USE OF SERUM PROTEINOGRAM TO DETECT INFLAMMATION IN GILTHEAD SEABREAM (*Sparus aurata*).

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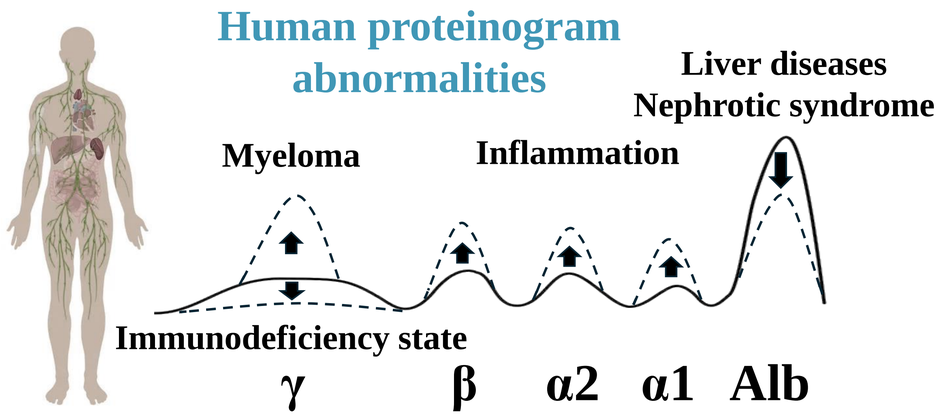
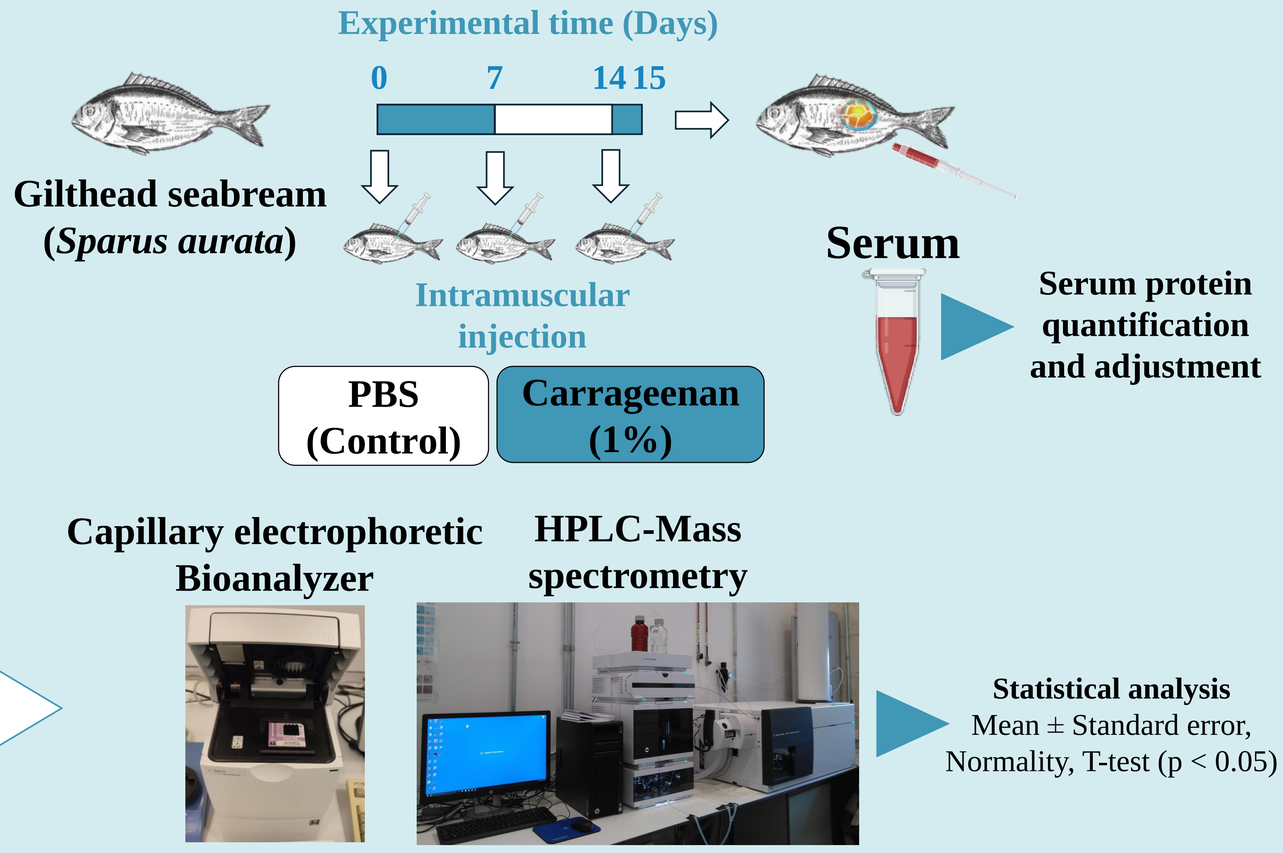
INTRODUCTION

