

Consumers' perception and liking of breast fillets from broiler chickens fed diets including dehydrated microalgae (*Arthrospira* spp.) and Black soldier fly (*Hermetia illucens*)

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ABSTRACT

Recent studies investigate the possibility of including alternative protein sources in broiler chicken diets, e.g., insect and algae. Given the important relationship between feed and sensory quality of the meat, it is crucial to assess consumer perception and preferences. To this aim, five chicken breast samples – i) one control ii) two from chickens fed diets containing 9 % and 18 % of *Hermetia illucens* larvae meal and iii) two from chickens fed diets including 3 % and 6 % of dehydrated microalgae meal (*Arthrospira* spp.) in partial replacement for soybean meal - were tested by 84 subjects. Each sample was portioned, individually packaged under vacuum and sous-vide cooked. Subjects were asked to provide hedonic (9-point hedonic scale) and Check-All-That-Apply (CATA) responses and to indicate the perceived intensities of four selected attributes. Instrumental analyses were carried out by means of image and texture analysis. No statistically significant differences were registered regarding flavor and texture, while breasts from chickens fed with microalgae meal had the highest visual liking. Cochran's tests, conducted on CATA results, revealed that six attributes were significant ($p \leq 0.05$). By Penalty Analysis, pink color and juiciness were found to be drivers of liking, while grey, white, dry and pale had a negative impact.

1. Introduction

The world population is expected to grow from 7.7 billion people in 2019 to 9.7 billion by 2050, according to the United Nations (Christensen et al., 2018). This rise will result in a 60 to 70 % increase in global demand for animal products by 2050, with developing countries playing a significant role in driving this growth (Makkar, 2018).

Approximately 800 million tons of cereals per year are used in animal feeding. By 2050, this figure is expected to have surpassed 1.1 billion tons (Makkar, 2018). Every year, the poultry industry consumes approximately 600 million tons of dry matter feed with a relatively high concentration of feed protein raw materials (Mottet and Tempio, 2017). Soybean meal is the most widely used protein source in poultry nutrition, due to its high protein content, excellent amino acid profile, and convenience (Ravindran et al., 2014). However, soybean cultivation has significant environmental impact, including increased greenhouse gas emissions due to extensive land resource requirements, deforestation,

and ecosystem disruption (Dreoni et al., 2022). Moreover, the expected rise in soybean feed production worsens various linked environmental, economic, and social issues tied to the entire soybean production process, spanning from cultivation to processing and transportation (Song et al., 2021). As a result, suitable and cost-effective alternative protein sources for animal nutrition are required. Recently, there has been growing interest in the use of microalgae and insect meals. These alternatives have the potential to be a cost affordable and more environmentally sustainable protein sources for livestock (Madeira et al., 2017; Akhtar and Isman, 2018). One of the most studied insect species is the black soldier fly (*Hermetia illucens*, L., HI). Dabbou et al. (2018) identified HI meal as a valuable and alternative protein source for poultry feeds. In fact, it has a lower environmental impact and can convert organic by-products into high-value proteins, thus being an excellent example of a circular economy (Meneguz et al., 2018). The larval and pupal stages are the most nutrient-rich phases. They typically contain approximately 18–33 % fat and 32–53 % protein (Lu et al., 2022).

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Spirulina (*Arthrospira platensis*, SP) is a high-nutritional-value edible microalga with a protein content ranging between 55 and 70 % (Ramírez-Rodriguez et al., 2021). Including SP in broiler chickens' diet can affect the performance, oxidative stability, and meat quality parameters of broiler chickens (Altmann et al., 2020; Gkarane et al., 2020; Zampiga et al., 2023). In detail, SP inclusion in broiler diet was found to remarkably affect yellowness (b^*), which resulted to be higher if compared to the control. In addition, higher levels of total carotenoids and saturated fatty acids, but lower levels of n-3 polyunsaturated fatty acids and α -tocopherol were observed (Pestana et al., 2020).

Currently, the available literature offers a fairly broad perspective on consumers attitude concerning the use of alternative ingredients in feed production (Baldi et al., 2021; Sogari et al., 2022), novel products (Michel and Begho, 2023), and meat alternatives (Fidder and Graça, 2023). These studies mainly focus on the decision-making process underlying the willingness to accept and consume these products. In fact, the role of information about more sustainable production methods (Napolitano et al., 2010), and the environmental benefits of alternative feed in meat and fish production (Altmann et al., 2022) were investigated. However, the importance of sensory analysis for assessing the quality of meat products remains crucial. In general, the concept of quality embodies all the features and characteristics that can comply with a request or a need (Ruiz-Capillas et al., 2021). Röhr et al. (2005) pointed out that quality can be studied from two points of view: i) an objective understanding, connected with features that can be objectively assessed, and ii) a subjective side, which is related to consumer perception of the sensory parameters such as color, odor, flavor, etc. (Grunert et al., 2004).

In the case of chicken meat (and meat in general), sensory descriptive tests are the most commonly used for the analysis of product profile. Several studies sought the evaluation of liking by using hedonic scales (e.g., the 9-point hedonic scale; Samant and Seo, 2016), which is one of the most well-used methods in the liking determination of food products (Moskowitz and Sidel, 1971). Usually, a list containing the attributes of interest for the study is drawn up by and/or provided to the panelists (Altmann et al., 2020). The goal is to find the features that better describe the sample using methods such as CATA (Check-All-That-Apply) or RATA (Rate-All-That-Apply) (Tan et al., 2017; Xu et al., 2020), and by evaluating their perceived intensity (Baston et al., 2010; Cullere et al., 2019). Dyubele et al. (2010) used an 8-point descriptive scale to assess the intensity of the proposed attributes, while Liu et al. (2004) preferred a 0–15 ranged scale for their analysis. Moreover, Tasoniero et al. (2016) made their panelists rank on a 150-mm unipolar continuous-line scale the sensory attributes of chicken breast affected by White Striping and Wooden Breast myopathies. Another method for assessing perceived intensity is the Generalized Labelled Magnitude Scale (gLMS), which proved to be a valuable approach for measuring flavor intensities (Bartoshuk et al., 2004).

Furthermore, there are some works that use a mixed-methods approach for their sensory evaluation sessions. Xu et al. (2020) combined CATA and RATA descriptive methods with a 9-point hedonic scale to discriminate the flavor differences in five parts of Chinese blanched chicken. To assess the sensory properties and consumers' acceptance of chicken meat from dual-purpose crossbreeds fed with an alternative feed, Escobedo del Bosque et al. (2022) mixed the CATA method, hedonic response and evaluation of perceived intensity. This mixed-methods approach was also used with other food products. Piochi et al. (2021) decided to combine both CATA and liking assessment using a 9-point hedonic scale to determine the sensory properties of olive oil.

Based on the aforementioned points, the current research aims primarily to investigate how feed may impacts final product quality by exploring consumers' liking and perception. Considering the reported background, to reach this goal, we asked our respondents to i) provide a hedonic evaluation using a 9-point hedonic scale, ii) describe the products by using the CATA method, iii) and indicate the perceived intensity of four selected attributes on the Generalized Labelled Magnitude

Scale (gLMS), under blind information conditions. This approach permitted us to outline which product characteristic better meets the consumers' preferences. Secondly, image and texture analysis were performed to obtain objective data related to color and tenderness and validate the relationship between sensory and instrumental results.

