3.1 INTRODUCTION

3.1.1 Mullets

The flathead mullet, scientifically known as *Mugil cephalus* L., is found worldwide in tropical, subtropical, and temperate coastal waters across all major oceans (Briggs 1960; Thomson 1966). This species inhabits diverse marine, estuarine, and freshwater environments, however reproduction takes place exclusively in the marine environment (Thomson 1955). *M. cephalus* is a robustly euryhaline species that can survive in waters ranging from fresh to hyperhaline, as would be expected based on the distribution pattern mentioned above (Wallace 1975; Cardona 2006). The flathead mullet is capable of inhabiting both clear and turbid environments, as well as sandy and muddy habitats. It is also able to thrive in waters with varying levels of dissolved oxygen, as documented by Thomson (1963) and Hoese (1985). This species is able to achieve a high population and large amount of living matter in many areas of its habitat due to its ability to adapt to a wide range of conditions and its feeding behaviour at the bottom of the food chain. These shared attributes have made it a valued species for fishing and aquaculture, particularly in countries like Greece and Taiwan where it holds significant economic importance (Maitland & Herdson 2009). Undoubtedly, the global demand for mullet roe has significantly increased in recent decades, leading to the grey mullet being known as 'grey gold' by fishermen (Hung & Shaw 2006). Since mullet are naturally abundant in temperate, subtropical, and tropical regions, there are several opportunities for mullet breeding, rearing, and farming. They serve as a readily accessible protein supply, which is highly regarded for human nutrition.

The first international symposium on grey mullet aquaculture, which took place in June 1974 in Haifa, Israel, appropriately demonstrated the significance of grey mullets for artificial rearing. Oren (1971,81) posits that *M. cephalus*, a species known for its widespread success, is likely the pioneering marine fish to have been cultivated. It is noticeable that this species has the most potential and is the most appropriate fish for artificial rearing due to its high growth rate, high energy content, high calorific value, and capacity to adjust to changes in food. Mullets are extremely euryhaline fish that have the ability to thrive across a broad spectrum

of salinities, ranging from 0 to 113 (Thomson 1966; Kutty *et al.* 1980). Mullets, being euryhaline, can be introduced into brackish water lagoons and coastal wetlands to enhance fish production (Ravagnan 1992). In order to enhance water quality and establish new fisheries, they can be raised in commercial freshwater fish ponds (Thomson 1966; Pillay 1993) and introduced into freshwater lakes and reservoirs (Thomson 1966).

3.1.2 Proximate Content

Since ancient times, fish has been regarded as a significant source of high-quality protein for human consumption (Weber 2007). Mullet is widely favoured and devoured by individuals across all economic strata due to its reputation as a delicious fish dish and its strong market demand (Brusle 1981). The mullet's diversified feeding habits, reliance on particulate organic matter and detritus, and non-predatory nature contribute to its high nutritional value. Mullet are efficient converters of natural food compared to other species (Whitfield *et al.* 2012).

Seafood offers several advantages, including high protein levels, excellent digestion, and low-fat content (Pigott & Tucker 1990). In general, "proximate composition" refers to the proportion composition of basic components, or macronutrients, such as water, protein, fats, carbohydrates, and minerals (Kumaran *et al.* 2012). An investigation of the seasonally-proximate composition of red mullet in Turkey was conducted by Tulgar & Berik in 2012. They discovered that the typical macronutrient composition was composed of water (75.25%), protein (17.75%), fat (5.25%), ash (1.19%), and carbs (1.01%). Boran & Karaçam (2011) examined the variations in the proximate composition of golden mullet over several seasons. They found that the average content of water was 76.67%, fat was 4.78%, protein was 16.19%, mineral was 1.90%, and carbs were 0.46%. In a study conducted by Kumaran *et al.* (2012), the proximate content analysis of *M. cephalus* from Tamil Nadu was undertaken. The *M. cephalus* had a moisture content of 75.27%, carbohydrate content of 1.2%, lipid content of 2.42%, protein content of 17.56%, and crude ash content of 1.15%. Mohanty *et al.* (2015) conducted a study in Chandipur that examined three distinct species, including *M. cephalus*. The analysis of *M. cephalus* revealed a moisture content of 55%, ash content of 1.5%, protein content of 18%, and lipid content of 4%. The findings

indicate that mullets possess a substantial amount of proteins that are of superior grade. No research on mullet has been conducted in Gujarat, despite the significant amount of mullet production in the region.

Moisture content refers to the quantity of water contained in fish tissue. The determination is made by measuring the reduction in weight when a sample of fish is subjected to the process of drying. The moisture content of fish is essential for comprehending its freshness and preservation properties. Protein content refers to the amount of protein that is found in the tissue of fish. The protein content of fish is a crucial nutritional component that directly affects its dietary value. Lipid content refers to the quantity of fats found in fish. Lipids play a crucial role in storing energy and are crucial for the well-being of humans. The ash content refers to the remaining residue that is left behind once all organic matter in the fish sample has been completely burned. It denotes the presence of inorganic minerals such as calcium, phosphorus, and magnesium. Fish tissue's mineral profile can be inferred from its ash content. Carbohydrate analysis is not commonly performed in fish proximate analysis due to the generally low quantities of carbs found in fish tissues. Nevertheless, the investigation was conducted to verify the carbohydrate content in *M. cephalus*.

The biochemical composition of fish exhibits significant variation, both across different species and within the same species. Differences in parameters can be observed among various parts of the same fish (Stansby 1954) and within the same tissue from different regions of the body (Sherni & Jafri 1978; Mustafa & Jafri 1978; Nair & Radhakrishnan 1988). The fluctuations in the biochemical components are a result of many physiological and other events, including maturation, spawning, and eating (Jacquot 1961).

Many fishes have been found to store nutritional elements in their muscles before their gonads mature. Before spawning occurs, there is a necessary process of metabolic reconfiguration in several tissues. It has been observed that muscle is the main location where these stored components accumulate and are later released (Chandran 1969). The study focused on investigating the seasonal variations in the organic composition of muscle, considering its significance as a storage organ.

The liver is widely recognised as the primary organ responsible for storage in vertebrates. Anukima (1965) observed that the liver's accumulation of fat was adaptively associated with survivability throughout the unfavourable summer season in the White Sea. Navaga, in contrast to Hoar (1953), documented a reduction in liver glycogen levels in the fish *Salmo salar* during the pre-spawning period. Fontaine & Hatey (1953) observed that gluconeogenesis from lipids, and possibly amino acids, played a crucial role in providing the necessary carbohydrates for the muscular activity of migratory salmons. The loss observed in the liver of the fish was nearly equivalent to the accumulation observed in the eggs. Evidently, the liver of fish is involved in both the pre-spawning activity and the overall metabolism of the body. Therefore, the alterations in the biochemical composition of the liver were associated with the changes in the gonads in order to clarify the liver's functional involvement in the animal's breeding cycle.

Baskaran (1993) conducted a comparative analysis of the feeding and reproductive cycles of mullet species in Tamil Nadu viz., *M. cephalus* and *Liza dussumieri* [Planiliza subviridis (Valenciennes 1836)]. The study found that, the hepatic index, gonado-somatic index, and gastro-somatic index of *M. cephalus* were lower than those of *P. subviridis*. *M. cephalus* is significantly larger than *P. subviridis*. Since they have larger muscles than *P. subviridis*, it makes sense that this fish would have lower values for these indices, which are expressed as a percentage of body weight. Maybe because this fish has more muscle on its body than other species, it is a better choice for aquaculture.

Significant contributions to the study of Indian fishes have been made by Chidambaram *et al.* (1952), Sreenivasan & Natarajan (1963), Hasan & Jafri (1964), Rao (1967), Chaturvedi *et al.* (1976), and Pandey *et al.* (1976). Nevertheless, our knowledge regarding the biochemical composition of mullets is restricted (Ghosh & Guha 1934; Marais & Erasmus 1977; Perera & De silva 1977). Hence, this study aims to analyse the fluctuations in moisture, fat, protein, carbohydrate, and ash content in the muscle and liver of *M. cephalus* from the Diu lagoon and Narmada estuary in Gujarat, India.

3.1.3 Aquaculture

In India, where per capita consumption of meat and milk is relatively low, fish serves as an essential source of supplementary nutrition due to the prevalent protein shortage. According to Sivakumaran (2002), protein malnutrition impacts approximately 30-40% of the world's population. Fish is highly regarded for its high digestibility, abundant supply of vital amino acids like lysine and methionine, and distinctive polyunsaturated fatty acids like eicosapentaenoic acid, which have the ability to lower blood cholesterol levels and prevent cardiovascular illnesses. In addition, fish is a rich source of essential vitamins (A, C, D, B-complex, B12) and minerals (calcium, phosphorus, iron, sodium, potassium, magnesium, sulphur). Aquaculture has evolved as an efficient method for producing animal protein, surpassing the production rate of traditional livestock methods. Aquaculture also aids in the recycling of food scraps and home wastes, so making a positive contribution to environmental conservation (Sivakumaran 2002).

Aquaculture, which refers to the controlled farming and care of economically valuable aquatic animals and plants, has notable benefits compared to conventional agricultural or veterinary methods. Sivakumaran (2002) brought attention to the fact that one can raise a tonne of fish or one hundred tonnes of shellfish in the same amount of space required to raise a few hundred kilogrammes of cattle. Sivakumaran (2002) also noted that fish have a food conversion rate that is 1.5 times higher than that of pigs, poultry, or cattle, and twice that of sheep or lambs. As a result, feed-supplied aquaculture produces fish at a continuously higher rate than livestock production. Furthermore, aquaculture contributes to environmental sustainability by offering a productive way to recycle household and agricultural waste. Aquaculture has the potential to promote integrated rural development by creating job opportunities in rural regions and reducing the migration of people to metropolitan areas. However, because to societal difficulties and habitat loss, it is subject to restrictions or prohibitions in some regions.

Aquaculture is a rapidly expanding method of food production on a global scale, seeing an annual growth rate of 9% (Tacon 1996). Developing regions, specifically in Asia, South America, Oceania, and Africa, are witnessing a far higher

pace of growth in aquaculture production compared to developed areas. India, being a leading aquaculture producer, is prioritising the development of brackish water fishing to fulfil the growing fish demand. Brackish water environments are very suitable for aquaculture due to their potential to greatly enhance fish productivity, create employment possibilities, and make beneficial use of otherwise unproductive coastal wetlands and mangroves.

Mullet serve as a base for fishery and aquaculture sectors in many regions across the globe. The FAO (2015) report said that the total production of Mugilidae in 2013 was 700,000 metric tonnes, with 80.2% coming from catch fishery and 19.8% from aquaculture. Asia is the leading producer of mullet, accounting for 70% of the global production. A total of 31 species from 10 genera constitute the main supply of mullet for the fisheries in India, Southeast Asia, and East Asia (Eschmeyer 2014). Mullets are a significant component of the brackish water fishery in India, with a total of 18 species belonging to seven taxa identified (Joshi *et al.* 2018). Eight of these species are part of the commercial catches. The species mentioned include *Mugil cephalus*, *Mugil cunnesius*, *Liza macrolepis*, *Liza parsia*, *Liza tade*, *Ellochelon vaigiensis*, *Valamugil seheli,* and *Rhinomugil corsula* (Luther 1973). The main areas that support mullet fishery are the estuaries of the Ganga, Mahanadi, Godavari, Krishna, and Cauvery rivers, as well as the brackish water lakes of Chilka and Pulicat on the east coast. On the west coast, the estuaries of the Narmada, Tapti, Gulf of Kutch, and backwaters of Kerala, particularly Vembanad Lake, Kayamkulam Lake, and Ashtamudi Lake, are notable locations for mullet fishing (Luther 1973). According to a report, only a small number of the main estuaries on the two coastlines of India have been studied for their biodiversity, out of the approximately 200 estuaries in total (Wafar *et al.* 2011).

Marine and freshwater fishing industries and the cultivation of aquatic organisms, Grey mullet are extensively harvested (Bacheler *et al.* 2005) by various fishing methods including beach seines, gill nets, purse seines, trammel nets, pegged nets, shoreline traps, barrage traps, dip nets, and cast nets (Thomson 1963). The composition of harvested mugilids will fluctuate based on the availability of different taxa in each region. However, *M. cephalus* is frequently a significant species in the catch (Katselis *et al.* 2003; Chaoui *et al.* 2006). Small-scale

fisheries mostly focus on catching adult fish, while larvae are specifically taken for aquaculture in specific regions. Nevertheless, the technique of artificially producing *M. cephalus* for the purpose of aquaculture was established over 40 years ago (Shehadeh & Ellis 1970). These young fish have since been utilised to populate aquaculture ponds in certain regions, such as Taiwan (Liao 1981; Chang *et al.* 2000). The significant mortality of wild fry that are captured is a critical issue that can have negative effects on the sustainability of aquaculture operations and the local populations of *M. cephalus* (Tang 1975; Ben-Yami 1981).

