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Seasonal Environmental Variability Drives Physiological Adaptations in the Thick-Lipped Grey Mullet (*Chelon labrosus*) of the Southern Mediterranean Sea

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***Chelon labrosus*; Environmental Variables; Mediterranean Sea; Physiological Adaptations; Thick-Lipped Grey Mullet**

Abstract: This study investigated the impact of seasonal variations in temperature, salinity, and dissolved oxygen in Ras Al-Tin lagoon (Eastern Libya) on hematological, liver, and kidney functions of Chelon labrosus. Surface water parameters ranged as follows: temperature (11–23°C), salinity (4.67–23‰), conductivity (11–27 mS/cm), and dissolved oxygen (9.5–11.9 mg/l). Blood samples from 40 fish collected seasonally in 2019 revealed significant physiological changes. Hematological results showed peak WBCs ($185.32 \pm 18.27 \times 10^3/\mu l$) and RBCs ($2.75 \pm 0.25 \times 10^6/\mu l$) in autumn, while summer exhibited the lowest HGB (5.86 ± 1.56 g/dl) and HCT (19.96 ± 6.04%). MCV and MCH peaked in spring (171.10 ± 4.80 fl; 61.15 ± 1.15 pg), whereas MCHC was highest in winter (36.04 ± 1.57 g/dl). Liver enzymes GOT and GPT spiked in autumn (229.75 ± 40.33 U/L; 23.25 ± 2.66 U/L), while LDH surged in spring (1331.00 ± 86.00 U/L). Kidney function markers urea and creatinine peaked in spring (39.00 ± 5.00 mg/dl; 0.75 ± 0.05 mg/dl), whereas uric acid was elevated in autumn and summer. These findings suggest that moderate temperatures in spring and autumn correlate with heightened metabolic and reproductive activity, supported by increased enzyme levels and hematological shifts. Seasonal extremes (summer/winter) likely induced stress, reflected in reduced erythrocyte indices and oxygen-carrying capacity. These results align with prior studies linking environmental variability to physiological responses in fish, emphasizing the lagoon's dynamic ecosystem as a driver of C. labrosus health. The study underscores the need for further research on **Libya's understudied aquatic ecosystems and their ecological impacts on native species.**

Introduction

Chelon labrosus (thick-lipped grey mullet) is a euryhaline, benthic detritivore widely distributed across coastal lagoons (e.g., Ras Al-Tin, Libya), estuaries, and shallow marine waters of the Northeast Atlantic and Mediterranean Sea. Adults typically reach 30–60 cm in length and 0.5–3 kg in weight, though larger specimens are occasionally recorded. It thrives in hypersaline to freshwater environments, tolerating significant temperature and salinity fluctuations (Whitfield *et al*., 2012; Koutrakis *et al*., 2004). As a key ecosystem engineer, *C. labrosus* recycles nutrients through detritus consumption, shapes benthic biodiversity by altering sediment composition, and serves as a critical prey species for larger fish, birds, and marine mammals, thereby linking energy transfer across trophic levels. Its ecological and economic importance, coupled with its adaptability, makes it an ideal model for studying physiological responses to environmental changes (Whitfield *et al*., 2012).

Seasonal variations in temperature and salinity critically influence marine organisms, impacting metabolic rates, osmoregulation, and overall health (Boeuf and Payan, 2001). These effects are particularly evident in key organs like the liver and kidney, which govern detoxification and metabolism, and in hematological parameters (e.g., hemoglobin, blood cell counts), which reflect stress responses (Hrubec *et al*., 2000; Witeska, 2013). Ras Al-Tin (Umm Hufayn) in Eastern Libya (Southern Mediterranean Sea), with its pronounced seasonal environmental shifts, provides an optimal setting to study these dynamics in *C. labrosus*, offering insights into Mediterranean ecosystem health.

Despite the species' significance, few studies have examined the combined effects of seasonal temperature and salinity variations on its liver/kidney functions and hematology. This study bridges that gap by analyzing these parameters across four seasons, generating baseline data for fisheries management and environmental monitoring.

Materials and methods

Study site description

The investigation was conducted in Ras Al-Tin Lagoon, Fig. 1a and 1b, (32°36'46"N–32°33'48"N, 23°07'19"E–23°05'29"E), a representative coastal lagoon ecosystem of the southern Mediterranean Sea, locally known as Umm Hufayn Lagoon. Located approximately 80 km east of Derna along the Tobruk corridor, this semi-enclosed water body exhibits distinctive hydrological features resulting from its unique geomorphology. The lagoon occupies a rocky promontory where the Libyan coastline curves southeastward into the Gulf of Bumba, accessible via a secondary road branching northward from the Darnah-Tobruk highway near Umm ar Rzam.