2. Materials and methods

2.1. Ethical approval

This study has been approved by the Alma Mater Studiorum – Università di Bologna Bioethical Committee (Prot. N. 0,173,440 date July 28th, 2022).

2.2. Experimental design for samples provisioning

The study was conducted at the Department of Agricultural and Food Sciences, University of Bologna. A total of 5 chicken breast samples of about 500 g each were collected from broilers included in the trial carried out within a project titled “NextGenProteins” (Transformation of Biomass into Next Generation Proteins for Food and Feed), which has received funding from the European Union's Horizon 2020 Research and Innovation Programme, Call H2020-LC-SFS-17–2019, grant agreement no 862,704 (<https://nextgenproteins.eu/>). When reached 42 days old and an average live weight of 2.8 kg, birds were slaughtered in a commercial plant and, after deboning, 15 *Pectoralis major* muscles were randomly collected 48 h post-mortem and stored at -40°C .

The 5 breasts used were obtained from chickens receiving different experimental diets, and categorised as follows: 1 control (C) from chicken fed conventional feed, 2 samples from those receiving 9 % (HI9) and the 18 % (HI18) of Black soldier fly (*Hermetia illucens*) diets (BSFL), and 2 samples from others on 3 % (SA3) and the 6 % (SA6) of Spirulina algae (SA; *Arthrospira* spp.) meals. All chickens received the same corn-soybean based diet during the starter phase. Then C group, received a commercial diet with soybean as main protein source during grower and finisher phases. Group HI9 and HI18 as well as SA3 and SA6 groups were fed the same basal diet as C birds but with *Hermetia illucens* and Spirulina Algae meal respectively included in partial replacement of soybean. Diets were formulated to meet nutritional recommendations (Aviagen, 2019), with analogous metabolizable energy content and with a similar amino acid profile, which was optimized by maintaining the same ratio of total essential amino acids to total lysine. Feeds were in a mash form and, as well as water, provided for *ad-libitum* consumption.

The chicken breasts were used for both the sensory analysis, and the subsequent instrumental analysis.

All tasting sessions were conducted in October 2022 at the sensory laboratory of the Department of Agricultural and Food Sciences of the Alma Mater Studiorum - Università di Bologna.

2.3. Sample preparation and cooking procedure

The whole chicken breast fillets were left to thaw in the refrigerator at 4°C within 24 h before each session. Then, each fillet was cut to obtain into approx. 4 cm x 4 cm and approx. 2 cm high sub-samples, with an average weight of 14 gs per portion (Fig. 1). Each sub-sample was then cooked under vacuum (Fig. 1). The consumer would have opened the sample directly in the sensory booth in order to perceive the flavor at its best, and to avoid manipulation by the operator after cooking. Thus, samples were vacuum-packed, and the sample code was written on the envelope. A small cut was made in each bag to facilitate easy opening.

Following the method reported by Park et al. (2020), several cooking tests were carried out to define the combination of temperature and time required to reach a core temperature of 72°C . In particular, samples have been cooked sous-vide at 75°C for 1 hour, at 85°C for 1 hour and at 92°C (the maximum temperature reachable by the used instrument) for 1 hour. The temperature of 72°C at the inner core of the product

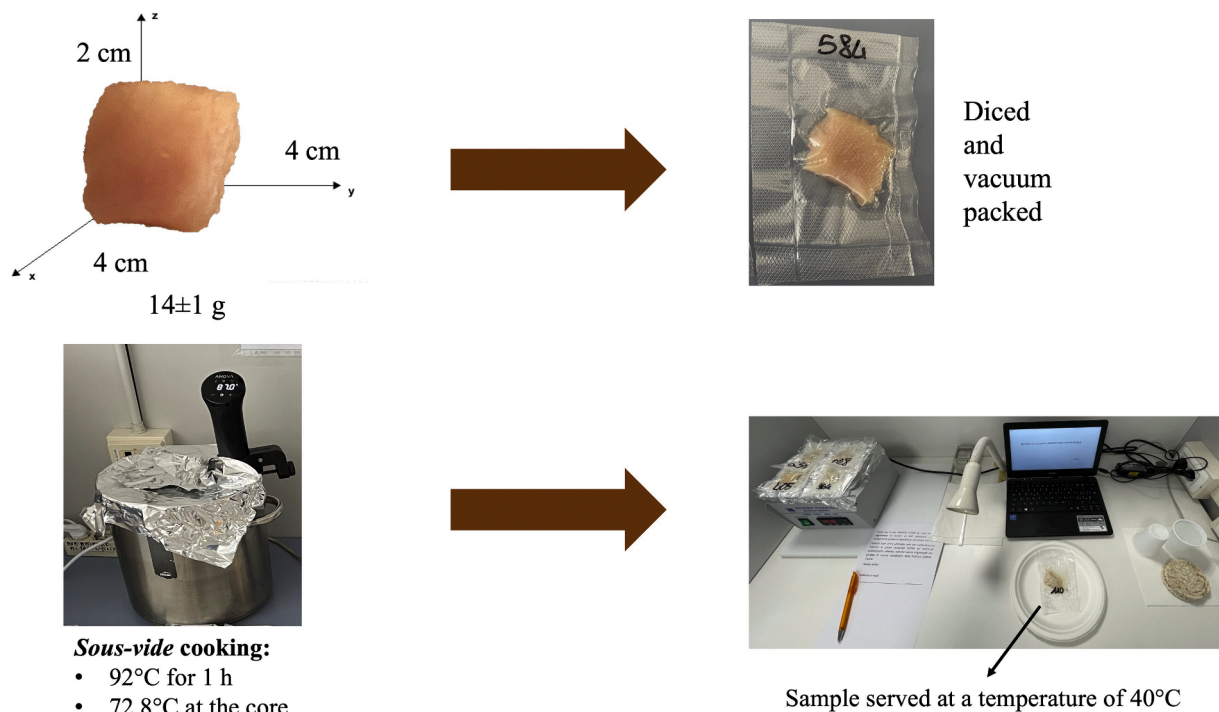


Fig. 1. Portioned chicken breast sample, vacuum packed for the sous-vide cooking, sous-vide cooking with Roner equipment and sensory booth ready for the test.

(measured with a food probe thermometer) was reached in the last case. Consequently, the chicken breasts to be tasted by participants were cooked at 92 °C for 1 hour (thus allowing to achieve a temperature of 72.8 °C at the core of the sample itself).

Roner equipment was used to perform the sous-vide cooking (Roner Anova Culinary Sous Vide Precision Cooker, 1000 Watts, Anova Applied Electronics, Inc., San Francisco, United States). All the samples were served at a temperature of 40 °C.

2.4. Participants

A total of 84 untrained individuals participated in the study. Participants were recruited via e-mail and with flyers at the Department of Agricultural and Food Sciences of Alma Mater Studiorum - Università di Bologna. The text of the email and on the flyer reported an email address through which it was possible to express the interest in participating by filling in the requested information (e-mail address, choice of date and time of the tasting session among those provided, declaration of absence of allergies and/or intolerances to crustaceans and/or soy, indication of age).

