

## Tracking the migration and distribution of Caspian Kutum (*Rutilus kutum*, Kamenskii, 1901) along the southern coastline of the Caspian Sea: Using stable isotope analysis

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#### Article

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## Abstract

Evaluating management strategies for fish catch development requires knowledge of movement patterns and their spatial distribution. The Caspian kutum (*Rutilus kutum*, Kamenskii, 1901) is an important commercial species throughout the southern coasts of the Caspian Sea. Stable isotope ratios are powerful indexes that simplify the understanding of the migration of aquatic animals. This research determined the stable isotope ratio of carbon ( $\delta^{13}$ C) and nitrogen ( $\delta^{15}$ N) for *Rutilus kutum* movement at 10 sites along the southern coastline of the Caspian Sea from January to December 2017. Spatial and temporal variations in stable isotope values of the coastal communities in the Caspian Sea remain poorly understood. These findings suggest that individual variation, but with a strong overall decline in  $\delta^{13}$ C and increasing in  $\delta^{15}$ N and  $\delta^{13}$ C/ $\delta^{15}$ N ratio with age, too. These isoscpes showed that the  $\delta^{15}$ N ratio increased and  $\delta^{13}$ C decreased from the east (Gomishan) to the west (Astara).

## 1. Introduction

The Caspian Sea is one of the most important closed seas in the world because of its unique natural environment, geographical location, extent, and the existence of biological and non-biological reserves (Karpinsky et al., 2005) and shows fluctuations at much shorter time scales than the world's oceans (Kroonenberg et al., 2008). The annual freshwater input into the sea is about 300 km3 by 130 rivers with various sizes and flows (Saleh et al., 2018). The general current of surface water in the south of the Caspian Sea is from west to east (Bohluly et al., 2018) which is influenced by wind direction, coastline morphodynamic, and bottom topography (Kostianoy et al., 2019). During the past several decades, the environmental condition of the Caspian Sea has changed significantly in response to the impacts of several factors, such as anthropogenic pressures, fluctuations in sea level, and introductions of non-native species (Pourang et al., 2005; Fazli et al., 2013).

In recent decades, technological advances in tools such as electronic tags for animal tracking (Hussey et al., 2017; Hays et al., 2019), chemical tracer analyses (Pethybridge et al., 2018), and 'meta data' analysis (Edgar et al., 2016; McCauley et al., 2015) greatly expanded our ability to understand of fish biology, feeding behavior, population dynamics, movement patterns, and behavior. By combining these technologies with traditional field research, we can better understand the ecological and environmental factors that influence fish populations, ultimately leading to more effective management practices and conservation efforts.

By utilizing Chemical measurements, such as stable isotope analysis (SIA), researchers can better understand the ecological and environmental factors that influence fish populations, ultimately leading to more effective management practices and conservation efforts (Finlay et al., 2008). Isotope ratios in organism tissues can serve as inherent markers of habitat because the environmental isotope ratios of one or more elements typically differ among geographic locations (Bowen, 2010). Specifically, stable isotope ratios of carbon ( $\delta^{13}$ C) and nitrogen ( $\delta^{15}$ N) are powerful tools for assessing fish dispersion and ecological relationships (Rodgers and Wing, 2008; Green et al., 2012). Isotopic fractionation ( $\Delta$ ) refers to the difference in the isotope ratio (e.g., <sup>15</sup>N/<sup>14</sup>N or <sup>13</sup>C/<sup>12</sup>C) between a source and a product, such as animal tissue. By measuring isotopic fractionation, researchers can gain insights into the underlying biogeochemical processes that influence the isotopic composition of an organism's tissues, ultimately leading to a better understanding of its ecological and environmental context (Peterson and Fry, 1987).

The fundamental aim of ecology is to determine the mechanisms that govern the influence of environmental factors, particularly temperature and anthropogenic effects, on population dynamics (i.e., the number, biomass, and distribution of species) and the coexistence of species within natural ecosystems (Thompson et al., 2016). By utilizing SIA and other tools, researchers can gain a more comprehensive understanding of how these factors impact marine ecosystems and the organisms that inhabit them. Marine ecologists face a significant challenge in quantifying the scale and magnitude of dispersal among populations (Cowen et al., 2007).

Movement, which is the shift in the spatial location of an individual over time, is driven by processes that operate across various spatial and temporal scales. This movement is a strategy organisms use to locate suitable breeding or feeding habitats, move towards or away from conspecifics, or simply relocate (Preisler et al., 2013; Nathan et al., 2008). Studying the movement patterns of geographically mobile animals is crucial for effective resource management, as highlighted by Peebles and Hollander (2020). Understanding the reasons behind how and why species move between accessible habitats in different regions and at different times is vital in determining the success of stock management efforts, as emphasized by Evans et al. (2015). By gaining a more comprehensive understanding of animal movement, we can make informed decisions about the appropriate management strategies required to maintain healthy and sustainable populations of these species.

Migratory behaviors are observed in a wide range of terrestrial and aquatic species, involving varying life habitats, distances traveled, migration frequency, and timing (Secor et al., 2020). Understanding animal movement is crucial to comprehending population dynamics, predator-prey relationships, nutrient and energy fluxes within food webs, and managing human-animal interactions (Truman et al., 2017). Furthermore, wildlife conservation planners aim to maintain connectivity between designated nature reserves over large geographic areas (Kays et al., 2015). By studying migratory patterns and behaviors, researchers can gain insights into the ecological and environmental factors that influence animal movement, as well as develop effective conservation strategies for preserving wildlife populations and their habitats. Understanding the migration ecology of a species is essential for effective management and assessment, as the stock structure of the species is influenced by both regional and seasonal movement (Kawazu et al., 2020). When organisms move from one habitat to another, their diet shifts, causing a change in the isotopic content of their tissues (Herzka, 2005). If the isotopic composition of available food sources differs following the move to a new habitat, this change will gradually be reflected in the organism's tissues (Hesslein et al., 1993). Stable isotopes, such as the carbon stable isotope ratio  $(\delta^{13}C)$  and nitrogen stable isotope ratio  $(\delta^{15}N)$ , are essential tools for studying food sources, habitats, and trophic dynamics in ecological communities (Bearhop et al., 2004; Layman et al., 2007). These tracers have a wide range of applications in fields such as geology, ecology, archeology, climatology, and

plant biology, as well as in metabolic flux analyses, proteomics, structural biology, pharmacology, and drug design (Lehmann, 2016). Historically, radioisotopes were exclusively used for such purposes, but stable isotopes have replaced them in many fields (Yang, 2016). Understanding the turnover rates of stable isotopes in a consumer species' tissues is crucial for accurately interpreting their isotopic ecology (Boecklen et al., 2011). In many stable isotope studies of fish, white muscle is the tissue of choice (Trueman et al., 2012).