The proteinogram is a semiquantitative analytical method that separates proteins into distinct bands using electrophoresis. This technique has been widely used in clinic to identify abnormal or elevated proteins in serum or other fluids, indicating possible liver, inflammatory, and immune diseases. However, this method has not been studied in fish for diagnostic use.

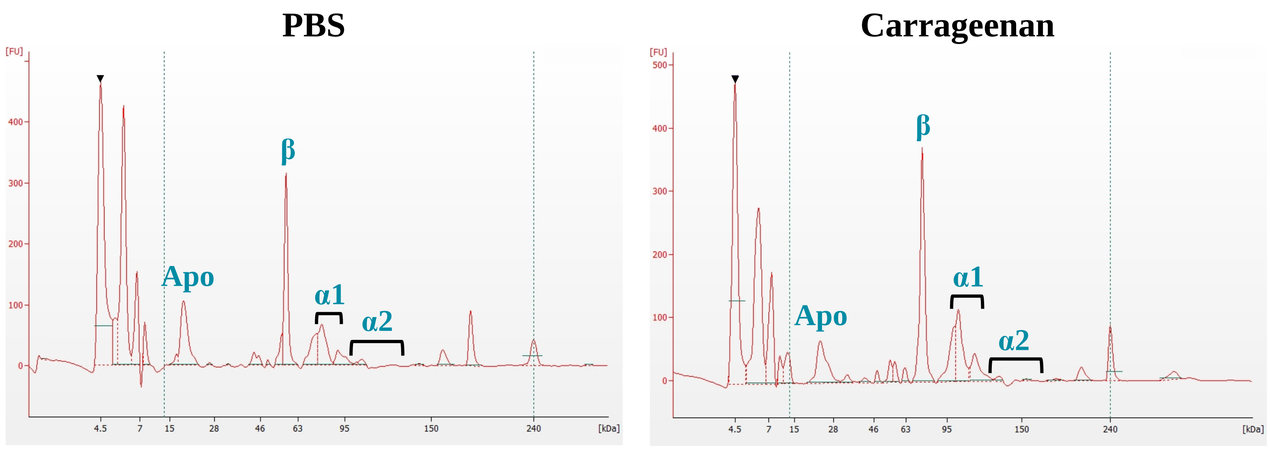


METHODOLOGY



RESULTS AND DUSCUSSION

Electropherograms



Protein identification

Electropherogram (Protein 230)				HPLC-Mass spectrometry			
		Concentration (% of Total)					
Peak number	Protein size [kDa]	PBS	Carrageenan	Protein size [kDa]	Accession number	Protein	Fraction
1	**19.5**	**24.2**	**20.4**	**20.0**	**A0A671WM58**	**Apolipoprotein A-II**	**Apo**
2	34.1	1.3	1.4	35.0	A0A671TQA7	Haptoglobin-like	
3	43.2	0.8	0.7	42.0	A0A671WGZ9	Protein AMBP	
4	48.3	4.9	4.1	48.5	A0A671UBA2	Hemopexin	
5	55.1	2.1	2.7	55.2	A0A671VGU4	Alpha-2-antiplasmin-like	
6	62.8	0.8	1.3	64.9	A0A671USN8	Ig-like domain-containing protein	
7	**76.3**	**31.6**	**29.3**	**74.3**	**F2YLA1**	**Serotransferrin**	**β**
8	88.5	0.5	1.9	86.9	A0A671TJ84	Complement C2	
9	92.8	10.2	5.5	89.3	A0A671V3B2	Plasminogen	
10	**104.4**	**13.2**	**17.2**	**102.2**	**A0A671YDN4**	**Inter-alpha-trypsin inhibitor heavy chain H3-like**	**α1**
11	**156.7**	**7.4**	**9.9**	**160.7**	**A0A671U9S6**	**Alpha-2-macroglobulin-like**	**α2**
13	186.9	1.7	1.2	186.5	A0A671TKG8	Complement C5	
14	212.0	4.8	1.3	193.0	A0A671TD78	Complement component c3b. tandem duplicate 2	

Figure 1. Serum Electropherogram profile of gilthead seabream post injection with phosphate buffer (control, PBS) or carrageenan (1%) by protein320 kit. Peak migration times were compared to an external size standard (ladder; dark arrowhead) to determine size, while peak areas were compared to a lower and an upper marker to determine concentration. The tables provides an overview of each protein peak found by electropherogram with its size (kDa) and relative concentration (%) with respect to total protein. These protein peaks correspond to the size of a similar protein determined by high-performance liquid chromatography-mass spectrometry. The proteins with the highest concentrations are shown in bold type.

