Design of a procedure for obtaining a protein concentrate prepared from Tuna Cooking Water

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Abstract: Analytical ultrafiltration using three TFF cartridges differing on the cut-off (10, 30 and 100 kDa) were used to desalination and recovery of the collagenous fraction from the tuna cooking water.

Best results were obtained with the 30 kDa membrane. The obtained concentrate had collagenous nature and provided functionality to be capable of gelling at temperatures close to the room temperature.

***Keywords*:** Tuna cooking water; Ultrafiltration; Collagenous fraction

1. Introduction

The main structural factors that determine the profitability of the tuna-canning sector is the low performance of the production process, which reaches losses than 50%. The losses are particularly high during the cutting, cooking and peeling stages.

In order to avoid this problem, it is necessary to optimize the manufacturing processes through the development of new presentations and products that will help to increase the overall performance of the transformation process.

One possibility to improve performance in canning has been the recycling of the wastes obtained before packing, particularly the protein recovery from the steam cooking effluents.

The tuna cooking water are brines resulting from the cooking process and therefore contains pieces of meat fish, sarcoplasmic proteins, and a small proportion of solubilized myofibrillar proteins. In greater proportion, it also contains gelatin resulting from the fusion of collagen during cooking. As a result, this residue presents a high organic load and a strong contaminat impact.

In the present study, desalination and recovery of the collagenous fraction from the tuna cooking water, mainly by filtering technologies (ultrafiltration), was valued.

2. Materials and Methods

2.1 Materials

Jealsa Rianxeira S.A. (A Coruña, Spain) kindly donated the tuna cooking water. The whole tuna fish was cooked and tuna cooking water was recycled during two or three days. Than liquid effluents obtained were filtered using glass wool to eliminate solid waste and frozen at -30ºC.

Table 1 shows the initial composition of tuna cooking water.

Table 1: Initial composition of the tuna cooking water obtained after 2 or 3 days cooking

	2 days	3 days
Salinity (g/L)	8,6	19,00
Protein (g/L)	23,20	51,60
Total Phosphorus (mg/L)	41,13	80,00

The water with three days cooking showed significant higher protein concentration and therefore it was selected for the following studies.

2.2 Analytical methods

Proteins were determined by the method of Lowry et al. [1], total nitrogen was measured by the method of Havilah et al. [2] and total phosphorus by Murphy et al. [3]

The salinity was measured by conductivity using a portable conductivity 524. The instrument measures the specific conductivity of the sample and then is converted to values of salinity.

2.3 Ultrafiltration of the cooking tuna water

Analytical ultrafiltration was carried out using three TFF cartridges differing on the cut-off (10, 30 and 100 kDa), using spiral polyethersulfone membranes (Millipore Prepscale) of 0, 56 m^2 and an assembly with full recirculation [4]. Process included two phases: ultrafiltration and diafiltration.

2.4 Determination of the melting point and hardness of the gels obtained from tuna cooking water

Gels were prepared from solutions of gelatin fractions adjusted to 6, 67% (w/w) of concentration [5]. Solutions were incubated at 45ºC during 30 min followed by cooling down to 8°C. The obtained gels were stable for 16-18h at that temperature.

3. Results and Discussion

3.1 Size exclusion of membranes

Ultrafiltration phase was performed with total recirculation and an initial volume was concentrated at approximately 1L. In particular, when using the 10 kDa membrane, 5L was concentrated to 970 ml, with the 30 kDa, 10L to 890 mL and in the case of the 100 kDa membranes, 5L was concentrated to 650 mL.

The stage of diafiltration was also performed with total recirculation, but adding distilled water to the retentate, at a flow rate equivalent to the permeate flow in order to maintain constant volume.

Experimental data (Figure 1, 2 and 3) for the balance of the solute, represented as percentages of initial concentrations (protein, total nitrogen and total phosphorus), were consistent with the model described by Murado et al. [4] following a first order kinetics. The adjustments were satisfactory in all cases.

$$C = C_f + C_0 \times exp^{-(1-s)\times D_r} \quad [a]$$

Where:

C: concentration of the permeable solute in the concentrate, with C0 as initial value. C_f is the final asymptotic value if only a part of a polydisperse solute is permeable. Thus, when we use normalized values (%): $C_0 + C_f = 100$, with $C_f = 0$ if all solute is permeable.

s: specific retention of the solute. It varies between 0 (the solute is filtered as the solvent) and 1 (the solute is totally retained).

