



An Evaluation of the Suitability of *Pangasius hypophthalmus* for Smoke-Drying: Assessment of its Nutritional Quality and Safety

Praveen Kumar G.¹, Amjad K. Balange^{2*}, Martin Xavier K. A.², Binaya Bhusan Nayak², Sanath Kumar H.², Gudipati Venkateshwarlu³

¹ Department of Fish Processing Technology, APFU-College of Fishery Science, Muthukur - 524 344, Nellore District, Andhra Pradesh, India

² Department of Post-Harvest Technology, ICAR-Central Institute of Fisheries Education, Versova, Mumbai-400 061, Maharashtra, India

³ ICAR-NAARM, Rajendranagar, Hyderabad 500 030, Telangana, India

Abstract

A study was conducted to evaluate the suitability of *Pangasius* as a raw material for smoke drying. In the present study, smoke drying was done similar to the methods followed by traditional fishermen but the experiments were conducted in controlled conditions in laboratory. *Pangasius* was hot smoked at 80°C for 3 hours, followed by mechanical drying at 60°C. The prepared product was cooled and quality of the product was evaluated using proximate composition, physicochemical parameters, benzopyrene content, microbial enumeration, analysis of fatty acids, analysis of amino acids and sensory evaluation. It was observed that the moisture content of smoked-dried *Pangasius* was below 15%. A decrease in pH to 5.96 and an increase in Total Volatile Basic Nitrogen (TVBN) to 24.45 mg N/100g was observed in smoked-dried *Pangasius*. After smoke-drying, the Peroxide Value (PV), Thiobarbituric Acid Reactive Substances (TBARS) and Free Fatty Acids (FFA) has increased to 2.98 meq O₂/kg fat, 1.35 mg MDA/kg fat and 1.29% oleic acid respectively. Total Viable Count (TVC) and *Staphylococcus spp.* were within the acceptable limit. Saturated fatty acids increased during smoke drying with palmitic acid dominating others. Among amino acids, glutamic acid, lysine and aspartic acid were dominant. The benzopyrene content of smoked-

dried *Pangasius* was observed to be 7 ppb. From the results, it was observed that the quality parameters of smoked-dried *Pangasius* were within the limit of acceptability with good sensory scores suggesting its acceptance. Hence, it was concluded that smoke-drying can be used as a method for preserving *Pangasius* by controlling the benzopyrene concentration in smoke during processing.

Key words: Smoking, drying, quality, fatty acids, aminoacids

Introduction

Fish is one of the widely preferred food all over the world. Fish plays an important role in human diet as it has high PUFA, animal protein and essential amino acids along with micronutrients like minerals (calcium, iron, zinc and selenium) and vitamins (A, B and D). Increased consumption of fish can reduce problems related to malnutrition (Anaemia in women and obesity) and nutritional deficiencies like iron, zinc, iodine and vitamin A can be treated (FAO, 2020). Marine capture fisheries resources are continuously depleting due to over exploitation and pollution. Aquaculture production has increased significantly but not all aquaculture produce fetch good price in the market. *Pangasius* (*Pangasius hypophthalmus* Sauvage, 1878) is one of such species, which has high production rate but it is not fetching a good price in the market. According to MPEDA (2018), Indian aquaculture production in the year 2017-18 was 7,05,600 MT of which *Pangasius* production was 26,293 MT with Andhra Pradesh as leading producer (24,845 MT) followed by

Received 16 May 2022; Revised 19 October 2022; Accepted 23 October 2022

*E-mail: amjadbalange@cife.edu.in

Maharashtra (573 MT). Therefore, there is a need to develop suitable processing methods for *Pangasius*.

Smoke-dried fishery products are popular mostly in the North-Eastern region and Lakshadweep Islands in India. Smoking is a traditional method of fish preservation of economic importance worldwide for thousands of years as it increases the shelf life of fish by depositing antioxidant and antimicrobial compounds generated from smoke along with dehydration of the product (Goulas & Kontominas, 2005; Djinojic et al., 2008). In recent days, smoking is being used to attain characteristic colour, odour, texture and taste for the product (Ciecėlik et al., 2017). Smoking is a widely applied method as smoke can inactivate harmful enzymatic compounds and microorganisms (Šimko, 2002). Though smoking increases the shelf life of the product, deposition of benzopyrene on the product may be carcinogenic to the consumers. So, an attempt was made to utilize marketable size *Pangasius* to prepare smoke dried product and evaluate its quality and safety.

Materials and Methods

25 kg of fish was purchased from Khar land Research Station in Panvel, Mumbai, India and immediately iced in the ratio of 1:1 (fish:ice) and brought to the laboratory within 4 hours. The length and weight of *Pangasius* varied from 37 - 42 cm and 850 - 1200 g respectively. After reaching laboratory, the fishes were washed with chilled water to remove slime and dirt. Later the fishes were eviscerated and were splitted open in butterfly style from dorsal side. The prepared fish were washed with chilled water and were hanged in head up position for smoking and drying.

Smoking was carried out similar to the method followed by traditional fish smokers in Amalapuram district in Andhra Pradesh, India. Smoking was carried out in two stages. Initially, the fillets were pre-dried in accelerated mechanical drier at 50°C for 30 min for surface drying of the fillets where no smoking was involved. Later, the smoking was carried out in Accelerated mechanical dryer (Yarrow & Co Ltd, Glasgow, Scotland) at temperatures of 80°C ± 5°C with 0.5 m/s air velocity for 3 hours. After 3 hours, smoking was stopped and *Pangasius* was dried at 60°C ± 5°C in the accelerated mechanical dryer for 36 h where the required moisture content of 20 % was attained as described by Kumar et al. (2017) (Unpublished data). In this

method, salt is not added in any step during preparation of smoked-dried *Pangasius*.

Four fishes were taken for sampling and 3 of them were used for microbial and biochemical analysis and one was used for sensory evaluation. For biochemical analysis, the muscle of fishes was mixed and was used for further analysis.

The moisture content, fat content, protein content and ash content were determined according to AOAC (2000) and the results were represented as percentage.

