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Mapping and characterization of dominant hydrolase enzymes in the digestive tract of Striped marlin (*Kajikia audax* Philippi, 1887)

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Abstract. *Ismail E, Prihanto AA, Sukoso, Kartikaningsih H. 2025. Mapping and characterization of dominant hydrolase enzymes in the digestive tract of Striped marlin (*Kajikia audax *Philippi, 1887). Biodiversitas 26: 1156-1163.* The digestive tract of Striped marlin (*Kajikia audax* Philippi, 1887): a fishery byproduct, contains several hydrolase subclass enzymes, such as protease, lipase, amylase, cellulase, and xylanase, which have practical applications in medicine, agriculture, and various industries, including pharmaceuticals, food, detergents, leather, and cosmetics. This study aimed to identify and characterize the enzymes in the digestive tract of Striped marlin. Enzymes from the hydrolase sub-class in different parts of the digestive tract, such as the stomach, pylorus, pancreas, and intestine, were analyzed separately using qualitative and quantitative methods. Each enzyme was further evaluated to determine its optimal pH and temperature. The results revealed that protease activity was the most dominant among the other enzymes. A clear zone with a diameter of >85 mm was observed on a casein substrate in the stomach and pylorus. The highest protease activity was recorded in the part of the stomach (1,044.71 U/mL): followed by the pylorus (937.21 U/mL): pancreas (492.06) U/mL, and intestine (362.21 U/mL). The optimal pH for enzyme activity was 4 in the stomach, 6 in the pylorus, 7 in the pancreas, and 10 in the intestine. The optimal enzyme activity temperature across all digestive tracts was 40°C. This study provides the first comprehensive mapping of hydrolase subclass enzymes in the digestive system of Striped marlin.

Keywords: Enzyme, digestive tract, hydrolase, marlin, protease

INTRODUCTION

The Striped marlin (*Kajikia audax* Philippi, 1887) is a species of billfish belonging to the Istiophoridae family. It can be found in the Indonesian region, particularly in the Indian Ocean and the Western Pacific. The population of Striped marlin accounts for 15 to 35% of the global population of this species (Mamoozadeh et al. 2020). As a bycatch in the longline tuna fishing industry, it accounts for 5% of the total catch (Fahmi et al. 2019), with a volume of 22,149,931 tons in Indonesia, according to a report in 2021. The gut content was removed before landing the catch for preservation and storage. These byproducts still hold considerable economic value due to their composition. Fish waste contains 15% to 30% protein, 0% to 25% fat, and 50% to 80% water (Ideia et al. 2020). Digestion of nutrients in fish requires enzymatic activity in the digestive tract, which could have added value with specific treatments and conditions. Industrial enzymes included in the hydrolase class are proteases, lipases, cellulose, and xylanases (Porto de Souza Vandenberghe et al. 2020). More than 70% of all enzymes in the industrial sector, such as detergents, leather, textiles, pulp and paper, food and animal feed, dairy, biofuels, and waste processing industries, all belong to the hydrolase class. Amylase, cellulase, and xylase are the second-largest groups of enzymes used in the industry (Shukla et al. 2022).

Commercial cellulase, which is widely applied in the food industry (Ejaz et al. 2021), is a complex mixture of several enzymes that are difficult to separate (Wang et al. 2023) and catalyzes the cellulose hydrolysis process (Nero et al. 2022). Cellulase is applied in fermentation to utilize food byproduct waste to convert cellulose fibers into soluble sugars through fermentation into bioethanol (Zou et al. 2020). One application of xylanase is the hydrolysis of xylan into xylose; in the pharmaceutical industry, xylanase is utilized as a sugar substitute for people with diabetes (Dhaver et al. 2022). Striped marlin, an opportunistic carnivore, preys on various pelagic fish containing fats, which are essential for synthesizing lipase in the pancreas in the form of bile salts (Tang et al. 2022). One of the applications of fat hydrolysis by lipase is in treating industrial wastewater, mainly from the milk and vegetable oil industries. Among the subclasses of hydrolase enzymes mentioned above, protease is the most dominant and versatile.