The economic significance of mugilids differs from one country to another. They are less valuable in parts of Spain, France, and Australia, but they are very valuable in Tunisia, Egypt, and Taiwan. Mugilids are highly valued as bait in certain areas and are commonly employed as live bait for huge piscivorous fish in nations such as South Africa. Particularly for *M. cephalus*, global mullet harvesting peaked in the early 2000s; however, yields have been declining since 2004. In the past, *M. cephalus* stocks in eastern Australia were deemed to be fully exploited by the mid-1900s, and by the 1940s, they were already depleted (Grey *et al.* 2004). The decrease in fish catches along the coast frequently results in an increase in the cultivation of the species through aquaculture to make up for the loss (Thomson (1963), Kesteven (1942)).

Flathead mullet (*M. cephalus*) account for approximately 50% of the total mugilid catch worldwide. However, the accuracy of this data is limited because fishermen and mariculture operations in certain regions are unable to distinguish between different species of mullet. The majority of mugilid fisheries are located in coastal lagoons and estuaries, while there are also some fisheries that focus on marine populations (Panfili *et al.* 2006). Post larvae and early juveniles of *M. cephalus* are harvested in specific Asian nations for the purpose of aquaculture. For instance, in Taiwan, young individuals are deliberately raised until they reach sexual maturity in order to collect the eggs, which are highly valuable in the local economy. Mullet is occasionally exported to countries where certain species hold more economic significance in other regions of the world.

Mugil cephalus is a commercially significant fish species, especially in India, where it is captured in substantial amounts during the winter season. Research on

the accessibility and excellence of mullet egg has been carried out over the coastal regions of India (Sarojini 1958; Joseph 1976). Prior research has documented the presence of multiple species of mullet, as well as their early life history and feeding behaviours, in the Pichavaram mangrove environment (Jeyaseelan & Krishnamurthy 1981). Although the fishery is commercially important, there has been a lack of research conducted on it. Previous studies include those by Thakur (1970), Pillay (1953), Breder (1940), Sarojini (1951), Devasundaram (1952), John (1955), Patnaik (1962), Luther (1963, 1973), Thomson (1963), Hickling (1970), Rengaswamy (1972), and Das (1978). Grey mullets are extensively utilised in both monoculture and polyculture systems for aquaculture in brackish water.

The presence of brackish water lakes and estuaries significantly contributes to the enhanced production of commercially valuable finfish species that are suitable for export. The importance and extent of expanding brackish water fisheries in India have been repeatedly emphasised in many forums. The government has acknowledged the need of increasing fish and fisheries product output and intensifying our exports. To address this issue, it is imperative to focus only on the advancement of brackish water fishery.

3.2 METHODOLOGY

3.2.1 Sample Collection

For the proximate content analysis, aquaculture potential and organoleptic study of *M. cephalus*, the samples were collected during July-2021 to June-2022 from the study sites described in Chapter 2.0 (Fig. 2.1).

3.2.2 Proximate Content Analysis

The proximate analyses (Moisture, Ash, Protein, Lipid, Carbohydrate) were typically performed using established laboratory protocols (Fig. 3.1), and the amount of each component is expressed as a percentage of the total weight of the fish sample. Proximate analysis results offer useful insights for nutritionists, food scientists, fisheries managers, and consumers, aiding in the comprehension of the nutritional value, freshness, and general composition of fish. In addition, proximate analysis is important for quality control and assuring adherence to food safety and labelling regulations.

Figure 3.1: Thematic representation of proximate content analysis methodology

3.2.2.1 Moisture Content Analysis

The oven drying method, as specified by AOAC (1995), opted for moisture content analysis. At first, a precise measurement of 5 gm of tissue was taken. Subsequently, the tissue was carefully positioned into a Petri plate, guaranteeing a uniform dispersion. Subsequently, the Petri dish containing the tissue was moved to a preheated oven set at a temperature of 100°C. The tissue was subjected to a

drying process in the oven for a period of 3 hours. After the completion of the drying process, the tissue was extracted from the oven. The moisture content was determined by recording the weight of the wet sample prior to drying. The moisture content calculation was subsequently conducted utilising the following formula:

$$\text{Moisture content (\%)} = \frac{\text{Weight of Wet sample} - \text{Weight of dried sample}}{\text{Weight of wet sample}} \times 100$$

3.2.2.2 Total Ash Content Analysis

This approach is based on the concept of burning a specific amount of fish until it turns into ash, and then measuring the weight of the ash as a percentage of the original weight. The essential equipment comprises a silica crucible, tongs, weighing scale, Bunsen burner, muffle furnace and desiccator. The process commences with the determination of the mass of an uncontaminated and dehydrated crucible. Subsequently, roughly 5 gm of the sample was taken into the crucible, and its mass was precisely measured. Subsequently, the crucible was placed atop a hob, with the lid slightly ajar, resulting in the sample undergoing charring and releasing smoke. Afterwards, the crucible was moved to a muffle furnace and subjected to a temperature of 600°C for a duration of 3 hours. This process guarantees the thorough oxidation of all organic substances, resulting in the only presence of minerals. Once the crucible had cooled down in a desiccator to reach room temperature, it was weighed again in order to ascertain the weight of the remaining ash residue. This approach offers a dependable way to evaluate the mineral content of the fish sample by removing biological matter and preserving just the mineral residue in the crucible.

$$\text{Ash content (\%)} = \frac{Z - X}{Y - X} \times 100$$

Where X = Weight of empty crucible in grammes, Y = Weight of crucible in grammes + sample in grammes, Z = Weight of crucible in gm after ashing + ash

3.2.2.3 Total Lipid Content Analysis by Folch et al, 1957

The lipids were extracted from the sample using a technique that utilised the reagents chloroform, methanol, and distilled water. At first, 2 g muscle

tissue was measured and homogenised using a mortar and pestle. Next, the homogenised tissue was combined with a solution consisting of 1-part chloroform and 2 parts methanol, with the total volume adjusted to 20 ml for every 2 g of tissue. After a comprehensive vortexing, an extra amount of chloroform was introduced to the homogenate and blended once more. Afterwards, an equivalent volume of distilled water was added, and the mixture was agitated for the given incubation period. The outcome of this procedure led to the creation of a suspension that contains non-extractable residues. The suspension was formed by mixing chloroform, methanol, and water in volumetric ratios of 2:2:1.8 (v/v/v). Following a medium flow filter paper filter, the suspension was placed into tubes and centrifuged for 20 minutes at 3000 rpm. Following centrifugation, the filtrate experienced thorough phase separation, facilitating the elimination of the upper aqueous layer. The lowermost stratum, comprising the lipid extract, was gathered in a separate Eppendorf tube that had been previously weighed, and subjected to incubation in a 25°C incubator for a duration of 24 hours. Afterwards, the Eppendorf tubes were measured in terms of weight, and the quantity of lipid was determined by subtracting the initial weight from the final weight of the tubes. The same procedure was employed to ascertain the lipid content of the liver tissue. However, the sample size for the liver was reduced to 0.5 g instead of the intended 2 g. As a result, the composition of the reagents was adjusted to accommodate the smaller sample size.

$$\text{Total lipid (\%)} = \frac{\text{W of lipid (mg)}}{\text{W of sample (mg)}} \times 100$$

3.2.2.4 Total Protein Content Analysis by Bradford Assay

The required reagents for the Bradford protein assay consist of dissolving 100 mg of Coomassie brilliant blue G250 in 50 ml of 95% ethanol, then adding 100 ml of concentrated orthophosphoric acid (H_3PO_4), and finally correcting the volume to 200 ml with distilled water. The solution should be stored in amber bottles and refrigerated. It maintains stability for a minimum of 6 months. To prepare the dye solution, combine one part of concentrated dye with four parts of distilled water and filter it using Whatman No. 1 paper.

The protein standard comprises 100 µg (0.1 mg) of bovine serum albumin (BSA) dissolved in 1 ml of phosphate-buffered saline (PBS). The optical density of a 1 mg/ml BSA solution in a 1 cm light channel is 0.66 at a wavelength of 280 nm.

50 mg (0.05 g) of the sample tissue was homogenised during the process using 2 ml of PBS or distilled water. Working standards were prepared by using a pipette to transfer 0.0, 0.2, 0.4, 0.6, 0.8, and 1 ml of the BSA standard into test tubes that have been marked. In addition, 1 ml of the provided sample was transferred using a pipette into a separate test tube, while another test tube containing 1 ml of pure water was used as a blank. Subsequently, a volume of 5 ml of diluted dye solution was introduced into each test tube, including those designated as 'blank' and 'unknown'. After agitating by vortexing or shaking, the colour was let to intensify for a duration of 5 to 30 minutes. Upon interacting with protein, the red dye undergoes a chemical transformation and changes its colour to blue. The measurement of absorbance at 595 nm was subsequently taken in comparison to the blank, and a standard curve is constructed with protein concentration represented on the X-axis and absorbance at 595 nm represented on the Y-axis. Ultimately, the protein concentration in the provided sample was determined by applying the formula y = mx + b to the standard curve. The outcome was represented as the unknown sample provided, which contains µg protein/ml. The parameter value was translated into a percentage using the dry weight basis. The formula for dry weight basis is as follows.

$$\text{Dry weight of tissue} = \frac{100 - \text{Moisture content (\%)}}{100}$$

$$\text{Protein (\%)} = \frac{\text{Protein}\left(\frac{\text{mg}}{100\text{mg}}\right)}{\text{Dry weight of tissue}}$$

3.2.2.5 Total Carbohydrate Content Analysis by Anthrone Method

The anthrone method, originally described by Hedge and Hofreiter in 1962, was employed to analyse the carbohydrate content in the tissue. Antrone reagent was prepared mixing 130 mg of anthrone with 4.03 ml of distilled water (DW), followed by the addition of 60.9 ml of 95% concentrated ice-cold sulfuric acid (H_2SO_4). The calibration process was carried out using standard glucose, which

was prepared by initially creating a concentrated solution by mixing 100 mg of glucose with 100 ml of distilled water. This concentrated solution was then diluted to create a working solution by adding 10 ml of the concentrated solution to 100 ml of distilled water. In addition, a 2.5 N hydrochloric acid (HCl) solution was created by combining 2.7 ml of HCl with 10.3 ml of distilled water.

The tissue analysis technique commenced by weighing 50 mg (0.05 g) of the sample tissue. The tissue was then mixed with 2.5 ml of 2.5 N HCl in a mortar and pestle. After complete homogenization, the liquid was transferred to a sterile test tube and exposed to a 3-hour boiling water bath. After the solution was cooled, sodium carbonate (Na_2CO_3) was added until the bubbling stopped, and then the volume was made up to 50 ml with purified water. The solution was subsequently subjected to centrifugation at a speed of 1000 revolutions per minute for a duration of 10 minutes, resulting in the collection of the supernatant. Five test tubes were set up for the analysis: three sample tubes, each containing one ml of the supernatant, one blank tube containing one ml of distilled water, and one standard tube containing one ml of standard working glucose. 4 ml of anthrone reagent was added to each test tube, ensuring complete mixing, and then incubated in a boiling water bath for a duration of 8 minutes. The test tubes were cooled and the green colour that appeared was measured at a wavelength of 630 nm using a spectrophotometer.

The carbohydrate content in sample tissue was initially estimated in terms of mg/100 mg using following formula.

$$\text{Carbohydrate}\left(\frac{\text{mg}}{100\text{mg}}\right) = \frac{\text{Sample OD} \times \text{Standard Concentration} \times \text{Dilution}}{\text{Standard OD} \times \text{Weight of tissue} \times \text{Aliquot}} \times 100$$

Where, Standard Concentration = 0.02, Dilution = 50, Weight of tissue = 50 mg, Aliquot = 1. The value of parameter was converted into % using dry weight basis. The formula for dry weight basis was given as follows.

$$\text{Dry weight of tissue} = \frac{100 - \text{Moisture content (\%)}}{100}$$

$$\text{Carbohydrate (\%)} = \frac{\text{Carbohydrate}\left(\frac{\text{mg}}{100\text{mg}}\right)}{\text{Dry weight of tissue}}$$

3.2.3 Aquaculture Potential

Sensory techniques are employed to evaluate the level of freshness by considering organoleptic attributes such as overall meat consistency, visual appearance, smell, texture, eye condition, and gill condition. A group of ten individuals who had received specialised training were chosen from the University community. The samples were given in a random manner. Five samples of *M. cephalus* were assessed using the hedonic scale, which consists of five points ranging from 1 (extreme dislike) to 5 (extreme liking). The evaluation continued until the samples deteriorated (Klein & Bardy 1984; Rana *et al.* 2020).

