

Flavor Aspects of Pulse Ingredients

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ABSTRACT

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Pulses (Fabaceae) have regained interest for their high protein level. However, food application of pulses and pulse ingredients is hampered by several issues around their off-flavor. Off-flavors in pulses are partially inherent and partially produced during harvesting, processing, and storage. Generally, volatile off-flavor compounds in pulses belong to the categories of aldehydes, alcohols, ketones, acids, pyrazines, sulfur compounds, and others, and off-taste is strongly correlated to the presence of saponins, phenolic compounds, and sometimes alkaloids. No systematic studies have been performed on the identification of the off-flavor compounds present in

pulses in relation to their contribution to the overall perception of the pulses. This review article aims to provide a concise overview highlighting the most important aspects of the knowledge available on the off-flavor compounds present in various pulses, their possible origins, and the technologies available to prevent, reduce, or mask these off-flavor compounds. Rather than attempting to make a full inventory of the literature in the field, this paper addresses the most relevant topics referring to a selected set of relevant papers on each topic to substantiate the observations and conclusions that may guide the reader toward additional literature.

The growing world population demands sustainably produced protein-rich foods. Pulses are an attractive crop for the production of protein-rich seeds, which can be eaten directly or used as starting material for the production of pulse ingredients, such as flours, protein concentrates or isolates, and starches. However, flavor is a major factor limiting the use of many vegetable protein ingredients in foods. Similar to soy ingredients, off-flavors in pulses are a barrier to consumption and limit the expansion of pulse ingredients into mainstream food applications. For pea protein, for example, Owusu-Ansah and McCurdy (1991) reported that the objectionable flavor of pea protein hampers broad application in foods and indicated that neither the source of the problem nor the solutions to it have been well documented.

Pulses belong to the family of legumes (Fabaceae). The term pulse refers to the dried seeds. The most common pulses are *Pisum sativum* (peas), *Cicer arietinum* (chickpeas), *Lens culinaris* (lentils), *Lupinus* spp. (lupins), *Vicia faba* (faba beans, broad beans), and *Phaseolus vulgaris* (common beans: kidney beans, navy beans, pinto beans, etc.). The legumes *Glycine max* (soybeans) and *Arachis hypogaea* (peanuts) are considered oil seeds and will therefore not be discussed in this review, although they are closely related to the pulses.

The words flavor, aroma, odor, and taste are not always used in the literature in a consistent way, which may lead to confusion. Taste is caused by nonvolatile compounds and is perceived via receptors on the tongue and in the oral cavity. There are five basic tastes, namely, sweet, bitter, sour, salty, and umami (~savory) (Chandrashekar et al. 2006). Aroma is perceived via a large number of receptors nasally and is caused by volatile compounds (Czerny et al. 2008). Flavor is mainly composed of taste and aroma. In addition to taste and aroma, there are also other effects in the mouth influencing flavor perception. The most important ones are described as astringent (~dry, rough), metallic, pungent (~spicy), cool, and fatty. Astringency is a puckering, dry, or rough sensation in the mouth (Lesschaeve and Noble 2005), which is caused by the interaction of a nonvolatile compound with the salivary proteins and mucins in the mouth, which leads to a loss of lubrication in the mouth (Gibbins and Carpenter 2013). The word off-flavor is used

to describe an unpleasant flavor that includes the perception of unpleasant taste, aroma, and other effects, such as astringency. The texture (e.g., smoothness, coarseness, hardness, thickness, slipperiness, viscosity, etc.) of a food product can also have a large influence on its flavor perception.

This review article does not aim to make a full inventory of the literature that addresses off-flavors in pulses. Instead, it is a concise overview highlighting the most important aspects of the knowledge available in this field covering the off-flavor compounds present in various pulses, their possible origins, and the technologies available to prevent, reduce, or mask these off-flavor compounds. The aspects that will be reported here will be substantiated by referring to a set of relevant papers without seeking completeness.

OFF-FLAVORS IN PULSES

Origin of Off-Flavors in Pulses. Off-flavors can be either inherent to the pulse or developed during harvesting, processing, and storage (Sessa and Rackis 1977). Off-flavors inherent to the pulse can only be removed, modified, or masked, but they cannot be prevented other than by breeding new cultivars with less off-flavor, whereas the developed off-flavor can be limited by tuning the processing of the seeds. Table I gives an overview of the most important families of compounds associated with off-flavors in soy and in peas, the latter being the most studied type of pulse.

