

Corn gluten high level inclusion in farmed trout nutrition: productive and hematological effects and quality of product

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Summary. *Background and aim of the work:* The substitution of fish meal in fish feeds has been thoroughly studied subject in aquaculture nutrition. Among cereals, corn gluten is one of the most used vegetal protein in fish feeds. This study investigated the effect of high level corn gluten inclusion in rainbow trout nutrition and related effects on productive and quality of product. *Methods:* Three isoproteic (crude protein: 41%) and isoenergetic (19 MJ kg⁻¹ of dry matter) diets were tested with graded level of corn gluten inclusion (40, 50 and 60%). Four hundred eighty rainbow trout (body weight : 50.7 ± 2.0 g) were utilised. At the end of experiment, productive traits, haematological analyses and quality analysis were carried out. *Results:* Considering the productive traits, statistically significant differences were found between fish fed with control and corn gluten diets. In all the measured parameters a similar pattern was visible showing main difference between highest level of corn gluten inclusion GLU50 and fish meal fed fish, CTRL (Specific Growth Rate, GLU50: 1.11 ± 0.1; GLU40: 1.14 ± 0.02^{ab}, CTRL, 1.36 ± 0.1^b; Food Conversion Rate, GLU50: 2.02 ± 0.1^{ab}; GLU40: 1.82 ± 0.1^{ab}, CTRL: 1.5 ± 0.1^b). *Conclusions:* Productive and haematological data indicated that the maximum corn gluten inclusion was reached with 40%. Quality of product analysis indicated that the fillets of fish fed with an inclusion of 50% of corn gluten showed similar aroma features of control fish fillet.

Key words: aquaculture, fish nutrition, rainbow trout, fish meal substitutes

Introduction

The substitution of fish meal in fish feeds and its effect on quality of final product have been thoroughly studied subjects in aquaculture (1-3). Among cereals, corn gluten meals (CG) are protein concentrates with low concentration of many of the anti-nutrients typically present in plant-derived ingredients. CG is one of the most commonly used vegetal source of protein in fish feeds (4, 5). Until this moment, the majority of researches in aquaculture investigated low – medium

levels of CG inclusion, ranging from 25 to 45% (1, 6, 7). However, considering that there is an urgent need to realize large substitution of fish meals in order to increase the aquaculture sustainability and its social acceptability (8-11), researchers often investigate high levels inclusion of new feedstuffs. It is clear that these large inclusions are desirable and possibly beneficial from the productive point of view, some aspects must be taken in account as potentially detrimental, in particular organoleptic quality of final products (12, 13). On the light of these considerations, the aim of this

work was to investigate the effect of the replacement of fish meal by high level of corn gluten in rainbow trout (*Oncorhynchus mykiss*) diet and the evaluation of final quality product.

Materials and Methods

Diets

Three different diets were formulated with graded level of corn gluten (GLU40, GLU50 and GLU60), and these diets were tested against a control fish meal based diet (CTRL). Diets analyzed by proximate composition according to standard methods (14) showed that all diets were isoproteic (Crude Protein: 41%) and isoenergetic (19 MJ kg⁻¹ of dry matter). Diets formulations and proximate composition are reported in table 1. All the dry ingredients and the oil were thoroughly mixed; water was then blended into the mixture to attain a consistency appropriate for pelleting using a 3.5 mm die meat grinder. After pelleting, the diets were dried at 50 °C and refrigerated at 6 °C until utilization.

Growth trial and "in vivo" digestibility trial

The growth trial was conducted at the Experimental Station of Agriculture Department of Turin University. 480 rainbow trout with a mean initial body weight of 50.7 ± 2.0 g were randomly allotted to 16 tanks (n = 30), with a water flow rate of 6 l/min. Trout were acclimated to experimental tanks for two weeks before to start the growth trial. Growth trial started on January 4th 2007 and ended in April 24th 2007. The feeding trial lasted for 110 days. The fish were fed twice a day by hand (feeding ratio 1.7% of body weight) for five days per week. Fish were bulk-weighted fortnightly in order to adjust the feeding rate. Water temperature and dissolved oxygen were measured 2 times per week and they were in the physiological range for rainbow trout along all the feeding trial. At the same time of the growth trial, an "in vivo" digestibility experiment was performed in order to determine the apparent digestibility coefficient (ADC) of diets. 64 rainbow trout from the batch of growth trial (initial body weight 168.0 ± 18.9 g) were randomly allotted to 16 cylindrical tanks. Four tanks were randomly allotted to each diet. After 15 days of acclimatization, fishes were fed by hand to satiety twice a day with the experi-

