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Review

Optimising the use of proteins from rich meat co-products and non-meat alternatives: Nutritional, technological and allergenicity challenges

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ABSTRACT

An exponential growth in the global demand for high quality proteins over the next 20 years is expected, mainly due to global population growth and the increasing awareness toward protein rich foods for more nutritive diets. Coupled with this, is the pressing need for more sustainable approaches within a bio-economy mindset. Although meat production is expected to increase to address this rising demand, a better use of the currently available resources provided by the food, and specially, the meat industry is required. In this regard, despite the high-quality proteins and other nutrients found in meat co-products; they are currently underused and their valorisation needs to be revisited. Also, emerging protein sources need to be investigated to alleviate the environmental pressure coming from the meat industry. In this review, the main focus was attributed to (i) the current and forthcoming challenges for the use of meat co-products as meat replacers to produce a new range of meat derived products (with high nutritional value, improved technological properties and better consumer acceptance); (ii) their performance regarding to the non-animal origin proteins currently used as meat protein replacers; and (iii) the allergenicity of the proteins that might fall into the category of novel protein sources.

1. Introduction

Meat is considered an important source of high quality proteins which coupled with its flavour, aroma, and texture profile, places meat as one of the most demanded foods at global scale (Bohrer, 2017). Its high nutritional value derives from the presence of a high content of essential amino acids; as well as being a valuable source of vitamin B12, zinc, phosphorous and iron, but low in carbohydrates (Godfray et al., 2018; Mullen & Álvarez, 2016). Furthermore, the specific functionalities (such as structure, emulsifying, gelling or water holding capacity) imparted by the meat proteins are of interest for the food industry (Kumar et al., 2017). These techno-functional properties of meat proteins are involved in the overall characteristics of meat and meat products (i.e. appearance, texture and mouth feel) that are challenging to reproduce in non-meat proteins.

Meat consumption, per capita, in Europe is twice the world average (Milford, Le Mouël, Bodirsky, & Rolinski, 2019). From an EU perspective, statistic indicates that around 16% of the global meat consumption takes place in EU, where 6% of the global population lives. Based on historical data, average EU consumption of meat, dairy and fish has increased strongly over the last 50 years (Westhoek et al., 2011) and is expected to increase another 50% by 2030 due to population growth and high demand for protein-based food.. The total global consumption of meat is expected to increase by almost 70% between 2000 and 2030 (Milford et al., 2019) and by another 20% between 2030 and 2050. Ritchie and Roser (2017) mostly because of the demand predicted in developing regions to fulfil their nutritional needs. Compared to previous figures of meat consumption is clear that one of the main meat industry challenges is to supply this growing demand.

Along with the production of meat there is a concomitant generation of large amounts of meat co-products and processing streams, which, in the interest of sustainable, ethical and economic demands, require optimal management and end use (Lynch, Mullen, O'Neill, Drummond, & Álvarez, 2018; Mullen et al., 2017). In Fig. 1 can be observed how the increase in the livestock production, brings a concomitant increased offal generation; with increments in the range of 110–170% of livestock production. Henchion, McCarthy, and O'Callaghan (2016), pointed that a possible way to reduce the environmental impact of meat production could be to reutilize in an effective manner these co-products as food ingredients, for protein fortification, additives and especially as meat protein replacers, since functionality performance is similar to meat proteins. In recent years,

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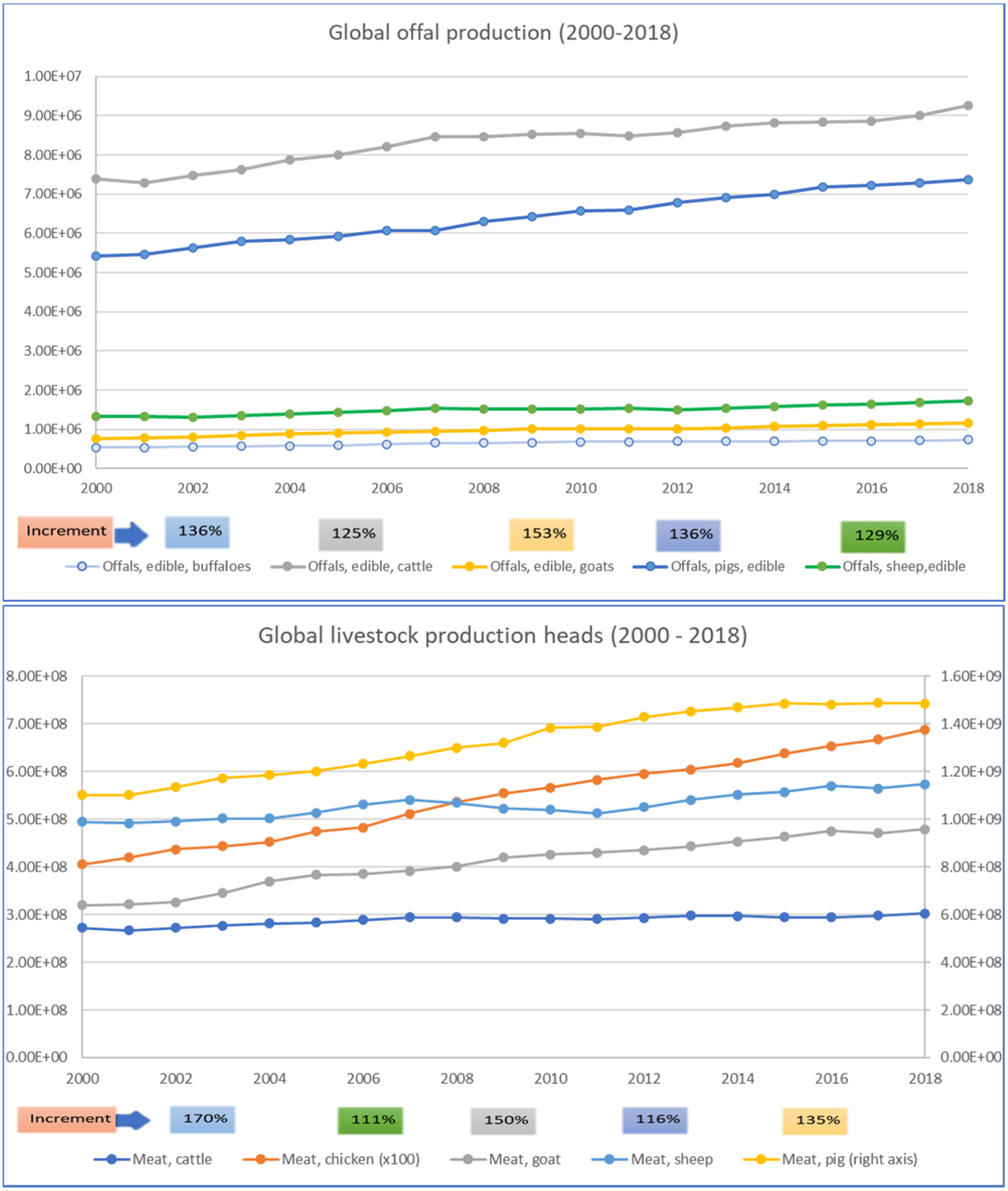