The main cause for off-flavor development during harvesting, processing, and storage is oxidation of unsaturated fatty acids (e.g., linoleic and linolenic acids) (Sessa and Rackis 1977). This oxidation can be enzymatic (Makower and Ward 1950; Lee and Wagenknecht 1958) or nonenzymatic (Pattee et al. 1983). Furthermore, off-flavors can be formed by the effect of heat on sugars and amino acids, such as Maillard reactions, by thermal degradation of phenolic acids, by oxidative and thermal degradation of carotenoids, and by thermal degradation of thiamine, or they can be derived as contaminants after solvent extraction (MacLeod et al. 1988). Off-flavor formation by oxidation of fatty acids has been reviewed thoroughly by MacLeod et al. (1988). Hydroperoxides produced by oxidation of fatty acids can be formed spontaneously by autooxidation of unsaturated fatty acids in the presence of atmospheric oxygen by free radical chain reactions. However, lipoxigenase (LOX)-catalyzed degradation of polyunsaturated fatty acids is believed to be a major cause of undesirable off-flavor development in legumes. More than 60 different isozymes of LOX have been identified in plants (Baysal and Demirdöven 2007). Generally, LOXs are classified into two different types (Baysal and Demirdöven 2007), namely, type 1 LOXs,

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which have an optimum pH in the alkaline region and are specific for oxidizing free fatty acids, and type 2 LOXs, which have optimum activity at neutral pH and cause co-oxidation of carotenoids (Maccarrone et al. 1994; Baysal and Demirdöven 2007). The initial products of LOX activity are hydroperoxides, which are further degraded into a wide range of compounds, among which many are responsible for off-flavors (MacLeod et al. 1988). Hexanal, 3-*cis*-hexenal, *n*-pentylfuran, 2(1-pentenyl)furan, and ethyl vinyl ketone are LOX-derived contributors to grassy, beany, and green flavors (Rackis et al. 1979). Lipase is another enzyme that contributes to off-flavor development of pulses. Lipase hydrolyses lipids into free fatty acids, which are then more susceptible to oxidation (MacLeod et al. 1988).

The off-flavors that occur in pulses not only are influenced by the species but also are variable depending on market class, cultivar, crop year, growing location, and storage conditions (Malcolmson et al. 2014), including volatile compounds and nonvolatile compounds. The bitterness of peas is related to the saponin content, which in turn is dependent on the pea variety (Heng et al. 2006). Several studies have shown that pea varieties differ in saponin content (Bishnoi and Khetarpaul 1994; Daveby et al. 1997), but the saponin contents reported vary greatly between different studies and often depend on the methods used for extraction and quantitative analysis (Heng 2005).

The presence of several phenolic compounds has been examined for extracts prepared from different legume seeds (red beans, adzuki beans, green lentils, red lentils, broad beans, faba beans, and peas) (Troszyńska et al. 2006). Among these phenolic compounds, the anthocyanidins delphinidin and cyanidin are typically known to be responsible for color (red, purple, and blue), but there are no sensory studies stating that any of them have an off-flavor. The aglycone cyanidin, however, has been shown to activate bitter taste receptors (Roland et al. 2013). The aglycone quercetin is a compound known for its bitterness, for example, in wine (Drewnowski and Gomez-Carneros 2000). The aglycone kaempferol has also been shown to activate bitter taste receptors (Roland et al. 2013). The glycosides of quercetin and kaempferol have been reported as astringent (Scharbert and Hofmann 2005; Hufnagel and Hofmann 2008). Vanillic, caffeic, *p*-coumaric, and ferulic acids as such have been described as astringent, whereas their ethyl esters have a bitter character, in addition to an astringent character (Hufnagel and Hofmann 2008). Sinapic acid was reported as bitter and astringent (Rubino et al. 1996). It is, however, not known for the different pulses whether these phenolic compounds contribute to bitterness and astringency in the pulse matrix.

Off-Flavors in Peas (*Pisum sativum*). Among pulses, off-flavors are best described for peas. A comprehensive overview of volatile and nonvolatile off-flavors was published by Heng (2005).

The volatile compounds in peas that cause off-flavors are partly inherent to peas themselves and partly developed during harvesting, processing, and storage. Three different 3-alkyl-2-methoxypyrazines have been identified as aroma constituents inherent to green peas, namely, 3-isopropyl-2-methoxypyrazine, 3-*sec*-butyl-2-methoxypyrazine, and 3-isobutyl-2-methoxypyrazine (Murray et al. 1970, 1976; Jakobsen et al. 1998). These compounds are present in extremely low concentrations but are the main compounds that contribute to the perceived green pea aroma in peas owing to their low threshold values (Murray et al. 1970, 1976). Besides 3-alkyl-2-methoxypyrazines, a combination of several classes of volatile organic compounds such as aldehydes, ketones, and alcohols has been reported to have impact on pea off-flavor. These volatile compounds are to a large extent generated by oxidation of unsaturated fatty acids (Sessa and Rackis 1977) and can also be produced via enzymatic reactions (Makower and Ward 1950). After harvesting, processing (e.g., dehulling), and storage, peas usually contain alcohols (methanol, ethanol, and hexanol) as the dominating volatile components (Murray et al. 1976). Aldehydes and ketones, which are present in smaller amounts, are usually of greater significance because they possess stronger aromas (Murray et al. 1976). Hexanal is responsible for the hay-like off-flavor in frozen peas

(Murray et al. 1976) and has also been described as beany, green, grassy, and leafy (Wilkins and Lin 1970; Kaneko et al. 2011). Sulfur-containing compounds (Ralls et al. 1965) and aliphatic and aromatic hydrocarbons (Murray et al. 1976) also contribute to off-flavor of peas.

