

Original Research Article
**VOLATILE FLAVOR COMPONENTS AND
AMINO ACID PROFILE OF
STEAMED INDIAN MACKEREL (*Rastrelliger* sp.)**

ABSTRACT

Heating method such as steaming could influence fisheries commodity chemical composition, notably its flavor components. This study aims to identify the volatile flavor compounds and amino acid composition of steamed Indian mackerel. The samples in this study were steamed (100°C, 30 minutes) Indian mackerel meat which their volatile compounds were extracted using Solid Phase Micro Extraction (SPME) method and identified by Gas Chromatography/Mass Spectrometry (GC/MS), while their amino acids profile were analyzed by High Performance Liquid Chromatography. The volatile components analysis result showed that there were 50 volatile compounds in steamed Indian mackerel. Most volatile compounds detected derived from hydrocarbons, aldehydes, alcohols and ketones groups of compounds. The amino acids profile analysis showed that glutamic acid is the highest quantity amino acids detected in steamed Indian mackerel (3.74% 3,74%). These results are basically influenced by many factors including heat treatment and various heat-induced reactions.

Keywords: amino acids; flavor; Indian mackerel; steaming; volatile components

1. INTRODUCTION

Indian mackerel (*Rastrelliger* sp.) is a pelagic fish that is economically important and well-known in Indonesia. In 2019, Indonesia's average catch production volume of Indian mackerel was 23,678,573.15 23.678.573,15 tons, indicating a 2.3% increase from 2018. West Java province contributed 1,514,995.71 1.514.995,71 tons of Indian mackerel national production volumes [1]. Indian mackerel, or "ikan kembung" locally, is a species of marine fish that is highly available and significant in Indonesia due to its tasteful flavor and affordable price compared to other pelagic and Omega-3 source fish, making it accessible to many groups of people.

In Indonesia, Indian mackerel is usually consumed directly as a dish by frying, baking, or steaming with light seasoning, or it can be used as raw material for various traditional food products such as "peda" and various other Indonesian culinary dishes. Almost all known fish processing methods could give specific characteristics due to the flavor, nutritional, and texture alteration of each processing result [2].

Among the processing methods, steaming is commonly used in Indonesia as a traditional cooking method. This method can relatively maintain the natural flavor of the food as convection heat transfers from the hot steam to the food that is being steamed. Steaming (water evaporation or steam as a heat source) has the advantage of lower loss risks of vitamins and other components sensitive to heat [3]. Steaming and other processing

methods cause complex chemical reactions to occur between volatile precursors from fat and lean tissue [4].

The overall perception (sensation) received by human senses, especially taste and aroma, when food and drink are consumed, is referred to as flavor [5]. Flavor compounds contained in fish usually come from derivatives of protein compounds and mostly fat. The main flavor of processed meat in the form of volatile and non-volatile components has a significant effect on the acceptance of processed meat, especially on the aroma [6]. The composition of volatile flavor compounds detected in fishery products usually comes from aldehydes, alcohols, ketones, and hydrocarbons [7].

Besides volatile compounds, flavor components also include nonvolatile compounds that impact the taste attributes of a product. These nonvolatile compounds are typically obtained from free amino acids, peptides, and nucleotides like IMP (disodium 5'-inosine monophosphate), GMP (disodium 5'-guanosine monophosphate), and AMP (disodium 5'-adenosine monophosphate) [7],[8]. Amino acid profile analysis in this study aims to provide general information concerning the overall taste characteristics of the samples that might be influenced by amino acids. Additionally, it provides information regarding the essential and non-essential amino acid composition of the samples. Amino acids and peptides contribute directly to seafood flavor [9].

Fishery products are a great source of nutrition due to their high protein and unsaturated fatty acid content. Knowledge of the chemical composition of a commodity, namely volatile components, and amino acids, is important for identifying nutritive values and chemical changes. A series of heat-induced changes that occur in the processing phase can influence the flavor composition of processed fish. Therefore, the purpose of this study is to identify volatile flavor components and amino acid profiles of steamed Indian mackerel meat samples.

