

International Journal of Advanced Engineering Research and Science (IJAERS) Peer-Reviewed Journal ISSN: 2349-6495(P) | 2456-1908(O) Vol-12, Issue-5; May, 2025 Journal Home Page Available: <u>https://ijaers.com/</u> Article DOI: https://dx.doi.org/10.22161/ijaers.125.1



Determination of the Rigor Mortis Index for the species *Eugerres brasilianus* (Cuvier, 1830), Captured in Itaipava, Espírito Santo - Brasil

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Received: 01 Apr 2025,

Receive in revised form: 03 May 2025,

Accepted: 08 May 2025,

Available online: 13 May 2025

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Keywords— Carapeba; freshness; rigor mortis.

Abstract— Fish has increasingly become an essential element in the search for sustainable protein sources in human nutrition. The diverse species available along the 8,000 km of our country's coastline provide a rich and varied source of food. To consolidate the fishing market, ensuring the quality of the food presented to the consumer is crucial. Among the existing ways to ensure the freshness of fish, the rigor mortis index reflects the biochemical aspects of the flesh and the stages it undergoes until it becomes unfit for human consumption. This study aimed to determine the rigor mortis index and its stages in the Carapeba species (E. brasilianus). The samples were captured on the beach of Itaipava, Espírito Santo, Brazil, and the experiment was conducted at IFES – Piúma Campus over a period of 30 hours. The measurements taken indicate that Carapeba exhibits pronounced and long-lasting rigor mortis indices, suggesting great potential in terms of quality and freshness. Specimens P2 and P3 reached the rigor mortis stage first, both at around 4.3 hours, while P1 took 5.3 hours. The full rigor mortis stage lasted 40 minutes for P1, whereas P2 and P3 remained in full rigor for 18.7 and 16.7 hours, respectively. The importance of this study for the species is notable, as even under stressful slaughter methods, the rigor mortis response time is significantly high, indicating an extremely attractive preservation potential.

I. INTRODUCTION

According to data from the FAO 2024 report, in the year 2022, fishery and aquaculture production reached 223.2 million tons, with approximately 83% of this amount related to the fishing of fish, crustaceans, and mollusks, and 17% related to the extraction and cultivation of algae. Of these 223.2 million tons, 130.9 million tons come from aquaculture, while 92.3 million come from fishing (FAO, 2024).

Fish and crustaceans are important sources of protein for the entire population, especially in coastal areas. Fishery products serve as a source of high-quality proteins essential for the maintenance and repair of human body tissues. Additionally, fish are rich in omega-3 fatty acids, which help reduce the risk of cardiovascular diseases, improve brain function, and reduce inflammation in the body. They are also important sources of vitamins, such as vitamin D, which is essential for bone health, and Bcomplex vitamins, which play a crucial role in energy metabolism. Minerals such as selenium, iodine, and zinc, present in fish, are fundamental for the proper functioning of the immune system and thyroid function control (Bito et al., 1983; Presenza et al., 2021).

Despite this nutritional significance, Brazil has low consumption rates, struggling to surpass the mark of 10 kilograms per capita (Sartori & Amancio, 2012), compared to the global average per capita fish consumption, which is around 20 kg per year (Trondsen et al., 2003). Fish consumption can be influenced by socioeconomic and social factors, cultural aspects, and personal consumption patterns, which can directly impact the amount of fish consumed by individuals (Trondsen et al., 2003). For decades, fish consumption levels in Brazil have remained below the global average, particularly in the southern and southeastern regions. The dissemination of knowledge about the nutritional benefits of fish consumption could significantly contribute to increasing this index (Rebelatto et al., 2022; Sartori & Amancio, 2012).

However, fish is a highly perishable food due to its high free water content, which accelerates bacterial action in its muscle tissue. After slaughter, a series of chemical, biochemical, and microbiological changes begin, which determine the degree of freshness of the fish. (Bito et al., 1983; Gonçalves, 2011; Minozzo, 2011).

