

Conference Paper

Preservation Effect of Grouper (*Epinephelus* sp) Fillet Against Survival of *Anisakidae*

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Abstract

This study was aimed to determine the effect of time and preservation method that the most effective in killing larvae *Anisakidae*, and still be able to maintain the quality post-preservation of fillet from three groups, combination of pickling with heating (50°C and 75°C); pickling with cooling (ice and dry ice); and pickling with salting (dry salting and brine salting). This study was a laboratory experimental research carried out in four phases, phase identification, preservation, organoleptic test and protein test. The results can be concluded that the time of preservation and preservation methods affect the survival of *anisakidae* larvae. The most effective heating method preservation to kill the larvae *Anisakidae* was heating at temperature of 75°C compared to 50°C, for cooling the use of dry ice is more effective than ice, and for methods of salting with 25% concentration, brine salting and dry salting effective in killing larvae *Anisakidae*, The effect of the method preservation for the quality of fillets in organoleptic test still considered adequate and still can be accepted by the public. As for the protein test a slight decline of the fillet quality post-preservation.

Keywords: Preservation; Fillet of Grouper; Larva *Anisakidae*.

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1. Introduction

One of the serious diseases that zoonotic in humans from eating raw fish is Anisakiasis caused by *Anisakis* sp (Adams et al., 1997). In the deployment Anisakiasis time processing and preservation methods of fish has also become one of the main factors that become important issues. There are many prefer method of preserving food that can kill the larvae of *Anisakidae*, but not necessarily the method is effective in killing

the larvae, and not all methods take attention to the quality of the products that have gone through the process of preservation, so that needs to be done a test to analyze the effect of time on each methods of preservation, that could find the most effective methods to kill larvae Anisakidae and still be able to maintain the quality of the product.

Anisakiasis is an infection due to larval genus *Anisakis* sp and *Pseudoterranova* sp in humans that can cause allergic reactions include urticaria, anaphylaxis, dermatitis, gastroenteritis, until the symptoms of asthma (Bircher et al. 2000), when the infection Anisakiasis appear 12-24 hours after eating fish raw or undercooked, the reactions that occur are epigastric pain accompanied by nausea and vomiting.

Meanwhile, when the infection occurred approximately 7 days after the larvae enter, then the reaction that arises is severe pain in the lower abdomen, nausea, vomiting, fever, diarrhea and sometimes there is blood in the stool, the infection of larvae occurs in the intestine (Ortega and Cocera, 2000). In 1968, Anisakiasis first reported in humans in 160 cases found in the Netherlands, then in 1976, in Japan with 487 cases. Anisakiasis most cases are caused by *Anisakis simplex* and the rest is *Pseudoterranova* sp. (Acha and Szyfres., 2003). Information on the incidence of the Anisakiasis disease is still very limited in Indonesia. One and the only study ever conducted in Indonesia on Anisakiasis is seroepidemiologi study on 244 patients who visited the Regional General Hospital in Sidoarjo, the study showed 11% of patients seropositive against antigen *Anisakis* sp. (Uga et al., 1996). Until now Anisakiasis not received special attention from the Indonesian government, because of that the good production management becomes very important to avoid this problem in the future.

Modernization is happening in Indonesia make people began to recognize the type of fresh foods (raw fish) without going through the cooking process such as sushi or sashimi, this course will be able to increase the probability of human transmission of the disease Anisakiasis and will degrade the quality of a fishery product. Groupers are one species of marine fish that are often found in the Indonesian market. As a carnivorous fish, groupers have a big role in the deployment process Anisakidae. According Arifudin and Nurul (2013) the prevalence of infected grouper fish larvae Anisakidae obtained in TPI Brondong, Lamongan, East Java reached 100% or almost be said that the grouper in TPI Brondong, Lamongan always infested by the larvae Anisakidae. This is in line with Maghgrabi et al (2010) which stated that 100% of grouper obtained from the Red Sea is also infected by *Anisakis* sp. Because of the high prevalence and danger of zoonosis of Anisakidae grouper, it would require a good handling to implement food safety and food quality to the sea-based food production, especially the production of fillet from fish grouper.