To evaluate pH of smoked-dried *Pangasius*, 10 g of muscle was taken and added to 50 ml distilled water and was homogenized in a homogenizer (Polytron system PT 2100, Kinematica, AG, Germany) for 30 s. The pH was measured using digital pH meter (Eutech Instruments, Singapore). TVBN was analysed by steam distillation method with slight modifications using trichloro acetic acid as described by Vyncke (1996). PV was analysed using the method of Jacob (1958) and the results were expressed as meq of O₂/ kg of fat. The FFA was determined according to the method described in AOAC (2000). TBARS were analysed by the method as described by Tarladgis et al. (1960).

Benzopyrene was analyzed by the GC-MS-MS based on the procedure of Chatterjee et al. (2016). Gas chromatograph (GC 7890A) coupled to a triple-quadrupole mass spectrometer (MS, 7000B) (Agilent Technologies, Palo Alto, CA, USA) was used for performing residue analysis. Analytical separation was done on HP-5MS (30 m × 0.25 mm, 0.25 μm) capillary column with helium as carrier gas at a constant flow rate of 1.2 ml/min with initial oven temperature set at 70 °C (1 min hold), later ramped to 150°C at 25°C/min, to 200°C at 3°C/min and finally to 285°C at 8°C/min (9 min hold), resulting in a total run time of 40.49 min and the transfer line temperature was maintained at 285°C. Multi-mode inlet (MMI) was used in solvent vent mode with injection volume of 5 μl.

The microbiological evaluation of Total Viable Count (TVC), *Escherichia coli* and *Staphylococcus* were carried out as per APHA (1992). The lipids were extracted according to the protocol of Folch et al. (1957). The fatty acid methyl esters (FAME) were prepared and the methylated fatty acids were analysed using GC-MS (QP2010, Shimadzu, U.S.A.)

as per the AOAC (1995) method. The operational parameters of the equipment were set as per Keer et al. (2018).

In a test tube, 100 mg of coarsely homogenized material was placed and 10 mL of 6 N HCl was poured. Further, it was infused with nitrogen for all amino acid analysis except for tryptophan. The contents were digested in an oven at 110°C for 24 h. After digestion, contents were cooled and filtered through Whatman No. 1 filter paper and vacuum flash evaporated until the content is acid-free (after three washings of 50 mL). The residue was dissolved in Buffer A (sodium citrate, ethanol, citric acid, NaOH and Brij with final pH 3.2) and filtered using 0.45 µm syringe filter. Then the sample (20 µL) was injected into high-performance liquid chromatography (Shimadzu chromatograph LC-10ATvp). The flow rate of mobile phase solvent A (sodium citrate + ethanol (pH 3.5)) and solvent 'B' (sodium citrate + NaOH (pH 9.8)) were maintained at 0.4 mL/min and the temperature of the column was set at 60 p C. The fluorescence excitation and emission wavelengths were 340 and 450 nm, respectively. After derivatization by O-phthalaldehyde, amino acids were identified and quantified by comparison of their retention times with those of standards (Sigma). The results were expressed in terms of percentage amino acid per total amino acids.

In case of tryptophan, a 300 mg sample was added with 10 mL of 5 % NaOH, digested and the contents were neutralized to pH 7.0 with 6 N HCl. A 0.1 mL of 2.5 % sucrose and 0.1 mL of 0.6 % thioglycolic acid were poured into the test tube containing 4 mL of 50 % H₂SO₄. Further, the tubes were kept in a water bath at 45-50°C for 5 min and cooled. Lastly, 0.1-0.8 mL of sample was added to test tubes and mixed well and made up to 5 mL with 0.1N HCl and kept aside for 5 min. Tryptophan content was measured spectrophotometrically at 530 nm (Sastry & Tammuru, 1985). The results were expressed in

terms of percentage amino acid per total amino acids.

The sensory evaluation of smoked and smoke-dried Pangasius was performed by 10 trained panelists in Department of Fish Processing, CIFE, Mumbai. 9 point hedonic scale was used to score the product from 9 to 1 viz., Like extremely (9), Like very much (8), Like moderately (7), Like slightly (6), Neither like nor dislike (5), Dislike slightly (4), Dislike moderately (3), Dislike very much (2), Dislike extremely (1). The scores from all panelists were collected and average was calculated.

In this study, all the analyses were done in triplicates and were subjected to statistical tests. To check whether there is a significant difference between the means at P<0.05, analysis of variance was performed by using one-way ANOVA and applying Duncan's multiple range tests and descriptive statistics in SPSS 16 (SPSS Inc., Chicago, IL, USA) software. The results in the tables were reported as mean values of determinations ± Standard Deviation.

Results and Discussion

In smoked and smoked-dried Pangasius, there was an increase in protein, fat and ash with a decrease in moisture. It was observed that moisture content decreased from 78.14 % (fresh Pangasius) to 67.34 % in smoked Pangasius and further decreased to 14.96 % in smoked-dried Pangasius (Table 1). Nahid et al. (2017) reported a decrease in moisture and an increase in lipid and protein contents with loss of water. Bhulyan et al. (1986) reported a decrease in moisture and increase in lipid content in Atlantic mackerel. Similar results were observed by Ünüsayın et al. (2001) in European eel, pike perch and rainbow trout. The prevalence of low moisture content in fish muscle becomes an unsuitable environment for spoilage organisms and chemical activities (Nahid et al., 2017). Lilabati & vishwanath (1996) suggested that the right or acceptable

Table 1. Changes in proximate composition (%) of smoked and smoke dried Pangasius

	Moisture	Protein	Fat	Ash
Fresh	78.14±0.45 ^a	18.23±0.04 ^a	2.29±0.36 ^a	1.44±0.85 ^a
Smoked	67.34±0.53 ^b	22.18±0.08 ^b	6.68±0.10 ^b	3.50±0.31 ^b
Smoked--Dried	14.96±0.34 ^c	66.55±0.12 ^c	9.57±0.29 ^c	4.72±0.18 ^c

* Each value is represented as mean ± SD (n =3).

Different superscripts in the same column indicates the values are significantly different (pd≤0.05)

moisture content of smoked-dried fish was 20 %. The moisture content of smoked-dried Pangasius was lower than the acceptable limit.