In a food matrix, compounds such as proteins, fats, and carbohydrates tend to adsorb flavor compounds (Plug and Haring 1993; Fischer and Widder 1997), resulting in their retention. Pea ingredients mainly used in food applications are pea protein concentrate and isolate. However, there is little knowledge on the off-flavor retention of these preparations, especially in relation to food matrices. Heng (2005) has studied the release of off-flavor in pea flour and different pea protein fractions: globulin legumin and vicilin, which are the two main proteins in peas (Derbyshire et al. 1976). Hexanal was the major compound released from pea flour and pea protein isolate, whereas 2-ethyl-1-hexanol was the major compound released from legumin and vicilin preparations. At acidic pH, less volatile compounds were adsorbed to pea flour than at basic pH; at the same pH, less volatile compounds were adsorbed to pea proteins than to pea flour (Heng 2005), indicating that protein purity also affects off-flavor retention. In addition to proteins, the presence of lipids and carbohydrates can also have significant influence on retention of volatile compounds (Heng et al. 2004). Lipids may possibly be the reason for the retention of endogenous volatile compounds in the protein preparations, which may lead to off-flavors if used as food ingredients, but lipids may also be off-flavors themselves (Heng 2005).

The bitterness of peas is related to their saponin contents (Heng et al. 2006). Two saponins, namely saponin β g (also called DDMP saponin) and saponin Bb, have been isolated from peas (Daveby et al. 1998). Saponin β g is the predominant saponin in peas. It differs from saponin Bb in the presence of a DDMP (2,3-dihydro-2,5-dihydroxy-6-methyl-4*H*-pyran-4-one) moiety at the C-22 position, which upon heating is released as maltol (Kudou et al. 1991). Both saponins in peas were perceived as significantly bitter. The threshold for saponin β g was determined to be <2 mg/L, whereas that of saponin Bb was 8 mg/L (Heng et al. 2006). Saponin β g has a similar threshold level to the common bitter reference compound quinine sulfate, which is also perceived at concentrations <2 mg/L. Besides being bitter, saponins in dried peas were also perceived as astringent and metallic (Price et al. 1985). Fractionation, isolation, and analysis revealed that the saponin in the samples was saponin Bb. It is, however, possible that saponin β g also caused this sensory impression, but it might have been degraded to saponin Bb during the isolation procedure. Saponins were reported to have affinity for proteins (Potter et al. 1993). Assuming that all the saponins present in pea flour would be retained during the protein extraction, the final protein isolate would be unsuitable for food consumption in a high amount because it would be perceived as highly bitter and/or metallic.

TABLE I
Overview of the Main Off-Flavors (Most Frequently Mentioned in Bold) and Associated Compound Families Causing Off-Flavor in Soy and in Peas

Off-Flavors	Soy	Peas	Compound	
Volatile	Beany	Green	Aldehydes	
	Fatty	Beany	Ketones	
	Green	Pea	Alcohols	
	Grassy	Earthy	Pyrazine	
	Rancid	Hay-like	Others	
	Leafy	Leafy		
	Earthy	Metallic		
	Cardboard	Brothy		
	Acrid	Acrid		
	Pungent	Pungent		
	Medicinal	Fatty		
	Nonvolatile	Bitter	Bitter	Isoflavones
		Astringent	Astringent	Saponins
Metallic		Metallic	Phenolic acids	
			Peptides/amino acids	

Therefore, when purifying proteins from peas, it is crucial to minimize the coextraction of the saponins together with the proteins.

Off-Flavors in Lupins (*Lupinus* spp.). The volatile off-flavor compounds in lupins (*Lupinus* spp.) have been well described.