The indication reported that it consisted of a sensory test, which would involve the tasting of chicken breast, and the place where it would take place (the sensory laboratory of the Department of Agricultural and Food Sciences of Alma Mater Studiorum - University of Bologna, Viale Fanin 40, 4th floor).

Participants were recruited from the department's student and staff communities and actively participated in the research sessions.

Participants provided informed consent for participation, as well as the privacy information sheet.

2.5. Evaluation procedure

For each sample, participants immediately provided the hedonic responses, followed by sensory CATA responses and finally the indication of perceived intensities of four selected attributes. The order of samples was randomized (Latin square design) among subjects.

Hedonic responses were always obtained on a 9-points labelled

category scale (1= "dislike extremely"; 9= "like extremely"). For each sample, four hedonic questions have been presented: "Please, indicate how much you like this chicken breast for color.", "Please, indicate how much you like this chicken breast for flavor.", "Please, indicate how much you like this chicken breast for texture.", and "Please, indicate how much you like this chicken breast for overall liking."

The used CATA terms were based on previous studies reported in the literature on this product category (Liu et al., 2004; Baston et al., 2010; Tasoniero et al., 2016; Xu et al., 2020; Escobedo del Bosque et al., 2022; Freire et al., 2022) and on an elicitation session performed with six people (50 % female; mean age 25.8 years) of staff from the Department of Agricultural and Food Sciences of the Alma Mater Studiorum - University of Bologna. Two CATA lists have been developed, the first one including attributes related to flavor, aroma and taste, while the second one lists the visual and texture descriptors. The first CATA list contained 24 attributes: i.e., *ammoniacal notes, animal, blood, beef broth, chicken, egg, feed, seaweed, wet cardboard, fermented, hay, insect/bait, mold, sulphurous notes, fish, wet feathers, rancid, metallic, wet soil, toasted; acid, bitter, sweet, salty*. The second one contained 16 descriptors: i.e., *white, yellow, grey, pale, presence of blood, presence of nerves, rose, brown, consistency (firmness), hard, fibrous, gummy, sandy, dry, juicy, tender*. Participants were asked to indicate all the attributes from each list that best describe the tasted sample.

The perceived intensity for four product characteristics, one for each sensory modalities i.e., tenderness (tactile), pink color (visual), acid taste (taste), typical chicken aroma (flavor), was evaluated using the Generalized Labelled Magnitude Scale (gLMS) ranging from 0 to 100 (Bartoshuk et al., 2004).

As latest task, participants were asked to respond to some questions about their chicken consumption habits (consumption frequency, most consumed cut of chicken, propensity for fresh meat or transformed products, reasons related to chicken consumption) and to provide socio-demographic data (age, gender, nationality).

Data were collected using the software Fizz 2.51 (Biosystèmes, Couternon, France) in individual computerized sensory booths prepared with a plate, a fork and a napkin to better manipulate the sample. In addition, water and unsalted rice cakes were made available for mouth

rinsing and cleaning and a paper with a pen to take notes were provided to each participant (Fig. 1). The average time to complete the test by the participants was 20 min.

2.6. Instrumental analysis

Samples were subjected to two types of instrumental analyses: image (three replicates for each sample) and texture analysis (five replicates for each sample). They were tested under the same cooking and preparation conditions (cut into approximately 4 cm x 4 cm and roughly 2 cm high sub-samples, each having an average weight of 14 gs per portion) as the sensory evaluation session: portioned, vacuum packed and cooked sous-vide at a temperature of 92 °C for 1 hour.

2.6.1. Image analysis

The aim of image analysis was to evaluate eventual color differences using an instrumental technique in addition to the subjective evaluation performed by consumers. Image analysis was carried out using an “electronic eye” (Visual Analyzer VA400 IRIS, Alpha MOS, Toulouse, France), a high-resolution (2592 × 1944 p) charge-coupled camera equipped with a photo camera (16 million colors). The instrument is furnished with two lights (2 × 2 fluorescent tubes) with a color temperature of 6700°K; only the light that shines from above has been used to take the pictures of the samples. Samples have been analyzed similarly to Tura et al. (2024) and Valli et al. (2022) in particular, they were placed on a white plastic tray, diffusing a uniform light inside the device’s closable light chamber, and the CCD camera took a picture. The instrument was calibrated with a certified color checker (ColorChecker Classic, x-Rite, Grand Rapids, MI, USA) before taking pictures. Both image analyses (RGB scale or CIE L*, a* and b*) and statistical analyses were carried out.

2.6.2. Texture analysis

Similarly, samples were submitted to a TA-XTplus Texture Analyzer in order to instrumentally measure mechanical parameters related to tenderness: max force of compression (F) and area under the curve (A). F and A are interrelated. Five replicates for each sample (C, HI9, HI18, SA3 and SA6) were analyzed. We settled the method following the available literature on texture analysis for meat product (Aguirre et al., 2018; Masoumi et al., 2018; Shin and Choi, 2021). All the samples were compressed using a cylinder probe with a diameter of 25 mm and a height of 76 mm. A 30 kg load cell at a test speed of 1.0 mm/sec was used to reach 4.0 mm of compression (50 % strain).

2.7. Data analysis

All data analyses were performed using XLStat 2018.1.1 (Addinsoft, Boston, MA, USA). The analyses were conducted in the same way for all the random subjects, categorized them by gender (male and female subjects), consumption frequency (high frequency and low frequency) and by excluding subjects who prefer transformed products to fresh meat.

Liking data were submitted to two-way ANOVA models (random factor: subjects; fixed factor: type of feed) to investigate the effect of feed on liking in the four different aspects (color, flavor, texture and overall liking). Tukey’s HSD Test ($p \leq 0.05$) was performed after two-way ANOVA models to test for significant differences between mean values. Liking data were also subjected to the definition of an internal preference map, representing each consumer as a direction of preference, via Principal Component Analysis (PCA) correlation.

For the CATA test data, a calculation of the number of times each descriptor was chosen by consumers for each sample (frequencies) was conducted to obtain two occurrence matrices, one for CATA related to flavor descriptors, and the other for CATA related to visual and texture descriptors. To test the significance of the attributes in discriminating among the five samples, Cochran’s Q tests were performed on

occurrence matrices. In the end, a penalty analysis (PA) was performed between the significant attributes ($p \leq 0.05$) and the overall liking to find the positive and negative drivers of liking for the samples. The PA included graphical representations of CATA data for visual and texture descriptors. These graphs displayed the percentage of participants selecting a specific attribute across the samples on the X-axis, while the Y-axis represented the average impact of these attributes on overall liking. Notably, the CATA matrix pertaining to flavor descriptors did not yield any statistically significant results ($p > 0.05$), thus it was excluded from the subsequent statistical analysis. In addition, two-way ANOVA models (random factor: subjects; fixed factors: type of feed and samples; two-way interaction model: feed x sample) were used to investigate the interaction between intensity perception and the type of feed for each sample. These ANOVA models were also followed by Tukey’s HSD Test ($p \leq 0.05$) when appropriate.

