complicated mixing processes between upstream water, coastal surface water, local rainfall and seawater. The mixing ratio between groundwater from shallow (SGW) and deep aquifers (DGW1, DGW2 and DGW3) and seawater showed a large variation. For instance, groundwater samples from SGW ( $q_h$ ,  $q_{p3}$ ) aquifers consist of from 0% to 82% seawater while DGW3 ( $n^1_2$ ) aquifer contains from 28% to 38% seawater. More noticeably, the mixing ratio between shallow groundwater and seawater a heterogeneous variety following depth of screened well and distance to the sea (Figure 3). High mixing ratios with seawater was observed up to 30 km inland and until 400 m depth of well, indicating that salt sources might originate from seawater trapped in Holocene period in shallow aquifers and paleo-seawater intrusion in depth aquifers. In contrast, groundwater in remaining aquifers were dominated by freshwater type, indicating the main rainfall recharge source with contribution ratios from 79% to 100%. However, groundwater in some locations contains from 5% to 21% of seawater, reflecting effects of seawater intrusion.

#### 4. Conclusion

This research aims to investigate the dynamics of seawater intrusion in both surface water and groundwater within a coastal tropical area of the Vietnamese Mekong Delta. The inflow of seawater into the coastal surface water system exhibits significant spatial and temporal variations, influenced by the interplay of upstream discharge and tidal-driven flow into the river system. The mixing proportion of seawater is notably higher at 29.5% in the dry season, contrasting with the rainy season's 4.12%. The study highlights the critical role of upstream discharge, contributing 62% and 68% to averting seawater intrusion in the dry and rainy seasons, respectively. Additionally, local rainfall-runoff in the rainy season contributes 28% to coastal surface water.

In contrast, seawater intrusion into the aquifer system demonstrates less seasonal fluctuation but presents a heterogeneous distribution based on well depth and proximity to the sea. The mixing ratio between seawater and groundwater varies widely, ranging from less than 10% in deep groundwater to a maximum of 38% in shallow groundwater. The intricate factors influencing seawater intrusion into the surface water system include upstream discharge, tidal patterns, and sluice gate operations. Conversely, seawater intrusion into the coastal aquifer system is influenced by geological characteristics, original recharge sources, human activities, and natural variations, particularly groundwater pumping and climate change.

While the operation of sluice gates may alleviate the impacts of seawater intrusion, it introduces significant water pollution to the area. Due to limited hydro-geochemical data in this region, the study emphasizes the necessity for further investigations to comprehend the interrelationships between surface and groundwater. Establishing a safe yield for sustainable groundwater use and management is crucial, taking into account factors such as the effects of seawater intrusion, especially considering the potential impacts of climate change.

**Author contribution statement:** Conceived and designed the experiments; Analyzed and interpreted the data; contributed reagents, materials, analysis tools or data; manuscript editing: T.A.N.; Performed the experiments; contributed reagents, materials, analyzed and interpreted the data, wrote the draft manuscript: T.A.N., V.T.H.T.; Revised and improved the manuscript: C.T.V.

**Acknowledgements:** The author would like to thank Dr. Tran Dang An for supporting the data and giving useful comments for improving the manuscript.

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