Unconventional protein sources for poultry feeding – opportunities and threats

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Abstract

In search for sustainable alternatives for fish meal and overseas vegetable protein sources some selected protein sources were evaluated. These included oil seed co-products, grain legumes and their concentrates, insects, leaf and aquatic proteins, etc. These sources differ substantially in terms of protein yield, environmental sustainability, nutritional value and availability. Products with a low dry matter content, i.e. lucerne, leaves, aquatic proteins are considered to be less sustainable due to the high energy costs for drying. Some values, however, are lacking for some of the protein sources.

Within the category of oil seeds, European produced soybean meal seems to be the most promising alternative for soybean meal from beans imported from South America. Nutritional value and especially protein digestibility of soybean meal is very good. Protein yield of soybean meal produced in Europe should be further increased to make this crop feasible for the farmer. To realize this, varieties have to be selected with an ultra-short growth season.

Within the category of grain legumes, peas seem the most promising alternative for soybean meal, at least for the short-term. The protein yield is reasonably high, but should be further improved. In long-term, leaf proteins and aquatic proteins probably might contribute to reduce soybean imports. Therefore, more knowledge regarding protein separating techniques and nutritional value of these products is necessary. Leafs, aquatic and insect protein sources are not in direct competition with the land use of other crops. Enhancing the protein content of these plant protein sources by processing needs further development; especially products such as duckweed and algae seems promising, but sustainability aspects should be studied. We also realize, that the nutritional value of products such as leaf proteins, some aquatic proteins and insects is not known sufficiently. For these sources, aspects of cell wall degradation, feed safety and legislation should definitely be covered. At this moment it is not possible to draw sound conclusions with respect to the environmental sustainability of the selected protein sources.

Introduction

The total EU protein crop production (e.g. protein from legumes seeds, soybeans, etc.) currently occupies only 3% of the EU's arable land. For this reason, large amounts of vegetable protein sources are imported in the EU, for a large part originating from South America. These imports are subject of increasing concerns, but the reasons of concern differ between stakeholders. A major concern of NGO's is the loss of natural ecosystems and its biodiversity, the increased water and soil pollution, and the ejection of small farmers and the native population in the producing countries. In addition, the European Parliament is worried about the increased dependency from imports from South America. Enhancing the EU protein

crop production enlarges the possibilities for crop rotation, having the advantage to reduce the risk of crop diseases and stabilising EU farmers' income. Moreover, more influence on socially desirable cultivation (e.g. non GMO soybean production) can be practised.
So, if there is a strive to be less dependent from non-European protein sources, this could be achieved by:

- The use of protein sources from EU, incl. new, unconventional diet ingredients
- Improved production efficiency
- Lowering the crude protein level in poultry diets, and/or increase of crystalline amino acids where possible.

In this manuscript, possibilities for a successful cultivation, processing and use of alternative protein sources in (organic) poultry diets under European climatic conditions have been summarized, thereby taking sustainability issues and legislative aspects into account. Because of new regulations with respect to regional cultivation of feed and the ban on the use of non-organic feed ingredients, the organic animal husbandry urgently needs regionally produced high quality protein sources. Therefore, the organic sector is an even larger appropriate sector for applying new protein sources and may also serve as an example for an increased use of novel protein sources in conventional intensive animal production systems. These novel, rather unconventional protein sources have been neglected up till now, owing to constraints imposed by different nutritional and technological considerations (Table 1).

Table 1. Factors limiting the use of novel feed resources in broiler feed formulation

Nutritional aspects

- variability in nutrient level and quality
- presence of naturally occurring anti-nutritional and/or toxic factors
- presence of pathogenic micro-organisms
- need for supplementation

Technical aspects

- seasonal and unreliable supply (need for storage)
- bulkiness, wetness and/or powdery texture
- processing requirements
- lack of research and development efforts

(Modified after Ravindran & Blair, 1991)

Present investigations, therefore, have to concentrate on the factors limiting to the use of these ingredients, originating from both:

- relative conventional ingredients, but implying novel technologies
- unconventional ingredients

The following information describes the potential of nutrient recovery from certain protein sources. For this purpose, the study of Van Krimpen al. (2013) was used as a general structure of the analysis with additional information on insects (Veldkamp et al., 2012) and special journal articles. The reader is referred to these studies to obtain references to a large number of issues presented in this paper.
In Van Krimpen et al. (2013), a long list of 62 feed ingredients was composed, containing a wide range of potential protein sources. Then, criteria were applied to select the protein sources that potentially might contribute to increase the European protein production:

- the protein source should be able to perform well in the climate conditions of North West Europe;

- the cultivation of the protein source in Europe is currently no common practice or its process technology is;
- In the long term (after 2020) the protein source is still applied in feed and not in food.