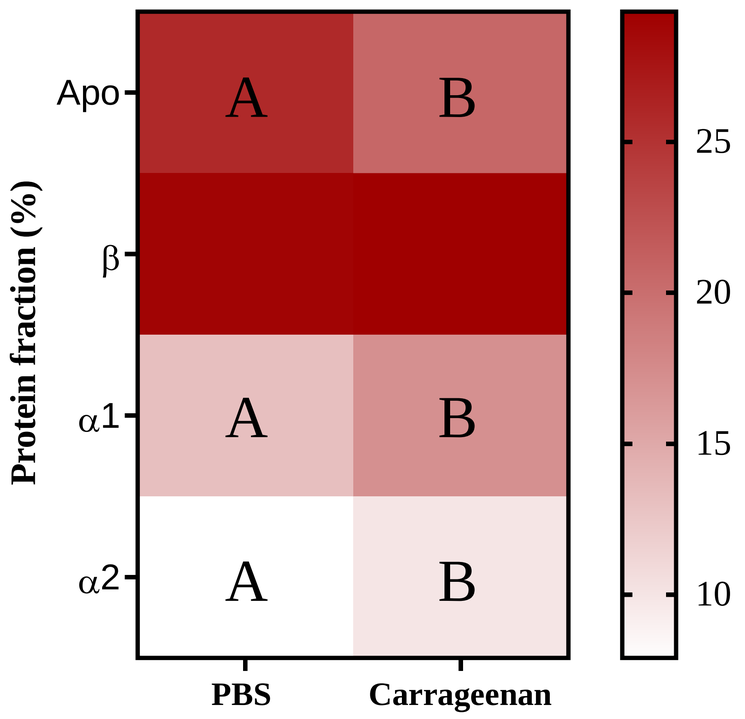
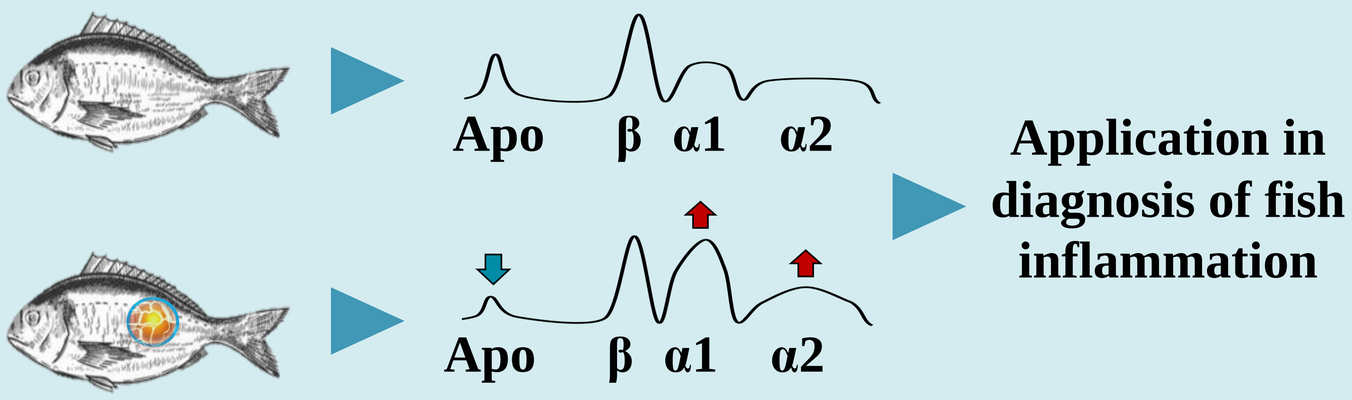


Figure 2. Heat map of serum protein fractions (%) of gilthead seabream post injection with phosphate buffer (control, PBS) or carrageenan (1%). Dark red squares denote increases, while white squares denote decreases in concentration. Different letters denote significant differences between the control and carrageenin groups (T-test; $p < 0.05$).

CONCLUSIONS



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