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Recipe Optimization to Produce Functional Food Based on Meat and Fish

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Abstract

Functional foods were produced from the combination of minced meat or fish with extra virgin olive oil and vegetable flour. Proper concentrations of oil and vegetable flours were selected to improve the nutritional properties of processed meat and fish burgers. Sensory evaluations were also carried out in different sessions by trained panel and generic consumers to assess product acceptability. The results of the work demonstrated that the two ingredients, selected for their well-known healthy benefits, enhance not only the nutritional value but also the sensory properties of food. In particular, the best formulation for meat patties contained 10% oil and 10% red pepper flour, whereas, 15% oil and 5% yellow pepper flour and 17.5% oil and 7.5% zucchini flour were the optimized ingredients for fish burgers. These recipes did not score significantly different from other CCD options but were preferred by consumers, when asked to compare these new products to traditional burgers. Moreover, analytical investigations carried out before and after cooking assessed the preservation of the health-giving compounds deriving from the pepper flour.

Keywords: Meat; Fish; Functional food; Oil; Whey protein; Vegetable flour

Introduction

Advances in food and nutrition sciences have highlighted the possibility of modulating some specific physiological functions in the organism through food intake [1,2]. This means that diet can provide consumers with components able to both modulate body functions, improve health status and wellbeing and/or reduce the risk of some diseases. This is the context of so-called functional foods that actually represents one of the chief factors driving the development of new products [3-5]. Market of functional foods is characterized by being dynamic and innovative with a quota of 10 to 15% and a growth rate of 20 to 30% per year, at world level.

All food are functional, in the general meaning of the term, insofar as they supply energy and nutrients necessary toward growth and maintenance. However, it can be stated that a food is functional if it allows the elimination of known components that cause deleterious effects when consumed (e.g., allergenic proteins); increases the concentration of a component naturally present in food to induce predicted beneficial effects (e.g., fortification with a micronutrient to reach a daily intake higher than the recommended daily intake but compatible with the dietary guidelines for reducing risk of disease); contains a component with known beneficial effects that is not normally present in most foods (e.g., non-vitamin antioxidant or prebiotic fructans); replaces a component (e.g., fats) whose intake represents a cause of deleterious effects, by a component for which beneficial effects have been shown (e.g., chicory inulin); increases the stability of a component known to reduce the potential disease-risk of food [6].

With the increasing popularity of functional products among consumers, food companies need to face the call for the manufacture of such products in order to appropriately meet constant market requests.

Meat-based functional foods are seen as an opportunity to improve their image and address the needs of consumers, as well as to update nutrient dietary goals [4,7-8]. Even though meat is frequently associated with a negative health image due to its high fat content and cancer-promoting food, meat ensures adequate delivery of essential micronutrients and amino acids. It is an important source for iron, selenium, vitamins A, B12 and folic acid. Therefore, being meat a protein rich and carbohydrate low product, a low intake, especially of red meat, is recommended to avoid the risk of cancer, obesity and metabolic syndrome [9]. One of the most important approaches to the development of potential meat-based functional foods is the partial replacement of meat fats with various non-meat fats of plant and marine origin [8]. Several technological options have been applied to fresh, cooked and fermented meat products to give healthier lipid formulation [10-13]. Numerous efforts have been also made to fortify meat products with various components [14-16].

Fish and shellfish are also excellent protein sources for human consumption; in addition, they have a high content of hydro-soluble and lipo-soluble vitamins, minerals and polyunsaturated fatty acids (PUFAs) of the n-3 family [17]. Of the n-3 PUFAs, α-linolenic acid is present in large quantities in plant oils, while other long chain fatty acids are found largely in seafood [18]. Being n-3 fatty acids capable to confer health benefits in humans, consumption of fish is advocated at least two times a week, as a safe and effective way to obtain health benefits. Coinciding with this, traditional fishing is undergoing a crisis caused most probably by overexploitation of fishing grounds and the interest has been redirected to augmenting the quality of the product offered [19]. It is employed not so much as a preservation method

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but as a food processing technique to diversify the supply of available products and enhance added value. This is among the factors that have stirred growing interest in fish farming, with notable success for sea bream and bass. Some efforts have been also made to extend shelf-life and improve the quality of smoked fish products [20].