Based on these criteria, the following protein sources were identified as potentially interesting alternatives (Table 2).

Table 2. Short list of potentially interesting protein sources to increase EU feed protein production

Category	Protein source
Oil seeds	Proteins of defatted soybeans, rapeseed and sunflower seed
Grain legumes	Peas, *Vicia faba*, lupines and their concentrates, chick peas
Forage legumes	Lucerne (alfalfa)
Cereals and pseudo cereals	Proteins from oat and quinoa or cereal co-products
Leaf proteins	Grass, sugar beet leaves
Aquatic proteins	Algae, both macro- (seaweed) and microalgae, duckweed Mussel meal
Insects	Mealworm, housefly, black soldier fly
Microbial proteins	Bacterial protein meal

From a plant cultivation point of view grain legumes, especially peas and beans, are very interesting due to their high protein content (17-35%) and because cultivation practises are already available and implemented. However, these crops are very sensitive to pests and pathogens. Soybeans might be interesting because of the high protein content, although the current yield is too low for making cultivation attractive for the European farmers. Breeding steps for high yielding cultivars with a short growing season still need to get more input. Rapeseed (meal) is already cultivated in considerable amounts in the EU with a reasonable protein yield per hectare.

Aquatic protein sources (duckweed, several micro-algae, some macro-algae and bacteria proteins) are very interesting because of a high protein content and very high yields (Table 3), but processing and feasibility for application as feed ingredients, however, still needs much research. Not only the high yield per hectare, as for duckweed, and the high protein level is interesting, but also the fact that these new putative protein sources do not need good agricultural soil for cultivation. For all protein sources with a high water content, such as (left-over) leaf material, duckweed, micro- and macro algae, a drying step for storage and transport is - in most cases -required.

Table 3. Protein content, yield/ha and protein yield/ha of some protein sources.

	Protein level (%)	Yield potential in EU (tons DM/ha/y)	Protein yield possible (tons/ha/y)
Soybeans, full fat	35	1.5 – 3	0.6 – 1.2
Legumes (pulses) e.g. peas	17-35	4-6	1-2
Sea weed	10-30	25	2.5-7.5
Micro-algae	25-50	15-30	4-15
Duckweed	35-45	30 -40	10-18

In the search for European protein sources, not only crop yield and nutritional value, but also environmental issues have to be taken into account for a complete consideration. Since feed production, including crop cultivation, feed processing and transport are responsible for about 54-73% of total GHG emissions per kg of pig. optimization of feed production and diet composition might help to reduce GHG emissions from animal husbandry. Calculation of GHG emission is based on emission factors during crop cultivation and transport, like input of N, phosphate, herbicides, pesticides, and diesel used by field machinery and transport. GHG emission is expressed in g CO_2-equivalents per kg of crop yield, also known as carbon footprint (CFP).
In addition to the emission of GHG directly related to feed production, GHG emission also occurs due to land use and land use change (LuLuc). Land use change relates to the conversion of land used for e.g. forestry or pasture, into cropland for cultivation. GHG emissions from land use change are distinguished in emissions from direct and indirect land use change. Direct land use change relates to the conversion of land attributed directly to a feed ingredient, e.g. soybeans. Indirect land use change relates to the conversion of land for other crops, induced by changes in the cultivation area of feed ingredients. If for instance pristine lands are cleared to fulfil the increased demand for crops, because part of these crops are used for the production of biofuels, the additional emissions of GHG due to this indirect land changes has to be adjusted to the GHG balance of the biofuel. Attribution of land use change emissions to crops is a complex issue and the methodology is still subject of debate.

In the following text, separate protein sources will be characterized for its properties with respect to its potential a novel poultry feed ingredients.

Oil seed co-products and grain legumes

Characterization

The processing of ingredients, thereby reducing the level of anti-nutritional factors (ANFs) and increasing the protein content to levels of 65% or higher, would fulfil the need for high quality proteins for application in all kind of organic diets and in conventional diets for young animals (piglets, broilers, rearing hens). The processing of the selected feed resources to enhance their protein content is generally still in development and not yet well established.
On the short term, attractive protein enriched resources might be:

- Regarding oil seeds: rapeseed protein concentrates. Protein enrichment of defatted sunflower meal seems to be less attractive.
- Regarding legumes: protein concentrates prepared by fine milling and subsequent dry fractionation from peas (Table 4) and faba beans. The former are already on the market.
- Regarding cereals, cereal protein concentrates as a results from front-end fractionation in ethanol production (Jung and Batal, 2009) can be considered.