D_r: relative diavolume: volume of added water / constant concentrates volume.

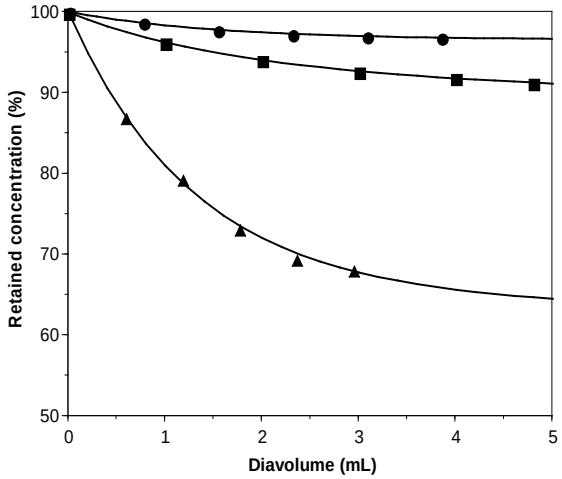

Figure 1: Graphical representation of the diafiltration phase. Protein concentration with 10 kDa membrane (●) 30 kDa membrane (■) and 100 kDa membrane (▲), in the concentrate, depending on the diavolumen (mL). Continuous lines represent experimental data adjustments to the model [1].

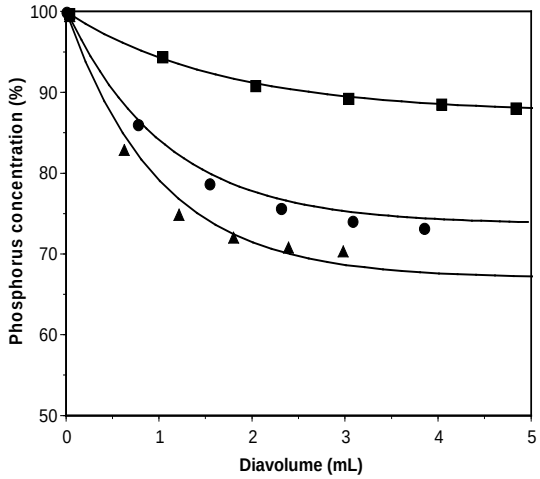

Figure 2: Graphical representation of the diafiltration phase. Phosphorus concentration with the membrane of 10 kDa (●) 30 kDa membrane (■) and 100 kDa membrane (▲) in the concentrate dependingon the diavolume (mL). Continuous lines represent experimental data adjustments to the model [1].

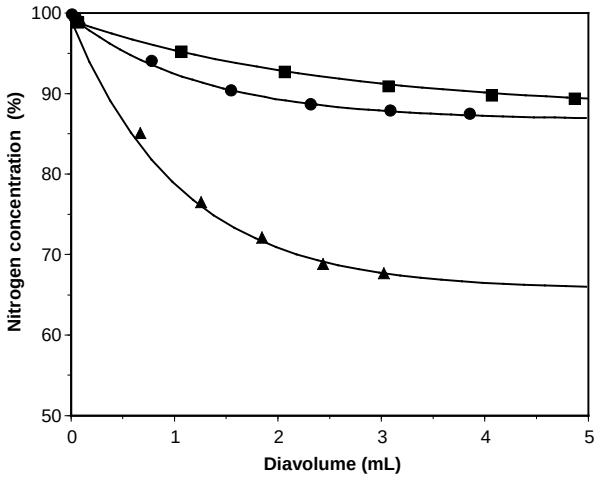

Figure 3: Graphical representation of the diafiltration phase. Niteogen concentration with the membrane of 10 kDa (●) 30 kDa membrane (■) and 100 kDa membrane (▲) in the concentrate depending on the diavolume (mL). Continuous lines represent experimental data adjustments to the model [1].

