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Inclusion of Hermetia illucens larvae meal on rainbow trout (Oncorhynchus mykiss) feed: effect on sensory profile according to static and dynamic evaluations

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4	RUNNING TITLE Sensory profile of rainbow trout fed insect meal
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25 ABSTRACT

26 BACKGROUND: Diet implementation with insect meal arouses increased attention in aquaculture considering the advantages of this new protein source. The 27 effect of Hermetia illucens meal (HI) inclusion in diets on rainbow trout physical-28 chemical and sensory properties was evaluated. Three diets were prepared: HI0, HI25, 29 HI50, with 0, 25 and 50% of HI replacing fish meal, respectively. Fillet sensory profiles 30 were described by descriptive analysis (DA) and Temporal Dominance of Sensation 31 (TDS) methods. Cooking Loss, WB-Shear Force, proximate analysis, fatty acid 32 composition were also determined. 33

34 RESULTS: Diets significantly affected fillets sensory profile. DA indicated significant changes in perceived intensity of aroma, flavour and texture descriptors as a 35 function of diet composition. TDS evaluations provided information on dominance and 36 37 evolution of sensations perceived in fillets from different diets. The first sensations perceived as dominant were related to texture attributes, followed by flavours. 38 39 Dominance of fibrousnesses decreased with the increasing of HI in diet. Boiled fish, algae flavours and umami taste clearly dominated the HIO dynamic profile. The onset of 40 metallic flavour dominance characterized HI25 and HI50. No differences in physical 41 42 parameters were detected. Principal component analysis highlighted the relationship between sensory attributes and physico-chemical parameters. 43

44 CONCLUSION: Sensory description of fillets indicated that HI inclusion45 induces significant differences in the perceived profile.

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47 Key words

Hermetia illucens, insect feeding, rainbow trout, Descriptive Analysis, Temporal
Dominance of Sensation.

50

51 **INTRODUCTION**

The rising demand and consumption for aquaculture feeds have generated a 52 rapid decline in the availability of fish meal (FM) and a concurrent price increase.¹ FM 53 is the optimal animal protein source used in commercial fish feeds.² However, the use 54 of FM is both environmentally and economically unsustainable.¹ Alternative protein 55 sources have been investigated to replace FM in livestock feeds, especially in 56 aquaculture. Nowadays, insects are being considered as a novel protein source both for 57 humans and livestock.³⁻⁵ Insects grow and reproduce easily, have high feed conversion 58 59 efficiency, can be grown on low quality organic waste, do not compete with humans and other farmed animals for nutrients and are particularly suitable for many freshwater 60 and marine fish feeding as a part of their natural diet.⁶ Moreover, they are a rich source 61 of protein, lipids, minerals and vitamins.⁷ Different insect species have been considered 62 for their possible use in fish feeds and some studies have been carried out.⁸⁻¹⁰ Among 63 the different species, Hermetia illucens seems to be very interesting as sustainable 64 alternative to replace FM in aquaculture feeds.⁸ H. illucens, is suitable to be reared on 65 organic wastes by converting them into protein-rich and lipid-rich biomass, therefore it 66 can be used for various purposes including animal feeding, biodiesel and chitin 67 production.⁶ 68

69 Changes in fish diet affect fish flesh sensory characteristics^{11,12} such as 70 texture^{13,14} and volatile compounds.^{15,16} Previous studies showed that the replacement of 71 FM with insect protein in aqua feeds determines changes of chemical composition of 72 fish muscle, especially for lipid content and fatty acid profile.^{17,18} In several studies 73 relationships were found between fish flavour, muscle chemical composition¹⁹ and fatty 74 acid profiles.²⁰⁻²² Considering the growing interest of *H. illucens* as alternative protein source to replace FM in the fish feeds, it can be highlighted that until now the studies on
the related effects on sensory properties of fish meat are scarce.^{17,18,23}

Modifying feeding practice without taking into account possible changes in fish sensory properties appears a risky option²⁴, since modifications on fish flavour and aroma can affect the perceived quality²⁵ and the acceptability by consumers.¹⁶

Descriptive Analysis (DA) and Temporal Dominance of Sensations (TDS) have been found as methods providing complementary information to describe food sensory properties.²⁶ DA permits identification, quantification, and description of food sensory attributes.²⁷ It is useful when a detailed description of the sensory properties is desired and provides a picture of the perceived differences among products in terms of intensity of sensory attributes.