The results of protein content indicated that protein content increased from 18.23 % in fresh Pangasius to 22.18 % in smoked Pangasius and again increased to 66.55 % in smoked-dried Pangasius (Table 1). Due to loss of moisture during smoking and drying, the proteins might have concentrated and led to an increase in the protein content (Koral et al., 2009; Kumolu-Johnson et al., 2010). Fapohunda and Ogunkoya (2006) reported an increase in protein and fat contents during smoke-drying of *Clarias gariepinus*. Nahid et al. (2016) reported that protein content increased in smoked-dried chapila, kaika and baim fish to 45.25, 74.85 and 70.85% respectively.

The fat content of smoked-dried Pangasius increased from 2.29 % (fresh Pangasius) to 6.68 % (smoked Pangasius) and later increased to 9.57 % (smoked-dried Pangasius) due to the removal of water from Pangasius during drying (Table 1). Nahid et al. (2016) reported an increase in fat content in smoked-dried chapila (*Gudusia chapra*, Hamilton-Buchanan; 1822), commonly called as Indian river shad, kaika (*Xenentodon cancila*, Hamilton-Buchanan; 1822) commonly called as Freshwater garfish and baim (*Mastacembelus pancalus*, Hamilton-Buchanan; 1822), commonly called as Barred spiny eel. Lipid content of smoked-dried chela fish (19.27 %) was higher than sun-dried chela fish (14.00 %) (Al-Reza et al., 2015). Huda et al. (2010) reported a fat content of 32.06 % and 8.02 % in smoked *Cryptopterus micronema* and *Macrones nemurus* respectively.

The ash content increased from 1.44 % in fresh Pangasius to 3.50 % (smoked Pangasius) and further increased to 4.72 % in smoked-dried Pangasius

(Table 1). Incineration removes the organic matter and the inorganic content will remain as ash (Clucas & Ward, 1996). Smoking results in concentrating of proteins and fat (Doe & Olley, 1990). Akintola et al. (2013) reported an increase in ash content during smoking and sun drying of *Penaeus monodon* from 12.66 to 13.57 %. The ash content of salted smoked-dried and dried batashi fish (Indian potasi) on the dry weight basis was 12.57 and 16.90, respectively (Rana & Chakraborty, 2016). Huda et al. (2010) reported that the ash content of smoked *Cryptopterus micronema* and *Macrones nemurus* was 5.41 % and 0.73 %, respectively.

A decrease in pH was observed during smoking of Pangasius from 6.67 to 6.13, which further decreased to 5.96 during smoke-drying of Pangasius (Table 2). The decrease in pH may be attributed to the effect of phenolic/ acidic constituents deposited on the fish muscle during smoking (Doe et al., 1998). Dhar et al. (2014) reported a pH of 5.99 and 5.92 in *Amblypharyngodon mola* and *Puntius sophore*, respectively.

An increase in TVBN from 4.95 mg N/100g to 6.46 mg N/100g was observed during smoking of Pangasius. The TVBN value further increased to 24.4 mg N/100g during smoke-drying of Pangasius (Table 2). According to Connell (1990), an increase in the TVBN was due to the bacterial spoilage of fillets and the limit of acceptability of TVBN is 35-40 mg N/100g. The TVBN values in the present study were lower than the maximum limit of acceptability indicating that the product was of good quality and was consumable. The high value of TVBN in smoked-dried fishes might be due to subsequent microbial and biochemical changes in fish muscle during smoking (Lilabati & Vishwanath, 2001). Dhar et al. (2014) reported a TVBN value of 23.1 and 10.6 in mechanically smoked *Amblypharyngodon mola* and *Puntius sophore*, respec-

Table 2. Physico-chemical changes of smoked and smoke dried Pangasius

	pH	TVBN (mg N/100g)	PV (meq of O ₂ /kg of fat)	TBARS (mg MDA/kg)	FFA (% oleic acid)
Fresh	6.75 ± 0.05 ^c	4.95 ± 0.18 ^a	0.07 ± 0.01 ^a	0.04 ± 0.00 ^a	0.09 ± 0.03 ^a
Smoked	6.13±0.01 ^b	6.46±0.35 ^b	0.35±0.04 ^b	0.57±0.04 ^b	0.35±0.00 ^b
Smoke-Dried	5.96±0.01 ^a	24.45±0.32 ^c	2.98±0.08 ^c	1.35±0.02 ^c	1.29±0.48 ^c

* Each value is represented as mean ± SD (n =3).

Different superscripts in the same column indicates the values are significantly different (p≤0.05)

tively. Akiba et al. (1967) reported that amino and volatile basic nitrogen compounds of fish increased during smoking.

The peroxide value of smoked Pangasius has increased from 0.07 meq of O₂/kg fat to 0.35 meq of O₂/kg fat, which further increased to 2.98 meq of O₂/kg of fat in smoked-dried Pangasius (Table 2). The meager increase of PV in smoked Pangasius can be attributed to low exposure time and the increase in PV of smoked-dried Pangasius may be due to extended exposure to higher temperature and air. Connell (1975) stated that a peroxide value of 10-15 meq/Kg of lipids indicates rancidity of the product. The PV values in the present study were within the limit of acceptability indicating that not much oxidation has occurred during smoke drying of Pangasius. Dhar *et al.* (2014) reported a PV of 11.46 and 9.85 meq O₂/kg in mechanically dried smoked samples whereas traditionally dried *Amblypharyngodon mola* and *Puntius sophore* samples had lower values. The oxidative deterioration of fat is due to smoking and exposure to oxygen during subsequent drying and storage (Bhuiyan et al., 1986).

The free fatty acid value increased in smoked Pangasius to 0.06 % oleic acid from an initial value of 0.04 % oleic acid and further increased to 1.29 % oleic acid in smoked-dried Pangasius (Table 2). The

hydrolysis of lipid during smoking and subsequent drying and storage may lead to the formation of free fatty acids (Huss, 1994). When the FFA value is 0.5 -1.5 % oleic acid, most fatty acids begin to be noticeable to the palate (Pearson, 1976). The value of FFA was below the maximum level of acceptance indicating that the product is of good quality. Dhar et al. (2014) reported an FFA value of 0.61 and 0.38% oleic acid in mechanically dried *Amblypharyngodon mola* and *Puntius sophore*, respectively, which was lower than the traditionally dried samples. Daramola et al. (2007) reported that the percentage FFA of five smoked fresh-water fish species, *Heterotis niloticus*, *Labeo coubie*, *Parachanna obscura*, *Oreochromis niloticus* and *Clarias gariepinus* ranged between 0.91-1.96% oleic acid.

