

# Composition of Flavour Non Volatile Compound Steamed Gourami (*Osphronemus gouramy*)

## ABSTRACT

This research aims to identify the composition of non-volatile flavor compounds contained in the steamed gourami (*Osphronemus gouramy*). The research was carried out experimentally with the treatment of steamed gourami samples (at 100°C for ± 30 minutes). This research was conducted at the Fisheries Product Processing Laboratory, Faculty of Fisheries and Marine Sciences, Padjadjaran University, Jatinangor; IPB Integrated Laboratory. A simple product description test was carried out on samples which included the appearance of the meat, aroma, texture, and taste. Identification of non-volatile flavor compounds using tools High Performance Liquid Chromatography (HPLC). The results showed that steamed gourami has 15 identified non-volatile flavor compounds of 15 amino acid compounds. Two umami-flavored amino acids are 4.09% glutamic acid and 2.67% aspartic acid. The four sweet tasting amino acids are 1.53% alanine, 1.19% threonine, 1.13% glycine and 1.01% serine. The nine bitter amino acids are lysine 2.54%, leucine 2.11%, arginine 1.64%, valine 1.30%, isoleucine 1.29%, phenylalanine 1.14%, tyrosine 0.91%, methionine 0, 79% and histidine 0.66%. Steamed gourami has an intact appearance and is pale white in color. The aroma of steamed gourami is fresh and tasty according to the specific gourami. The texture of the meat is dense, soft and very compact. The taste of steamed gourami is savory according to the specific type.

*Keywords: non volatile flavor compounds, steaming, gourami*

## 1. INTRODUCTION

Indonesia is a maritime country that has vast territorial waters, very large land, and sea fisheries resources. Inland aquaculture is one of Indonesia's fishery production potentials which has increased every year. Types of fish farming developed in Indonesia include gourami, carp, catfish, tilapia, and milkfish [30]. One of the very potential commodities is gourami (*Osphronemus gouramy*). Gourami is one of the main commodities of freshwater aquaculture with a production volume of 149,170 tonnes in 2021, as well as cultivated catfish, carp, tilapia [59].

Gourami has a high economic value, when compared to other freshwater fish commodities [4];[40]. According to [79], many gourami are traded fresh, both alive and dead. The market demand for gourami is quite high, because gourami has a good taste of meat, relatively easy maintenance, and a relatively stable price. In addition, gourami is a commodity with high nutritional content which is beneficial for humans for growth and formation of energy. Traditionally, gourami is usually processed by frying, steaming, or baking, with added spices.

Traditional processing that is commonly carried out can affect product flavor characteristics, such as high temperature thermal processing in the manufacture of pepes which is a typical food menu of West Java. This thermal process is one of the most important methods in food processing because it results in the ripening and formation of specific flavors required for food quality, product shelf life, improved nutrient availability, and relatively easy to control processing conditions [17];[20]. The thermal process that is often used is the steaming method (wet heating). According to [65], steaming is a high-temperature processing method using water vapor as a heat-conducting medium. The use of water vapor as a heat source can reduce the risk of possible nutritional loss or loss of vitamins and food compounds that are sensitive to high temperatures [20]. The heating process can affect and change the basic characteristics of fresh fish used as raw materials. Raw materials that are heated will experience changes in physical, chemical, and product characteristics, especially flavor and texture.

38 Flavor is all impressions or sensations that can be felt by the human senses, especially through taste and smell when  
39 consuming a food product [53]. According to [8], flavor is formed from a combination of experiences and sensations  
40 received on the characteristics of the material. Flavor is an important factor in fresh and processed food, especially in  
41 products made from fish. This happens because flavor can affect the level of consumer acceptance, preference, and  
42 consumption of a product. According to [49], the chemical composition and flavor of various types of fishery commodities  
43 have differences both in fresh and processed forms. The flavor characteristics of a commodity are influenced by  
44 compounds which are divided into two groups, including volatile and non-volatile flavor compounds.