TDS tracks the evolution of "dominant" sensations (the ones catching the attention) during the product evaluation.²⁸ The dynamic of perception has important consequences for a better understanding of the processes used by consumers to assess acceptability and sensory properties of food products.²⁹

In this work, DA and TDS were utilised to investigate the effect of replacing part of diet proteins with *H. illucens* on sensory properties of rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792). The physical-chemical characteristics and sensory properties of rainbow trout fed regular diet with FM as exclusive source of protein or fed diet including *H. illucens* in partial replacement of FM were described and compared. Moreover, the relationships between physico-chemical characteristics and sensory profile of fillet samples were investigated.

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98 MATERIALS AND METHODS

99 Sample characteristics

100 <u>Diet formulation</u>

101 Three isoproteic and isolipidic diets were formulated (Table 1), in which 102 *Hermetia illucens* prepupae meal (HI; Hermetia Deutschland GmbH & Co. KG 103 (HDKG), Baruth/Mark, Germany) substituted 0% (HI0), 25% (HI25) and 50% (HI50) 104 of FM

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106 <u>Fish feeding and sampling</u>

107 The experimental trial was performed at the experimental facility of the 108 Department of Agricultural, Forest, and Food Sciences (Italy). A total of 360 rainbow 109 trout (*Oncorhynchus mykiss*), with an average initial weight of 178.9 ± 9.81 g, was 110 individually weighed and randomly distributed in twelve fiberglass tanks (1 m³; T: 111 $13\pm1^{\circ}$ C; dissolved oxygen: 7.6 - 8.7 mg L⁻¹; water flow: 8 L min⁻¹), in an indoor 112 openwater system (30 fish per tank). Experimental diets were randomly assigned to the 113 tanks.

Fish were fed by hand twice a day at 1.5% of body weight, and fish wereweighted in bulk every 15 days to adjust the feeding rate.

At the end of the trial (92 days), fish were individually weighed and 30 fish for each diet were slaughtered, accurately packaged inside polystyrene boxes with ice covering, and brought to the Department of Agri-Food Production and Environmental Sciences (Florence, Italy). After the arrival, all the fish were weighed, dissected and filleted; the fillets were vacuum packaged, and frozen at -80°C until analyses.

121

122 <u>Sample selection and preparation for sensory evaluations</u>

Fish were ranked within each diet according to their weight. Individuals with the highest weights within the diets and with comparable weights across diets were selected for sensory evaluations (Table 2). In total, 15 individuals were selected from each diet,

four were utilised for Descriptive Analysis (DA) and three were utilised for Temporal
Dominance of Sensation (TDS) analysis. The remaining individuals were used to set up
the evaluation conditions and for sensory panel training purpose.

Before evaluations, fillets were thawed at 4°C for 24 h, washed and accurately dried with paper, and skinned. The part close to tail was discharged and the ventral fish bones removed. Samples were prepared by cutting the cleaned fillets in several portions of 4 ± 0.2 g each, and around 22 portions from each individual were obtained. Each portion was wrapped in an aluminium foil, and stored at 6-8°C until the evaluation session started.

Wrapped samples were steam cooked for approximately 1.30 min, until reaching
a temperature of 62°C at the heart and immediately presented to subjects for evaluation.

