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Partner in livestock innovations

Cultivation, processing and nutritional aspects for pigs and poultry of European protein sources as alternatives for imported soybean products

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Abstract
In this study, the conditions are described for
successfully cultivation, processing and
applying alternative protein sources in (organic)
pig and poultry diets under European climatic
conditions, thereby taking sustainability
characteristics, and legislative aspects into
account.

Keywords
Alternative proteins, cultivation, nutrition,
processing, soybeans

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Cultivation, processing and nutritional aspects for pigs and poultry of European protein sources as alternatives for imported soybean products

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Samenvatting

De totale eiwitproductie in de EU, o.a. via de teelt van leguminosen en sojabonen, bedraagt slechts 3% van het areaal Europese landbouwgrond. Daarom worden in de EU grote hoeveelheden eiwit geïmporteerd, hoofdzakelijk vanuit Zuid-Amerika. In toenemende mate is er bezorgdheid over deze eiwitimporten, hoewel de redenen van bezorgdheid verschillen tussen stakeholders. NGO's zijn met name bezorgd vanwege het verlies van natuurlijke ecosystemen en biodiversiteit, de toenemende water- en bodemvervuiling en het verdrijven van kleine boeren en inheemse bevolking in de producerende landen. Het Europese Parlement is met bezorgd vanwege de toenemende afhankelijkheid van Zuid-Amerika. Bevordering van de Europese eiwitproductie geeft akkerbouwers in de EU meer mogelijkheden voor gewasrotatie, waardoor de kans op plantenzieken vermindert, met uiteindelijk een grotere mate van economische stabiliteit. Door gewassen in Europa te telen, is er meer gelegenheid om dit op maatschappelijk gewenste wijze te doen. Een voorbeeld hiervan is de teelt van GMO-vrije soja.

Op voordracht van de Task Force Voer – Mest kringlopen, en in opdracht van het Ministerie van Economische Zaken hebben onderzoekers van Wageningen UR Livestock Research (WLR), Plant Research International (PRI) en Food & Biobased Research (FBR) gezamenlijk de mogelijkheden van meer Europese eiwitproductie onderzocht. Het doel van dit onderzoek was om de voorwaarden te beschrijven die nodig zijn voor het onder Europese omstandigheden succesvol telen, bewerken en toepassen van alternatieve eiwitbronnen in (biologische) varkens- en pluimveevoeders, rekening houdend met aspecten van duurzaamheid en wetgeving. De biologische veehouderijsector staat voor nieuwe uitdagingen als gevolg van gewijzigde regelgeving met betrekking tot regionale teelt van voedermiddelen en het volledige verbod op het gebruik van niet-biologisch geteelde grondstoffen in het voer. Om hieraan te kunnen voldoen is er in de biologische veehouderij dringend behoefte aan regionaal geteelde hoogwaardige eiwitbronnen. Daarom is de biologische veehouderij een geschikte sector voor het op korte termijn toepassen van nieuwe eiwitbronnen en in dit opzicht een voorloper functie vervullen voor de gangbare intensieve veehouderij.

Allereerst is een 'long list' met 62 uiteenlopende eiwitrijke diervoedergrondstoffen opgesteld (Bijlage 1). Vervolgens is een aantal criteria toegepast, waarna de grondstoffen zijn overgebleven die in potentie een bijdrage kunnen leveren aan het vergroten van de Europese eiwitproductie. Deze criteria waren:
- Het gewas moet in staat zijn om een goede opbrengst te leveren onder de klimaatomstandigheden die in Noordwest Europa voorkomen;
- Het telen van het gewas in Europa is op dit moment nog geen gangbare praktijk;
- Het gewas is ook op de lange termijn (na 2020) nog steeds beschikbaar als grondstof voor diervoeders en niet uitsluitend als grondstof voor humane voeding.

Op basis van deze criteria zijn de eiwitbronnen die weergegeven zijn in Tabel 1 geïdentificeerd als potentieel interessante alternatieven.

Tabel 1 'Short list' van potentieel interessante eiwitbronnen voor het verhogen van de Europese eiwitproductie ten behoeve van de diervoedersector

Categorie	Eiwitbron
Oliehoudende zaden	Eiwitten na ontvetting van sojabonen, raapzaad en zonnebloemzaad
Vlinderbloemige zaden	Erwten, veldbonen, lupinen en concentraten hiervan, kikkererwten
Vlinderbloemigen (blad)	Lucerne
Blad eiwitten	Gras, bietenblad
Aquatische eiwitten	Algen, zeewier (macro algen) en eendenkroos (micro algen)
(Pseudo) granen	Eiwitten van haver en quinoa
Insecten	O.a. meelworm, huisvlieg en huiskrekel[1]

[1]) De voedingskundige aspecten van insecten zijn onderzocht in een apart project. De resultaten zijn gepubliceerd in het rapport 'Insects as a sustainable feed ingredient in pig and poultry diets – a feasibility study' (Veldkamp et al., 2012).

Teeltkundige aspecten

Vanuit teeltkundig oogpunt zijn vlinderbloemige zaden, met name erwten en bonen, interessante alternatieve eiwitbronnen. Ze hebben een hoog ruw eiwitgehalte (17 – 35%) en teeltkundige technieken zijn reeds beschikbaar en geïmplementeerd. Een nadeel is echter dat deze gewassen erg gevoelig zijn voor ziekten en plagen. Het hoge eiwitgehalte maakt de teelt van Europese sojabonen interessant, hoewel het opbrengstniveau op dit moment te laag is om het gewas concurrerend te laten zijn met tarwe. Veredeling is noodzakelijk voor het verhogen van het productieniveau, waarbij in Europa met name behoefte is aan een variëteit met een extreem kort groeiseizoen. Raapzaad wordt al behoorlijk veel verbouwd in Europa met ook een redelijke eiwitopbrengst per hectare is, maar in Nederland is deze teelt nog zeer bescheiden. Haver en quinoa bevatten kwalitatief goed eiwit, maar in vergelijking met tarwe is het opbrengstniveau van deze gewassen op dit moment veel te laag. Verwacht wordt dat het opbrengstniveau door middel van intensieve veredeling wel op het niveau van tarwe gebracht kan worden. Aquatische eiwitten zijn erg perspectiefvol, omdat sommige bronnen een hoog eiwitgehalte hebben (o.a. eendenkroos, sommige algensoorten en zeewiersoorten) en een zeer hoge opbrengst per hectare. De bewerkingsstappen die nodig zijn voor het daadwerkelijk kunnen toepassen van deze grondstoffen in diervoeder vragen echt nog veel onderzoek. Niet alleen de hoge opbrengst per hectare, zoals bijv. bij eendenkroos, en het hoge eiwitgehalte, maar ook het feit dat ze geen beslag leggen op land met een agrarische bestemming maakt deze potentiële eiwitbronnen interessant. Voor alle eiwitbronnen met een hoog watergehalte, zoals bladeren, eendenkroos, algen en zeewieren, is een droogstap voorafgaand aan transport en opslag noodzakelijk.

Bewerking

Het verder bewerken van grondstoffen, zodat onder andere anti nutritionele factoren (ANFs) verwijderd worden of het eiwitgehalte verhoogd wordt tot 65% of meer, kan een manier zijn om te voorzien in de vraag naar hoogwaardig eiwit in allerlei soorten biologische voer of in gangbare voeders voor jonge dieren (biggen, vleeskuikens, opfokhennen). Dit rapport beschrijft diverse bewerkingsmethoden voor het verhogen van het eiwitgehalte van de potentiele eiwitbronnen en gaat ook in op de economische aspecten hiervan. De duurzaamheidsaspecten ten aanzien van het water- en energieverbruik worden globaal besproken. In zijn algemeenheid kan gesteld worden dat bewerkingsmethoden voor het verhogen van het eiwitgehalte van de geselecteerde eiwitbronnen verder doorontwikkeld dienen te worden en op dit moment nog niet standaard toegepast worden. Op korte termijn lijkt verdere verhoging van het eiwitgehalte bij de volgende grondstoffen perspectief te bieden:

- Met betrekking tot oliehoudende zaden: raapzaadeiwitconcentraat. Verdere verhoging van het eiwitgehalte van ontvet zonnebloemzaadschroot lijkt minder perspectiefvol.
- Met betrekking tot vlinderbloemigen: eiwitconcentraten die verkregen worden door het droog fractioneren van erwten en veldbonen. Deze concentraten zijn overigens al wel commercieel verkrijgbaar. Lupineconcentraat lijkt minder perspectiefvol.

Op lange termijn kan het concentreren van eiwit uit gras en andere bladeren waardevolle voedermiddelen opleveren. Dit geldt vooral voor graseiwit, omdat de ontwikkeling hiervan zich al in het pilotstadium bevindt, terwijl de technieken voor het extraheren van eiwit uit lucerne en bietenloof op dit moment minder ver ontwikkeld zijn. De technieken voor het concentreren van het eiwit uit aquatische grondstoffen zoals eendenkroos en algen staan op dit moment nog in de kinderschoenen. Verwacht wordt dat er op de langere termijn (> 10 jaar) ook voor deze grondstoffen perspectiefvolle bewerkingsmethoden zullen komen.