mental diets. The "in vivo" digestibility experiment was carried out in April and May 2007. The ADC of dry matter of each diet was determined using the indirect method. 1% of celite® (Fluka, Buchs, Switzerland) was included as inert marker in fish feeds and the feed formulation was consequently corrected. Faeces of each tank were collected using a continuous automatic collector 24 h after feeding over a period of six days per week. Faeces were collected every morning and frozen daily (-20 °C).

Sampling and chemical analysis

Five fish per tank were sampled in order to determine the somatic indexes.

The gut and liver were separated from the rest of the body and weighed. The dorsal muscle tissues from the same fish body were sampled, frozen and freeze-dried until the successive chemical determinations. All the fish diets and fillets were analyzed to determine the proximate composition according to standard methods (14). The gross energy content was determined using an adiabatic calorimetric bomb (IKA C7000, Staufen, Germany). The total nitrogen content was determined using a nitrogen analyzer (Rapid N III, Elementar Analysen systeme GmbH, Hanau, Germany) according to the Dumas method and the crude protein was calculated as total N×6.25.

Haematology

Blood samples were collected from all the randomly selected fish (n = 7), within 20 min (range 10-20 min) from capture and after anaesthesia in 3-aminobenzoic acid ethyl ester solution (100 mg/l) by caudal vein puncture. Fresh blood was placed in heparinized tubes for haematology determination. The rest of the blood was left to clot at 4 °C for 2 h, the clot was removed after centrifugation, and the serum was aliquoted and deep-frozen (-80 °C) for biochemistry determination. Total white blood cells were counted by direct observation in a Bürker chamber, after diluting a sample of fresh blood (1:20) with Shaw's double staining fluid (15). Red blood cells count (RBC) was manually counted after dilution, x 200, in modified Dacie's fluid using a counting chamber. Values were expressed as 10⁶ µL. Haematocrit (Hct) values were determined in duplicate by using a microhaematocrit

capillary tubes, a microhaematocrit centrifuge (2500 g for 5 min) (Tomos Biotoools Co., Changsha, China) and a microhaematocrit reader. The values were expressed as the percentage of erythrocytes. Haemoglobin (Hb) concentration (100 g/mL) was estimated spectrophotometrically (540 nm wavelength) (U-5100 Ratio Beam Spectrophotometer, Hitachi, Tokyo, Japan) using the cyanomethaemoglobin method with Drabkin's reagent. Haematochemical analysis were performed using commercially available kits for veterinary use (Biochemical Systems International s.r.l., Arezzo, Italy) with an automated biochemical analyser (VETSCREEN, Biochemical Systems International S.r.l., Arezzo, ITALY) for the following variables: glucose (Glu) and urea.

Flesh quality analysis

Forty samples of fish fillet were divided into 8 groups (5 samples of fish flesh per treatment) for final quality analysis. The internal sample temperature was measured before baking (0 min). Cooking was performed at 165°C to reach 70 °C internal temperature in an electric forced-air convection oven. By forcing air into the oven, after passing through an active charcoal filter, regular cooking odour out flow was guaranteed. PEN 2 (AIRSENSE Analytics GmbH, Hagenower, Germany) is a portable electronic nose (EN) with 10 metal oxide sensors (MOS) that change their resistance in the presence of oxidising and reducing gaseous compounds (16). The ten PEN2 sensors analyse 10 classes of chemicals: 2 sensors for aromatic (W1C and W3C), broad range (W5S), hydrogen (W6S), aromatic-aliphatic (W5C), broad-methane (W1S), sulphur-organic (W1W), broad-alcohol (W2S), sulphur-chloride (W2W) and methane-aliphatic (W3S). This instrument was utilized in the non-stop monitoring of volatile compounds on raw and cooked fish flesh. The temperature probe was put into the sample at the start of cooking and the internal temperature was checked while the sample reached 70°C. For statistical elaboration the average of the final 30 sec of electronic nose detection was considered. In order to correctly measure the aromas, the electronic nose samples were analysed following an increasing corn gluten inclusion, as follow: CTRL, GLU40, GLU50 and finally, GLU60.

