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RESEARCH ARTICLE.....

Standardization of thermal processing of cuttlefish nidamental gland

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ABSTRACT..... Cuttlefish nidamental gland was blanched in boiling 5 per cent brine for 2, 5, 10 and 15 minutes to standardise the blanching condition. The sample blanched for 5 minutes was found to have the best sensory qualities. Sub-sequently, glands blanched under these standardised conditions were packed in cans in brine and were subjected to thermal processing in a retort. Thermal processing conditions were standardised by processing the cans under different retorting conditions. Out of these, the retorting temperature and the process time of 121.1° C and 25 minutes, respectively was found to be the best with respect to sensory qualities and could produce a commercially sterile product. The F_o value attained by the process was 11.25 minutes with a total process time of 30.54 minutes. The storage study of the product over a period of three months showed that it has good stability at ambient condition and has a shelf-life of more than three months. The present investigation delineates that cuttlefish nidamental gland can be used for the production of ready-to-eat thermally processed product, which can be stored at room temperature for long periods.

KEY WORDS..... Cuttlefish, Nidamental gland, Heat penetration, Thermal processing, Sensory evaluation, Cephalopod

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INTRODUCTION.....

Thermal processing is one of the most important methods of food preservation. At present, the demand for thermally processed foods is on the rise due to the increasing attraction of the consumers towards convenience ready-to-eat foods. The main cause of their increasing demand is that they are shelf-stable, due to which they can be conveniently kept in room condition without the need of refrigeration during their storage and distribution. With respect to the preservation of fish, among the different methods available, thermal processing stands next only to freezing. In 2012, out of the total world fish catch, about 11 per cent was preserved by thermal processing (FAO, 2014). The demand for thermally processed products is predicted to increase considerably in the future due to its superior quality, ease of use and safety of consumers' health.

Starting from the discovery of thermal processing of foods by Nicholas Appert in 1804, a lot of research has been conducted on this aspect to prevent its spoilage, assure its safety and improve its quality (Russell, 1895; Prescott and Underwood, 1897; Barlow, 1913 ; Esty and

Meyer, 1922). Attempts have also been made to include diversified food commodities in the list of thermally processed products. The important categories of food commodities include fruits, vegetables, meat and fish. The present investigation was carried out to evaluate the suitability of cuttlefish nidamental gland for thermal processing.

Cuttlefish are cephalopods, which are exclusively marine molluscs. There are about 660 species of cephalopods in the world oceans (Voss, 1977 and Worms, 1983). Among them, less than a hundred species are of commercial importance. In the Indian seas, there are about 80 species of commercially important cephalopods (Oommen, 1977 and Sarvesan, 1974). In the recent years, the importance of cephalopods has increased due to their increasing demand in the export trade next to shrimp. Among the cephalopods, squid and cuttlefish are two commercially important species. Squid is particularly important for its superior nutritional quality, as its meat forms a high-protein, low-fat food material (Suryanarayanan *et al*., 1973 and Joseph *et al*., 1977). An excellent account on the utilization of squid for human consumption has been given by Kreuzer (1984). However, very little work has been done on thermal processing of these species in contrast to their counterparts such as fish and shrimp. It has been found that squid constitute a good raw material for preparation of thermally processed foods due to their firm texture after blanching (Varma and Joseph, 1980 and Parshwanath, 1989). The canning procedure for squid mantles in brine has been developed by Raghunath and Solanki (1986). The canning procedure for ready-to-eat squid masala in indigenous polymer-coated tin-free steel cans was developed by Sreenath *et al*. (2007).