Enzyme applications are increasingly gaining attention in medicine, agriculture, and industry, including pharmaceuticals, food, detergents, leather, and cosmetics. Enzymes are utilized in these industries to enhance product durability, reduce process time, improve efficiency and stability, and manage waste effectively (Dahiru et al. 2024). However, from an economic perspective, enzyme production requires large-scale biomass, the availability of raw materials, regulatory and market considerations, and an assessment of potential environmental impacts (Jenol et al. 2024). Several challenges in enzyme production include the bitter taste of enzymes produced from plants (Jioe et al. 2023), the

consideration of halal standards, and the use of non-halal enzyme sources, such as those derived from pigs or improperly slaughtered cows (Prihanto et al. 2016). Using proteases derived from microbes containing recombinant pig and human DNA poses further obstacles to applying these enzymes in Indonesia (Jaswir and Mahfudh 2022).

Previous studies carried out to obtain alternative enzyme sources from fish include Yellowfin tuna (*Thunnus albacares* Bonnaterre 1788) (Pasaribu et al. 2018; Nurjanah et al. 2021), Yellow pike conger (*Congresox talabon* Cuvier 1829) (Putra et al. 2021), and Mackerel (*Euthynnus affinis* Cantor 1849) (Nurhayati et al. 2020). The need for the availability of enzymes in large quantities and efforts to utilize the digestive tract of fish, which has been considered waste, makes it essential to find alternative enzyme sources. Therefore, this study aimed to identify and characterize the enzymes that dominate the digestive tract of Striped marlin.

MATERIALS AND METHODS

Research material

In this study, the materials used were the digestive tract of Striped marlin randomly taken from suppliers in various regions in Indonesia, namely Tanggamus District, Lampung Province; Sorong City, West Papua Province; and Bone District, South Sulawesi Province, Indonesia. The material in this study includes distilled water, Skim Milk Agar, Nutrient agar, casein, Virgin Coconut oil (VCO): amylum, Carboxymethyl Cellulose (CMC): Xylan, buffer-HCl 0.05 M pH 9.0, and other analytical materials written in the research procedure. All materials used analytical grade from Sigma-Aldrich with the distribution of Merck Chemical and Life Sciences-Indonesia.

Sampling and preparation

The length of the fish was measured (cm) and weighed (kg) to determine the average length and weight. The digestive tract consisting of the stomach, pylorus, pancreas, and intestines were washed with clean water, packed in clean polyethylene plastic bags, and weighed to obtain the ratio between the total weight and the weight of the digestive tract in percent. After that, the ice was added with a 1:2 sample/ice (w/b) to reach a temperature of 0°C, then frozen at -20°C and stored for no more than 3 months. Samples were brought to the advanced laboratory of the Faculty of Fisheries and Marine Science, Universitas Brawijaya, Malang, Indonesia.

Research procedures

Extraction

Frozen samples were thawed with cold water until the temperature reached 0°C and cut with a size of ± 1 cm to be extracted. The extraction method was followed (Borges et al. 2023). The samples were homogenized with 0.01 M cold Tris-HCl buffer (pH 8) and 0.01 M $CaCl_2$ with a sample:buffer ratio of 1:2 (b/v). After homogenization in a cold atmosphere (4°C): the sample was filtered with Whatman paper no 42. The sample was centrifuged at 4°C at 10,000 rpm for 15 minutes. The supernatant containing the enzyme was stored at -20°C before being analyzed as a crude extract.

Qualitative test

The enzymatic activity was determined by using a 6 mm diameter paper disc soaked in 50 µL of enzyme crude extract, deposited aseptically on agar media containing substrate with the Kirby-Bauer disk diffusion method (Santajit et al. 2022) with the composition presented in Table 1.

Quantitative test

Analysis of protease activity

Protease activity was conducted following the protocol outlined by Yi et al. 2023). 0.5 mL crude extract was mixed with 0.5 mL of 2% casein in 0.05 M HCl buffer at pH 9.0. The mixture was incubated at 40°C for 20 minutes. One mL of 0.4 M trichloroacetic acid (TCA) was added to terminate the reaction. For the blank control, TCA was added to distilled water before incubation. The mixture was centrifuged at 5,000 rpm for 15 minutes and subsequently allowed to stand at room temperature for another 15 minutes. Next, 0.5 mL of the supernatant was mixed with 2.5 mL of 0.4 M sodium carbonate (Na_2CO_3) and 0.5 mL of Folin-Ciocalteu reagent. The solution was incubated statically with flow to maintain temperature at 40°C for 20 minutes, and the absorbance was measured at a wavelength of 680 nm. One protease activity (U) unit is required to hydrolyze casein and produce 1 µg of tyrosine.