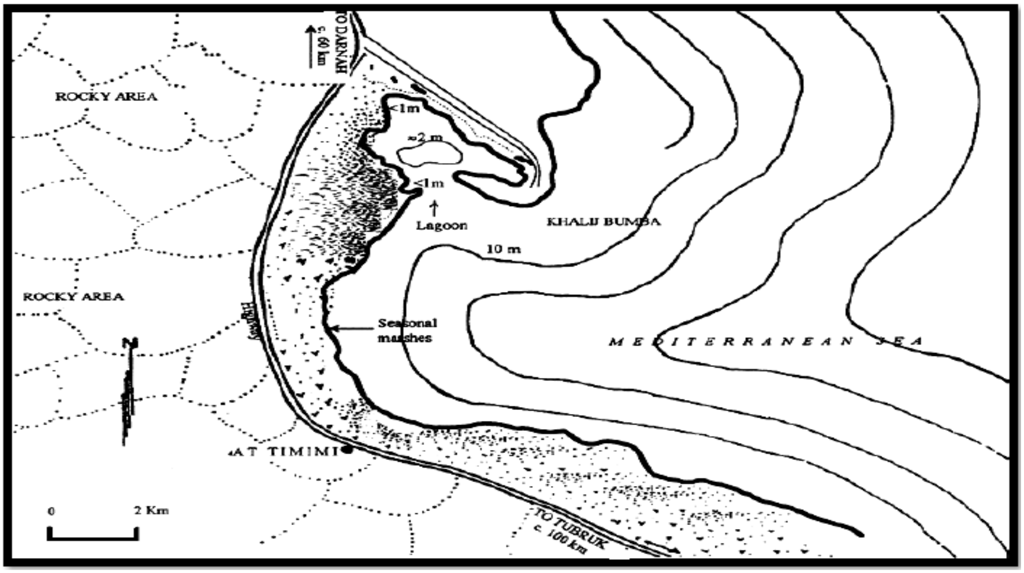

Fig. 1a. Map represents Ras Al-tin (Eastern Libya).

Fig. 1b. Close-up view of Ras Al-tin in Eastern Libya.

The site comprises two principal habitats: (1) a wave-protected sandy beach sheltered by a natural fringing reef barrier that harbors artisanal fishing operations, and (2) a shallow lagoon basin (maximum depth <5 m) connected to the open Mediterranean through a 500-meter-wide tidal inlet. This connection facilitates regular seawater exchange while subterranean freshwater springs along the lagoon's inner margins create localized brackish conditions, as characterized by Abd AL Hamid *et al*. (2017). These dual freshwater-marine inputs create dynamic salinity and temperature gradients that exhibit marked seasonal variation, making this an ideal system for studying euryhaline species adaptation

Water characteristics measurements

Surface water parameters were measured in situ during midday sampling sessions to characterize the ambient conditions during fish collection. Temperature was recorded using a calibrated mercury thermometer, while salinity determinations were made through electrical conductivity measurements taken with a Beckman RS-10 inductive salinometer, and dissolved oxygen was assessed using a digital oxygen meter. These measurements were conducted concurrently with biological sampling to ensure accurate correlation between environmental conditions and physiological data.

Collection of fish and blood samples

The study employed a seasonal sampling design, collecting forty adult specimens of *Chelon labrosus* (Mugilidae) during each of the four seasonal periods (autumn 2019 through summer 2019) from Ras Al-Tin. Blood samples were obtained immediately after capture using sterile techniques, with collection from either the caudal vasculature or via cardiac puncture using 21-gauge needles and disposable syringes, following the protocols established by Hrubec *et al.* (1997). Following extraction, blood samples were transferred to anticoagulant-treated vials and placed on ice for one hour to facilitate clotting. Serum separation was achieved through centrifugation at 14,000 × g for five minutes, with subsequent storage at -80°C to preserve sample integrity until biochemical analysis.

Measuring CBC, hepatic, and renal biomarkers

Laboratory analyses focused on CBC (Complete Blood Count) and key hepatic and renal biomarkers. CBC was established with Siemens automated hematology analyzer. Liver function was assessed through quantification of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), acid phosphatase (ACP), lactate dehydrogenase (LDH), and bilirubin using established spectrophotometric methods (Henry *et al*., 1979; Patton and Crouch, 1977; Pearlman and Lee, 1974). Renal function evaluation included measurements of urea and creatinine concentrations following standardized protocols (Patton and Crouch, 1977).