Besides meat and fish, fruit and vegetables represent other important sources of phytochemical components, such as vitamins, minerals, folates, fiber, carotenoids, polyphenols, phytosterols [21, 22]. These compounds exerted an important antioxidant protection against the free radicals that are generally implicated in the development of various health disorders [23]. Tomatoes have received much attention for the potential role of lycopene in cancer risk reduction. Garlic (*Allium sativum*) contains an odorless amino acid, which is converted enzymatically into allicin that, in turn, decomposes spontaneously to form numerous sulfur-containing compounds that can inhibit tumor genesis. The tea, particularly green tea, is rich in polyphenols. Epidemiological evidence has also associated the frequent consumption of cruciferous vegetables with decreased cancer risk for the high content of glucosinolates. Several epidemiological studies have also shown that citrus fruit are protective against a variety of human cancers due to nutrients as vitamin C, folate, fibre and phytochemicals known as the limonoids. Red sweet peppers may provide beneficial effects on health because are generally reach in capsanthin that is an effective antioxidant, more active than β-carotene, lutein and zeaxanthin [24, 26]. Carotenoids are synthesized by all plants and many microorganisms, but not by animals, including humans, who therefore rely on dietary uptake. Besides the well-known provitamin A activity of some carotenoids, further potential health beneficial properties have moved into the focus of investigation, especially those related to the prevention of a number of chronic diseases.

Giving the above considerations, the aim of the current work is to develop new functional food based on meat and fish, by combining the beneficial effects of these matrices to the health promoting effects of extra-virgin olive oil and flours of vegetable origin. The nutritional and sensorial aspects were taken into account. To this aim, a preliminary screening of flours and of different concentrations of oil were carried out; subsequently, a fine optimization of the product formulation was achieved by elaborating a central composite design (CCD). The preservation of the health-giving compounds in the product after cooking was also evaluated.

Materials and Methods

Production of functional meat and fish-based burgers

Hamburgers of beef were produced with meat purchased from a local farm in Foggia (Italy). Different amounts of whey protein-based crumb (DAVISCO Food International, Inc., USA) soaked in commercial available extra-virgin olive oil, were also added to meat formulation. The whey protein crumb was prepared according to Del Nobile and others [10], by mixing 100 g whey protein, 4 g NaCl, 10 g Na_2CO_3 and 120 ml distilled water. This mixture was cooked at 160 °C for 40–50 min in the oven (Moulinex Activys, France). After cooking, the whey protein-based foam was minced by a Sterilmixer (PBI International, USA), reduced to crumbs that were soaked in the extra virgin olive oil in a 1:4 ratio and mixed at different amounts (5, 10, 15 and 20% w/w) with minced meat. Each mixture was homogenized for a few minutes and then shaped by means of a burger maker to obtain hamburgers with extra virgin olive oil. As control samples, hamburgers with sole minced meat were also realized.

Hamburgers enriched with oil and vegetable flour were also produced. To this aim, flours obtained from eggplants, carrots, red peppers, pumpkins, fennels, cauliflowers, zucchini, tomatoes, artichokes, broccoli, yellow peppers and asparagus were used, separately. All the flours were purchased from a local farm, Farris (Foggia, Italy). Each flour was mixed with the minced meat formulation containing whey protein soaked in oil (15% w/w), to reach a final concentration of vegetable flour of 10% (w/w). As control samples, hamburgers with sole meat and hamburgers without flour were also realized.

After the screening of the several flours, the research was aimed to determine the best formulation of functional hamburgers in terms of amount of meat, oil and flour. To reduce the number of possible combinations to a manageable size, hamburgers were prepared as above, according to a Central Composite Design (CCD). The effects of the three ingredients (meat, oil and flour) were investigated by modulating the levels of meat and oil according to a two-factor, five-level CCD [27] and by setting the amount of flour as completion to 100%. The five levels chosen for each factors (independent variables) are listed in Table 1.