Diervoedingsaspecten

Eiwitten afkomstig van oliehoudende zaden zijn zeer goed bruikbaar in varkens- en pluimveevoeders. Op dit moment worden sojaschroot, raapzaadschroot en zonnebloemzaadschroot ook al breed toegepast in deze voeders. De chemische samenstelling en nutritionele waarde van deze grondstoffen zijn inmiddels ook al goed onderzocht. We veronderstellen dat de nutritionele kenmerken van sojaschroot van Europees herkomst niet verschilt van die van Zuid-Amerikaanse herkomst, hoewel hierover nog geen onderzoeksgegevens beschikbaar zijn. Over de diervoedingskundige eigenschappen van de eiwitconcentraten van deze grondstoffen is echter nog niet veel informatie beschikbaar. Uit het enige experiment dat hierover beschikbaar is blijkt dat raapzaad (canola) eiwitconcentraat tot een niveau van 10% probleemloos toegepast kan worden in voeders van gespeende biggen.

Vlinderbloemigen, zoals veldbonen, lupinen en erwten, en kikkererwten kunnen een aanzienlijke bijdrage leveren aan de eiwitvoorziening van varkens en pluimvee, hoewel er wel rekening gehouden dient te worden met de in deze producten aanwezige ANF's. Uit een onderzoek met gespeende

biggen bleek dat de verteerbaarheid van erwtenconcentraat vergelijkbaar of zelfs beter was dan van erwten zelf. Ook deden de biggen die voer met erwtenconcentraat kregen het erg goed met betrekking tot voeropname, groei en voederconversie. Op basis hiervan, en rekening houdend met het feit dat eiwitconcentraten uit vlinderbloemigen duurzaam verkregen kunnen worden én ook al commercieel beschikbaar zijn, concluderen we dat deze concentraten een perspectiefvolle categorie van hoogwaardig Europees eiwit vormen, met name voor toepassing in biologische voeders.

Op dit moment zijn de nutritionele eigenschappen van bladeiwitten nog niet bij varkens en pluimvee onderzocht.

Bepaalde aquatische eiwitten, zoals microalgen en eendenkroos, lijken waardevolle eiwitbronnen voor varkens en pluimvee te zijn, terwijl intact zeewier hiervoor minder geschikt lijkt. Naast het eerder genoemde onderzoek naar gewenste methoden om het eiwit uit deze grondstoffen te verwijderen, is het ook noodzakelijk om onderzoek te doen naar de voedingskundige aspecten, naar technieken voor het ontsluiten van celwanden en naar aspecten van voedselveiligheid en wetgeving.

Havereiwit is een hoogwaardige eiwitbron voor eenmagigen en is geschikt voor toepassing in voeders voor jonge biggen. Hoewel quinoa in potentie geschikt lijkt als hoogwaardige voedingsbron is er op dit moment onvoldoende informatie beschikbaar om betrouwbare uitspraken te doen over toepassing van deze grondstof in varkens- en pluimveevoeders.

Conclusies

Er zijn aanzienlijke verschillen tussen de geselecteerde eiwitbronnen met betrekking tot de beschreven duurzaamheidsaspecten. We gaan ervan uit dat producten met een hoog vochtgehalte, zoals lucerne, bladeren en aquatische eiwitten, vanwege de hoge energiekosten voor het drogen van deze producten minder duurzaam zijn. Van sommige eiwitbronnen zijn echter nog geen ecologische voetafdrukwaarden bekend. Ook dient nog onderzoek plaats te vinden naar de duurzaamheidsaspecten van eiwitextractie technieken. Daarom is het op dit moment nog niet mogelijk om weloverwogen conclusies te trekken ten aanzien van de duurzaamheidsaspecten van deze eiwitbronnen.

Binnen de groep van oliehoudende zaden lijkt sojaschroot van Europese herkomst het meest perspectiefvolle alternatief voor Zuid-Amerikaanse sojaschroot. Sojaschroot kenmerkt zich door een hoge nutritionele waarde, met name vanwege een hoog gehalte aan goed verteerbaar eiwit. Om het gewas aantrekkelijk te maken voor een akkerbouwer dient de opbrengst per hectare echter aanzienlijk verhoogd te worden. Hiervoor is veredeling van het gewas noodzakelijk, waarbij met name geselecteerd dient te worden op rassen met een ultrakort groeiseizoen.

Binnen de categorie van de vlinderbloemige zaden lijken erwten, in elk geval op de korte termijn, het beste alternatief voor sojaschroot. Het gewas heeft een redelijke eiwitopbrengst per hectare, hoewel dit verder verhoogd zou moeten worden. Op langere termijn kunnen ook bladeiwitten en aquatische eiwitten bijdragen aan vermindering van de soja importen. Daarvoor is eerst wel meer onderzoek nodig naar bruikbare technieken voor het extraheren van het eiwit en naar de diervoedingskundige waarde van deze grondstoffen.

Bladeiwitten en aquatische eiwitten leggen geen beslag op de bestaande landbouwgronden. Als er meer eiwitrijke gewassen in Europa geteeld worden, kan dit ten koste gaan van de teelt van granen en knolgewassen (aardappelen en suikerbieten). Op dit moment is de EU ruim zelfvoorzienend voor aardappels en precies zelfvoorzienend voor suikerbieten. Experts verwachten echter een afname van de teelt van suikerbieten met 30% op het moment dat de vaste (hoge) prijs van suiker wordt losgelaten. Dit geeft aan dat er binnen het beschikbare areaal landbouwgrond in Europa nog enige ruimte is voor de teelt van eiwitrijke gewassen. Daarnaast wordt aangenomen dat er nog 1,8 miljoen ha braakliggend land beschikbaar is in de Donau-regio in het Zuidoosten van Europa. De totale hoeveelheid land die in Europa in potentie beschikbaar is voor de teelt van eiwitrijke gewassen wordt geschat op ten minste 2,4 miljoen ha.

Summary

The total EU protein crop production (e.g. legumes, soybeans) currently occupies only 3% of the EU's arable land. Therefore, large amounts of protein are imported in the EU, mainly 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 biodiversity, the increased water and soil pollution, and the ejection of small farmers and the native population in the producing countries. The European Parliament is worried about the increased dependency from South America. Enhancing the EU protein crop production enlarges the possibilities for crop rotation, thus reducing 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.

On the recommendation of the Dutch Task Force Feed – Manure Cycles, and on request of the Dutch Ministry of Economic Affairs, scientists from Wageningen UR Livestock Research (WLR), Plant Research International (PRI) and Food and Biobased Research (FBR) have collaboratively investigated the options to increase European protein production. The aim of this study was to describe the conditions for successful cultivation, processing and use of alternative protein sources in (organic) pig and poultry diets under European climatic conditions, thereby taking sustainability characteristics, 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 appropriate sector for applying new protein sources and may also serve as an example for increased use of novel protein sources in conventional intensive animal production systems.

First of all, a long list of 62 feed ingredients (Annex 1) was composed, containing a wide range of 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;
- In 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 1).

Table 1.   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)
Leaf proteins	Grass, sugar beet leaves
Aquatic proteins	Algae, both macro- (seaweed) and microalgae, duckweed
Cereals and pseudo cereals	Proteins from oat and quinoa
Insects	E.g. mealworm, housefly, house cricket[1]

[1]) The nutritional aspects of insects were studied in a separate project and presented in the report 'Insects as a sustainable feed ingredient in pig and poultry diets – a feasibility study' (Veldkamp et al., 2012).

Plant cultivation
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, but is not cultivated at a large scale in the Netherlands. The protein quality of oats and quinoa are interesting, but at this moment the yield per hectare is low compared to wheat. It still needs intensive breeding input, but with adequate attention from breeders, the production level could reach that of wheat. Aquatic protein sources are very interesting because of a high protein content (duckweed, several micro-algae and some macro-algae) and very high yields, but processing and feasibility for application as feed 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 required.

Processing
Further 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). Processing to enhance protein content of these potential protein sources is described and economic aspects of this processing are discussed. The sustainability of the processes in terms of energy and water use is briefly addressed. 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 dry fractionation from peas and faba beans. The former are already on the market. Lupines are less attractive.
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.
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).

Nutritional aspects
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.
Legumes, e.g. *Vicia faba*, lupines and peas, and chickpeas, can significantly contribute to the protein supply of pigs and 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. Moreover, piglets that were fed a diet supplemented with pea protein concentrate performed very well. 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.
The nutritional value of leaf proteins for pigs and poultry has not been studied yet.
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.
Oat protein has a good nutritional value for monogastrics and can be used as high quality protein in diets for young piglets. Although quinoa might have some promising nutritional properties, current knowledge is not sufficient for accurate supplementation of this ingredient in diets of pigs and poultry.

Conclusions
The selected protein sources differ substantially in terms of environmental sustainability. 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. Values are lacking for some of the sources. Besides, more research is necessary to determine if protein extraction processes are sustainable. At this moment it is not possible to draw sound conclusions with respect to the environmental sustainability of the selected 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 seam 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 and aquatic protein sources are not in direct competition with the land use of other crops. Protein crops have to compete with cereals and root crops (potato, sugar beet) in Europe. Currently, the EU is more than self-sufficient for potatoes. We are self-sufficient for sugar beets as well, but experts expect a drop of 30% in production after the soon expected release of the fixed (high) price for sugar. Therefore, some arable land in the EU might be available for protein crop cultivation. Moreover, it is estimated that 1.8 million ha of fallow land is available in the Danube region, the South-East part of Europe. The estimated minimum land potential for cultivation of protein crops in Europe is 2.4 million ha.