Statistical analyses

All statistical analyses were performed using SPSS version 15.0 (IBM Corporation). Data are presented as means ± standard error (SE) following verification of normal distribution assumptions. Multivariate analysis of variance (ANOVA) was employed to examine seasonal effects, with post-hoc comparisons conducted using Tukey's HSD test where appropriate. Statistical significance was determined at $\alpha = 0.05$ for all tests.

Results

physicochemical water characteristics surface water parameters in Ras Al-Tin

These parameters exhibited marked seasonal variation (Table 1), with three key parameters showing a consistent winter-summer gradient:

Temperature increased from 11 ± 1.73°C (winter) to 23 ± 0°C (summer)

Salinity rose from 4.67 ± 0.88‰ (winter) to 23 ± 1.15‰ (summer)

Conductivity climbed from 11 ± 1.15 mS/cm (winter) to 27 ± 1.73 mS/cm (summer)

Dissolved oxygen, in contrast, showed an inverse pattern, peaking in autumn (11.9 ± 0.02 mg/L) and reaching its lowest summer concentration (9.5 ± 0.01 mg/L).

Table 1. seasonal variation in surface water characteristics at Ras Al-Tin (mean ± se)

season / parameter	summer (Jun.-Aug.)	autumn (Sep.-Nov.)	winter (Dec.-Feb.)	spring (Mar. – May)
temperature °c	(23 ±0)	(18 ±0)	(11 ±1.73)	(14 ±1.15)
salinity ‰	(23 ±1.15)	(12± 0.58)	(4.67± 0.88)	(9 ±1.732)
conductivity ms/cm	(27±1.73)	(24±1.70)	11±1.15)	(17±1.15)
oxygen mg/l	(9.5±0.01)	(11.9±0.02)	(10.7±0.03)	(11.4±0.01)

Seasonal hematological variation in *Chelon labrosus* (Table 2, and Figs. 2 to 9)

Leukocytes (WBCs): Highest in autumn (185.32 ± 18.27 × 10^3/µL), lowest in summer (171.44 ± 6.12 × 10^3/µL)

Erythrocytes (RBCs): Peak counts in autumn (2.75 ± 0.25 × 10^6/µL), minimal in spring (1.21 ± 0.41 × 10^6/µL)

Hemoglobin (HGB): Maximum in autumn (11.17 ± 0.51 g/dL), minimum in summer (5.86 ± 1.56 g/dL)

Hematocrit (HCT): Highest in autumn (40.77 ± 3.03%), lowest in summer (19.96 ± 6.04%)

Erythrocyte indices:

MCV peaked in spring (171.10 ± 4.80 fL)

MCH and **MCHC** reached maxima in spring (61.15 ± 1.15 pg) and winter (36.04 ± 1.57 g/dL), respectively

Platelets (PLT): Highest in spring (283.00 ± 276.00 × 10^3/µL), lowest in autumn (20.25 ± 3.09 × 10^3/µL)

Table 2. Seasonal hematological parameters of Chelon labrosus (mean ± SE)

	Parameters	Summer	Autumn	Winter	Spring
CBC	**WBCs (10^3 /µl)**	171.44± 6.12^a	185.32± 18.27^a	171.50± 13.85^a	183.85± 32.85^a
	RBCs (10^6 /µl)	1.63± 0.45^{ab}	2.75± 0.25^b	1.55± 0.22^{ab}	1.21± 0.41^a
	HGB (g/dl)	5.86±1.56^a	11.17±0.51^b	8.86±1.14^{ab}	7.45±2.65^{ab}
	HCT (%)	19.96±6.04^a	40.77±3.03^b	24.52±2.80^a	20.90±7.60^a
	MCV (fl)	122.70±6.15^a	149.97±8.50^b	160.64±4.85^b	171.10±4.80^b
	MCH (Pg)	36.50±1.68^a	41.70±4.20^a	57.78±2.78^b	61.15±1.15^b
	MCHC (g/dl)	29.98±1.84^a	27.65±1.41^a	36.04±1.57^b	35.75±0.35^b
	PLT (10^3 /µl)	22.60±17.44^a	20.25±3.09^a	88.60±36.74^{ab}	283.00±276.00^b