Fig. 1. Evolution of offal (tonnes) and livestock (heads) production at global scale from 2000 to 2018.
Source: FAOstat (http://www.fao.org/faostat/en/#home).

the interest in recovering proteins from meat co-products for the production of functional ingredients or bioactive peptides has increased (Aspevik et al., 2017; Lafarga, Álvarez, & Hayes, 2017; Toldrà, Parés, Saguer, & Carretero, 2020). In meat processing, the highest production of underutilised co-products and losses arise during slaughtering and carcass processing; however, the whole chain is subjected to the same hurdles, from meat processors through to meat product manufacturers and retailers (Spang et al., 2019). The amount of co-products produced depends on the animal type, and the specific index (ratio of live animal weight and weight of co-products generated) for slaughter houses is 0.56 for cow, 0.2 for pig and 0.1 for sheep (Russ & Meyer-Pittroff, 2004). Jayathilakan, Sultana, Radhakrishna, and Bawa (2012), found that 11.4% of the gross income from the beef industry, and 7.5% of the gross income from the pork industry, is generated by the re-utilization of meat co-products at all levels of the production chain. There is an ongoing pressing need to examine opportunities to extract further value from meat co-products (Mullen et al., 2017). These co-products are, depending on species, composed of a variety of organs, tissues and trimmings including hides, skin, feathers, hoofs, head, lung, tongue, heart, fat and meat trimmings. Also, other liquid streams (e.g. blood, drip loss, cook-out juices, glue water or stick water, brines and exudates) are a common co-products of meat processing. Various compounds of high nutritive value and/or valuable functional properties are present in these products and offer potential for revalorisation. For example, depending on the source, protein content and amino acid profile can be comparable, or even better, than lean meat (Alvarez, Drummond, & Mullen, 2018; Aspevik et al., 2017). These products can be processed to extract the target component and as co-products vary in terms of organic, mineral and biogenic matter, their extraction and purification will require different recovery strategies (Bustillo-Lecompte & Mehrvar, 2015). As with the consumption of meat, there are recommendations regarding daily intake values and consequently, attention should be also given to meat co-products in which high levels of fat and cholesterol are reported, as for example kidney or spleen (Mullen & Álvarez, 2016). Extracts from meat co-products can be processed in such a way that fat content can be almost negligible in the final extract, following several strategies such as pH shift (Li et al., 2018; Zou et al., 2018); enzymatic extraction (Chiang, Loveday, Hardacre, & Parker, 2019; Mora, Toldrá-Reig, Reig, & Toldrá, 2019) or emerging technologies, for instance PEF (pulsed electric fields), US (ultrasounds) or HHP (high hydrostatic pressures) (Borrajo et al., 2019).

However, meat co-products are not the only available alternative for meat protein replacement. There is increasing awareness towards more sustainable approaches to supply the global market with low footprint protein, through considering alternative sources. The reasons behind the interest in these alternative protein sources are various, as reported by Parniakov et al. (2018), especially those related with healthier diets and more sustainable production (low water/carbon foot printing). Plant-based proteins are of particular relevance, as they are considered to have lower production costs (Gorissen et al., 2018) and are readily available in undeveloped countries. Moreover, there is a wide variety of sources, such as legumes, oilseeds, cereals, and fungi, which provide alternatives for protein rich food products (Ganeshan & Chibbar, 2019). A further advantage is the suitability of the non-meat-based proteins for kosher and halal markets (Rahim, Muhammad, & Hassan, 2017). However, allergies are a main concern, since many people have been found to be allergic to soy proteins, cereals and other potential replacers. Furthermore, replacing meat proteins with non-meat alternative sources needs to address the technological challenges faced while trying to mimic meat proteins behaviour in food matrices, and to ensure that consumer demands and expectations are met.

Therefore, the aim of this review is to provide an updated overview on the use of either meat co-products or alternative non-animal protein sources as meat replacers. As well as to discuss the challenges for optimal utilisation of natural resources (livestock), mitigating the environmental impact derived from the projected increase in meat production and responding to consumers' needs for high quality protein rich products.

2. Meat co-products, current uses

Despite the fact that the main component of meat co-products, excluding water, is protein with a high percentage of essential amino acids, they are underused as protein sources for human consumption (Mullen et al., 2017). Nevertheless, some co-products have an important role in the diet in certain countries of the world. However, as previously mentioned, due to the fact that such products usually have a high content in cholesterol and saturated fatty acids (specially brain, kidney or spleen), it is recommended to restrict its consumption according to a healthy diet observing recommended daily intakes as it is done for red meat consumption. Liver, heart, kidney, tongue, thymus, brain and tripe are the most consumed animal co-products; and some even command a moderate market value on their own in specific regions. For example, tongue can reach an appreciable value of 12 €/kg, or cheek meat 6 €/kg; while others as lung (1.5 €/kg) or heart (1.2 €/kg) possess very low market value (Co-product Market Report, 2017). There is a strong linkage between culture, tradition and consumer demands with the different use of co-products for direct consumption. For instance, those that are judged as inedible in one country can be considered as delicacy products in others. Regardless of the increase in meat consumption, there has been a decrease of meat co-products consumption in the recent years, which have a negative impact on the greenhouse gas (GHG) emissions. According to Xue et al. (2019), re-utilising the 50% of the current offal production as food, will reduce the GHG emissions by 18.8%. Consequently, the disposal, processing and commercialisation are becoming a challenge for the overall meat industry. For many co-product producers, the easy route is to categorize them as for non-human consumption since the regulations are less strict; even though the economic profitability they can obtain is not substantial. Depending on the country, meat co-products are often used as ingredients for pet food, animal feed, and in very low amounts for other uses as in pharmaceutical industry, or biomedical and other industrial applications with some going for landfill (Lynch et al., 2018). It is the point of view of the authors of this review that meat manufacturers are the providers of animal origin protein to fulfil the dietary requirements of the global population. In this sense, such high nutritive value proteins, found in co-products, must be harnessed and used in food products to keep up with the increasing global protein demand; even more considering that the cost of producing one unit of meat is almost equivalent to the cost of producing the same weight of the fifth quarter. In this sense, it has been reported that the cost of dietary energy density of raw material varies from 90 to 780 kcal/ 1US$ (beef flank and turkey thigh respectively) for meat products. The average values for beef and pork were estimated to be 120 kcal/1US$ and 250 kcal/1US$ respectively. For the same cost (1 US$) it is possible to generate 644 kcal from peanuts, 780 kcal from chicken eggs, or 1000 kcal from beans (Bohrer, 2017). As these products are produced alongside meat production the production cost has already been somewhat carried by the meat production chain. Certainly, there will be some additive additional cost to making co-products ready to use by industry or consumers, however given the volumes under consideration and the global need for protein there are clear gains to be capitalised upon for such protein rich products. Depending on the source co-product and the market under consideration the co-product may be used in its entirety e.g. kidney, heart, tongue etc. However, another possible avenue of use is when protein rich extracts are prepared and this is used as a component in a final product. One school of thought suggests this approach helps remove the 'yuck' factor which some consumers have for offal etc (Henchion et al., 2016). These products can be used in conjunction with meat proteins to provide additional protein in the end product assuming the appropriate legislation is followed (see below) and levels of inclusion do not have any negative impact on sensory or technological aspect of the end-product. With this in mind, a number of studies have been focused on evaluating the behaviour of the meat co-products as meat replacers or as protein enhancers.