As regards fish burgers, sea bass *(Dicentrarchus labrax)* was used. Specimens of sea bass were purchased from a local farm (Cariglia, Manfredonia, Foggia, Italy). Fishes were slaughtered by immersion in ice-cold water (hypothermia) and packed in insulated expanded polystyrene boxes with ice. Then, they were delivered to the laboratory within 2 h from the moment of the harvest. Once at the laboratory, fishes were decapited, cleaned, filleted and skinned. Skin-off fillets of species were weighted and minced by a domestic food processor (Multichef, Ariete, Firenze, Italy) to produce a fish patty. As for hamburgers, also for fish burgers, different amounts of whey protein, prepared and soaked in extra virgin olive oil as described for meat, were added to the formulation (5, 10, 15 and 20% w/w). Each mix was homogenized for a few minutes and then formed in a burger maker to obtain a fish burger

Samples	Meat [%]	Oil-soaked-whey protein-foam [%]	Red Pepper Flour [%]
A	80	12,50	7,50
B	75	12,50	12,50
C	85	7,50	7,50
D	80	7,50	12,50
E	80	10	10
F	75	10	15
G	85	10	5
H	75	15	10
I	85	5	10
L	80	10	10
M	80	10	10

Table 1: Combinations of meat, oil-soaked-whey protein-foam and pepper flour, according to a two-factor/five-level CCD. The amount of flour was set as completion to 100%.

with extra virgin olive oil. As control samples, burgers with sole fish were also produced.

In a subsequent step, different flours were added to the fish formulation containing extra virgin olive oil (10% w/w). As above, flours obtained from eggplants, carrots, red peppers, pumpkins, fennels, cauliflowers, zucchini, tomatoes, artichokes, broccoli, yellow peppers and asparagus were used, separately, to reach a final concentration of 10% (w/w). The mix was homogenized in a bowl mixer with a spiral dough hook during 5 min. As control samples, burgers with sole fish and burgers without flour were also realized.

To select the final best combinations of fish, oil and vegetable powder, a CCD was conducted. As above, a two-factor, five-level CCD was used, by setting the amount of flour as completion to 100%. The five levels chosen for each factors (independent variables) are listed in Table 2.

Sensory evaluations

Panel and consumer tests were carried out on both hamburgers and fish burgers. To select the approximate amount of oil and the best flour to be added to minced meat and fish, trained panelists were used. On the other hand, the final acceptability of products with both oil and flour was assessed by consumer tests. A number of 7 judges were used for panel test, according to previous works dealing with sensory evaluation of meat and fish-based products [28-31]. Our panelists had at least several years of experience in evaluation of meat and fish products prior to this study. The panelists were retrained for this study in four sessions held over two days (2sessions/day, 2 h/session). Retraining samples included uncooked and freshly cooked meat and fish patties. Appropriate descriptive terms for sensory evaluation were decided during the retraining sessions. After retraining, experienced graders were able to evaluate product appearance, flavor, juiciness and texture as main quality parameters of these types of foodstuffs [30,32-33]. Panel tests were conducted in different sessions for meat and fish, using individual booths (located away from the sample preparation area) under red-filtered incandescent light to avoid bias, due to potential color differences among samples. Each patty was cut into four wedges, assigned with three-digit random numbers and served to each panelist. A glass of water and unsalted crackers were provided to cleanse the palate among samples. The evaluators were asked to give a judge on color, odor, juiciness, texture, taste and on the whole quality of each type of product before and after cooking in a microwave oven to an internal temperature of about 57 °C. A 9-point scale was used, where the category definitions were: 1, extremely dislike; 2, dislike very much; 3, dislike moderately; 4, dislike slightly; 5, neither like nor dislike; 6, like slightly; 7, like moderately; 8, like very much; 9, like extremely [28, 34]. According to the scale, a score of 5 was taken as the lower limit of acceptability. Experimental samples were evaluated in a total of four sessions held over 2 days (2 sessions/day, one for meat and one for fish products, with a 2h break between sessions). Samples serving order in each session was randomized.

Samples	Fish [%]	Oil-soaked-whey protein-foam [%]	Vegetable Flour* [%]
A	75	17,50	7,50
B	70	17,50	12,50
C	80	12,50	7,50
D	75	12,50	12,50
E	75	15,00	10,00
F	70	15,00	15,00
G	80	15,00	5,00
H	70	20,00	10,00
I	80	10,00	10,00
L	75	15,00	10,00
M	75	15,00	10,00

**Vegetable flours: zucchini or yellow pepper flour.*

Table 2: Combinations of fish, oil-soaked-whey protein-foam and vegetable flour, according to a two-factor/five-level CCD. The amount of flour was set as completion to 100%.