Not much change has occurred in TBARS value during smoking of Pangasius. But there was increase in TBARS value of smoked-dried Pangasius from 0.04 mg MDA/kg (smoked) to 1.35 mg MDA/kg (Table 2). The lower TBARS value in smoked Pangasius can be correlated to lower PV values of smoked Pangasius. The increase in TBARS value indicates that oxidation has occurred and the increase in TBARS of smoked-dried Pangasius might be due to the production of free radicals during an extended period of drying. TBARS values higher than 3-4 mg MDA/kg indicates a loss of product quality (Cakli et al., 2006). The TBARS

Table 3. Microbial changes of smoked and smoke dried Pangasius

	TVC (log cfu g ⁻¹)	<i>Staphylococcus spp.</i> (log cfu g ⁻¹)	<i>E. coli</i> (MPN/g)
Fresh	4.69	<1.39	<3.0
Smoked	3.78	<1.39	<3.0
Smoked-Dried	2.56	<1.39	<3.0

* Each value is represented as average of 2 samples

Table 4. Sensory changes of smoked and smoke dried Pangasius

Days	Colour	Appearance	Odour	Texture	Taste	Smoke flavor	overall
Fresh	8.82±0.25 ^c	8.86±0.19 ^b	8.80±0.19 ^c	8.70±0.19 ^b	8.86±0.28 ^c	0.00±0.00 ^a	8.94±0.38 ^c
Smoke	8.62±0.45 ^b	8.80±0.07 ^b	8.70±0.07 ^b	8.62±0.04 ^b	8.68±0.08 ^b	8.46±0.05 ^b	8.65±0.02 ^b
Smoked-dried	8.12±0.68 ^a	8.22±0.18 ^a	8.18±0.04 ^a	8.06±0.05 ^a	8.3±0.10 ^a	8.1±0.07 ^a	8.17±0.07 ^a

* Each value is represented as mean ± SD (n =10).

Different superscripts in the same column indicates the values are significantly different (p≤0.05)

values in the present study were within the limits of acceptability stating that the tertiary oxidation of product was low. Bouriga et al. (2012) reported a TBARS value of 1.29 and 0.76 mg MDA/kg in traditionally and industrially smoked tilapia fillets, respectively in which the initial value was 0.47 mg MDA/kg. According to Adenike (2014), the TBARS value of smoke-dried sliced catfish ranged from 0.14 to 0.24 mg MDA/kg.

The total viable counts of smoked Pangasius decreased from 4.69 log cfu g⁻¹ (fresh Pangasius) to 3.78 log cfu g⁻¹, which has further decreased to 2.56 log CFUg-1 in smoked-dried Pangasius (Table 3). It was observed that the TVC of smoked-dried Pangasius was lower than smoked Pangasius, which may be due to the increased exposure of microbes to high temperatures. The lower TVC value in smoked and smoked-dried Pangasius may be attributed to the bactericidal properties of smoke (Huss, 1994). According to Eklund et al. (1988), smoking imparts a high degree of microbial stability to the product due to reduced water activity, heating and smoking. According to FSSAI (2017), the maximum limit of TVC in dried fish product is 5 log CFU/g. The TVC value of smoked-dried Pangasius was within the limit of acceptability indicating that the product was microbiologically acceptable. The bacterial counts of salted smoked-

dried and ring tunnel dried batashi fish (*Neotropius atherinoides*) decreased from 2.72×10⁵ CFU/g to 1.14×10⁴ and 1.88×10⁴ CFU/g respectively (Rana & Chakraborty, 2016). The total viable count of smoke-dried chapila, kaika and baim fish also decreased during smoking (Nahid et al., 2016). The decline in total viable count may be due to the bactericidal effect of constituents of smoke namely acids, aldehydes and phenols (Eyo, 2006). The *E. coli* and *Staphylococcus spp.* was not detected in smoked and smoke dried Pangasius.

The changes in fatty acids of smoked-dried Pangasius are presented in Table 5. The saturated fatty acids (SFA) decreased from 60.80 (fresh) to 58.64 % during smoking and again increased to 60.84 % in smoked-dried Pangasius respectively. Regulska-Illow et al. (2013) while analyzing fatty acid of smoked halibut, mackerel, bloater and sprat reported that SFA accounted for one-fourth of fatty acids analysed with palmitic acid as dominant fatty acid and the highest percentage of palmitic acid was observed in sprats and the lowest in halibut. Usydus et al. (2009) reported that palmitic acid was dominant fatty acid among SFA in the fat of smoked sprats. Monounsaturated fatty acids (MUFA) in Pangasius increased with smoking (39.11%) and smoke-drying (36.52%) from an initial value of 35.08%. Polyunsaturated fatty acids (PUFA) of Pangasius decreased

Table 5. Changes in fatty acids (%) of smoked and smoked-dried Pangasius

	Common name	Fresh fish	smoked	Smoked-dried
C14:0	Myristic acid	8.07	8.25	8.92
C15:0	Pentadecyclic acid	0.53	0.51	0.83
C16:0	Palmitic acid	50.52	49.25	50.07
C17:0	Margaric acid	0.37	0.36	0.48
C18	Stearic acid	0.22	0.21	0.07
C20:0	Arachidic acid	0.04	0.06	0.11
Σ Saturated fatty acids		60.8	58.64	60.84
C18:1(n-9)	Oleic acid	33.69	36.87	35.25
C20:1(n-9)	Eicosenoic acid	0.93	0.76	0.23
C22:1(n-9)	Erucic acid	0.46	1.48	1.04
Σ Mono unsaturated fatty acids		35.08	39.11	36.52
C18:2(n-6)	Linoleic acid	1.36	0.95	0.29
C20:4(n-6)	Arachidonic acid	1.76	1.12	2.02
C22:6(n-3)	Docosapentaenoic acid	0.2	0.18	0.33
Σ Poly unsaturated fatty acids		4.12	2.25	2.64