3. Technological impact of meat co-products as replacers in meat products

Proteins recovered from meat co-product have different physicochemical characteristics, amino acid profile and functional properties compared to meat proteins, and therefore, it is crucial to assess the impact of their inclusion or use as replacers on the final product characteristics (summarised in Table 1). Several factors need to be taken into consideration while using meat co-products, and in general any protein source, as meat protein replacers:

– Amino acid profile: from a nutritional point of view it is important to ensure that the nutritional value is not compromised. For example, collagen rich extracts are not valuable sources of essential amino acids; meanwhile on the other hand, plasma has a higher content of essential amino acids compared to meat (Mullen & Álvarez, 2016). Meat cuts generally reach Protein Digestibility-

Table 1
Main properties and characteristics of meat co-products used as meat replacers.

Co-product	Protein (%)	EAA (%)	Fat (%)	Nutrients	Properties	Utilization
Blood	18.5	58.0	0.4	Fe	Emulsifying, stabilizer, clarifier, colour additive, water and fat binder	Blood sausages, blood pudding, biscuits and bread
Brain	10.3	46.8	9.2	Melatonin hormone vitamin A, carotene and cholesterol, Ca and P	Emulsifying used in cosmetics	Sausage ingredient, broiled, braised and cooked in liquid, poached, scrambled
Ear	22.3	25.6	15.5	Collagen	Gelling, water and fat binding, emulsifying, stabilizer and film forming	Smoked and salted, stewed with feet
Feet	21.2	21.6	22.0	Collagen	Gelling, water and fat binding, emulsifying, and stabilizer	Jelly, pickled, cooked in liquid, boiled, fried
Hides	30–35*	–	2.5–3-.0*	Collagen	Gelling, water and fat binding, emulsifying, and stabilizer	Athletic equipment, reformed sausage casing and cosmetic products, sausage skins, edible gelatine and glue
Heart	17.0	47.7	4.4	Niacin and vitamin B12, Fe	Viscosity and creaminess	Braised, cooked in liquid, luncheon meat, patty, loaf
Kidney	15.3	48.0	3.2	Vitamin A, B12, B6 and folacin, Fe, Cu	Mild water binding	Broiled, cooked in liquid, braised, in soup, grilled, in stew
Liver	19.0	48.9	3.7	Vitamin A, B2, B12, B6, niacin, folacin, ascorbic acid, heparin, Fe, P, Zn, Cu	Mild water binding and good cooking yield	Braised, broiled, fried, in loaf, patty and sausage
Lung	15.0	37.8	2.7	Ascorbic acid and vitamin A, collagen, Fe, Na, K	Emulsifying and foaming	Blood preparations, pet food
Spleen	17.9	41.9	2.6	Ascorbic acid and cholesterol Fe, K	Mild emulsifying	Fried, in pies, in blood sausage
Tongue	16.3	40.2 17.2		Carbohydrates, Ca		Cooked in liquid, cured, sausage casing, sausage ingredient
References	(Li, Yang, & Li, 2008; Mullen & Álvarez, 2016)			(Duarte, Carvalho Simões, & Sgarbieri, 1999; Jayathilakan et al., 2012; Jiménez-Colmenero & Cassens, 1987; Mullen & Álvarez, 2016; Ünsal & Aktaş, 2003)	(Darine, Christophe, & Gholamreza, 2010; Devatkal, Mendiratta, Kondaiah, Sharma, & Anjaneyulu, 2004; Ejike & Emmanuel, 2009; Ionescu, Aprodu, Darabă, & Porneală, 2008; Jayathilakan et al., 2012; Nuckles, Smith, & Merkel, 1990; Rivera, Sebranek, & Rust, 2000; Silva & Silvestre, 2003)	(Gómez-Guillén, Giménez, López-Caballero, & Montero, 2011; Jayathilakan et al., 2012; Ockerman & Hansen, 1999)

Corrected Amino Acid Score (PDCAAS) scores from 0.9 to 1.0 which is considered high or very high quality (Boateng, Nasiru, & Agyemang, 2020). In case of meat co-products, the amino acid composition is variable, as shown in Table 1. Thus, by creating new formulations, tailored blends of different co-products can fulfil the requirements of essential amino acids, even if collagen is used in it. Recent attempts have assessed that blends of different co-products (tripe, ears, heart or lips) can increase the biological value by balancing the content of essential amino acids reaching values of 78% of "in vitro" digestibility (Vietoris et al., 2019). From a sensory point of view, the abundance of hydrophobic amino acids (as happens with haemoglobin), can impart bitter taste to the final product; which may need to be masked.

– Impact on the most relevant technological properties: depending on the final product, the requirements might vary, but in general terms the most essential properties from a commercial perspective include emulsifying capacity and emulsion stability, gelling ability, water holding capacity and oil holding capacity. Such functionalities will provide improved product stability, a better cook yield, less thaw and drip losses and hence, increased profitability. Also, the presence of pro-oxidants needs consideration as these can lead to an increase in oxidation. For example, haemoglobin and derivates are considered as pro-oxidants, which can influence product stability and can impart a rancid flavour. Also, texture can be manipulated: even though collagen and plasma increase hardness, after hydrolysis of these proteins their use can result in softer products. This can be attributed to the fact that less protein-protein interactions are taking place and therefore the gel structure is weakened. Finally, shelf-life and microbial stability should be analysed in order to assess the safety of the new products.
– Percentage of replacement: as it will be discussed later in this review, the replacement level is essential in creating new formulations. It is of the upmost importance to find the balance between replacement level and final properties of the end product.
– Safety: to comply with all regulation regarding collection, processing and storage of meat co-product intended for human consumption.
– Consumer acceptance: the consumer has to be informed about the type of meat co-product employed, which has to be properly labelled according to the legislation. Rejection might be a main challenge since these products may impart a strong flavour, darker colour; or even more, they can be perceived as low-quality protein. Trying to overcome this last factor, it is important that the consumer becomes well informed about protein quality and how co-products can play a role in their diets.