137

138 <u>General sensory evaluation conditions</u>

Samples were presented monadically and identified by a three-digit code. The 139 140 presentation order was randomized between subjects and sessions. The order of 141 attributes was balanced between subjects to minimize a possible "proximity" effect and was always the same for a given assessor. After each sample, subjects rinsed their 142 mouth with water for 30 s, had some plain crackers for 30 s and finally rinsed their 143 144 mouth with water for a further 30 s. Subjects took a fifteen min break after every 145 session. Data were collected with the software Fizz (ver. 2.47.B, Biosystemes, 146 Couternon, France).

147

148 Subjects of the sensory evaluation

Ten subjects, 8 males and 2 females, aged from 20 to 30 years, regular fish consumers, were recruited. The subjects were informed that the aim of the evaluation was the description of sensory properties of fish fed diets containing also proteins from

152 insects. Before starting with the experiment, a written informed consent was obtained

153 from each subject. The subjects had no history of disorders in oral perception.

154

155 Sensory evaluation by Descriptive Analysis

156 Training sessions: sensory vocabulary development and subject training

Panellists participated in three training sessions of about 60 min each. The 157 subjects developed a vocabulary describing differences and similarities between 158 159 experimental samples in two different sessions, according to a simplified version of the repertory grid method.³⁰ The initial list of attributes was reduced to achieve a list that 160 161 comprehensively and accurately described the product space: redundant and/or lesscited terms were grouped on a semantic basis and/or eliminated according to the 162 subjects' consensual decisions. A main list of attributes was developed (Table 3) which 163 164 described the texture, taste and flavour of fish samples. Some standards were prepared, 165 as reported in Table 4, to induce a weak/moderate intensity. A nine point category scale 166 labelled at the extremes with "extremely weak" (corresponding to 1) and "extremely strong" (corresponding to 9) was utilised for evaluation. Two repetitions of the whole 167 set of samples were performed in individual booths. Assessor and panel performance 168 were validated by evaluating two sets of samples used for the study. Panel and assessors 169 170 data were analysed using Panel Check software (ver. 1.4.0, Nofima, Trømso, Norway).

171

172 <u>Evaluation</u>

The evaluation of fish meat from each diet was replicated four times in four sessions. In each session, each panellist evaluated three individuals, one from each diet. Two samples from each individual were tested. The first sample was utilised for aroma (ortho-nasal odour) and texture assessment, while the second sample was utilised for taste and flavour evaluation. The overall aroma and flavour descriptors were alwayspresented as the last attribute of the relevant list.

179

180 Sensory evaluation by Temporal Dominance of Sensations

Subjects participated in three training sessions. In the first session, the concepts 181 182 of dominance and temporal evolution of sensations were explained to the subjects. 183 Then, the most relevant attributes for describing the temporal evolution of sensory 184 properties were selected from the attribute lists used for DA. Nine attributes were selected: Melt in mouth, Tenderness, Juiciness, Fibrousness, Metallic, Boiled fish and 185 186 Algae flavours, Umami, and Astringency. Two sessions were performed for training subjects with the use of the computer system for TDS data acquisition. Panellists were 187 trained to click on the "Start" button as soon as the sample was in the mouth and to 188 189 immediately start the evaluation. Performance of panellists and eventual artefacts were 190 evaluated by visual inspection of individual out-put of training session evaluations.

Panellists participated in six evaluation sessions. Two sessions per day were performed and three individuals, one from each diet, were evaluated twice in the same day, in two independent sessions. In total, three individuals per each diet were evaluated. The total evaluation time was 90 sec.

195 Sample presentation and evaluation conditions were the same described for DA196 evaluations.

197

198 Physical analyses

A number of 4 fish for each diet was randomly weighed and slaughtered. Analyses on physical and chemical properties were performed on the cooked fillets of each sample. The cooking loss (CL) was calculated by measuring the difference in weight of the fillet before the cooking process and after, according to the formula:

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203

205	Texture analyses were carried out using a Zwick Roell [®] 109 texturometer (Ulm,
206	Germany) with the Text Expert II software, equipped with a 1 kN load cell. The
207	Warner-Bratzler shear force test (WB-SF) was performed on the cranial part of the fillet
208	epaxial region (two measurement for each fillet). A straight blade (width of 7 cm),
209	perpendicular to muscle fibre direction, was utilised at a crosshead speed of 30 mm/min
210	to 50% of total deformation. Maximum shear force, defined as maximum resistance of
211	the sample to shearing ³¹ was determined from the plot of force (N) compared with
212	deformation (%) and expressed as mean.