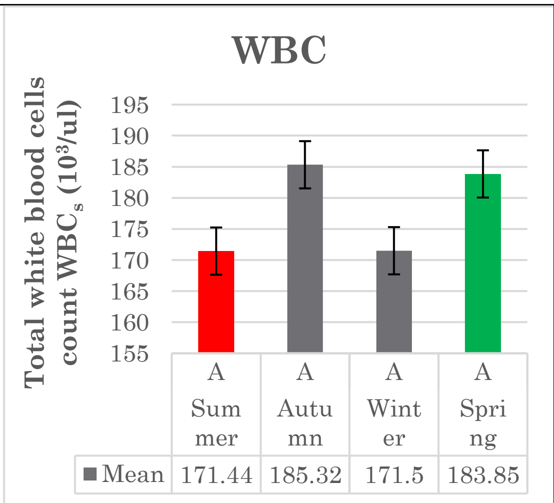

Fig. 2 Effect of seasonal variation on WBCs (10^3 /µl) in *Chelon Labrosus*

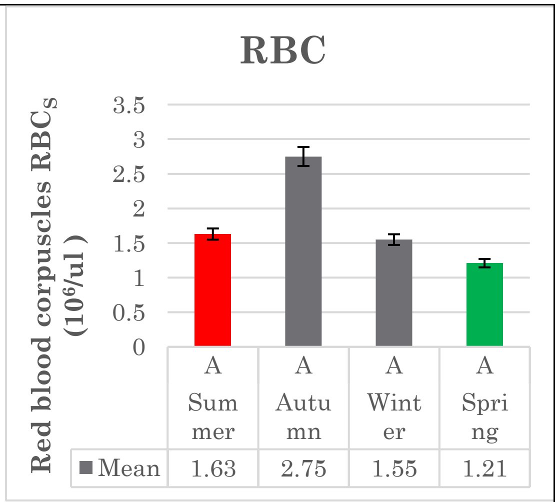

Fig. 3 Effect of seasonal variation on RBCs (10^6 /µl) in *Chelon Labrosus*

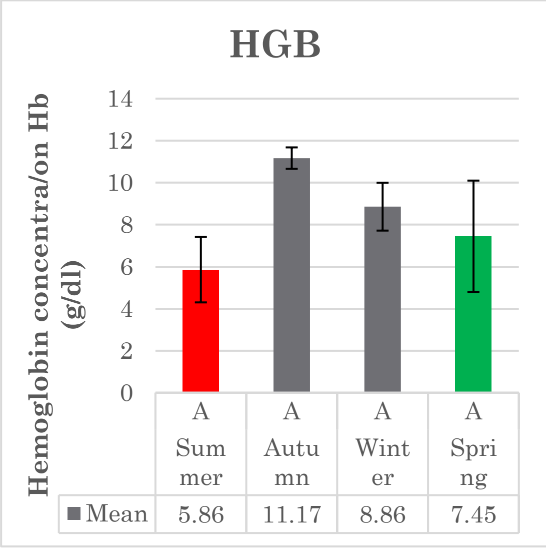

Fig. 4 Effect of seasonal variation on HGB (g/dl) in *Chelon Labrosus*

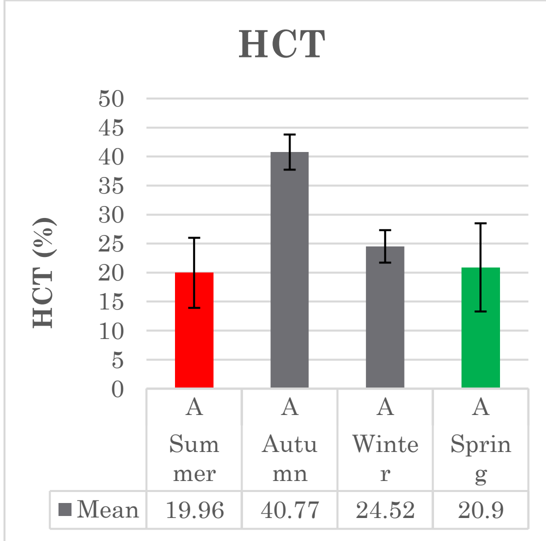

Fig. 5 Effect of seasonal variation on HCT (%) in *Chelon Labrosus*

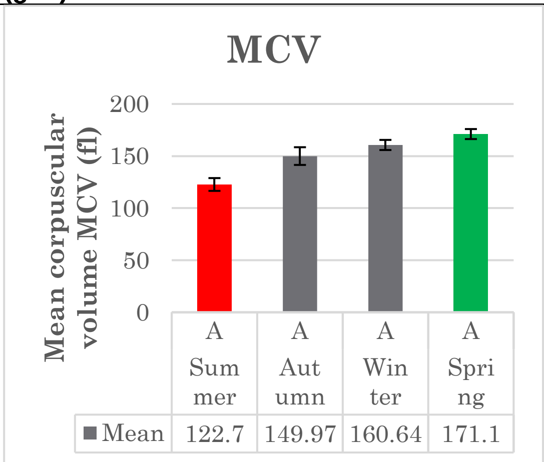

Fig. 6. Effect of seasonal variation on MCV (fl) in *Chelon Labrosus*.