The strongest odor impressions found in lupin protein extracts were described as green/milky (*n*-pentanal), sperm (1-pyrroline), grassy (*n*-hexanal), and sulfurous (dimethyl trisulfide) (Schindler et al. 2011). *n*-Pentanal and *n*-hexanal originate from lipid oxidation of linoleic acid and have low odor thresholds of 12 ppb (*n*-pentanal) and 4.5 ppb (*n*-hexanal) in water (Buttery et al. 1988, 1999). 1-Pyrroline is known as a Strecker degradation product of proline and has a low odor threshold of 22 ppb in water (Amoore et al. 1975). With its low odor threshold of 0.008 ppb in water (Milo and Grosch 1996), dimethyl trisulfide is suggested to play an important role as a key aroma compound in lupin protein extracts. Bader et al. (2009) investigated the most important volatile compounds for *Lupinus angustifolius* cv. Boregine by means of aroma extract dilution analysis (AEDA). This technique expresses sensory importance of a compound using the flavor dilution (FD) factor (Grosch 2001). The most important flavor attributes for lupin flour were cheese-like, metallic, green or grassy, meat-like, fruity, fatty, and hay-like. AEDA, in combination with the identification experiments, revealed the sweaty and cheese-like substances 2- and 3-methylbutanoic acid to have the highest FD factor of 2,048. High intensities (FD factors 512–1,024) were also found for *trans*-4,5-epoxy-(*E*)-dec-2-enal (metallic), vanillin (vanilla-like), and β -ionone (violet-like, flowery). β -Ionone was reported to be derived from the oxidation of carotenoids, for example, β -carotene (Belitz et al. 2001). Nine compounds were reported to have FD factors of ≥ 256 , such as 3-isopropyl-2-methoxypyrazine, a set of unsaturated aldehydes, and for example, compounds such as vanillin, maltol, sotolon, and ethyl vanillin (although this last compound is a synthetic compound, it was reported to be present in the extract). The methoxypyrazines probably originate from a secondary metabolic pathway in plants, as demonstrated in some raw vegetables such as peas and beans (Murray and Whitfield 1975). Eleven substances revealed medium-high intensities (FD factors 32–128) and were identified, for example, as unsaturated and saturated carbonyl compounds, most likely derived from LOX activity in lupin flour. Lupin flour contains high amounts of polyunsaturated fatty acids, especially linoleic acid (18:2) and linolenic acid (18:3) (Chiofalo et al. 2012). Five odorants with low odor thresholds were reported to derive from oxidation of linoleic and linolenic acids (Belitz et al. 2001). The carboxylic acids most likely derive from the degradation of amino acids owing to the metabolism of microorganisms present on the hulls of the lupin seeds upon storage (Bader et al. 2009). The most potent odorants in lupin flour seem to derive from LOX activity, secondary metabolism, or activity of microorganisms. However, this study described the sensorially most important flavors but did not classify whether all the detected flavors were perceived as off-flavors or whether some of the flavors were perceived as positive flavors. Furthermore, the taste aspect of lupin flour was not investigated. In contrast to the off-flavors in fresh lupin flour (which were cheese-like, metallic, green or grassy, meat-like, fruity, fatty, and hay-like) (Bader et al. 2009), the main volatile off-flavors for stored spray-dried lupin protein extract have been described as green/milky, sperm, grassy, and sulfurous (Schindler et al. 2011), indicating that the processing and the storage of the *Lupinus angustifolius* seeds resulted in very different off-flavors.

The LOX activities of different lupin species and varieties were determined and ranged from 50 to 1,004 units/mg of protein (Stephany et al. 2015). In contrast to soy and other legumes, LOX from lupin only converted free fatty acids, whereas trilinolein and β -carotene were not oxidized, which might result in less off-flavor formation. Consequently, according to the established classification, lupin LOX activity may be assigned to the type 1 LOX (Stephany et al. 2015).

All species of lupin contain quinolizidine alkaloids to a greater or lesser extent (range: 0–4% of the seed, w/w) (Blancher et al. 1981).

Wild lupins have higher levels of alkaloids than domesticated lupin varieties. These quinolizidine alkaloids impart a bitter taste even when present in low concentrations; they make the seed unpalatable and sometimes toxic (Cristian et al. 2010).

Off-Flavors in Lentils (*Lens culinaris*). Mainly alcohols, aldehydes, and ketones have been identified as volatile compounds in lentils (Lovegren et al. 1979). To the best of our knowledge, there is no literature that establishes the importance of particular volatile compounds in generating off-flavors in lentils.

The presence of off-flavors in lentils is described in the literature (Troszyńska et al. 2011), but the identity of the compounds sensorially responsible for the off-flavor is, to the best of our knowledge, still lacking. Because lentils have a high LOX activity (second to soy), off-flavor compounds originating from lipid oxidation were expected and have been reported (Bhatty 1988). Troszyńska et al. (2011) detected the off-flavors beaniness and aftertaste in lentil in the beginning of a seven-day germination period. At the end of the seven-day germination period, a general off-odor, pea-pod flavor, bitterness, and astringency had developed. The beaniness had almost vanished by then. The bitterness and astringency of the lentil sprouts were ascribed to catechin gallate and different forms of kaempferol glycosides (Troszyńska et al. 2011). However, there are phenolic compounds present in lentils that are known to be bitter, astringent, or both (Troszyńska et al. 2011). Also, the isoflavones daidzein (0.01 mg/100 g) and genistein (0.05 mg/100 g) have been identified in raw lentils (USDA 2008). However, these are low concentrations compared with the isoflavone concentrations in soy, which are about 1,000 times higher (USDA 2008). Therefore, it is likely that isoflavones do not cause off-flavors in lentils. Lentils contain considerable amounts of saponin β g (e.g., *Lens culinaris* Magda 20, 0.7 g/kg, and *Lens culinaris* Lyda, 1.1 g/kg) (Ruiz et al. 1996b). Saponin β g has been shown to cause bitterness in peas, with a threshold value in water of < 2 mg/L (Heng et al. 2006). Also, after cooking, lentils still contain a high amount of saponins, but with increasing cooking time, saponin β g is more and more converted into saponin Bb (Ruiz et al. 1996a). Saponin Bb is less bitter than saponin β g but is still considerably bitter. It can, therefore, be assumed that saponins β g and Bb may also cause bitterness in lentils; however, lentils are not known as especially bitter or astringent.