213

214 Chemical analyses

Proximate composition of HI meal, experimental diets and cooked freeze-dried fillets from the three groups of fish differently fed was determined according to AOAC procedures.³² Dry matter, ash, crude protein and ether extract were determined according to 950.46, 920.153, 976.05, and 991.36 methods, respectively.

The fatty acid (FA) group composition was analysed on total lipid extract³³ of HI 219 220 meal, experimental diets samples and cooked muscle samples obtained from fish fed 221 different diets. The FAs composition was determined by gas chromatography (Varian 222 GC 430; Agilent, Palo Alto, CA, USA) equipped with a flame ionization detector and a Supelco Omegawax[™] 320 capillary column (30 m × 0.32 mm i.d., 0.25 µm film and 223 polyethylene glycol bonded phase; Supelco, Bellefonte, PA, USA). FAs were identified 224 225 by comparing the FAME retention time with the standard Supelco 37 component FAME mix (Supelco). Individual FAs were quantified using tricosanoic acid (C23:0) 226 (Supelco) as internal standard. FAs were expressed as a percentage of total FAME. 227

229 Statistical analysis

Intensity data from the trained panel were analysed by multi-block PCA (Tucker-1) and by *P**MSE plot (Panel Check software, ver. 1.4.0, Nofima, Norway) to assess panel calibration and assessor performance, respectively.³⁴ Based on the *P**MSE plots and Tucker-1 plot, 2 out of 10 subjects were considered unreliable and were taken out from further data analysis.

Principal Component Analysis (PCA) was carried using the mean data for each repetition, in order to geometrically represent the variability associated to diets and individuals by using The UnscramblerX 10.3 software (Norway). Samples were included as dummy variables (downweighted in the data matrix) to improve the visual interpretation.³⁵ The full cross validation was computed to validate the interpretation of the first two components.

Every sensory attribute was analysed following a two-way factorial design in which the diets and panellist were treated as a fixed effect and as a random variable, respectively.

244 The mean dominance curves for each treatments and six repetitions were computed from raw software coding (1 selected; 0 not selected). The data of TDS were 245 analysed by the software Fizz (ver. 2.47.B, Biosystèmes, Couternon, France). When the 246 247 TDS curves were plotted, two additional lines were drawn for the chance and significance 248 levels. The chance level refers to the dominance rate that an attribute could obtain by chance. Its value is inversely proportional to the number of attributes ($P_0=1/p$, where p is 249 250 the number of attributes). The significance level (Ps) is the minimum value this proportion should be equal if it is to be considered significantly (p<0.05) higher than P_0 . 251 Rosner³⁶ recommended that np (1–p)>5 (where n=number of trials and p=probability of 252 success). In the present study, 10 panellists performed six replications of each product and 253 nine attributes were utilised, thus the number of observation was satisfied (np = 5.87). 254

Normality of data distributions was tested by the Kolmogorov-Smirnov test on cooking loss, WB-shear force, proximate analysis and fatty acid composition. One-way ANOVA was performed on physical-chemical results, considering the diets as main effect. The Bonferroni post-hoc test was applied to check the significance of the differences among diets, using SPSS version 17.0 software (SPSS Inc. Illinois, USA).

A PCA was calculated after standardization of variables in order to assess the relationship among the sensory, physical and chemical dataset, using The UnscramblerX 10.3 software (Norway).