One solution could be to obtain assurance on food safety and food quality from grouper fish fillet is using effective preservation techniques. Some of the usual method for use in preservation is pickling, heating, cooling, and salting. Pickling is known to maintain the quality of the fish meat, but had no effect on Anisakidae (FDA, 2015)^b. Because of it, pickling can be used as a combined preserve for the other preservation method. The next method is heating, according to Adams et al (1999) the temperature of 50°C - 75°C in a few minutes is enough to kill parasites Anisakidae. So we need a study to compare the level of the effective time of preservation at temperatures of 50°C and 75°C. Further cooling methods. The use of dry ice are still very rare in Indonesia, so research to see the level of effective preservation time between the used of ice or dry ice in killed Anisakidae was done. The latter method is salting. Dry salting indeed could kill the parasite Anisakidae (Baladin, 2006), but brine salting could be an effective alternative as a way of salting and control of Anisakidae.

Because of the importance of the influence of time and the need for methods of preservation effective from pickling, heating, cooling and salting to prevent Anisakiasis, and also maintains the quality organoleptic value and the protein content of the products fillet of grouper post-preservation, the necessary research to determine the effect of time preservation fillet of grouper with a combination of pickling with heating, cooling and salting against survival of Anisakidae larvae in order to find the most effective methods to kill larvae Anisakidae, which is still able to maintain the quality of the value of the organoleptic and protein content of the fillet of grouper as a basis for healthy food and the control of Anisakiasis zoonotic diseases.

2. Materials and Methods

This study was a laboratory experimental study and carried out in four stages, the first stage was examination Anisakidae larvae, were identified used dissecting microscope and some samples were colored with carmine. The second stage Anisakidae larvae survival test by insert the larvae into fillet of grouper, immersed with 5% acetic acid, and preserved used three methods which had been prepared (heating, cooling and salting). The third stage was organoleptic test of the preservation resulted from grouper fillet, which was tested used the scoring test and hedonic test. The fourth stage was protein test used Kjeldahl method.

3. Results and Discussion

Analysis of the results of the study were divided into four groups, namely the identification of larvae Anisakidae, preservation to determine the survival of Anisakidae larvae, organoleptic test results from preservation grouper fillet and protein test from grouper fillet after preservation.

3.1. Larvae Identification

Identification of larvae was conducted in the Department of Parasitology, Faculty of Veterinary Medicine, University of Airlangga. Identification is done by two methods, namely the method of staining methods using native and Carmine. From the results of the nematode larvae originating from grouper (*Epinephelus* sp), obtained mukron on the posterior and booring tooth on anterior. The two structure, is a specific characteristic of the family Anisakidae.

3.2. Preservation

The results of the analysis test influence between subject at the time of preservation, preservation methods, and the interaction between these two factors on mortality of *Anisakidae* larvae shows that there is a real effect ($p < 0.01$). The result of the influence of time preservation of the survival of the larvae Anisakidae divided into three group. Results of preservation of each subgroup was analyzed using F test (ANOVA) followed by Duncan multiple range test. With a temperature of 75°C for 1 minute was able to kill the larvae Anisakidae, with a best time of 7 minutes to kill all the larvae up to 100%. While the method of heating with a temperature of 50°C only can kill larvae Anisakidae 4% within 5 minutes, and can not kill all larvae at 7 minutes. Preservation methods using dry ice for 12 hours can kill all larvae up to 100%, while the preservation methods using ice only can kill larvae in 36 hours with a percentage of 8% larval mortality, and not be able to kill all larvae within 48 hours. Preservation method using dry salting technique and brine salting technique for 24 hours can kill all larvae up to 100%. Brine salting method works more fast than dry salting method, this can be seen as the 3 hour of preservation brine salting can kill 36% of larvae Anisakidae. As for dry salting larvae mortality rates can be seen in the 6 hour reservation with larval mortality rate of 40%.