The assessment of fish quality and freshness can be determined through sensory, physicochemical, and microbiological methods. However, due to the subjectivity of analyses, delays in results, and high costs, other methodologies have gained prominence, such as rigor mortis (Presenza et al., 2021). This method is based on muscle contraction and the loss of the elastic capacity of fish muscle. Using simple materials and an easy evaluation process, it is possible to determine the quality and freshness of the fish in question (Juliana Antunes, 2017).

In the initial phase, also categorized as pre-rigor, the fish's musculature remains flaccid for varying periods depending on thermal and intrinsic characteristics. The second phase, known as rigor mortis, is characterized by muscle stiffness and a reduction in the characteristic fish odor. The duration of this phase is determined both by the environmental characteristics of the storage location and by the slaughter method used during the capture of the fish. However, muscle conditions change over time, making the musculature flaccid again. This third phase is known as post-rigor. In the post-rigor phase, conditions favor microbial growth, which, combined with chemical reactions, become perceptible both in physical and sensory aspects (Nunes et al., 2007).

Fish has high nutritional value and is an extremely perishable product. Therefore, ensuring the

commercialization of raw materials that are safe for consumption is essential in the fishery production chain. Consequently, understanding the physicochemical and biological changes in fish before subjecting it to any processing or commercialization is of vital importance. From this perspective, the present study aimed to observe and calculate the rigor mortis index of Carapeba in order to assess the potential shelf life of this fish's derivatives.

II. METHODOLOGY

For the evaluation of the rigor index, three specimes of carapeba (*Eugerres brasilianus*) were selected. These specimens were collected throug beach seine fishing conducted in Itaipava (Fig 1), located in the municipality of Itapemirim, in the southern region of the state of Espírito Santo (Fig 2).



Fig.1: Record of Beach Seine Fishing Activity being carried out in the municipality of Itaipava, ES.



Fig.2: Geographical Location of the Collection Point.

The collected individuals were removed alive from the net and immediately placed on ice for transport to the Fish Processing Laboratory at the Federal Institute of Espírito Santo, in Piúma, a municipality bordering Itapemirim (SISBIO: 87028-2). The icing process was carried out at approximately 9:20 a.m., and the rigor analysis process began at 10:00 a.m. The individuals were identified as P1, P2, and P3 (Fig 3).



Fig.3: Eugerres brasilianus specimens arranged on the bench, with a square ruler used for measurements beside them. From left to right, the individuals P1, P2, and P3 are displayed, respectively.

The specimens were placed on an aluminum bench, and using a square ruler (Figure 3), the initial measurement of each specimen's length was taken. These measurements continued to be taken every 20 minutes.

The evaluation of the rigor index was conducted using a methodology adapted from Bito et al.(1983), positioning the square ruler beside the fish on a horizontal table, with half of its body (the tail section) suspended beyond the edge of the table. At 20-minute intervals, the rigor index was measured using equation 1.

Rigor Index (%) = $[(D0 - Dt)/D0] \times 100$ (1).

Where D_0 is the distance between the base of the caudal fin and the reference point immediately after death, and D_t is the distance between the base of the caudal fin and the reference point measured at the predetermined time intervals, which in this study were set at every 20 minutes. The experiment lasted approximately 30 hours. Ambient temperature measurement was not conducted, as the experiment took place in a climate-controlled environment with an average temperature of 23°C.

The rigor mortis index values were analyzed using the statistical software JASP (version 0.18.3.0). P1, P2, and P3 were subjected to an Analysis of Variance (ANOVA) test, and the samples were compared using Tukey's method at a 5% significance level.

III. RESULTS AND DISCUSSIONS

The rigor mortis stage was first observed in samples P2 and P3, 4.3 hours after slaughter, while sample P1 reached this stage one hour later than the others (Fig 3). Regarding

the duration of full rigor, a difference between sample P1 and the others was again observed. P2 and P3 remained in full rigor for 18.7 and 16.7 hours, respectively, while P1 remained in this stage for only 40 minutes.

Based on the above findings, when analyzing the organoleptic factors of the fish, the first visual signs of decomposition began to appear at 23 hours into the experiment. The fish became opaque, with a concave eye appearance, dry skin, and a foul odor, no longer meeting the ideal conditions for commercialization.



Fig.4: Rigor Mortis Index (%) Over Time for Eugerres brasilianus (Carapeba).