263

264 **RESULTS**

265 Descriptive analysis

266 The analysis of principal components (PCA) was performed using all tested 267 individual as independent samples, in order to evaluate the differences due to both the 268 different diets and those relevant to biological variability among of individuals reared 269 following the same diet. The PCA correlation loading plot (Figure 1) showed that 270 samples were mainly discriminated along the first component (PC1: 42% explained variance) according to the diets. Fish fed with the control diet were positioned on the 271 left side of the map, while those with 50% of HI meal were located on the right. The 272 273 HI25 samples were located closed to the origin of the component. Along the second 274 component (18% of explained variance), sample position reflects the sensory variability due to the biological variability of individuals within the same diet. Considering the 275 276 distribution of samples in the perceptual space, it appears that the differences among samples due to different diets are more evident than those perceived between different 277 278 individuals fed the same diet. Thus, the individual evaluations belonging to the same 279 diet were treated as repetition.

The mixed ANOVA model on the intensity data of the sensory attributes were 280 281 performed, in order to estimate the sample effect (three levels: HI0, HI25, HI50) (Table 5). A significant sample effect of the diets was found for 12 out of 19 attributes 282 283 evaluated. No significant effects of assessor \times product interactions were found for the significant attributes (data not reported). Results reported in Table 5 showed that the 284 285 main differences were found between the control (HI0) and HI50 samples, while HI25 286 expressed some similarities with HI0 for some attributes and with HI50 for the others. 287 Considering the aroma-related attributes, the perceived intensities of boiled fish, algae and overall aroma were significantly higher in HI0 than HI50 samples (p <0.001). On 288 289 the other hand, the fresh fish aroma showed a significantly higher intensity in HI25 than HIO and HI50 (p <0.05). Metallic aroma was higher in samples from FM partially 290 replaced with insect proteins diets. Texture attributes resulted significantly more intense 291 292 in HI50 than HI0 and HI25 (p <0.05). Indeed, samples obtained by fish fed the 50% of 293 insect meal inclusion diet were juicier, more tender and melting more in mouth than HIO 294 samples. Overall aroma intensity tended to significantly decrease with the increasing of 295 insect protein inclusion.

Boiled fish flavour and sweet taste were perceived as more intense in HIO samples, with respect to individuals fed insect meal diets (p <0.05). The addition of *Hermetia illucens* prepupae meal also induced a significant increase in overall flavour intensity, independently from HI concentration. Moreover, metallic flavour intensity increased with the increase of HI meal content in the diets.

301

302 Temporal Dominance of Sensations

Mean TDS curves of the samples from the three diets are reported in Figures 2–4 for HI0, HI25 and HI50 groups, respectively. In general, the curves showed that the texture attributes dominated the first part of evaluation (0 to 15 seconds), followed by 306 flavour and taste attributes. In HIO samples (Figure 1) tenderness and fibrousnesses 307 clearly dominated the first part of evaluation. On the other hand, only tenderness clearly dominates the dynamic profile of HI25 (Figure 3) and HI50 samples (Figure 4). 308 309 Furthermore, it appears that the dominance of juiciness is mainly related to FM partial replacement with insect proteins. Flavour of HIO samples was dominated by boiled fish 310 and algae flavours even at a lower extent, umami taste was the sensations mostly 311 dominating the after taste. In samples HI25, boiled fish clearly dominated the profile 312 313 together with algae, metallic flavours and umami. The dynamic profile of HI50 appears complex with several descriptors perceived as dominant at the same time (boiled fish, 314 315 algae, metallic, umami). Even though umami resulted as the most important attribute in the aftertaste of all samples however some differences among diets have been observed. 316 317 Indeed, while in HIO the umami was the only dominant attribute, in HI25 the metallic 318 flavour was dominant and in HI50 the boiled fish flavour persisted until the end of 319 evaluation.