Proteins derived from oil seeds are very useful for application in pig and poultry diets, while there is already a widespread use of soybean, rape seed, and sunflower seed meal in these diets. These protein sources are well known in terms of chemical composition and nutritive value. It is assumed that the nutritional characteristics of European cultivated soybean meal are similar to the ones cultivated in South America, but until now this has not been proven. Less information is available with respect to concentrates of these protein sources. Results of one experiment showed that rape seed (canola) protein concentrate can be used up to 10% in piglet diets.

Dry fractionation of legumes seeds include fine milling and separation of particles on the basis of differences in the density of protein and starch granules in an air flow, the process of air classification. In this way, a fineness fraction is derived with protein levels up till 55-60% (Table 4; Van der Poel & Zandstra, 2013; unpublished results).

Table 4. Yield and separation efficiency of pea (cv Mulder Marne) after milling and air classification (cut point 20 µm).

Fraction	Yield (% of M)	Moisture level (%)	CP in DM (%)	PSE* (%)
M**	100	11.9	21.0	
P1	20.4	9.5	56.7	
S1	79.6	--	--	
P2	6.4	9.1	53.3	
S2	73.2	11.0	11.8	
P1	20.4			55.1
P1+P2	26.8			71.4

* Protein separation efficiency
** M = whole pea; P is protein (fineness) fraction; S = starch fraction

From Table 4 it can be deduced that fractionation of this pea cultivar resulted in a pea protein concentrate (PPC; P1 + P2 fractions) that comprises 26% of the pea mass, has a protein content of ~ 55%; about 71% of the total original protein level was included in the PPC. Protein concentrates from feed legumes are commercially available, however, together with the enrichment of the protein in the fineness fraction, also its proteinaceous enzyme inhibitors (trypsin inhibitor; TIA) are enriched in these fractions.

Peas differ greatly in their TIA level which means that, depending on the TIA level and its application, an additional heating step is necessary before (treatment of peas) or after (treatment of the concentrate) dry fractionation (Table 5).

Table 5. Apparent ileal digestibility in broilers of methionine and cysteine in peas with low and high trypsin inhibitor activity (TIA)

	Pea cultivar A		Pea cultivar B	
	High TIA	Low TIA	High TIA	Low TIA
TIA (TIU/mg DM)	8.73	1.45	7.40	1.78
CAID cysteine	0.738	0.812	0.721	0.804
CAID methionine	0.887	0.930	0.885	0.929

TIU: trypsin inhibitor units; CAID: coefficient of apparent ileal amino acid digestibility.
WISEMAN et al. (2003)

Legumes, e.g. *Vicia faba*, lupines and peas, and chickpeas, can significantly contribute to the protein supply of poultry, although their anti-nutritional factors have to be taken into account. Results of a piglet trial showed that the digestibility of pea protein concentrate was similar or even better than that of whole peas. Based on these results, and considering that the production process of protein concentrates from legumes is sustainable and already commercially available, it was concluded that these concentrates are a promising category of European produced high quality protein, especially for application in organic diets.

An example of the more conventional ingredients (corn) combined with more or less unconventional processes is front-end fractionation sieving DDG. In this procedure, corn fractions (endosperm, germ, bran) are separated before fermentation takes place, giving rise to a high protein corn distillers dried grain (corn DDG). This process eliminates non-fermentable fractions (germ/bran) to result in a DDG that contains more protein (45% in DM) and amino acids (but is lower in phosphorus and fat) than the traditional distillers dried grains and solubles (DDGS) from corn.
This high protein DDG has been proven to be an acceptable feed ingredient when up to 12% is included in a laying hen diet during peak production (Table 6).

Table 6. Effect of feeding high-protein corn DDG to laying hens on production characteristics (21-41 d of production).

Variable	Control	3% DDG	12% DDG
Hen-day egg production (%)	90.9b	93.8a	93.3a
Egg weight (g)	55.5ab	56.2a	55.0b
Egg mass* (g)	50.4b	52.7a	51.3b
Feed intake g/d*hen	92.6b	95.8ab	94.9ab
Feed intake/g egg mass	1.84ab	1.82b	1.85ab

*Egg mass = hen-day egg production * egg weight / 100
Adapted from Jung & Batal, 2009

Opportunities & threats

Cereals, grain legumes including European soybeans and their protein concentrates are a promising category of European produced high quality protein – level and digestibility – also for its application in organic diets. For legumes, their anti-nutritional factors have to be considered but there is a lot of knowledge available on this issue from European research programmes, some twenty years ago.