Table 1 summarises the main meat co-products that can be used as meat replacers, including the nutrients that will be incorporated to the food product, the main functionality imparted by the co-product and in which end-products they have been tested.

Pork head meat (which is composed of 21% tongue and 79% boneless meat) was used to replace pork meat in frankfurters with inclusion levels of up to 10% showing similar characteristics to a control formulation and levels above this leading to negative product quality (Choi et al., 2016a,2016b). Nevertheless, higher replacement levels of pork meat head (20%) increased the cook loss and the emulsion instability; whereas frankfurters formulated with 10% of pork head meat showed similar properties than the control. However in patties replacement at levels up to 20% had a negative impact on many product characteristics (Choi et al., 2016a,2016b). In a more recent study (Alvarez et al., 2018), proteins recovered from two co-products (blood and meat exudates) and two processing streams (brines and glue water from rendering) were employed to investigate the effect of 10% and 20% replacement level in a model meat system: Irish breakfast type sausages. It was found that each one of the replacers employed had different effect on cook loss, texture profile and colour; compared to controls. In general, 10% replacement level did not exert a significant impact. It was suggested that tailored blends of such ingredients can be designed in order to impart specific properties to the final products. The use of blood as protein replacer has been investigated in sausages at different levels (15, 20, 25 and 30%) (Choi et al., 2015). It was observed that 20% of replacement improved the quality characteristics of the final products. A fraction of the blood, plasma, has been employed as phosphate replacer, based on its excellent water holding capacity (Hurtado et al., 2011). Overall, no impact on technical properties was observed, and the final product was generally well accepted by the panelists but off-flavours were detected.

Meat co-products have also been employed as proteins extenders; in this case, lean protein is not replaced, but meat co-products proteins are added to the formulation to increase the protein content. The effect of the addition of collagen protein from pork in sausages and burgers was investigated (Carvalho, Milani, Trinca, Nagai, & Barretto, 2017; Lee & Chin, 2016). In both products, the additional collagen significantly improved the cooking yield and water holding capacity; besides a slightly firmer structure resulted at high level of collagen addition.

Mechanically deboned poultry meat (MDPM) has been reported as a raw material coming from poultry industry. According to Massingue et al. (2018), it can be used as a source of valuable functional and nutritional proteins. The percentage of replacement of MDPM is an important factor that might influence the mechanical properties of the sausages (Daros, Masson, & Amico, 2005). Different levels (0, 20, 40, 60, 80, and 100%) of replacement significantly altered the structure of the final product. The structure of the sausage maintained its integrity up to 40%, but at 60% the structure was compromised; with a higher negative impact at 80% (weak structure to resist the normal packaging). Also, the texture was affected negatively with replacements levels higher than 60%. The effect of replacing mechanically deboned chicken meat with its hydrolysate (10, 20 and 30%) in mortadella-type sausages was analysed; it was found that higher level of replacement lead to softer, more oxidised and darker product, so a threshold of 10% of replacement was found as the optimum one (Cavalheiro et al., 2014). MDPM was also employed as ingredient in lamb and mutton sausages (Massingue et al., 2018); results showed that best results were obtained where MDPM was employed at a level of 30% in the batter. As in previous reports, a softer texture was found at higher inclusion levels; whereas oxidation, pH or water activity were not affected. A possible solution to avoid these issues in the textural properties might be the simultaneous addition of deboned chicken meat with collagen fibres. In this case, the negative effect of the MDPM were minimized on cooking yield, hardness and lightness of the final product (Pereira et al., 2011).

A further ultilization of poultry co-products is as fat replacers. The effect of duck feet gelatine in low-fat frankfurters lead to higher textural parameters, and similar satisfaction scores to control (20% back fat) were obtained when 5% back fat – 15% duck gelatin were used (Yeo et al., 2014). The replacement of pork back fat by bovine heart surimi-like materials resulted in the increase of hardness, whereas no differences in color, odor, or tenderness were observed by panelist at 20% and 40% of fat replacment (Seo, Yum, Kim, Jeong, & Yang, 2016). The increasing level of hydrolysed collagen (25%, 50% and 75%) as fat replacer in frankfurters improved the water holding capacity, stability and texture, and no differences in the overall acceptance were observed (Sousa et al., 2017).

Because of the different characteristics and properties of each single co-product, there is no general trend regarding the impact of the co-product replacement/addition in a meat-based product. Nevertheless, some conclusions arise if the collagen content is considered. Generally, a higher content of collagen-type replacers leads to harder and brighter products with lower cook loss; whereas non-collagen-type replacers generate softer, darker and with higher cook loss products. Moreover, in terms of texture, it was observed that mechanically deboned meat additions result in a decrease in the shear force, compared to collagen rich co-product that create the opposite effect on the final product. In general terms, a 10% of replacement level has not an impact on the technical properties of the final product; whereas higher replacement levels generate opposite results depending whether the replacer employed is collagen rich or not. Also, when protein hydrolysates are used as replacers, there is a marked trend towards obtaining softer products, showing higher cook loss and, in general, poor performance. As an exception, collagen hydrolysates, could have the opposite effect, as long as the degree of hydrolysis is not excessive, allowing the collagen to act as a texturizing and gelling agent. In order to expand the use of meat co-products as meat replacers or protein extenders, using tailored blends of collagen rich extracts combined with extracts low in collagen seems to be a promising approach.

4. Alternative protein sources for meat protein replacers and analogues

Despite meat is an excellent source of high biological value protein there has been increased interest and demand for reduced-meat or non-meat alternative products. Various values have driven this demand and include for example sustainability, health and animal welfare. It has been estimated that the percentage of people choosing not to consume meat ranges from 2 to 10% in developed nations (Bohrer, 2017). As well, added to those consumers choosing not to eat meat, there are others who either are looking to a flexitarian diet or looking for products with reduced animal protein content (i.e. increased plant protein) (Kemper, 2020). In this sense, novel products must be designed to address consumers' demands and fill the market gaps. A key challenge in this regard is in developing a meat-like end-product, or meat analogue, is emulating meat texture and flavour. Legumes and oilseed proteins have high protein content and, with due regard paid to anti-nutrients in these sources, are often considered. One of the more promising approaches is to employ texturized plant-based proteins, which have been considered as a suitable alternative to animal proteins for many decades for particular types of products. Meat analogues share some similarities with meat e.g. appearance, colour, flavour and texture while hydrated or cooked.

