# Nutritional potential and Functional properties of Autolysates of Sturgeon (Acipenserbaeri) Caviar Extraction by products from Madagascar.

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## Abstract

Sturgeon byproducts (viscera) generated after caviar production are usually discarded by the industry. The purpose of this study was to determine the nutritional and functional potential of sturgeon (Acipenserbaeri) viscera autolysats. By applying enzymatic autolysis of these byproducts, it may be possible to produce autolysis fractions that are very rich in nutrients with very interesting functional properties. Biochemical characterizations of these fractions showed that up to 90% of proteins were concentrated in the supernatant and that more than 50% of the lipids were released in the oil phase. The phosphorus, iron and calcium levels in autolysates deserve alsomore focus on the nutritional benefit from these byproducts. The peptides of the derivatives have very interesting functional properties in the food industries (emulsifying activities, oil and water retention capacities).

Keywords: Co-products, autolysis, autolysates, proteins, lipids, functionalproperties

## Introduction

Fisheries and aquaculture make a significant contribution to the food security and livelihoods of millions of people around the world. For Madagascar, overall fish production reached129,365 tons in 2013 (Ministry of Fisheries Resources and Fisheries, 2014).Sturgeon fish farming is both a conservation technology and a source of economic development, by satisfying international market demand (QIWEI, 2015).It began in the former USSR in the 1970s with the Siberian sturgeon (FAO, 2006). The sturgeon is currently recognized as one of the most valuable commercial fish in the world, especially appreciated for its eggs, caviar.

Sturgeon farming was established in Madagascar by a company called ACIPENSER SAin 2013. This activity reflects the diversity of Malagasy aquaculture practice, which is one of the source of employment for the local population.

Currently, the first African and Indian Ocean caviar is produced in Madagascar thanks to the possible adventure of this company. However, caviar production generates several types of co-products, with the most abundant viscera. These non-edible co-products are present in huge quantities in the industry and their management is a problem. Yet these co-products are good sources of nutrients, including proteins (IBRAHIM et al., 1999),

lipids (DUMAY et al., 2006), minerals, vitamins (HEU et al., 2003), and other bioactive compounds (KIM et al., 2008). Indeed, the objective of this work is to valorize sturgeon viscera produced after caviar production as a new source of proteins and other substances that can be used in relation to food and nutrition. Enzyme autolysis is used as a recovery technique.

### **Materials And Methods**

#### **Biological materials**

The sturgeon viscera used in this work were kindly provided by ACIPENSER SA, the only sturgeon farm in Africa. Sturgeon farming is carried out in Lake Mantasoa, 45 kilometres from Antananarivo (the capital of Madagascar), and caviar production is carried out in its factory located in the rural commune of Ambatolaona, nearby the lake. The samples we recollected to ACIPENSER in 2018 and then were separated into 200g aliquots prior to their storage in a freezer until to use. Before any analysis, they were thawed overnight at 4°C.

#### **Biochemical characterization of raw materials**

#### **Determination of protein content**

The method used for the determination of crude protein (N x 6.25) in the sample was that of KJELDAHL, which consists of determining the mineral nitrogen content after destruction of the organic matter in the sample (GODON et al, 1991).

#### **Determination of the water content**

To determine the water content, a drying at  $103^{\circ}\pm 2C$  for 24 hours was carried out. The water contained in the sample is lost during this operation (MALEWIAK et al, 1992).

#### Fat determination

The method used was that of WOLF, which uses hexane extraction. Lipids are soluble in organic solvents. At the end of the extraction, the extracted lipids are weighed

#### Determination of the raw ash content

The crude ash composed of mineral elements is contained in the residue after calcinations at 550°C in a muffle furnace (LAURENT, 1991).