Finally, the software Alphasoft version 14.0 (Alpha MOS, Toulouse, France) was used to explore the data from the image analysis using principal component analysis (PCA). Texture data on max force (F) were analyzed by non-parametric Kruskal-Wallis test (one-way ANOVA analysis, $p \leq 0.05$) since we were dealing with heteroskedastic data. The Dwass-Steel-Critchlow-Fligner pairwise comparisons test was used in post-hoc analysis.

3. Results

3.1. Panel description, chicken consumption habits and preferences

Participants were mostly from Italy (94 %), while the remaining were from Spain (3 %), Portugal (1 %), Mexico (1 %) and Costa Rica (1 %). All participants were aged 18–55 years old (mean age 25.3), and 48.8 % of them were female. Most participants were regular chicken consumers with 56 % declared to consume it once or twice per week. As for the remaining, 10 % declared to consume it from 3 to 4 times per week, 19 % less than once a week, 6 % once a month or less, 6 % a few times per year, and only 1 % declared they never consumed chicken meat.

Moreover, two third of the participants (67.8 %) indicated that they most frequently consumed chicken breast, while 15.4 % of subjects preferred chicken legs, 8.3 % opted for the entire chicken, 7.1 % favored chicken thighs, and only one subject went for chicken wings. Finally, 91.7 % of participants preferred fresh chicken cuts (such as breast, thighs, wings, whole chicken to cook) over processed products (such as chicken cutlets, nuggets, ready-made meatballs, pre-cooked roast chicken, etc.).

When subjects were asked to indicate the reasons why they like to consume chicken meat, 61.9 % responded that chicken is a versatile ingredient for cooking, while 36.9 % of participants indicated that it is for the affordable price and because chicken is lean meat.

3.2. Liking

Significant differences among samples regarding color and overall liking have been highlighted ($p \leq 0.05$). In particular, the two samples of breast from chicken fed with 3 % and 6 % of SP were the most liked (mean value: SA3 = 6.143 and SA6 = 6.262) (Two-way ANOVA, $p < 0.001$) (Fig. 2). Moreover, significant differences have been evidenced concerning the overall liking ($p \leq 0.05$); in particular, the sample of chicken fed SA3 was the most liked (mean value of overall liking = 6.226) (Fig. 2).

An internal preference map of the five chicken breast samples was obtained (Fig. 3). The first two component of the model explain 62.6 % of the variance with the first-dimension accounting for 32.9 % of the variance and the second for 29.7 %. The biplot in Fig. 3 reports scores and loadings of the model.

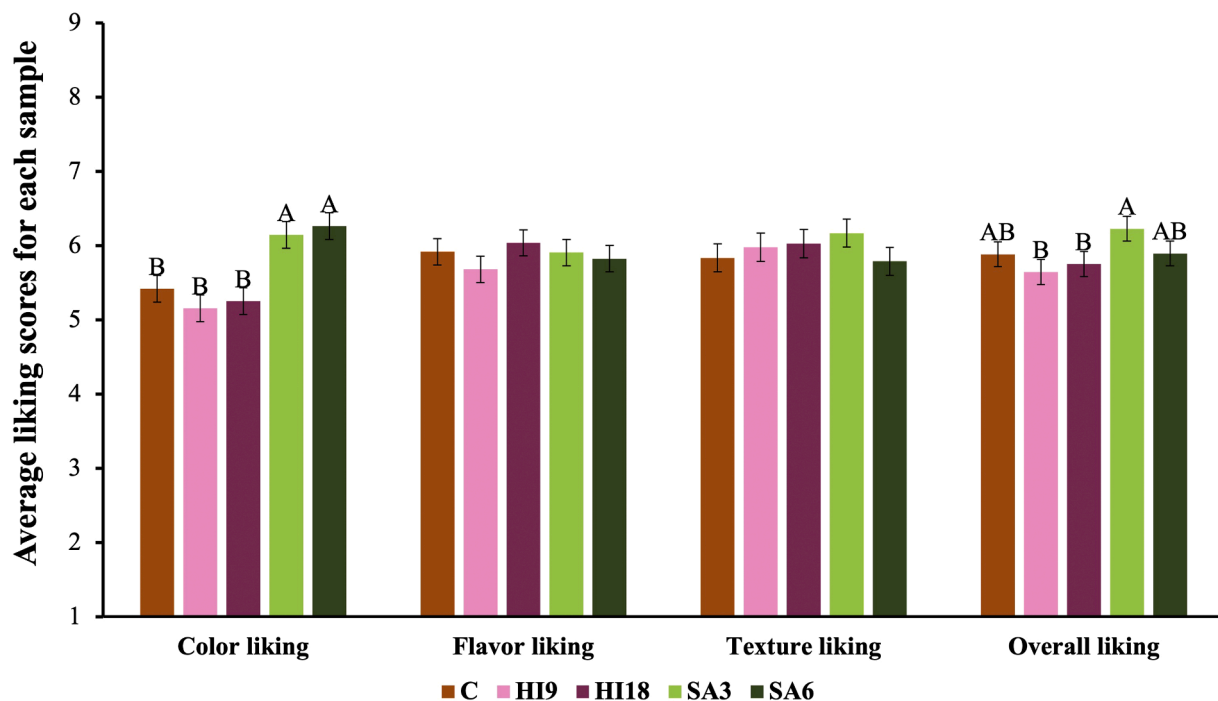


Fig. 2. Histogram depicting the results relating to liking in terms of color, flavor, texture and overall liking for the five chicken samples. C = chicken fed with conventional feed; HI9= chicken fed 9 % of Black soldier fly meal (*Hermetia illucens*); HI18= chicken fed 18 % of Black soldier fly meal (*Hermetia illucens*); SA3= chicken fed 3 % of Spirulina algae; SA6 = chicken fed 6 % of Spirulina algae. The y-axis shows the average liking scores for each sample, measured using the 9-point hedonic scale. Different letters indicate statistically significant differences (Two-way ANOVA, $p < 0.001$, Tukey's HSD).

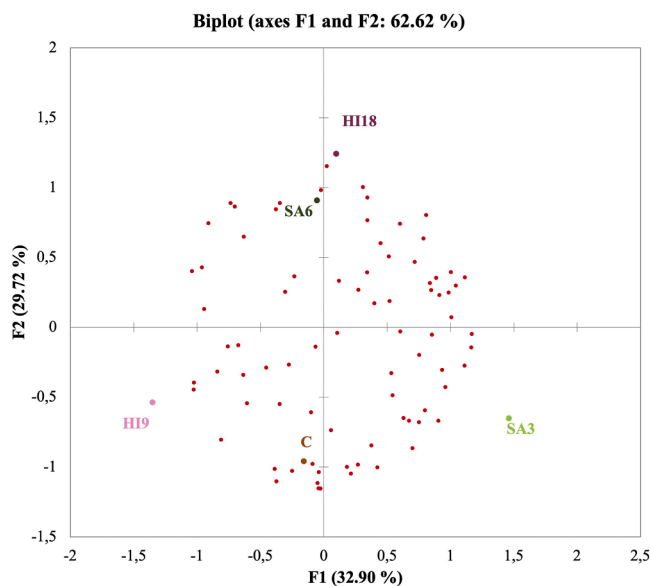


Fig. 3. Internal preference mapping score and loading biplot. C = chicken fed conventional feed; HI9= chicken fed 9 % of Black soldier fly meal (*Hermetia illucens*); HI18= chicken fed 18 % of Black soldier fly meal (*Hermetia illucens*); SA3= chicken fed 3 % of Spirulina algae; SA6 = chicken fed 6 % of Spirulina algae to.