Differently, the addition of texturized vegetable proteins as extenders is of benefit for those consumers willing to reduce their meat consumption. Plant protein based extenders will improve the overall functional properties of the final product such as water binding, fat emulsification and texture (Smetana et al., 2018). However, these properties are not displayed if the extenders are hydrated or cooked on their own. Moreover, proteins from vegetables are appropriate for consumption in food preparations, because of their structural integrity, which withstands hydration, cooking and other usual food processing procedures.

Table 2 illustrates the most significant high-quality protein products used in food industry as meat extenders or analogues. Particularly soy, legume, oilseed proteins, cereal proteins, and mycoproteins were taken into consideration. Whey proteins have been included for comparison purposes, since they are widely employed in the food industry as protein fillers and a high quality protein source, as shown by the high PDCAAS (Protein digestibility-corrected amino acid score) (> 1) (Hess & Slavin, 2016). For each class, the protein content, main nutrients, and functional properties, as well as the advantages and disadvantages of their use, incorporation/replacement level and the possible solutions to overcome the negative aspects have been included.

In summary, soy, mycoproteins and whey proteins had a PDCAAS equal or even better to that reported for meat (0.92); contrarily to legumes, oilseed and cereal proteins, (0.40–0.70) (Bohrer, 2019). The lack of some essential amino acids lowers the PDCAAS value for cereal proteins. For this reason, some meat replacers can reduce the nutritional value of the final product, when compared to the original one. Nevertheless, combinations of plant proteins can reach higher PDCAAS values, as for example grains and legumes or grains, nuts and seeds (Brennan, Brennan, Mason, & Patil, 2016).

From nutritional point of view an added advantage is that soy, legume and cereal are beneficial source of fibres, which are totally absent in meat. It is commonly known that plant-based products contain anti-nutritionals which are naturally synthesized by plant such as phytates, tannins, lectins, oxalates, etc. The downside of these are the effects generated in the nutrient's absorption, like reducing nutrient intake, digestion, and utilization and may produce other adverse effects (Popova & Mihaylova, 2019). A part from lack of antinutrients, it is well established that meat is a valuable source of minerals and vitamins. Sources such as soy, legume, mycoproteins and whey proteins also demonstrate high nutritional profiles in this regard. Additionally, from the techno-functional point of view soy, cereal and whey protein demonstrated water and fat binding capacity. Moreover, whey, soy and legume showed emulsifying properties, contrary to cereal proteins. When plant protein are intended for use as meat analogues, soybean, rapeseed, pea and chickpea are the ones with the best functional properties in terms of texture, water and oil binding capacity, and elastic gel formation (Jones, 2016). However, the functionality of plant derived proteins is still poorly understood, as well as their impact when introduced in meat formulations.

If added to products, plant-based proteins can perform very well as water binding agent, resulting in a reduced cook loss compared to controls, as reported in Table 1. A comparison about the effect of pea protein, rice protein and lentil flour on fortified and restructured beef steaks was carried out (Baugreet, Kerry, Allen, Gallagher, & Hamill, 2018). It was found that as the protein content was increased, by increasing plant protein content, resulting products had reduced cook loss and increased binding strength and textural properties (hardness, chewiness, cohesiveness and gumminess); with no impact on the colour of cooked product. When the same proteins were employed to fortify beef patties, a similar trend was observed in terms of textural properties, and improvement in cook loss. However, lentil fortified patties were found to be softer than standard beef patties, probably because of the lower protein content of this raw material. Interestingly, rice proteins significantly increased redness values (Baugreet, Kerry, Botineştean, Allen, & Hamill, 2016).

In recent years, the use of novel protein sources in human diet has become more appealing. The most emerging sources of high-quality protein sources, from a nutritional point of view, are algae (distinguished as microalgae and seaweeds) and insects. Particularly in the case of microalgae, *arthrospira* (usually denoted as *spirulina* in the market) (blue-green algae) has been used in food industry for years (Saranraj & Sivasakthi, 2014). Microalgae could have the potential to become a sustainable animal-based protein because some species are a valuable source of high-quality proteins with balanced amino acid profile (Caporgno & Mathys, 2018). The protein content of *spirulina* ranges from 60 to 70 wt% (Saranraj & Sivasakthi, 2014) and it is also a valuable source of vitamins, essential amino acids, minerals and essential fatty acids (Sotiroudis & Sotiroudis, 2013). Both *chlorella* and *spirulina* were used in developing chicken Roti recipes (Parniakov et al., 2018). Inclusion of algae protein was reported to improve the amino acid profile since the detection of higher proportions of essential amino acids. Despite this, it has been observed that this has generated softer products, compared to those formulated with soy proteins. On the other side, the acceptability of recipes including algae was reported to be lower than when soy, lentil, pea or beans were employed, possibly due to the effect on flavour and colour. The effect of three seaweeds extracts (wea spaghetti (*Himanthalia elongata*), wakame (*Undaria pinnatifida*), and nori (*Porphyra umbilicalis*)) on meat emulsions was studied (Cofrades, López-López, Solas, Bravo, & Jiménez-Colmenero, 2008). Surface colour was altered with reduced lightness, redness in all cases, and increasing yellowness when wakame was employed. Emulsion stability, measured as water and fat release after cooking, was greatly improved at both 2.5 and 5% inclusion level, with the exception of nori at 2.5% which had a negative effect. In this case, the improvement was attributed to both protein and fibre present in the seaweed extracts.

Table 2
Most relevant sources of proteins employed as meat replacers or analogues.