with smoking (2.25%) and smoke-drying (2.64%) from an initial value of 4.12%. The MUFA of smoked halibut, mackerel, bloaters and sprats were 39.8 %, 26 %, 31.1 % and 30.4 %, respectively with oleic acid, the primary fatty acid in MUFA was 16.67 %, 9.72 %, 12.54 % and 18.05 %, respectively (Regulska-Ilow et al., 2013). The decrease in PUFA in smoked-dried Pangasius may be due to oxidation of fat during drying for a longer period and at high temperature. Usydus et al. (2009) reported that n-3 PUFA of smoked mackerel, herring and sprats were 10.6, 19.2 and 22.3 %, respectively. Regulska-Ilow et al. (2013) have reported the percentage of PUFA in smoked halibut, mackerel, bloaters and sprats were 31.9, 45.4, 40.8 and 37 % of total fatty acids respectively. The decrease in PUFA, MUFA and increase in SFA may be due to the unstable nature of unsaturated fatty acids and presence of oxygen, which accelerates oxidative rancidity and decrease in moisture content during smoking and drying (Little et al., 2000; Kaya et al., 2008; Bouriga et al., 2020). Similar results were observed by Ciecelik et al. (2017) in smoked common carp, Rainbow trout and Northern pike

and Bouriga et al. (2020) in hot smoked zander fish.

The changes in the amino acid profile of smoked-dried Pangasius is shown in Table 6. The essential amino acids of smoked-dried Pangasius increased from 38.8 % to 47.5 %. The non-essential amino acids decreased in smoked-dried Pangasius from 61.19 % to 52.49 %. Among the essential amino acids, major changes were observed in phenylalanine, histidine, lysine and tryptophan, which have shown an increase during smoke drying, whereas Leucine decreased after smoke drying. Among essential amino acids, lysine (13.36 %) was the most dominant amino acid followed by leucine (7.59 %). Among non-essential amino acids, major changes were seen in glutamic acid, alanine and arginine, which have declined. But there was an increase in proline and glycine after smoke-drying of Pangasius. Glutamic acid (19.37 %) was the dominant amino acid in non-essential amino acids followed by aspartic acid (12.26 %). Among all amino acids, the glutamic acid was dominant followed by lysine and aspartic acid. Aremu et al. (2013) reported that glutamic acid and

Table 6. Changes in amino acid (%) of smoked and smoke dried Pangasius

Amino acid Name	Abbreviation	Fresh	Unsalted Smoked-dried
Threonine	Thr	2.64	2.77
Valine	Val	6.44	6.33
Methionine	Met	1.17	1.64
Isoleucine	Ile	2.71	2.85
Leucine	Leu	8.65	7.59
Phenylalanine	Phe	4.4	6.46
Histidine	His	3.14	4.33
Lysine	Lys	9.65	13.36
Tryptophan	Try	0	2.17
Σ Essential amino acids		38.8	47.5
Aspartic acid	Asp	13.09	12.26
Serine	Ser	3.74	3.37
Glutamic acid	Glu	24.04	19.37
Proline	Pro	0	1.47
Glycine	Gly	2.93	3.82
Alanine	Ala	5.82	4.44
Cysteine	Cys	0	0.1
Tyrosine	Tyr	1.92	0
Arginine	Arg	9.65	7.66
Σ Non-essential amino acids		61.19	52.49

aspartic acid were the dominant amino acids ranging between 12.87 to 14.24 g/100 g crude protein (CP) and 8.93 to 9.71 g/100 g CP, respectively in catfish (*Clarias gariepinus*) smoked using sawdust, melon husk, electric oven and rice bran. Lysine with a range of 7.02 to 7.78 g/100 g CP was the most concentrated essential amino acid followed by leucine ranged from 6.80 g/100 g CP in rice bran heat treatment to 7.32 g/100 g CP in electric oven (Aremu et al., 2013). Essential amino acids in smoked *Cryptopterus micronema* were higher than smoked *Macrones nemurus* and the content of glutamic acid was greater than other amino acids (Huda et al., 2010). Swastawati et al. (2012) reported an increase in lysine during smoking from an initial value of 0.28 and decreased during storage. Similar results were observed by Ciecelik et al. (2017) in smoked common carp, rainbow trout and northern pike and Ayeloja et al. (2020) in smoked *Oreochromis niloticus*.

The overall sensory score of smoked Pangasius was higher than the smoked-dried Pangasius with values 8.65 and 8.17, respectively (Table 4). It indicates that smoked Pangasius was highly preferred by panelists than the smoked-dried Pangasius. The probable reason for this preference may be due to its cooked effect and succulence of smoked fish. The colour characteristics of smoked fish not only depend on pigmentation of skin but also on quantity and composition of smoke deposits and their interactions with tissue components (Sikorski et al., 1998). Phenolic compounds deposited during smoking process attributed to the colour, taste and flavour of

smoked fish (Dhar et al., 2014). From the results, it was observed that the prepared smoke-dried Pangasius was acceptable and have a good appearance, odour, taste and texture making the product delicious. The high value for overall acceptance in smoked Pangasius may be due to its succulence, golden colour and taste. In smoked-dried Pangasius, it was observed that there was a shift in colour from golden (smoked Pangasius) to brown (smoked-dried Pangasius) due to drying.

From the results, it was observed that benzopyrene concentration of 7 ppb was detected in the smoked-dried Pangasius (Figure 2) whereas the benzopyrene content was below detectable level in smoked Pangasius (Figure 1). According to the European Union, 5 ppb of benzopyrene is the maximum retention limit in food. In the present study, it was observed that the levels of benzopyrene in smoked-dried Pangasius was higher than the acceptable limit indicating that the product was carcinogenic to consumers. It was observed that in smoked Pangasius, benzopyrene was not detected, but it was detected in smoked-dried Pangasius. Concentration of benzopyrene in the muscle during drying process might be the probable reason for the increase in benzopyrene levels of smoked-dried Pangasius.