Once defined the general formulation to be used, consumer tests were carried out for both the new meat and fish products. Hamburgers and fish burgers were produced according to the above reported CCD and the different samples were judged by 80 subjects, composed of students and staff of the University of Foggia, usual consumers of meat and fish (48 females, 32 males; age range 21-50). As above, the sensory evaluations were conducted in individual booths, under red-filtered incandescent light, in different session for meat and fish. Practicing sessions were performed before tests to enable the consumers to familiarize with the use of the hedonic scale. Moreover, consumers were instructed to take a small bite of cracker first and after a sip of water. For the sensory analyses, the assessors were presented simultaneously peaces of randomly coded samples. The consumers were asked to evaluate colour, odour, consistency, drip and taste of products on the basis of a 9-point hedonic scale. Moreover, considering the weight of the single attribute on the global quality, each consumer was asked to give information on the whole quality of functional meat and fish-based products. A score equal to 5 represented the threshold for the acceptability.

Determination of health-giving compounds in the flour and after product cooking

Yellow peppers (1 Kg) were purchased from a local store. Each fresh pepper was immersed for 5 minutes in a 50% ethanol solution and then cut in half and immersed again in the same solution. Then, each half pepper was cut into small pieces and put to dry in an oven (CDL, Milan, Italy) at 35 °C for 72h. Well dried peppers were ground using a wiring blender (LB20EG, Waring commercial, Torrington, USA), to obtain a flour. The carotenoids were extracted as described by Sun and others [26,35] with slight modifications.

10 g of this flour were mixed with 50 mL of n-hexane 95% (Carlo Erba, Milan, Italia) and the mixture was gently stirring at 50 °C for 20 min. Then, it was centrifuged at 5000 × *g* for 10 min at 4°C (Eppendorf 5804 R, Italy) to obtain clear supernatant. The supernatant was transferred to a clean flask. The residue was mixed with another 50 mL of n-hexane 95% to repeat the extraction. The resulting supernatant was combined with the previous one. This operation was repeated 5 times. The extract was obtained after the n-hexane in the supernatant was completely evaporated under vacuum at 35 °C using a Rotavapor (BUCHI R-200). The residue was dissolved in 15 mL of n-hexane 95%

and then applied to the DSC-DIOL SPE 1 g cartridge (Supelco, Milan, Italy), which was activated with 5 mL of methanol (Carlo Erba, Milan, Italia) and 5 mL of n-hexane. β-carotene fraction was eluted with 10 mL of n-hexane by HPLC, while for the other carotenoids (capsanthin, lutein and zeaxanthin) acetone (Carlo Erba, Milan, Italia) was used. β-carotene, capsanthin, lutein and zeaxanthin standards were from Extrasynthese. Each eluted was evaporated to dryness and dissolved in 3 mL of dichloromethane and filtered through a 0.45 μm syringe filter (Teknokroma PTFE 0.45-μm) and then injected into the HPLC.

A HPLC 1200 (Agilent Technologies, USA) equipped with degasser (model G1379B), binary pump solvent delivery (model G1312B), auto sampler (model G1367B), column oven (model G1316B) and DAD system (model G1315C) was used. Samples were injected onto a reversed stationary phase column, C_{18} Aqua 5µ 200A (150 × 2.00 mm) and 5 µm particles diameter (Phenomenex, Milan, Italia). The injection volume was 10 μL for each sample. The composition of mobile phase was 30% ammonium acetate (Sigma Chemical e Co.) 1M in methanol (eluent A) and methanol (eluent B); the elution program was as following: at 0 min 5% B, 25 min 95% B, 40 min 95% B at the flow rate of 0.5 mL/min. The HPLC was controlled by the LC ChemStation 3D (Hewlett-Packard, USA) chromatography software. The wavelength was set at 450 nm for quantifying β-carotene, capsanthin, lutein and zeaxanthin. The concentrations were calculated using corresponding standard calibration curves and expressed as mg/Kg using their peak areas.

The same procedure reported above for carotenoide evaluation in the flour was also applied for the extraction and the separation of carotenoids in the burgers before and after cooking.