In addition, the specimens were transported to the Animal House located in the Department of Zoology at the Faculty of Science, The Maharaja Sayajirao University of Baroda, Vadodara. To facilitate the adjustment of the fish, they were transferred to a pre-prepared aquarium tank with a salinity level similar to that of the location where they were originally collected. Following a 24-hour period, a trio of individuals were transferred to separate aquariums with varying levels of salinity (10ppt, 20ppt, 30ppt) then after their length and weight measurements were recorded. They were kept in tanks for six months with an equal amount of food to monitor their growth and biochemical changes in various salinity ranges.

Figure 3.2: Methodology for aquaculture potential study

3.3 RESULTS

3.3.1 Proximate Content Analysis of Mugil cephalus

3.3.1.1 Total Moisture Content

Moisture content in *M. cephalus*, ranges from 73.13±0.66% (Apr-22) to 79.93±0.16% (Aug-21) of males and from 75.67±0.52% (Apr-22) to 80.97±0.93% (Dec-21) of females among all the study sites. For male, the average moisture content recorded was 78.14±1.09% from Ambetha, followed by 77.79±0.83% from Diu, 77.46±1.24% from Bhadbhut and 75.36±1.35% from Bharuch in descending manner. Whereas in females, the moisture content (%) was found high as compare to males. The mean moisture content recorded in females was 79.21±0.89% Diu, followed by 78.70±0.95% from Ambetha, 78.17±1.11% Bhadbhut and 78.12±1.27% from Bharuch. The highest moisture content was observed during spawning and post-spawning months whereas the lowest were found during summer in both the species. The minimum and maximum values recorded from all the study sites are shown in Table 3.1.

Table 3.1: Total Moisture content (%) analysis of male and female *M. cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

Moisture Content (%) - MALE				
	Bharuch	**Bhadbhut**	**Ambetha**	**Diu**
Min	73.13±0.66 Apr-22	75.77±0.17 May-22	76.37±0.17 May-22	76.37±0.18 May-22
Max	77.27±0.96 Nov-21	79.63±0.19 Aug-21	79.93±0.16 Aug-21	79.13±0.15 Aug-21
Moisture Content (%) - FEMALE				
Min	75.67±0.52 Apr-22	76.47±0.54 Apr-22	77.37±0.58 Apr-22	77.97±0.52 Apr-22
Max	79.87±0.96 Nov-21	80.07±0.87 Dec-21	80.57±0.82 Dec-21	80.97±0.93 Dec-21

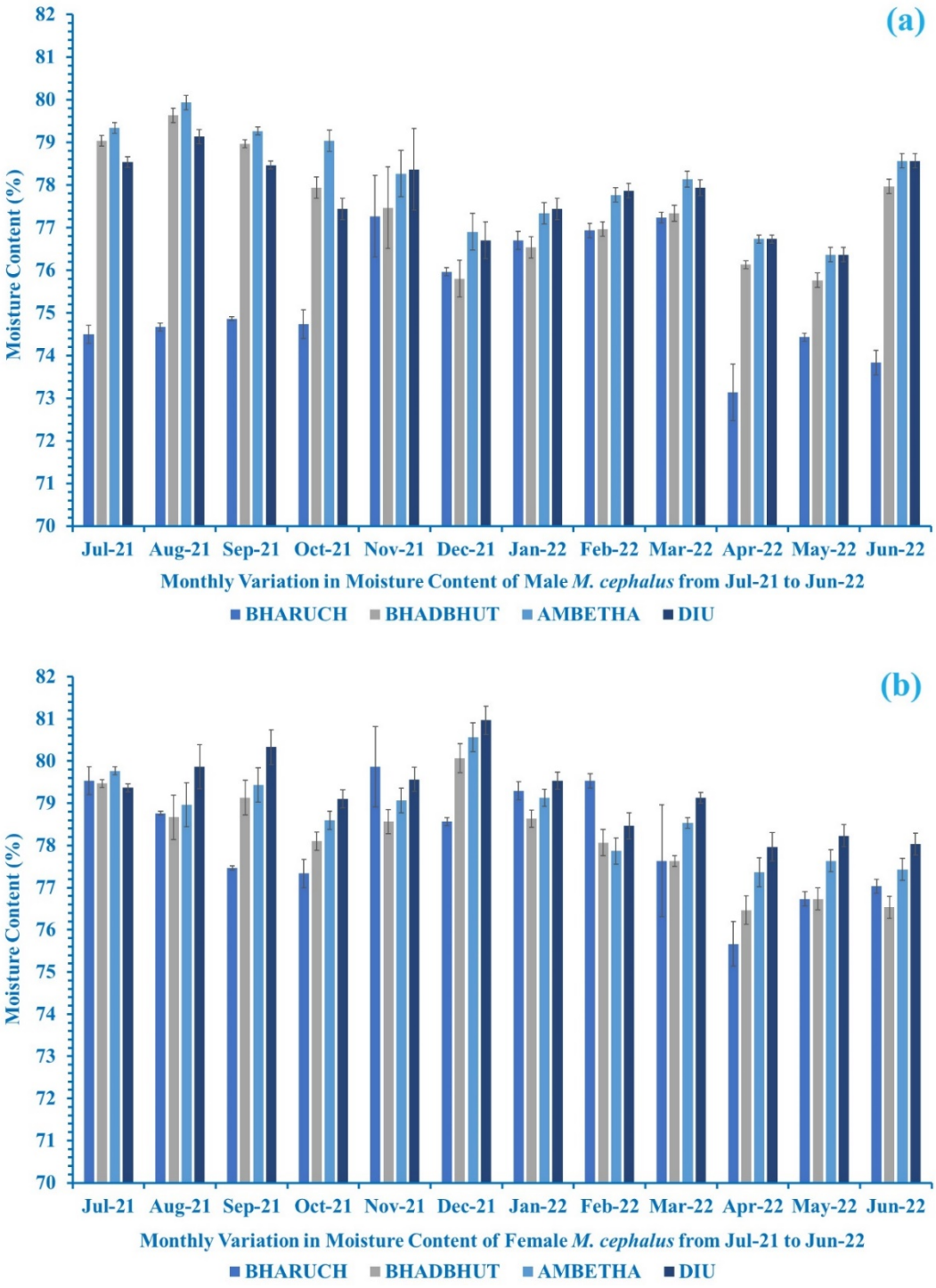


Figure 3.3: Monthly variation in Moisture Content (%) analysis of (a) Male and (b) Female *Mugil cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

3.3.1.2 Total Ash Content

The total ash content (%) were found to range from 0.81±0.02% (Jul-21) to 1.28±0.11% (Nov-21) of dry weight of males and from 0.10±0.01% (Feb-22) to 1.84±0.06% (Jan-22) of dry weight of females among all the study sites. For male, the average ash content recorded was 1.08±0.08% from Ambetha, followed by 0.99±0.09% from Bhadbhut, 0.92±0.08% from Bharuch and 0.10±0.09% from Diu in descending manner. Whereas in females, the total ash content (%) was found high as compare to males. The mean ash content observed in females was 1.40±0.24% from Bhadbhut followed by 1.20±0.12% from Ambetha, 1.16±0.14% from Diu and 1.14±0.15% from Bharuch. The highest ash content in females was observed during post-spawning (Jan) and summer (May-Jun), whereas, the lowest was observed in winter (Feb-22) and onset of gonadal development (Jul-Aug). In case of males, the highest content was observed in post-spawning (Nov-Jan). The lowest was observed in the beginning of the gonadal maturation (Jul-Aug) except the males of Bhadbhut which were showing lowest content in spawning season (Oct-21). The minimum and maximum values recorded from all the study sites are shown in Table 3.2.

Table 3.2: Total Ash content (%) analysis of male and female *M. cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

Ash Content (%) - MALE				
	Bharuch	**Bhadbhut**	**Ambetha**	**Diu**
Min	0.81±0.02 Jul-21	0.87±0.03 Oct-21	0.99±0.02 Jun-22	0.82±0.02 Aug-21
Max	1.05±0.06 Jan-22	1.17±0.05 Nov-21	1.28±0.11 Nov-21	1.13±0.03 Dec-21
Ash Content (%) - FEMALE				
Min	0.10±0.01 Feb-22	1.13±0.01 Jul-21	1.09±0.06 Jul-21	0.88±0.09 Aug-21
Max	1.61±0.03 Jun-22	1.84±0.06 Jan-22	1.53±0.04 Jun-22	1.51±0.07 Jun-22

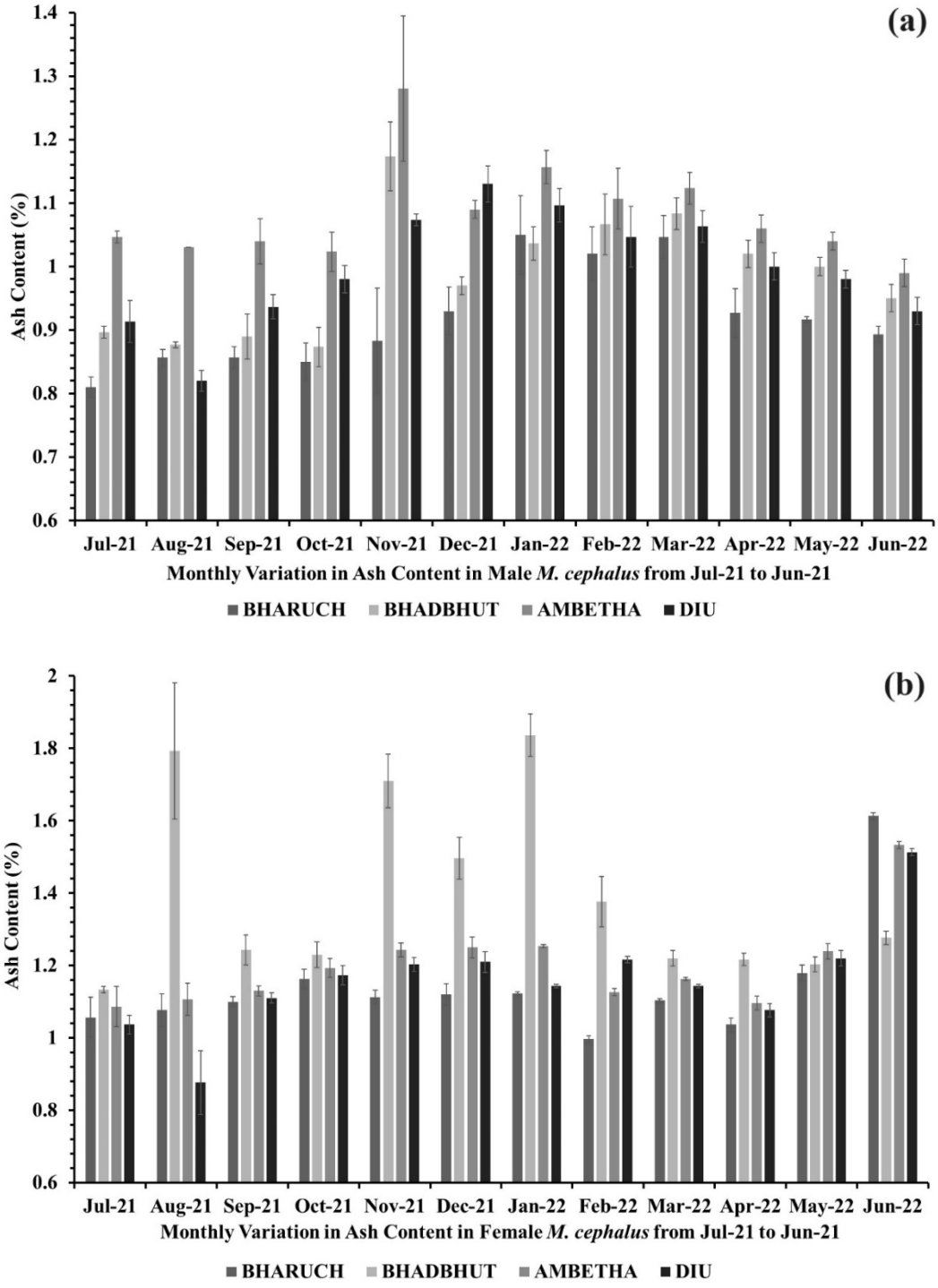


Figure 3.4: Monthly variation in Ash Content (%) analysis of (a) Male and (b) Female *Mugil cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

3.3.1.3 Total Lipid Content

Total lipid content (%) was measured in muscle and liver tissues of male and female individual of *M. cephalus*. In males, it was found to vary from 9.10±1.19% (May-22) to 16.71±1.72% (Oct-21) of dry weight of muscles and from 13.95±1.00% (Sep-21) to 24.95±4.24% (Jan-22) of dry weight of liver from all the study sites. For muscles, the average lipid content (%) was recorded as follows: 15.01±1.19% (Bhadbhut), 14.78±1.18% (Diu), 13.03±1.19% (Bharuch) and 12.79±1.15% (Ambetha). The mean lipid content in liver was found higher compare to muscles and it was recorded as follows: 20.10±2.47% (Bhadbhut), followed by 19.13±2.46% (Ambetha), 19.03±2.42% (Diu) and 17.55±2.48% (Bharuch). The highest content of lipid in muscles was observed during breeding month (Oct-21) and the lowest was observed in summer (May-22). Whereas in liver the highest content was recorded post-spawning months (Dec-21, Jan-22) and the lowest was observed during spawning season (Sep-21). The minimum and maximum values recorded from all the study sites are shown in Table 3.3.