Furthermore, the compound arbutin has been identified in lentils (Tsoptom and Muir 2010) but not quantified. Arbutin is a well-known bitter-tasting compound with a sensory threshold of 0.9mM in water (Bufe et al. 2002). Because no arbutin concentration in lentils was given, it cannot be said whether its concentration is above or below the sensory threshold. Furthermore, it is not known whether potentially off-flavor-containing compounds exhibit their off-flavor in the lentil matrix, because other components may mask potential off-flavors.

Off-Flavors in Chickpeas (*Cicer arietinum*). Literature about off-flavors in chickpeas is scarce, and the literature found is mainly on chickpea applications, for example, to what extent wheat flour can be replaced by chickpea flour without off-flavor.

Chickpeas are not reported to be bitter or astringent, to the best of our knowledge. However, several phenolic compounds are present in chickpeas that are known to be bitter, astringent, or both (Simons 2011; Troszyńska et al. 2011). Also, the relatively uncommon isoflavones formononetin and biochanin A are present in chickpeas. They have not been investigated in sensory tests, but they activate the same bitter receptors that other isoflavones, such as daidzein and genistein, do (Roland et al. 2011). Therefore, it is likely that they also taste bitter. It is not known, however, whether these compounds taste bitter in the chickpea matrix.

Phosphatidylcholine has been identified in defatted chickpea flour (Sánchez-Vioque et al. 1998). Sessa et al. (1974) identified oxidized phosphatidylcholine as a contributor to the bitter taste in soybeans. It might, therefore, be possible that phosphatidylcholine can also be oxidized in chickpeas and lead to bitterness.

Off-Flavors in Faba Beans (*Vicia faba*). Volatile compounds in faba beans (also called fava or broad beans) have been

shown to consist of aromatic hydrocarbons, aldehydes, alkanes, alkenes, alcohols, ketones, furans, and other compounds (Oomah et al. 2014). The off-flavors reported for faba bean protein isolates were dependent on the pH. At neutral pH, a dried pea-like flavor was reported, whereas at acidic pH an unpleasant fruity flavor was reported (Schultz et al. 1988). It was speculated that one or more volatile organic acids produced this specific off-flavor in the faba bean products. Regular faba beans contain up to 8–9% tannins, which can explain their perceived bitterness. As a result of plant breeding, there are low-tannin or zero-tannin faba beans, which contain only 1% tannins (Oomah et al. 2011).

Off-Flavors in Common Beans (*Phaseolus vulgaris*). Literature reporting off-flavor compounds in common beans is also scarce. A total of 62 volatile compounds have been detected in uncooked common beans (*Phaseolus vulgaris* L.) of the three market classes black bean, dark red kidney bean, and pinto bean (Oomah et al. 2007). They belonged to aromatic hydrocarbons, aldehydes, alkanes, alcohols, and ketones. The bean cultivars differed in abundance and profile of the volatile compounds. No sensory tests were performed in this study, but the presence of compounds previously known to have impact on flavor could be shown. Hexanal, 3,5-octadien-2-one, 1-penten-3-ol, and benzaldehyde were established as volatile marker compounds for common beans. Volatile compounds in common beans have also been reported by Buttery et al. (1975), including aliphatic ketones, aliphatic alcohols, aromatic oxygenated and heterocyclic compounds, aromatic hydrocarbons, terpenes, furans, pyrazines, pyridines, thiazoles, thiazolines, other sulfur heterocyclics, and benzene derivatives. The information regarding impact on off-flavor of common beans is, however, limited. Geosmin, an oxygenated hydrocarbon, has been reported as responsible for the musty, moldy, earthy flavor of dry white navy beans (Buttery et al. 1976). A cooked bean aroma has been reported for pinto bean paste. The paste was low in raw beany flavor, nutty flavor, bitterness, and astringency (Song et al. 2009).

TECHNOLOGIES TO IMPROVE THE FLAVOR OF PULSES

Systematic studies on off-flavor improvement of pulses are largely lacking in the literature. Therefore, technologies that can be used to remove specific compounds in specific pulses were investigated. An acceptable flavor is a crucial aspect for products to be successful in the food industry. One way to obtain products with acceptable flavors is to start with raw materials that are low in flavors and to make sure that development of off-flavors remains as low as possible during processing and storage. The following technologies have been described for pulses to prevent formation of off-flavors.