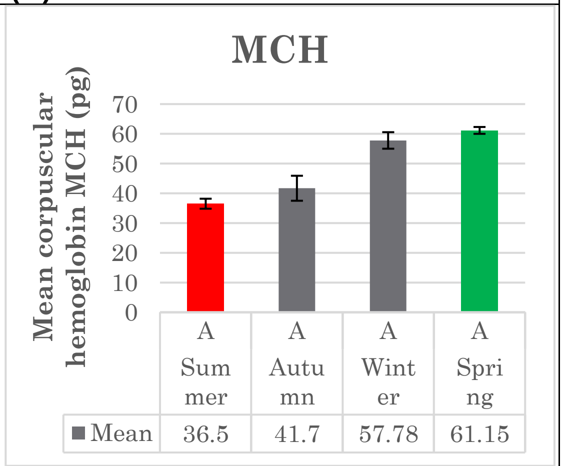

Fig. 7. Effect of seasonal variation on MCH (Pg) in *Chelon Labrosus.*

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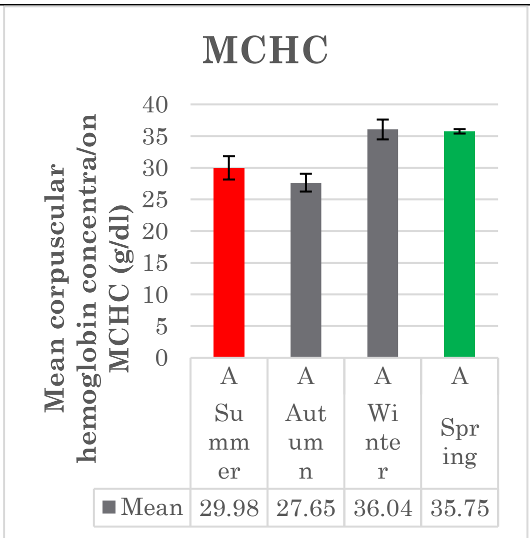

Fig. 8. Effect of seasonal variation on MCHC (g/dl) in *Chelon Labrosus*.

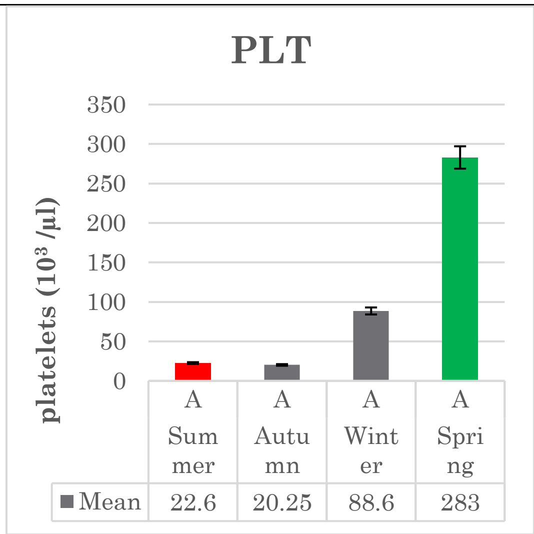

Fig. 9. Effect of seasonal variation on PLT (10^3 /µl) in *Chelon Labrosus*.

Effect of seasonal variation on liver function parameters (GOT, GPT, ALP, LDH) in *Chelon Labrosus*

The assayed GOT and GPT levels increased markedly during autumn compared to winter, while ALP levels remained relatively stable. In contrast, LDH levels peaked during the spring season (Table 3 and Figs. 10–13; mean ± SE).

Table 3: Effect of seasonal variation of liver function parameters (GOT, GPT, ALP, LDH) in *Chelon Labrosus* (mean ± SE; significant at p. < 0.05), means with similar superscripts across rows were not statistically different**.**

	Parameters	Summer	Autumn	Winter	Spring
Liver functions	GOT	130.20 ± 21.4^{a}	229.75 ± 40.33^{b}	90.60 ± 4.99^{a}	101.50 ± 2.50^{a}
	GPT	13.60 ± 0.93^{ab}	23.25 ± 2.66^{c}	18.60 ± 1.21^{bc}	12.00 ± 1.00^{a}
	ALP	86.00 ± 5.37^{a}	89.50 ± 4.48^{a}	83.80 ± 2.37^{a}	95.00 ± 6.00^{a}
General	LDH	373.60 ± 18.4^{a}	309.25 ± 6.02^{a}	359.40 ± 17.80^{a}	1331.00 ± 86.00^{b}