Statistical analysis

Same experimental plan was adopted for growth trial and for "in vivo" digestibility trial. The experimental design was balanced, monofactorial with randomized blocks, four levels of treatment and four replicates (4x4). Fish diet was the experimental factor tested. All data were analysed by one-way ANOVA, except fillet composition data that were elaborated with non parametric ANOVA (Kruskall – Wallis test), in consideration of low number of replicates in these experimental parameters. After the ANOVA, differences among means were determined by the Tukey test multiple comparisons of means, using the significant level of $P < 0.05$ (17). For haematological data, Pearson multiple correlation matrix was calculated. For electronic nose statistical analyses, 40 data sets (4 experimental groups, i.e. diets; 5 replicates, i.e. fish; 2 levels of cooking, i.e. raw and cooked) containing 14 columns (parameters) and 30 observations (the last 30 seconds of aroma detection) each, were initially considered. The data were successively summarized and analyzed as two set of data for raw and cooked fishes, with 24 rows (8 replicates per 3 treatments) and 10 columns corresponding to the sensors used for electronic nose. PCA multivariate analysis was performed for statistical elaboration of electronic nose data.

Results

In the growth trial the mortality was less than 5% in all the experimental groups, except for GLU60 group where the final mortality was 41%. The water temperature was $13.5 \pm 0.1^\circ\text{C}$, and dissolved oxygen was $7.0 \pm 0.2 \text{ mg/l}$.

Productive results

Fish diets showed high digestibility and there was a progressive increase in digestibility of the diet, proportional with the corn gluten inclusion (Tab. 1). Productive traits (Tab. 2) resulted statistically different between fish fed with fish meal based diet and CG diets.

In all the measured parameters at the end of the growth trial similar pattern was visible: the highest level of corn gluten inclusion was the most different group from fish meal based diet.

A clear difference was also detected between higher corn gluten inclusion level groups (GLU60 and GLU50) and CTRL group, while productive parameters of GLU40 group were similar to those of control group. Somatic indexes showed similar pattern of productive indexes and the increase of volume proportional with dietary gluten inclusion was evident in liver and intestine (Tab. 2). Proximate analyses of fish fillet were strongly influenced by diet and there was a relevant fat deposition on GLU60 fillets and also a decrease of protein content was observed (Tab. 3).

Haematological results

The experimental treatment did not strongly affect the haematological parameters (Tab. 4). Only the glucose level showed a statistically significant increase in the fish fed with higher gluten inclusion level. This difference was more evident if the fish meal based diet group and the corn gluten groups were grouped in only two separate groups. In general, a progressive reduction of white blood cell (WBC) was evident, proportional with dietary corn gluten inclusion. A correlation ($r = 0.64$) was observed between WBC and Hb, while other parameters were not correlated.

Table 1. Fish feed composition and diet digestibility

Ingredients (%)	GLU60	GLU50	GLU40	CTRL
Herring fish meal	6	13	20	54
Corn gluten	60	50	40	3,5
Fish oil	6	6	6	5,5
Corn oil	1	1	1	1
Soybean oil	1	1	1	1
Barely meal	7.5	8.5	9.5	7.5
Corn meal	7	9	10	0
Corn starch	9	9	10	18
bentonite	0	0	0	7
Lygnumsulphite	0.5	0.5	0.5	0,5
Mineral mixture ¹	0.5	0.5	0.5	0.5
Vitamine mixture ²	1	1	1	1
Liver-protector integrator ³	0.5	0.5	0.5	0.5
Proximate composition (% on DM) (n=4)				
Crude protein	42.9 ± 4.1	41.9 ± 2.4	39.6 ± 4.1	41.1 ± 1.2
Ether extract	11.4 ± 0.3	11.5 ± 1.4	11.7 ± 0.4	12.1 ± 1.3
Ash	2.8 ± 0.3	3.7 ± 0.4	4.9 ± 0.2	14.8 ± 0.1
Gross energy (MJ kg ⁻¹ DM)	20.7 ± 0	20.7 ± 0.3	20.3 ± 0	18.2 ± 0.1
Digestibility				
ADCDM ⁴	96 ± 0.3 b	95 ± 0.9 b	94 ± 0.2 b	87 ± 0.2 a

In the rows, different letters mean statistical difference at $P \leq 0.05$.