Statistical analysis

Experimental data were compared by one-way variance analysis (ANOVA). A Duncan's multiple range test, with the option of homogeneous groups (P 0.05), was used to determine significance among differences. STATISTICA 7.1 for Windows (StatSoft, Inc, Tulsa, OK, USA) was used for this purpose.

Results and Discussion

New functional foods were produced from the combination of minced meat or fish with extra virgin olive oil and vegetable flour. The products were developed by following different steps to balance the nutritional and sensory properties. For this reason, sensory evaluations were carried out in different sessions by trained assessors to define the approximate amounts of ingredients to be used. Afterwards, the final acceptance of the new functional burgers was assessed by consumer tests. Moreover, to verify the preservation of the health-giving compounds derived from the vegetable flour, analytical investigations were also carried out before and after cooking. In the following, results obtained for meat and fish patties were presented separately.

Functional hamburgers

Results of sensory evaluation carried out on minced patties enriched in oil-soaked-whey protein-foam are reported in Figure 1 for cooked products. Data underlined that, even if there is a peak at 10%, no statistically significant differences were recorded between samples in terms of whole quality. Slight differences were reported for specific product attributes as taste and juiciness (data not shown), being the samples with highest concentrations of oil more tender and not leathery. Evaluators judged with a similar intensity color, odor and texture, thus assessing final scores for whole quality very comparable to each other. The same happened with uncooked products. Very comparable results in sensory properties were also found for fermented sausages where pork back-fat was substituted with olive oil [36, 37]. Even though in the current study no significant differences were recorded, a slight better result was measured for hamburger with 10% of oil. For this reason, in the subsequent step, meat patties with vegetable oil at this selected concentration (10%) were used for the screening of flours.

In Figure 2 the comparison between samples can be observed. As one could infer, the combination of meat with flours was not always very prized. Taste and texture varied substantially between samples, thus affecting the final whole quality. The worst flour was that obtained from broccoli, that compromised all attributes of hamburgers, most

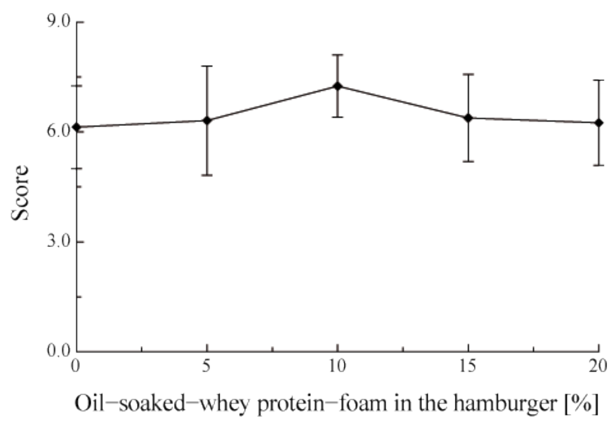

Figure 1: Score from sensory evaluation carried out on hamburgers enriched in different concentrations of oil-soaked-whey protein-foam.

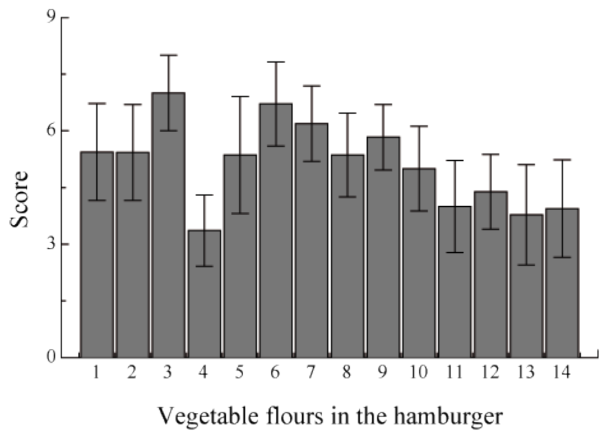

Figure 2: Score from sensory evaluation of hamburgers containing oil-soaked-whey protein-foam (10%) and different vegetable flours. (1) traditional hamburger with sole meat; (2) hamburger containing tomato flour; (3) hamburger containing red pepper flour; (4) hamburger containing broccoli flour; (5) hamburger containing pumpkin flour; (6) hamburger containing yellow pepper flour; (7) hamburger containing oil-soaked-whey protein-foam without flour; (8) hamburger containing zucchini flour; (9) hamburger containing fennel flour; (10) hamburger containing carrot flour; (11) hamburger containing cauliflowers flour; (12) hamburger containing asparagus flour; (13) hamburger containing eggplant flour; (14) hamburger containing artichoke flour.

probably due to the high content in glucosinolates of cruciferous vegetables [38]. Contrary, among the acceptable flours, the most interesting were represented by red and yellow peppers. The same trend was recorded with the corresponding uncooked hamburgers (data not shown).