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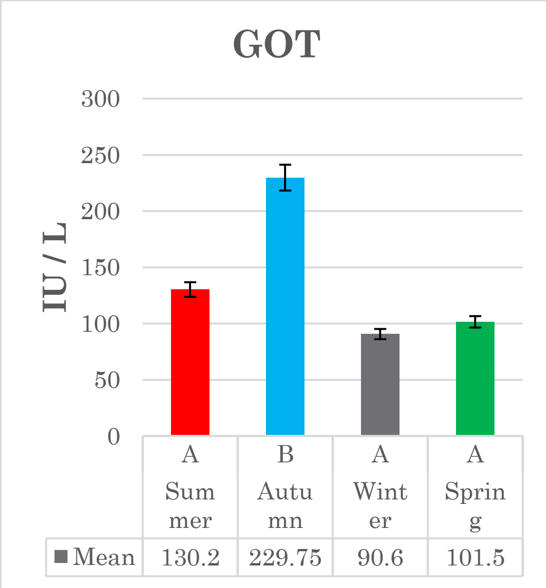

Fig. 10: Effect of seasonal variation on GOT levels in *Chelon labrosus*.

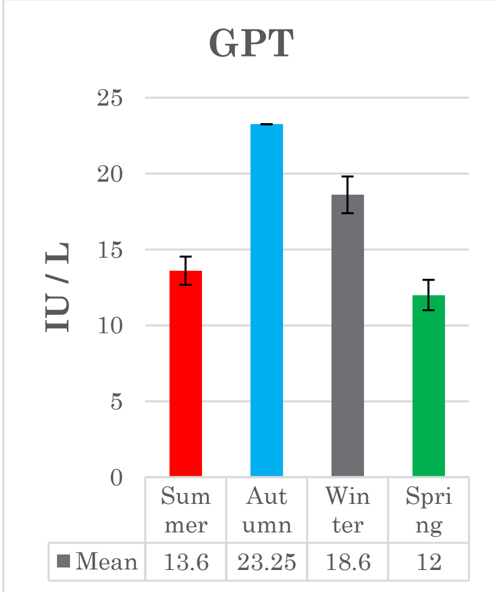

Fig. 11: Effect of seasonal variation on GPT levels in *Chelon labrosus*.

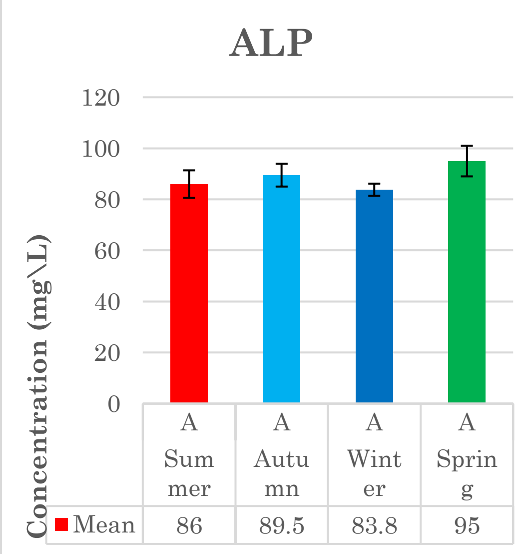

Fig. 12: Effect of seasonal variation on ALP levels in *Chelon labrosus*.

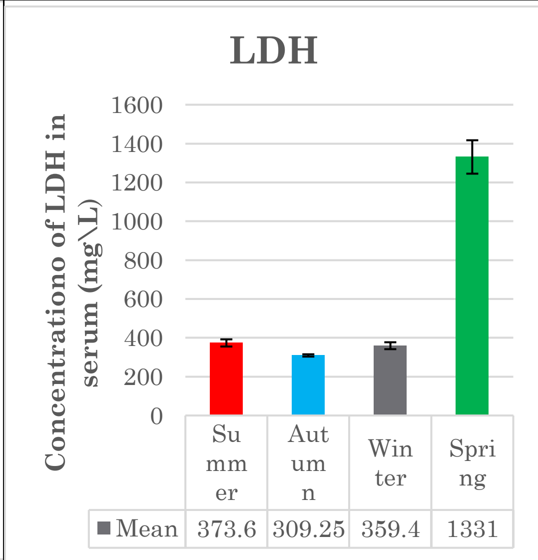

Fig. 13: Effect of seasonal variation on LDH levels in *Chelon labrosus*.

Effects of seasonal variation on kidney function parameters (urea, uric acid, and creatinine) in *Chelon labrosus* (Table 4 and Figs. 14 to 16).

Urea level in spring was significantly higher than in summer, autumn, and winter, which showed no statistical differences.