3.3. Check-All-That-Apply

Table 1 shows the number of citations for each of the attributes of the CATA question used to describe the evaluated chicken breast samples and the relative p-value (Cochran's Q tests). The most frequently used terms were "chicken flavor", "meat broth", "animal" and "metallic" in relation to flavor perception, while "pale", "white", "tender" and "pink"

were visual and texture attributes. Significant differences ($p \leq 0.05$) were found in the frequencies of 6 out of the 16 terms of the CATA test related to appearance and texture used to describe samples (Table 1), mainly for the terms related to the color of the chicken breast. No significant differences ($p > 0.05$) were found for the terms related to flavor and taste; this could be related to the relatively small differences between the samples.

3.4. Perceived intensity of selected descriptors

Each participant has rated the perceived intensity of four selected attributes on a gLMS scale. Each one of the four descriptors was related to a different sensory modality (i.e., tactile/tenderness, sight/pink, taste/sour, smell/chicken flavor). Statistically significant differences ($p \leq 0.05$) were found among the samples in terms of pink color. In particular, samples of chicken breasts from birds fed with the substitution of algae were perceived with a higher pink intensity ($p < 0.00001$) both with respect to the control sample (C) and to those fed with insect meal substitution (HI9 and HI18). While, for the descriptors "tenderness", "sour" and "chicken flavor" no statistically significant differences have been highlighted among the samples ($p \leq 0.05$) (Fig. 4).

3.5. Influence of attributes perception on liking

According to the Cochran's Q test results, five visual attributes and one related to the texture of the chicken breasts tested were significant, i.e., to discriminate the samples. Furthermore, the visual attribute "brown" and the texture attribute "juicy" showed p-values very close to significance ($\alpha=0.05$), i.e., 0.058 and 0.053, respectively (Table 1). Thus, a total of 8 descriptors (6 related to appearance and 2 to texture) have been included in the PCoA elaboration, together with overall liking scores (Fig. 5). As reported in Fig. 5, it has been highlighted that liking is strictly related to the pink color and to the juiciness of the chicken breast.

Table 1
Results of the Check-All-That-Apply (CATA) question. Number of citations for each attribute and evaluated samples and relative p-value (Cochran's Q tests).

Attribute	p-value	Sample						
		C	HI9	HI18	SA3	SA6		
Flavor and taste list	Mold	0.695	0	1	1	1	2	
	Chicken flavor	0.412	69	65	71	73	70	
	Animal	0.356	23	26	24	25	32	
	Meat broth	0.897	34	34	38	38	36	
	Wet earth	0.841	3	6	6	5	6	
	Egg	0.995	9	8	9	8	9	
	Bug/bait	0.478	0	1	1	2	0	
	Fish	0.303	6	3	1	5	5	
	Ammonia	0.406	0	2	3	1	3	
	Sulphur notes	0.209	8	3	3	4	6	
	Seaweed	0.600	4	6	4	5	2	
	Wet cardboard	0.980	12	12	10	12	11	
	Blood	0.255	6	14	10	9	8	
	Metallic	0.583	19	24	25	20	21	
	Toasted	0.417	9	5	3	5	5	
	Hay	0.995	7	7	8	8	8	
	Wet feathers	1.000	0	0	0	0	0	
	Rancid	0.763	13	15	14	16	11	
	Feed	0.755	11	9	10	8	13	
	Fermented	0.565	2	3	5	2	4	
	Salty	0.305	10	15	9	7	12	
	Bitter	0.219	2	5	1	4	1	
	Sweet	0.126	14	7	12	9	7	
	Sour	0.148	8	16	15	10	10	
	Appearance and texture list	Yellow	< 0.0001	9	7	4	25	36
		Presence of blood	0.579	2	5	5	4	2
		Gray	< 0.0001	19	28	32	10	7
		White	< 0.0001	46	54	49	23	19
		Brown	0.058	1	2	6	4	8
		Pale	< 0.0001	58	61	56	40	27
		Pink	< 0.0001	24	21	20	41	48
		Presence of nerves	0.192	5	9	5	12	6
Juicy		0.053	12	19	28	20	20	
Fibrous		0.361	25	27	27	33	34	
Gummy		0.273	9	9	16	15	13	
Firm		0.450	32	22	27	24	26	
Tender		0.207	40	40	32	38	29	
Sandy		0.605	8	9	13	8	8	
Hard	0.103	10	3	5	4	4		
Dry	0.021	30	27	16	19	30		

3.6. Instrumental analysis

The application of the software available with the instrument (Alphasoft, version 14.0, Alpha MOS, Toulouse, France) allowed color spectra to be grouped in bins of 16 bits for each RGB coordinate, resulting in 4096 variables that were analyzed. The proportion of each color in the analyzed image, on a fixed scale of 4096 colors, is represented as a percentage. Specifically, variables such as “color 4076” ($L^*=92.679$, $a^*=1.916$, $b^*=17.697$), “color 4059” ($L^*=88.457$, $a^*=8.016$, $b^*=20.152$), “color 4058” ($L^*=88.202$, $a^*=6.513$, $b^*=28.057$), and “color 3803” ($L^*=87.040$, $a^*=1.983$, $b^*=17.941$) are very pale yellow-orange colors and they characterize sample SA6. While samples SA3 and C are related to a more intense pink shades, such as “color 3240” ($L^*=71.156$, $a^*=8.649$, $b^*=21.187$) and “color 3787” ($L^*=83.055$, $a^*=9.845$, $b^*=12.424$). Sample HI18 were mainly characterized by dark colors, such as “color 3514” ($L^*=77.289$, $a^*=10.040$, $b^*=12.642$), “color 3531” ($L^*=81.615$, $a^*=3.835$, $b^*=10.158$), “color

3258” ($L^*=75.815$, $a^*=3.918$, $b^*=10.324$) and “color 3257” ($L^*=75.524$, $a^*=2.147$, $b^*=18.497$). On the other hand, sample HI9 appears to be characterized by lighter coloration, particularly “color 3786” ($L^*=82.772$, $a^*=8.202$, $b^*=20.463$) (Fig. 6).

In terms of max force, measured in kg for texture analysis, during the compression test, no significant differences were outlined by Kruskal-Wallis test ($p = 0.38$). The type of feed, from an instrumental point of view, had no effect on compressive force, and consequently on samples' level of tenderness. Table 2 displays these results.

4. Discussion

We investigated the effect of feed on the sensory quality of chicken breast. In particular, we researched how two different types of alternative protein sources in partial replacement of soybean can influence consumer perception and liking of the final product.