45  
46 Volatile components contained in flavors are components that give the sensation of aroma or smell, give an initial  
47 impression, and can evaporate quickly. Non-volatile components produce sensations of taste, such as sweet, sour, salty  
48 and bitter. This component does not provide an aroma sensation but becomes a medium for volatile components, and  
49 holds back the evaporation of volatile components. The taste characteristics of a product are influenced by non-volatile  
50 flavor compounds which usually come from peptide group compounds, amino acids, and also nucleotides [9];[50];[51].  
51 Non-volatile extractive components containing nitrogen such as free amino acids play an important role in flavoring fishery  
52 products [77]. Based on opinion [13], amino acids and peptides play a direct role in the flavor of processed fishery  
53 products. The types of amino acids that influence the flavor formation process can be identified from the amino acids  
54 contained in fishery products [50].

55  
56 Research on the non-volatile flavor of fishery commodities has been widely carried out abroad, but research on the non-  
57 volatile flavor of fishery commodities has not been widely carried out in Indonesia. An example of research on the non-  
58 volatile flavor composition of fishery commodities abroad was conducted by [6], namely regarding the effect of cultivating  
59 seawater with different salinities on non-volatile active flavor compounds in pacific oysters (*Crassostrea gigas*), as well as  
60 research [36] regarding the influence of high hydrostatic pressure and storage temperature on fatty acids and non-volatile  
61 flavor active compounds crayfish red claw (*Cherax quadricarinatus*).

62  
63 Research on the identification of the composition of non-volatile flavor compounds in gourami (*Osphronemus gouramy*)  
64 has not been found in Indonesia. Characteristics of non-volatile flavor compounds of a commodity are identified to be  
65 used as mapping data for the flavor composition of a food product. Information regarding non-volatile flavor compounds is  
66 important to analyze the effect of the steaming method on the composition of non-volatile flavor compounds that affect the  
67 taste of a product. This data can be used as basic data as well as a reference in various applications, including for the  
68 manufacture of a product, one of which is flavor extract. In addition to data analysis of non-volatile flavor compounds, it is  
69 also necessary to carry out organoleptic tests. Organoleptic test is also called subjective measurement and has a high  
70 relevance to product quality and quality control. The organoleptic test used is a descriptive test which functions to identify  
71 and get a complete picture of the important sensory characteristics of a product, as well as provide information about the  
72 degree or intensity of these characteristics. Based on the background that has been described, it is necessary to conduct  
73 research on the composition of non-volatile flavor compounds in fresh gourami and its steamed products.

## 74 75 **2. METHODOLOGY**

### 76 77 **2.1 Time and Place of Research**

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79 This research was conducted from December 2022 to February 2023, taking place at the Fishery Product Processing  
80 Laboratory, Faculty of Fisheries and Marine Sciences, Padjadjaran University for sample preparation and organoleptic  
81 tests. While the analysis of the amino acid profile using the method High Performance Liquid Chromatography (HPLC)  
82 was conducted at the Integrated Laboratory, Bogor Agricultural University.

### 83 84 **2.2 Materials**

85  
86 The main ingredient used is gourami (*Osphronemus gouramy*) as much as 2.5 kg from the fish cultivation site in the Cirata  
87 Reservoir, Purwakarta. The chemicals used for the analysis of amino acid compounds were Orthophthaldehyde or (OPA)  
88 50 mg, sodium hydroxide, 10 g boric acid, 1 ml Brij-30 30% solution, 1 ml 2-mercaptoethanol, 0.5 standard amino acid  
89 solution.  $\mu\text{mol/ml}$  1 ml, Na-EDTA 5 g, Methanol 200 mL, tetrahydrofuran (THF) 10 mL, Na-acetate 5 g and HP 2 L water.  
90 The tools used in this study were steamer, HPLC Ultra Techspere, erlenmeyer, knife, plastic wrap, aluminum foil, scales  
91 (Tanita) with an accuracy of 0.1 gr, plastic zip-lock, stove, label paper and cool box.

### 92 93 **2.3 Research Methods**

94  
95 The method used in this research is an experimental method including analysis of non-volatile flavor compounds and  
96 simple descriptive tests. Data analysis of non-volatile flavor compounds with the method of analysis of amino acid  
97 compounds through High Performance Liquid Chromatography (HPLC) which will then be analyzed descriptively  
98 comparatively.

99  
100 **2.3.1 Sampling**

101  
102 The first stage of this research was taking gourami samples from fish farms in the Cirata Reservoir, Purwakarta. The 2.5  
103 kg gourami samples obtained were transported alive and fresh in plastic filled with water and oxygen to the FPIK UNPAD  
104 Fishery Products Processing Laboratory, Jatinangor.