Leaf proteins

Characterization

For the longer term, protein enrichment of leafs/grasses might deliver attractive feed ingredients. Particularly grass protein concentrates seem to be promising because their development is already in the pilot stage. Lucerne and sugar beet leafs processing is in a less advanced stage than grass processing.
For the use of leaf proteins, only scarce information is available for the application in poultry diets, contrary to pigs diets. In organic broiler husbandry, access to a grass-clover covered pasture might substantially contribute to the protein supply of the broilers. Broilers were able to realize 7% of the recommended amount of protein by the intake of grass-clover from the pasture. Laying hens are able to consume considerable amounts of fresh grass, which might contribute for 12 – 13% of the total dry matter intake.
Protein content and digestibility of fresh grass depends on a number of factors, among others stage at harvest. Protein content of fresh grass, that was harvested at either a young or older (3 weeks later) stage, was 148 and 108 g/kg DM.

Extraction of protein from grass, thereby separating proteins from fibres, might increase the applicability in poultry diets. These proteins can be valorised as alternatives to extracted soybeans. Until now, hardly any information regarding nutritional value of grass proteins for monogastric animals is available. Extraction of protein may influence its properties and nutritive value. For example, protein digestibility can be reduced as a consequence of Maillard-like reactions, the formation of lysinoalaline, reactions with oxidized polyphenols, and racemization, finally resulting in a reduced uptake of lysine and other amino acids. Moreover, further development of protein extraction techniques is necessary to increase protein yield and to make these techniques economically feasible.

Opportunities & threats

Grass and lucerne hay to some extent can contribute to the protein supply of monogastric animals as described in literature for gestating sows and growing finishing pigs. The fibre content of these ingredients, however, may limit their use in monogastric diets. Bio-refinery might increase possibilities to use grass and lucerne by separating the protein and fibre fractions, but techniques should be further developed before application in practice. Consequently, at the moment, no firm conclusion regarding their potential use in poultry diets can be drawn.

Aquatic proteins

Characterization

Some aquatic proteins, e.g. micro algae and duckweed, might be valuable protein sources for pigs and poultry, whereas intact seaweed seems less suitable. In addition to the necessary development regarding protein extraction from these sources, more research is required to determine the nutritional characteristics of these ingredients, cell wall degradation characteristics, feed safety, and legislative aspects.

Processing to enhance the protein content of the aquatic resources algae and duckweed is still in its infancy. They may offer new opportunities on the long term (> 10 years).

Algae

Aphanizomenon flos-aquae (blue-green alga), Spirulina (blue-green alga), and Chlorella (green alga) are the most prominent protein-rich algae, which are commercially produced. Blue-green algae (also called cyanobacteria) are micro-organisms, because of their simple cellular structure. Some *Aphanizomenon* and *Spirulina* are toxic, however, the strains cultured for consumption do not contain toxins. In contrast to *Spirulina*, *Aphanizomenon* flos-aquae is able to fix nitrogen. Apart from their high protein content, health promoting properties are attributed to blue-green algae, such as the supply of unsaturated fatty acids, stimulation of the immune system, and protection from cancer. Intake of Chlorella extracts is associated with enhanced immune responses and antitumor effects.
Algae meal from *Spirulina platensis* was evaluated as a poultry feed ingredient in an experiment with broilers. The algae meal contained 344 g total ash and 423 g crude protein per kg dry matter. The ash content in that algae meal was high, compared to the 83 g/kg mentioned. The most dominant minerals in the ash fraction were sulphur, potassium, sodium and chloride, and these mineral contents have to be taken into account for the feed formulation. Amino acid composition showed a lower concentration of lysine, histidine and phenylalanine in proteins of the *Spirulina* meal than in protein of soybean meal, whereas the other essential amino acids were present in higher concentrations. Four diets, containing 0, 50, 100 and 150 g algae meal per kg diet were fed to male broiler chicks from 6 to 34 day of age. Feed intake was reduced in birds fed diets containing 100 and 150 g algae meal, whereas weight gain in birds fed these diets was decreased to less than 80% or the control group. Feed conversion ratio for diets with 0, 50 and 100 g algae meal was within the range of good commercial production, whereas birds fed the diet with 150 g algae meal consumed significantly more feed per unit weight gain. ME content was similar for the 4 diets, which suggests that the ME-concentration in the *Spirulina* meal was not much different from the mixture of maize, soybean meal, and wheat-starch.