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1 Introduction

The total EU protein crop production (e.g. legumes, soybeans) currently occupies only 3% of the EU's arable land (Euractiv, 2011). In 2009, 42 million tonnes of soybeans and soybean cakes, equivalent to 17.5 million ton protein, were imported in the EU (FAOSTAT, 2009). These protein sources mainly originated from South America. In terms of land use abroad, these imports represent 10% (20 million ha) of the EU's arable land (Euractiv, 2011). Although it is stated that 80% of EU's consumption of protein crops is imported (Euractiv, 2011), on a nitrogen base 14% (import is 1.7 billion kg; total use in feed is 12.2 billion kg) of all feed proteins originate from non-EU countries (Westhoek et al., 2011), as shown in Figure 1.
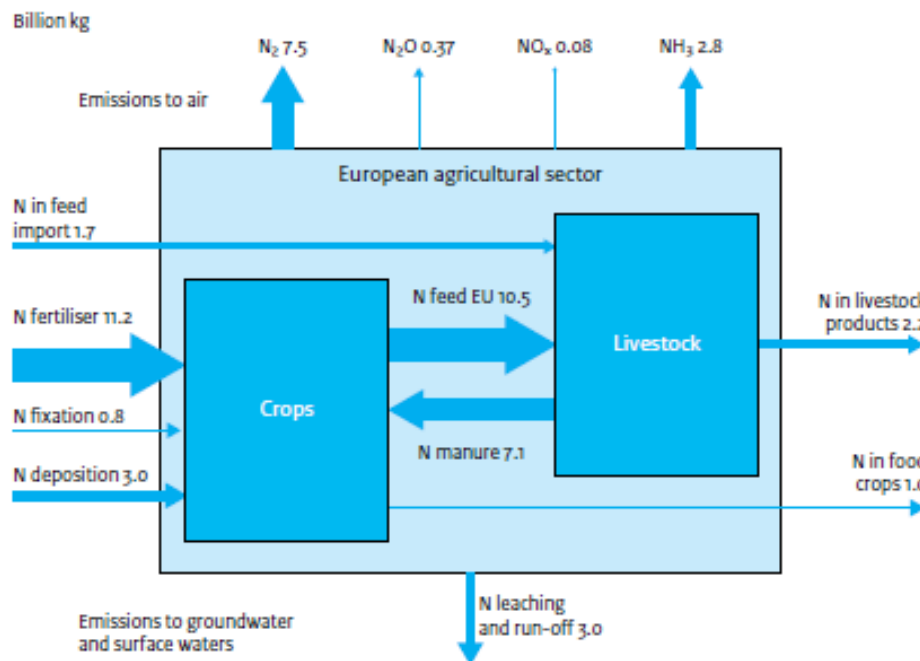
Figure 1. Nitrogen flows in agricultural sector in EU_{27}, 2005 (Westhoek et al., 2011).

There are increasing concerns about the amount of imported feed proteins from outside the EU. The reasons of concern differ between stakeholders, e.g. governments, NGO's, and consumers. In 2011, The European Parliament adopted a resolution on 'the EU's protein deficit', putting forward a series of measures to reduce the dependency on imports of protein crops for animal feed, primarily from the US, Argentina, and Brazil (Euractiv, 2011). The European Parliament is concerned that such massive dependency on imports makes the EU livestock sector extremely vulnerable to price volatility and trade distortions, causing feed price to rise, thereby increasing farmers' production costs and reducing the sectors' profitability. A major concern of NGO's is the deforestation of tropical rain forest, to fulfil the need of arable land for soybean cultivation (WNF, 2011 ; Van Gelder and Kuepper, 2012). As a consequence of conversion of natural ecosystems into agriculture, the rate of biodiversity loss (proportion of extinct species) increases, whereas the current status has already more than ten times exceeded the proposed boundary (Rockström et al., 2009). Moreover, large scale soybean cultivation may increase water and soil pollution, and drive small farmers and the native population out of business (WNF, 2011). Other reasons for enhancing the EU protein crop production are the increased possibilities for crop rotation, which might reduce sensibility for crop diseases and might stabilise farmers' income, and an increased influence on socially desirable cultivation (e.g. non GMO soybean production) (Westhoek et al., 2011). According to Westhoek (2011), however, EU protein crop cultivation will not positively affect the EU phosphate balance, whereas there is a risk on increased land use and consequently a higher use of phosphate fertilizer. The world stock of phosphorus is rather limited, and therefore a very conscious use of this mineral is required. A too high level of phosphate fertilization might result in phosphate emission to ground and surface water, consequently affecting aquatic and marine and biodiversity.

As a follow up on a public debate regarding sustainable livestock production, the Provincial Executive of Brabant (the Netherlands) installed a commission that has to develop concrete ambitions for sustainability of livestock in 2020 (Commissie Van Doorn, 2011). One of the formulated goals of this commission was that in 2020 at least 50% of the protein-rich feed should originate from Europe (27% in 2011), under the condition that this results in a more sustainable feed production compared to the current situation. If Europe still should import protein-rich ingredients from outside Europe, from January 2014 onwards these ingredients should be produced according to sustainable standards. In case of soybean meal and Palm oil, these standards are according to the requirements of RTRS (Round Table on Responsible Soy) and RSPO (Round Table on Sustainable Palm Oil), respectively. In the Netherlands, also the Task Force Feed – Manure Cycles exists. In this Task Force, the Dutch Ministry of Economic Affairs, the feed industry, NGO's, farmer representatives, and scientists are developing practical solutions for creating more balance in mineral cycles, and for enhancing the European protein production.

On the recommendation of the mentioned Task Force and on request of the Dutch Ministry of Economic Affairs, Agriculture, and Innovation, scientists from Wageningen UR Livestock Research (WLR), Plant Research International (PRI) and Food and Biobased Research (FBR) have collaboratively studied the options to increase European protein production. The aim of this study was to describe the conditions for successful cultivation, processing and applying use of protein sources in (organic) pig and poultry diets under European climatic conditions, thereby taking sustainability characteristics, and legislative aspects into account. Because of new regulations with respect to regional cultivation of feed and the ban on the use of conventional feed ingredients including synthetic amino acids, the organic livestock production has an urgent need for regionally produced high quality protein sources. Therefore, the organic sector seems to be an appropriate sector for applying new protein sources and may also serve as a pilot for increased use of novel protein sources in conventional intensive animal production systems.

2 Criteria for selected protein sources

First of all, a long list of 62 feed ingredients (Annex 1) was composed, containing a wide range of protein sources. Then, a list of considerations was developed with the final aim to select the sources that potentially contribute to increase the European protein production. These considerations were:
- Are the climate and other conditions in North West Europe appropriate for adequate crop performance?
- Are the climate and other conditions in South East Europe appropriate for adequate crop performance?
- Is the protein source applicable in diets of young animals (piglets, broilers)?
- Is the protein source applicable in diets of adult animals (growing finishing pigs, sows, laying hens, turkeys)?
- Is it allowed to include the protein source in organic diets?
- Is further processing of the protein source before adding it into the diet necessary?
- Is the protein source sustainable in terms of CO_2 footprint?
- What legislative aspects may need to be solved before the protein source can be used in animal diets?
- Is the protein source expected to be available for feed in long term (> 2020) or is the use limited to food applications?
- Is the protein source already commonly used in feed already common practice so that no further attention is necessary, or not?

Based on these considerations, the following criteria were formulated to assess which ingredients have the potential to contribute to increase the European feed protein production:
- Protein sources should be able to perform well in climate conditions of North West Europe;
- The cultivation of the protein source in Europe is currently no common practice;
- In long term (after 2020) the protein source is still applied in feed and not in food.
Sources that meet these criteria were selected for the short list and discussed in more detail in the following parts of this report.

The main reason for excluding other protein sources from the short list was that the use of these sources is already common practice (n=28, e.g.. maize gluten meal, potato protein, rapeseed meal). Another important reason for exclusion was the absence of appropriate cultivation conditions in (Northwest) Europe (n=4, sorghum, jatropha, rice meal and rice protein concentrate).
The short list of new putative interesting protein sources for pigs and poultry was subdivided in groups, based on their application and the biological classification (Table 2).
The plant protein sources are been discussed in more detail in the following parts of this report. The nutritional aspects of insects, however, were studied in a separate project and presented in the report 'Insects as a sustainable feed ingredient in pig and poultry diets – a feasibility study' (Veldkamp et al., 2012).