Table 3.3: Total Lipid content (%) analysis from muscle and liver tissues of Male *M. cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

Lipid Content (%) – MALE - MUSCLES				
	Bharuch	**Bhadbhut**	**Ambetha**	**Diu**
Min	9.10±1.19 May-22	11.97±1.18 May-22	9.75±1.29 May-22	11.74±1.93 May-22
Max	14.73±1.74 Oct-21	16.71±1.72 Oct-21	14.49±1.83 Oct-21	16.48±1.73 Oct-21
Lipid Content (%) – MALE - LIVER				
Min	13.95±1.00 Sep-21	17.39±1.06 Sep-21	15.53±0.98 Sep-21	15.43±0.91 Sep-21
Max	21.50±2.49 Dec-21	24.95±4.24 Jan-22	23.08±2.47 Dec-21	22.98±2.41 Dec-21

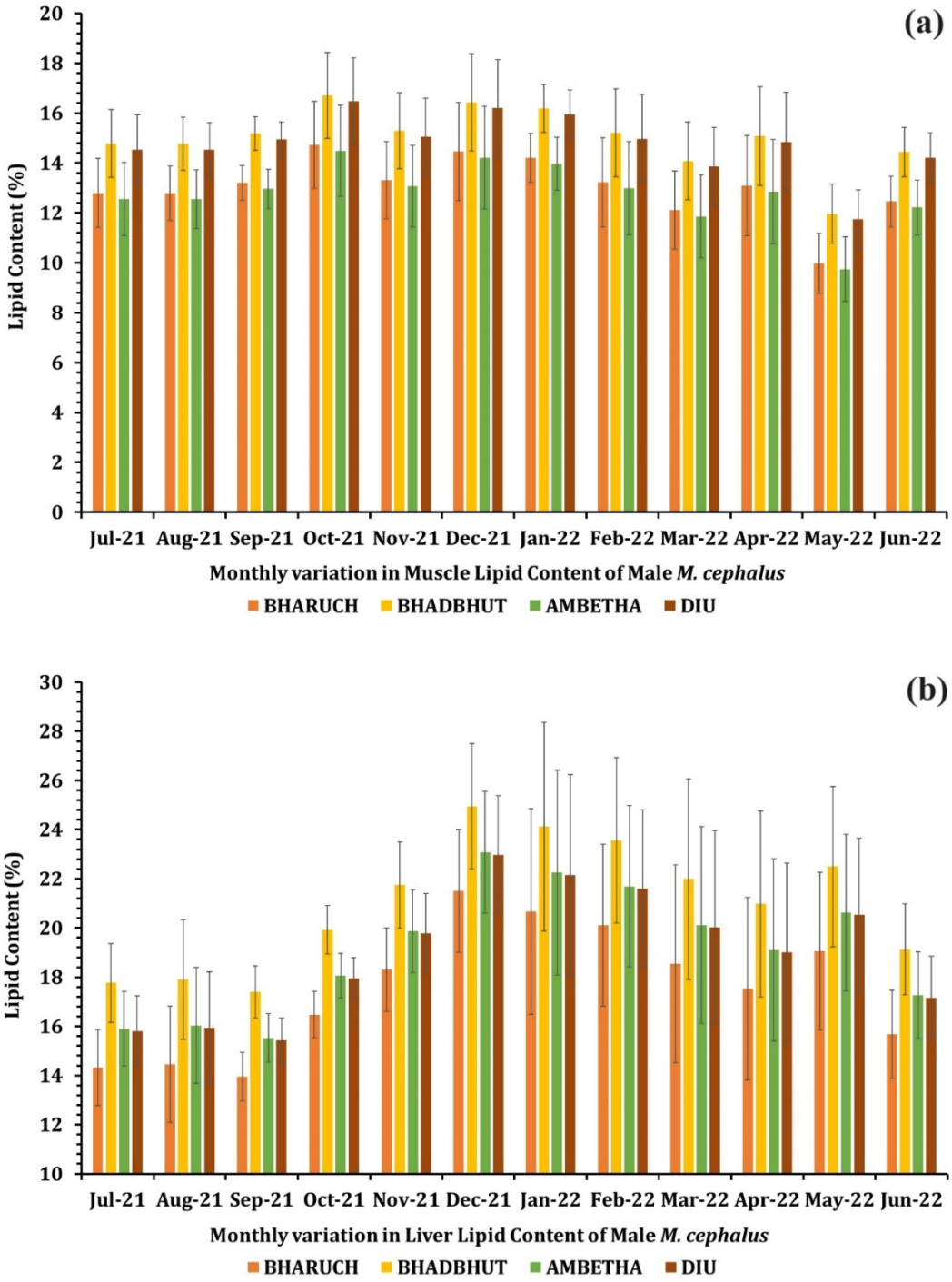


Figure 3.5: Monthly variation in Lipid Content (%) analysis of (a) Male – Muscle tissue and (b) Male – Liver tissue of *Mugil cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

In females, the lipid content (%) was ranged from 13.14±1.75% (Jun-21) to 17.59±1.32% (Feb-22) of dry weight of muscles and from 15.58±1.78% (Oct-21) to 23.51±2.42 (Jan-22) of dry weight of liver. For muscles, the average lipid content (%) was recorded as follows: 16.09±0.66% (Bhadbhut), 15.09±0.67% (Diu),

15.01±0.67% (Ambetha) and 14.29±0.68% (Bharuch). The mean lipid content in liver was found higher compare to muscles and it was recorded as follows: 20.70±1.64% (Bhadbhut), followed by 19.61±1.63% (Diu), 18.93±1.61% (Ambetha) and 18.73±1.67% (Bharuch). The highest content of lipid in muscles was observed during post-spawning (Feb-22) and the lowest was observed in summer (Jun-22). Whereas in liver the highest content was recorded post-spawning months (Jan-22) and the lowest was observed during spawning season (Oct-21). The minimum and maximum values recorded from all the study sites are shown in Table 3.4.

Table 3.4: Total Lipid content (%) analysis from muscle and liver tissues of Female *M. cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

Lipid Content (%) – FEMALE - MUSCLES				
	Bharuch	**Bhadbhut**	**Ambetha**	**Diu**
Min	13.14±1.75 Jun-22	14.94±1.27 Jun-22	13.87±1.10 Jun-22	13.94±1.18 Jun-22
Max	15.79±1.81 Feb-22	17.59±1.32 Feb-22	16.52±1.16 Feb-22	16.59±1.24 Feb-22
Lipid Content (%) – FEMALE - LIVER				
Min	15.58±1.78 Oct-21	17.54±2.22 Oct-21	15.78±1.71 Oct-21	16.46±1.67 Oct-21
Max	21.55±1.99 Jan-22	23.51±2.42 Jan-22	21.75±1.91 Jan-22	22.42±1.86 Jan-22

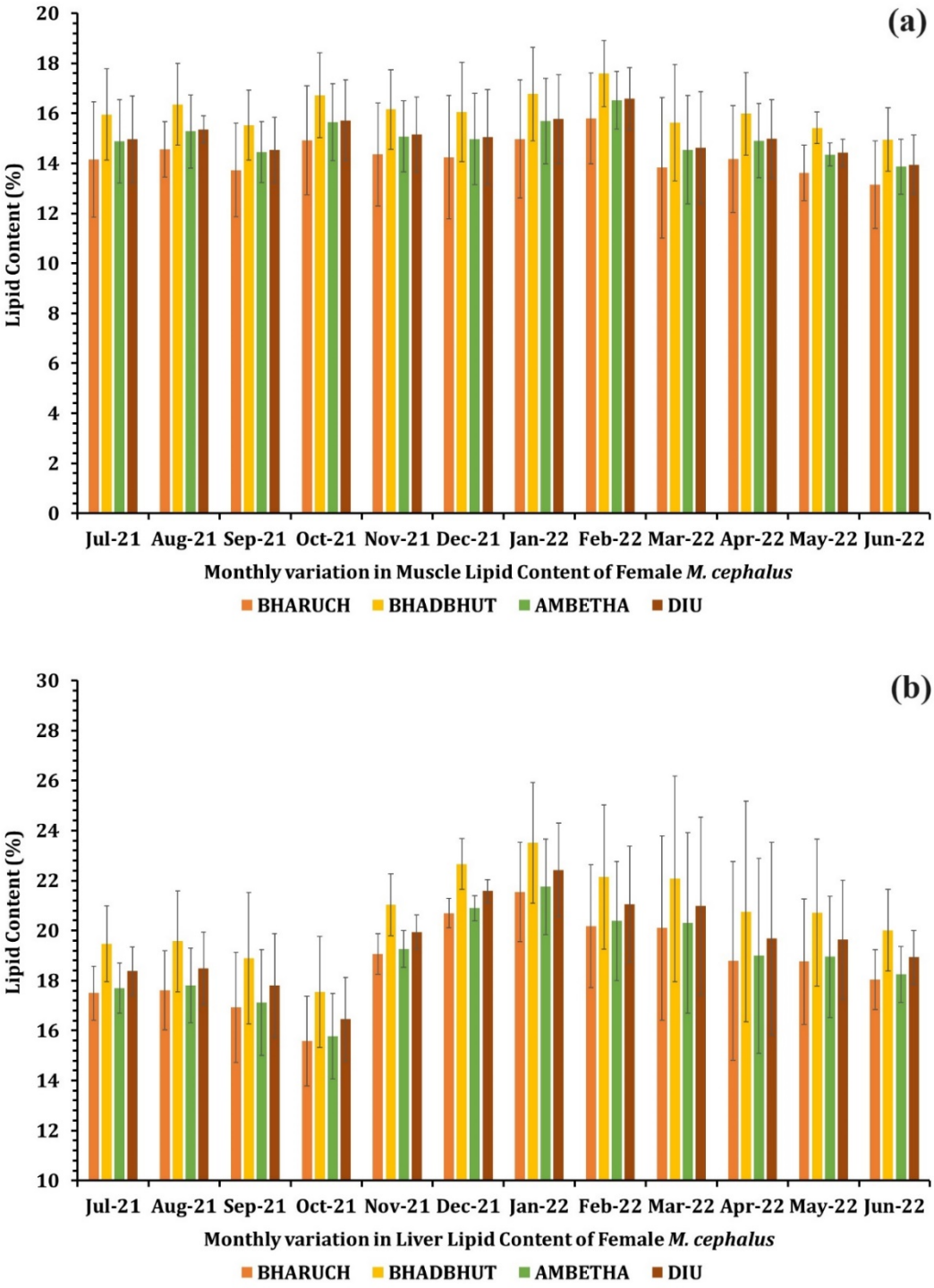


Figure 3.6: Monthly variation in Lipid Content (%) analysis of (a) Female – Muscle tissue and (b) Female – Liver tissue of *Mugil cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

3.3.1.4 Total Protein Content

Total protein content (%) was investigated in muscle and liver tissues of male and female *M. cephalus*. In males, it was ranged from 49.25±1.51% (Sep-21) to 65.37±2.32% (Feb-22) of dry weight of muscles and from 51.24±1.77% (Oct-21) to 65.78±1.74% (Feb-22) of dry weight of liver from all the study sites. For muscles, the average protein content (%) was recorded as follows: 59.07±3.66% (Bhadbhut), 58.05±3.66% (Diu), 56.60±3.66% (Ambetha) and 54.09±3.66% (Bharuch). The mean protein content in liver was found higher compare to muscles and it was recorded as follows: 60.42±3.39% (Bhadbhut), followed by 59.34±3.27% (Diu), 57.66±3.41% (Ambetha) and 56.50±3.27% (Bharuch). The highest content of protein in muscles was observed during winter (Feb-22) and the lowest was observed in breeding (Sep-21). Whereas in liver the highest content was recorded post-spawning/winter months (Feb-22) and the lowest was observed during spawning season (Oct-21). The minimum and maximum values recorded from all the study sites are shown in Table 3.5.