In comparison to squid, very little work has been done on cuttlefish, though its importance with respect to human consumption and world seafood trade is similar to those of the former. Cuttlefish is exported in different forms such as whole, whole cleaned, double skinned, strips, beaks, wing and roe. The seafood product exported under the name cuttlefish roe is, in fact, its nidamental gland, which is a pair of flattened glands associated with the female reproductive system. The glands are extracted from mature females and exported in frozen condition. In India, the nidamental glands obtained from the entire catch is frozen and exported to major importers like Italy, Belgium, Portugal and other European countries. In Italy, it is taken after boiling and with oils as in salads, which is a cheaper substitute for oysters. However, very little work has been done on the nidamental gland of cuttlefish. Santhosh Kumar *et al*. (1999) have studied the nutrient composition of this gland from the cuttlefish, *Sepia pharaonis*. In view of the above facts, the present investigation was taken up to develop a procedure for the thermal processing of cuttlefish nidamental gland, so as to prepare a shelf-stable ready-to-eat product that would require no refrigeration for its storage and distribution and no further processing before its consumption.

RESEARCH METHODS.....

Raw material :

Mature female cuttlefish, *Sepia pharaonis* procured from the fish landing center at Kochi were iced quickly and transported in iced condition to the Fish Processing Laboratory of College of Fisheries, Panangad, Kerala, India. The cuttlefish were dissected with a sharp stainless steel knife and the nidamental glands were removed from the visceral cavity manually. The collected glands were washed with chilled potable water to remove blood and other adhering tissues. The washed glands were packed in polythene pouches and kept at 4.0 ± 0.5 °C in the chilling cabinet of a domestic refrigerator (Kelvinator, India) until further processing in the same day.

The container :

Three-piece tin cans of 4½ oz. fluid capacity having trade dimension of 301 x 203 with internal lining of sulphur resistant lacquer were used in the present study for packing of the glands and further thermal processing.

Standardization of blanching conditions :

The washed glands were blanched in boiling 5 per cent brine as recommended by Saralaya (1978) for canning of other mollusks like clam and green mussel. Blanching was done for different time periods of 2, 5, 10 and 15 minutes. After blanching the samples were subjected to sensory evaluation to find out the optimum period of blanching. The sensory evaluation was done, using a 10-point hedonic scale, by a group of 10 experienced panelists who are accustomed to the consumption of blanched and thermally processed seafood. The panelists were requested to evaluate the

products separately without biasness. The scoring of the products was done based on the score card given in Appendix 1.

Appendix 1 : Score card for sensory evaluation of cuttlefish nidamental gland

Quality grade description	Score
Excellent	9-10
Very good	7-8
Good	5-6
Fair	3-4
Borderline of acceptability	1-2
Poor/unacceptable	0
Remarks if any	

Name of the panelist: Date of sensory evaluation:
Sample number:

Standardization of process temperature :

Twenty tin cans were washed properly using detergent solution followed by thorough rinsing with potable water. About 128 ± 2 g of blanched glands was packed into each can followed by filling it with 68 ± 2 ml of 2 per cent hot brine so as to get a standard net weight of about 197 ± 2 g, a gland: brine ratio of 65:35 and a head space of 6 mm as per Saralaya (1978). To ensure perfect hermetic sealing of the cans, utmost care was taken to avoid the contamination of their seal area. The cans with their lids on were subjected to steam exhausting for 10 minutes in an autoclave followed by sealing immediately using a can double seamer (Metal Box Company, Kolkata, India). The sealed cans were manually washed using detergent solution followed by adequate rinsing with potable water.

For application of two different thermal processes, which are commercially adopted for thermal processing of canned seafood, the cans were grouped into two sets of ten numbers each. Then the cans were subjected to thermal processing in a laboratory model overpressure retort (John Fraser and Sons Ltd, Newcastle-upon-Tyne, UK). One set was subjected to a thermal process of 115.0 °C for 60 minutes and the other to a process of 121.1 °C for 30 minutes. After thermal processing, the cans were quickly removed from the retort and subjected to water cooling by dipping in chilled (4±1°C) potable water chlorinated to a residual chlorine level of 5 ppm. After cooling, the cans were removed from the cooling water and subjected to air drying.

To determine the optimum thermal process with respect to sensory quality, the cans were opened using a can opener (Metal Box Company, Kolkata, India) and the contents poured onto cleaned glass plates. The sensory evaluation of the thermally processed products was done by the same sensory evaluation process as mentioned earlier for the standardization of blanching condition.