The Caspian kutum, a member of the Cyprinidae family, is an endemic anadromous fish found in the brackish water habitats of the Caspian Sea and its freshwater branches (Gharedaashi et al., 2013). This species is mainly distributed in the southern part of the Caspian Sea, accounting for more than 60% of total catchments in the Iranian region (Abdolhay et al., 2010), and is rarely found in other areas of the Sea, such as the North part and Volga River (Afraei Bandpei et al., 2010). The Caspian kutum migrates to rivers for spawning during thermal changes and water currents (Shikhshabekov, 1979; Abdoli, 1999). However, recent years have seen a significant decline in the wild stocks of Caspian kutum due to various factors, including overfishing, industrial and agricultural sewage influx, sand mining, and unfavorable conditions for natural spawning (Valipour and Maghsoodieh Kohan, 2018).

In this study, we utilized the temporal variation of carbon and nitrogen stable isotopes to examine distribution patterns and species habitat scales, with the aim of inferring migratory movement histories of Caspian kutum populations. Stable isotope analysis provides a powerful approach for reconstructing migratory histories of individual fish, offering valuable insights into life history variability, habitat use, and trophic interactions. Specifically, our study sought to use isotopic signatures in Caspian kutum scales to achieve three main objectives: (1) identify migratory contingents; (2) assess the extent of movements across the southern coasts of the Caspian Sea; and (3) examine whether these characteristics could be used to understand ecological connectivity among *R. kutum* populations along the southern coasts of the Caspian kutum migratory patterns and informing effective long-term conservation strategies for this species.

## 2. Materials and Methods

# 2.1. Study area

The study was conducted in ten sites along the southern coast of the Caspian Sea: Gomishan, Khajeh Nafas, Miyankaleh, Sari, Babolsar, Nowshahr, Tonekabon, Anzali, and Astara (Fig. 1.). Seasonal environmental parameters for ten regions on the southern coast of the Caspian Sea were obtained from the Iran meteorological organization. Extracted environmental variables were Chlorophyll a concentration (Chl a;  $\mu$ g/m3) and Sea surface temperature (SST; °C), which were observed by the National Aeronautics and Space Administration (NASA) Moderate Resolution Imaging Spectrodiometer's Aqua sensor).

# 2.2. Sample collection

To explore the movement of fish communities over a large scale (the southern coasts of the Caspian Sea), collection sites monthly from the local catches of the commercial fishery from January 2016 to March 2017. The fish obtained from each location were from the same age/ cohort by choosing fish of similar length. Fish were euthanized by an overdose of eugenol (200 ppm) followed by a sharp blow on the head, measured and muscle tissue was extracted in the field. All were immediately placed in vials in liquid nitrogen containers for transport to Gorgan University of Agricultural Sciences and Natural Resources where they were held at -80 °C freezer.

## 2.3. Sample preparation and analysis

For the examination of the  $\delta^{13}$ C and  $\delta^{15}$ N isotopic values of Caspian kutum dorsal muscle tissues, samples were obtained from 230 specimens estimated to be 3–4 years old, with a mean (± SD) total length of 71.1 cm (± 11.7) and mean (± SD) weight of 1876.4 g (± 903.4). Samples were cleaned individually for 5 min with distilled water and freeze-dried for 48 h, ground and homogenized to a uniform consistency using an automatic agate mortar (Retsch, Germany). Weighted all samples (ca. 0.6–0.7 mg) were loaded into 3.5 × 5 mm tin capsules for stable isotope analysis. The reference material used was an internal standard of known relation to the international standards of Vienna Pee Dee belemnite for carbon isotopes and atmospheric nitrogen for nitrogen isotopes. Standard samples were run repeatedly in each sequence. Standard deviations within reference samples were less than 0.2‰ for carbon and nitrogen in each sequence.

Nitrogen and carbon contents, as well as nitrogen ( $\delta^{15}N$ ) and carbon ( $\delta^{13}C$ ) stable isotopic composition, were analyzed using an Isotope Ratio Mass Spectrometer interfaced with an Elemental Analyzer (EA-IRMS). Isotope  $\delta$  values are presented as the per mil (‰) differences from international reference materials. Stable isotope analysis was conducted by the Marine Ecology within Ocean and Earth Science, National Oceanography Centre Southampton at the University Of Southampton, United Kingdom. The stable isotope ratios are calculated as deviation standards following the formula:

 $\delta^{15}$ N or  $\delta^{13}$ C = ((R <sub>sample</sub>- R<sub>standard</sub>)/ R<sub>standard</sub>) ×10<sup>3</sup>

Where  $R_{sample}$  and  $R_{standard}$  are the heavy-to-light isotope ratios (<sup>15</sup>N:<sup>14</sup>N and <sup>13</sup>C:<sup>12</sup>C) of the samples and standards, respectively (Fry, 2006). The international standard for nitrogen is atmospheric N<sub>2</sub> and for carbon is a marine limestone called Peedee Belemnite (Fry, 2006).