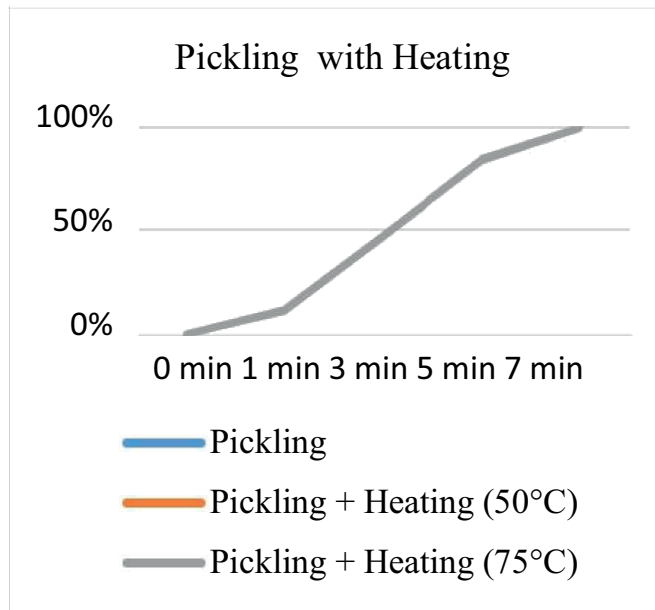


Figure 1: Result of preservation pickling with heating.

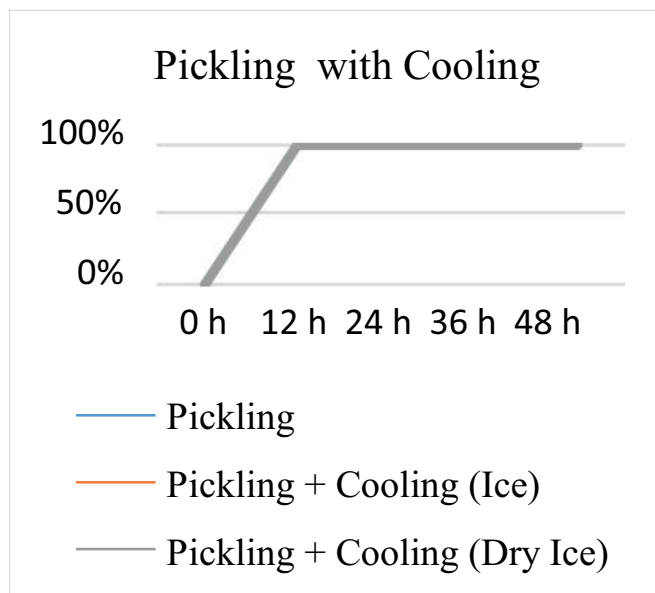


Figure 2: Result of preservation pickling with cooling.

3.3. Organoleptic test

The data obtained from the assessment sheet tabulated and determined the value of quality by finding the mean of each panelist results at the 95% confidence level. To calculate the average interval quality score of each panelist used the formula that has been set by the Indonesian National Standard (2006). The results of the test scoring showed that the method of preservation fillet with heating 50°C, Ice, Dry Ice, Dry Salting and Brine Salting have the same score in the organoleptic test that is 6, while fillet