¹ Mineral mixture (mg/kg diet): bicalcium phosphate 500 g, calcium carbonate 215 g, sodium salt 40 g, potassium chloride 90 g, magnesium chloride 124 g, magnesium carbonate 124 g, iron sulphate 20 g, zinc sulphate 4 g, copper sulphate 3 g, potassium iodide 4 mg, cobalt sulphate 20 mg, manganese sulphate 3 g, sodium fluoride 1g, (Granda Zootechnica, Cuneo, Italy).

² Vitamin mixture (IU or mg/kg diet): DL- α tocopherol acetate, 60 IU; sodium menadione bisulphate, 5 mg; retinyl acetate, 15000 IU; DL-cholecalciferol, 3000 IU; thiamin, 15 mg; riboflavin, 30 mg; pyridoxine, 15 mg; B12, 0.05 mg; nicotinic acid, 175 mg; folic acid, 500 mg; inositol, 1000 mg; biotin, 2.5 mg; calcium pantothenate, 50 mg; choline chloride, 2000 mg (Granda Zootechnica, Cuneo, Italy).

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⁴ ADCDM apparent digestibility coefficient of diets dry matter (DM) express as percentage

Table 2. Productive traits (n=4; mean \pm sd) and somatic indexes (n=5; mean \pm sd)

Diets	CTRL	GLU40	GLU50	GLU60
SGR1	1.36 \pm 0.1b	1.14 \pm 0.0ab	1.11 \pm 0.1ab	0.96 \pm 0.1a
FCR2	1.5 \pm 0.1b	1.82 \pm 0.1ab	2.02 \pm 0.1ab	2.3 \pm 0.2a
PER3	1.49 \pm 0.1b	1.23 \pm 0.1ab	1.1 \pm 0.1ab	0.97 \pm 0.1a
NPU4	0.6 \pm 0.0b	0.49 \pm 0.0ab	0.44 \pm 0.0ab	0.39 \pm 0.0a
BG5	117.74 \pm 10.3b	89.27 \pm 7.5ab	76.63 \pm 5.4 a	60.25 \pm 7.5a
HSI6	1.19 \pm 0.2b	1.58 \pm 0.2ab	1.56 \pm 0.3ab	1.89 \pm 0.2a
VSI7	9.05 \pm 1.3	10.22 \pm 3.4	10.78 \pm 1.5	11.41 \pm 1.3

In the columns, different letters mean statistical difference at $P \leq 0.05$.

¹Specific Growth Rate (%) = $(\ln \text{ final weight} - \ln \text{ initial weight}) \times 100 / \text{feeding days}$; ²Feed Conversion Rate = $\text{total feed supplied (g of DM)} / \text{BG (g)}$; ³Protein Efficiency Ratio = $\text{WG (g)} / \text{total protein fed (g)}$; ⁴Net Protein Utilization = $\text{utilised protein (g kg}^{-1}) / \text{protein gain (g kg}^{-1})$; ⁵Biomass gain (g) = $\text{final total weight} - \text{initial total weight}$; ⁶Hepato Somatic Index = $(\text{liver/body weight}) \times 100$; ⁷Viscero Somatic Index = $(\text{viscera/body weight}) \times 100$

Table 3. Fish fillet proximate analysis (n = 4; mean \pm sd)

Diets	Dry matter (%)	Crude protein (% of DM)	Crude fat (% of DM)	Ash (% of DM)	Gross energy (Mj kg ⁻¹ DM)
GLU60	73.2 \pm 1.3	63.3 \pm 8.1a	23.7 \pm 42.0a	4.4 \pm 1.4a	26.0 \pm 0.2a
GLU50	75.3 \pm 1.3	72.6 \pm 4.2ab	19.3 \pm 12.4a	5.1 \pm 2.3b	24.9 \pm 0.1a
GLU40	74.4 \pm 2.2	77.8 \pm 13.3ab	14.9 \pm 16.2ab	5.3 \pm 0.4b	24.5 \pm 0.1ab
CTRL	76.7 \pm 2.9	86.6 \pm 7.2b	7.1 \pm 1.1b	5.7 \pm 0.9b	22.9 \pm 0.2b

In the columns, different letters mean statistical difference at $p \leq 0.05$.