Cultivar Selection. Between cultivars there can be a large variation in flavor composition. Depending on the intended application, it can be useful to use cultivars low in off-flavor precursors, off-flavors, or enzymes that support off-flavor development. LOX is regarded as one of the main causes of off-flavor development in legumes, mainly of volatile off-flavors. In soybeans, cultivars lacking or overexpressing one or more of the LOX isoenzymes have been identified (Boyington et al. 1993). Analogously, the occurrence of differing LOX activities within different lupin cultivars and species may be assumed (Stephany et al. 2015). However, LOX has also been reported to play a beneficial role for several functional properties in baking technology (Rackis et al. 1979). The choice of LOX-free or low-LOX cultivars should thus be well considered.

The same holds for cultivars low in other off-flavors, such as saponins, isoflavones, or phenolic compounds. Regarding tannins in faba beans, for example, there are faba bean cultivars that have, as a result of plant breeding, a low tannin content. These cultivars are lower in bitterness than high-tannin cultivars (Oomah et al. 2011).

In addition to the variety, the location and the crop year (different weather conditions) at which the crop is harvested may have an

impact on the off-flavor compounds found, although the literature available for pulses on this topic is limited. A study showed that the crop year is the dominating factor in isoflavone concentrations in soybeans (Wang and Murphy 1994).

Control of Oxidation and Temperature. A simpler way to control the formation of off-flavor compounds is the inactivation of LOX and other enzymes. This is often achieved by thermal treatment such as blanching, steam heating, or dry heating. Tissue wounding owing to mechanical harvesting triggers lipid oxidation pathways. For example, bruised peas that are not blanched develop strong off-flavors within a few hours (Dörnenburg and Davies 1999). Chickpeas that were cooked and parched showed a marked drop in LOX activity with complete inhibition of lipase activity (Attia et al. 1996). Another way to inactivate LOX is alcohol or acid treatment (Rackis et al. 1979). Storage of peas at 4°C has been shown to lead to less volatile compound development than at ≈22 or 37°C (Azamia et al. 2011). However, low storage temperature alone might not be sufficient to prevent off-flavor development. Consequences of the treatments here described can be accelerated chemical reactions upon heating, development of new flavors (among which possibly off-flavors), and possible change of protein functionality owing to the influence of heat, alcohol, or acid.

TECHNOLOGIES TO REMOVE OR MODIFY OFF-FLAVOR IN PULSES

Soaking and Thermal Treatment. Soaking is a treatment often applied to leach unwanted compounds into the soaking water. Thermal treatment is usually used to remove solvent in defatted flour production, to inactivate enzymes, or to destroy antinutritional factors (Chango et al. 1995).

Debittering of lupin (bitterness caused by the presence of quinolizidine alkaloids) is an operation that involves leaching in water and boiling in water (Yáñez et al. 1986; Santana and Empis 2001). This processing unfortunately leads also to a partial loss of the soluble protein, together with the undesirable bitter alkaloids (mainly lupanine), oligosaccharide flatulents, and phytate. Another treatment described to eliminate bitterness in lupins is thermal coagulation (Chango et al. 1995). The effects of aqueous acid and alkaline thermal debittering treatments were evaluated for *Lupinus campestris* seed flour. The alkaline treatment was most effective in eliminating quinolizidine alkaloids, and the sample obtained with alkaline treatment had the best protein quality (Cristian et al. 2010).

Soaking, cooking, and germinating of lentils (*Lens culinaris* var. *vulgaris*) were performed in distilled water, citric acid, and sodium bicarbonate solutions to investigate changes of antinutritional factors such as the trypsin inhibitor activity and the phytic acid content (Vidal-Valverde et al. 1994). Soaking did not modify the trypsin inhibitor activity, decreased the phytic acid content, and increased the tannin and catechin contents. Cooking the presoaked seeds and germination both resulted in large decreases of trypsin inhibitor activity and phytic acid level and in increased contents of tannins and catechins. Soaking and cooking processes liberate these complexes, and therefore, the tannin levels increase (Vidal-Valverde et al. 1994). Tannins and catechins can act as off-flavors, because tannins are mainly astringent and slightly bitter, and catechins are mainly bitter and slightly astringent (Peleg et al. 1999). However, lentils are not known as especially bitter or astringent. Fernandez et al. (1993) described an increase in the tannin and catechin contents after soaking faba beans in distilled water, 0.1% citric acid, and 0.07% bicarbonate solutions. As explained for lentils earlier, an increase of the tannin and catechin contents might affect bitterness and astringency.