# 2.4. Data analysis

As the majority of the sample sets with two well-known Kolmogorov-Smirnov and Shapiro Wilk normality tests examine variables (a significance level of 0.05) were used to check differences between groups. To establish whether  $\delta^{13}$ C and  $\delta^{15}$ N differed significantly between sites, we used Mann-Whitney U tests with Bonferroni corrected p values (P < 0.05). Temporal variations in  $\delta^{13}$ C and  $\delta^{15}$ N values in fish species between different sites and seasons were tested by one-way analysis of variance (ANOVA). We then used Dunkan's honest significant difference (HSD) multiple comparison post hoc test to distinguish significant

differences between ages and locations. Pearson's correlation (r) was undertaken to assess the correlation between  $\delta^{13}$ C and  $\delta^{15}$ N values between the length and age or weight of individuals. All data are demonstrated as mean ± standard deviation. Statistical analyses were performed using the software Microsoft Excel, SPSS (version 24 for Windows), and R (R Core Team, 2020). Maps and figures were created in R using the package 'ggplot2' (Wickham, 2016).

**Approval for animal experiments**: All experimental protocols were approved by the Gorgan University of Agricultural Sciences and Natural Resources (GUASNR/96003041). All experiments were performed in accordance with relevant guidelines and regulations. Our reporting of research involving animals follows the recommendations of the ARRIVE guidelines (Animals in Research: Reporting In Vivo Experiments) guidelines. https://doi.org/10.1371/journal.pbio.1000412.

## 3. Result

# 3.1. Spatial and temporal variation in SST and OC

Our comparisons are based on data collected at the station located on the southern coasts of the Caspian Sea. The lowest mean OC  $(1.23 \pm 0.2\mu g/m3)$  was recorded in Gomishan (summer), while the highest mean OC  $(4.77 \pm 0.8\mu g/m3)$  was recorded in Anzali (winter). Mean temperature ranged from 6.94  $\pm$  0.2 °C in Astara (winter) to  $30.85 \pm 2.5$  °C in Khajeh-Nafas (summer). (Two- way ANOVA) (Figs. 2 and 3).

## 3.1. Stable isotope analysis

Isotope ratios were measured in Marine Ecology within Ocean and Earth Science, National Oceanography Centre Southampton at the University of Southampton, United Kingdom. Descriptive statistics for stable isotope ( $\delta^{13}$ C and  $\delta^{15}$ N) results by the assemblage as a whole are presented in Table 1. A visual display of all stable isotope results combined and organized chronologically suggests oscillations in ranges of both isotope ratios (Fig. 4), as well as in the relationship between  $\delta^{13}$ C and  $\delta^{15}$ N values (see Table 1); statistical analysis of all specimens confirms this pattern Boxplot showed the distribution of  $\delta^{13}$ C and  $\delta^{15}$ N ratios in different stations. Results of  $\delta^{13}$ C and  $\delta^{15}$ N analyses of fish tissue from ten stations of the Caspian kutum we sampled are presented in Table 1. Overall  $\delta^{13}$ C values for fish samples ranged from a minimum of -20.2 ± 0.5‰ (Astara) to a maximum of -19.76 ± 0.5‰ (Gomishan). In Astara  $\delta^{13}$ C were significantly less than those in Gomishan, Khajeh-Nafas and, Nowshahr (ANOVA, n = 231, p > 0.05).

 $\delta^{15}$ N values for Caspian kutum on the southern coasts of the Caspian Sea ranged from a minimum of -7.78 ± 0.5‰ (Gomishan) to a maximum of 9.15 ± 0.5‰ (Astara). In Astara  $\delta^{15}$ N were significantly more than in other stations (ANOVA, n = 231, p > The carbon Carbon to nitrogen (C:N) mass ratio ranged from 3.19 to 3.37 (mean = 3.27, SD = 0.13; Table 1).

Table 1Comparison of mean ± SD stable carbon and nitrogen isotopic compositions between stations in the<br/>southern coasts of the Caspian Sea

Station	Ν	Total Length (cm) Min, Max	δ <sup>13</sup> C (‰)	δ <sup>15</sup> N (‰)	C:N mass ratio
Gomishan	20	35.9, 62.8	-19.76 ± 0.5 <sup>a</sup>	7.78 ± 0.5 <sup>d</sup>	3.19 ± 0.09 <sup>d</sup>
Khajeh-Nafas	8	35.8, 43	-19.77 ± 0.3 <sup>a</sup>	7.81 ± 0.2 <sup>d</sup>	3.21 ± 0.06 <sup>cd</sup>
Miyankaleh	22	34.6, 59	-19.86 ± 0.5 <sup>ab</sup>	8.00 ± 0.7 <sup>d</sup>	3.30 ± 0.17 <sup>abc</sup>
Sari	22	36, 48.9	-19.90 ± 0.3 <sup>ab</sup>	8.02 ± 0.7 <sup>d</sup>	3.28 ± 0.12 <sup>abcd</sup>
Babolsar	25	37, 49.5	-19.98 ± 0.8 <sup>ab</sup>	8.16 ± 0.7 <sup>cd</sup>	$3.22 \pm 0.10^{bcd}$
Nowshahr	32	29, 50.2	-19.81 ± 0.5 <sup>a</sup>	8.33 ± 0.8 <sup>cd</sup>	$3.25 \pm 0.12^{bcd}$
Tonekabon	26	34, 62	-20.01 ± 0.6 <sup>ab</sup>	8.65 ± 1.1 <sup>bc</sup>	$3.37 \pm 0.2^{a}$
Roudsar	25	32.5, 51	$-20.04 \pm 0.4^{ab}$	8.69 ± 0.7 <sup>bc</sup>	3.30 ± 0.17 <sup>abc</sup>
Anzali	28	29, 47.7	$-20.02 \pm 0.4^{ab}$	9.15 ± 0.9 <sup>ab</sup>	3.31 ± 0.14 <sup>ab</sup>
Astara	22	31, 54.5	$-20.2 \pm 0.5^{b}$	9.40 ± 0.9 <sup>a</sup>	3.31 ± 0.12 <sup>ab</sup>

Spatially continuous isoscapes are illustrated by smooth changes in isotope ratios across the southern coasts of the Caspian Sea, giving continuous isotopic gradients along map transects. These isoscpes showed that  $\delta^{13}$ C and  $\delta^{15}$ N ratios increased from the east (Gomishan) to the west (Astara) (Fig. 5).