According to the analysis of variance at a 5% significance level, the mean of sample P1 compared to P2 and P3 showed a significant difference, while no difference was observed between P2 and P3. Considering that a p-value less than 0.05 indicates a significant difference between the means of the analyzed samples, otherwise, the difference between the means is considered negligible (Table 1). Tabela 1 - Análise comparativa das amostras de acordo com o teste de Tukey.

 Table. 1: Comparative Analysis of Samples According to the Tukey Test.

Comparison		Mean Difference	SE	t	Ptukey
P1	P2	-10.894	3.415	-3.190	0.005
P1	P3	-12.322	3.415	-3.609	0.001
P2	P3	-1.429	3.415	-0.418	0.908

Note: Adjusted p-value for a family of 3.

Correlating the fact that the Carapeba was captured alive and immediately placed on ice, the autolysis process was significantly reduced, as after capture, the organism undergoes a series of biochemical, physical, and microbiological changes driven by enzymatic and bacterial actions (Gonçalves, 2011; Medeiros et al., 2024).

The concentration of ATP in the post-mortem muscle and muscle contraction are decisive factors for observing the changes in fish, with rigor mortis being the stage where muscle stiffness has already occurred due to the lack of energy reserves for muscle activity. Because of this, this stage is crucial for extending the shelf life of the fish, especially when associated with the temperature and storage binomial (Gonçalves, 2011).

Several factors, such as sexual maturity, nutritional aspects, stress, method of slaughter, fish size, and others, influence the rigor mortis indices of a species. Statistically, sample P1 was the one that presented results divergent from the other samples.

It is essential to state that the method of capture has a direct influence on the rigor mortis index between the samples. As an active fishing technique, trawl fishing causes greater stress to the fish compared to other capture methods. In this type of fishing gear, the fish must exert significant effort to escape the net, leading to a rapid depletion of its energy reserves, significantly increasing the consumption of ATP and glycogen. This rapid energy demand increases the production of lactic acid, which consequently lowers the pH of the flesh (Academy, 2023; Gonçalves, 2011). Moura (2018) reports in his studies that Acaris bodós specimens subjected to stressful slaughter methods entered and exited the rigor mortis stage more quickly than those slaughtered through other methods. It is worth noting that all samples were subjected to the same capture method, meaning that stress cannot be considered the sole hypothesis for sample P1 deviating so much from the others.

However, the stress factor cannot be overlooked in this study. Mendes et al. (2015) demonstrated through their experiments that *Colossoma macropomum* specimens subjected to slaughter methods after a certain recovery period showed a delayed onset of rigor mortis compared to those subjected to stressful slaughter methods. Therefore, the importance of the capture method for the target species cannot be ignored, as the physiological stress load to which the species is subjected is extremely high when struggling or exerting effort to escape capture. Although each individual may have different responses to stressful stimuli, intrinsic factors such as nutritional, reproductive aspects, and the habitat in which the organism is situated contribute to changes in the post-mortem rigor index.

IV. CONCLUSION

The species *Eugerres brasilianus* (Couvier, 1830) remained in full rigor for an average of 12 hours, indicating a greater resistance of the fish to deterioration. Thus, it is a species with significant conservation potential, which opens up a range of possible studies for its conservation and processing methods.

More studies are needed regarding rigor mortis in this species, as its behavior when subjected to less stressful slaughter methods is still unknown. Therefore, it is recommended that future studies conduct analyses with greater detail on the post-mortem biochemical changes in the fish, aiming to understand the behavior of the rigor index in this species when subjected to other slaughter conditions. This study highlights the potential factors affecting the quality and shelf life of Carapeba meat.

ACKNOWLEDGEMENTS

We extend our deepest and most respectful gratitude to the Federal Institute of Espírito Santo – Piúma Campus, for its invaluable institutional support, academic excellence, and intellectually stimulating environment that greatly contributed to the successful development of this research. We are sincerely grateful for the dedication of the faculty, the support provided by the technical staff, and the infrastructure made available, all of which were essential to the accomplishment of this work. Our heartfelt appreciation goes to everyone who, directly or indirectly, contributed to this achievement.

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