In the case of protein concentration (fig.1), with a 10 kDa membrane, the values of the coefficients were C_f = 96, 51 % and *s*=0,332, that means a practically full retention, as well as a specific retention that would demand a relative diavolume of 3 (i.e., a relationship between the volume permeate and the initial volume) would be enough, in theory, to finish the process at asymptotic. An *s*= 0,332 it means that the 3, 40% of solutes is practically filtered as the solvent.

With a 30 kDa membrane it is possible to retained a 92, 43 % and a 7, 57 % is filtered with a coefficient s=0,441, finding more limitations when compared with the 10 kDa membrane. Finally the 100 kDa membrane, the coefficient values are C_f= 65, 22% and s=0, 228, losing a 34, 78 % of protein.

In the case of 30 and 100 kDa membranes, asymptotic level is not reached with the used diavolumen; however, given the goodness of fit to the model [a], percentages are acceptable.

In the case of total phosphorus (Fig 2.), highest retained were achieved with the 30 kDa membrane, with values close to 88%. In the 10 kDa membrane, 72, 5 % of phosphorus it is completely retained and 27, 5 % goes out in permeate with few restrictions on the transfer as seen in the value of *s*= 0,064, very close to zero. Finally, in the 100 kDa membrane, the *s* is null, what means that 32, 7% is lost with any restriction on transfer.

In the case of nitrogen, the membrane of 30 kDa also retains a higher percentage (Cf=88, 73%) when compared with the 10 and 100 kDa membrane. What is lost, an 11, 26% have some difficulty in their transfer across the membrane, with an s= 0,601 very close to 1.

3.2 Final composition of Tuna cooking water

After the analytical ultrafiltration the final composition of the tuna cooking water with three days cooking is different depending the size of the membrane (10, 30 and 100 kDa).

Table 2: Final composition of tuna cooking water

	10 kDa		**30 kDa**		**100 kDa**	
	Inicial	**Final**	**Inicial**	**Final**	**Inicial**	**Final**
Salinity (g/L)	19,0	0,35	19,0	0,53	19,0	0,10
Protein (g/L)	50,31	291,28	51,02	134,07	54,63	17,88
Total Phosphorus (mg/L)	37,0	7,00	86,0	13,0	73,0	5,00
Volume (L)	5,00	0,97	10,0	0,89	5,0	0,65
Concentration factor		5,16		11,2		7,70

3.3 Melting point and hardness strength gel obtained from tuna cooking water

The melting point and hardness of the gel were determined based on an aqueous solution of 6, 67 % gelatin.

Table 3: Values of the melting point and hardness

Membranes UF (kDa)	**Melting point (ºC)**	**Hardness (Bloom nominal)**
100	**20,2**	**22**
30	**19,6**	**91**
10	**18,8**	**40**

Pig: Type A. Sigma G-2500. Bloom nominal ~ 300
Bovine: Type B. Sigma G-6650. Bloom nominal ~ 75

Hardness of the gel obtained with the faction coming from the 10 kDa membrane was expected to be higher than the gels prepared from the 30 and 100 kDa, as only 3,5% of protein was lost with the first membrane. A possible explanation might be that the retentate obtained with the 10 kda membrane contains a relative higher content of medium size polipeptides (MWs between 10 and 30 kDa) whose chain length is not enough to stabilize the gel structure, thus giving rise to the formation of weak gels. These peptides fractions would be eliminated with the 30 kDa membrane.

4. Conclusion

We have evaluated membrane technologies for desalting and recovery of concentrated protein fractions differing on molecular weight from tuna cooking water. Concentrates obtained from ultrafiltration using a 30 kDa membrane has a collagenous nature and form gels at room temperature. The hardness of the gel was higher than that of a commercial bovine gelatin.

The developed method to obtain a hydratable polymer capable of gelling opens up the possibility to improve the performance of the tuna canned manufacture. Thus, this collagen fraction, alone or in combination with other agents (salt, pieces of muscle), is being used as a brine injection to reincorporate constituents of the cooking tuna effluents in the process prior to tuna packaging.

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