To extend our analyses beyond a single population and consider Caspian kutum isotopic patterns on a broader scale, bulk values of  $\delta^{13}$ C and  $\delta^{15}$ N were plotted from Caspian kutum samples collected the southern coast of the Caspian Sea (Fig. 6). The results indicated broad patterns of population clustering between stations. Within these strong groupings, we noted that all specimens sampled from stations clustered together. The bulk SIA values from each region haven't a relatively broad range.

It seems that the samples in Gamishan and Astara stations, by separating stations, have two different populations. The samples of Sari, Babolsar and Nowshahr in the east of the Caspian Sea and the samples of Rudsar, Anzali in the west of the Caspian Sea are more mixed together. Probably, samples from the west and east of the Caspian Sea are mixed due to the samples from Tonekabon. The results showed that some Gamishan fish migrated from the east part of the sea to Anzali. Some samples belonging to Miankala-Sari are from the Gamishan place and some fish are from the west of the sea. Samples from Babolsar to Rudsar-Anzali have migrations from east to west and vice versa. Furthermore, the Caspian kutum from Astara to Babolsar also offshore migrate. So, the samples of these regions were not observed in Rudsar-Anzali, Gamishan and Miankale-Sari samples (Fig. 6).

For Caspian kutum ontogenetically, was employed to estimate the contribution of different age groups, with individuals grouped into four age classes. When age 2-5 fish were analyzed, there were significant differences in fish age (ANOVA One-Way). The  $\delta^{15}$ N ratio showed a comparatively greater proportion of the numbers of Caspian kutum at age 5, and fewer fish at age 2. For the  $\delta^{13}$ C ratio, the greatest proportion showed at ages 3 and 5. These results reveal considerable individual variation, but with a strong overall decline in  $\delta^{13}$ C and increasing in  $\delta^{15}$ N and  $\delta^{13}$ C/ $\delta^{15}$ N ratio with age (Table 2,).

Table 2 Comparison of mean ± SD stable carbon and nitrogen isotopic compositions between ages in Caspian kutum							
Age	δ <sup>13</sup> C	δ <sup>15</sup> N	δ <sup>13</sup> C/δ <sup>15</sup> N				
2	-19.76 ± 0.36 <sup>b</sup>	$7.65 \pm 0.45^{c}$	3.23 ± 0.11 <sup>b</sup>				
3	-19.82 ± 0.39 <sup>a</sup>	8.07 ± 0.87 <sup>bc</sup>	3.26 ± 0.12 <sup>ab</sup>				
4	-19.97 ± 0.47 <sup>ab</sup>	$8.52 \pm 0.92^{b}$	3.28 ± 0.14 <sup>ab</sup>				
5	-20.16 ± 0.82 <sup>a</sup>	9.12 ± 0.70 <sup>a</sup>	3.33 ± 0.19 <sup>a</sup>				

	2	$-19.76 \pm 0.36^{\circ}$	$7.65 \pm 0.45$
	3	-19.82 ± 0.39 <sup>a</sup>	8.07 ± 0.87
	4	-19.97 ± 0.47 <sup>ab</sup>	8.52 ± 0.92

### Conclusion

Using stable isotope compositions are being utilized to study the migratory behavior of fish populations by several researchers (Trueman et al., 2012; Redding et al., 2020; Ghosh et al., 2022; Sheehan et al., 2022). In summary, this study quantified the movement of the *Rutilus kutum* community on the southern coasts of the Caspian Sea, while movement varied between sites, it was not limited to species that move to reproduce. Our results are not consistent with the hypothesis that a single stock unit of the Caspian kutum occurs on the Southern coasts of the Caspian Sea. In recent years, stable isotopes have increasingly received attention for their ability to provide complementary information (García-Vernet et al., 2022). This is a common result in studies of this type conducted of the Caspian kutum, and the fluctuations have been usually attributed to migratory movements that implied crossing different isotopic baselines, seasonal variation in feeding patterns and diet composition, or a combination of both (Eisenmann et al., 2016; Eerkes- Medrano et al., 2021).

Nowadays, the utilization of marine biological resources is increasing due to the increase in the world population. Meanwhile, environmental changes are one of the effective factors in the exploitation of these resources (Behrenfeld et al., 2006). The environmental changes, sea surface temperature (SST), and chlorophyll a, are widely used for biochemical and biological analysis and description of the life cycle of oceans and seas. Changes in water temperature are diving significant effects on the biological conditions of many marine organisms that have Consequences for marine ecosystems (Fazli et al., 2022). Knowledge of marine aquatic species distribution is one of the most important agents for understanding the effects of climate and other environmental factors on population ecology (Asante et al., 2017). The environmental parameters include season changes, geographical location, weather, wind, nutrition, depth, light, temperature, water salinity, etc (Froese, 2004) and have a significant effect on the catch and migration of the Caspian kutum (Patimar, 2008). With the increasing temperature in March and April, there have been more migration, because adult fishes come to the coasts for spawning. Spawning of South Caspian kutum occurs at a temperature of 9 in April (Afraei Bandpei et al., 2010). Differences in spawning peak and migration months are due to geographical location and water temperature changes.

The biggest challenge is investigating how environmental parameters are closely linked to other factors and play different roles during migration. The environmental variables are often interrelated, making it difficult to identify which variables are most important in influencing migration. For example, there is a strong relationship between photoperiod and water temperature in moderate climates.