From the results, it was observed that the quality parameters of smoked and smoked-dried Pangasius was within the limit of acceptability indicating that smoked-dried Pangasius is suitable for consumption. Presence of high protein, fat, along with higher

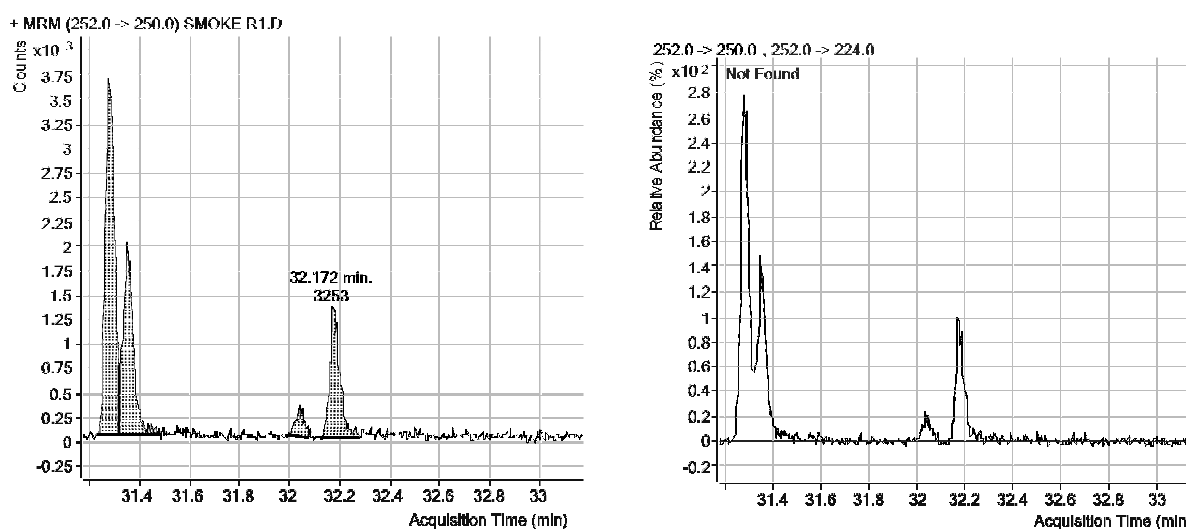


Figure 1a. Chromatogram showing retention time and peak for benzopyrene 1 and 2 of smoked Pangasius

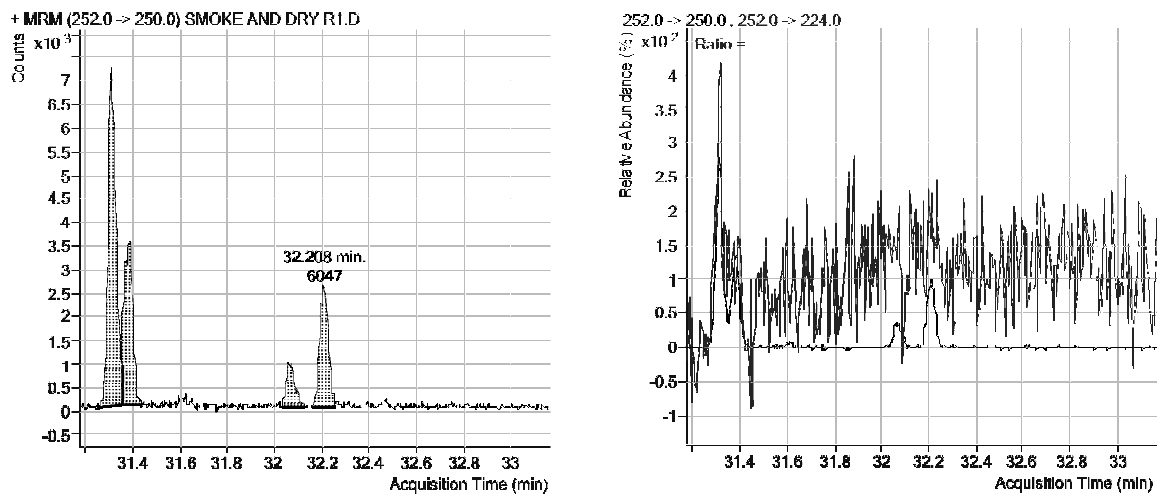


Figure 1b. Chromatogram showing retention time and peak for benzopyrene 1 and 2 of smoked-dried Pangasius

saturated and monounsaturated fatty acids and high essential amino acids in smoked-dried Pangasius indicate its high nutritional quality. It was also observed that benzopyrene content in smoked-dried Pangasius exceeded the maximum retention limit whereas it was not detected in smoked Pangasius. So, it can be concluded that Pangasius can be used as a candidate species for preparation of smoked and smoked-dried products with good nutritional quality only if proper precautions are taken during the manufacturing to reduce the benzopyrene content in smoke generated by using filters.

Acknowledgements

The first author is thankful to Indian Council of Agricultural Research (ICAR) and Director, ICAR-Central Institute of Fisheries Education, Mumbai for providing necessary facilities for conducting this study. The first author also remains grateful to University Grants Commission (UGC) for providing financial assistance in the form of fellowship (RGNF). Authors are thankful to Dr. Kaushik Benerjee, Principal Scientist, National Referral Laboratory, ICAR-National Research Centre for Grapes, Pune for helping us in analysing Benzopyrene.