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)
Leaf proteins	Grass, sugar beet leaves
Aquatic proteins	Algae, both macro- (seaweed) and microalgae, duckweed
Cereals and pseudo cereals	Protein concentrates from oat and quinoa
Insects	E.g. mealworm, housefly, house cricket

3 Plant cultivation aspects

3.1 Description of plant groups

Several crops or plant species of the short list have a relatively high protein content in their grains or seeds which are harvested, especially the Legumes (soybean, lucerne, pea, chick pea, *Vicia faba*, lupines), up to 40%. The oil seed crops, including the Legume soybean, rapeseed and sunflower, are grown for their oil content but the protein content of the seeds is relatively high (about 25%) and the defatted fractions are generally used in feed (and food). Cereals grains like wheat, barley and oat contain about half of the protein content of Legume grains (12-15%). Pseudo cereal, quinoa, has a kernel protein content of about 15-18%.
Leaf material as a by-product from a crop, or grass, could also be used as a source of protein for feed. The protein content is relatively low (about 12%), but refinery of plant rest material for protein production could be a sustainable way of valorising all biomass from a crop.
The group of the aquatic protein sources contains putative interesting protein sources: duckweed, seaweed and algae, but these need several steps of research into the yield, cultivation practices and nutritional value of the proteins produced. Especially micro-algae and duckweed species can have a very high protein content (45% or higher). The aquatic protein sources do not compete for agricultural land use.

These groups of putative new plant protein sources will be discussed for their agronomic feasibility in Europe, their present cultivation in the Netherlands and Europe, their protein content and crop yield, the presence of anti-nutritional components, and the potential improvement in protein yield and use by plant breeding. The cultivation areas and yield of the different protein crops in the Netherlands and Europe are shown in Table 3, whereas possible crop yield and protein yield per hectare are shown in Table 4.

3.2 Oil seeds: soybeans (a legume), rapeseed, and sunflower seed

Oil seeds in the list of potential protein sources include soybean (a Legume), rapeseed and sunflower seed. These crops are grown for the production of oil that is extracted from the seeds. The seeds of rapeseed, and sunflower contain up to 40-45% of crude oil, and 20-25% of protein. Soybeans have a higher protein content of about 40% and 20% oil. After extraction of the oil from the oil seeds, the remaining product still contains the proteins. The concentration of protein depends on the protein level of the starting material, and the processing steps of the oilseeds, e.g. de-hulling and extraction method.

3.2.1 Soybean

Soybeans have a very high protein level (about 40%) with a good amino acid profile for animal feed, although the concentration of sulphur containing amino acids, methionine and cysteine, are relatively low, and a relatively high fat level (20% oil). Soybeans are used both as a protein crop and an oil crop. Soybean is a legume (or pulse) and belongs to the family of the fabaceae, although the FAO classes the plant as an oilseed rather than a pulse. Like other legumes, soybeans can fixate nitrogen due to a symbiotic relationship with Rhizobium bacterium strains. Soybeans produce significantly more protein per hectare than other large oilseed crops: 940 kg/ha, versus 792 kg/ha for rapeseed and 280 kg/ha sunflower seed (Vahl, 2009).
Cultivation of soybean is successful in regions with hot summers, with optimum growing conditions at a mean temperature of 20 to 30°C. Soybean is hardly cultivated in the EU and not in the Netherlands. For 2011 there are no data in Eurostat on soybean cultivation in Europe. In the past, there was more cultivation of soybean in Europe, but it reduced from 2000 until 2008 from 501 (*1000) ha to 236 (*1000) ha, but seems to increase again (Engwerda, 2011). The cultivation area in Europe in 2010 was 2,740 (*1000) ha and the yield accounted $4,8 \times 10^6$ tons. The main countries within the EU-27 with soybean cultivation are Italy and Rumania. There are no numbers in the databases for cultivation in the Netherlands. The climate in North-West-Europe is less optimal for cultivation of soybean, having a long growing season under the European climatological conditions. The crop needs to be sawn early in the year (before half of April) in order to ripen in time, but night frost in April might harm the crop. Cultivation in Europe will need breeding for cultivars with a short growing season (Kamp et al., 2008;

Vahl, 2009). In the Netherlands, field experiments with the cultivation of soya have been performed in the southern part of the province Limburg (Paauw, 2007). In 2011, the company Agrifirm performed trials with soybean cultivation in the Netherlands, in the provinces Limburg, Flevoland and Brabant. The yield results were positive (~3 ton per ha), but not adequate to compete with production of wheat. Trials in Poland, Denmark and Sweden resulted in a yield of 2 ton/ha, which showed that cultivation in Northern Europe is possible, but not competitive with wheat cultivation, which can yield in Europe 8-10 t/ha. (Engwerda, 2011). In the US the yield of soybean cultivation is comparable (2-3 ton/ha), but the yield of wheat is less than in Europe, 3-4 ton/ha, therefore soya cultivation is more preferred by farmers in the US whereas the European farmers prefer cultivation of wheat.

The main producers of soybeans are USA, Argentina, Brazil, China and India. The average worldwide yield in 2010 was 2.5 tons per hectare. The EU is the biggest importer of soybeans after China. Within the EU the Netherlands is the biggest importer, especially from South America and the US (about 4 Mt. of beans). A quarter of this import is exported to other EU countries, and the remaining import is processed for the production of soybean oil and soybean meal. The largest part of the imported soybeans is used for the production of animal feed (93%, Vahl 2009) and part of the produced animal feed is exported to other countries. For the production of Dutch feed 1.8 Mt of soybeans is used.

Soybeans, like other legumes, contain anti-nutritional factors such as phytic acid (phytate) and protease inhibitors, (Stegeman, 2010). Phytate is a source of inositol phosphate, only digestible by ruminants and not bioavailable for non-ruminants in the absence of the phytate degrading enzyme phytase. Phytate can reduce the absorption of important trace elements, such as zinc and iron. Protease inhibitors reduce the activity of proteases which are essential for digestion and absorption of protein. To destroy protease inhibitors (e.g. trypsin inhibitor), soybeans must be heated, preferably in moist conditions. Raw soybeans are toxic to all monogastric animals, including humans.

The relative low yield in Europe, in combination with a long growing season, consequently enhances the water content of the beans, which is unfavourable for long storage of the beans. This makes soybean cultivation in the Netherlands less attractive than cultivation of peas and beans.

3.2.2 Rapeseed

Rapeseed, which belongs to the family of the Brassicaceae (cabbage), is grown for its oil content in the seed (up to 40%) and within the EU it is the most important oilseed followed by sunflower. The seeds contain approximately 23% protein. The production area in Europe is 6.743 (*1000) ha, but in the Netherlands only 2.026 ha. The production yield in EU is 23.080 (*1000) t, and for the Netherlands 11,5 (*1000) t. It is mainly produced in France and Germany. The oil is used for human consumption and as biodiesel. The rapeseed press cake contains about 30-40% protein and is used for animal feed. Anti-nutritional compounds in rapeseed meal are glucosinolates, phytates and phenolic compounds. Breeding resulted in cultivars that are low in glucosinolates (double-zero lines). The anti-nutritional factors all work in different ways, but all tend to impede the uptake of nutrients such as vitamins and minerals (Stegeman et al., 2010). Rapeseed meal, although it contains high-quality protein, has high levels of anti-nutritional factors and high levels of fibre, which makes it less valuable than soybean meal as a feed ingredient. Several groups (and industry) are working on extraction methods to isolate the protein from those negative factors.

3.2.3 Sunflower

Sunflower seed is the second major oil producing crop in Europe. Production area in EU is 3.700 (*1000)ha and yield is 12.002 (*1000)t. In the Netherlands it is not cultivated. Oil and proteins are the main components of the sunflower seed. The seeds contain about 23% proteins. Compared to proteins from Legumes and other oilseed crops, sunflower seeds have a high content of phenolic compounds, which attribute to the dark colour of protein products and interact with the proteins thereby reducing their bio-availability. This can affect the protein properties in several ways, such as reducing protein digestibility and functionality, prolonging or shortening its storage life and stability (Mulder, 2010).

3.3 Legumes or pulses (grain): peas and Vicia faba, lupines (beans)

Farmed legumes can belong to many agricultural classes, including forage and grain species. Most commercially farmed species are used for several purposes, depending upon their degree of maturity when harvested.

Grain legumes, also called pulses, are cultivated for their seeds. The seeds are used for human and animal consumption or for the production of oils for industrial use. Grain legumes include beans, lentils, lupines, peas, and peanuts.

Pulses are important food crops due to their high protein and essential amino acid content, although the methionine content of legumes is relatively low. Like many leguminous crops, pulses play a key role in crop rotation due to their ability to bind nitrogen. The FAO distinguished eleven groups of pulses that all belong to the family of the Leguminosaea: beans (*Phaseolus ssp* and some *Vigna* species, such as kidney bean, Lima bean, mung bean), broad bean (*Vicia faba*; such as horse bean and field bean), peas (*Pisum ssp*), chickpea (*Cicer arietinum*), cowpea (*Vigna unguiculata*; blackeye bean), pigeon pea (*Cajanus cajan*; cajan pea), lentils (*Lens culinaris*), Bambara groundnut (*Vigna subterranea*; earth pea), vetch (*Vicia sativa*), lupines (*Lupinus ssp*), and minor pulses such as Jack bean, winged bean and Yam bean.

The global pulse market is estimated 60 million ton.