Opportunities & threats

Results available from literature suggest that algae can be considered as a useful protein source in mono-gastric diets, especially when mass-cultured systems are considered. Further investigation, however, is needed concerning productive systems, contamination by bacteria, its nutrient composition (the cellulosic cell wall digestibility) and its nutritive value in poultry nutrition.

Threats: toxins/heavy metals, variable composition. At the moment, the nutritional benefits of algae cannot justify a systematic inclusion in poultry diets due to higher costs than that of conventional protein ingredients such as soybean meal.

Seaweed

The moisture content of fresh seaweed (marine algae) is very high and might amount up to 94% of the biomass. The nutritional composition of seaweeds varies, depending on strain, season and area of production. The total protein content varies among different seaweed strains in North-western Europe and is rather small in brown seaweed (10 – 24% of dry weight), whereas higher protein contents are observed in green and red seaweed species (up to 44% of dry weight).

The major components of unwashed/washed seaweeds are crude protein (11.14 and 12.42%) and crude ash (43.30 and 37.25%). Washing resulted in a change of the contents of some minerals and amino acids, e.g. Ca (2.20 and 38.6 g/kg), lysine (4.62 and 3.86), methionine (1.21 and 1.36 g/kg), cysteine (0.76 and 0.90 g/kg), threonine (3.48 and 3.77 g/kg) and tryptophan (1.17 and 1.03 g/kg) were determined. The main minerals in the ash fraction of the washed seaweed were magnesium (122 mg/g seaweed), chlorine (61 mg/g), calcium (39 mg/g)sodium (39 mg/g), potassium (33 mg/g), and phosphorus (28 mg/g). These high mineral contents have to be taken into account during feed optimization. No anti-nutritional factors were observed, except tannins at a very low level (1.50 and 1.67 g/kg). In vitro digestibility of dry matter was low (29.0 and 24.7%), probably due to the high content of inorganic matter and the presence of complex polysaccharides in seaweed. These authors concluded that the unwashed seaweed due to its high content of minerals and amino acids provides perspective for use as a supplement for animal feeding. This perspective, however, could not be confirmed in a digestibility experiment.

Ventura et al. (1994) stated that crude *Ulva rigida* seaweed is not a suitable ingredient for poultry diets, at least at inclusion rates of 100 g/kg or higher. These authors found a low AME_n value (2.9 MJ/kg DM) in a chick growth trial using diets containing 0, 100, 200 and 300 g seaweed per kg. As the content of seaweed was increased, feed intake and growth rate decreased ($P< 0.05$).

Opportunities & threats

Only a limited number of experiments has studied the nutritional value of seaweed for monogastric animals; seaweed in its present form is unsuitable as energy and protein source for poultry. Until now, no results are available of the nutritional value of extracted seaweed protein concentrates. Moreover, applying cell wall degrading techniques (e.g. pulse electric field) and enzymatic break down of structural carbohydrates and proteins might improve energy and protein digestibility of seaweeds. Further research is necessary to investigate these aspects.

Duckweed

On a dry matter base, duckweed has a high protein content with a valuable amino acid composition. Chemical composition of different duckweed species is shown in Table 7. Protein content, however, is also affected by harvest date and location.

Table 7. Chemical composition of different duckweed species (% of DM) (Rusoff et al., 1980).

Species	*L. Gibba*	*S. punctate*	*S. polyrhiza*	*W. columbiana*
Dry matter (%)	4.6	5.2	5.1	4.8
Crude protein	25.2	28.7	29.1	36.5
Crude fat	4.7	5.5	4.5	6.6
Crude fibre	9.4	9.2	8.8	11
Ash	14.1	13.7	15.2	17.1

The amino acid composition of several duckweed species is shown in Table 8.

Table 8. Production levels of egg-laying birds at 18 weeks fed diets containing different levels of dehydrated *lemna gibba* meal from sewage water (33% N x 6.25 in DM).

	Level of dehydrated duckweed, %		
	0	25	40
Feed consumption (g/d)	131	131	125
No of eggs/week/hen	5.9	5.9	5.6
Mean egg weight (g)	64.2	63.1	63.6
Feed conversion (g DM/g egg)	2.41	2.47	2.38
Live weight (g)	40	114	-118

(Haustein et al., 1988)

Obviously, very high levels of duckweed may decrease performance but substantial levels can be used to replace protein sources as SBM or fish meal. However, some variable responses are reported depending on the source of duckweed which can be high protein/low fibre or low protein/high fibre depending on the nutrients in the growth medium.