Table 1. Agar medium and substrates were used to qualitatively analyse protease, lipase, amylase, cellulase, and xylanase

Enzyme	Media	Substrate	Dye	Reference
Protease	Skim milk agar 2%	Casein 10 g/L	-	Zhang et al. (2019); Nadaf and Hivrale (2023)
Lipase	Nutrient agar 2%	Virgin Coconut Oil (VCO) 20 mL/L	Rhodamin B	Molitor et al. (2020)
Amylase	Nutrient agar 2%	Starch 30 g/L	Lugol	Hashemzahi et al. (2020)
Cellulase	Nutrient agar 2%	Carboxylmethyl Cellulose (CMC) 5 g/L	Congo red	Hashemzahi et al. (2020)
Xilanase	Nutrien agar 2%	Xylan 1 g/L	Congo red	Ghosh et al. (2020)

Analysis of lipase activity

Lipase activity was determined using the spectrophotometric method, as described by Van Gaelen et al. (2021). The substrate used for this assay was VCO. An emulsion was prepared using 50 g of gum arabic dissolved in 1 L of a 0.05 M KH_2PO_4/K_2HPO_4 buffer at a pH of 8, then heating at 50°C for 3 hours. If possible, include β-cyclodextrin at a concentration of 3 g/L. Use a glass fiber-coated filter with a pore size of 0.6 µm. Dissolve 25 mL of VCO in 1 L of gum arabic emulsion and mix uniformly at 10,000 rpm for 10 minutes. Adjust the mixture to a pH of 7 by adding 1 M NaOH. Add 1 mL of rhodamine B solution (prepared by dissolving 1 g of rhodamine B in 1 L of distilled water). Store the emulsion in a glass container at 4°C for 2 days. Use an enzyme-to-substrate ratio of 3:10 and maintain the mixture at ambient temperature for 10 minutes with continuous stirring. The absorbance of the solution was quantified using a 580 nm spectrophotometer. A unit of lipase is defined as the quantity of protein that converts 1 µmol of palmitate into 1 mL of lipase per minute, expressed as Unit/mL.

Assessment of amylase activity

The procedure for measuring amylase activity established by Gómez-villegas et al. (2021) and Nolasco-Soria (2021) was as follows: A 100 µL crude extract was introduced into a test tube containing a 500 µL starch solution (0.4 mg per mL in a 0,02 M phosphate buffer, pH 7). After incubating at 37°C for 30 minutes, the reaction was halted by adding 500 µL of a 0.01 M iodine solution and 3000 µL of distilled water. A blank solution of 100 µL distilled water was added with the iodine solution. Absorbance was measured at a wavelength of 660 nm. A unit of amylase activity is the quantity of amylase present in 100 mL of Crude Extract (CE) that breaks down 10 mg of starch substrate in 30 minutes at 37°C.

Quantification of cellulase activity

100 µL of crude extract was mixed with 4.9 mL of CMC solution (1% in 0.1 M acetate buffer, pH 5.0) and incubated at 25°C for 70 minutes. After incubation, approximately 1 mL of the mixture was taken, and 3 mL of 1% 3,5-dinitrosalicylic acid (DNS) reagent was added. The mixture was then heated at 90°C for 10 minutes (Deshavath et al. 2020). The absorbance was measured using a UV-Vis spectrophotometer at 540 nm.

Measurement of xylanase activity

Xylanase activity was evaluated using the DNS method to quantify reducing sugars. The protocol described by Dhaver et al. (2022) adding 600 µL of a 1% (w/v) xylan solution (prepared by dissolving 1 g of xylan in 100 mL of 0.05 M citrate buffer, pH 5.0) was put into a 15 mL test tube. Subsequently, 100 µL of the supernatant was added to the tube. The reaction mixture was incubated at 55°C for 15 minutes, followed by adding 1 mL of DNS reagent. The mixture was then heated at 100°C for 5 minutes. After cooling, the solution was diluted with 3 mL of distilled water. The concentration of reducing sugars in the solution was measured using a spectrophotometer at a wavelength of 575 nm. One unit of xylanase activity is defined as the amount of enzyme required to release 1 µmol of reducing sugar per minute per milliliter of enzyme solution under the assay conditions (Miller 1959).