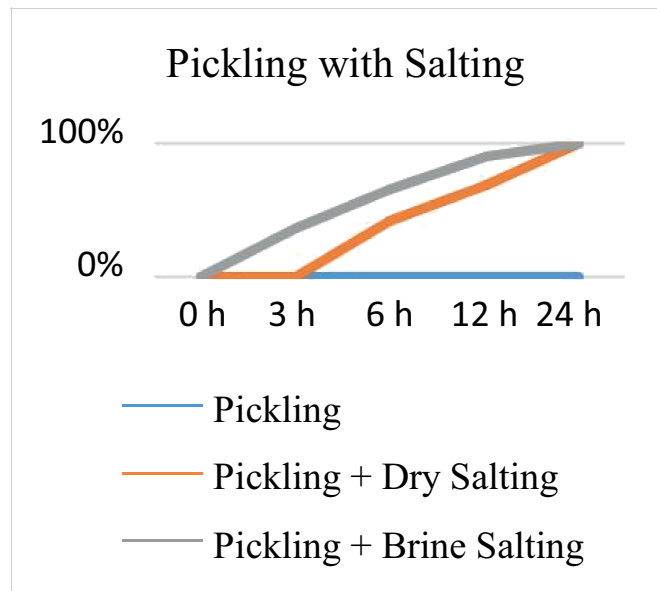


Figure 3: Result of preservation pickling with salting.

heating with temperature 75°C have different values from methods of preservation of others, namely 6, 5. This score indicates that the fillet still feasible for public consumption. As for the hedonic test results show that people are still able to receive the results of grouper fillet preservation.

3.4. Protein Test

The results of the best preservation of every method that can kill larvae anisakidae taken on sample for later examined the proteins that are found in fillets, **Fig 4** is the result of using the protein test with kjeldahl method. From the results can be seen for the control of grouper fillet without going through the process of preserving the protein content 19.51%, fillet of grouper through pickling process has slightly decrease of value of 18.03%, little difference with the protein content of the fillet in preservation using dry ice 18.12%. For protein content value from grouper fillet in preservation using ice has a value of 17.60%, higher than the protein content grouper fillet in preservation using heating methods, ie 17.26% for heating 50 °C and 17, 11% for heating 75 °C. The result of the lowest protein content is the protein content from grouper fillet in a preservation method of salting, which is 16.38% at salting using the technique of dry salting and 16.56% at salting using brine salting techniques.

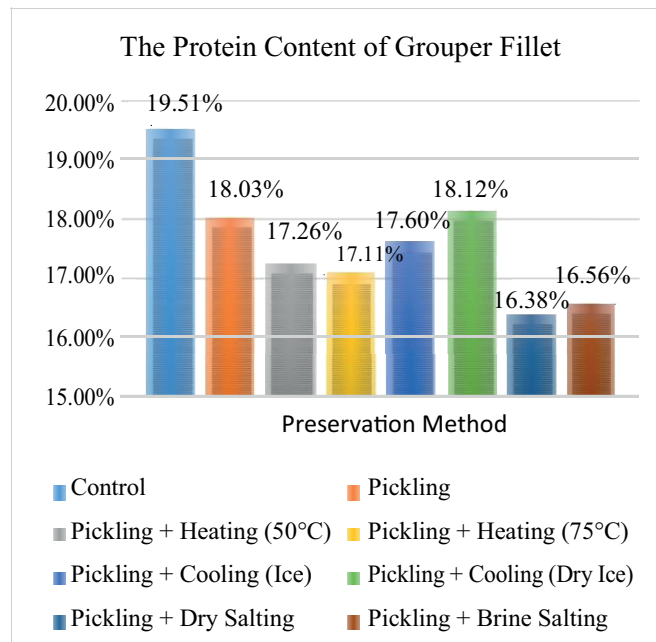


Figure 4: Graphic protein test results using kjeldahl.

4. Conclusion

Time and method preservation of grouper fillet by combination of pickling with heating, cooling and salting have significant effect for the survival of Anisakidae larvae. Method of heating with a temperature of 75°C most effective in killing larvae Anisakidae when compared to the method using a heating temperature of 50°C that ineffective. Cooling methods using dry ice most effective in killing larvae Anisakidae. when compared with the use of cooling method using ice that ineffective. Dry salting methods and brine salting effective in killing larvae Anisakidae. Effect of the preservation method for the quality of organolepti and protein content of grouper fillet for organoleptic test is still considered adequate and still can be accepted by the public. As for the protein test a slight decrease of the fillet quality post preservation.

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