In vitro digestibility of duckweed was rather low and ranged from 60 – 63%, whereas analysed levels of dioxin and arsenic were above European legal standards (Hoving et al., 2012). Two duckweed species (*Lemna gibba and Wolffia arrhiza*) were added to diets of laying hens, thereby replacing soybean meal and fish meal. Duckweed was first sun dried to 40% DM, followed by forced air drying for 15 to 30 min to 90% DM. The optimal Lemna level in the diet of laying hens was 15%, but even at an inclusion level of 40% egg quality remained unaffected. The authors concluded that duckweed can be used to replace soybean meal and fish meal in diets of laying hens.

In contrast to adult laying hens, broiler chickens that were fed diets containing various levels (0 – 400 g/kg) of *Lemna gibba* showed a decreased feed intake and growth rate as the level of *Lemna gibba* increased.

Opportunities & threats

There is a tremendous potential for the production of duckweeds (10-18 ton/ha/y; Table 3). Despite this potential and its nutritive value, there are numerous impediments to these plants being incorporated into western farming systems. Large genetically determined variations in growth in response to nutrients and climate, apparent anti-nutritional factors, concerns about sequestration of heavy metals and possible transference of pathogens raise questions about the

safety and usefulness of these plants when grown under natural conditions and not cultivated under controlled conditions. A clear understanding of how to address and overcome these impediments needs to be developed – as are drying procedures and storage (sensitivity for rotting) and legislation – before duckweed is widely accepted for nutrient reclamation and as a source of animal feed. Genetic variations in in growth and responses to nutrients and climate conditions, however, provides the possibility for further selection of duckweed species that optimally fits in European farming systems.
Duckweed has the ability to accumulate heavy metals, so pollution in reservoirs has to be taken into consideration whereas also the presence of tannins/phenolic compounds was shown by Fasakin (1998). However, duckweed species *L. Gibba* can replace conventional protein sources up to 15% of the diet inclusion level without affecting production characteristics and body composition in laying hens (Haustein et al., 1994). However, variable responses to duckweed proteins are reported depending on the nutrients in the growth medium.

Insect proteins

Characterization

Already in 1919, Lindner suggested that larvae of the common housefly could be used for the production of proteins and fat using human waste as a substrate (Lindner, P., 1919: Extraction of fat from small animals. *Z. Tech. Biol.* 7: 213-220).

The use of insects or its protein fraction as a sustainable protein rich feed ingredient in poultry diets is technically feasible. Insects can be reared on low-grade bio-waste and can turn bio-waste into high quality proteins, but opinions differ whether this is possible within 5 years. Insects therefore can be an interesting link in the animal feed chain to fulfill the globally increasing demand for protein.
Cultivation and processing insects and their inclusion in feeds for poultry production seems a promising innovation because of their high efficiency in making valuable nutrients. Insects have a well-balanced nutrient content; they have the same or an even better amino acid profile compared to soybean meal and fishmeal for use in pig and poultry diets. A rich content of polyunsaturated fatty acids, micronutrients and vitamins can be attained too. Furthermore, one must not forget the beneficial properties of the polysaccharide chitin, which is also found in insects. The use of insects has already been analysed for poultry because insects are already part of their natural diet and poultry is the second mostly eaten meat in the world.
Three insect species are most suitable to use in poultry diets due to their high amount of protein and their ability to degrade organic waste. These species are the Black Soldier Fly (BSF; *Hermetia illucens*), the common Housefly (*Musca domestica*) and the yellow Mealworm (*Tenebrio molitor*).
To concentrate on the BSF, to use larvae as a protein source in feed, certain steps within the process need to be taken: the larvae have to be separated from the substrate on which they grow. Manure seems to be a good substrate for larvae due to the high growth rate of the insects. After separation, the larvae need to be killed, washed and dried. During drying by heated air, temperatures should not exceed 70°C to prevent denaturation of the protein and with it the essential amino acids. After drying, the larvae could be milled to obtain larvae meal. Fat can be extracted from the full larvae meal to increase the protein content. One of the drawbacks is that using insects for animal feed is currently not possible in Europe due to legislation, which prohibits the use of processed animal proteins in feed for pigs, poultry, and ruminants. At the moment, discussions about revising legislation are kept and a clarification is expected in 2014.
Safety studies have shown that insects represented no health issues because microorganisms that infect insects are not able to contaminate vertebrates such as poultry, and processing techniques are available to prevent any pathogens or toxins from appearing within the feed.