Characterization

Impact of varying pH levels and incubation temperatures

The effect of various pH in incubation on the activity of crude enzyme extracts was evaluated using appropriate substrates within a pH range of 4-11 at a temperature of 40°C for 20 minutes. The buffers used included 0.1 M glycine-HCl (pH 4,0): 0.1 M Potassium phosphate (KH_2PO_4 and K_2HPO_4) (pH 5.0-6.0): 0,1 M Tris-HCl (pH 7.0-9.0) and 0,1 M glycine-NaOH (pH 10-11). The effect of different temperatures (30-70°C) of incubation was determined based on the method described by Nalinanon et al. (2010).

Data analysis

Enzyme activity values are expressed as the mean ± standard deviation, and their Relative Standard Deviation (RSD) values were calculated. The results were evaluated using one-way ANOVA, with differences between groups considered significant at a 95% confidence level ($p<0.05$). All experiments were performed in three replicates.

RESULTS AND DISCUSSION

The length and weight of the samples

The body length, body weight, total weight, and the weight of each part of the digestive tract of Striped marlin from three different locations, namely Tanggamus District, Lampung Province; Bone District, South Sulawesi Province; and Sorong City, West Papua Province, Indonesia have the ratio of the total weight of the digestive tract to body weight was 6.78%, in sample 1, 8.02% in sample 2, and 6.03% in sample 3 (Table 2 and Figure 1.A).

Table 2. Composition of length and weight for each compartment of the Striped marlin

Samples	Length (cm)	Body weight (kg)	The weight of each part (kg)				Average weight (kg)
			Stomach	Pyloric caeca	Pancreas	Intestine	
1	167	39	0.940	1.450	0.166	0.087	2.643
2	201	53	1.750	2.133	0.265	0,105	4.253
3	154	29	0.653	0.922	0.098	0.076	1.750
		$\bar{x}$ =40.33	$\bar{x}$ =1.11433	$\bar{x}$ = 1.50157	$\bar{x}$=0.17633	$\bar{x}$ =0.8933	$\bar{x}$=2.299

Table 2 shows that the mean weight of the digestive tract in Striped marlin is as follows (i) the pyloric caeca (1.501 kg): stomach (1.114 kg); (ii) pancreas (176.33 g); and (iii) intestine (89.33 g). The proportion of the average weight of the digestive tract to the overall weight of the fish is 7,145%. It is higher than the proportion of Yellowfin tuna (*T. albacares*) (6.1%) (Krishnan et al. 2024). Swordfish have an average stomach-weight to total body-weight ratio of 3% (Hatzonikolakis et al. 2021). The ratio of stomach to body weight varies widely depending on species, age, and the type of diet (e.g., the size and quantity of prey).

The characteristics of the digestive tract in Striped marlin are illustrated in Figure 1.B. The esophagus is connected to the stomach (i): which is higher in volume than other parts. The pyloric caeca (ii) is adjacent to and attached to the stomach. A connecting channel exists between the pylorus and the pancreas (iii). This section is parallel to the initial part of the intestinal tract (iv). The intestine is small and short, with gonads attached asymmetrically at the end, a distinctive characteristic of the Striped marlin species.

Qualitative test of enzyme activity

The diameter of the clear zone in Table 3 represents the enzyme activity of crude extract on each substrate. The findings showed that protease has a wider clear zone than lipase, amylase, cellulase, and xylanase.

Quantitative analysis of enzymes activity

Table 4 shows that the protease subclass exhibited the highest activity, with a value of 1,013.82 U/mL in the stomach, 939.27 U/mL in the pylorus, 487.50 U/mL in the pancreas, and 361 U/mL in the intestine. The second-highest activity in the digestive tract of Striped marlin was observed for lipase, with a value of 17.47 U/mL in the pylorus, 16.31 U/mL in the intestine at 16, 14 U/mL in the stomach and 15.59 U/mL in the pancreas. Meanwhile, the activity of amylase, cellulase, and xylanase ranged from 2.85 U/mL to 5.22 U/mL.

Characterization

Variation of pH on enzyme activity

The effect of pH changes on protease optimal activity in the digestive tract of the Striped marlin (Figure 2.A) were as follows (i) pH 4 in the stomach, the pylorus at pH 5; (ii) the pancreas at pH 6; (iii) and the intestines at pH 9. Lipase activity (Figure 2.B) in the stomach, pylorus, pancreas, and intestines was optimal at pH 4, 8, 9, and 7, respectively. Optimal amylase activity (Figure 2.C) was observed in the stomach at pH 4, in both the pylorus and pancreas at pH 10, and in the intestines at pH 5. For cellulase activity (Figure 2.D): the optimal pH was 4 in the stomach and intestines, pH 5 in the pancreas, and pH 9 in the pylorus. Finally, overall xylanase activity (Figure 2.E) was optimal at low pH, specifically in the stomach at pH 5 and other parts at pH 4.