105  
106 **2.3.2 Sample Preparation**

107  
108 Sample preparation was carried out at the FPIK UNPAD Fisheries Processing Laboratory. Samples of steamed gourami  
109 were cleaned, weeded and then weighed as much as 100 grams to identify non-volatile flavor compounds and 1 fresh  
110 gourami for organoleptic testing then steamed until cooked for  $\pm 30$  minutes at 100°C.

111  
112 **2.3.3 Packaging**

113  
114 Samples of steamed gourami that have been weighed are packed using aluminum foil, labeled and re-coated using cling  
115 wrap then put in plastic zip-lock. The packed sample is put inside a coolbox containing ice, then taken to the Integrated  
116 Laboratory, Bogor Agricultural University. Samples were analyzed using High Performance Liquid Chromatography  
117 (HPLC).

118  
119 **2.4 Parameters Observation**

120  
121 **2.4.1 Amino Acid Profile Analysis**

122  
123 The first step of amino acid analysis is to dissolve 2 g of the sample that has been measured in a 50 mL flask, then filter it  
124 with millipore paper. After that, potassium borate buffer pH 10.4 was added in a 1:1 ratio. a 10  $\mu$ L sample was put into a  
125 clean empty vial and added 20  $\mu$ L OPA reagent and left for 1 minute so that the derivatization was complete. The sample  
126 was then injected into the HPLC column as much as 5  $\mu$ L and then waited until the separation of all amino acids was  
127 complete. The time required is about 25 minutes.

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129 **2.4.2 Gourami Fish Product Description Test**

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131 Simple descriptive sensory analysis is a sensory analysis method in which the attributes of a product or food ingredient  
132 are identified and described. The simple descriptive test procedure was a sample of steamed gourami was prepared, a  
133 questionnaire was prepared, the sensory properties of the product were identified, and the results of the product sensory  
134 identification were described.

135  
136 **2.5 Data Analysis**

137  
138 Quantitative data from the analysis of amino acid profiles will be discussed in a comparative descriptive manner.  
139 Comparative descriptive research is research that compares one or more variables in two different samples or at different  
140 times [61].

141  
142 **3. RESULTS AND DISCUSSION**

143  
144 **3.1 Identification of Non-volatile Flavor Compounds of Gourami Fish**

145  
146 The results of liquid chromatography analysis of non-volatile flavor compounds showed that there were 15 types of amino  
147 acids detected in the steamed gourami samples. The amino acids contained in the steamed gourami samples were  
148 divided into two, namely essential amino acids and non-essential amino acids. The dominant amino acid compounds  
149 identified in the steamed gourami sample were glutamic acid of 4.09%, aspartic acid in steamed gourami of 2.67%,  
150 arginine 1.64% and alanine 1.53% which belong to the **non-essential amino acids**. The dominant essential amino acids in  
151 steamed gourami were lysine (2.54%), leucine (2.11%), and valine (1.30%).

152  
153 There are several factors that affect the amino acid content in fish meat such as season, type of fish, body size, age,  
154 stage of maturity, environmental temperature, level of feeding, digestible content in feed, stages of the organism's life  
155 cycle, and availability of food ingredients [35];[46];[60]. According to [43], the formation of free amino acids is influenced  
156 by parameters of fish species, feed, habitat, freshness of raw materials, storage and **processing**. The heating process on  
157 gourami samples can increase the amount of free amino acids formed compared to the amount in fresh gourami. The  
158 formation of free amino acids can be increased due to a proteolytic reaction during the heating process [37];[68].

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**Table 1. Amino Acid Composition of Steamed Gourami Sample**

Amino Acid	Value (%)
Aspartate	2,67
Glutamate	4,09
Serine	1,01
Histidine	0,66
Glycine	1,13
Threonine	1,19
Arginine	1,64
Alanine	1,53
Tyrosine	0,91
Methionine	0,79
Valine	1,30
Phenylalanine	1,14
Isoleucine	1,29
Leucine	2,11
Lysine	2,54
<b>Total Amino Acid</b>	<b>23,98</b>

*Amino acid composition (mg/100g) of steamed snakehead fish sample*

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Changes in protein levels in fish are associated with reduced water content in fish during the steaming process. The higher the temperature and the longer the processing time can result in the protein content in the food being damaged and decreasing [14]. According to [54], the protein content of gourami is 18.93% greater than goldfish (16.04%) [50], and tilapia (17.33%) [19].