2. MATERIAL AND METHODS

2.1 Sample Preparation

Indian mackerel fish samples were taken from fish landing site at Karangsong, Indramayu, West Java. Samples were then brought in a cool box with ice to fish ratio of 2:1 in the morning time so that the freshness of the fish can be maintained. The samples were then transported to the Laboratory of Fishery Products Processing, Padjadjaran University to be handled and prepared. The fish were washed clean, eviscerated, weighed adequately and steamed using a temperature of 100°C for 30 minutes [7]. The three-layered packaging was done to minimize the changes and damage to the flavor of samples to be analyzed were caused by air, light and temperature [10]. Subsequently, the samples were transported to several analytical laboratories in Sukamandi and Bogor, West Java.

2.2 Analysis Method

Volatile compounds analysis procedures used in this study was a modification of analysis procedure carried out by Guillen & Errecalde [11]. The analysis was performed using a series of Gas Chromatography (GC Agilent Technologies 7890A System) and Mass spectrometry (Agilent Technologies 5975C inert XL EI CI / MSD) apparatus. Samples extraction was conducted by Solid Phase micro extraction (SPME) using fiber DVB/Carboxen/Poly Dimethyl Siloxane with a heating temperature of 40°C for fresh samples and 80°C for steamed samples for 45 minutes (in water bath). GC column used was HP-INNOWax (30m x 250µm x 0,25µm), helium carrier gas, initial temperature was 45°C (hold 2 minutes), temperature escalation as much as 6°C/minute, a final device temperature 250°C (hold 5 minutes) with overall time 45 minutes. Mass spectra of compounds detected were then compared with the mass spectra patterns available within the computer database or NIST (National Institute of Standards and Technology) library version 0.5a. The data of volatile flavor component were analyzed further using Automatic Mass Spectral Deconvolution and Identification System (AMDIS) software [12].

Amino acids profile analysis was carried out based on modification from [13] and [14]. The analysis was performed using High Performance Liquid Chromatography (HPLC) (Shimadzu CBM-20A, Shimadzu Corporation, Japan) apparatus and the parameters setting in general were: Ultra Techsphere column, 1mL/minute mobile phase flow rate with fluorescence detector. The resulting data from volatile compounds analysis were discussed descriptively based on identification and semi quantification intensity of the compounds detected from analyzed samples [10]. The amino acids profile result was identified using 15 standards of amino acids and quantified in μmol concentration unit based on their peak areas and amino acids standards peak areas.

3. RESULTS AND DISCUSSION

3.1 Volatile Components

The results from volatile flavor component analysis showed that there are 50 volatile compounds successfully detected in steamed samples (Table 1) with wide variation of compounds. The identified volatile flavor compounds were categorized into several categories such as hydrocarbons, aldehydes, alcohols, and ketones.

Table 1. Volatile Compounds Identified in Indian Mackerel Steamed Meat

Group	Compounds	Area	Proportion (%)
Hydrocarbons	Pentadecane	38414618	46.678
	Pentadecane, 2,6,10,14-tetramethyl-	14826520	18.016
	Heptadecane	1833407	2.228
	Tetradecane	1406377	1.709
	Tridecane	375072	0.456
	Hexadecane	277542	0.337
	Oxirane, tetradecyl-	254166	0.309
	Bicyclo[4.2.0] oct-7-ene	139967	0.170
	1-nonadecene	120621	0.147
	Benzocycloheptatriene	119144	0.145
	Undecane	109455	0.133
	1,3,6-Heptatriene, 5-methyl-	85177	0.104
	Limonene	72756	0.088
	1H-Indene, 1-methylene-	56131	0.068
	Undecane	37847	0.046
	Oxirane, hexadecyl-	35515	0.043
Toluene	9055	0.011	
Aldehydes	Octanal	2255192	2.740
	Hexadecanal	2153464	2.617
	Nonanal	2101000	2.553
	Decanal	747768	0.909
	Heptanal	705118	0.857
	Dodecanal	699139	0.850
	Hexanal	611328	0.743
	2-Undecenal	353748	0.430
	Benzaldehyde, 4-ethyl-	327170	0.398
	2,4-Heptadienal, (E, E) -	285125	0.347
	2-nonenal, (E) -	250132	0.304
	2-Octenal, (E) -	168360	0.205
	2-Heptenal, (E) -	144215	0.175