Table 3.5: Total Protein content (%) analysis from muscle and liver tissues of Male *M. cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

Protein Content (%) – MALE - MUSCLES				
	Bharuch	**Bhadbhut**	**Ambetha**	**Diu**
Min	49.25±1.51 Sep-21	54.23±1.96 Sep-21	51.77±1.94 Sep-21	53.21±2.01 Sep-21
Max	60.39±1.86 Feb-22	65.37±2.32 Feb-22	62.91±2.30 Feb-22	64.35±2.37 Feb-22
Protein Content (%) – MALE - LIVER				
Min	51.24±1.77 Oct-21	55.00±1.89 Oct-21	52.14±1.31 Oct-21	54.12±1.74 Oct-21
Max	61.46±1.62 Feb-22	65.78±1.74 Feb-22	63.00±1.16 Feb-22	64.54±1.59 Feb-22

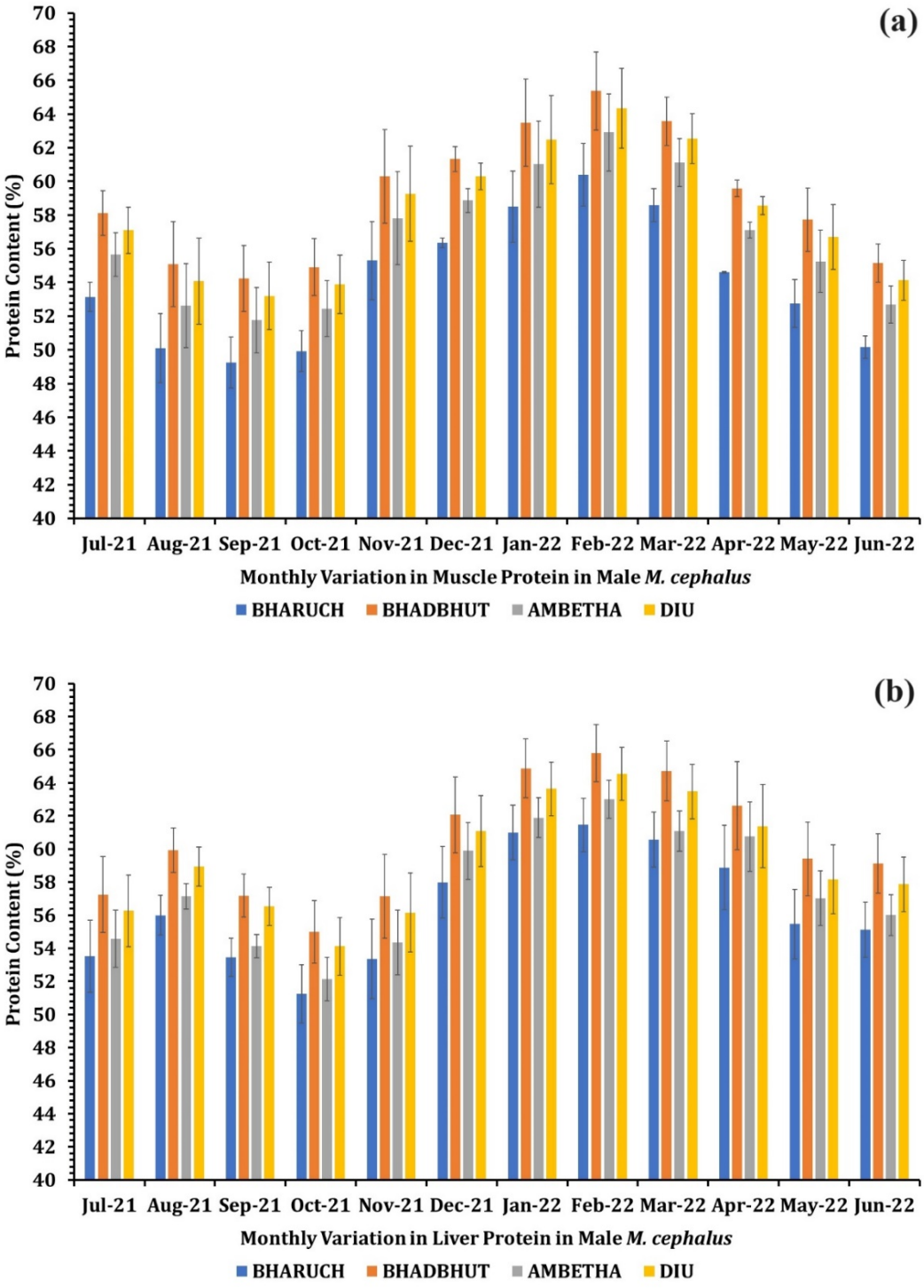


Figure 3.7: Monthly variation in Protein Content (%) analysis of (a) Male – Muscle tissue and (b) Male – Liver tissue of *Mugil cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

In females, the protein content (%) was ranged from 50.16±2.54% (Aug-21) to 62.92±2.80% (Feb-22) of dry weight of muscles and from 52.62±2.49% (Sep-21) to 64.72±1.43% (Feb-22) of dry weight of liver. For muscles, the average protein content (%) was recorded as follows: 59.21±3.03% (Bhadbhut),

58.22±3.03% (Diu), 57.25±3.03% (Ambetha) and 55.75±3.03% (Bharuch). The mean protein content in liver was found higher compare to muscles and it was recorded as follows: 60.56±2.68% (Bhadbhut), followed by 60.29±2.56% (Diu), 58.55±2.68% (Ambetha) and 56.55±2.69% (Bharuch). The highest content of protein in muscles was observed post-spawning (Feb-22) and the lowest was observed during the onset of gonadal maturation (Aug-21). Whereas in liver the highest content was recorded post-spawning months (Jan-22, Feb-22) and the lowest was observed during spawning season (Sep-21). The minimum and maximum values recorded from all the study sites are shown in Table 3.6.

Table 3.6: Total Protein content (%) analysis from muscle and liver tissues of Female *M. cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

Protein Content (%) – FEMALE - MUSCLES				
	Bharuch	**Bhadbhut**	**Ambetha**	**Diu**
Min	50.16±2.54 Aug-21	53.62±3.50 Aug-21	51.66±3.44 Aug-21	52.63±3.04 Aug-21
Max	59.46±1.85 Feb-22	62.92±2.80 Feb-22	60.96±2.74 Feb-22	61.93±2.35 Feb-22
Protein Content (%) – FEMALE - LIVER				
Min	52.62±2.49 Sep-21	56.63±2.57 Sep-21	54.63±2.21 Sep-21	56.42±2.56 Sep-21
Max	60.71±1.35 Feb-22	64.72±1.43 Feb-22	62.72±1.08 Feb-22	64.21±2.12 Jan-22

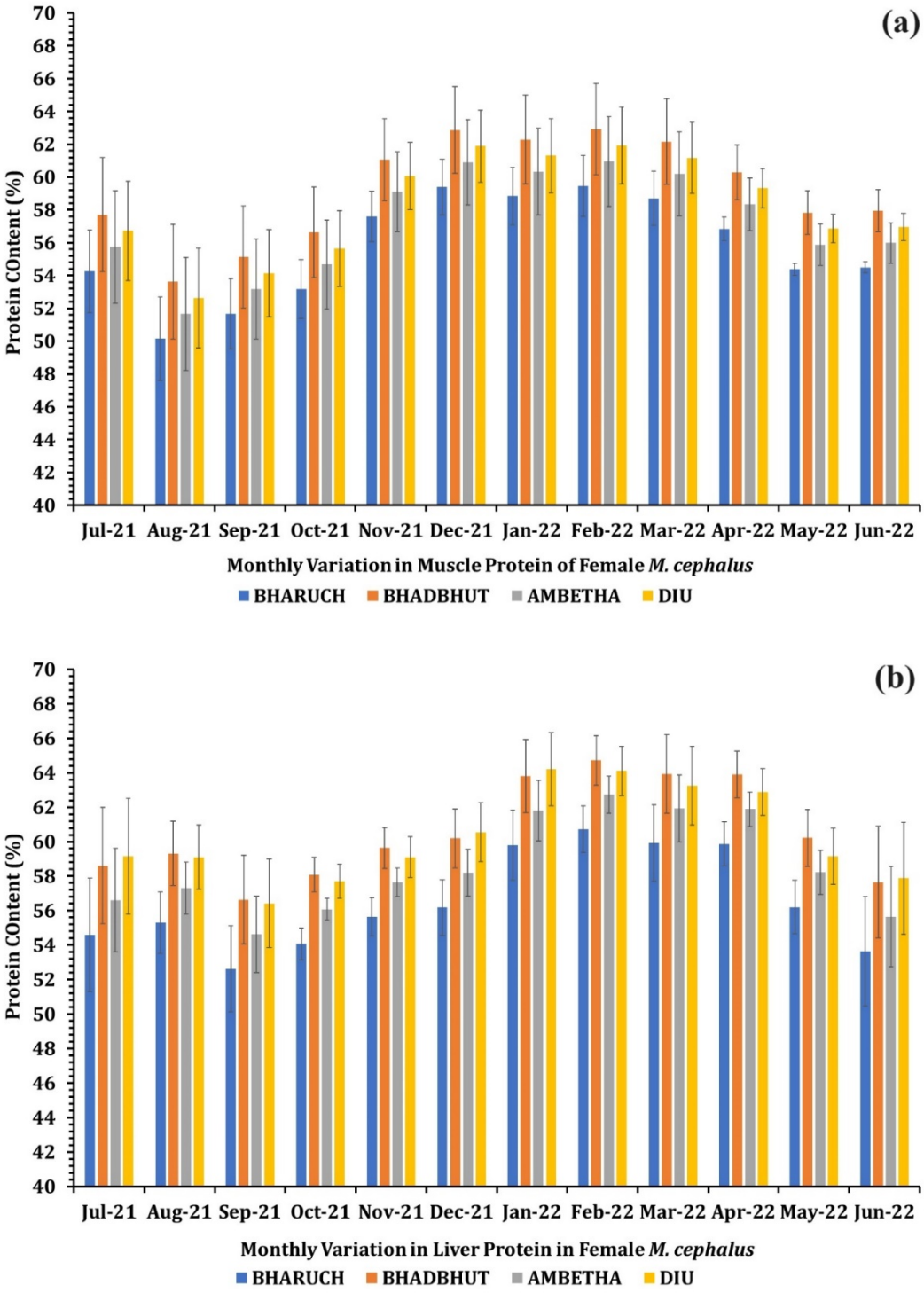


Figure 3.8: Monthly variation in Protein Content (%) analysis of (a) Female – Muscle tissue and (b) Female – Liver tissue of *Mugil cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

3.3.1.5 Total Carbohydrate Content

Total carbohydrate content (%) was investigated in muscle and liver tissues of male and female *M. cephalus*. In males, it was found to vary from 5.63±0.83% (May-22) to 8.46±0.89% (Jan-22) of dry weight of muscles and from 5.75±1.31% (Oct-21) to 7.20±1.25% (Nov-21) of dry weight of liver from the selected study sites. The average carbohydrate content in muscle tissue was recorded as 7.42±0.49% (Bharuch) followed by 7.34±0.53% (Bhadbhut), 7.25±0.46% (Diu) and 6.46±0.48% (Ambetha) in descending order. Whereas in liver tissue, it was recorded as 6.27±0.42% (Diu) followed by 6.14±0.40% (Bhadbhut), 5.17±0.39% (Bharuch) and 5.14±0.41% (Ambetha). It was observed that compare to lipid and protein, the carbohydrate content in liver was law compare to muscles of M. cephalus from all the study sites. The highest carbohydrate content in muscles was observed post-spawning (Jan-22) and the lowest was observed during summer (May-22). Whereas in liver, the highest was observed during offset of the spawning season (Nov-21) and the lowest was recorded during onset of the spawning season (Oct-21). The minimum and maximum values recorded from all the study sites are shown in Table 3.7.