## **Determination of mineral elements**

Most common mineral element assay methods are performed on liquid media. The ashes contained in the capsule were moistened with distilled water. Depending on the amount of ash, 5 to 25ml of HCl could be added. Then, the solution transferred into a beaker was brought to a boil on a hot plate for 10 minutes. After cooling, the solution was filtered and rinsed several times with distilled water to recover more mineral elements. The volume was then reduced to 100ml. The mineral elements contents were determined from this solution.

Iron, magnesium, manganese, zinc and calcium were measured by flame atomic absorption spectrophotometer. Phosphorus was measured using a Beckman UV/VIS spectrophotometer at 430nm wavelength

#### Identification of amino acids

Amino acids were identified by using thin layer chromatography. The samples, deposited on a solid fixed phase (stationary phase), were carried by a liquid mobile phase (moving phase). The amino acids then migrate at varying distances depending on their solubility in the solvent and their affinity for the fixed phase and their molecular size. The revelation is made using a ninhydrin solution (MOORE and STEIN, 1948).

## Autolysis and determination of the degree of hydrolysis

Autolysis was performed according to the method described by CAO et al in 2008. It consists of breaking the peptide bonds of the substrate under the effect of endogenous enzymes activated by temperature.

The thawed sample was ground before being mixed with distilled water, twice the weight of the shred. The resulting mixture was heated on a hot plate at 40°C. During the three hours of reaction, the temperature was increased by 5°C, every 30 minutes. When the temperature had reached 70°C, the endogenous enzymes were inactivated by heating the mixture to 100°C for 10 minutes. The autolysate were then cooled and centrifuged for 20 minutes at 5000 rpm. The recovered phases were then dried at 70°C in an oven and biochemically analyzed.

The degree of hydrolysis was obtained from the ratio between the amount of soluble protein and the amount of total protein.

## Determination of the emulsifying capacity

The method used is the one described by NACZK et al (1985).

A quantity of 350 mg of sample powder and 5 ml of distilled water were homogenized for 30 seconds by using a vortex. 2.5 ml of soybean oil were added, then the mixture was homogenized again for 30 seconds. The same volume of oil was poured into the mixture before being homogenized on the vortex for 90 seconds. The whole was finally centrifuged for 5 minutes at 5,000 rpm. The emulsifying activity (AE) was calculated by the formula: AE= (volume of emulsion in ml/total volume in ml) x100The stability of the emulsion is the proportion of emulsion remaining after 25 minutes of rest (GBOGOURI, 2005)

## <u>Results</u>

#### Nutritional value of sturgeon viscera per 100 grams dry weight:

Analyses showed that sturgeon viscera are rich in protein, with a content of  $62.77\pm0.63\%$  based on dry weight. Lipids are present at  $35.59\pm0.01\%$  and the water content is  $65.60\pm026\%$ . The analysedviscera samples are also a good source of minerals elements with a crude ash content of  $1.69\pm0.01\%$ .

#### **Biochemical characteristics of autolysates**

Two fractions were obtained after autolysis of the sturgeon viscera: a supernatant and an oily phase. The maximum hydrolysis degree reached after 3 hours was of 46.43%.

The nutritional compositions of the autolysis fractions in relation to fresh and dry matter are summarized in the following table:

	Content in g/100g of freshmaterial		Content in g/100g of dry matter		
Parameters	± standard deviation		± standard deviati	on	
	Supernatant	Oily phase	Supernatant	Oily phase	
Water content	4,99±0,69	12,00±0,48	4,99±0,69	12,00±0,48	
Proteins	88,36±0,04	48,15±0,05	93,98±0,04	55,10±0.05	
Fats	0,11±0,04	56,42±0,88	0,50±0,00	64,12±0,88	
Rawashes	1,57±0,00	0,71±0,00	1,67±0,00	0,81±0,00	

#### Table 01: Nutritional composition of autolysis fractions

Biochemical characterizations showed that autolysis fractions are excellent sources of protein with a content of 93.98% in the supernatant and lipids of 64.12% in the oil phase.