Group	Compounds	Area	Proportion (%)
	Dodecanal	67196	0.082
	2,4-Hexadienal, (E, E) -	32881	0.040
	2,6-Nonadienal, (E, Z) -	19200	0.023
	Pentanal	1465	0.002
Ketones	2-decanone	3471248	4.218
	2-Heptanone	3418798	4.154
	2-Nonanone, 3- (hydroxymethyl) -	1055851	1.283
	3,5-Octadien-2-one	538187	0.654
	5-Hepten-2-one, 6-methyl-	75498	0.092
	2,3-Octanedione	13525	0.016
Alcohols	1-Octanol	2661693	3.234
	1-Nonanol	124695	0.152
	1-Heptanol	59013	0.072
	2-Ethyl-1-Dodecanol	21935	0.027
	1-Pentanol	15509	0.019
Other	Sulfurous acid, octadecyl 2-propyl ester	1448119	1.760
	Furan, 2-pentyl-	273126	0.332
	Octyl thioglycolate	8559	0.010
	Oxalic acid, nonyl isobutyl ester	8326	0.010
	Acetaldehyde	6686	0.008

The hydrocarbon group of compounds detected in steamed Indian mackerel contained 17 types of compounds, with pentadecane (46.6782%) and pentadecane, 2,6,10,14-tetramethyl (18.0159%) being the most abundant. Similarly, 17 types of aldehydes were detected in the fish, with octanal (2.7403%) being the most abundant. The ketone group contained 6 types of compounds, with 2-decanone (4.2180%) having the largest proportion. The alcohol group had 5 types of compounds, with 1-Octanol (3.2343%) being the most abundant. Other compounds were also detected but in very low amounts, and they belong to other groups of compounds and organic acids that are rarely identified in fish meat and require further identification.

Pentadecane, 2,6,10,14-tetramethyl, also known as pristine, is formed from an auto-oxidation process that is typical for fish [15] and is also detected in various local smoked fish such as *fufu* (smoked skipjack), *salai* (smoked catfish), *kayu* (cold smoked skipjack), cooked krill, and crayfish [10]. Octanal, the most abundant compound in the aldehyde group, is considered a fatty aldehyde lipid molecule that occurs naturally in citrus oils. Hexadecanal, also known as palmitaldehyde, is another compound from the aldehyde group that is a fatty aldehyde. Ketones such as 2-decanone can be produced or generated by thermal oxidation or degradation of fatty acids, degradation of amino acids, or microbial oxidation. Alcohol compounds, such as 1-octanol, occur naturally in the form of esters in some essential oils and are generally present in low concentrations in food [11], [15], [16], [17], [18].

The other groups of compounds detected in both samples derive from many classes of compounds. For example, 2-pentylfuran is a heterocyclic compound that has been detected in steamed Patin catfish [19], smoked black bream [11], and steamed silver carp [7]. This compound is known to originate from the dehydration of glucose (cellulose thermal degradation), but could also derive from the Maillard reaction [20], [21]. The sulfurous acid, octadecyl 2-propyl ester, detected in the Indian mackerel samples is an ester compound that most likely derives from the esterification of acids and alcohols formed from lipid metabolism

or lipids thermal degradation products [11], [20].

3.2 Amino Acid Profile

In this study, amino acid profile analysis was carried out to provide information concerning essential and non-essential amino acids. Other than that, this analysis also provides general picture regarding their contribution to overall sample's taste characteristic. From fish flavor perspective, the importance of amino acids has been well recognized [22]. According to [23], the presence of amino acids in food played an important role on most seafood taste. This study used 15 amino acids standards to quantify individual amino acids present in samples. These standards were aspartic acid, glutamic acid, serine, histidine, glycine, threonine, arginine, alanine, tyrosine, methionine, valine, phenylalanine, leucine, lysine and isoleucine. As we can see from Table 2, glutamic acid is the highest amino acids detected in steamed Indian mackerel samples (3.74% 3,74%), while other amino acids values show variation in content. Some amino acids, aside from glutamic acids, are detected higher than other amino acids in steamed samples, such as aspartic acid (2.41% 2,41%), lysine (2.38% 2,38%), leucine (2.08% 2,08%) and arginine (1.98%). Fish meat that has less savoury taste could be caused by less glutamic acid contain in it [24]. As point out before, each individual amino acid could contribute to product's basic taste. Bitter taste in peptide could be contributed by proline and the presence of phenylalanine, alanine, glycine, leucine, tyrosine, and valine while glutamate affects umami taste [23], [25], [26], [27].