320

321 Physical and chemical characterization of fish fed different diets

Table 6 reports results of physical and chemical parameter analyses. No 322 significant effect of diets was observed for both cooking loss and WB shear force, 323 324 indicating that, from an instrumental standpoint, the samples lose the same amount of 325 water during cooking and were equally soft. Proximate composition of cooked samples 326 was not significantly affected by diets of fish (p>0.05). On the contrary, the sum of the 327 principal groups of the fatty acids showed differences associated with the experimental diets. It is of note that HI50 fish have the significantly highest level of saturated fatty 328 329 acids (SFA), followed by HI25. The HI0 had the significantly lowest content of SFA 330 and the significantly highest content of PUFA ω 3, showing an inverse relationship with

fish fed Hermetia illucens inclusion diets. MUFA and PUFA showed a similar trend,

332 significantly decreasing with the increase of HI meal concentration in diets.

333

Relationship between instrumental and sensory analyses

The correlation loadings plot in Figure 5 summarizes the main trend of sensory, 335 336 chemical and physical variables of the samples obtained from fish fed different diets, 337 highlighting the relationship between sensory and instrumental parameters. The explained variance after the first two components (PC) account of 53%. PC1 (37% of 338 339 explained variance) separated samples without Hermetia illucens inclusion in diet from 340 samples fed including HI prepupae meal. PC2 seemed to further separate samples that 341 have different content of HI. The predominant differences between the samples were 342 due to the fatty acids (FAs), mainly SFA that were negatively related to PUFAs. SFA resulted positively correlated to metallic aroma/flavour, overall flavour and tenderness 343 344 (negative part of PC1), as well as protein and ash content. The positive part of PC1 345 showed the relationship between PUFA, PUFA₀3, MUFA and boiled fish flavour and overall aroma. PUFA₆₆ seemed highly related to algae flavour, loaded on the positive 346 part of the second component. At the same time, juiciness and melt in mouth attributes 347 348 were strongly related to PUFA₆₆ and moisture content. WB-shear force did not play a relevant role in this PCA, as expected considering the lacking of significant differences 349 detected with analysis of variance. 350

351

352 **DISCUSSION**

353 Sensory evaluation

354 Terms freely generated by assessors to describe fish sensory properties are not 355 associated to negative hedonic valence, thus indicating that FM partial replacement with

356 insect meal did not induce the perception of sensory defects or off-flavours. According 357 to sensory results, differences among diets have been observed. DA showed significant differences in terms of aroma, flavour and texture. These results disagree with previous 358 findings reported in literature. For example, no sensory significant differences have 359 been found with inclusion of insect meal in diets on Atlantic salmon.¹⁸ Performing a 360 triangle test on rainbow trout fed diet with different content of insect meal, Sealey et 361 al.¹⁷ did not find any significant differences. In these studies, differences in FA 362 363 composition were detected, and the lacking of significant differences in sensory proprieties was quite unexpected since differences in FA composition affect the sensory 364 profile.^{20,21} Possibly these results reflect the lack of power of the adopted sensory 365 366 techniques. The results of the present work further confirm DA as a powerful sensory descriptive technique, providing the accurate description of sample sensory properties. 367

368 The dynamic analysis of sensory proprieties confirmed the differences between 369 the groups of fish fed different diets. TDS results partially confirmed the results 370 obtained by DA, and allowed a better understanding of the perception of sensory 371 proprieties during all the chewy process. Fibrousnesses intensity was not significantly different amongst trout samples but this sensation appears to be much more important in 372 HIO than in HI25 and HI50 samples. The inclusion of HI prepupae meal in diets led to 373 374 the perception of a more complex sensory profile with several flavour sensations 375 dominating the perception at the same time. Dominance of metallic flavour 376 characterized HI25 and HI50 samples in respect to HI0. This sensation can be seen as unfamiliar or as unexpected in fish thus catching the assessor attention despite its 377 moderate intensity value. Dominance values indicated that the importance of a sensation 378 379 during food consumption is not necessarily the same as that indicated by intensity ratings from static sensory profiles.³⁷ Thus, the use of the TDS method for the sensory 380

381 characterization of fish samples provides information which complements those from382 DA studies.