Furthermore, the relative importance of each environmental parameter may depend on the local habitat specification which occurs during the migration (Torregroza-Espinosa et al., 2021). Despite these challenges, several studies have been shown repeatedly that a few environmental factors affect fish migration. Water temperature can trigger and synchronize migratory activity in fish. This situation can occur under two conditions. First, in heterogeneous thermal environments; for a specific population, the temperature may overstep the range of thermal tolerance. Hence, the fish are forced to quest for a new suitable thermal habitat. Second, the population's thermal needs may change. As an example, the optimal temperature for growth may not be suitable for reproduction. Nonetheless, the temperature is known as a coordinator factor during reproduction migration. This is true, especially since it happens in a relatively short time- migrations (Bernal, 2011).

Using Bayesian stable isotope mixing models provided valuable insights into permit ecology and conservation (Brownscombe et al., 2022). Quinzan *et al.* (2016) studied the influence of the environment on the migration pattern of Atlantic pomfret (*Brama brama*) in North-eastern Atlantic waters; results show the key factor of this species' migration is a strong influence of temperature at 200 m depth along with the upwelling in the Galician (NW Iberian) waters. Length alternation distributions indicate an increase in size between successive seasons supporting the hypothesis that migration is a feeding strategy and a return to tropical waters of origin for spawning. The present study shows that the temperature of the water is a key variable in appointing migration patterns of the Caspian kutum.

Maruyama et al. (2001) investigated Change in stable nitrogen isotope ratio in the muscle tissue of a fluvial-lacustrine amphidromous goby, *Rhinogobius* sp., in the Lake Biwa natural system. Researchers expressed that because the  $\delta^{15}N$  of age-0 + fish greatly decreased after their upstream migration from the lake, about 80% of the variation in  $\delta^{15}N$  was ascribed to growth. The results of this study showed that the half-shift period for  $\delta^{15}N$  was estimated as being longer than 1 month with the portion of growth and metabolic turnover in the field. Growth is primarily responsible for isotopic variations in fish muscle and was needed to examine the role of metabolic turnover using slow-growing fish. The present results showed; the  $\delta^{15}N$  values vary between stations and increase from Gomishan to Astara. Hence, the relationship between muscle  $\delta^{15}N$  and fish size with environmental parameters and life history can be

used as a proxy to study the preferred habitat, in defining the preference of individuals tracking the migratory behavior with an age of *Rutilus kutum* across its life history stages. MacKenzie et al. (2012) study showed that marine location can be inferred from the carbon isotope composition of Atlantic salmon (*Salmo salar*) which changes with sea surface temperature. In addition, applying this technique can be used to identify the place of any pelagic animals for which tissue archives and matching records of sea surface temperature are available. Hoffman et al. (2011) identified trophic linkages of larval fish in Lake Superior coastal habitats using naturally occurring differences in the stable isotope ratios of nitrogen ( $\delta^{15}$ N) and carbon ( $\delta^{13}$ C). For all species,  $\delta^{13}$ C was increasingly negative with increasing weight. Trends in  $\delta^{15}$ N with increasing weight varied among species; an increase, decrease, and no change in  $\delta^{15}$ N was observed. The result showed fish obtained a constant signature after a 10-fold gain in body mass, implying their tissue was at isotopic equilibrium with their diet. MacKenzie et al. (2014) tested the temporal stability of isoscapes of carbon and nitrogen isotopes across the North Sea over a ten-year period; using jellyfish tissues. They showed that hydrodynamic and biogeochemical processes control the distribution of carbon and nitrogen isotope values, and thus that the underlying isoscapes are temporally stable.

Trueman et al. (2017) assess the accuracy and precision that can be obtained through such as carbon and nitrogen dietary-isotope-based location methods known as individual locations, and herring with wellunderstood population-level distributions in the North Sea. In this study, the spatial patterns of  $\delta^{13}$ C and  $\delta^{15}$ N ratio in the pelagic food webs of the North Sea are well explained with a combination of river nutrient inputs, hydrology, and winter regeneration of sediment nutrients.

We expected a better level of isotopic separation across the southern coast of the Caspian Sea and a larger role of  $\delta$ 13C in explaining differences between stations, but our results are consistent with the hypothesis that a single stock unit of *Rutilus kutum* occurs on the Southern coasts of the Caspian Sea. In total, clear reasons explaining the lack of separation between stations be that fish move freely and regularly between sub-basins in the Caspian Sea and maps didn't show the  $\delta^{13}$ C differences between stations are not obviously pronounced; but, we observed  $\delta^{13}$ C ratio was increased from Gomishan to Astara. That  $\delta^{15}$ N value appears to be unaffected by Caspian kutum size or diet probably reflects the relatively uniform length of fish in the study. Nevertheless, increasing the  $\delta^{15}$ N ratio from Gomishan to Astara is likely due to riverine inputs and physical circulation patterns in the Caspian Sea. The eastern parts of the Caspian Sea coasts are comparatively low compared to central and western draining into proper with significant agricultural and sewage inputs; which can explain the differing  $\delta^{15}$ N patterns observed between stations.

The results suggest that the muscles  $\delta^{13}$ C and  $\delta^{15}$ N composition which ordinally reflects environmental conditions can prove to be useful to analyze the groups of the Caspian kutum population across its distribution. Furthermore, fish movement patterns in the Caspian Sea are not comprehensively identified. As this study, the temperature and chlorophyll-a effects are factors that cause an impact on movement behavior in the *Rutilus kutum* of the southern coasts of the Caspian Sea. Whereas, it is necessary that a

repeated sampling at larger spatial and temporal levels and coverage of different growth stages should be assumed so that results can be used with confidence to make suitable management strategies. In fact, more study is required to reduce the scientific gap of the migration routes and to resolve the indeterminacy in recognizing the niche or habitat of their maximum growth or aquaculture. Conservation strategies and stock assessment can be evaluated with isotopic signatures to raise the fish species' production and total size increase.

## Declarations

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#### Data availability:

The datasets generated and analyzed during the current study are not publicly available due to the funding responsibility but are available from the corresponding author upon reasonable request.