Standardization of process time (B) :

For the Standardization of process time (B), the nidamental glands processed and packed in tin cans as described above, were retorted at the selected process temperature for different time periods of 20, 25 and 30 minutes. For the three time periods, three batches of ten cans each were prepared. Evaluation of the thermal processes was performed by testing the cans for commercial sterility as per IS 2168 (1971). For this, six cans were selected at random from each batch. Three cans from each batch were incubated at 37 °C for 15 days and the other three at 55 °C for 5 days. The incubated cans were opened aseptically and about 1 g sample from each can was taken by a sterilized forceps. The samples were inoculated into two sets of sterilized fluid thioglycollate broth tubes. A layer of sterilized liquid paraffin was put on the top of the broth so as to create anaerobic condition. One set of tubes was incubated at 37 °C for 48 hours and the other at 55 ºC for 5 days. The tubes were then observed for development of any turbidity. Nonappearance of turbidity indicates ability of the thermal process to impart commercial sterility. The rest of the four cans from each batch were subjected to organoleptic evaluation to find out the process time most suitable for producing organoleptically acceptable products.

Thermal process evaluation :

For the evaluation of thermal process, the nidamental glands processed and packed in tin cans as described above, were retorted at the selected process temperature and process time. To obtain the thermal process data, thermocouple needle (Ellab Co., Roedovre, Denmark) was inserted into the can so that its tip reached the slowest heating point (SHP) or cold spot of the can. The SHP of a round can is known to be located along its vertical axis at 1/3rd of its height from the bottom. The temperature output data of the thermocouples were recorded in an Ellab CTF 9008 data recorder (Ellab Co.,

Roedovre, Denmark). The recorded data were analyzed using an MS-Excel Software. The heat penetration data were plotted on a semi-logarithm paper with temperature deficit in logarithmic scale on Y-axis against time in linear scale on X-axis. Lag factor for heating (J_h), lag factor for cooling (J_c), slope of the heating curve (f_h) and time in minutes for sterilization at retort temperature (U) were determined. Then, the total process time (T) was obtained by adding the process time (B) and 58 per cent of the come-up-time (CUT). Finally, the F_o value was calculated by the mathematical method of Stumbo (1973).

Storage studies :

The nidamental glands retorted at the selected process temperature and time as described above were stored at room temperature and subjected to chemical and sensory analysis at a monthly interval for a total period of three months. For chemical analysis the parameters determined were pH (IS 2168, 1971), thiobarbituric acid (TBA) value (Tarladgis *et al*., 1960) and salt content (AOAC, 2000) of the glands excluding the filling medium. Sensory analysis of the processed glands during storage was done by 10 experienced panelists using a ten-point hedonic scale as per IS 6273 [II] (1971). The parameters considered in the sensory analysis were appearance, colour, odour, taste, texture and overall acceptability.

Statistical analysis :

The SPSS (2000) statistical package was used for the analysis of the experimental results. Analysis of variance (ANOVA) and Duncan multiple range test were used to find out significant difference ($P < 0.05$) among the products.

RESEARCH FINDINGS AND ANALYSIS.....

The standardization process essentially involved determination of optimum blanching condition as well as retorting temperature and time to produce thermally processed nidamental gland which is safe, shelf-stable and appetizing.

Standardization of blanching conditions :

The sensory evaluation scores and salt concentration of blanched cuttlefish nidamental gland are given in the Table 1. It was observed that, blanching in 5 per cent brine for 5 minutes gives the best sensory scores as compared to the other blanching conditions. The salt content in this product was found to be 2.15 per cent, which is very close to the recommended value of 2 per cent that should remain in any processed food preparation like fish curry (Vijayan *et al*., 1998), fish cutlet (Gopakumar, 2002), fish sausage and curry for canning (Ninawe and Rathnakumar, 2008). Blanching of the glands also resulted in the development of a firm texture. Similar phenomenon has also been reported by Varma and Joseph (1980) and Parshwanath (1989) in blanching of squid.