References

- Adenike, O.M. (2014) The effect of different processing methods on the nutritional quality and microbiological status of catfish (*Clarias lezera*). J. Food Process. Technol., 5(6): 333-340
- Akiba, M., Motohiro, T. and Kudo, S. (1967) Studies on smoked fish products. Bull. Fac. Fish. Hokkaido Univ., 157-161.
- Akintola, S.L., Brown, A., Bakare, A., Osowo, O.D. and Omolola, B. (2013) Effects of hot smoking and sun drying processes on nutritional composition of giant tiger shrimp (*Penaeus monodon*, Fabricius, 1798). Pol. J. Food Nutr. Sci., 63(4):227-237
- Al-Reza, S., Karmaker, S., Hasan, M., Roy, S., Hoque, R. and Rahman, N. (2015) Effect of traditional fish processing methods on the proximate and microbiological characteristics of *Laubuka dadiburjori* during storage at room temperature. J. Fish. Aquat. Sci., 10(4): 232-243
- AOAC (1995) AOAC Official Methods of Analysis. 13th ed., Vol. 15, Association of Official Analytical Chemists, Arlington, VA., pp.132-320
- AOAC (2000) AOAC Official Methods of Analysis, 18th Edition Association of Official Analytical Chemists, Maryland, U.S.A
- APHA (American Public Health Association), (1992) *Compendium of Methods for the Microbiological Examination of Foods*. 3rd ed. ed. D C. Vander dent, and splittstoesser. washington Dc 2
- Aremu, M.O., Namu, S.B., Salau, R.B., Agbo, C.O. and Ibrahim, H. (2013) Smoking methods and their effects on nutritional value of African catfish (*Clarias gariepinus*). Open Nutraceuticals J., 6: 105-112
- Ayeloja, A.A., Jimoh, W.A., Adetayo, M.B. and Abdullahi, A. (2020) Effect of storage time on the quality of smoked *Oreochromis niloticus*. Heliyon, 6(1), e03284
- Bhuiyan, A.K.M.A., Ratnayake, W.M.N. and Ackman, R.G. (1986) Effect of smoking on the proximate composition of Atlantic mackerel (*Scomber scombrus*). J. Food Sci., 51(2): 327-329

- Bhulyan, A.K.M.A., Ratnayake, W.M.N. and Ackman, R.G. (1986) Stability of lipids and polyunsaturated fatty acids during smoking of Atlantic mackerel (*Scomber scombrus* L.). *J. Am. Oil Chem. Soc.*, 63(3): 324-328
- Bouriga, N., Bejaoui, S., Jemmali, B., Quignard, J.P. and Trabelsi, M. (2020) Effects of smoking processes on the nutritional value and fatty acid composition of Zander fish (*Sander lucioperca*). *Grasas y Aceites*, 71(1), e340-e340
- Bouriga, N., Ben, I.H., Gammoudi, M., Faure, E. and Trabelsi, M., (2012) Effect of smoking-method on biochemical and microbiological quality of Nile Tilapia (*Oreochromis niloticus*). *Am. J. Food Technol.*, 7(11): 679-689
- Cakli, S., Kilinc, B., Dincer, T. and Tolasa, S. (2006) Comparison of the shelf lives of MAP and vacuum packaged hot smoked rainbow trout (*Onchoryncus mykiss*). *Eur. Food Res. Technol.*, 224(1): 19-26
- Chatterjee, N.S., Utture, S., Banerjee, K., Shabeer, T.A., Kamble, N., Mathew, S. and Kumar, K.A. (2016) Multiresidue analysis of multiclass pesticides and polyaromatic hydrocarbons in fatty fish by gas chromatography tandem mass spectrometry and evaluation of matrix effect. *Food Chem.*, 196: 1-8
- Cieœlik, I., Migda³, W., Topolska, K., Mickowska, B. and Cieœlik, E. (2017) Changes of amino acid and fatty acid profile in freshwater fish after smoking. *J. Food Process. Preserv.*, 42(1), e13357
- Clucas, I.J. and Ward, A.R. (1996) Post Harvest Fisheries Development; A Guide to handling, preservation, processing and quality. Natural Resources Institute. U.K. pp.5- 428
- Connell, J.J. (1975) Control of Fish Quality. Fishing News Books Ltd., UK., pp: 137
- Connell, J.J. (1990) Control of fish quality, 3rd edn. Fishing News Books, Oxford
- Daramola, J.A., Fasakin, E.A. and Adeparusi, E.O. (2007) Changes in physicochemical and sensory characteristics of smoke-dried fish species stored at ambient temperature. *African J. Food, Agric. Nutr. Dev.*, 7(6): 1-15
- Dhar, B., Karthikeyan, M. and Roy, D. (2014) Quality characteristics of two freshwater fishes smoked under traditional and mechanical methods. *Fish. Technol.*, 51(3): 187-193
- Djinovic, J., Popovic, A. and Jira, W. (2008) Polycyclic aromatic hydrocarbons (PAHs) in different types of smoked meat products from Serbia. *Meat Sci.*, 80(2): 449-456
- Doe, P. and Olley, J. (1990) Drying and Dried Fish Products. In: *Seafood: Resources, Nutritional Composition, and Preservation* (Sikorski, Z.E. Ed.) CRC Press, Boca Raton, Florida. pp.125-145
- Doe, P.E., Sikorski, Z., Haard, N., Olley, J. and Pan, B.S. (1998) Basic principles. In: *Fish Drying and Smoking: Production and Quality* (Doe, P. E. Ed.), Technomic Publishing Co. Inc., USA, pp 13-46
- Eklund, M.W., Peterson, M.E., Paranjpye, R. and Pelroy, G.A. (1988) Feasibility of a heat-pasteurization process for the inactivation of nonproteolytic *Clostridium botulinum* types B and E in vacuum-packaged, hot-process (smoked) fish. *J. Food Prot.*, 51(9): 720-726
- Eyo, A.A. (2006) *Fish processing technology in the tropics*. University of Ilorin Press., pp.104-189
- FAO (2020) *State of World Fisheries and Aquaculture. Sustainability in Action 2020*. FAO Technical publication, Rome, Italy
- Fapohunda, O.O. and Ogunkoya, M. (2006) Effect of smoke-drying on the proximate composition of *Tilapia zillii*, *Parachanna obscura* and *Clarias gariepinus* obtained from Akure, Ondo-State, Nigeria. *Anim. Res. Int.*, 3(2): 478-480
- Folch, J. (1957) A simple method for the isolation and purification of total lipids from animal tissues. *J. biol. Chem.*, 226, 497-509
- FSSAI (2017) Final Gazette Notification of Food Safety and Standards (Food Product Standards and Food Additives) Third Amendment Regulation, 2017 related to microbiological standards for fish and fish products. Dt: 13.02.2017, Food Safety and Standards Authority of India, New Delhi
- Goulas, A.E. and Kontominas, M.G. (2005) Effect of salting and smoking-method on the keeping quality of chub mackerel (*Scomber japonicus*): biochemical and sensory attributes. *Food chem.*, 93(3): 511-520
- Huda, N., Dewi, R.S. and Ahmad, R. (2010) Proximate, color and amino acid profile of Indonesian traditional smoked catfish. *J. Fish. Aquat. Sci.*, 5(2): 106-112
- Huss, H.H. (1994) Assurance of Seafood Quality, FAO Fisheries Technical Paper. 334, Rome, FAO, 169 pp
- Jacob, M.B. (1958) *The Chemical Analysis of Foods and Food Products*. Kreiger Publishing Co. Inc. New York, USA. pp.393-394
- Kaya, Y., Turan, H. and Emin Erdem, M. (2008). Fatty acid and amino acid composition of raw and hot smoked sturgeon (*Huso huso*, L. 1758). *Int. J. Food Sci. Nutr.*, 59(7-8): 635-642
- Keer, U., Alim, H., Xavier, M., and Balange, A.K. (2018). Quality Changes during Ice Storage of Acetes Species. *Int. J. Curr. Microbiol. Appl. Sci.*, 7(1), 2063-2071
- Koral, S., Köse, S. and Tufan, B. (2009) Investigating the