Table 3.7: Total Carbohydrate content (%) analysis from muscle and liver tissues of Male *M. cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

Carbohydrate Content (%) – MALE - MUSCLES				
	Bharuch	**Bhadbhut**	**Ambetha**	**Diu**
Min	6.58±0.97 May-22	6.51±0.76 May-22	5.63±0.83 May-22	6.42±0.78 May-22
Max	8.46±0.89 Jan-22	8.38±0.68 Jan-22	7.50±0.75 Jan-22	8.29±0.71 Jan-22
Carbohydrate Content (%) – MALE - LIVER				
Min	4.65±0.58 Oct-21	5.62±0.33 Oct-21	4.62±1.22 Oct-21	5.75±1.31 Oct-21
Max	6.11±1.52 Nov-21	7.08±1.28 Nov-21	6.07±1.16 Nov-21	7.20±1.25 Nov-21

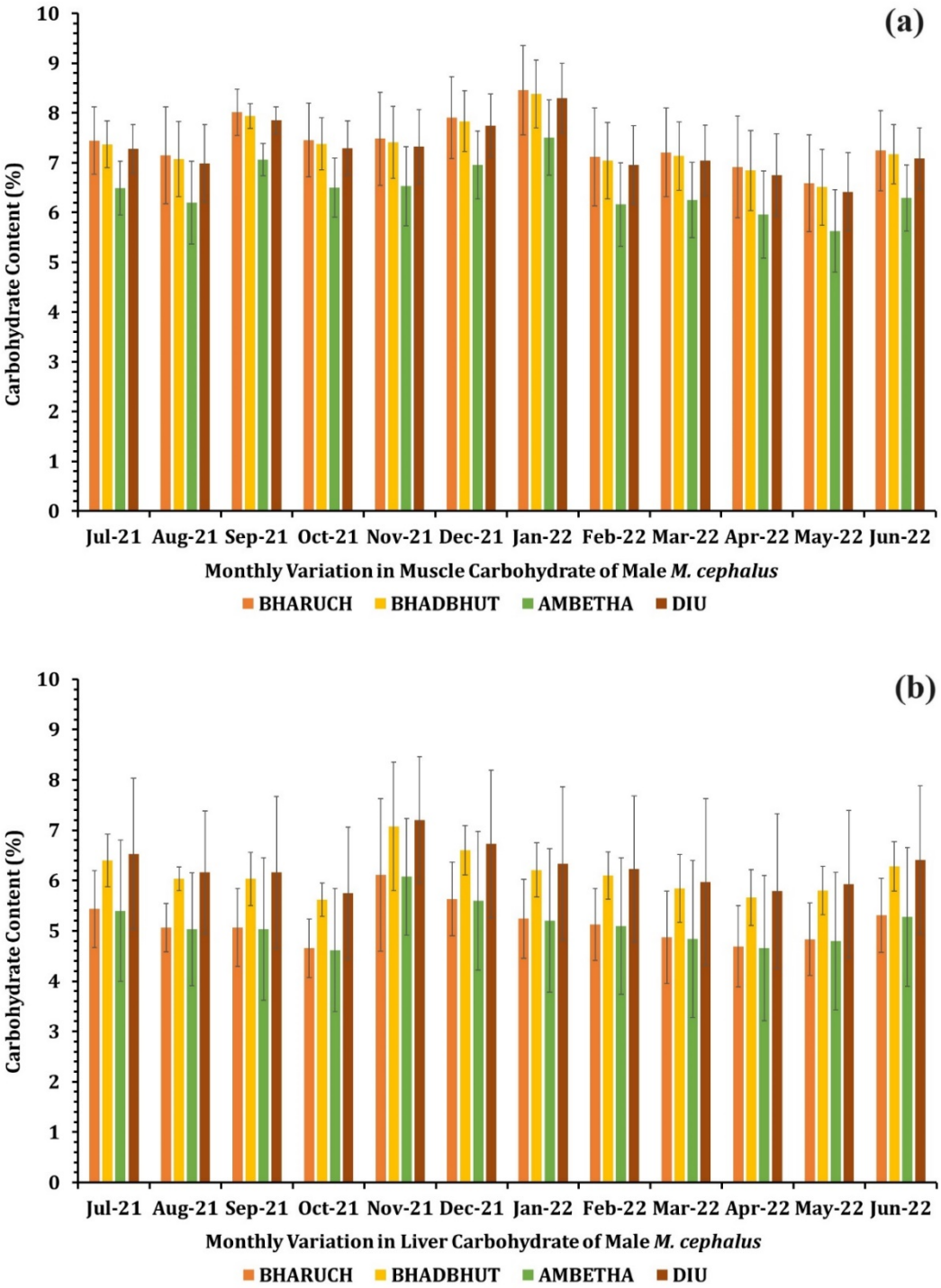


Figure 3.9: Monthly variation in Carbohydrate Content (%) analysis of (a) Male – Muscle tissue and (b) Male – Liver tissue of *Mugil cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

In females, the carbohydrate content (%) was ranged from 4.77±0.81% (Jun-22) to 8.67±0.49% (Aug-21) of dry weight of muscles and from 4.58±0.73% (Mar-22) to 7.49±1.40% (Sep-21) of dry weight of liver. For muscles, the average carbohydrate content (%) was recorded as follows: 7.54±0.84% (Bhadbhut),

7.49±0.78% (Diu), 6.57±0.81% (Ambetha) and 6.48±0.83% (Bharuch). The mean carbohydrate content in muscle was found higher compare to liver. In liver, it was recorded as follows: 6.75±0.45% (Diu), followed by 6.15±0.39% (Bhadbhut), 5.71±0.43% (Ambetha) and 5.19±0.38% (Bharuch). The highest content of carbohydrate in muscles was observed during the onset of spawning season (Aug-21) and the lowest was observed during summer (Jun-22). Whereas in liver the highest content was recorded during spawning months (Sep-21) and the lowest was observed during summer (Mar-22). The minimum and maximum values recorded from all the study sites are shown in Table 3.8.

Table 3.8: Total Carbohydrate content (%) analysis from muscle and liver tissues of Female *M. cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

Carbohydrate Content (%) – FEMALE - MUSCLES				
	Bharuch	**Bhadbhut**	**Ambetha**	**Diu**
Min	4.77±0.81 Jun-22	5.83±0.36 Jun-22	4.86±0.61 Jun-22	5.78±0.38 Jun-22
Max	7.61±0.95 Aug-21	8.67±0.49 Aug-21	7.71±0.75 Aug-21	8.63±0.52 Aug-21
Average				
Carbohydrate Content (%) – FEMALE - LIVER				
Min	4.58±0.73 Mar-22	5.54±0.69 Mar-22	5.10±1.21 Mar-22	6.14±1.20 Mar-22
Max	5.93±0.92 Sep-21	6.89±0.89 Sep-21	6.45±1.41 Sep-21	7.49±1.40 Sep-21

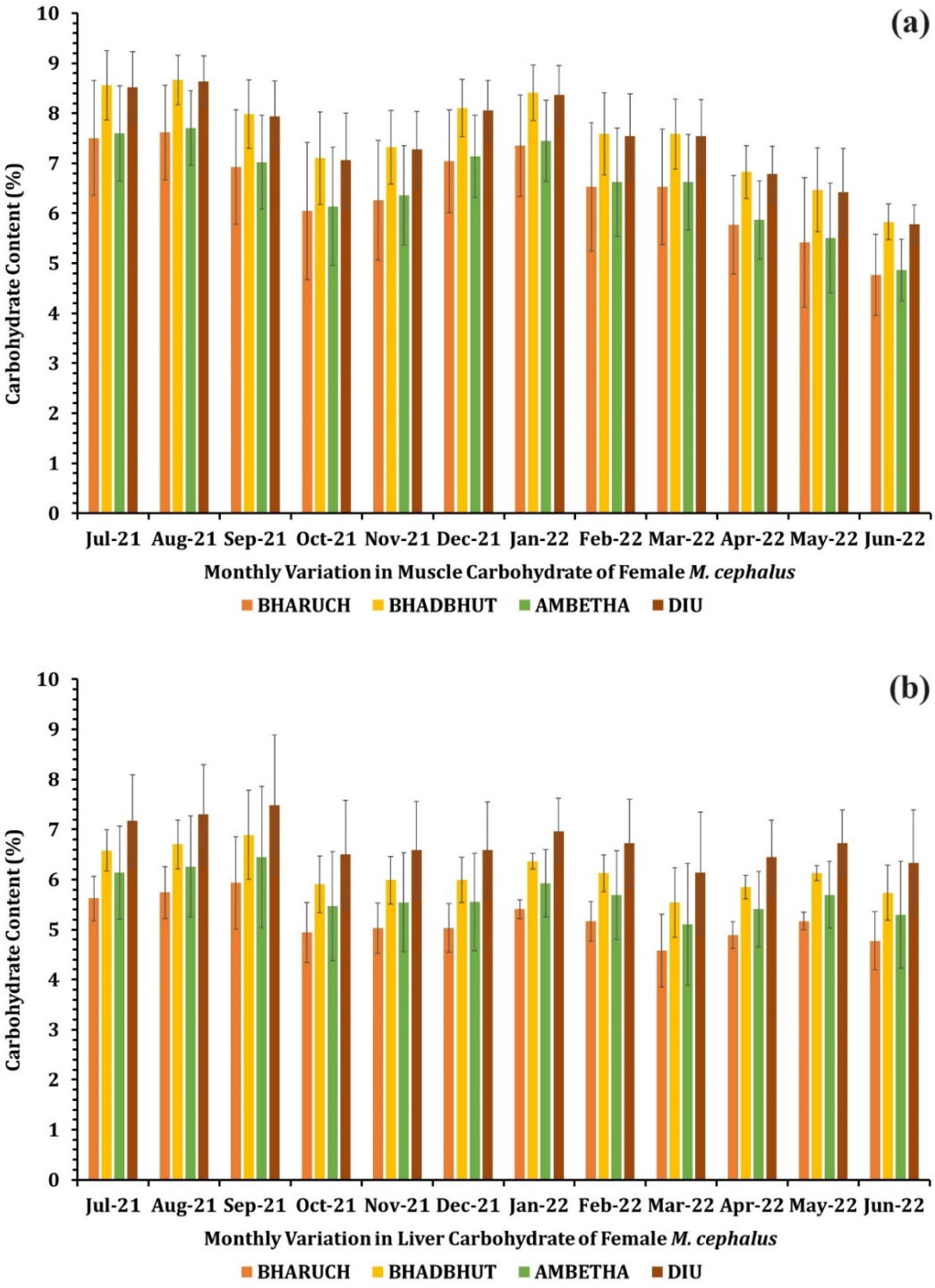
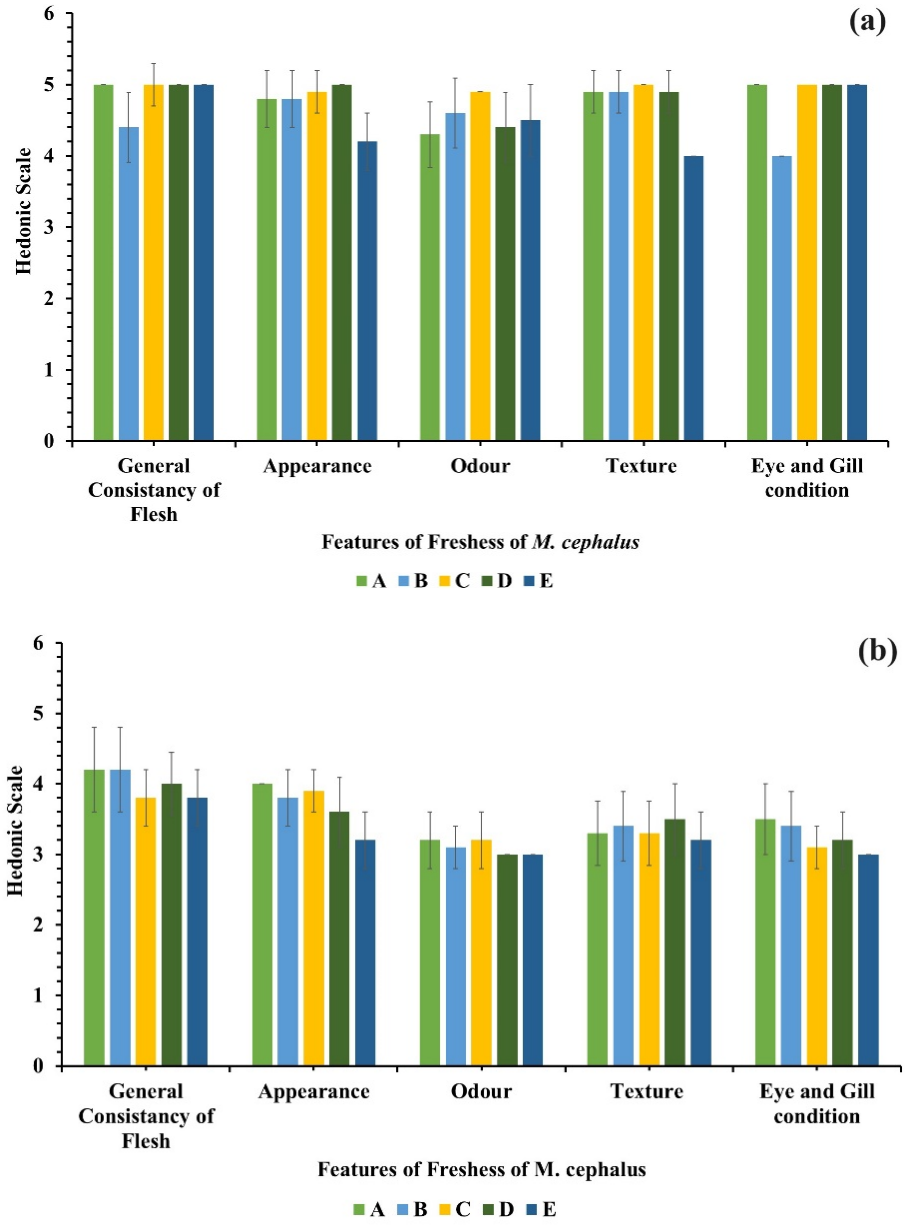


Figure 3.10: Monthly variation in Carbohydrate Content (%) analysis of (a) Female – Muscle tissue and (b) Female – Liver tissue of *Mugil cephalus* from Diu lagoon and Narmada estuary of Gujarat, India

3.3.2 Aquaculture Potential Study

Fig. 3.11 provides the information of the hedonic scale points given to the quality of the fish at room temperature. On the first day, majority of the trained individuals gave the 5 points of general consistency, appearance, texture and eye and gill condition to first four specimens (A, B, C, D) and the rest others received average score between 4 to 5. On day 2, score decreases in odour followed by texture, eye and gill condition, appearance and general consistency which scored between 3 to 4.5 on hedonic scale base. On day 3, the overall condition of the specimen showed marked changes in organoleptic qualities as evidenced by faded appearance, cloudy and depressed eyes, dull gill colour, and soft belly etc.