The value of glutamic acid content detected in steamed gourami was 4.09%. Glutamic acid can be found in foods that have a high protein content such as meat, seafood, boiled meat (broth) and soy sauce without the addition of any components, glutamate can be used to add to the taste of food such as increasing the savory taste [67]. In fishery products, glutamic acid is an amino acid that is commonly found and can create aroma and taste characteristics in food ingredients [47]. According to [10], aspartic acid and glutamic acid greatly determine the taste of umami. Glutamic acid can provide flavor umami if the concentration in the food product is above the taste threshold [31];[32];[74];[80]. Meanwhile, serine, glycine, alanine, and tyrosine contribute to the sweet taste. In addition, the interaction of several sweet amino acids such as serine, glycine and alanine with IMP can improve umami taste [33].

Glutamate ions stimulate several types of nerves that are on the human tongue. This property is used as a flavor enhancer in food [73]. According to [64], if the glutamic acid content found in fish meat is low, then the taste of fish meat will be less savory. Several fish processing methods, such as steaming can increase glutamic acid levels in fish meat. The steaming method can increase the glutamic acid content in gourami meat because the steaming process uses high heat which can change the protein structure in gourami and change the amino acids contained therein. There is an increase in the content of glutamic acid and aspartic acid during the processing at certain temperatures and cooking times [2]. The increase in glutamic acid content in meat is caused by deamination between the amino acids glutamine and asparagine which form glutamic acid so that glutamic acid levels in meat increase [55].

The content of aspartic acid identified in steamed gourami was 2.67%. In addition to glutamic acid compounds, aspartic acid also contributes to the taste of umami. Aspartic acid is a non-essential amino acid which is found in fish protein. According to [66], aspartic acid and glutamic acid work together to create a savory taste in food, especially the flavor of the seafood product. Taste of umami which is perfectly produced through a combination of glutamic acid (savory-salty) and aspartic acid (savory-sweet) [15].

194 The leucine compound detected in steamed gourami was 2.11%. Leucine is an amino acid that contributes to a bitter  
195 taste but not as bitter as phenylalanine compounds [31]. Leucine is an essential amino acid that belongs to the class of  
196 ketogenic amino acids, namely amino acids that can produce ketone compounds in the liver [38]. Leucine is able to  
197 increase insulin secretion and improve glycemic control in patients with uncontrolled blood glucose levels [44];[58].  
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199 Lysine compound identified in steamed gourami was 2.54%. Lysine has a contribution in giving a bitter taste [42];[78]. The  
200 most destructive processing method is heating and the most affected amino acid is lysine [5]. There are several factors  
201 that affect the lysine content of fish in processing processes such as bacterial activity, changes in pH, oxygen, heat and  
202 light or a combination thereof [27]. Lysine is an amino acid that has a function as a basic material for blood antibodies,  
203 becomes a framework in the formation of niacin (Vitamin B3), strengthens the circulatory system, maintains normal cell  
204 growth, works together with proline and vitamin C to form collagen tissue, lowers blood triglyceride levels. excess.  
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206 The content of arginine compounds in steamed gourami is 1.64%. Arginine is part of the essential amino acids which are  
207 alkaline and give food a bitter taste [48];[57]. Arginine is an amino acid that belongs to the group of glucogenic amino  
208 acids, namely amino acids that can be converted into glucose and glycogen [38]. According to [80], concentrations of  
209 arginine compounds that are below the threshold increase saltiness and contribute to the taste of umami on pig's hair [32].  
210 The high content of arginine compounds can provide a sweet taste and distinctive flavor of seafood on crabs and scallops  
211 [74]. Arginine is an essential amino acid that the body needs to make seminal fluid (semen), as well as increase growth  
212 hormone production [69].  
213