At this moment, producing insects as an alternative protein source is still too expensive compared to conventional protein sources but several improvements or valorisations of insects properties could be achieved in order to make it more attractive. Within processing, costs could be reduced by increasing the level of automation and mechanization, and reducing energy consumption of the installations. Moreover, chitin and residues of manure have an economical value and could be separated and used for other applications. Finally, companies rearing insects could benefit from subsidies because of the reduction of nitrogen in manure and water pollution. Information on the nutritive Value is available for a large number of insect species, often about the common house-fly mealworm larvae. The protein content of insects widely varies across insect species and also within insects species and life stages (Figure 1).

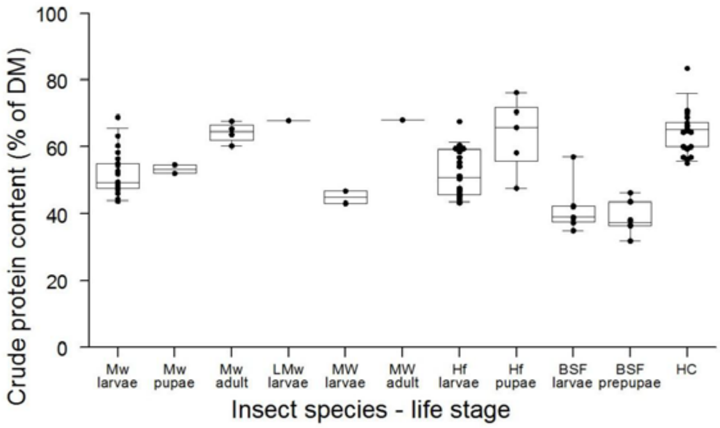

Figure 1. Insect protein levels (E. Ottevanger & G. Bosch, 2013; pers. communications)

Highest median crude protein content was found for common house-fly pupae (65.7% of DM) and lowest for the black soldier fly larvae (38.9% of DM) and pre-pupae (37.4% of DM).

Diener et al. (2009) concluded from their small-scale experiments. that biomass produced black soldier flies (BSF) could be a low-tech based waste treatment. These converters of organic wastes can serve as a nutritious ingredient for chicken feed, resulting in 42-45% protein and 31-35% fat. Yellow mealworm larvae and house-fly were comparable in CP content (median around 50% of DM). few data were available for the yellow mealworm pupae (n=2; 53.4% of DM), lesser mealworm larvae (n=1; 67.9% of DM) and super worm larvae (n=2; 45.0% of DM). General data on amino acid composition and its digestibility are promising although there is a need for evaluating nutrient digestibility of (processed) insects as feed ingredients before formulating insect-containing poultry feeds.

Opportunities & threats

To introduce insects as a feed ingredient in the poultry feed chain, additional research is recommended on its feeding value, functional properties, safety when using bio-wastes as a rearing substrate, extraction/use of nutrients (e.g. chitine) and use of left-overs/residue products after production. A low carbon footprint facilitates insects such as meal worms and black soldier flies to be used as feed ingredients.

Main bottlenecks were identified in the area of legislation, both approval in poultry diets and environmental being an obstacle for large-scale production of insects. Moreover, product safety (dioxin, toxic metals) and the achievement of a low cost price by an automation of the production process should be guaranteed.

Microbial proteins

Characterization

Øverland et al. (2010) reported that methanotrophic bacteria are able to efficiently convert natural gas (methane; methanol) into high-quality proteins by anaerobic fermentation and, hence, may be a good alternative for sustainable protein sources. Bacterial protein meals (BPM) or autolysates (BPA) were evaluated in poultry diets where the proteins meals especially showed to be a suitable protein source for broiler chicken. With BPM, improved growth performance of broiler chicken (Table 9) and litter quality was observed.

Table 9. BPM concentrations and growth performance of broiler chicken

	BPM g/kg			
	0	40	80	120
Weight gain (g)	2213	2146	2154	2167
Feed intake (g)	3404[a]	3174[b]	3209[b]	3171[b]
Feed conversion efficiency	0.65[a]	0.676[b]	0.672[b]	0.684[b]
Mortality (*n*)	3	1	4	1

Øverland et al. (2010)

Adding BPM to diets reduced feed intake but improved weight gain:feed overall up to 35 days. No effects on viscosity of diets was observed and minor effects on litter quality.