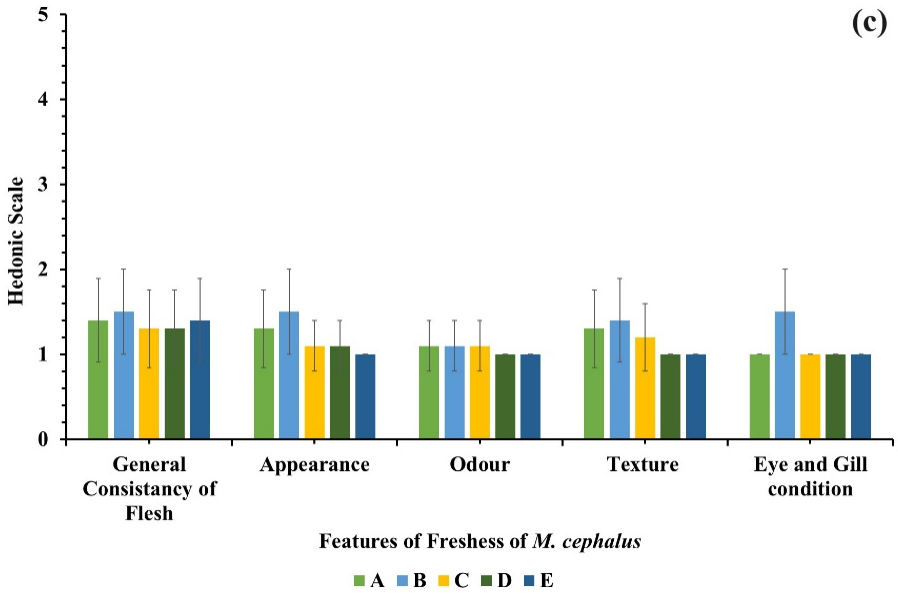


Figure 3.11: Organoleptic Study of *Mugil cephalus* based on hedonic scale of five points

The collected specimens were kept in three different aquarium tanks (10ppt, 20ppt, 30ppt) and their initial length in cm and weight in g were taken (Table 3.9). The final values of all three tanks are shown in table 3.9 after the period of six months. In tank 1, the initial average growth was 15.50±0.08 (TL) and 42.99±0.78 (TW). Where as in tank 2 and tank 3, the initial growth rate was recorded 15.57±0.12 (TL), 43.62±1.29 (TW) and 15.33±0.09 (TL), 41.27±0.79 (TW) respectively. Over a period of six month, it was observed that the average growth rate was found more in tank 2 (20ppt) followed by tank 3 (30ppt) and tank 1 (10ppt). In tank 2, the average growth was recorded 17.27±0.09 (TL), 51.19±1.02 (TW) followed by 17.93±0.17 (TL), 51.84±1.02 (TW) in tank 3 and 17.27±0.09 (TL), 51.19±1.02 (TW) in tank1 respectively. The growth rate analysis suggests that the salinity range near 20ppt provides the optimal condition to *M. cephalus* for their survival.