According to an investigation conducted by the BVA Doxa institute in 2021 (*L'indagine BVA Doxa per Unaitalia, 2021*) on a representative sample of the Italian population aged 18–74, 72 % consume chicken meat at least once a week, which is in line with our results. Moreover, the BVA Doxa survey also confirms our result regarding chicken breast, which is the preferred cut for the majority of Italians. In addition, the trend to prefer fresh cuts over processed products (as reported by 91.7 % of participants) is consistent with research conducted in the US (Wideman et al., 2016); these preferred cuts need further investigation in the European area as well. In general, chicken meat is mostly considered a versatile ingredient in food preparations.

For this reason, poultry is one of the main sources of food of animal origin in the world (Mottet and Tempio, 2017), to which are added grounds of religion (Barbut and Leishman, 2022), convenience (Kennedy et al., 2004) and nutrition (Farrell, 2013). A large part of our panel stated that the main reason why they like to consume chicken is the adaptability as ingredient, which is in line with Leroy and Degreef (2015) findings. Furthermore, our respondents' preferences align with the trends investigated by Michel et al. (2011), particularly in terms of their preferences for the affordable price and low-fat intake of chicken meat. However, when consumer behavior is under investigation, we cannot just consider stated preferences (Carlsson et al., 2022), and the impact of other factors, such as flavor and visual appearance, needs to be deepened. In particular, visual perception is a key factor in making decisions related to eat or not to eat something (Altmann et al., 2023). In the assessment of meat's sensory quality, attention has been largely focused on other most common related attributes such as tenderness, chewiness, juiciness, flavor, and odor (Braghieri et al., 2012).

Our approach has included visual perception in this list, aiming to include this parameter in the analysis of cooked meat. While the decision-making process when purchasing meat is significantly influenced by the visual appearance of the raw product (Altmann et al., 2022), much less information is available regarding the visual characteristics of cooked meat. Nevertheless, as highlighted in this study, the evaluation of color emerged as a critical factor in the assessment of liking for the cooked samples, emphasizing the pivotal role that visual cues, even after cooking, play in shaping consumer preferences. The characteristic color of chicken breasts depends on several factors, such as bird sex, age, strain, method of processing, exposure to chemicals, cooking method, irradiation, freezing and feed (Fletcher, 2002; Tomasevic et al., 2021). It is essential to remind that color represents one influential visual trait usually involved in the decision-making process by consumers when purchasing meat (Tomasevic et al., 2021). Image analysis allowed the color spectrum of each sample to be obtained in RGB coordinates (red, green, blue). The analysis is rapid, and the equipment is easy-to-use, being potentially useful also for routine quality control, given that no sample preparation is required.

Even if color is strongly perceived as an indicator of the freshness of meat (Tomasevic et al., 2021), in the case of chicken, a flesh-colored white (i.e., pale or light grey) or yellow or pink does not imply

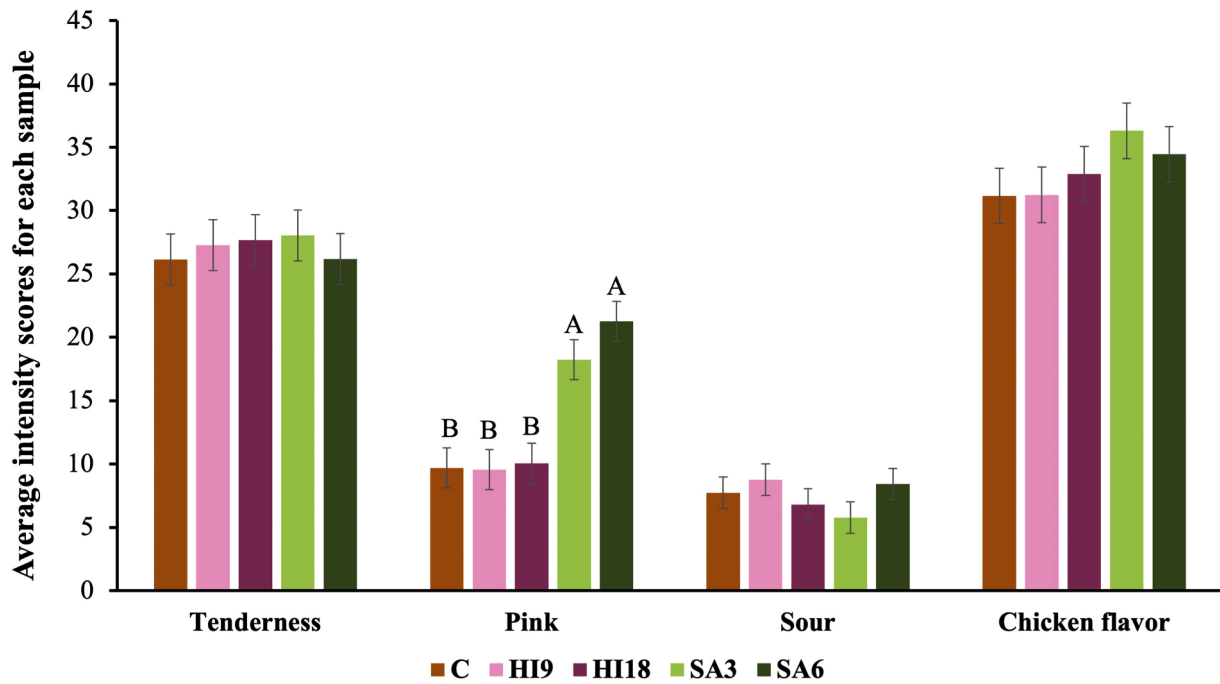


Fig. 4. Histogram depicting the results relating to perceived intensity of four preselected attributes for the five chicken samples. C = chicken fed with conventional feed; HI9= chicken fed 9 % of Black soldier fly meal (*Hermetia illucens*); HI18= chicken fed 18 % of Black soldier fly meal (*Hermetia illucens*); SA3= chicken fed 3 % of Spirulina algae; SA6 = chicken fed 6 % of Spirulina algae. The y-axis shows the average intensity scores for each sample, measured using the Generalized Labelled Magnitude Scale (gLMS). Different letters indicate statistically significant differences (Two-way ANOVA, $p < 0.0001$, Tukey’s HSD).