Roasting has been reported as a processing step to reduce the beany off-flavor from lupin seeds (Yáñez et al. 1986). The effect of roasting lupin seeds (*Lupinus albus* cv. Multolupa) for 10, 20, 30, and 40 min at 80–90°C was studied. Roasting for 20, 30, and 40 min caused a severe reduction in protein quality. Therefore, the authors

suggested heating lupin seeds for 10 min may help to remove the beany flavor of this legume. However, they did not perform a sensory study on whether the beany flavor was really reduced.

Germination. Germination improves the nutritional quality of legumes by decreasing antinutritional factors, such as trypsin inhibitors and phytates, and undesirable beany flavor caused by LOX, along with increasing the levels of phytonutrients such as vitamins, phytosterols, saponins, and phenolics (Simons 2011). Because certain saponins and phenolics are known to be bitter, astringent, or both (Okubo et al. 1992; Soto-Vaca et al. 2012), the benefit of decreasing beany off-flavor by germination might be undone by the disadvantage of increasing bitterness and astringency.

Troszyńska et al. (2011) studied the sensory impact of germination on green lentil seeds. Their objective was to investigate whether the change of phenolic compound composition owing to germination might modify or improve the sensory quality of the resulting sprouts. After seven days of germination, the intensity of the attributes beany and green decreased, but unfortunately, germination of lentil sprouts resulted in an increased intensity of the attributes off-odor, green, bitterness, and astringency. A strong correlation was found between two of the negative sensory attributes (bitterness and astringency) and the content of some of the phenolic compounds (catechin gallate and four forms of kaempferol glycosides). Despite the improvement on some sensory attributes, germination did not improve the overall sensory quality of lentils.

However, in the case of chickpeas, it was shown that germination can increase the sweetness (Bellaio et al. 2013), and thereby bitterness might be masked.

Solvent Extraction. To remove off-flavors from pea protein preparations, one may need to apply an efficient lipid extraction step prior to protein extraction and purification (Heng 2005). Hexane extraction may not sufficiently remove lipids compared with chloroform/methanol extraction (Heng et al. 2004). Because chloroform/methanol is not food grade, this solvent cannot be used to remove (polar) lipids (and together with this, off-flavors) from materials to be used as food ingredients. Aqueous ethanol (e.g., 70%) could be used as an alternative, but the efficiency with respect to removal of lipids and off-flavors remains to be established. In addition, this treatment may lead to a change in protein conformation, which may result in a protein that is not functional for gelling; this requires further investigation as well (Heng 2005).

Organic solvents are effective in removing phenolic compounds from legumes. The solvent (aqueous solutions of acetone, methanol, or ethanol) with the highest extraction yield depended on the kind of phenolic compound and on the kind of pulse (Xu and Chang 2007). This study aimed at a high extraction of phenolic compounds and therefore did not investigate the effect of organic solvent treatment on off-flavors or protein functionality after the treatment. It can be expected that the best extraction solvent is not necessarily the best method to maintain protein functionality.

A combination of defatting and isoelectric washing of faba bean flour was the most effective technique in reducing dried pea flavor of faba bean protein isolates (Schultz et al. 1988).

Fermentation. It is known that there are some saponins that are sweet (Kitagawa 2002; Heng 2005). Sweetness or bitterness of saponins is not dependent on the saponin skeleton but on the type of substituents (both sugar residues and other functional groups) attached to the skeletons, as well as the stereochemistry of the attached substituents. Heng (2005) suggests by theoretical explanation that saponin Bb might be modified in such a way that it becomes sweet or at least neutral in taste, for example, by addition of a carbonyl at C11, which can possibly be obtained by fermentation. Fermentation of saponins might thus be a future area to explore in debittering of pulses.

An alternative debittering method to leaching and boiling is the bacterial removal of quinolizidine alkaloids in lupins. Bacterial debittering has been described for lupin seed flours, using two bacterial strains able to catabolize lupanine and to degrade other lupin alkaloids (Santana and Empis 2001).

Lactic acid fermentation studies were performed for the improvement of the aroma of pea and lupin protein (Schindler et al. 2011, 2012). Fermentation considerably amended the aroma profile of pea protein preparations, resulting in a reduction or a masking of undesirable flavors. The aroma profile of untreated lupin (*Lupinus angustifolius*) protein extracts (LPE) was compared with that of fermented protein extracts (LPEF), both after storage for three months in the dark. Most of the identified substances were lipid degradation products or derived from the Strecker degradation of amino acids (Ballance 1961). Although the fermented extract (LPEF) was not free of off-flavors, the aroma profile was significantly modified by the fermentation process, and the off-flavors were reduced or masked by newly formed compounds when compared with the LPE. Similar results were obtained by lactic acid fermentation of pea protein (Schindler et al. 2012).