214 The value of the alanine compound identified in steamed gourami was 1.53%. Alanine is an amino acid that contributes to  
215 the sweet taste of food [57]. Alanine is a compound with a nonpolar R group that is used as a glucogenic precursor and  
216 nitrogen carrier from surface tissues for nitrogen excretion [34]. The high alanine content is found in seafood like snow  
217 crab, clam, and scallop [22];[56];[75]. Alanine is an important source of energy that can boost immunity, supply energy to  
218 muscle tissue, the brain, and the human central nervous system.  
219

220 Valine compound identified in steamed gourami was 1.30%. Valine is an amino acid compound that gives a bitter taste  
221 but not as bitter as phenylalanine [31];[57]. Valine is a branched-chain amino acid that functions as a glucogenic  
222 precursor, so it is needed for the growth and maintenance of muscle tissue, stimulates mental abilities, muscle  
223 coordination, helps repair damaged tissue, and maintains nitrogen balance [26]. According to [70], deficiency of valine  
224 compounds can cause loss of muscle coordination and the body becomes very sensitive to pain, cold and heat.  
225

226 The isoleucine compound detected in steamed gourami was 1.29%. Isoleucine is an amino acid that can give food a bitter  
227 taste [57]. Isoleucine compounds are needed for optimal growth, repairing damaged tissue, developing intelligence,  
228 maintaining the body's nitrogen balance, forming other non-essential amino acids, forming hemoglobin, and stabilizing  
229 blood sugar levels [18]. Deficiency of isoleucine compounds can trigger symptoms of hypoglycemia.  
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231 The content of glycine compounds in steamed gourami is 1.13%. According to [52], glycine is classified as a non-essential  
232 amino acid for humans. Glycine works together with glutamic acid and aspartic acid to give fish products a distinctive  
233 taste. Glycine is an active flavor component that gives characteristic sweet taste to various aquatic products [48]. Glycine  
234 plays an important role in stimulating the release of growth hormone, helping muscle development and growth, and wound  
235 healing. Glycine is needed in Hb synthesis and can be used to reduce stomach acidity.  
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237 Threonine compound identified in steamed gourami was 1.19%. Threonine is an amino acid compound that gives a sweet  
238 taste to foodstuffs [57]. According to [28], threonine is able to improve the ability of the intestines and digestive processes,  
239 maintain protein balance, is important in the process of forming collagen and elastin, helps the function of the heart and  
240 central nervous system and prevents epileptic seizures. Threonine compounds also function to maintain the proper  
241 protein balance in the body and are important in the maintenance of the intestinal mucosa [39].  
242

243 The detected value of the phenylalanine compound in steamed gourami was 1.14%. Phenylalanine is a compound that  
244 contributes to giving food a bitter taste [57]. Phenylalanine is an amino acid that has a dominant function in the body's  
245 work system. The amino acid phenylalanine also plays a role in relieving pain and alleviating depression. Phenylalanine is  
246 needed by the thyroid gland to produce thyroxine which has the benefit of preventing goiter, playing a role in increasing  
247 basal metabolism, and regulating body temperature [3]. A deficiency in phenylalanine amino asthma can cause the body  
248 to become weak, lethargic, liver damage, and stunted growth.  
249

250 The content of serine compounds in steamed gourami was 1.01%. Serine compounds play a role in giving a sweet taste  
251 [57];[76]. Serine is one of the amino acids found in fish meat which functions as a component of protein, plays a role in  
252 various biochemical processes in the body, is needed for fatty acid metabolism, and boosts the immune system [18].  
253 According to [34], serine is a component of phospholipids containing hydroxyl groups. Serine is used as a precursor to  
254 ethanolamine and choline.

255 Tyrosine compounds identified in the steamed gourami sample was 0.91%. The amino acid tyrosine has a similar function  
256 to phenylalanine which produces a bitter taste [76]. Tyrosine is an amino acid that has a phenol group and is weakly  
257 acidic. Tyrosine belongs to a group of non-essential amino acids which function to reduce stress, antidepressants, drug  
258 detoxification, and cocaine [1];[34]. According to [41], tyrosine also acts as a high antioxidant against peroxy free radicals.  
259 Lack of tyrosine compounds can cause hypothyroidism [18].  
260

261 The methionine compound of steamed gourami was 0.79%. The methionine compound is an amino acid that gives a bitter  
262 taste [57]. Methionine can be found in large quantities in animal food sources. Methionine plays an important role in fat  
263 metabolism, maintains liver health, prevents accumulation of fat in the liver and main arteries, prevents allergies and  
264 osteoporosis [64].  
265