Table 3. Results of qualitative tests on the activity of hydrolase enzyme sub-classes

Enzyme	Parts of the digestive system	Clear zone
Protease	Stomach	+++
	Pyloric caeca	+++
	Pancreas	++
	Intestine	++
Lipase	Stomach	++
	Pyloric caeca	++
	Pancreas	++
	Intestine	++
Amylase	Stomach	+
	Pyloric caeca	+
	Pancreas	+
	Intestine	+
Cellulase	Stomach	+
	Pyloric caeca	+
	Pancreas	+
	Intestine	+
Xylanase	Stomach	+
	Pyloric caeca	+
	Pancreas	+
	Intestine	+

Notes: +++: diameter > 85 mm, ++: 75-85 mm, +: 60-74 mm

Table 4. The activities of protease, lipase, amylase, cellulase, and xylanase from the hydrolase sub-class in the stomach, pyloric caeca, pancreas, and intestine in the digestive tract of Striped marlin

Organ	Activity (U/mL)				
	Amylase	Cellulase	Xylanase	Lipase	Protease
Stomach	3.96	4.03	4.52	16.14	1,013.82
Pyloric caeca	4.03	2.85	3.1	17.47	939.27
pancreas	5.22	3.76	3.1	15.96	487.5
Intestine	3.93	4.37	4.01	16.31	361.32

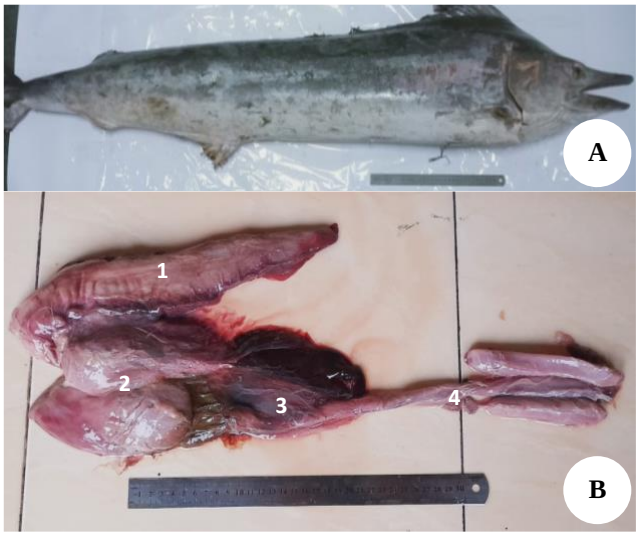

Figure 1. A. Striped marlin with complete removal of the beak and tail; B. The anatomical segment of the digestive tract. 1: Stomach, 2: Pyloric caeca, 3: Pancreas, 4: Intestine

Effect of temperature on enzyme activity

The effect of temperature on protease activity (Figure 3.A) throughout the digestive tract of the optimum Striped marlin showed that the optimum temperature is 40°C, which is also the optimal temperature for lipase activity (Figure 3.B): amylase activity (Figure 3.C) and cellulase activity (Figure 3.D). However, xylanase activity (Figure 3.E) was different, with its optimal temperature being 50°C in the stomach, pylorus, and intestines, while the pancreas has an optimum temperature of 60°C.

Discussion

There is a positive correlation between the length and weight of the fish (Figure 1.A and Table 2). The samples collected represent different stages of fish development. During the development period, an increase in length ranges from 140 cm to 210 cm (Chang et al. 2018). The digestive system of carnivorous fish exhibits several distinct characteristics, such as a shorter digestive tract than herbivorous and omnivorous fish, a large stomach volume, and the pyloric caeca located adjacent to the stomach, which delivers nutrients. Carnivorous fish typically have a large stomach volume and a pylorus near the relatively small intestines. The proportion of the digestive tracts to average weight is 7.145%, much higher than that of Yellowfin tuna and Swordfish. The ratio of stomach to body weight varies widely depending on factors such as species, age, and the type of diet (e.g., the size and quantity of prey).