3.3.1 Pea

Pea *(Pisum sativa)* is the most cultivated grain legume in Europe. It is an established and significant crop in Europe, but its yield per hectare is relatively low. In 2010 green pea was cultivated in Europe on 20.1 (*1000) ha with a yield of 4-5 ton/ha. In the Netherlands, the cultivated area was relatively small, up to 500 ha (Kamp et al., 2008), but increased according to FAOSTAT up to 4,000 ha in 2010. The yield per hectare in the Netherlands (5-6 ton/ha) is high compared to other countries in Europe. In recent years, this crop receives increasing interest to be used as protein source for feed and human nutrition. It contains about 25% protein and 50% starch. The properties and composition of pea protein are quite similar to those described for soy protein (Mulder, 2010). Anti-nutritional factors in pea are protease inhibitors, lectines and phytate (Stegeman et al., 2010). Pea as a crop is very sensitive to pathogens and pests, and the plants lodge (fall down) easily. Different fungal diseases, insects, birds and weeds can attack the crop. Therefore, peas can be cultivated only once every five years on the same location. Breeding can improve yield and pest resistance, but as a crop it is still lagging behind cereals.

3.3.2 Vicia faba

Beans *(Vicia faba)* are the second most cultivated legumes in Europe. Beans are cultivated for human consumption and animal feed. For high yield and good growth, beans are very dependent on enough water supply during the growing season. Cultivation area in the Netherlands was very low (400 ha; Kampman et al., 2008), but increased according to FAOSTAT up to 5 (*1000) ha in 2010. The yield is higher compared to pea (5-6.5 tons/ha), but like peas, beans are very sensitive to pathogens and pests, lodging and drought. Due to fungal diseases, the crop can only be cultivated once every 5-6 years at the same location.

3.3.3 Chickpea

Chickpea (*Cicer arietinum*) is a legume also belonging to the family fabaceae, like *Vicia faba*. Its seeds are high in protein and it is one of the oldest cultivated legumes. It is grown best in a tropical or subtropical climate with sufficient water, but can also be cultivated in temperate climates resulting in lower yields. Main producer is India, followed by Pakistan and Turkey. Chickpea is not cultivated in the Netherlands. The total area of cultivation in Europe is 60.000 ha in 2010, yielding 80.000 tonnes per year. Chickpea has the lowest yield per hectare of all pulses (on average 1.4 tons/ha), and is very sensitive to several pathogens and pests, like other grain legumes. Protease inhibitors, lectins and polyphenols are the anti-nutritional factors present in chickpea. The protein content is 21%.

3.3.4 Lupine

*Lupine (*Lupinus*)* is cultivated to harvest the beans for food and feed, and as forage. Lupine is an interesting crop because of the high protein content in beans (35%), but the yield of beans per hectare is relatively low (1-5 ton/ha). Under optimal conditions the yield can be 6 to 8 tons per hectare. It can contain anti-nutritional compounds, such as toxic alkaloids which are bitter tasting and may reduce food intake. Breeding resulted in 'sweet lupine' cultivars with a lower alkaloid level. Lupine is sensitive to several plant pests, such as fungal pathogens (e.g. *Colletotrichum gloeosporiodes*). Lupine is not cultivated in the Netherlands. The main producers in the EU are Germany, Poland, Spain and France. The total area of cultivation in Europe is 148 (*1000) ha, and the production yield in Europe is 200 (*1000) tonnes per year.

3.4 Legumes (forage): lucerne (alfalfa)

Alfalfa (*Medicago sativa*; legume family fabaceae) is the most cultivated forage legume in the world. Worldwide production was around 436 million tons in 2006 (FAO, 2006). The US is the largest alfalfa producer in the world, but considerable production is found in Canada, Argentina (primarily grazed), Southern Europe including France, Australia, South Africa, and the Middle East. Yields are around 8 tons per hectare, but maximum yields of 20 t/ha have been recorded. Like other Legumes it has the ability to fix nitrogen due to symbiosis with bacteria. The production area in Europe is 7.12 million ha (25% of the world production area), and it is mainly cultivated in France and Italy. Alfalfa is the Legume crop that produces the highest amount of protein per hectare: 2,400 kg of protein per hectare on the basis of 13 tonnes of dry matter with an average of 19% of crude protein as compared to 800 kg of protein per hectare for soybean. The draw-back compared to pulses and (oil)seed protein sources is that the leaf material contains a lot of water and generally a drying process is required prior to storage.
Most of the improvements in alfalfa production over the last decades have consisted of better disease resistance on poorly drained soils in wet years, better ability to overwinter in cold climates, and the production of more leaves. Multi-leaf alfalfa varieties have more than three leaflets per leaf, giving them a higher nutritional value because there is more leafy matter for the same amount of stem. Still, alfalfa is very sensitive to several bacterial, fungal, viral and parasitic diseases.

3.5 (Pseudo)cereals: oats, quinoa

3.5.1 Oats

Oats are cultivated in the Netherlands on a very low scale, only 1700 hectares, for the food industry and as feed for horses. The protein content is comparable to wheat protein content, 12-15%. The yield per hectare is lower compared to wheat: about 3-5 tons per hectare (wheat 6-9 tons per hectare). Oat is very well adapted to cultivation in the Netherlands and North-West and Eastern Europe. Breeding for higher yield, as has been performed with wheat, could improve the yield per hectare to the same level as for wheat. Breeders have been focussing on wheat instead of oat as a grain cereal because of the wheat gluten functionalities for food applications. Oat is a very robust crop that fits good in an organic farming system. In general, oat requires lower fertilisation doses than wheat and due to its vigorous growth habits, oat can compete with most weeds. It is sensitive to lodging, although some cultivars, such as naked oats, are more resistant (Kamp and van Reeuwijk, 2011). In Europe (EU-27) about 7.400 (* 1000) tons oats were produced on about 2.700 (*1000) hectares in 2010. Six countries produce two-third of the total oat production in Europe: Poland and Spain are the highest producers (both 20%), followed by Finland (10,3%). United Kingdom, Germany and Sweden together account for 15,7% of the total EU-production.

3.5.2 Quinoa

Quinoa (*Chenopodium*) is a grain-like crop belonging to the Chenopodia, not to the cereals (Gramineae), and therefore called a pseudo-cereal. The protein content is 15-18%, and the nutrient composition is very good compared with common cereals. Quinoa contains a higher level of lysine and also calcium, phosphorus, and iron. Depending on the variety, it's optimal growing conditions are in

cool climates with temperatures that range from −3°C during the night, to near 35°C during the day. Optimal for Quinoa growth is rainfall during early growth and development of the plant and dry conditions during seed maturation and harvesting. The average yield per hectare is 3-5 tons.
The main anti-nutritional factor is the presence of saponins. After harvest, the grains need to be processed to remove the bitter-tasting saponins, by e.g. alkaline water washing or mechanically via abrasion. It has been attempted to lower the saponin content of quinoa through selective breeding to produce sweeter, more palatable varieties. The first commercial supply chain for European quinoa has been established in the Loire Valley of France. In 2009 the department of Maine-et-Loire produced 140 tons of quinoa on 100 hectares. Yields varied from 0.7 to 3 tons per hectare. It was the first large-scale quinoa production trial in Europe. In 2011 quinoa was cultivated on 250 hectares, mainly in South Europe. It is not cultivated in the Netherlands.

3.6 Leaves from sugar beet and grass

3.6.1 Sugar beet leaves

Sugar beet is cultivated in the Netherlands on 71 (*1000) hectares and in Europe on 3229 (*1000) hectares. The crop yields 65 tons of taproots per hectare and about 45 tons of leaf and stem material. The latter is generally cut in pieces and spread on the arable land during the harvesting process. Collection would result in 3500 (*1000) tons remaining leaf material for the Netherlands per year and 150×10^6 tons for Europe. This huge amount of left-over material could be a source of proteins. For all green leafy biomass including sugar beet leaves, a drying step is necessary for preservation, storage and transport and to concentrate the protein content.

3.6.2 Grass

The area of grassland in the Netherlands is 99,5 (*1000) ha, and in Europe 238.000 (*1000) ha. The yield is expected to be 10-15 tons DM per hectare per year, but there are no production data available in Eurostat or CBS StaLine. Agricultural practises for cultivation in Europe are already in place. The cultivated grassland is already used for grazing of ruminants and horses and the production of silage and hay. All grass from the total grassland area could be harvested and used as source for proteins. This would need a drying step, as holds for all leafy material.

3.7 Micro- and macroalgae (seaweed)

3.7.1 Microalgae

Microalgae are of main interest because of the opportunities to use them for biodiesel production. A biorefinery concept, however, is essential to make the production of biofuel from microalgae cost-effective. Besides oil and secondary metabolites, microalgae can contain large amounts of proteins (25-50%) depending on the strain used (Becker, 2007; Mulder, 2010). In growth experiments and pilot plants, yields could be reached of 15-30 tons ds/ha per year. Research is still needed for development of the best cultivation conditions and selection of the best performing algae species and strains depending on their application, such as production of bio-fuel or production of high-value compounds, or production of proteins. High production costs might be a hurdle for cultivation of micro-algae solely for protein production.
Cultivation on high industrial scale of microalgae is at this moment in the Netherlands and in Europe not yet applied.