266 Histidine detected in steamed gourami was 0.66%. Histidine is known to give a bitter taste but not as bitter as  
267 phenylalanine [31]. Histidine gives a very weak taste and it is thought that this amino acid does not play a direct role in  
268 taste characteristics [51]. Histidine content can be reduced due to fat oxidation [7]. Histidine is one of the essential amino  
269 acids needed by the human body for protein synthesis and various physiological functions. Histidine plays a role in the  
270 nervous system and carnosine is an amino acid [63].  
271

### 272 **3.2 Test Description of Steamed Milk Fish**

273 The test data for the description of steamed gourami showed that steamed gourami had a complete appearance with a  
274 pale white color. The aroma of steamed gourami is fresh and tasty according to the specific type of gourami. The texture  
275 of the steamed gourami meat is dense, soft and very compact. The taste of steamed gourami is savory according to the  
276 specific type.  
277

278 Color or shape is one of the important parameters in food ingredients, both those that go through a manufacturing process  
279 and those that do not go through a manufacturing process. According to [45], color can provide information about the  
280 conditions of chemical changes in a food. A food ingredient will not be consumed if it has an unsightly color, so that color  
281 can be used to determine quality [72]. Discoloration of fish meat is caused by physical and chemical changes during  
282 storage [24]. [25] stated that the binding of the solution by the meat tissue resulted in a change in the color of the meat.  
283 The color of the fish meat fades because the protein in the meat experiences aggregation which can inhibit the formation  
284 of milking in the spine [16]. According to [11], meat color can affect meat quality, consumer preferences, and indicators of  
285 maturity of the cooking process. The cooking process using high temperatures can affect the color of fish meat [62].  
286

287 Aroma is one of the parameters that determine the good taste of a food product [23]. Aroma or smell is one of the  
288 delicious parameters of a food and has its own charm to determine the good taste of a food product. The fishy smell is a  
289 characteristic fish odor caused by nitrogen components, trimethylamine oxide (TMAO), guanidine, and imidazole  
290 derivatives.  
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292 The gourami meat that has been steamed has a dense, soft and very compact texture. Changes in the texture of steamed  
293 fish became soft and decreased elasticity due to protein denaturation which resulted in the release of large amounts of  
294 water. According to [29], the texture of food ingredients is influenced by several things such as the ratio of protein content,  
295 type of protein, fat, water content, water activity, and processing temperature. The texture of a food is mostly determined  
296 by the water content contained in the product [21]. The higher the water content in the fish makes the texture softer and  
297 not crunchy.  
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299 Taste is the tongue's response to stimuli provided by a food and can influence consumers on a food product so that it has  
300 an important role in determining the final decision [12]. The savory taste of gourami is related to the presence of amino  
301 acid compounds such as glutamic acid or its salts in a food ingredient, for example monosodium glutamate and 5-  
302 nucleotide types such as guanidine 5-monophosphate (GMP), inosine 5-monophosphate (IMP) [71]. Taste is influenced  
303 by several factors such as chemical compounds, temperature, concentration, and interactions with other taste  
304 components. According to [77], free glutamic acid is contained in all fish species and is the most important flavor  
305 contributor.  
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### 307 **4. CONCLUSION**

308 Based on the description of the discussion, it can be concluded that steamed gourami has identified non-volatile flavor  
309 compounds as many as 15 amino acid compounds. Two umami-flavored amino acids are 4.09% glutamic acid and 2.67%  
310 aspartic acid. The four sweet tasting amino acids are 1.53% alanine, 1.19% threonine, 1.13% glycine and 1.01% serine.  
311 The nine bitter amino acids are lysine 2.54%, leucine 2.11%, arginine 1.64%, valine 1.30%, isoleucine 1.29%,  
312 phenylalanine 1.14%, tyrosine 0.91%, methionine 0, 79% and histidine 0.66%. Steamed gourami has an intact  
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316 appearance and is pale white in color. The aroma of steamed gourami is fresh and tasty according to the specific  
317 gourami. The texture of the meat is dense, soft and very compact. The taste of steamed gourami is savory according to  
318 the specific type.  
319

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## 325 COMPETING INTERESTS

326 Authors have declared that no competing interests exist.  
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