Principal Coordinate Analysis (axes F1 and F2)

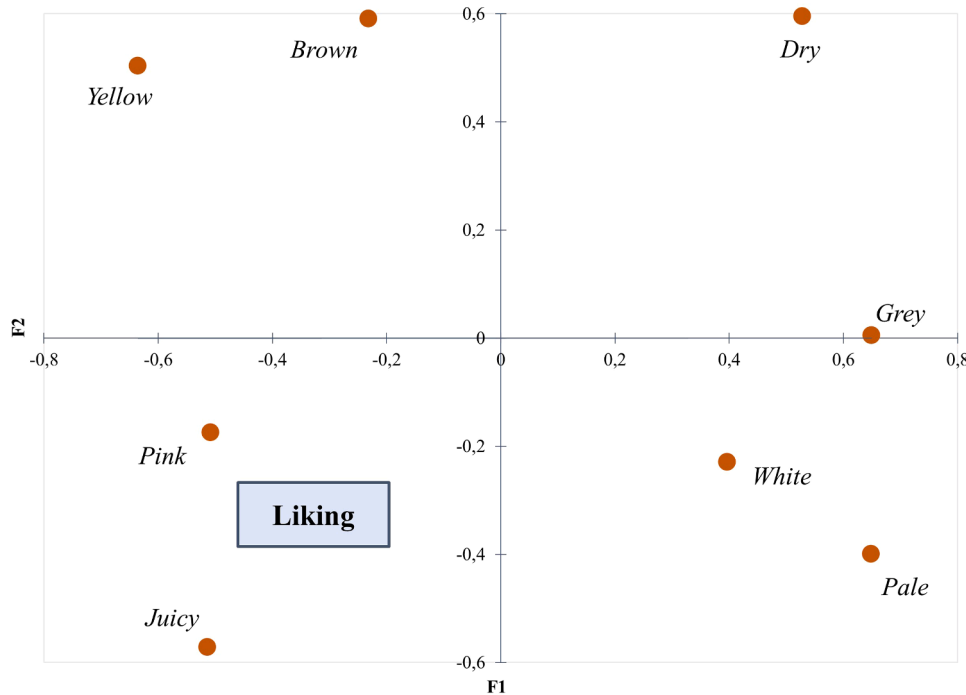


Fig. 5. Principal Coordinate Analysis (PCoA) of the statistically significant ($p \leq 0.05$) CATA descriptive attributes related to appearance and texture (Cochran’s Q test) and “juicy” and “brown” descriptors which p-value was slightly above $\alpha=0.05$ with the overall liking score.

differences in terms of overall quality. In fact, the color of raw meat can depend not only on the genetics and breed of the animal (Escobedo del Bosque et al., 2022), but also on the feeding process (Smith et al., 2002).

When animals are fed with a fodder containing, for instance, carotenoids, and this is the case of feed with the substitution of Spirulina

algae, they tend to accumulate pigments in the flesh (Altmann and Rosenau, 2022). The result is a product (i.e., chicken breast) more yellow- or pink-shade. In the case of insect-based feed, the majority of studies have found no significant differences in raw meat color compared to a product obtained from animals fed with conventional

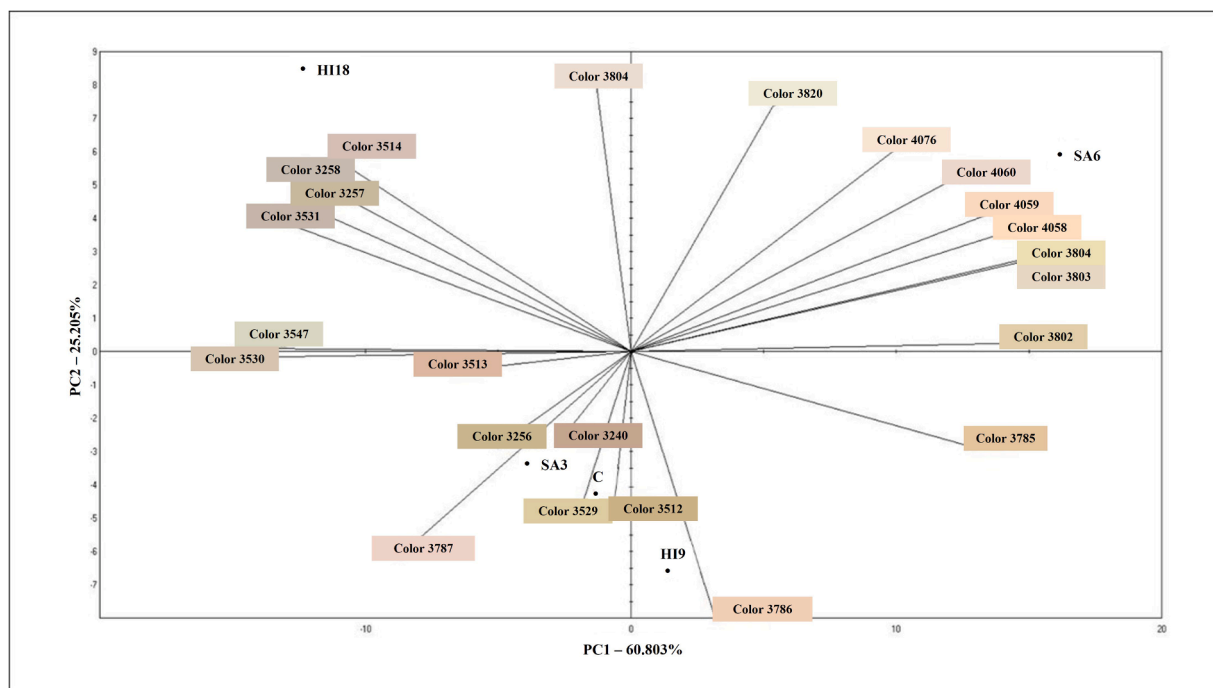


Fig. 6. Biplot of scores and loadings (intensities and shades of colors) on a plane (PCA). C = chicken fed with conventional feed; HI9 = chicken fed 9 % of Black soldier fly meal (*Hermetia illucens*); HI18 = chicken fed 18 % of Black soldier fly meal (*Hermetia illucens*); SA3 = chicken fed 3 % of Spirulina algae; SA6 = chicken fed 6 % of Spirulina algae.

Table 2

Group descriptives in terms of number of replicates (N), mean (M) and SD (standard deviation), for max force of compression (F) measured by texture analysis and analyzed by Kruskal-Wallis test (one-way ANOVA model, $p \leq 0.05$).

Type of Feed	N	M	SD
C	5	2.46	1.36
HI9	5	2.11	1.11
HI18	5	3.08	1.96
SA3	5	3.58	1.18
SA6	5	2.47	1.09

soybean-based feed (Gasco et al., 2019). The final product tends to exhibit a white/pale or light grey appearance, similar to that of a conventional product. The cooking process reduces the degree of color variation (Fletcher et al., 2000), but the initial color of the chicken breast does not undergo significant changes. This is confirmed by our results. Our respondents perceived the pink color more intensely in samples from animals fed with the two levels of substitution of Spirulina algae in the feed. This could be related to the presence of carotenoids in Spirulina algae included in the feed, which influence the color of the meat (El-Shall et al., 2023).

No differences in terms of perceived color intensity were shown between control and insect-based feed samples. At the same time, the samples where the perceived intensity of the pink color was higher, were the most preferred with regard to color liking. Furthermore, SA3 was the most preferred sample when it came to overall liking. This confirms a trend already underlined by Kennedy et al. (2004) and Droval et al. (2012), which investigated preferences related to poultry meat color among Northern Ireland and Brazilian consumers. A pink color is generally preferred by consumers for raw meat, and color uniformity leads to higher acceptance (Geronimo et al., 2022). Moreover, preferences for poultry color vary by region and by culture (Kennedy et al., 2004). Thus, the relationship between liking and pink color might be culture specific. Since the research on the color of chicken mainly focused on raw meat, the investigation of preferences related to the color

of cooked meat needs to be deepened. As we highlighted, color is one of the main drivers associated with consumer preferences, and our research could be a starting point for the analysis of the Italian market.