3.7.2 Macroalgae

Macroalgae (seaweed) are cultivated on a large scale by Asian producers (China, Japan), which account for the total global year market of 10.000 (*1000) ton. Current applications of seaweeds include the use of fresh seaweed for human consumption, primarily in Asia, and production of alginates/agars, extracts for personal care, fertilisers but also feed. In Europe, traditionally a limited amount of seaweed from natural production is harvested in Northern Europe, especially in Norway

(EU NETALGAE network on macro-algae industry). Recently, several seaweed species native in the North Sea are being cultivated in Ireland (36 (*1000) ton; Morrissey et al., 2011). Pilot projects as the 'Zeeboerderij' in the Netherlands, province Zeeland study the potential of cultivation of seaweed. Seaweeds typically contain a high amount of polysaccharides (up to 60%), but also high value compounds such as colorants, omega fatty acids and bioactive compounds. Depending on the species, seaweeds contain between 10-30% of protein (Mulder, 2010).

3.7.3 Duckweed

Another potential protein source is duckweed. The protein content of duckweed is very high, 35-45%, and the yield per hectare is also very high due to its capacity to grow vegetative and exponentially, up to 30-40 tons DM per hectare. The protein content depends very strongly on the nutrient availability (particularly nitrogen and phosphorous). The protein content of *Lemna gibba* can be as high 40% in a high nutrient lagoon (dairy cattle waste lagoon) and as low as 9% in a low nutrient one (Hasan and Chakbari, 2009).

In North America, Australia and developing countries duckweed cultivation already has been used for organic wastewater treatment and to purify agricultural wastewater. If the waste water do not contain high levels of heavy metals or other toxic compounds, the produced duckweed biomass can be used as feed, either directly (as fish feed) or after a pressing and drying step as dry fodder (Pillai, 2010). Especially in developing countries duckweed is processed for feeding farm animals and fish or to serve as fertilizer. In the Netherlands, only small scale trials are conducted and a duckweed value chain has not been developed yet. Duckweed can be a suitable protein source that can be locally produced in basins. It could become the most productive protein source in Europe (protein production per ha almost ten times higher than soybean) which does not compete for arable land use (Leng et al., 1995; Derksen and Zwart, 2010; Nieuwenhuis and Maring, 2009; Yu et al, 2011). Duckweed can have a 50% biomass increase every two days (Nieuwenhuis and Maring, 2009; Derksen and Zwart, 2010; Zondervan, 2012). There is no commercial duckweed cultivation on a high scale yet in Europe and the Netherlands. Duckweed is a putative new protein source with very high potential, but it needs more (scientific) input on the level of cultivation, processing and the application in feed. The processing of the duckweed biomass needs a drying step.

Table 3. Cultivation areas and yield in the Netherlands and Europe for the different alternative protein sources.

	Area production in NL (ha x1000)	Area production in EU (ha x1000)	Yield in NL (t x 1000)	Yield in EU (t x 1000)
Oil seeds – soybean	-	2.740	-	4.790
Oil seeds – rapeseed	2,6	8.770	11,5	23.080
Oil seeds – sunflower	-	3.700	-	12.002
Legumes (pulses) – peas/beans/lupine	9,3	2.480	67,9	6.530
Legumes (pulses) - chickpea	-	60	-	80
Legumes (forage) – lucerne	6,4	7.120	-	78.320
Cereals – oat	1,7	2.700	8	7.400
Pseudo cereals – quinoa	-	0,25	-	0,27
Leaves – grass	99,5	238.000	995*	2.380.000*
Leaves – (e.g. sugar beet leaves)	71	3.229	3.500*	149.800*
Macro algae – seaweed	-	-	-	-
Micro algae	-	-	-	-
Duckweed	-	-	-	-

Sources: Eurostat (http://epp.eurostat.ec.europa.eu/portal/page/portal/statistics/search_database); FAOSTAT (http://faostat.fao.org) ; CBS StatLine (http://www.cbs.nl/en-GB/menu/themas/landbouw); * estimation, because no data available

3.8 Comparison protein content and yields

Protein content of the seed of many Legumes (pea, horse bean, lupine etc) is high and ranges from 30-40% in the dry matter. Especially soybean has a very high protein level: 40%. The same holds for several species of Duckweed. For some micro algae species the protein content is reported to be as high as 55%, whereas for macro-algae (seaweeds) the protein level can vary between 10-30%. Oilseed crops such as rapeseed and sunflower seed have a protein content in the range of 20-26% of DM. Leaf material of Legumes (Lucerne) can reach up to 15% protein. Cereals such as wheat and oats have a protein content of 15-19% of DM, which also holds for the pseudo grain quinoa. Leaves and grasses contain relatively the lowest protein level, up to 10-12 % of DM.

Table 4 shows the protein content for the various putative protein sources, the yield per hectare in Europe, and the protein yield per hectare. For the algae (micro and macro) and Duckweed, the numbers are still based on pilot cultivation trials.

Table 4.   Protein content, yield/ha and protein yield/ha of the various protein sources.

	Protein content	Yield in EU possible (tons ds/ha/y)	Protein yield possible (tons/ha/y)
Oil seeds – soybean	40%	1.5-3 tons	0.6-1.2 tons
Oil seeds – rapeseed	25%	3 tons	0.75 ton
Oil seeds – sunflower	23%	3 tons	0.7 ton
Legumes (pulses) – peas/beans/ lupine	17-35%	4-6 tons	1-2 tons
Legumes (forage) – lucerne	19%	13 tons	2.5 tons
Cereals – oat	12-15%	3-5 tons	0.4-0.75 ton
Pseudo cereals – quinoa	12-18%	3 tons	0.4-0.5 ton
Leaves – grass	12%	10-15 tons	1.2-2 tons
Leaves – (e.g. sugar beet leaves)	12%	4.5 tons	0.5 ton
Macro algae - seaweed	10-30%	25 tons	2.5-7.5 tons
Micro algae	25-50%	15-30 tons	4-15 tons
Duckweed	35-45%	30-40 tons	10-18 tons
Wheat (as reference)	11%	10 tons	1.1tons

3.9 Conclusions

Peas and beans are very interesting due to the high protein content (17-35%) and because cultivation practises are already available and implemented. However, these crops are very sensitive to pests and pathogens and crop rotation is necessary meaning that cultivation on the same field can take place once every five years.
Soybeans might be interesting because of high protein content and new cultivars are being selected for growth in Europe having a short growing season. The yield per hectare in Europe of 3 tons/ha/year is still much lower than wheat with a yield of 8 tons per year, making soybean cultivation less attractive for farmers in Europe. Breeding steps for high yielding cultivars with a short growing season still need to get more input. Rapeseed (meal) is interesting because of high production in the EU, with a reasonable protein yield per hectare, but not in the Netherlands.
Oats are interesting, but at this moment the yield per hectare is lagging behind compared to wheat. It still needs intensive breeding input, but with adequate attention from breeders, the production level could reach that of wheat.
Aquatic proteins are very interesting because of high protein content (duckweed and several micro-algae) and very high yields, as has been shown in pilot experiments. However, large scale cultivation, processing and feasibility for application as feed still needs more 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 arable land for cultivation.
For all the protein sources with a high water content, such as (left-over) leaf material, duckweed, micro- and macro algae holds that a drying step for protein extraction or concentration is required.

4 Plant processing aspects to enhance protein content

Further processing of potentially interesting crops, thereby reducing the ANF contents and increasing the protein content to levels of 65% or higher, might 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). Processing to enhance protein content of these sources will be described and the economics of this processing will be discussed. The sustainability of the processes in terms of energy and water use will be briefly addressed.

4.1 Technology

A wide range of procedures and techniques have been developed to enhance the protein content of raw materials. In the next paragraphs general procedures that are commonly used by companies working in this field, will be described. In addition, academic and applied research is being performed to develop new procedures that better meet demands with respect to sustainability aspects like water and energy use and regulatory issues, e.g. avoidance of organic solvents. These procedures will not be described, because such a comprehensive overview is beyond the scope of this project. Focus is on the raw materials in the short list, for which processing would be an option to enhance their use in relevant feed systems.

4.2 Oil seeds

Soybeans, a legume with a relatively high oil content (around 20%), have set the scene for developing the processing of oil seeds, particularly with respect to preparing protein enriched fractions such as protein concentrates with a protein content for soy > 65% and isolates with a protein content for soy >90%. Hence soybean processing is described as the example for oil seed processing (see for more information on processing of soybeans e.g. Liu (2005). Emphasis is on the usual type of soybean processing to prepare protein enriched fractions . This type of processing includes a defatting step by solvent (usually hexane) extraction. Other defatting procedures include a defatting step by expelling using screw presses or combinations of expelling and solvent extraction. These processes are more common when using oil seeds other than soybeans. Solvent extraction results in the lowest fat content of the remaining meal, expelling results in the highest oil content.

Consequently, for commercial defatted oil seed meals, three types can be distinguished:
- Solvent extracted meals
- Expeller extracted meals (the only one allowed for organic feed)
- Expeller/solvent extracted meals)

Furthermore, it should be emphasized that in soybean processing in general dehulling is applied. This is not the case for the processing of many other oil seeds, often rapeseed and sunflower seeds are not or only partly dehulled. Dehulling leads to a separate stream of hulls that may or may not be added to the defatted flour. Presence of the hulls results in a reduction of the protein content in the defatted material and to an increase in the fibre content.