For the subsequent steps, red peppers flour was taken into account. To assess the optimal combination of ingredients from a sensory point of view, meat, extra virgin olive oil and red pepper flour were varied according to a CCD and derived hamburgers were evaluated by a consumer test. In Figure 3 results obtained for the overall quality demonstrated that samples were very comparable and, even if no combination seems to be particularly interesting, all of them were acceptable. For this reason, another consumer test was performed with fewer samples. Each panelist was provided with two coded half hamburgers and was asked to order the two products according to the own preference. The two samples represented a common hamburger (neither oil nor flour) and the functional meat patty containing 10% oil and 10% of pepper flour, being the combination of the CCD that recorded the upper score. Results of this last test demonstrated that most of the consumers preferred the functional hamburgers, that were judged tender, more gradable to taste and very appetizing compared to the traditional hamburger (Figure 4).

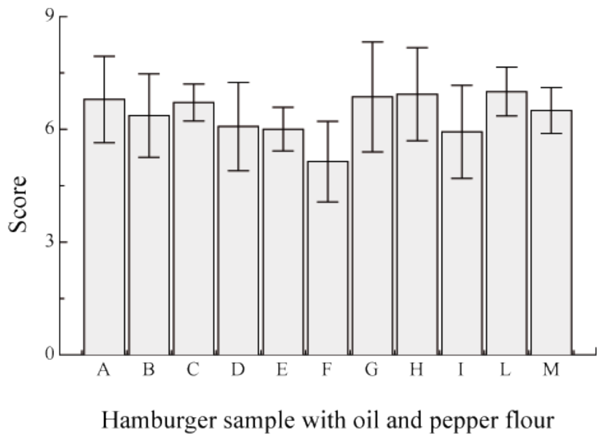

Figure 3: Score from sensory evaluation carried out on hamburgers enriched in oil-soaked-whey protein-foam and red pepper flour, according to a two-factor/five-level CCD. The letters correspond to the combinations reported in Table 1.

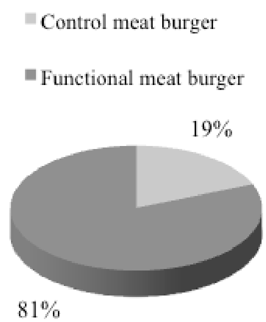

Figure 4: Results of consumer preference between a traditional burger and the new functional meat burger with olive oil and red pepper flour.

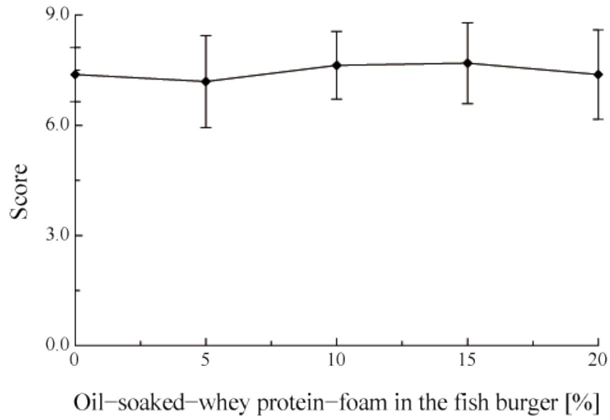

Figure 5: Score from sensory evaluation carried out on fish burgers enriched in different concentrations of oil-soaked-whey protein-foam.