Table 2. Amino acids profile of fresh and steamed Indian mackerel (%)

Amino acids	Value (%)
Aspartic acid	2.41
Glutamic acid	3.74
Serine	0.72
Histidine*	0.79
Glycine	0.80
Threonine*	1.11
Arginine	1.98
Alanine	1.35
Tyrosine	1.17
Methionine*	1.13
Valine*	1.57
Phenylalanine*	1.10
Isoleucine*	1.52
Leucine*	2.08
Lysine*	2.38

Essential amino acids which steamed Indian mackerel has from highest quantity to the lowest were lysine, leucine, valine, isoleucine, methionine, threonine, phenylalanine and histidine. Lysine has the highest quantity in both samples and has an important role in human body as basic composition needed for blood antibody, strengthen the circulation and maintaining normal cells growth and lysine will decrease excessive blood triglycerides if together with proline and vitamin C [24]. The content of amino acids is affected by heat treatment time length and heating method [19], [28]. Free amino acids forming are increase due to proteolytic reaction which take place throughout heating process [7], [29]. Taste quality of some amino acids, which are the building block of protein, would depend on their side chain structure. Glutamate and aspartate which are classified as acidic L-amino and

both contributed to umami taste and sweet taste is dominantly contributed by mainly D-amino acids [23], [25]., while bitter taste is contributed by aromatic, basic and branched amino acids [27].

4. CONCLUSION

Most of the volatile flavor components detected in steamed Indian mackerel samples derived from the hydrocarbons, aldehydes, ketones and alcohols groups. As many as 50 compounds were detected and identified in the sample. Compounds that have the highest proportion in is pentadecane (46.6782%) which belongs to hydrocarbons group. The highest amount of amino acids detected in steamed samples is glutamic acid which commonly contributes to umami taste characteristics.

REFERENCES

1. Ministry of Marine and Fisheries Affairs. Produksi Perikanan. Statistik KKP. 2023. Accessed 1 May 2023. Available: <https://statistik.kkp.go.id/home.php?m=total&i=2#panel-footer>
2. Pratama RI, Rostini, I and Awaluddin, MY. Flavor compound composition of fresh and steamed common carp (*Cyprinus carpio*). Jurnal Akuatika. 2013;4(1), 55-57.
3. Fellows P. Food Processing Technology, Principles and Practice. Cambridge: Woodhead Publ. Ltd. 2000
4. Purba M. Pembentukan Flavor Daging Unggas Oleh Proses Pemanasan dan Oksidasi Lipid. Wartazoa. 2014; 24(3):109-118.
5. Rothe M. Introduction to aroma research. Dordrecht (Netherlands): Kluwer Academic Publisher. 1989.
6. Joo ST, Kim DD. Meat quality traits and control technologies. In: Joo ST, editor. Control of meat quality. Kerala (India): Research Signpost. 2011.
7. Liu JK, Zhao SM, Xiong SB. Influence of re-cooking on volatile and non-volatile compounds found in silver carp *Hypophthalmichthys molitrix*. Fish Sci. 2009;75: 1067-1075.
8. Chen DW, Zhang M. Non-volatile taste active compounds in the meat of Chinese mitten crab (*Eriocheir sinensis*) Food Chemistry. 2006; 104, 1200-05
9. Deng Y, Luo Y, Wang Y, Zhao Y. Effect of different drying methods on the myosin structure, amino acid composition, protein digestibility and volatile profile of squid fillets. Food Chemistry 2014;171: 168-176
10. Pratama RI. Flavor Characteristics of Several Indonesian Smoked Fish. Thesis. School of Postgraduate Bogor Agricultural Institute. Bogor. 2011
11. Guillen M, Errecalde M. Volatile components of raw and smoked black bream (*Brama raii*) and rainbow trout (*Onchorhynchus mykiss*) studied by means of solid phase micro extraction and gas chromatography/mass spectrometry J Sci Food Agric. 2002; 82: 945-952
12. Mallard GW, Reed J. Automatic Mass Spectral Deconvolution and Identification System (AMDIS) User Guide. Gaithersburg: U.S. Department of Commerce. 1997
13. Ishida Y, Fujiwara M, Kinoshita T, Nimura N. Method of amino acid analysis. United States Patent 4670403. 1987. Accessed: March 8, 2011. Available online: <http://www.freepatentsonline.com//4670403>
14. Toppe J, Albrektsen S, Hoppe B, Aksnes A. Chemical composition, mineral content and amino acid and lipid profiles in bones from various fish species Comparative Biochemistry and Physiology B. 2007;146: 395-401