4.2.1 Soybean processing

A schematic overview of the usual soy protein processing is presented in Figure 2. The processing starts with cleaned soybeans that have been dried, cracked (i.e. broken in small pieces to ease dehulling and flaking), dehulled, conditioned with steam and flaked to increase the surface. The flakes are then conveyed to an extractor where the oil is removed by counter current solvent extraction, usually with hexane as the solvent. After this oil extraction, the flakes still contain some hexane that has to be removed (termed desolventizing). The way in which this is done in terms of time, temperature and moisture during the process, has a profound effect on protein denaturation. The hexane from flakes intended to be used for feed is usually removed by passing the flakes through a desolventizer-toaster (termed DT flours), which uses steam. The flakes are subjected to a high-temperature, moist heat and hence the proteins denature depending on the intensity of the treatment. For most food applications, other types of desolventizing procedures are used resulting in less protein denaturation (the resulting flakes are termed " white flakes"), a common one being the flash

desolventizing system. By adjusting processing parameters defatted flakes can be obtained with a wide range in protein denaturation, and hence also in inactivation of proteinaceous anti-nutritional factors (ANF's) such as trypsin inhibitors and lectins. The extent/intensity of protein denaturation affects the digestibility of the protein: a denatured and hence unfolded protein structure is advantageous for the accessibility to digestive (proteolytic) enzymes. However unfolding also increases the ability of the protein to interact with itself and other proteins, so called aggregation, and with other components. This needs not to be disadvantageous, however very strong and tight aggregation may inhibit the accessibility to enzymes.

```
                          ┌──────────────┐
                          │   Soybeans   │
                          └──────────────┘
                                 │
                                 │  Cleaning, Cracking, Dehulling, Flaking
                                 ↓
┌──────────────┐          ┌──────────────┐
│ Full Fat Meal│ ◄────────│ Full Fat Flakes│
└──────────────┘          └──────────────┘
            Grinding             │
                                 │  Oil extraction
                                 ↓
┌──────────────┐          ┌──────────────┐
│ Defatted Meal│ ◄────────│Defatted Flakes│
└──────────────┘          └──────────────┘
            Grinding          ╱       ╲
 Protein extraction,        ╱           ╲   Soluble carbohydrate extraction, Drying
 Precipitation, Drying    ╱               ╲
                    ┌──────────┐    ┌──────────────┐
                    │ Isolate  │    │ Concentrate  │
                    └──────────┘    └──────────────┘
```
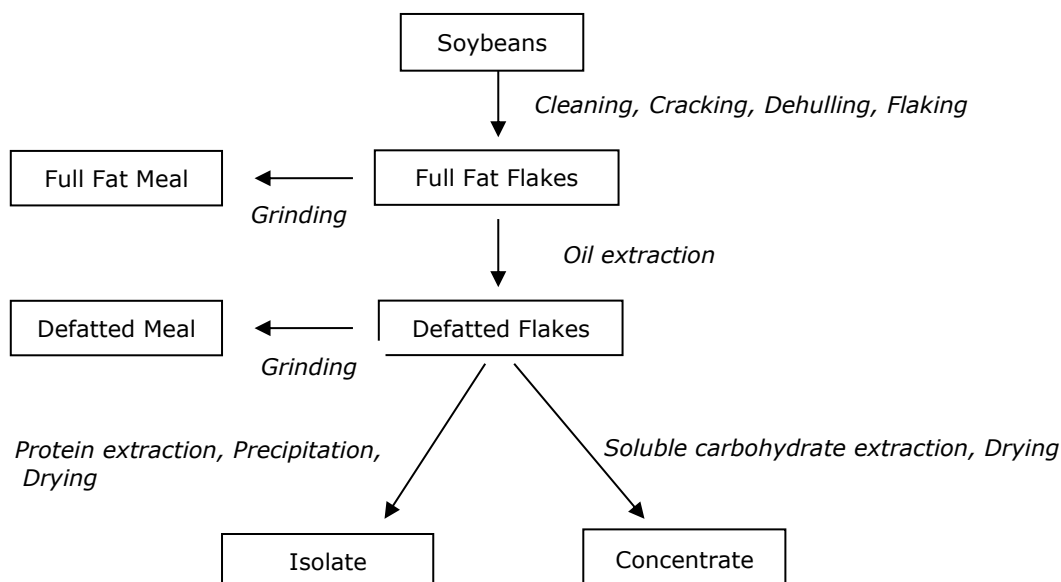
Figure 2. A schematic overview of soybean protein processing.

From the defatted flakes with a protein content <50%, protein concentrates are being prepared by removal of non-protein components, especially soluble carbohydrates. (a side-stream of relative low value but its removal is beneficial for nutritional purpose) The resulting product is mainly composed of proteins and larger (cell wall) polysaccharides. Processes to remove the non-protein components include:
- Water washing (leaching) of heat-denatured defatted soy flour
- Diluted-acid washing of defatted flours at the isoelectric pH (4.5)
- Washing with aqueous alcohol (ethanol or when warranted isopropanol).

The latter process, using warm 60-70% ethanol, is the most common one. In contrast to acid washing, the proteins become denatured but the yields are higher and less waste is generated.

Soybean protein isolates are being manufactured from defatted soy flakes by separation of the protein from both soluble and insoluble carbohydrates. This is achieved by extracting the proteins at alkaline pH. After centrifugation to remove the insolubles (mainly cell wall polysaccharides), the solubles (mainly carbohydrates and minerals and some proteins including trypsin inhibitors)) are removed by precipitating the proteins at the iso-electric pH. The precipitated proteins are spray dried, usually after neutralization. To increase protein yield, membrane filtration is used to remove the non-protein solubles, in this way recovering the part of the protein that is soluble at the isoelectric pH. However, the most common process is iso-electric precipitation.

Soybean protein concentrates are used both in food products (e.g. in the meat industry) and in higher-value feed products. Soybean protein isolates are not used in feed, apart from exceptions, but as a functional protein ingredient in a large variety of food products.

It should be emphasized that during soybean processing, heating steps are used to inactivate enzymes such as lipoxygenase (catalysing the oxidation of unsaturated fatty acids and thereby responsible for off flavours) and proteinaceous ANF's such as trypsin inhibitors and lectins. A common treatment of soybeans, particularly aimed at inactivating lipoxygenase, is roasting (dry heating) to inhibit the development of rancidity in the beans/meals (full fat roasted soybeans).

4.2.2 Other oil seeds

Meals, derived from all other oil seeds mentioned in the short list (i.e. rapeseed, sunflower), protein concentrates and isolates are or can be prepared by processes more or less similar to the one described for soybeans (e.g. Gonzalez Perez and Vereijken, 2003). Of course, processing needs to be optimised for the specific type of seeds. The optimization does not only includes specific conditions of temperature, pH etc. but also the removal/inactivation of ANF's (enzymes, inhibitors, lectins, polyphenols, etc). For feed applications, the latter is facilitated by the fact that there is no need for avoiding denaturation of the bulk of the protein. However, very intensive energy costly treatments, such as excessive heat treatments as used for the production of DDGS as a co-product of the bio ethanol production, should be avoided because they can result in reduction of nutritional value and/or digestibility, e.g. loss of bioavailability of lysine due to Maillard reactions, and too strong aggregation. Specifically for sunflower seeds, it should be mentioned that the production of protein concentrates and isolates is hampered by high levels of polyphenols in sunflower seeds, leading to crosslinking of proteins, and colour formation. This makes the production of sunflower protein concentrates less attractive, particularly since the meal already contains high levels of protein: expeller extracted meals about 40%, solvent extracted meal about 50% but when using comparable dehulled seeds these figures are 53 and 66% (e.g. Gonzalez Perez and Vereijken, 2003).

Rapeseed contains polyphenols, although lower levels than in sunflower seeds, and also two other ANFs: eruca (erucic) acid and glycosinolates. Varieties have been bred that are low in these compounds: they are termed double-null rapeseed or canola. Because canola is low in ANF's, they are the preferred starting materials.

Rapeseed concentrates usually have a somewhat lower protein content than soybean concentrates, thereby reflecting differences in the ratio of the main components of the seeds. The protein content of sunflower concentrates can be higher or lower, because of the higher variation in the ratios of the main components in the seeds, strongly depending on varieties.

4.3 Legumes

Many legumes contain carbohydrates, particularly starch, instead of oil, as in soybeans as the energy source for the growing seedling. Examples of starch containing legumes are peas, Vicia faba and chickpeas, all mentioned in the short list. Protein preparations from this type of seeds can be manufactured by two types of processes termed:

- Wet fractionation, a process similar to the one described for the preparation of soybean isolates.
- Dry fractionation, usually milling followed by air classification.

Using wet fractionation, protein preparations are obtained with a protein content of about 80 to above 90%. Such preparations are intended for use in food products. Therefore, wet fractionation will not be further discussed in this study.

Dry fractionation includes fine milling and separation of the particles on the basis of their density in a flow of air (so-called air classification or wind sifting). Three fractions are obtained: fractions enriched in fibres, starch and proteins. The enrichment depends on the type of seed and the processing conditions. Common figures for peas and faba beans indicate an enrichment to about 60-70% of protein (e.g. Gueguen, 1983).