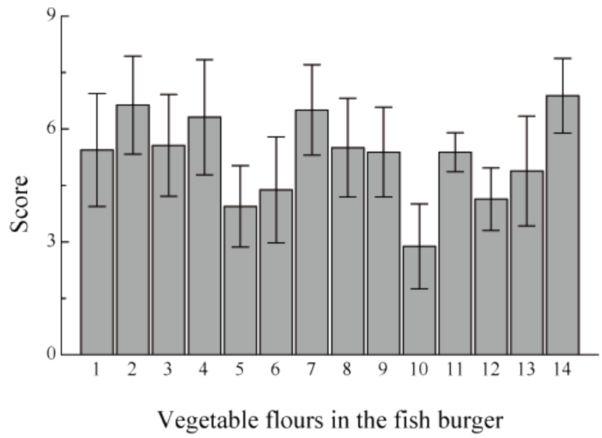

Figure 6: Score recorded by the sensory evaluation of the fish burgers containing oil-soaked-whey protein-foam (15%) and different vegetable flours. (1) traditional hamburger with sole fish; (2) fish burger containing zucchini flour; (3) fish burger containing asparagus flour; (4) fish burger containing oil-soaked-whey protein-foam without flour; (5) fish burger containing artichoke flour; (6) fish burger containing broccoli flour; (7) fish burger containing red pepper flour; (8) fish burger containing tomato flour; (9) fish burger containing pumpkin flour; (10) fish burger containing eggplant flour; (11) fish burger containing cauliflowers flour; (12) fish burger containing carrot flour; (13) fish burger containing fennel flour; (14) fish burger containing yellow pepper flour.

Functional fish-burgers

Panel test on fish burgers with different concentrations of oil-soaked-whey protein-foam highlighted that sensory properties of sea bass dough were improved by the new ingredients addition. Figure 5 reports data of cooked fish burgers global quality, as average between the specific scores given by the seven trained panelists for each attribute. While the uncooked products were very similar, cooked samples with 15% of oil received a slight higher score, even if no significantly difference between was highlighted. Juiciness and taste were the two attributes more appreciated at 15% (w/w) of oil, thus suggesting selecting 15% as the best concentration for the next steps.

The subsequent experimental trial was aimed to screen different vegetable flours. To this aim, several flours were separately added to fish burgers containing also 15% oil-soaked-whey protein-foam. While uncooked fish burgers seemed very comparable, some interesting differences were recorded between cooked samples (Figure 6). As can be inferred from the Figure 6, a few flours provoked complete unacceptability; most of them generated prized burgers. As in the case of hamburgers, also for the new fish formulation, pepper flours represented the most appreciated vegetable ingredients. In addition, flour obtained from zucchini also received a good score. Therefore, for the further product optimization yellow peppers flour and zucchini flour were both used.

In the next trial, consumer tests were performed to assess the best combination between sea bass, oil-soaked-whey protein-foam and flours. Hence, fish burgers with yellow pepper and zucchini flours were respectively prepared according to the CCD reported in Table 2. In Figure 7 the results of judges recorded with each flour were also reported. Even though some peaks in the graph appeared for both zucchini and yellow pepper flours, no statistically significant differences were underlined between samples, thus making no possible to determine the best formulation. Therefore, as in the case of the hamburgers, a different approach was adopted by carrying out a further consumer test. Each assessor was asked to indicate the preferred sample between a fish burger containing neither oil nor flour, and two functional fish burgers, containing the concentrations of oil and flour that recorded the upper score in the previous evaluation (see Figure 7). The concentrations are 15% oil and 5% yellow pepper flour and 17.5% oil and 7.5 zucchini flour. The last sensory evaluation assessed that fish burgers were ranked in the following order: burgers with pepper flour, burgers with zucchini flour and common fish burgers (Figure 8).

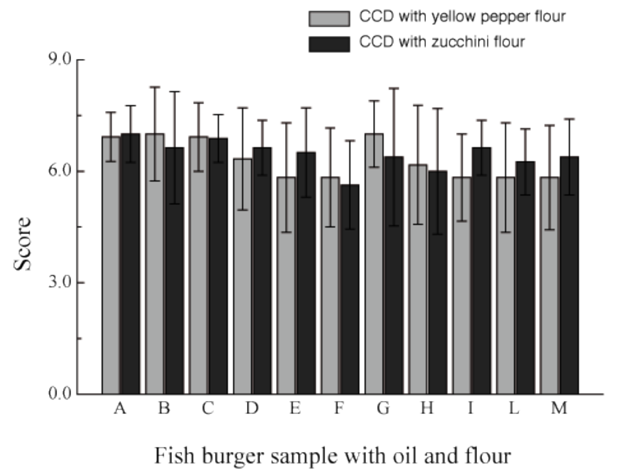

Figure 7: Score from sensory evaluation carried out on fish burgers enriched in oil-soaked-whey protein-foam, yellow pepper flour or zucchini flour, according to a two-factor/five-level CCD. The letters correspond to the combinations reported in Table 2.