Opportunities & threats

Bacterial protein meals have a tremendous potential for protein production (50 tons/d ...), have a high nutritional value and balanced amino acids composition and a high digestibility. A positive effect on carcass quality was observed (Hellwing et al., 2006). It is a low risk ingredient, free of dioxins, with no anti-nutritive factors and GMO-free. Bacteria generally have a high content of nucleic acid, non-selective optimal pH, and high risks of toxicity by endotoxins and membrane substances. Because of their small size, they are most difficult to separate.

Considerations for the use of unconventional protein sources.

In Table 10 (next page), the characteristics of the selected protein sources with respect to protein yield, nutritional value, CFP, LuLuc, N-requirement, estimated period of coming commercially available in the EU, and applicability in organic diets are summarized. Wheat gluten meal is added as a reference ingredient.

Conclusions

As general conclusions, recommendations are given for plant breeding aspects to improve the yield of production of European soybeans and oats; large scale cultivation, processing and feasibility of aquatic, leave and insect proteins for application as feed still needs more research.

Enhancing plant protein content by processing needs further development, especially for duckweed and algae, including sustainability aspects.

Finally, the nutritional value of products such as leaf proteins, some aquatic proteins and insects is not known sufficiently. Moreover, aspects of cell wall degradation, feed safety and legislation should be covered.

Table 10. Some characteristics of selected protein sources under EU-conditions

	Protein yield/ha[1]	Nutritional value for the animal[2]	Carbon Footprint[3]	Luluc[4]	N-Require-ment[5]	Availability In EU on short term[6]	Applicable in organic diets[7]
Cereal co-products							
Wheat gluten meal	+	+/+	+	+/+	+/-	+/+	+
Defatted European oil seed products							
Soybean meal	+	+/+	+/-	+	+	+/-	-
Soybean concentrate	+	+/+	+/-	+	+	+/-	+
Rapeseed meal	+/-	+/-	+/-	+/+	-	+/+	-
Rapeseed concentrate	+/-	+/-	+/-	+/+	-	+/+	+
Sunflower meal	+/-	+	+	+	+	+/+	-
Sunflower concentrate	+/-	?	+	+	+	-	+
Grain legumes							
Pea	+	+/+	+/-	+/-	+	+/+	+
Pea concentrate	+	+/+	+/-	+/-	+	+/+	+
Vicia faba	+	+/-	+/-	+	+	+/+	+
Vicia faba concentrate	+	+/+	+/-	+	+	+	+
Lupine	+/-	-	+/-	-	+	+/+	+
Lupine concentrate	+/-	?	+/-	-	+	+	+
Chickpea	-	?	?	?	?	+/-	+
Forage legumes							
Lucerne	+	-	-	+	+	+/+	+
Leaf proteins							
Grass protein	+	-	-	?	?	+	+
Sugar beet protein	-	-	-	+/+	+/+	-	+
Aquatic proteins							
Algae	++	?	-	+/+	?	+/-	?
Seaweed	+/+	-	-	+/+	+/+	-	?
Duckweed	+/+	?	-	+/+	?	+/-	?
Cereal protein							
Oat protein	+/	+/-	+	+	-	+/-	+
Quinoa protein	-	?	?	?	?	?	+
Insects	+/+	?/+/-	?	+/+	+/+	+/-	?
Microbial proteins	+/+/+	?	?	?	?	?	?

[1]) - = < 500 kg/ha; +/- = 500 – 1000 kg/ha; + = 1000 – 2000 kg/ha; ++ = > 2000 kg/ha
[2]) - = Ileal ??Protein digestibility < 75%
+/- = Protein digestibility > 75% and < 80%
+ = Protein digestibility > 80% and < 85%
++ = Protein digestibility > 85%
[3,4,5]) - = CFP > 1000 CO_2-eq; LuLuc > 1000 CO_2-eq; N-requirement > 50 g N/kg yield
+/- = CFP > 500 CO_2-eq; LuLuc > 500 CO_2-eq; N-requirement > 25 g N/kg yield
+ = CFP > 250 CO_2-eq; LuLuc > 250 CO_2-eq; N-requirement > 10 g N/kg yield
+/+ = CFP < 250 CO_2-eq; LuLuc < 250 CO_2-eq; N-requirement < 10 g N/kg yield
[6]) - = > 10 years +/- = 5 – 10 years + = 0 – 5 years +/+ = currently available
[7]) - = no

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