Table 02: Mineral element composition per 100 grams of autolysates											
	Samples	Fe (mg/100g)	Mn (mg/100g)	Mg 1g/100g)	Ca (mg/100g)	P (mg/100g)	Zn (mg/100g)				
	Supernatant	11,00±0,00	0,40±0,000	23,00±0,00	8,00±0,00	6,00±0,00	0				

The mineral elements measured in the autolysis fractions are summarized in the table below: Table 02: Mineral element composition per 100 grams of autolysates

 $0.70\pm0.00$ 

The results revealed that the mineral elements are distributed in both fractions. However, zinc is absent in the supernatant. Iron and magnesium are the main mineral elements present in the two fractions.

22.00±0.00

 $18.00 \pm 0.00$ 

 $1.30\pm0.00$ 

 $2.00\pm0.00$ 

#### Amino acids identified in autolysis fractions

 $26.00 \pm 0.00$ 

Eleven amino acids were revealed on the chromatogram, six of which were found in the dried supernatant (Methionine, Histidine, Asparagine, Alanine, Tyrosine, Tryptophan) and five amino acids in the oil phase (Arginine, Glycine, Threonine, Valine, Phenylalanine).

## Emulsifying activity and emulsion stability

The emulsifying activity of the supernatant is 42.85% and the stability of the emulsion is 40.85%. For the oil phase, the emulsifying activity is much lower than that of the supernatant because it represents 21.24%. On the other hand, the emulsion obtained with this fraction is largely stable (62.24%) compared to that of the supernatant.

## Discussions

Oily phase

The viscera correspond to all the internal organs of sturgeons (liver, pancreas, intestines, gills...). Based on the results obtained, these sturgeon co-products are a good source of nutrients. Despite its high water content ( $65.60\pm026\%$ ) which often complicates the preservation of the products, the very high protein ( $62.77\pm0.63\%$ ) and fat ( $35.59\pm0.01\%$ ) contents deserves their valorisation. These values are similar to those of sardine viscera (DUMAY, 2006), and also to those of tuna viscera (NGUYEN, 2009).

The biochemical characterization of dried autolysis fractions has the advantage of using the autolysis technique to solubilize proteins in the supernatant and to release lipids in the oil phase in addition to the release of mineral elements in these fractions through the activation of several endogenous enzymes in the viscera.

The dried supernatant contains twice as much protein  $(93.98\pm0.04\%)$  as the soluble phase of the shrimp heads found by RANDRIAMAHATODY and his collaborators in 2011. The lipid content  $(64.12\pm0.88\%)$  of the oil phase is twice as high as the insoluble phase obtained with the enzymatic hydrolysis of shrimp heads. The ash content obtained is low compared to that of the salmon heads found by GBOGOURI and its teams in 2005. However, mineral elements, in particular iron, magnesium and calcium, are present in significant quantities.

Most of the amino acids identified are essential amino acids, including histidine, which is very essential for the growth of children. Indeed, the constituent proteins of sturgeon viscera are proteins of good nutritional quality. Concerning the functional properties of these proteins, they have a very interesting emulsifying activity (emulsifying agent) in the food industry (chocolate, biscuits and pastries). Its ability to stabilize the emulsion is not negligible. However, its properties are low compared to the hydrolysates of salmon heads, which were 87 to 88% (GBOGOURI, 2005). The next perspectives, will focus on the exploitation of these functional properties in order to contribute to the food diversification of Malagasy consumers.

#### Conclusion

This studyallowed us to show that the sturgeon (viscera) co-products studied deserve to be promoted in different areas, particularly in the food and nutrition sector. Compared to other recovery techniques such as enzymatic hydrolysis, chemical hydrolysis which is expensive and leads to products that could contain chemicals, autolysis is the best recovery technique for the sturgeon co-products. It solubilizes the proteins in the supernatant and recovers the lipids in the oily phase. The amino acids identified are generally essential amino acids, hence the proteins of sturgeon viscera and their derivatives are of good quality. The emulsifying capacity of autolysates is interesting in the agri-food industry sector

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