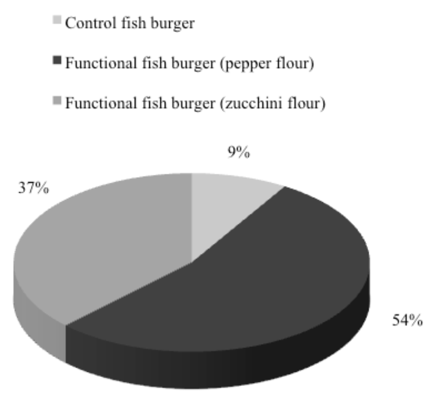

Figure 8: Results of consumer preference between a traditional fish burger and the two new functional fish burgers with olive oil and red pepper flour or zucchini flour.

	Zeaxanthin (mg/Kg)		Capsanthin (mg/kg)		Lutein (mg/kg)	
	Mean	SD*	Mean	SD	Mean	SD
Yellow pepper flour	90	1ᵃ	19	0.4ᵃ	4.0	0.3ᵇ
Hamburger before cooking	55	5ᵇ	19	4ᵃ	5.3	1.1ᵇ
Hamburger after cooking	63	13ᵇ	25	5ᵃ	11.6	3.0ᵃ
	Mean	SD	Mean	SD	Mean	SD
Yellow pepper flour	90	1ᵃ	19	0.4ᵃ	4.0	0.3ᵃ
Fish-burger before cooking	52	9ᵇ	21	2ᵃ	5.0	0.7ᵃ
Fish-burger after cooking	61	12ᵇ	17	4ᵃ	6.0	1.5ᵃ

**Data are presented as mean ± standard deviation (SD).*

Data with different superscripts in the same column are significantly different ($P < 0.05$).

Table 3: Analytical data of carotenoids in yellow pepper flour and in the burgers before and after cooking.

Health-giving compounds determination

Among the three vegetables used to prepare the new burgers (red pepper, yellow pepper and zucchini) the analytical investigation was focused on yellow pepper, as one of the notable vegetables with anti-oxidant properties [39-41].

Cooking process, while avoiding microbial hazards, results in certain changes of nutritional attributes and in particular, it can destroy thermolabile food components [42]. Therefore, to prove the preservation of the antioxidant content in the developed functional burgers after cooking, main carotenoids of pepper flour were monitored along the different steps of the production: in the flour, in the burgers before cooking and in the products after cooking. Table 3 shows the results obtained for carotenoids in the pepper flour and in each food matrix before and after cooking. It is worth noting that the amount of carotenoids recorded in each product after cooking are equal or higher than the level in the product before cooking, thus proving that thermal process did not affect the antioxidant content. The significant higher amount of carotenoids recorded in the samples compared to the quantity measured in the flour or fresh burger, could be ascribed with high probability to the variable capacity of the solvent to extract carotenoids in three different matrices; however, the lack of works dealing with the same topic does not allow to make further speculations, thus suggesting the need for additional investigations.

Anyhow, the results obtained in the current work confirm the healthy function of the developed products and bearing in mind the amounts of carotenoids recorded in each burger, a double advantage could be achieved. The consumption of these burgers could also represent an alternative solution to fruit and vegetables that generally represent the main sources for the daily intake of carotenoids but very often are not prized products [43].

Conclusions

New functional burgers based on meat or fish were developed. The added value of burgers derived from the beneficial effects of extra virgin olive oil and pepper flour that were added to the formulation in proper amounts, as assessed by trained panelists and generic consumers over several panel sessions. The results of the work demonstrated that the two ingredients, selected for their well-known heath benefits, can be incorporated in the dough without affecting product acceptability. The health effects can be expected also after cooking the products because results confirmed the presence of pepper flour carotenoids also in cooked burgers.

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