### References

- Abdolhay, H. A., Khalijah, D. S., Pourkazemi, M., Shapor, S. S., Rezvani, S., Satar, M. K. A. and Hosseinzadeh Sahafi, H. 2010. Morphological studies of Mahisefid (*Rutilus frisii kutum*, Kamensky, 1901) from selected rivers in the southern Caspian Sea. Iran J Fishe Sci. (9):1-18.
- 2. Abdoli, A. 1999. The inland water fishes of Iran. Natural and Wild Life Museum of Iran, Tehran, Iran. 198–200. (In Persian).
- 3. Afraei Bandpei, M. A., Mashhor, M., Abdolmaleki, S. and El-Sayed, A. M. 2010. Population dynamics of Caspian Kutum, *Rutilus frisii kutum* (Cyprinidae) in southern Caspian Sea, Iran. Cybium. 34(3): 285–292.
- Asante, C. K., Jardine, T. D., Van Wilgenburg, S. L. and Hobson, K. A. 2017. Tracing origins of waterfowl using the Saskatchewan River Delta: Incorporating stable isotope approaches in continentwide waterfowl management and conservation. The Condor: Ornithological Applications. 119(2): 261-274.
- 5. Bearhop, S., Adams, C. E., Waldron, S., Fuller, R. A. and MacLeod, H. 2004. Determining trophic niche width: a novel approach using stable isotope analysis. Journal of animal ecology. 73(5): 1007-1012.
- 6. Behrenfeld, M. J. and Boss, E. 2006. Beam attenuation and chlorophyll concentration as alternative optical indices of phytoplankton biomass. Journal of Marine Research. 64(3): 431-451.
- 7. Bernal, D. 2011. Physiological Specializations of Different Fish Groups: Pelagic Fishes. Di dalam: Farrell AP, Cech JJ, Richards JG, Stevens ED, editor. Encyclopedia of fish physiology: from genome to

environment. (1): 1887-1902.

- 8. Boecklen, W. J., Yarnes, C. T., Cook, B. A. and James, A. C. 2011. On the use of stable isotopes in trophic ecology. Annual Review of Ecology, Evolution and Systematics. 42(1):411-440.
- 9. Bohluly, A., Esfahani, F. S., Namin, M. M. and Chegini, F. 2018. Evaluation of wind induced currents modeling along the Southern Caspian Sea. Continental Shelf Research. (153): 50-63.
- 10. Bowen, G. J. 2010. Isoscapes: spatial pattern in isotopic biogeochemistry. Annual review of earth and planetary sciences. 38(1):161-187.
- 11. Brownscombe, J. W., Shipley, O. N., Griffin, L. P., Morley, D., Acosta, A., Adams, A. J., Boucek, R., Danylchuk, A. J., Cooke, S. J. and Power, M. 2022. Application of telemetry and stable isotope analyses to inform the resource ecology and management of a marine fish. Journal of Applied Ecology. 59(4):1110-1121.
- 12. Cowen, R. K., Gawarkiewicz, G., Pineda, J., Thorrold, S. R. and Werner, F. E. 2007. Population connectivity in marine systems an overview. Oceanography. 20(3):14-21.
- Edgar, G. J., Bates, A. E., Bird, T. J., Jones, A. H., Kininmonth, S., Stuart-Smith, R. D. and Webb, T. J. 2016. New approaches to marine conservation through the scaling up of ecological data. Annual Review of Marine Science. (8):435-461.
- Eerkes-Medrano, D., Aldridge, D. C. and Blix, A. S. 2021. North Atlantic minke whale (*Balaenoptera acutorostrata*) feeding habits and migrations evaluated by stable isotope analysis of baleen. Ecology and evolution. 11(22):16344-16353.
- 15. Eisenmann, P., Fry, B., Holyoake, C., Coughran, D., Nicol, S. and Bengtson Nash, S. 2016. Isotopic evidence of a wide spectrum of feeding strategies in Southern Hemisphere humpback whale baleen records. PLoS One. 11(5): p.e0156698.
- Evans, M. L., Hori, T. S., Rise, M. L. and Fleming, I. A. 2015. Transcriptomic responses of Atlantic Salmon (*Salmo salar*) to environmental enrichment during juvenile rearing. PloS one. 10(3): p.e0118378.
- 17. Fazli, H., Daryanabard, G. R., Abdolmaleki, S. and Bandani, G. 2013. Stock management implication of Caspian kutum (*Rutilus frisii kutum* Kamensky, 1901) in Iranian waters of the Caspian Sea. Ecopersia. 1(2): 179-190.
- Fazli, H., Khoshghalb, M.R.B. and Abdolmaleki, S. 2022. A Multi-species modeling approach to consider the effects of environmental parameters on Caspian sturgeon fishes stock status. Regional Studies in Marine Science. (56): p.102666.
- 19. Finlay, J.C., Kendall, C., Michener, R. and Lajtha, K. 2008. Stable isotopes in ecology and environmental science. Stable Isotope tracing of temporal and spatial variability in organic matter sources to freshwater ecosystems. 283-324.
- Froese, R., Stern-Pirlot, A., Winker, H. and Gascuel, D. 2008. Size matters: how single-species management can contribute to ecosystem-based fisheries management. Fisheries Research. 92(2-3): 231-241.
- 21. Fry, B., 2006. Stable isotope ecology. New York: Springer. (521): 317p.