With regards to CATA analysis, besides color, two attributes related to texture, “juicy” and “dry”, were indicated to be able to discriminate between samples. It is worth to mention that the two attributes are strongly anticorrelated, thus indicating the same sensory perception we can summarize in the “juiciness”. This confirms the importance of texture-related attributes for the determination of a sensory profile for meat products, as pointed out by Braghieri et al. (2012). If visual parameters, such as color, are critical for the first decision-making buying process, flavor and texture characteristics are essential to assess preferences and liking (Baston et al., 2010; Napolitano et al., 2010; Xu et al., 2020). In the case of meat, juiciness (i.e., the ability of meat to retain moisture) remains a fundamental criterion for liking assessment (Huff-Lonergan and Lonergan, 2005). It is crucial to provide a product that is able to maintain this characteristic, even after different preparation and cooking processes. If both sensory and instrumental analyses did not find any significant differences ($p \leq 0.05$) between samples in terms of tenderness, the attribute “juicy” was mostly associated with SA3 and SA6 by our respondents. This is another confirmation of the potential of feed partially substituted with Spirulina algae to provide a product that meets the tastes of consumers. As concerns flavor, no differences were perceived by the participants, confirming that the use of alternatives in feed composition basically has no or low impact on the final product flavor profile (Cullere et al., 2019; Altmann et al., 2020).

We performed this sensorial analysis session using a blind test, which did not provide information on the alternative feed submitted to the animals the samples came from. The results clearly underlined the preference of our panel for the product fed with a substitution of Spirulina algae.

But does this stated preference correspond with a clear intention to buy the product? Kyto et al. (2018) demonstrated how purchase intentions are better predictable in the context of informed tasting. If, in fact, our study confirms the effect of feed on consumer perception and liking, to completely understand consumers purchase intentions, it is

necessary to investigate the role of information (Endrizzi et al., 2021). Already proven to be essential in the acceptance process of novel products (Laureati et al., 2016; Baldi et al., 2021; Sogari et al., 2022), information can also help to reduce some effects that may become barriers to consumption. With regards to insects, food neophobia (Tuccillo et al., 2020) and disgust (Spartano and Grasso, 2021) are the main barriers to the development and diffusion of this ingredient in the Western countries, even if it is used in feed preparation (Higa et al., 2021). At the same time, the unfamiliarity with an ingredient such as Spirulina algae (Grahl et al., 2020) could be critical in communicating with final consumers. As reported by Asioli and Grasso (2021), informed consumers of the nutrition and environmental benefits of upcycled ingredients are more willing to pay for innovative products. In the case of chicken meat, Escobedo del Bosque et al. (2021), demonstrated how consumers consider feedstuff origin a driver of their preferences. Consequently, from a future perspective, it would be interesting to combine sensory evaluation in both blind and informed conditions, to further investigate the role of feed on perception and liking, as already done by Grasso et al. (2022) for hybrid beef and plant-based burgers. Even Schouteten et al. (2016) performed an investigation on insect-, plant- and meat-based burgers under blind, expected and informed conditions, finding how information actually influenced overall liking for insect-based burgers.

Preference for spirulina feed chicken breast is an intriguing phenomenon that can be attributed to several key factors. Firstly, spirulina is known to be a rich source of high-quality proteins, vitamins, and essential minerals, including vitamin B12 and iron, which can enhance the nutritional value of chicken meat (AlFadhly et al., 2022). This feature is particularly appealing to consumers seeking healthier and more nutrient-dense food options. The increasing consumer awareness regarding the significance of a sustainable and environmentally friendly diet may have played a crucial role in the preference for chicken breast from animals fed with Spirulina algae. Raising these animals can help reduce the ecological footprint and contribute to addressing issues linked to intensive farming (Tso et al., 2020). In summary, the decision to consume chicken breast from animals fed with Spirulina algae may encompass a blend of nutritional, sensory, and ecological factors that align with the changing needs and preferences of today's consumers.

In the end, the forthcoming studies could use this research as a starting point to explore the correlation that exists between production methods and the final sensory profile of meat, under different consumers' decision-making conditions. The consumer preference for poultry products sourced from animals fed with spirulina algae can have significant implications for the poultry industry. As consumers increasingly prioritize health-conscious and sustainable food choices, poultry producers may find it beneficial to explore and invest in sustainable feeding practices that incorporate nutrient-rich ingredients like spirulina into the diets of their poultry. This could lead to the development of premium poultry products with enhanced nutritional profiles and potentially unique flavor profiles, catering to a growing market segment. Moreover, aligning with sustainable and environmentally friendly practices in poultry farming may not only appeal to conscious consumers but also help poultry producers reduce their ecological footprint and comply with evolving industry standards and regulations. Overall, understanding and responding to consumer preferences for spirulina-fed poultry can be a strategic move for the poultry industry to meet changing market demands and maintain competitiveness in the evolving landscape of food production.

5. Conclusions

The effect of alternative protein sources in chicken feeding on consumer's perception and liking for breast fillets was evaluated by sensory analysis. Liking data on color, texture and flavor profile were collected, as well as descriptive attributes, using the CATA method.

This approach allowed us to identify the meat characteristics that are

strictly related to liking in a blind test: in the case of chicken breast fillets, a pink color and a juicy texture. No less, this study demonstrated that these specific features clearly define the sensory profile of chicken breasts from animals fed diets containing Spirulina algae meal in partial substitution of soybean. The combination of these factors enables us to consider the product derived from chicken receiving Spirulina algae supplementation as potentially valuable to meet consumer preferences. In future research, it may be beneficial to consider incorporating this method along with preference analysis conducted under well-informed conditions regarding the environmental advantages of using alternative protein sources in chicken feeding. This would enable the acquisition of a comprehensive consumer profile.

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Ethical statements

Ethical approval for the involvement of human subjects in this study was granted by the Alma Mater Studiorum – Università di Bologna Bioethical Committee, Reference number 0,173,440, dtd 07/28/2022.

Participants gave informed consent via the statement "I am aware that my responses are confidential, and I agree to participate in this survey" where an affirmative reply was required to enter the survey. They were able to withdraw from the survey at any time without giving a reason. The products tested were safe for consumption.

The study was explained to consumers previous to the questionnaire. They were informed that all data will be anonymously recorded and only reported in the aggregate. All participants acknowledged an informed consent statement in order to participate in the study.

CRediT authorship contribution statement

Rosalba Roccatello: Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Matilde Tura:** Writing – original draft, Visualization, Methodology, Investigation, Formal analysis, Data curation. **Eugenio Aprea:** Writing – review & editing, Supervision, Resources, Project administration, Methodology, Data curation, Conceptualization. **Sihem Dabbou:** Writing – review & editing, Writing – original draft, Visualization, Supervision, Data curation, Conceptualization. **Francesca Soglia:** Writing – review & editing, Writing – original draft, Visualization, Supervision, Data curation. **Federico Sirri:** Writing – review & editing, Supervision, Resources, Funding acquisition, Data curation. **Tullia Gallina Toschi:** Writing – review & editing, Supervision, Resources, Project administration, Methodology, Data curation, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The data that support the findings of this study are available from the corresponding author upon request.

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