Uric acid and creatinine levels did not vary significantly across the four seasons.

Table 4: Effect of seasonal variation on kidney function (urea, uric acid, creatinine) in *Chelon labrosus*.

	Parameters	Summer	Autumn	Winter	Spring
Kidney function	Urea (mg/dl)	14.00±1.30[a]	14.50±0.87[a]	11.60±1.21[a]	39.00±5.00[b]
	Uric acid (mg/dl)	3.98±0.44[a]	4.87±0.20[a]	3.24±0.85[a]	3.55±0.65[a]
	Creatinine (mg/dl)	0.48±0.07[a]	0.45±0.06[a]	0.58±0.12[a]	0.75±0.05[a]

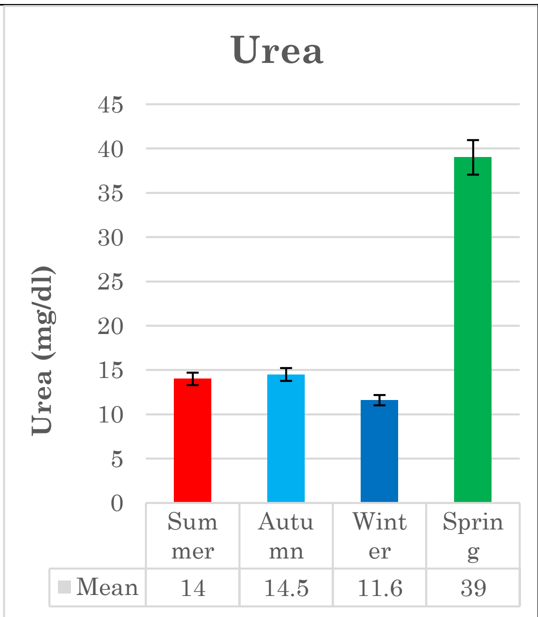

Fig. 14: Seasonal variation in urea (mg/dL) levels in *Chelon labrosus*.

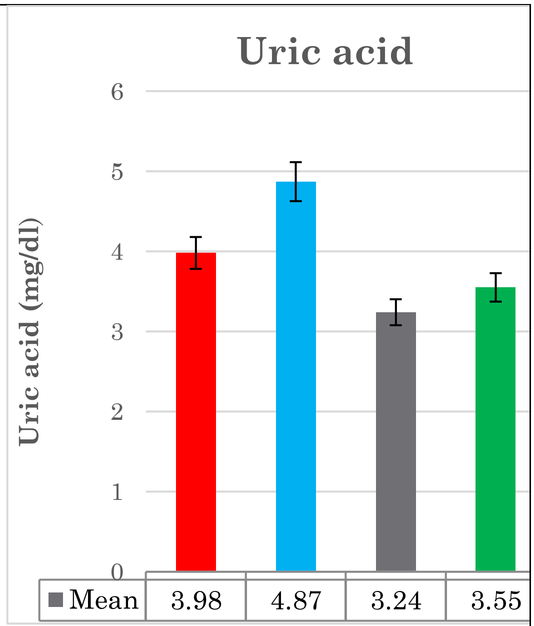

Fig. 15: Seasonal variation in urea (mg/dL) levels in *Chelon labrosus*.

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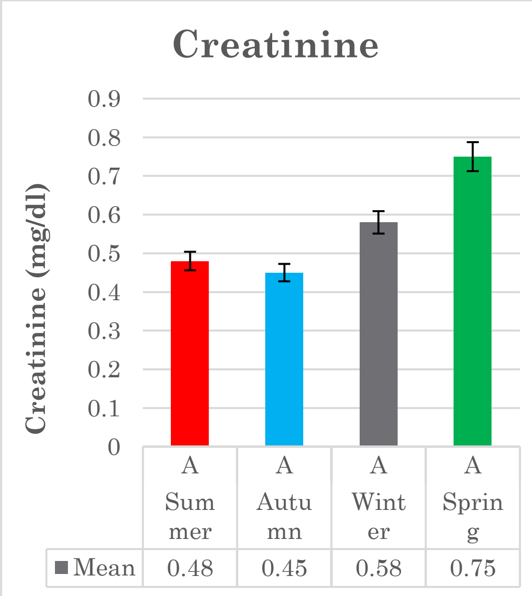

Fig. 16: Seasonal variation in creatinine (mg/dL) levels in *Chelon labrosus*.