15. Guillen MD, Errecalde MC, Salmeron J, Casas C. Headspace volatile components of smoked swordfish (*Xiphias gladius*) and cod (*Gadus morhua*) detected by means of solid phase micro extraction and gas chromatography–mass spectrometry Food Chem. 2006; 94: 151-156.
16. Alasalvar C, Taylor KDA, Shahidi F. Comparison of volatiles of cultured and wild sea bream (*Sparus aurata*) during storage in ice by dynamic headspace analysis/gas chromatography-mass spectrometry J. Agric. Food Chem. 2005; 53: 2616-2622
17. Cha YJ, Baek HH, Hsieh CY. Volatile components in flavour concentrates from crayfish processing waste J Sci Food Agric. 1992; 58: 239-248
18. Thelestam M, Curvall M and Enzell CR. Effect of Tobacco Smoke Compound on the Plasma Membrane of Cultured Human Lung Fibroblasts. Toxicology. 1980; 15(3): 203-217
19. Pratama RI, Rostini I, Rochima E. Amino Acid Profile and Volatile Flavour Compounds of Raw and Steamed Patin Catfish (*Pangasius hypophthalmus*) and Narrow-barred Spanish Mackerel (*Scomberomorus commerson*). 3rd International Conference on Tropical and Coastal Region Eco Development. IOP Conf. Series: Earth and Environmental Science 2017; 116:1-17
20. Chung HY, Yung IKS, Ma WCJ, Kim J. Analysis of volatile components in frozen and dried scallops (*Patinopecten yesseonensis*) by gas chromatography/mass spectrometry. Food Research International 2002; 35: 43-53
21. Maga JA. The flavor chemistry of wood smoke. Food Review International 1987; 3: 139-183
22. Antoine FR, Wei CI, Littell RC, Quinn BP, Hogle AD, Marshall MR. Free amino acids in dark and white muscle fish as determined by O-phthaldialdehyde Precolumn derivatization. Journal of Food Science. 2001; 66: 72-77
23. Kawai M, Uneyama H, Miyano H. Taste-active components in foods, with concentration on umami compounds. Journal of Health Science. 2009; 55: 667-673
24. Suryaningrum DT, Muljanah I, Tahapari E. Sensory profile and nutrition value of several types of patin catfish and nasutus hybrid Jurnal Pascapanen dan Bioteknologi Kelautan dan Perikanan 2010; 5: 153-164
25. Wongso S, and Yamanaka H (1998) Extractive components of the adductor muscle of Japanese baking scallop and changes during refrigerated storage. Journal of Food Science 63, 1-5
26. Zhao CJ, Scheber A, Ganzle MG. Formation of taste-active amino acids, amino acid derivatives and peptides in food fermentations. Food Research International. 2016; 89, 39-47
27. Kato H, Rhue MR, Nishimura T. Role of free amino acids and peptides in food taste. in Flavor Chemistry; Trends and Developments, R. Teranishi (Ed.), p. 158-174. American Chemistry Society, Washington DC. 1989
28. Oluwaniyi OO, Dosumu OO, Awolola GV. Effect of local processing methods (boiling, frying and roasting) on the amino acid composition of four marine fishes commonly consumed in Nigeria. Food Chemistry (2010); 123: 1000-1006
29. Toth L, Potthast K. Chemical aspects of the smoking of meat and meat products (Advances in Food Research) ed CO Chichester Academic Press Inc. New York. 1984