	protein % PDCAAS	Nutrients	Functional properties	Advantages	Disadvantages	Solution	Product replacement
Legume (lentils, beans, mung beans, chick beans and peas)	20–30% 0.70	Source of energy, minerals and B vitamins, carbohydrates and fibres iron (Fe), calcium (Ca), zinc (Zn)	Emulsifying and foaming properties, which can be enhanced by acylation	Isolates with low cost, high protein content and wide acceptability	Low in sulphur-containing amino acids, Contain antinutrients like protein inhibitors, lectins, polyphenols and phytic acid	Supplementation with cereals, several strategies to remove and inactivate nutritional factors in seeds (dehulling, soaking, cooking and thermal treatment)	Meat balls, sausages and meat extenders
References	Riascos, Weissinger, Weissinger, and Burks (2010), Bhat and Karim (2009), Serdaroğlu, Yıldız-Turp, and Abrodímov (2005), Lawal (2005), Klupšaitė and Juodeikienė (2015), Suárez-López, Kizlansky, and Lopez (2006), Strahm and Harrison (2006), Sánchez-Chino, Jiménez-Martínez, Dávila-Ortiz, Álvarez-González, and Madrigal-Bujaidar (2015)						
Oilseeds proteins (Safflower, cottonseed, rapeseed, peanut and tree nuts)	13–17% safflower, 23% cottonseed 25% rapeseed and peanuts 0.86 (canola seeds)	Source of antioxidant tocopherols and carotenoids, iron (Fe), copper (Cu), Calcium (Ca), Magnesium (Mg), and polar lipids, source of fibres	Viscosity increase with increasing of protein concentration, good solubility, no good foaming properties	Low level of unsaturated fatty acids, vitamins	Low in sulphur-containing amino acids, Contain antinutrients like protein inhibitors, lectins, polyphenols and phytic acid	Supplementation with cereals or mycoproteins, several strategies to remove and inactivate nutritional factors in seeds (fermentation soaking, germination and thermal treatment)	Meat extenders
References	Prakash and Narasinga Rao (1986); Sunilkumar, Campbell, Puckhaber, Stipanovic, and Rathore (2006), Moure, Sineiro, Domínguez, and Parajó (2006), Serraino and Thompson (1984), Sarwar (2013), Sharma, Su, Joshi, Roux, and Sathe (2010), Strahm and Harrison (2006), Riascos et al. (2010), Bhat and Karim (2009), Lawal (2005), Singh and Bhalla (2008), Sánchez-Chino et al. (2015)						
Soy (flour, concentrates, isolates)	35–40% 0.95–1.00	high quality protein, fat, low content of saturated fat, carbohydrates, rich in fibres, iron (Fe), calcium (Ca), zinc (Zn) and B vitamins	Water binding ability, fat binding ability, emulsion stability, increase yield, elasticity, colour control, decrease cooking loss	Low cost, source of high-quality proteins, well balance composition of amino acids	Strong off-flavours associated with saponins and isoflavones	Development of soybean cultivars with low undesirable flovours	Argentina sausage “Chorizo”, frankfurters, pressed loaves and poultry rolls
References	Golbitz and Jordan (2006), Singh, Kumar, Sabapathy, and Bawa (2008), Lindsay and Claywell (1999), Sun, Li, Li, Dong, and Wang (2008), Porcella et al. (2001), Katayama and Wilson (2008), Vallejo-Cordoba, Nakai, Powrie, and Beveridge (1987) Bhat and Karim (2009), Serdaroğlu et al. (2005)						
Cereal proteins (wheat, maize, barley, rice, oats, rye)	Wheat (8–17.5%), maize (8.8–11.9%), barley (7–14.6%), rice (7–10%), oats (8.7–16%), rye (7%–14%) 0.59	Good source of carbohydrates, linoleic, oleic and palmitic acids, source of fibres	Film forming, adhesive properties, increase water holding capacity and decrease cooking loss	Wheat gluten an economically co-product in the recovery of wheat starch, improvement of the rheological properties of flours and meat products	Low protein contents and imbalanced essential amino acids composition, lower water solubility of wheat gluten limits its potential as food ingredient	Mixture of legume proteins and cereals provide a balance mixture of amino acids, limited hydrolysis improves gluten solubility	Smoked sausages with mechanically separated poultry meat, frankfurters, ground meat patties, sausage products
References	Guerrieri (2004), Zhang, Xiao, Samaraweera, Lee, and Ahn (2010), Singh et al. (2008); Xiong, Agyare, and Addo (2008)), Kong, Zhou, and Qian (2007), Asgar, Fazilah, Huda, Bhat, and Karim (2010), Wondimu and Malleshi (1996), Agyare, Xiong, and Addo (2008), Li, Carpenter, and Cheney (1998), Gnanasambandam and Zayas (1992), Suárez-López et al. (2006)						
Myco-proteins From *Fusarium venatum*	Freeze-dried powder 11.8% frozen chicken style chunks 42.9% 0.91	Low sodium content, good source of zinc (Zn), selenium (Se), calcium (Ca), iron (Fe), copper (Cu), good source of essential amino acids and vitamins (thiamin, riboflavin and niacin)	replacement does not affect the appearance, flavour, texture and aroma	Presence of all the essential amino acids, low in saturated fatty acids, potential, valuable supplement of cereal-based and legume-based diets, approved by Halal Food Authority	Harvested hyphae have similar morphology to animal muscle cells but they need to be held together like the connective tissue in muscle cells, morphological changes from mutants over the parental strain,	Fungal biomass is mixed with a binding agent (egg albumin) to make a meat product texture, application of low dilution rates, changes in the nitrogen source or decrease the selected pressure	Fillets, cold-cut style slices, nuggets, burgers, sausages, pastries and pies
References	Rodger (2001), Miller and Dwyer (2001), Denny, Aisbitt, and Lunn (2008), Asgar et al. (2010), Turnbull, Leeds, and Edwards (1990), McIlveen, Abraham, and Armstrong (1999), (Weiss, Gibis, Schuh, and Salminen (2010); Wiebe (2002)), Burley, Paul, and Blundell (1993)						
Whey	34, 50 and 80% 1.00	Good source of essential amino acids in particular sulphur amino acids, phosphorus (P), potassium (K), sodium (Na), calcium (Ca) and magnesium (Mg)	Gelling, foaming, water binding properties, Improvements in colour, emulsion formation, reduce cook loss and springiness, increase brittleness	A by-product from cheese makers and casein manufacturers, cost-effective unit processes for concentration, transformation, fractionation and dehydration of whey	Lactose intolerance, imbalance mineral in the bones (loss of bones density), increase pH of blood and problems in kidneys metabolism with high amount of whey	Soybean proteins can be a substitute	Frankfurter-type sausages, poultry raw and cooked meat butter
References	Riedel (1994), Yetim, Müller, and Eber (2001), Hongsprabhas and Barbut (1999), Shoveller, Stoll, Ball, and Burrin (2005), Smithers (2008), Gangurde, Patil, Chordiya, and Baste (2011), Tunick (2008)						

PDCAAS: Protein Digestibility Corrected Amino Acid Score.

Researchers reported that even at low salt content (where emulsion tends to be weaker) the hardness of the product could be maintained by adding these extracts.

The most consumed types of insects are mealworms, buffalo worms and locust, which they are usually freeze-dried and marketed via wholesale (Gravel & Doyen, 2019). Insects are a sustainable source of proteins with low production of greenhouse gases and ammonia compared with conventional livestock (van Huis, 2016). Moreover, they are valuable sources of essential amino acids and micronutrients like iron and zinc. However, to the best of our knowledge, very few reports employing insect proteins as meat replacers have been published. A recent paper reported that black soldier fly larvae was employed as ingredient in Vienna sausages formulations (Bessa, Pieterse, Sigge, & Hoffman, 2019). Authors reported that, sausages made with a 28% of insect protein behave similarly to control in terms of texture and proximate analysis. Another recent paper (Azzollini, Wibisaphira, Lakemond, & Fogliano, 2019), describes the impact of temperature and $CaCl_2$ concentration on protein-protein interaction of insect protein to form gel-like structures with a view to be used as meat analogue. It was concluded that final textural properties can be tailored by adjusting protein processing parameters. High moisture extrusion was employed to prepare meat analogues from *Alphitobius diaperinus* (tenebrio) combined with soy protein and fiber (Smetana et al., 2018). Results proved that a blend of insect protein (40%) combined with soy dry matter (60%) generated a product with a texture similar to meat products. Several efforts are underway to examine more closely the opportunities raised from insect production for food uses. Current research is focused on increasing utilisation of insects as food protein source and on the economic viability of the insect value chain and potential opening markets (Lombardi, Vecchio, Borrello, Caracciolo, & Cembalo, 2019).