Table 4. Haematology and serum biochemistry (n=7)

	RBC (106m/l)	WBC (103m/l)	Hct (%)	Hb (gd/l)	Glu (mmol/l)	Urea (mmol/l)
GLU60	2.18 \pm 0.7	12.93 \pm 6.5	48.57 \pm 3.9	8.39 \pm 0.6	14.44 \pm 8.0a	0.19 \pm 0.06
GLU50	2.41 \pm 0.4	15.86 \pm 7.1	47.57 \pm 7.0	8.63 \pm 1.8	9.52 \pm 3.8ab	0.26 \pm 0.16
GLU40	2.39 \pm 1.4	16 \pm 15.7	51.57 \pm 5.9	9.61 \pm 3.2	7.07 \pm 4.7b	0.36 \pm 0.5
CTRL	1.83 \pm 0.4	22.21 \pm 17.4	47.14 \pm 5.9	8.92 \pm 1.9s	6.41 \pm 4.1b	0.27 \pm 0.14

In the columns, different letters mean statistical difference at $P \leq 0.05$.

Quality results

In this study PCA has been used to investigate the experimental diets on flesh aroma in raw and cooked

fish. In this study first three PCA axes were considered, in order to facilitate figures interpretation. Figure 1 and Figure 2 represent respectively the samples of

flesh aroma of raw and cooked final samples. It has been observed that the samples CTRL and GLU 40 were more different from other groups.

PCA1 axis explains the greater part of the variance for raw (explained variance 67.9%) and for cooked fish (explained variance 61.6%) and the principal electronic noise sensors related with this axis were aromatic compound sensors (Tab. 5). Some of these sensors are correlated with PCA2 in cooked fish. For raw and cooked fish PCA3 is influenced only by W6S sensor, hydrogen sensor. It is clear that the use of the sole electronic noise in this experiment represents a preliminary result in the context of quality of final product, as these analyses would need an external analysis to be fully interpreted, however it is clear that between the final fish fillets of different experimental groups there are detectable differences in raw and cooked fish.

Discussion

Productive traits

The growth and other productive parameters studied in this research showed that CG inclusion in rainbow trout diet could reach 40% without any adverse effects. Between alternative foodstuffs used in fish nutrition studies CG is one of the most used, considering its large availability and balanced amino acid composition. CG inclusion has been successfully used in several species of fish as gilthead sea bream (*Sparus aurata*) (6) or European sea bass (*Dicentrarchus labrax*) (1) and the inclusion level in these researches was comprised between 14 and 57 % in fish diet. CG inclusion has been also tested in rainbow trout diets: Mente et al. (2003) (7), Palti et al. (2006) (5), and Satoh et al. (2003) (18) used near to 40% of inclusion, 33.42 % CG inclusion, and 25% CG inclusion respectively. In the present study, the CG dietary protein