Enzymatic Treatment and Ultrafiltration. Enzymatic treatment and ultrafiltration were investigated to reduce the off-flavors present in pulses (Roozen and Pilnik 1979; Song et al. 2009). Even though the enzymatic treatment could reduce some off-flavors, it also increased the presence of some other ones (Song et al. 2009). The use of membranes in combination or not with enzymes to remove off-flavors in pulses was not successful (Roozen and Pilnik 1979). Two points must be noted, however: 1) membrane filtration has changed enormously in the last two decades, and 2) the number of examples was insufficient to conclude that filtration is inadequate.

TECHNOLOGIES FOR MASKING OFF-FLAVORS IN PULSES

There is not much literature about masking of off-flavors in pulses. Traditional masking methods in the food industry involve the addition of sugars, salts, acids, and flavorings. For faba bean protein isolates, addition of strawberry flavor to gels made with faba bean protein isolates was effective in reducing dried pea flavor (Schultz et al. 1988). Masking of faba bean flavor in faba bean protein with vanillin was not successful, which was explained by increasing binding of vanillin to proteins upon heat treatment or upon increasing protein concentration, and only free vanillin contributed to the flavor perceived (Ng et al. 1989).

Heng (2005) suggested masking of pea off-flavor by addition of volatile compounds. It should, however, not be overlooked that if different off-flavor compounds are present these might interfere with each other. Heng (2005) suggested that the presence of saponins in pea protein preparations might have an effect on not only the bitterness of the pea protein preparation but also indirectly on the aroma of the pea protein preparation. Saponins are likely to interact hydrophobically with pea proteins. This means that they may occupy the hydrophobic pockets on the proteins and thus may compete with volatile compounds for the same interaction sites, or create more sites on the proteins. The retention of added aroma compounds could, therefore, be reduced or be enhanced owing to the presence of saponins (Heng 2005). Thus, the removal of saponins might also affect more than only bitterness.

Pulse ingredients are often used as partial replacers of traditional ingredients such as wheat flour (often in the context of gluten avoidance). The extent to which the pulse ingredient is used is mainly limited by flavor. Up to a certain percentage, the off-flavor of the pulse ingredient is masked by other compounds present in the food matrix, such as positively associated flavor. The extent to which replacement is possible can vary a lot depending on the application and probably also on the quality of the pulse ingredient, which is often not described. Most literature in this field reports the usage of chickpea ingredients, suggesting that chickpeas are the most suitable pulse for these applications.

Another method used for masking off-flavors is fermentation. As mentioned earlier, fermentation of pea protein extracts led to the formation of, for example, 2,3-octanedione, 2-nonanone, 3-octen-2-one,

2,4-heptadienal, 2-nonenal, 2-undecanone, 2-octen-1-ol, 2,4-nonadienal, and 1-decanol (Schindler et al. 2012), which could be potentially responsible for masking off-flavors in pea protein extracts.

CONCLUSIONS

Although pulses and pulse-derived ingredients are of increasing importance in quickly growing numbers of applications in the food industry, there is still a strong need to improve the quality of these ingredients in relation to their flavor profile. Having said that, this literature review still concludes that although pulses strongly gain in economic interest there is a gap in the knowledge regarding off-flavor compounds in pulses, most specifically for the lentils, faba beans, chickpeas, and common beans. This even holds for peas, which are the most commonly used pulse ingredient in foods. Moreover, the majority of the studies regarding peas were performed on green peas and not on yellow peas, whereas the latter variety is the most used in industrial applications.

This review shows that there are some general principles dominating the type of off-flavors present in pulses. For example, the most important off-flavor compounds in pulses mainly originate from oxidation of unsaturated fatty acids. On the other hand, the oxidative products generated are also dependent on the type of LOX present (specificity and activity). In addition, off-flavor compounds such as, for example, phenolic compounds and saponins present in pulses were observed to be more variety-specific or were reported to vary owing to differences in environmental conditions.

Because expanding the applications of pulse protein ingredients is commercially interesting but hampered because these materials still have an off-flavor, developing more bland variants of the currently marketed materials will deliver new opportunities, especially in protein-enriched foods and beverages with a mild taste and flavor. To be able to develop such improved ingredients, insight into the nature of the flavor system is crucial: whether the current nonbland flavors are the result of off-flavor molecules that are an intrinsic property of the pulses or are generated during the production and isolation processes of these ingredients. Next to that, knowing the identity of these compounds will give strong leads in developing methods for preventing production of these off-flavor systems during the manufacturing process. Knowing the off-flavor molecules' identities will provide opportunities to remove them from the materials or even, if still required, mask their sensory effect. Irrespective of the growing commercial importance, the scientific background behind the origin and identity of these off-flavors in pulses is scarce. Moreover, the data available from the literature are fragmented in the sense that not all relevant topics in production or reduction of off-flavors are sufficiently well covered for the most important pulses. Setting up and executing systematic comprehensive studies is crucial to increase the understanding of what drives off-flavor perception in pulse-derived materials. The information gathered for the latter set of materials still falls short most particularly when compared with what is available for soy already.

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