Standardization of retorting temperature :

The sensory evaluation scores for standardization of retorting temperature are presented in the Table 2. In the present study, two retorting temperatures *viz*., 115.0° C and 121.1° C were selected for standardization as per the common practice adopted in the commercial food canning industry where, thermal processing of low acid foods (pH $\geq$ 4.5) including seafood is generally done

Table 1: Sensory evaluation score and salt concentration of the gland after blanching **(Mean ï SD, n=10)**

Concentration of brine (%) /Time (minutes)	Texture	Saltiness	Flavour	Salt (%)
5/2	4.94± 0.25^c	6.06± 0.07^b	5.31± 0.19^c	1.57± 0.16^d
5/5	5.69 ±0.31^a	6.19± 0.19^a	6.13± 0.28^a	2.15± 0.15^c
5/10	5.63± 0.23^b	5.81± 0.07^c	5.69± 0.17^b	2.38± 0.17^b
5/15	4.94± 0.20^c	3.94± 0.11^d	5.00± 0.11^d	3.93± 0.15^a

Values with the same superscript are not significantly different (P>0.05)

Table 2 : Sensory evaluation score for standardisation of retorting temperature **(Mean ï SD, n =10)**

Temperature (°C)/ Time (minutes)	Appearance	Colour	Odour	Taste	Texture	Overall acceptability
115 /60	6.17±0.15	5.67±0.14	6.33±0.06	6.17±0.10	6.33±0.14	6.33±0.16
121.1/30	6.38±0.12	6.00±0.08	5.75±0.12	6.75±0.08	6.56±0.12	7.19±0.18

at two temperatures *viz*., 240° F (= 115.6° C) and 250° F (=121.1 °C) (Saralaya, 1978; Warne, 1988; Ninawe and Rathnakumar, 2008). According to McLay (1982) fish packs are usually processed at temperatures of 110° C - 121° C and in general, a short time at a high temperature is to be preferred. Between the two retorting temperatures used in the present study, nidamental glands retorted at the higher temperature of 121.1° C were found to have superior sensory quality as compared to those processed at 115.0° C. Therefore, the retorting temperature of 121.1° C was selected in the present study for further standardization of process time.

Standardization of process time :

For the standardization of process time, three time periods of 20, 25 and 30 minutes were chosen for the processing of the cans at the previously selected temperature of 121.1°C. The choice of the time periods was based on two facts. Firstly, retorting time of 30 minutes was already found to give a product with superior sensory quality in the present study. Secondly, a shorter processing time has economic benefits as well as better retention of nutritional value (Balachandran, 2002) for which two lower temperatures of 25 and 20 minutes were also selected for the comparative evaluation of the process time.

The results of the commercial sterility test indicated that processing at 121.1°C for all the three processing times of 20, 25 and 30 minutes were able to produce commercially sterile products (Table 3). Subsequent organoleptic evaluation of the cans indicated that thermal processing of the glands for 25 minutes gave a product with superior sensory quality as compared to those for 20 and 30 minutes (Table 4). Therefore, the process time of 25 minutes at 121.1

Table 3: Commercial sterility test of canned nidamental gland processed for different time at 121.1 °C

Time (minutes)	Observation for turbidity	Inference
20	No turbidity	Commercially sterility attained
25	No turbidity	Commercially sterility attained
30	No turbidity	Commercially sterility attained

Table 4 : Sensory evaluation score for standardisation of process time at 121.1 °C **(Mean ï SD, n =10)**

Time (minutes)	Appearance	Colour	Odour	Taste	Texture	Overall acceptability
20	6.14±0.18^{a}	5.86±0.18^{a}	6.71±0.19^{b}	6.86±0.21^{b}	7.14±0.17^{a}	6.43±0.19^{c}
25	6.14± 0.21^{a}	5.71±0.17^{b}	6.86±0.21^{a}	7.00±0.18^{a}	6.93±0.21^{b}	6.71±0.22^{a}
30	6.21± 0.19^{a}	5.57±0.20^{c}	6.57±0.22^{c}	6.86±0.19^{b}	6.71±0.18^{c}	6.57±0.20^{b}