5. Allergens in meat protein replacers

When employing alternative protein sources to be used either as meat extenders or meat analogues, it has to be considered the allergens that might be present. Overall, the best way to manage an allergy is to avoid consuming the products triggering the allergic reaction. Therefore, despite the important nutritional and technological role played by nonmeat protein replacers, it is essential to inform the consumers through adequate and complete labelling, and if possible finding an "allergic-free" alternative, as if offered by most meat processing co-products.

Food allergy is defined as an adverse reaction of the human immune system to an otherwise harmless food component (Verhoeckx, Broekman, Knulst and Houben, 2016). Allergic reaction to food can affect the skin, the gastrointestinal tract, the respiratory tract, and, in the most serious cases, the cardiovascular system (Brotons-Canto et al., 2018).

5.1. Meat

Allergy to meat has been traditionally been considered infrequent and, unlikely many other alternative protein sources detailed below, there is no allergenic status attributed to meat proteins. However, recent studies are beginning to shed some light on potential allergies. For example IgE-mediated hypersensitivity to meat products (Wilson & Platts-Mills, 2018), and identification of pork-cat syndrome and delayed anaphylaxis to red meat (i.e. the α-Gal syndrome) (Fischer, Yazdi, & Biedermann, 2016; Hilger et al., 2016; Wilson, Schuyler, Schroeder, & Platts-Mills, 2017).

5.2. Legumes

Although legumes are rich sources of proteins, vitamins and fiber, they are also sources of food allergens (Foschia, Horstmann, Arendt, & Zannini, 2017; Sicherer & Sampson, 2014). Legume allergies are some of the most common food-related allergies. Few examples are lentil (*Lens culinaris Medikus*), chickpea (*Cicer arietinum L.*), black gram (*Vigna mungo (L.) Hepper*) and lupin (Lupinus) (Brotons-Canto et al., 2018; Cabanillas, Jappe, & Novak, 2018; Chan, Greenhawt, Fleischer, & Caubet, 2019). Lupine allergies frequently cause acute and severe reactions including anaphylactic shock and fatality. Because legume proteins might have similar structures, including similar epitope regions (Koeberl et al., 2018), it is possible that patients sensitized to one legume allergen could cross-react to a structurally similar protein from another legume (De Jong et al., 2010; Hoffmann, Münch, Schwägele, Neusüß, & Jira, 2017; Jappe & Vieths, 2010).

5.3. Oilseed proteins

Peanut (Arachis hypogaea L.) and soybean (Glycine max (L.) Merr.), despite the fact that are botanically legumes, are classified as oilseeds. As per lupine, peanuts also cause severe allergic reaction (anaphylactic shock and death). Soy, a product of soybeans, is one of the most common food allergens. Soybeans are assumed not to cause severe reactions; its symptoms are mainly hives, itching, swelling, eczema, vomiting, diarrhoea and nausea (Piccolo et al., 2016). Oilseed rape pollen allergies have been previously described as the result of cross-sensitization with various pollens (Puumalainen et al., 2015). The major allergens are napins (2S albumins) (Puumalainen et al., 2015), and cruciferin (11S globulin) (L'Hocine, Pitre, & Achouri, 2019). However, the clinical relevance of allergy to oilseed is controversial. Indeed, no evidence exists reporting that the intake of rapeseed oil might cause or worsen symptoms in oilseed rape- and turnip rape-allergic patients (Poikonen et al., 2006). However, evidence exists on the allergenicity of oilseeds such as peanut oil based on the response of peanut-allergic individuals in skin prick testing (Jappe & Schwager, 2017).

5.4. Cereal proteins

Wheat proteins are associated with a spectrum of diseases. The most common are gluten-related disorders, which occur for genetically predisposed subjects (carrying HLA-DQ2 or HLA-DQ8) upon ingesting gluten (Elli et al., 2015). Celiac disease is a genetically determined chronic inflammatory intestinal disease induced by gluten, affecting approximately 1% of people in the world (Jnawali, Kumar, & Tanwar, 2016). Gastrointestinal symptoms are mucosal inflammation, small intestine villous atrophy, increased intestinal permeability and malabsorption of macro- and micronutrients (Pasha et al., 2016). Gluten could have a direct 'toxic' (innate) effect on the intestinal mucosa epithelial cells (IECs) (Barone, Troncone, & Auricchio, 2014). As for the adaptive immune response, it involves CD4 + T cells in the lamina propia that recognize processed gluten epitopes called "immunogenic" (du Pré & Sollid, 2015). Wheat allergies can be triggered by several allergens including α-amylase/ trypsin inhibitors, non-specific lipid transfer protein (nsLTP), gliadins, HMW glutenin's germ agglutinin and peroxidase (Christensen, Eller, Mortz, & Bindslev-Jensen, 2014; Elli et al., 2015; Volta, Caio, Tovoli, & De Giorgio, 2013). Baker's asthma is a serious occupational obstructive airway disease affecting 4% to 25% of bakery workers worldwide (Bittner, Peters, Frenzel, Müsken, & Brettschneider, 2015). Major allergens have been identified in soluble proteins including serpins (serine proteinase inhibitors), thioredoxin, agglutinin, α- and β-amylases, peroxidase, acyl CoA oxidase, glyceraldehyde-3-phosphate dehydrogenase and triosephosphate isomerase) (García-Molina, Giménez, Sánchez-León, & Barro, 2019).

5.5. Mycoproteins

Mycoprotein, which springs from the mould *Fusarium venenatum*, is a novel cause of allergic and gastrointestinal reactions, but little information has been available on its associated symptomatology (Koeberl et al., 2018). Mycoprotein shares multiple common allergenic determinants with environmental moulds including *Aspergillus fumigatus, Cladosporium herbarum* and *Alternaria alternate* (Dzeladini, Chan, & Kummerow, 2017).