Discussion

The study of *Chelon labrosus* in Libyan waters, particularly concerning its physiological responses to environmental changes, has been limited, despite the species' ecological and economic importance. This research aimed to address this gap by examining how seasonal variations in the Ras Al-Tin lagoon (indicative of the Southern Mediterranean Sea) influence the hematology, liver, and kidney functions of this euryhaline fish, providing valuable insights into its adaptability.

Seasonal physiological variations

Our findings indicate that the moderate temperatures (14–18°C) and optimal salinity levels (9–12‰) observed during spring and autumn correlate with heightened liver enzyme activity (GOT, GPT, LDH) and hematological parameters (WBCs, RBCs, HGB). These conditions likely support increased metabolic and reproductive demands. It's also noteworthy that autumn exhibited the highest dissolved oxygen levels (11.9 ± 0.02 mg/L). The higher oxygen availability in autumn - likely due to increased oxygen solubility in the cooler, less saline water - may further explain the observed peak in RBCs and HGB, as greater oxygen-carrying capacity supports active metabolic processes. Similar patterns have been observed in other Mediterranean estuaries, where increased enzyme activity is associated with spawning periods and higher metabolic rates in fish like *Mugil cephalus* (Cardona, 2006; Koutrakis *et al*., 2004). The significant surge in LDH levels during spring (1331.00 ± 86.00 U/L) may be indicative of increased anaerobic metabolism, potentially linked to gonadal maturation, as seen in other fish species (Boeuf and Payan, 2001).

Environmental Stress Responses

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In contrast, the extreme conditions of summer (23°C, 23‰ salinity) were associated with reduced erythrocyte indices (HGB, HCT) and elevated urea levels. These changes suggest that *C. labrosus* experiences osmoregulatory and thermal stress during the summer months. Notably, summer also had the lowest dissolved oxygen concentrations (9.5 ± 0.01 mg/L) - likely due to reduced oxygen solubility in the warmer, higher saline water. Lower DO levels can exacerbate the effects of high temperature and salinity, as oxygen solubility decreases with increasing temperature and salinity, potentially contributing to the observed reduction in oxygen-carrying capacity (HGB, HCT). Research on other fish species, such as *Oreochromis niloticus*, has shown that high salinity can impair oxygen-carrying capacity and renal function, supporting our observations (Hrubec *et al*., 2000). Additionally, the decline in WBCs during winter (171.50 ± 13.85 × 10^3/μL) may point to immunosuppression due to low temperatures (11°C), a phenomenon also observed in *Schizothorax niger* (Witeska, 2013).

Comparative Regional Studies and Ecological Implications

The Ras Al-Tin lagoon's fluctuating salinity (4.67–23‰) highlights the remarkable euryhaline nature of *C. labrosus*, which is consistent with findings in other estuarine environments like those in Portugal (Almeida, 2003; Whitfield *et al*., 2012). However, the lower hemoglobin levels observed in *C. labrosus* during summer (5.86 ± 1.56 g/dL) compared to other Mediterranean mullets under similar conditions suggest potential genetic or habitat-specific adaptations. The interplay between temperature, salinity, and dissolved oxygen is crucial here. The combined stress of high temperature, high salinity, and low DO in summer likely creates a challenging environment for *C. labrosus*. The unique microhabitats created by freshwater inputs and tidal exchange in Ras Al-Tin lagoon are crucial for the species. It is essential to consider the potential impacts of reduced freshwater inflow and increasing salinity fluctuations, driven by climate change, on these lagoon ecosystems, as well as the potential for these changes to alter DO dynamics.

Management and Future Directions

The results of this study have implications for the regional fisheries management of *C. labrosus*. Integrating these findings into management plans can aid in mitigating overharvesting and habitat degradation, ensuring the long-term sustainability of the species, as emphasized by the FAO (2020) guidelines for Mediterranean fisheries. Future research should focus on the genetic diversity of *C. labrosus* populations and the long-term consequences of climate change on Mediterranean lagoon ecosystems. Further investigations into the molecular mechanisms underlying the observed physiological adaptations, including the specific effects of varying dissolved oxygen levels and their interactions with other environmental variables, are also warranted.

Conclusion

Liver and kidney functions, along with other physiological parameters, peaked during spring and autumn, coinciding with moderate temperatures and abundant food resources (seaweeds, phytoplankton, zooplankton) and increased dissolved oxygen. Elevated enzyme activities (e.g., ALP, LDH) during these seasons align with gonadal maturation, increased adrenaline/cortisol secretion, and metabolic demands (Olaoluwa *et al*., 2015; Abdelhafid, 2014).

Seasonal environmental changes directly influence physiological parameters in *Chelon labrosus*, underscoring the need for further research to elucidate adaptive mechanisms.

Conflict of Interest: The authors declare that there are no conflicts of interest.

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