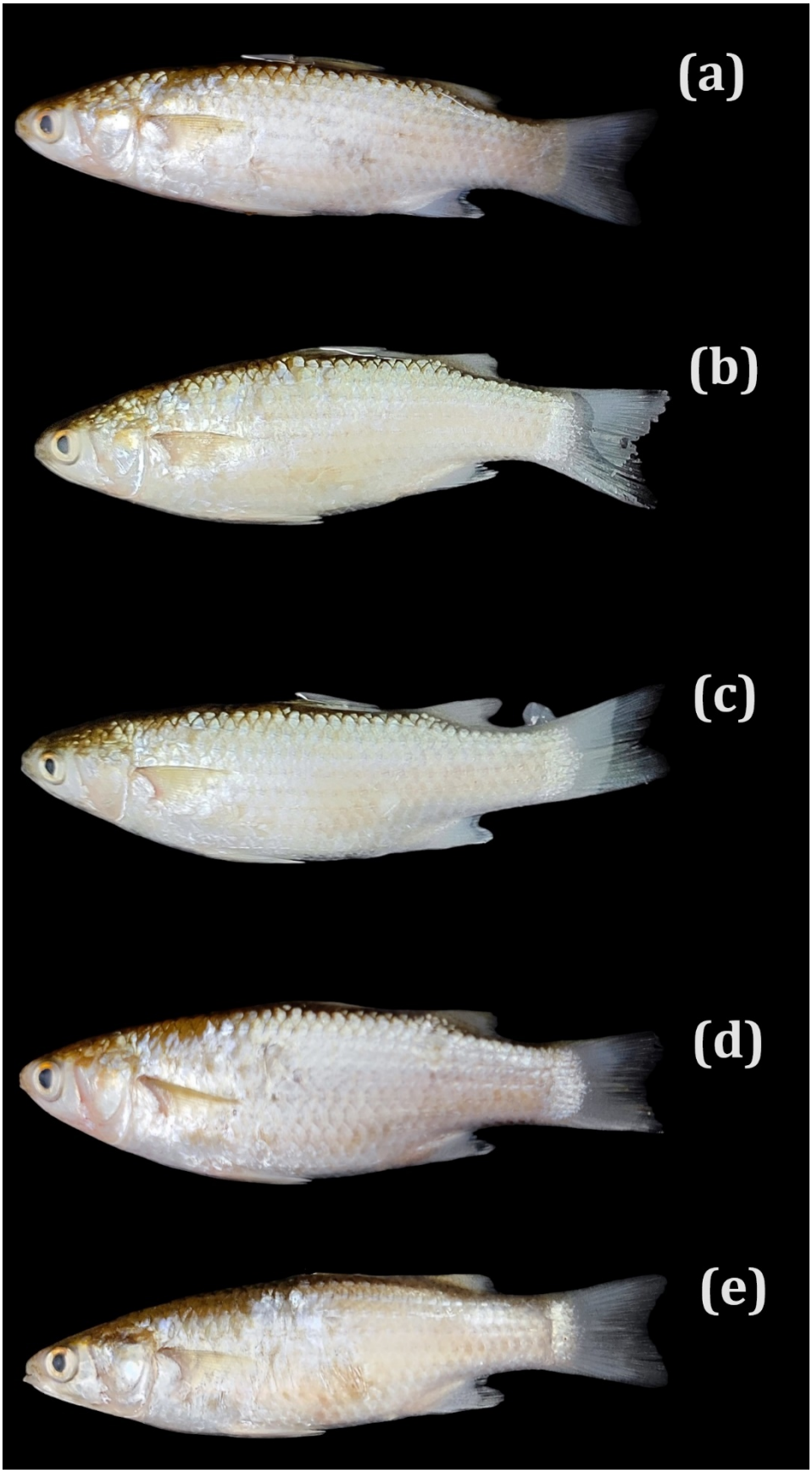


Figure 3.12: Observations of freshness index on Day-1

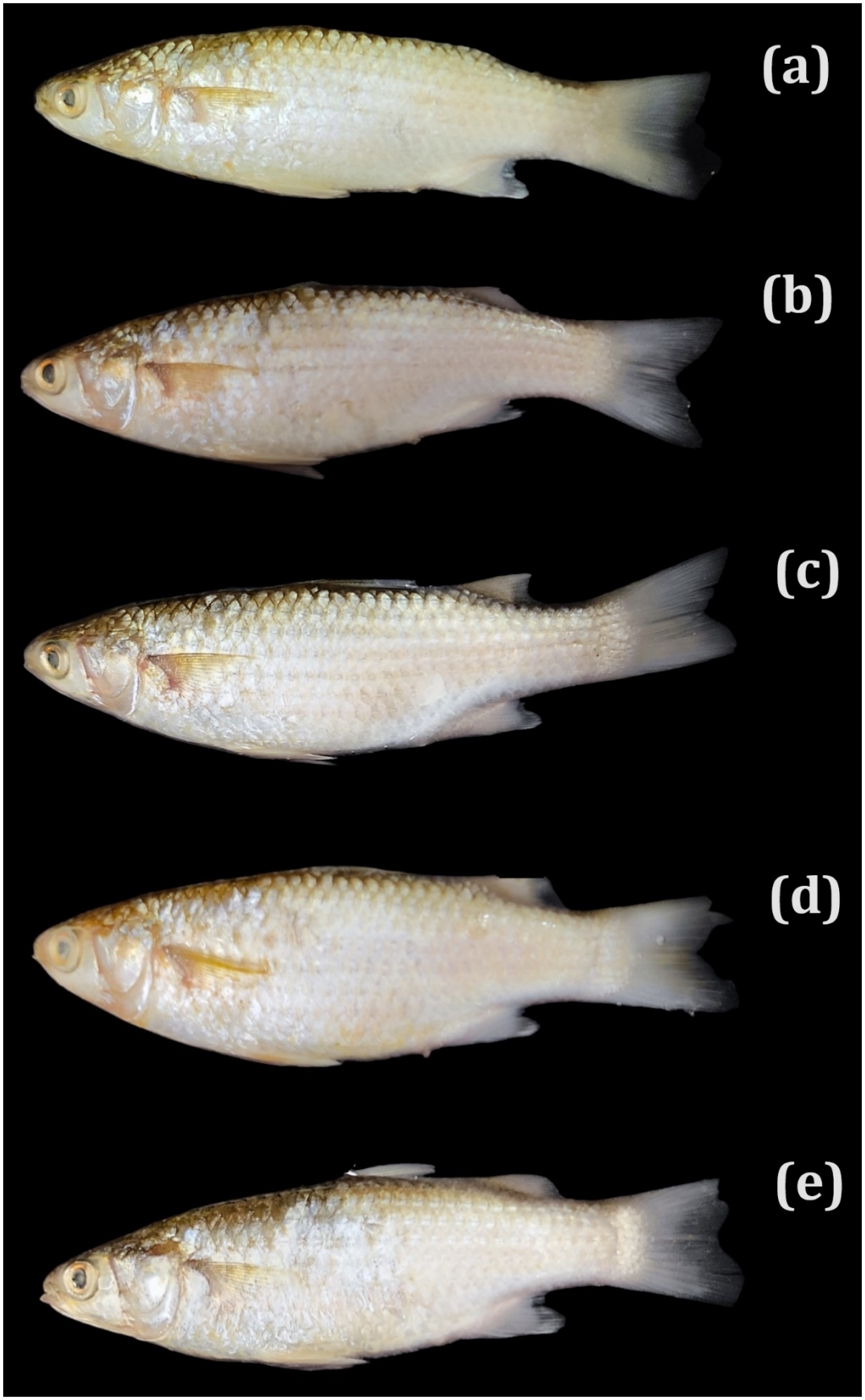


Figure 3.13: Observations of freshness index on Day-2

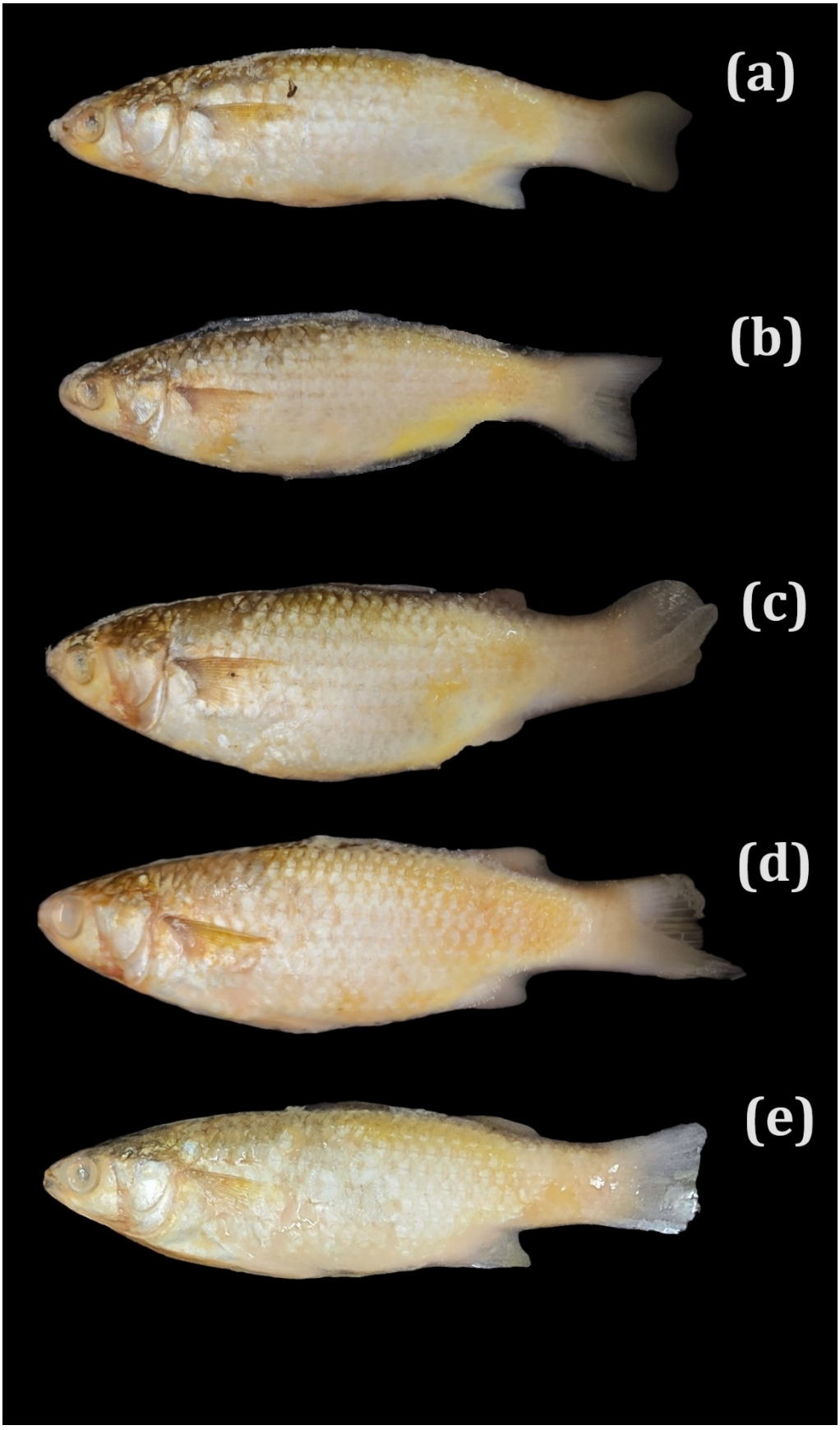


Figure 3.14: Observations of freshness index on Day-3

The proximate content analysis data revealed that the maximum conversion of the food was found in the T2 having a salinity of 20ppt followed by T3 (30ppt) and T1 (10ppt). Carbohydrate content showed variation and it was higher in T1 compare to T3 but remained low compare to T2 (Table 3.10). The average moisture content recorded from highest to lowest as follows: 78.10±0.41 (T2), 75.83±0.46 (T3) and 74.87±0.25 (T1). The average ash content was varied from highest to lowest as follows: 1.07±0.03 (T2), 0.97±0.04 (T3), and 0.91±0.07 (T1). The average lipid content was varied from highest to lowest as follows: 13.06±0.40 (T2), 12.53±0.38 (T3), and 12.41±0.36 (T1). The average protein content was varied from highest to lowest as follows: 54.70±2.21 (T2), 53.17±0.93 (T3), and 49.91±0.63 (T1). The average carbohydrate content was varied from highest to lowest as follows: 7.18±0.18 (T2), 7.09±0.20 (T1), and 6.55±0.24 (T3).

Table 3.9: Initial and final changes in growth of *M. cephalus* reared in various salinity range (10ppt, 20ppt, 30ppt) tanks for the period of six months

Growth Rate				
	Initial		**Final**	
Salinity (ppt)	**TL (cm)**	**TW (g)**	**TL (cm)**	**TW (g)**
TANK 1 (T1)				
10ppt	15.50	42.32	17.20	50.12
	15.60	44.08	17.40	52.57
	15.40	42.58	17.20	50.87
TANK 2 (T2)				
20ppt	15.60	43.76	18.10	50.86
	15.70	45.12	18.00	53.24
	15.40	41.98	17.70	51.42
TANK 3 (T3)				
30ppt	15.40	42.39	17.50	49.85
	15.40	40.75	17.50	50.36
	15.20	40.67	17.40	50.07

Table 3.10: Proximate content analysis of *M. cephalus* reared in various salinity range (10ppt, 20ppt, 30ppt) tanks for the period of six months

Proximate Content Analysis (%)					
Salinity	**Moisture**	**Ash**	**Lipid**	**Protein**	**Carbohydrate**
TANK 1 (T1)					
10ppt	74.60	0.89	12.39	49.25	7.01
	75.20	1.01	12.85	50.76	7.36
	74.80	0.84	11.98	49.71	6.89
TANK 2 (T2)					
20ppt	78.10	1.04	13.05	53.96	6.98
	78.60	1.11	13.56	57.71	7.42
	77.60	1.06	12.57	52.44	7.14
TANK 3 (T3)					
30ppt	76.30	1.01	13.01	53.01	6.35
	76.00	0.99	12.58	54.38	6.89
	75.20	0.92	12.09	52.11	6.41

3.4 DISCUSSION

The nutrient composition of fish, whether gathered from the wild or from aquaculture, exhibits significant variation overall. In intensive aquaculture, fish are given nutrient-rich compounded diets, which allows them to accumulate significant amounts of nutrients, especially lipids. Significant alterations in environmental conditions may result in variations of the availability and composition of food, which in turn can impact the nutrient composition of wild fish (Orban *et al.* 2007). The composition of nutrients varies across distinct body sections in various species. A study conducted by Baskaran (1993) found that the muscle serves as the primary storage organ for *Planiliza subviridis* (Valenciennes 1836), while the liver serves as the primary storage organ for *M. cephalus*.

In this study, the total ash content was shown to be greater in females compared to males. During the period from November 2021 to March 2022, there was an increase in the ash content in men. However, starting from April 2022, there was a dramatic decrease in the ash content. Females had elevated ash concentration from October 2021 to January 2022, followed by a decrease starting from February 2022. An additional surge in ash content was reported in June 2022.

The moisture content was found to be greater in females compared to males. The moisture content decreased after the monsoon and during the winter season, but increased during the monsoon season, which is when the *M. cephalus* spawns. According to Polat *et al.* (2007), there is a higher availability of nutrients during the post-monsoon and post-winter seasons compared to winter. This leads to an increase in the amount of dry matter and a decrease in moisture content during these seasons.

In the case of the gonads, proteins were the primary components of the liver, followed by lipids, and carbohydrates were the least abundant. Despite the common belief that the liver is a storage organ for glycogen, carbohydrates were found to be present in lower quantities. The muscle protein content was shown to be greater in males compared to females. During the post-spawning and winter season (November 2021 to March 2022), the protein content was shown to be greater. A little increase in protein content was detected in April-22 and June-22

prior to the onset of gonadal maturation. The deposition of protein in muscle served as the preliminary phase for the gonadal maturation cycle. Protein content experienced a decrease in July-August, but once the breeding season resumed, there was an increase in protein content. The increment was greater in females compared to males. A similar pattern was detected in the protein level of the liver, with the liver having a higher protein concentration. During the beginning of the breeding season, the protein content in females drops to a greater extent than in males. Analysis revealed a significant rise in the protein content of red mullet and a decrease in moisture content during the spring season ($P < 0.05$). The protein level in fish muscle varied based on the amount of fish food, and there exists a negative correlation between moisture and protein content (Hall & Ahmad 1997; Özyurt *et al*. 2005; Polat *et al.* 2007).

The lipid content in the muscles of *M. cephalus* remained constant compared to the other proximate components. There was a slight decrease in the lipid content of the muscle during the beginning of gonadal development, and this decrease was more pronounced in females compared to males. A significant decrease in liver lipid content was found in females starting in July 2021, which was larger compared to the fall in muscle lipid content. Sargent (1995) found that the fluctuations in lipid levels in fish were primarily linked to their reproductive cycle. The spawning period of red mullet in the Mediterranean took place from May to August, as reported by Özyurt (2003). In the current study, it is believed that lipids in the muscle of red mullet were utilised for gonadal development during the spring season (3.68%) and subsequently increased after spawning (5.76% in autumn, 5.33% in winter). Bandarra *et al.* (2001) found similar outcomes for horse mackerel (*Trachurus trachurus*), Grigorakis *et al.* (2002) for sea bream (*Sparus aurata*), and Özyurt & Polat (2006) for sea bass (*Dicentrarchus labrax*).

The carbohydrate content was detected in only a small quantity in both the male and female populations of *M. cephalus*. A little variation was observed in the composition of muscle carbohydrates. In females, muscle glycogen levels rise during the onset of the gonadal maturation cycle and decline during the spawning season. No significant alterations in muscle glycogen content were detected in

males, with the exception of a slight reduction during the summer season. This may be attributed to limited dietary availability and environmental conditions (Kumaran *et al.* 2012). The liver's glucose content exhibited variations when compared to muscles. The female population exhibited a rise from July to September, followed by a decline throughout the breeding season. Male liver carbohydrate content increased after monsoon, suggesting a surplus of food materials, which began to decline in the summer and showed a slight increase before to the onset of the breeding season.

According to Baskaran (1993) a high hepatic index is often accompanied with a high proportion of proteins, indicating a stronger contribution of proteins to the enhancement of the liver. In addition to proteins, lipids also appear to play a significant role in the initial stage of liver development. Although the liver is typically considered a crucial organ for storing carbs in the form of glycogen in vertebrates, in this particular situation, it did not seem to play a significant role, except for a slight decrease in glucose levels when proteins and lipids were low (Baskaran 1993).

Several studies have demonstrated the correlation between hepatosomatic index and gonadal maturation. According to Bailey (1952) report, female Baltic herring with higher fecundity possessed livers that were more developed than those of females with lower fecundity. It has been shown in certain species that the liver size grows as the size of the ovaries increases, and reduces when the ovaries reach maturity (Baskaran, 1993). Indian scientists have conducted proximate content study on *M. cephalus* in many regions of India, including south India and the east coast (Miranda *et al.* 2011; Kumaran *et al.* 2012; Rowshan-Ali *et al.* 2014; Mohanty *et al.* 2015; Ali *et al.* 2017).

The freshness index and organoleptic study are essential instruments for guaranteeing quality assurance in aquaculture. Aqua culturists can maintain stringent quality standards by closely monitoring freshness indicators and conducting sensory assessments. This ensures that only fresh and high-quality fish products are sold to consumers (Huss 1995). The quality of fish has a direct impact on how satisfied consumers are and the choices they make when buying it. Consumers are more inclined to prefer products that have exceptional sensory

qualities, which in turn leads to higher sales and more market demand. Aqua culturists can increase customer contentment and develop brand loyalty by giving priority to freshness and sensory quality. Evaluating the state of being fresh is of utmost importance in guaranteeing the safety of food in aquaculture (Huss 1995). Multiple research has been conducted on the sensory analysis of *M. cephalus* and other species, proposing different preservation strategies (Afolabi 1984; Mohamed & Mohamed 2019; Tsogas *et al*. 2019; Yehia *et al*. 2022).

Several studies have examined the impact of varying degrees of salt on fish physiology, including research conducted by Boeuf and Payan (2001), Aragão *et al.* (2010), and Gholampoor *et al.* (2011). However, there is only one previous study specifically focused on *M. cephalus* (Rabeh *et al.* 2010). Fazio *et al.* (2013) conducted a study that documented various alterations in the haematological and biochemical parameters of the commonly cultivated mullet species, *M. cephalus*. They observed a reduction in red blood cells (RBC), white blood cells (WBC), haemoglobin (Hgb), hematocrit (Hct), glucose levels, etc. as a result of osmoregulatory dysfunction. Girling *et al*. (2003) and Montero *et al.* (1999) also reported similar findings. Amino acids are crucial for fish to adapt to varying ambient salinities, serving as both energy sources and essential osmolytes for regulating cell volume (Aragão *et al.* 2010). The animals grown in tank 2 with a salinity of 20ppt exhibited the highest level of growth in the current investigation. The same study in Chapter 2 found similar results, with *M. cephalus* of Bhadbhut exhibiting comparable optimal growth conditions and biochemical compositions. The environmental parameters of the Bhadbhut area, particularly the salinity, were measured and found to fluctuate approximately 20 parts per thousand (ppt), which is consistent with the experimental settings described in chapter 2.