383

384 Physical and chemical characteristics and relationship with sensory profile

The partial replacement of FM with HI meal in diets for fish feeding, as 385 alternative source, seems to have effects on qualitative aspects of fillet, in terms of 386 sensory and physico-chemical characteristics.^{16,17,23} In the present study, the 387 388 instrumental physical differences concerning the parameters investigated were not identified. The *H. illucens* meal is a high-value feed source, rich in protein and fat. The 389 390 fat amount of black soldier fly larvae is extremely variable and depends on the feeding 391 substrate and development stage of the insect. Further, their FA composition depends on the FA composition of the diet utilised for larvae rearing.³ The lipid content of HI 392 393 affected the chemical composition of fish fillet, when the FM was partially replaced by 394 the insect meal. In this study, it seems that the inclusion of HI in diets implies a change 395 in FA profile of fish fillet, especially increasing the incidence of SFAs. On the other 396 hand, the PUFA incidences diminish when HI inclusion increases compared to control samples. This trend was also observed in previous studies on Atlantic salmon¹⁸ and 397 rainbow trout,¹⁷ where the amount of whole-body SFAs increased employing diets 398 399 containing increased amount of HI. Regarding proximate composition, in our trial no 400 significant differences in fillets from different groups of fish were noted. Contrariwise, 401 Sealey et al. observed that fillet moisture and lipid composition were significantly 402 altered by replacement of dietary FM with black soldier fly prepupae meal in rainbow trout.¹⁷ They reported that fish fed diets containing HI had significantly greater moisture 403 404 and lower lipid in muscle in comparison with fish fed the control diet. Even though these analyses were conducted on raw muscle, while in our study the analysis was 405 performed on cooked fillets, the results of this work are partly in line with these 406

407 previous findings, since a trend for a lower lipid content in fillet with increasing408 inclusion of insect meals, even if not significant, was also observed.

409

410 Relationship between sensory and instrumental analyses

The compositional differences of the diets, i.e. lipid content, FA profile and 411 proximate composition, can have affected the sensory properties. These variations in 412 diet modify, in particular, lipid content and composition of fish muscle. Overall aroma 413 and flavour intensity are both dependent on final product lipid content.¹² This study has 414 highlighted the relationship between sensory and physico-chemical parameters (Figure 415 5). FAs, flavour and texture attributes showed the main relationships. SFA increase in 416 the fish fillet with the increased inclusion of HI in the diets, and it seems to be 417 correlated to flavour and texture in fish flesh, in agreement with previous study.¹⁵ The 418 rise of fatty acids had an effect on tenderness of fish meat as confirmed also by 419 Grigorakis et al.¹¹ and Rincón et al.¹² findings. Valente et al.³⁸ found a significant 420 421 relationship between lipid content and both fatty flavour and perception of fatty texture. Additionally, lipid content and FA profile of fillets have a connection to flesh texture³⁹ 422 and they affect texture attributes, mainly juiciness and tenderness.¹¹ However, this 423 relationship with tenderness measured with Warner-Bratzler shear force was not 424 425 revealed in the case of this trial samples. Water contents contributed to juiciness and melting in mouth of the fish samples, in agreement with a previous work findings.¹² 426 Concerning the flavour modification, Grigorakis et al.¹¹ showed that fat content strongly 427 affects the mouth impression and volatile compounds that were also correlated to 428 differences in sensory taste. 429

430

431 CONCLUSION

Sensory description of fish samples indicated that HI inclusion induces 432 433 significant differences in the perceived profile. Furthermore, HI inclusion in the diet did not induce the perception of sensations relevant to defects or off-flavours. The effect of 434 435 diets was highlighted both by DA and TDS descriptions and the information obtained appear complementary. The strict relationship between sensory profile and fatty acid 436 437 composition was also confirmed by the results obtained. Further study will be necessary 438 to understand if the highlighted differences in sensory properties of fish fed diets 439 characterized by different protein sources would be reflected in liking judgements by 440 consumers.

441

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