Values with the same superscript are not significantly different (P>0.05)

Table 5 : Heat penetration data for thermally processed nidamental gland in brine in tin plate can

Parameters	Value
Fo value (minutes)	11.25
J_h	0.54
J_c	1.03
f_h (minutes)	12.50
U (minutes)	11.26
f_h /U	1.11
g (°C)	0.37
B (minutes)	24.74
Come- up time (minutes)	10.00
Total process time (minutes)	30.54
Cook value (minutes)	81.54

Note: f_h= Slope of heating curve, J_h = Lag factor of heating, J_c = Lag factor of cooling, U = Time in minutes for sterilization at retort temperature, g = Final temperature deficit, B = Ball's process time

°C was selected in the present study for the evaluation of thermal process.

Determination of F_0 value :

The rate of heat penetration into the cans packed with nidamental glands in brine during processing at a retort temperature of 121.1 °C for a process time of 25 minutes is given in Fig. 1 and 2. The process parameters and F_0 value calculated by formula method (Stumbo, 1973) are presented in the Table 5.

Although the process time was targeted for 25 minutes, a slight deviation in the targeted value was observed. The CUT should be as shorter as possible (Anonymous, 1968). The CUT was 10 minutes, the heating lag factor (J_h) was calculated to be 0.54 minutes and the cooling lag factor (J_c) was found to be 1.03 minutes. The f_h value was reported 12.50 minutes. The cook value obtained was 81.54 minutes. The total process time was determined by adding process time and 58 per cent of the CUT. The value was calculated to be 30.54 minutes. Bratt (1995) recommended an F_0 in the range of 5 to 20 minutes for fish and fishery products. In the present study an F_0 value of 11.25 was calculated for the product, which is well in between the recommended range.

Storage studies :

The results of the chemical analysis of the processed cuttlefish nidamental gland during the storage period are given in Table 6. During the storage period the salt content increased from 1.62 per cent to 1.82 per cent. However, the values were close to the recommended value of 2 per cent that should remain in any processed food

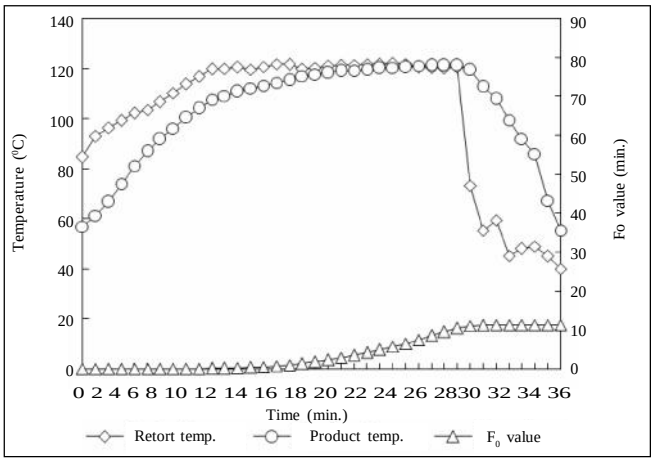

Fig. 1 : Heat penetration characteristics of nidamental gland in brine medium with respect to F_0 value

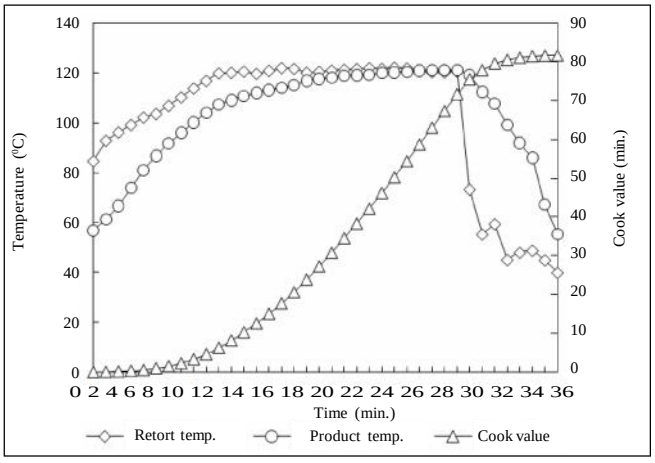

Fig. 2 : Heat penetration characteristics of nidamental gland in brine medium with respect to cook value