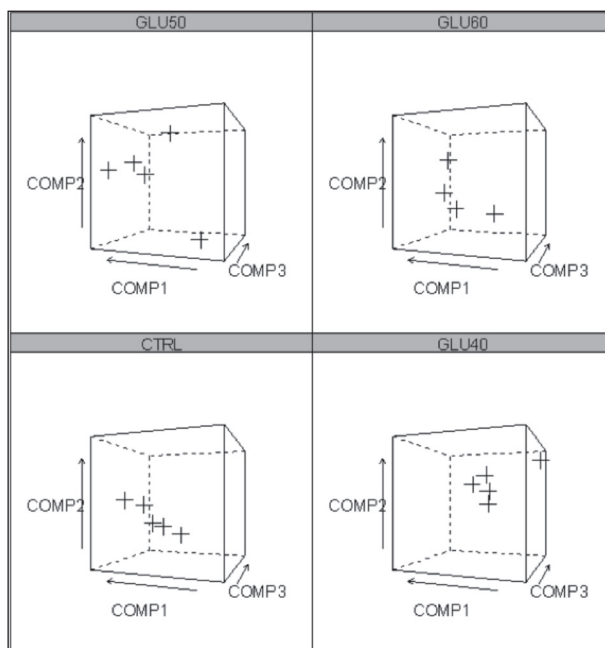


Figure 1. Aroma analysis on raw fish (first three PCA axes), 4 experimental groups: CTRL: control diet; GLU40 40% corn gluten inclusion; GLU50 50% corn gluten inclusion; GLU60: 60% corn gluten inclusion. COMP1 = first PCA axis; COMP2 = second PCA axis; COMP3 = third PCA axis

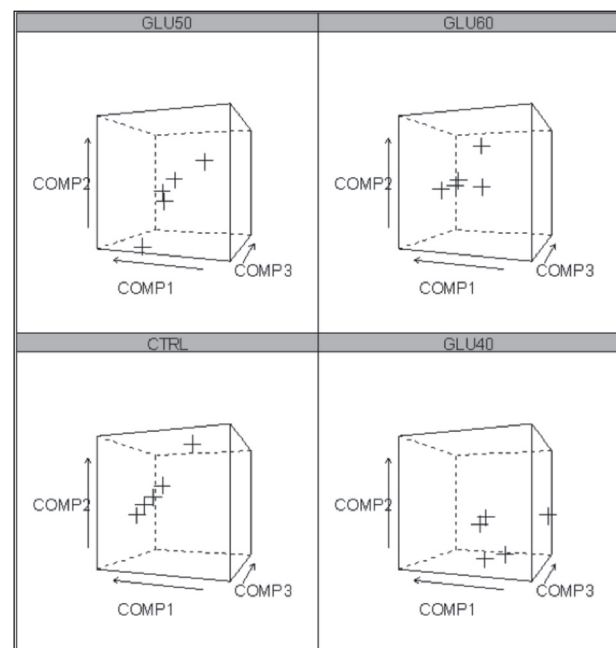


Figure 2. Aroma analysis on cooked fish (first three PCA axes), 4 experimental groups: CTRL: control diet; GLU40: 40% corn gluten inclusion; GLU50: 50% corn gluten inclusion; GLU60: 60% corn gluten inclusion. COMP1 = first PCA axis; COMP2 = second PCA axis; COMP3 = third PCA axis

Table 5. PCA scores on first 3 PCA axes: raw and cooked fish.

Sensor	RAW FISH			COOKED FISH		
	PCA1 (67.9%) ¹	PCA2 (16.1%)	PCA3 (9.4%)	PCA1 (61.6%)	PCA2 (25.0%)	PCA3 (9.5%)
W1C	0.36	-	- 0.27	0.28	-0.44	-
W5S	-0.36	0.19	-	- 0.33	- 0.26	0.33
W3C	0.36	- 0.11	- 0.26	0.29	-0.42	-
W6S	- 0.22	-	-0.71	- 0.11	- 0.26	-0.88
W5C	0.36	- 0.21	- 0.20	0.35	- 0.29	-
W1S	-0.36	0.27	-	-0.36	- 0.25	0.17
W1W	- 0.13	-0.54	0.45	- 0.34	0.27	- 0.17
W2S	-0.37	0.13	- 0.22	-0.35	- 0.28	0.18
W2W	- 0.26	-0.53	- 0.12	-0.37	-	- 0.11
W3S	- 0.27	- 0.48	- 0.19	- 0.29	-0.43	-

In bold the highest values for each PCA component.

¹ explained variance

PCA1 = principal component analysis first axis; PCA2 = principal component analysis second axis; PCA3 = principal component analysis third axis; W1C = electronic nose first sensor for aromatic compounds; W3C = electronic nose second sensor for aromatic compounds; W5S = electronic nose sensor for broad range compounds; W6S = electronic nose sensor for hydrogen; W5C = electronic nose sensor for aromatic-aliphatic compounds; W1S = electronic nose sensor for broad-methane compounds; W1W = electronic nose sensor for sulphur-organic compounds; W2S = electronic nose sensor for broad-alcohol compounds; W2W = electronic nose sensor for sulphur-chloride compounds; W3S = electronic nose sensor for methane-aliphatic compounds