Protease activity is higher than other enzyme activities, indicated by a wider clear zone. The protease activity is higher than other enzyme activities because the casein substrate is more suitable for detecting protease activity (Zhang et al. 2021). The structure of the digestive tract corresponds to the feeding habits of fish, where food digested by gastric acid moves to the anterior section, including the pyloric membrane, and then to the midsection of the intestine (Cho et al. 2023). The dimensions of each part of the digestive tract (Figure 1.B) are in line with the results of Young et al. 2018) and Liu et al. 2022), which show that the Striped marlin is a carnivorous fish with a denser and wider pyloric caeca compared to other parts. These factors are affected by the feeding habits of fish, which shape the structure of the digestive tract and gut microflora (Jiao et al. 2023).

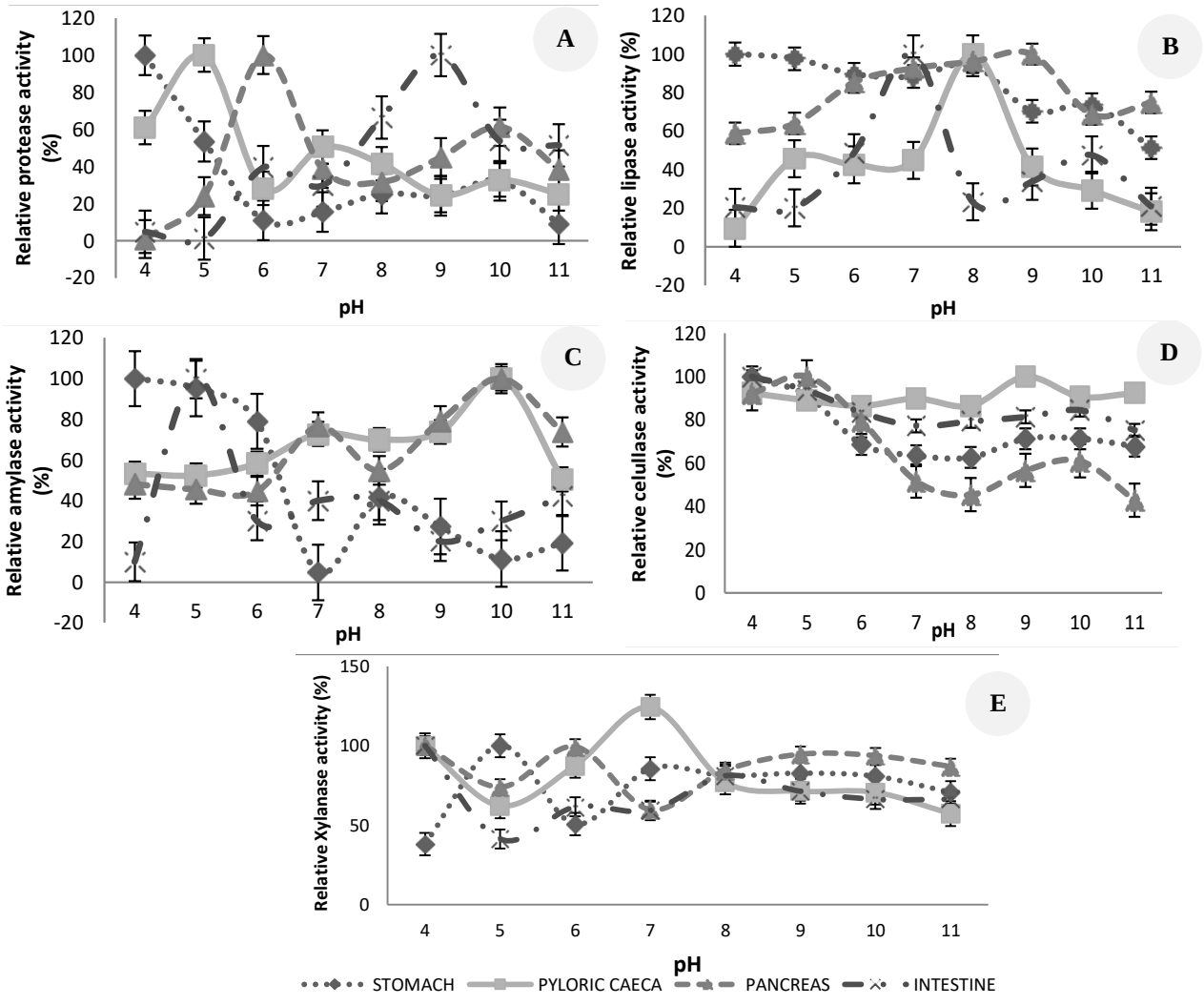

Figure 2. Relative enzyme activity with pH variations. A. Protease; B. Lipase; C. Amylase; D. Cellulase and e-xylanase