- 22. García-Vernet, R., Aguilar, A., Zafra, J., Víkingsson, G., Halldórsson, S. D. and Borrell, A. 2022. Sulfur stable isotope ratios provide further insight into movements of the fin whale, an oceanic long-range migrant. Marine Ecology Progress Series. (692):185-194.
- Gharedaashi, E., Nekoubin, H., Imanpoor, M. R. and Taghizadeh, V. 2013. Effect of copper sulfate on the survival and growth performance of Caspian Sea kutum, *Rutilus frisii kutum*. SpringerPlus. 2(1): 1-5.
- 24. Ghosh, P., Rajawat, V. S., Nazir, A., Banerjee, Y., Nath, A. K. and Sakthivel, T. 2022. Stable isotope on hilsa shad (*Tenualosa ilisha*) otoliths revealed migratory behavior of a population found in Hooghly River, West Bengal, India. Environmental Biology of Fishes. 1-10.
- Green, B. C., Smith, D. J., Grey, J. and Underwood, G. J. 2012. High site fidelity and low site connectivity in temperate salt marsh fish populations: a stable isotope approach. Oecologia. 168(1): 245-255.
- 26. Hays, G. C., Bailey, H., Bograd, S. J., Bowen, W. D., Campagna, C., Carmichael, R. H., Casale, P., Chiaradia, A., Costa, D. P., Cuevas, E. and de Bruyn, P. N. 2019. Translating marine animal tracking data into conservation policy and management. Trends in Ecology and Evolution. 34(5): 459-473.
- 27. Herzka, S. Z. 2005. Assessing connectivity of estuarine fishes based on stable isotope ratio analysis. Estuarine, Coastal and Shelf Science. 64(1): 58-69.
- 28. Hesslein, R. H., Hallard, K. A. and Ramlal, P. 1993. Replacement of sulfur, carbon, and nitrogen in tissue of growing broad whitefish (*Coregonus nasus*) in response to a change in diet traced by δ<sup>34</sup>S, δ<sup>13</sup>C, and δ<sup>15</sup>N. Canadian Journal of Fisheries and Aquatic Sciences. 50(10): 2071-2076.
- 29. Hoffman, J. C., Cotter, A. M., Peterson, G. S., Corry, T. D. and Kelly, J. R. 2011. Rapid stable isotope turnover of larval fish in a Lake Superior coastal wetland: implications for diet and life history studies. Aquatic Ecosystem Health & Management. 14(4): 403-413.
- 30. Hussey, N. E., Hedges, K. J., Barkley, A. N., Treble, M. A., Peklova, I., Webber, D. M., Ferguson, S. H., Yurkowski, D. J., Kessel, S. T., Bedard, J. M. and Fisk, A. T. 2017. Movements of a deep-water fish: establishing marine fisheries management boundaries in coastal Arctic waters. Ecological Applications. 27(3): 687-704.
- 31. Karpinsky, M. G., Shiganova, T. A. and Katunin, D. N. 2005. Introduced species. The Caspian Sea Environment. 175-190.
- 32. Kawazu, M., Tawa, A., Ishihara, T., Uematsu, Y. and Sakai, S., 2020. Discrimination of eastward trans-Pacific migration of the Pacific bluefin tuna *Thunnus orientalis* through otolith  $\delta^{13}$ C and  $\delta^{18}$ O analyses. Marine Biology. 167(8):1-7.
- 33. Kays, R., Crofoot, M. C., Jetz, W. and Wikelski, M. 2015. Terrestrial animal tracking as an eye on life and planet. Science. 348(6240): p.aaa2478.
- 34. Kostianoy, A. G., Ginzburg, A. I., Lavrova, O. Y., Lebedev, S. A., Mityagina, M. I., Sheremet, N. A. and Soloviev, D. M. 2019. Comprehensive satellite monitoring of Caspian Sea conditions. In Remote sensing of the Asian Seas. Springer, Cham. 505-521.

- 35. Kroonenberg, S. B., Kasimov, N. S. and Lychagin, M. Y. 2008. The Caspian Sea, a natural laboratory for sea-level change. Geography, environment, sustainability. 1(1):22-37.
- 36. Layman, C. A., Arrington, D. A., Montaña, C. G. and Post, D. M. 2007. Can stable isotope ratios provide for community-wide measures of trophic structure? Ecology. 88(1): 42-48.
- 37. Lehmann, S. B., Braun, D. R., Dennis, K. J., Patterson, D. B., Stynder, D. D., Bishop, L. C., Forrest, F. and Levin, N. E. 2016. Stable isotopic composition of fossil mammal teeth and environmental change in southwestern South Africa during the Pliocene and Pleistocene. Palaeogeography, Palaeoclimatology, Palaeoecology. (457): 396-408.
- 38. McCauley, L. A., Anteau, M. J., van der Burg, M. P. and Wiltermuth, M. T. 2015. Land use and wetland drainage affect water levels and dynamics of remaining wetlands. Ecosphere. 6(6): 1-22.
- MacKenzie, K. M., Longmore, C., Preece, C., Lucas, C.H. and Trueman, C. N. 2014. Testing the longterm stability of marine isoscapes in shelf seas using jellyfish tissues. Biogeochemistry. 121(2): 441-454.
- 40. MacKenzie, K. M., Trueman, C. N., Palmer, M. R., Moore, A., Ibbotson, A. T., Beaumont, W. R. and Davidson, I. C. 2012. Stable isotopes reveal age-dependent trophic level and spatial segregation during adult marine feeding in populations of salmon. ICES Journal of Marine Science. 69(9):1637-1645.
- 41. Maruyama, A., Yamada, Y., Rusuwa, B. and Yuma, M., 2001. Change in stable nitrogen isotope ratio in the muscle tissue of a migratory goby, *Rhinogobius* sp., in a natural setting. Canadian Journal of Fisheries and Aquatic Sciences. 58(11):2125-2128.
- 42. Nathan, R., Getz, W. M., Revilla, E., Holyoak, M., Kadmon, R., Saltz, D. and Smouse, P. E. 2008. A movement ecology paradigm for unifying organismal movement research. Proceedings of the National Academy of Sciences. 105(49):19052-19059.
- 43. Patimar, R., 2008. Some biological aspects of the sharpnose mullet *Liza saliens* (Risso, 1810) in Gorgan Bay-Miankaleh wildlife refuge (the southeast Caspian Sea). Turkish Journal of Fisheries and Aquatic Sciences. 8(2): 225-232.
- 44. Peebles, E. B. and Hollander, D. J. 2020. Combining isoscapes with tissue-specific isotope records to recreate the geographic histories of fish. In Scenarios and responses to future deep oil spills. Springer, Cham. 203-218.
- 45. Peterson, B. J. and Fry, B. 1987. Stable isotopes in ecosystem studies. Annual review of ecology and systematics. 293-320.
- 46. Pethybridge, H. R., Choy, C. A., Polovina, J. J. and Fulton, E. A. 2018. Improving marine ecosystem models with biochemical tracers. Annu. Rev. Mar. Sci. 10(1): 199-228.
- Pourang, N., Tanabe, S., Rezvani, S. and Dennis, J. H. 2005. Trace elements accumulation in edible tissues of five sturgeon species from the Caspian Sea. Environmental Monitoring and assessment. 100(1):89-108.
- 48. Preisler, H. K., Ager, A. A. and Wisdom, M. J. 2013. Analyzing animal movement patterns using potential functions. Ecosphere. 4(3): 1-13.