- quality changes of raw and hot smoked garfish (*Belone belone euxini*, Günther, 1866) at ambient and refrigerated temperatures. *Turk. J. Fish Aquat. Sci.*, 9(1): 53-58
- Kumolu-Johnson, C.A., Aladetohun, N.F. and Ndimele, P.E. (2010) The effects of smoking on the nutritional qualities and shelf-life of *Clarias gariepinus* (BURCHELL 1822). *Afr. J. Biotechnol.*, 9(1): 73-76
- Lilabati, H. and Vishwanath, W. (1996) Biochemical, nutritional and microbiological quality of three smoked fishes (*Wallago attu*, *Tor putitora*, *Puntius jayarami*) sold in the Imphal market, Manipur. *J. Freshwater Biol.*, 8(4): 217-223
- Lilabati, H. and Vishwanath, W. (2001) Biochemical and microbiological changes during storage of smoked *Puntius sophore* obtained from market. *J. Food sci. Technol.*, 38(3): 281-282
- Little, S.O., Armstrong, S.G. and Bergan, J.G. (2000) Factors affecting stability and nutritive value of fatty acids: culinary practices. *Fatty acids in foods and their health implications.* (Ching Kuang Chow, Ed.), pp.427-437. CRC Press, Boca Raton, Florida
- MPEDA (2018) MPEDA Annual Report 2017-18. The Marine products Export Development Agency, Ministry of Commerce and Industry, Government of India
- Nahid, M.N., Farid, F.B. and Begum, M. (2017) The determination of shelf life quality of three smoke-dried freshwater fishes in refrigeration storage. *Int. J. Adv. Sci. Res.*, 2(1): 17-24
- Nahid, M.N., Latifa, G.A., Chandra, S., Farid, F.B. and Begum, M. (2016) Shelf-life quality of smoke-dried freshwater fish/ ; chapila (*Gudusia chapra*, Hamilton-Buchanan; 1822) kaika (*Xenentodon cancila*, Hamilton-Buchanan; 1822) and baim (*Mastacembelus pancalus*, Hamilton-Buchanan/ ; 1822) stored at laboratory. *J. Agri. and Vet. Sci.* 9(3): 23-32
- Pearson, D. (1976) *The Chemical Analysis of Foods.* 7th edition, Churchill, Livingstone, Edinburgh, London and New York
- Rana, M.M. and Chakraborty, S.C. (2016) Effect of salt and smoke on quality and shelf life of salt-smoke-dried batashi (*Neotropius atherinoides*) kept at different storage condition. *Res. in Agric. Livest. and Fish.*, 3(3): 443-451
- Regulska-Ilow, B., Ilow, R., Konikowska, K., Kawicka, A., Rózańska, D. and Bochińska, A. (2013) Fatty acid profile of the fat in selected smoked marine fish. *Rocz. Panstw. Zakl. Hig.*, 64(4): 299-307
- Sastry, C.S.P. and Tummuru, M.K. (1985) Spectrophotometric determination of tryptophan in proteins. *J. Food Sci. Technol.*, 22(2), 146-147
- Schulze, K. and Zimmermann, T. (1983) Untersuchungen an Raucherfischen unter besonderer Berücksichtigung von makrelen. *Arch. Lebensmittelhyg.*, 34(3): 67-70
- Sikorski, Z., Haard, N., Motohiro, T. and Pan, B.S. (1998) Quality. In: *Fish Drying and Smoking: Production and Quality* (Doe, P.E. Ed.), Technomic Publishing Co. Inc., USA, pp.106-113
- Šimko, P. (2002) Determination of polycyclic aromatic hydrocarbons in smoked meat products and smoke flavouring food additives. *J. Chromatogr. B.*, 770(1): 3-18
- SPSS (2010) *SPSS for windows.* Release 16. Chicago, 1: SPSS Inc
- Swastawati, F., Susanto, E., Cahyono, B. and Trilaksono, W.A. (2012) Sensory evaluation and chemical characteristics of smoked stingray (*Dasyatis bleekeri*) processed by using two different liquid smoke. *Int. J. Biosci. Biochem. Bioinformatic.*, 2(3): 212-216
- Tarladgis, B.G., Watts, B.M., Younathan, M.T. and Dugan, L. (1960) A distillation method for the quantitative determination of malonaldehyde in rancid foods. *J. AM. Oil Chem. Soc.*, 37(1): 44-48
- Ünlüsayın, M., Kaleli, S. and Gulyavuz, H. (2001) The determination of flesh productivity and protein components of some fish species after hot smoking. *J. Sci. Food Agric.*, 81(7): 661-664
- Usydus, Z., Szlinder-Richert, J., Polak-Juszczak, L., Komar, K., Adamczyk, M., Malesa-Cieciewicz, M. and Ruczynska, W. (2009) Fish products available in Polish market—Assessment of the nutritive value and human exposure to dioxins and other contaminants. *Chemosphere*, 74(11): 1420-1428
- Vyncke, W. (1996) Comparison of the official EC method for the determination of total volatile bases in fish with routine methods. *Arch. Lebensmittelhyg.*, 47: 110-111