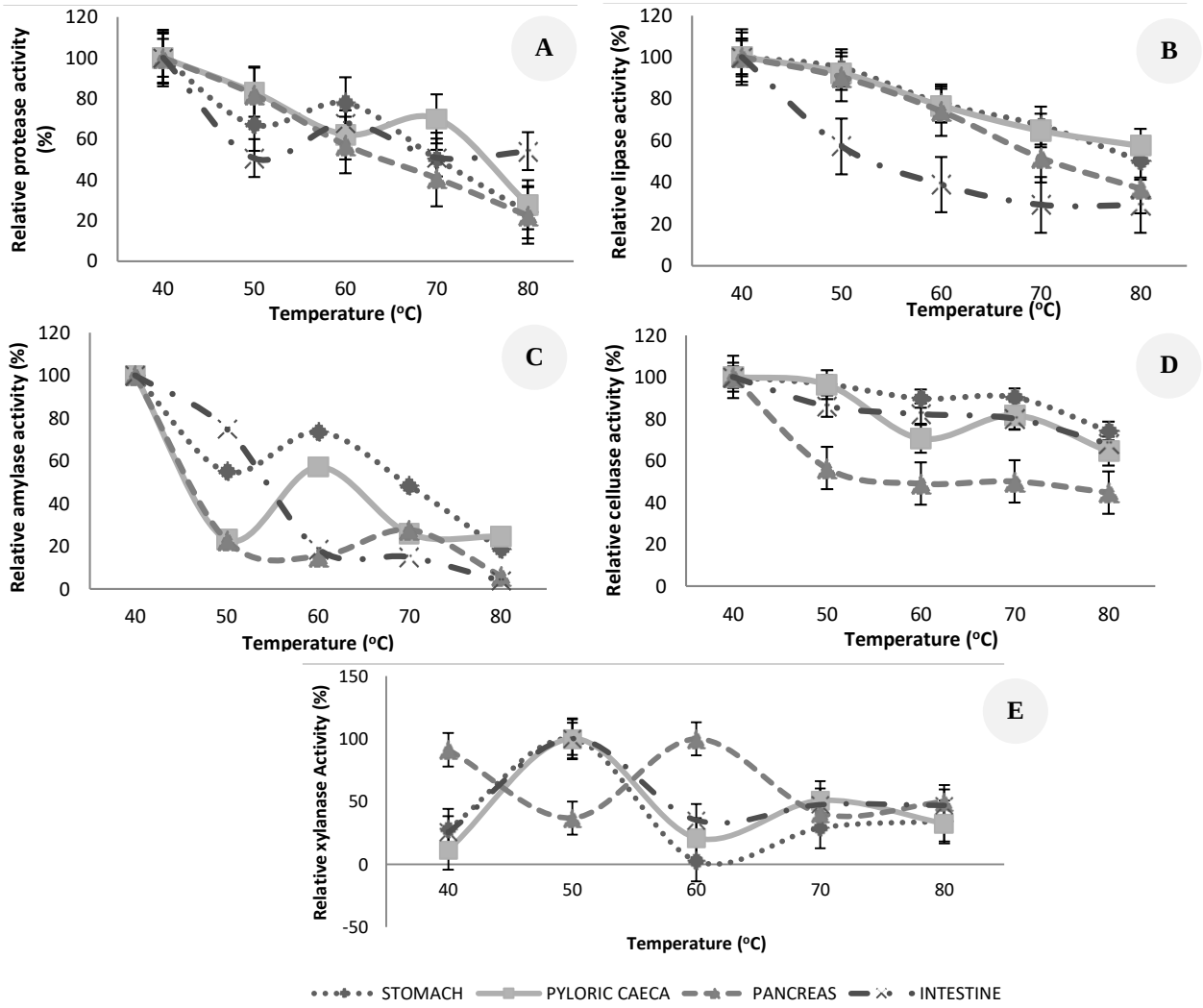

Figure 3. Comparative enzymatic activity at different temperatures. A. Protease; B. Lipase; C. Amylase; D. Cellulase; E. Xylanase

The hydrolase activity in the digestive system of fish is affected by the feeding behaviors of Striped marlin, as opportunistic carnivores that generally eat fish such as small pelagics, cephalopods, and crustaceans that are mainly composed of protein, fat, and carbs (Young et al. 2018, Hansen et al. 2022). The function of the digestive enzymes is to rapidly break down food into simpler constituents that can be easily absorbed for growth (Zhou et al. 2020; García-Meilán et al. 2023; Skvortsova et al. 2025). Table 4. shows that protease activity is higher than lipase, amylase, cellulase, and xylanase; this result aligns with qualitative analysis of enzyme activity (de la Parra et al. 2007, Lal et al. 2024).