- 49. R. Core. Team. 2020. R: A language and environment for statistical computing. R Foundation for Statistical Computing.
- 50. Redding, S. G., Cooper, L. W., Castonguay, M., Wiernicki, C. and Secor, D. H. 2020. Northwest Atlantic mackerel population structure evaluated using otolith δ180 composition. ICES Journal of Marine Science. 77(7-8): 2582-2589.
- 51. Rodgers, K. L. and Wing, S. R. 2008. Spatial structure and movement of blue cod *Parapercis colias* in Doubtful Sound, New Zealand, inferred from δ<sup>13</sup>C and δ<sup>15</sup>N. Marine Ecology Progress Series. (359): 239-248.
- 52. Saleh, A., Hamzehpour, A., Mehdinia, A., Bastami, K. D. and Mazaheri, S. 2018. Hydrochemistry and nutrient distribution in southern deep-water basin of the Caspian Sea. Marine pollution bulletin. (127): 406-411.
- 53. Secor, D., O'Brien, M., Rothermel, E., Wiernicki, C. and Bailey, H. 2020. Movement and habitat selection by migratory fishes within the Maryland Wind Energy Area and adjacent reference sites. Sterling (VA): US Department of the Interior, Bureau of Ocean Energy Management, Office of Renewable Energy Programs. OCS Study BOEM. (30): 109p.
- Sheehan, K. L., Dorr, B. S., Clements, S. A., Christie, T. W., Hanson-Dorr, K. C., Rush, S. A. and Davis, J. B. 2022. Predicting consistent foraging ecologies of migrating waterbirds: Using stable isotope and parasite measurements as indicators of landscape use. Ecological Indicators. (140): p.109038.
- 55. Shikhshabekov, M. M. 1979. On the Reproductive Biology of Kutum *Rutilus frisii kutum* (Kamensky), Asp *Aspius aspius* (L.), Vimba *Vimba vimba* persa (Pallas) and Rudd *Scardinius erythrophthalmus* (L.) in the Water Bodies of Dagestan. Vopr. Ikhtiol. (19):495-502.
- Thompson, M. S., Bankier, C., Bell, T., Dumbrell, A. J., Gray, C., Ledger, M. E., Lehmann, K., McKew, B. A., Sayer, C. D., Shelley, F. and Trimmer, M. 2016. Gene-to-ecosystem impacts of a catastrophic pesticide spill: testing a multilevel bioassessment approach in a river ecosystem. Freshwater Biology. 61(12): 2037-2050.
- 57. Torregroza-Espinosa, A. C., Restrepo, J. C., Escobar, J., Pierini, J. and Newton, A. 2021. Spatial and temporal variability of temperature, salinity and chlorophyll-a in the Magdalena River mouth, Caribbean Sea. Journal of South American Earth Sciences. (105): p.102978.
- 58. Trueman, C. N., MacKenzie, K. M. and Palmer, M. R. 2012. Identifying migrations in marine fishes through stable-isotope analysis. Journal of Fish Biology. 81(2):826-847.
- Trueman, C. N., MacKenzie, K. M. and St John Glew, K. 2017. Stable isotope-based location in a shelf sea setting: accuracy and precision are comparable to light-based location methods. Methods in Ecology and Evolution. 8(2): 232-240.
- 60. Valipour, A. and Maghsoodieh Kohan, H., 2018. The Study of Stocking Density Effect on Growth and Survival of Fingerlings Kutum, *Rutilus kutum*, with Caspian Sea Water. Journal of Applied Ichthyological Research. 6(1):131-144.
- 61. Wickham, H., Chang, W. and Wickham, M. H. 2016. Package 'ggplot2'. Create elegant data visualisations using the grammar of graphics. Version. 2(1):1-189.

62. Yang, J., Chen, H., Nie, Y., Zhang, W. and Wang, K. 2016. Spatial variability of shallow soil moisture and its stable isotope values on a karst hillslope. Geoderma. (264):61-70.

### **Figures**



#### Figure 1

Map of the southern coast of the Caspian Sea and depicting geographic position of the Caspian kutum sampling location sites



Seasonal changes of Sea surface temperature in southern coasts of the Caspian Se



Seasonal changes of OC in southern coasts of the Caspian Sea



Boxplot (mean± standard deviation) of muscle  $\delta^{13}$ C and  $\delta^{15}$ N ratios measured for Caspian kutum between different stations in the southern coasts of the Caspian Sea. Points represent outliers.



Kriging map of the southern coast of the Caspian Sea: Spatial variation of nitrogen and carbon isotopes from Caspian kutum between stations. Spatial patterning of an environmental property underlying variation in Caspian kutum isotope ratios in the southern coasts of the Caspian Sea. (A) The spatial distribution of  $\delta^{15}$ N isotope ratios between stations; (B) The spatial distribution of  $\delta^{13}$ C isotope ratios between stations.



Relationship between carbon and nitrogen values for *Rutilus kutum* collected from different stations along the southern coasts of the Caspian Sea. Sites including: G: Gomishan, K: Khajeh-Nafas, M: Miyankaleh, S: Sari, B: Babolsar, N: Nowshahr, T: Tonekabon, R: Roudsar, Z: Anzali and A: Astara. A special pattern was not shown for this species.