was respectively 62% in the GLU40 feed, 75% in the GLU50 feed 75% and 88% in the GLU60 feed. All the productive parameters showed similar pattern already observed in Atlantic salmon (19): the highest level of CG inclusion (GLU60) was the most different group from fish meal based diet (CTRL). CG digestibility for rainbow trout was higher than turbot and yellow-tail and comparable with CG digestibility for carp, thus suggesting that this feedstuff was more digestible for freshwater rather than marine fish. Moreover fish diet digestibility decreased proportionally with increase in gluten inclusion, as expected and observed in other studies (4, 19, 6). Somatic indexes showed similar pattern to productive indexes and the increase of volume was particularly evident in liver size, as reported in similar studies on Atlantic salmon (4, 19). Dietary CG inclusion negatively affected fillet proximate composition, primarily on high fat deposition resulted in fillets of CG fed fish. This result confirmed that 40% CG inclusion was the highest inclusion level in rainbow trout diets. Similar results were reported in Atlantic salmon CG fed (19, 4) and gilthead seabream (6). The 60% of CG inclusion in trout feed is clearly

not recommendable following our findings, however the recorded mortality for GLU60 group was probably not induced by experimental diets, in fact in a previous similar work on Siberian sturgeon (*Acipenser baeri*), no mortality appeared at the 60% of CG meal dietary inclusion (20).

Haematology

The haematological parameters investigated in this research are often studied as indicator of general state of health in fish (21, 22). Corn gluten inclusion exerted a positive effect on erythropoiesis. Haematocrit values were in the physiological range for Salmonid (22), while red cells were higher, probably indicating nutritional stress conditions. The measured values for haemoglobin and erythrocytes were slightly higher than reference (22-24), even if there was no difference between experimental treatments (23). Rehulka and Minarik (2003) showed that the increase in erythrocyte count was dependent on the fish meal dietary inclusion level. In our case the erythropoiesis stimulation was related to corn gluten inclusion, but haematocrit and haemoglobin was unaffected. As

regard as carbohydrate metabolism parameters are considered glucose is one of the most investigated parameters, (25-27) and this experimentation showed a high glucose blood level caused by CG inclusion that confirmed the negative effect registered on productive traits. In fish, as well as in other vertebrates, the immune system is activated in environmental change (24) and it is generally manifested as variation in number of WBC. White cell count was higher than in similar research ($9-16 \times 10^3$ cells /mm³) (28) in CTRL group, while a small decrease was also observed in the corn gluten groups. This situation can indicate a dietary protein deficiency. Immune system could be affected by essential amino acid scarcity that are used for main physiological functions rather than for white cell synthesis.

Aroma analysis

Quantitative studies on aroma are common in research on food quality and the large number of data utilised needs accurate statistical method in order to extract useful information from multivariate data (12, 29). Multivariate statistics as PCA is often used in quality study in aquaculture (26, 30-32). PCA1 axes in this study synthetically represents a linear combination of considered variables, the electronic nose sensors. In this study the experimental groups were well separated from aroma detection and this fact was visible both in raw than cooked fish. PCA1 represented vegetable aroma and the effect of cooking determines a kind of ordination, following a corn gluten gradient, that is opposite as expected considering GLU60 fish fillet is more similar to control fish than GLU40 fish, which showed an aroma more different from others group. The effect of cooking was particularly evident between GLU40 and GLU50 aroma. Electronic nose analyses were relevant on cooked fish, considering that fish are normally consumed in cooked form. From these results it was possible to see that fish diet affected fish fillet aroma, but the most interesting result was that high levels of corn gluten inclusion did not affect fish aroma more than low levels (GLU40), in fact the highest level of corn gluten inclusion, GLU60, showed similar aroma with fish meal fed fish.

In conclusion, productive and haematological data indicated that in this experimentation the maximum corn gluten inclusion was reached with 40% and the corn gluten inclusion could not be used in rainbow trout without compromising productive performances to a major extent. The negative effect on productive traits was partially counterbalanced by quality of product, in fact aroma analysis shows that the 60% corn gluten inclusion level had similar aroma of control fish fillet, although opposite was expected.

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