The stomach, pyloric caeca, and pancreas exhibit ideal protease activity (Figure 2.A) at pH 4, 5, and 6, respectively. Meanwhile, the ideal pH in the intestine is 9. This finding is supported by the study conducted by Taghizadeh Andevari et al. (2019) and Nurhayati et al. (2024), which found that rainbow trout (*Oncorhynchus mykiss* Walbaum 1792) viscera activity was optimum at pH 8. Furthermore, Chong et al. (2002) showed a similar trend, where the stomach of the discus (*Symphysodon aequifasciatus* Pellegrin 1904) had an optimum pH of 2, while the intestine had a pH of 8. Enzyme activity in the digestive system of Striped marlin is characterized by the dominance of protease over lipase, amylase, cellulase, and xylanase in the stomach, pyloric caeca, pancreas, and intestines. Different parts of the gastrointestinal tract in fish had different optimum pH for the same enzyme due to the complex nature of digestive physiology and adaptation to diverse dietary habits. The stomach typically exhibits an acidic environment, with pH values ranging from 3.5 to 4.5 in species like perch and zander. This low pH is optimal for pepsin activity, which shows maximum activity at pH 2-3. The intestinal regions maintain a more alkaline environment, with pH values between 6.5 and 7.2 (Solovyev et al. 2015). This pH gradient along the digestive tract allows for the optimal functioning of different enzymes at various stages of digestion. Interestingly, the optimum pH for enzyme activity does not always correlate with the actual pH in the digestive tract.

For instance, alkaline protease and nonspecific lipase show maximum activity at pH 8-9, higher than the physiological pH observed in the intestine (Solovyev et al. 2015). Similarly, in Atlantic salmon, the pH optimum for

pepsin (3.0) is lower than the average stomach chyme pH (4.8). The pH optimum for total proteolytic activity in the proximal intestine (8.8) is higher than the prevailing chyme pH (Krogdahl et al. 2015). This discrepancy suggests that fish have evolved mechanisms to maintain enzyme activity across pH values, allowing for efficient digestion throughout the gastrointestinal tract. In short, the varying pH optima for the same enzyme in different parts of the fish gastrointestinal tract reflect the complex interplay between enzyme function, dietary adaptations, and physiological conditions. This diversity in pH optima enables fish to efficiently digest various nutrients under varying environmental and nutritional conditions, contributing to their adaptability and survival in diverse aquatic ecosystems. Exploring further purification and characterization of protease as the predominant enzyme is highly valuable for future investigation into protease's features and functional properties for application in diverse sectors. The impact of temperature on fish digestive enzyme activities appears to be species-specific, as the optimal temperature for enzyme activity generally aligns with the temperature range of the fish's natural habitat (Volkoff and Rønnestad 2020). The effect of temperature on protease activity (Figure 3.A): lipase activity (Figure 3.B): amylase activity (Figure 3.C) and cellulase activity (Figure 3.D): showed an overall optimum at 40ºC. Meanwhile, xylanase activity (3.E) was optimal at 50ºC in all parts except the pancreas, where peaked at 60ºC. This way slightly lower than the findings of Dhaver et al. (2022): who reported that xylanase activity from *Trichoderma harzianum* purified at different concentrations exhibited optimal activity at 65°C.

In conclusion, protease activity in the digestive tract of Striped marlin was higher than that of lipase, amylase, cellulase, and xylanase activity. Protease activity in the stomach and pylorus was optimum at acidic pH; however, the optimum pH of protease activity in the pancreas and intestines was alkaline. Lipase activity was optimum at alkaline pH in all parts, amylase in the stomach and intestines was optimum at acidic pH, and pylorus and pancreas were optimum at alkaline pH, cellulose activity in all parts was optimum at acidic pH except pylorus at alkaline pH, overall xylanase activity was optimum at acidic pH.

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