preparation like fish curry (Vijayan *et al*., 1998), fish cutlet (Gopakumar, 2002), fish sausage and curry for

Table 6: Changes in chemical parameters of processed nidamental gland during storage at room temperature (Mean ï SD, n =3)

Storage period (days)	Salt (%)	TBA no. (mg malonaldehyde/kg)	pH
0	1.62± 0.07^{d}	0.561± 0.02^{d}	6.35± 0.14^{a}
30	2.06± 0.13^{a}	0.776± 0.05^{c}	6.32± 0.13^{ab}
60	1.73± 0.08^{c}	0.868± 0.03^{b}	6.16± 0.11^{c}
90	1.82± 0.11^{b}	1.092± 0.04^{a}	6.24± 0.07^{bc}

Values with the same superscript are not significantly different (P>0.05)

Table 7: Sensory evaluation score of the product during storage at room temperature (Mean ï SD, n =10)

Storage period (days)	Appearance	Colour	Odour	Taste	Texture	Overall acceptability
0	7.11±0.14^{a}	6.56± 0.26^{a}	7.33 ±0.22^{c}	6.78 ±0.14^{b}	6.61± 0.21^{a}	6.78 ±0.18^{a}
30	7.36± 0.08^{b}	6.71± 0.15^{c}	7.71± 0.15^{a}	7.14±0.23^{b}	7.00± 0.29^{b}	7.57 ±0.12^{a}
60	6.75 ±0.08^{a}	6.00± 0.13^{b}	7.10 ±0.28^{b}	7.40 ±0.24^{a}	6.95± 0.32^{a}	7.40 ±0.12^{a}
90	6.56± 0.33^{a}	5.94± 0.19^{b}	7.22±0.25^{b}	6.94±0.29^{a}	6.83± 0.29^{a}	6.83± 0.21^{a}

Values with the same superscript are not significantly different (P>0.05)

canning (Ninawe and Rathnakumar, 2008). TBA value of the products increased significantly from 0.561 mg malonaldehyde/ kg to 1.092 mg malonaldehyde/ kg during the storage period. Such increase could be due to the autoxidation of lipids present in the gland by the residual oxygen which might be left over in the can during exhausting. However, the TBA values were much lower than 2 mg malonaldehyde/ kg, which is the recommended limit for fish and fishery products (Gopakumar, 2002). The pH of the canned products varied marginally between 6.35 and 6.24 throughout the period of storage, which indicates that nidamental gland is a low acid food. Similar observations have been made by Chand (1998) in canned fried mackerel in brine (pH=6.8) and by Sreenath *et al.* (2007) in canned squid in masala (pH= 5.5).

The result of the organoleptic evaluation of the thermally processed glands carried out during the storage period for the attributes such as appearance, colour, odour, taste, texture as well as overall acceptability is given in Table 7. The scores for all these attributes remained more than 6 throughout the period of storage indicating that the product is of 'very good' quality and can attract the consumers. This also implies that the thermally processed nidamental gland of cuttlefish has good stability at ambient condition and has a shelf life of more than three months.

Conclusion :

The value addition of cuttlefish nidamental gland can be done by thermal processing which gives a product having good organoleptic qualities and optimum stability at ambient conditions of storage. The optimum conditions for the processing of the gland are blanching in 5 per cent brine for 5 minutes followed by retorting at 121.1 °C for 25 minutes which ultimately results in a required F_0 value of 11.25 minutes. Hence, the technology can widely be adopted by the canning industry for commercialization of the product.

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