5.6. Whey proteins

Caseins account for 80% and whey proteins for 20% of the total amount of proteins present in cow's milk (O'riordan, Kane, Joshi, & Hickey, 2014). Major allergens in cow's milk are whey proteins α-lactalbumin, β-lactoglobulin, BSA and lactoferrin as well as 4 caseins (Solinas, Corpino, Maccioni, & Pelosi, 2010). These allergens can be responsible of the onset of IgE-meditated type I hypersensitivity and T cell-mediated delayed-type hypersensitivity reactions (Xu, Gong, Gern, Ikeda, & Lucey, 2018). Symptoms involve skin, gastrointestinal and respiratory tracts and even systemic anaphylaxis may occur, for this reason patients should follow a strict diet to avoid such symptoms (Linhart et al., 2019).

5.7. Insects

Food allergy to insects has been reported for silkworm, mealworm, caterpillars, *Bruchus lentis*, sago worm, locust, grasshopper, cicada, bee and *Clanis bilineata,* (de Gier & Verhoeckx, 2018). Insects are mainly associated with sting or inhalant allergies (Pomés, Mueller, Randall, Chapman, & Arruda, 2017; Stanhope, Carver, & Weinstein, 2015). Furthermore, it has been suggested that shrimp-allergic patients, might present allergy to insects as mealworm since IgE biding to muscular proteins was detected (Broekman et al., 2016). Allergic symptoms following the consumption of insects might affect the skin (e.g. urticaria, pruritus, rash, flushing, angioedema), the gastrointestinal tract (e.g. abdominal pain, nausea, vomiting, diarrhoea) and the respiratory system (e.g. asthma, dyspnoea) (de Gier & Verhoeckx, 2018).

5.8. Microalgae

Although microalgae have been consumed for centuries, there is still unknows regarding the presence of allergens and this may affect the time to market such products as protein sources (Caporgno & Mathys, 2018). Allergic reactions to proteins from edible microalgae spirulina have been recently reported to be involved in several symptoms and in few rare cases anaphylaxis (Le, Knulst, & Roeckmann, 2014). On the other hand, much more research has been done on microalgae as a source of antiallergic compounds (Fleurence & Levine, 2018); nevertheless, some of the same species generating those compounds, can be also described for having allergic activity. Same authors report how species as Chlorella or Arthrospira can change from one to another depending on the environmental conditions of production and harvesting.

6. Relevant regulations for meat protein replacers

The main purpose of the many regulations and controls related to the production, distribution and supply of foods is to ensure that food is safe to eat and has the content and quality promised and expected by the consumer. In the European Union, food derived from animal sources is subjected to special requirements regarding the harvesting of the raw material, under regulations for food of animal origin also known as the "Hygiene Package" Regulations (Regulation (EC) 852/2004, Regulation (EC) 853/2004, Regulation (EC) 854/2004). Additionally, Regulation (EC) 1169/2011 for the provision of food information to consumers establishes the general principles, requirements and responsibilities related to food information and in particular food labelling. The main aim is to provide consumers which clear information which will lead to informed choices to suit their dietary needs. Accordingly, the Regulations also prohibit the use of information that: "would mislead the consumer in particular as to the characteristics of the food, food effects or properties, or attribute medicinal properties to foods" (Regulation (EC) 1169/2011). Of particular relevance, Regulation (EC) 1169/2011 restricts the definition of "meat" to skeletal attached muscles. Any other parts of the animal must be declared separately in the list of ingredients and the meat species must be identified on the label (e.g.: "beef heart" or "bovine heart"). Additionally, these parts must be excluded from the meat and protein content calculations. Regulation 1333/2008 on food additives (latest consolidated version published February 2016) applies to proteins that have a functional role in the final product. Blood plasma, edible gelatine, protein hydrolysates and their salts, are not considered to be food additives, but proteins having properties such as emulsifying, gelling, water holding capacity, etc. and incorporated into foods for this purpose would be considered as food additives. In Australia and New Zealand foods containing offal must include these in the label or directly inform the consumer if the food in question does not have a label (Australia New Zealand Food Standards Code - Standard 2.2.1 - Meat and Meat Products, 2016). In the US, the Federal Code of Regulations states that labelling should not mislead consumers. In addition to parts belonging to the list of specified risk materials from cattle, the Regulations also excludes detached spinal cords, testicles and tonsils from any animal to be used as ingredients of meat food products. Other parts may be used if in line with particular product standards, always accompanied by the name of the species. Blood may also be used if permitted by a product standard, or if it is a traditional or typical ingredient in the product. The term "blood" and the species from which it is derived must be included in the ingredient list. Extracts from any non-meat part of the carcass should be listed in the label including the part name and species from which it is prepared (Regulation 9 CFR 318.6).

For alternative (non-animal) protein sources, regulations are usually related to either novel (have no history of widespread and safe consumption) or allergenic protein ingredients. In most jurisdictions, protections are in place requiring novel foods to be approved before entering the market. In the European Union, the novel food regulation (Regulation (EU) 2017/2470) requires novel protein sources to undergo a comprehensive safety assessment. Once approved, these ingredients may be subjected to specific conditions of use (e.g. specified food category and maximum levels) and/or additional specific labelling requirements, including, if relevant, potential allergenic reactions. Canada, New Zealand and Australia have similar requirements for safety assessment prior to authorisation of foods considered novel, while the United States operates on the principal that any food ingredient needs to be assessed for safety unless it is generally recognised as safe (GRAS) (Van Putten, Kleter, Gilissen, Gremmen, Wichers & Frewer, 2011).

Some examples of novel protein ingredients evaluated and authorised for use in the European Union are lucerne or alfalfa protein, potato proteins (coagulated and hydrolysated) and rapeseed proteins.

7. Conclusions

This review highlights the potential of meat co-products as an excellent source of high-quality proteins that can be incorporated in meat products as meat replacer or protein extenders. In addition, it provides a summary of other alternative non-meat proteins that can be used either a part or whole replacement of meat proteins. In order to meet consumers and producers' expectations, the percentage of replacement must be investigated for each particular combination of replacer and meat product with due regard to researching effects at technological, nutritional and sensory levels. Alternative sources of proteins have the potential to be used as meat analogues, protein replacers or extenders. However, their impact on texture colour, amino acid profile and the presence of antinutrients and allergens must be carefully considered. In all cases, clear labelling of products providing information on the benefits of these approaches is a necessity not a choice. The use of tailored blends of proteins from different sources is a promising strategy that can provide meat products with specific functionalities and nutritive profiles using sustainable resources.

Future research strategies can be directed towards methods of ensuring the pre-treatment of the meat co-products and other alternative proteins prior to their inclusion in food formulation. In this light, pretreatments should have a dual objective: (i) obtaining a product with appreciated techno-functional properties (able to compete with those conventional) and (ii) not hindering the nutritional properties. A multifactorial design will help to identify optimal treatment and inclusion levels for meat by product to maintain both the nutritional and techno-functional properties.

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.

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