In addition to peas and beans also other legumes can be used as starting material, e.g. lentils and lupines. An important criterion is the oil content, because oil hampers the dispersion of the milled particles into the air. Therefore, it has to be experimentally tested whether for instance a given variety of lupines with a fat content of 5-15%, depending particularly on variety, can be directly fractionated by air classification. If not, air classification is possible after defatting, e.g. by solvent extraction (see oils seeds processing). Furthermore, lupines contain relative high amounts of alkaloids. Breeding programmes have resulted in reducing these levels. These species, including blue, (*Lupinus angustifolius*), white (*Lupinus albus*) and yellow (*Lupinus luteus*) lupines are called sweet lupines in contrast to the other lupines called bitter lupines.

To our knowledge pea protein concentrates are commercially available. For many other legumes, concentrates have been produced at more smaller or even "pilot" scale and on similar scale by wet procedures (extracting the proteins and precipitating them by heat or at their isoelectric pH).

4.4 Cereals and pseudo cereals

On the short list are both oats, a cereal, and quinoa, a pseudo cereal. Just as with the starch containing legumes, the protein content can be enhanced by dry fractionation. However, compared to many cereals and starch containing legumes, the size of the starch granules of oats and quinoa is much smaller. This may hamper the separation of the starch from the proteins, and hence may result in lower protein content of the protein enriched fraction. There is very limited information on the dry fractionation of oats and quinoa, aimed at obtaining a protein enriched fraction.

4.5 Leaf processing

In literature, processing to produce protein feeds from quite a number of different leaves (e.g. cassava, alfalfa/lucerne, grass) has been described. The basic steps are grinding the leaves, squeezing out the protein rich juice by mechanical pressing and recovering the protein from the juice by heating it to precipitate the protein (e.g. Dale et al., 2009). The protein content of the resulting leaf protein concentrates (LPC) range from 50 to nearly 90%, depending on the plant species and conditions. A serious draw-back for this technique is that freshly harvested, green leaves are required, which are usually only available for a few months per year. Another disadvantage is that the co-product – the fibrous leaf material – is depleted from protein and hence has a relatively low value in feed products. These draw-backs are the main reasons that the economics of LPC production are not yet competitive with other protein sources. These challenges are addressed in research on so-called integrated biorefinery systems that allow operating all year round, not only using fresh material but also dried or ensiled material, and add value to the residual cellulose and hemi-cellulose rich stream. An example of leaf processing on pilot scale is the Dutch Grassa! project, aimed at producing both protein for feed and fibres for paper (www.grassanederland.nl). Recently, the added value that grass refining could have for the Dutch livestock farming has been evaluated. This evaluation clearly shows that grass refining has potential but still a large number of questions including ecological and societal ones, have to be addressed (Van den Pol-Dasselaar et al., 2012).

4.6 Algae

Algae are regarded as a very promising source for the production of biofuels, feed and food (e.g. Wijffels and Barbosa, 2010). Two types of algae are distinguished, i.e. micro- and macroalgae or seaweed. They are not yet used for the production of (relatively cheap) bulk components such as oil and protein. With respect to protein refinery, particularly micro-algae are very promising because their protein content can be in some cases as high as 50%, depending amongst others on species and growing conditions (Becker, 2007; Hasan and Chakbari, 2009). The protein content of seaweeds is lower, up to about 30% and higher in green and red seaweeds than brown ones Bulk scale production of microalgae will take 10-15 years (Wijffels and Barbosa, 2010) and serious problems to overcome include cultivation/fermentation (increasing yield per hectare), harvesting (because of their small size) and biorefining (open up the very firm cellulosic cell wall).

4.7 Duckweed

Because of their larger size, harvesting is much less of a problem than harvesting microalgae. Biorefinery of duckweed is – as for algae – still in its infancy.
In contrast to legumes and oil seeds, algae and duckweed are very wet starting materials. Leaves also have a relatively high water content but their processing procedure – squeezing out the protein rich juice –requires the presence of water. For the production of feed containing algae and duckweed as such, a drying step will usually be required.

4.8 Economics

The market for vegetable protein concentrates derived from oil seeds and legumes, is dominated by soybean protein concentrates. These products set the price and all other protein concentrates will have comparable economic value unless they have very specific (dis)advantages e.g. in terms of digestibility. Most of the processes to enhance the protein content of the sources mentioned in the short list are still in development or only used at a pilot scale. A thorough analysis of the economics of such processes is beyond the scope of this desk study. However, one important aspect, without going into the details of the process (and investments), is discussed in this paragraph.

To assess economic feasibility of enhancing the protein content of a given source it should be emphasized that all resulting products should be taken into account. Oil seed processing is economically feasible due to the fact that two valuable products are obtained: oil and a meal enriched in protein. Further enrichment of the meal is often economically feasible due to the fact that the protein enrichment is quite high, for soybeans from about 45% up to 65% in the concentrate, and undesired components may be reduced in the concentrate. The added value of the protein enrichment compensates the loss in value of the protein depleted fraction, the soluble carbohydrate fraction.

During the fractionation of starch containing legumes such as peas and Vicia faba, also two valuable fractions are obtained: a fraction enriched in protein and one enriched in starch, in addition to a less valuable fibre fraction. For lupines the picture is different: they have no or very little starch and their fat content is not very high (ranging from 5 to 15%, depending particularly on variety) as compared to oilseeds such as soybeans (20% oil) and rapeseed (40%). This clearly hampers the economic feasibility of enhancing the protein content of lupines. Furthermore, lupines already have a relatively high protein content (30 up to 50%) compared to peas and Vicia faba (20-30%).

The economics of leaf processing is also hampered by the fact that next to the valuable protein fraction a fibre fraction depleted in protein is obtained. The feasibility of leaf processing can change if the prices of protein rich feed ingredients increase and/or the value of the fibre fraction for feed and non-food (e.g. paper, textile).

4.9 Sustainability

Enhancing the protein content will require resources such as energy and water. The choice of the process technology has a large effect on these sustainability aspects.

In a recent review, Schutyser and van der Goot (2010) argued that for protein fractionation, separation technologies are needed that:

- Use less or no solvents (water or organic solvents)
- Require no stabilization process after separation (e.g. no drying)
- Focus on functionality rather than on purity
- Are easily scalable and have the possibility to be located close to the potential application.

Dry separation technologies meet all these criteria. Compared to wet separation technologies, they need no solvents and the input of energy is much less as is exemplified by Schutyser and Van der Goot (2010) using the separation of wheat in starch and protein by wet and dry processing.

The only commercial operating dry fractionation technology is milling combined with air-classification. This technology is very well suited for cereals and pulses; oil containing raw materials still face severe challenges (Schutyser and Van der Goot, 2010). New developments (improved milling technology, electrostatic separation) will enlarge the potential of dry fractionation. Furthermore, combinations of wet and dry fractionation (e.g. enhancing the protein content by dry fractionation prior to wet separation) offer interesting opportunities to enlarge the sustainability of protein enhancement.

With respect to oil seed processing the literature on data regarding Life Cycle Analysis is very limited. We found only few literature data on the energy (expressed in terms of equivalents CO_2 per kg material) required for the preparation of concentrates from defatted soybean meals (Mustakas and Sohns, 1976; Dalgaard et al., 2008). This limited set indicates that the amount of CO_2 per kg of material for the production of the concentrate from a defatted meal is comparable to that required for the production of the defatted meal. Unfortunately, we lacked the time for a more profound analysis. Very carefully we assume that by processing of defatted meals to concentrates, the energy load may increase with a factor of two.

The processing of leaves, algae and duckweed to protein preparations is still in its infancy. Because assessing sustainability parameters such as energy and water input very much depends on factors such as material, the type of processing (apparatus, temperatures, concentration, etc.), the type of products to be delivered (particularly dry or wet product), the scale of operation etc., even no indicative

conclusions regarding these parameters can be made without many assumptions. This requires much time and cannot be performed within the framework of this screening study.

We recommend that if protein enrichment of certain sources is researched, at an early stage a thorough analysis is made of the energy that this process entails, and this is compared with the gain in environmental impact when using this enriched material in animal feed.

4.10 Conclusions

Processing of the selected feed resources to enhance their protein content is generally still in development and not yet well established. This makes it very tricky to draw conclusions about the sustainability aspects involved in this additional processing and how the sustainability impact should be divided among the various (by)products obtained by this processing. Therefore, only the processing and economic aspects are taken in consideration in this paragraph.

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 dry fractionation from peas and faba beans. The former are already on the market. Lupines are less attractive.

For the longer term protein enrichment of leafs/grasses might deliver attractive feed stocks. 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.

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).

Regarding the new resources such as leafs and particularly the aquatic ones, it should be remarked that they have a relatively high water content, which may require drying before their use as feedstock as such (for their processing to enrich protein this is not required but then they should processed fresh). Seeds of legumes and oil seeds are harvested in a relatively dry state, often not requiring any additional drying step for their as such in feed.

5 Nutritional aspects for pigs and poultry

The nutritional aspects of the potential European protein sources for monogastrics will be considered in this chapter.

5.1 Concentrates and isolates from oil seeds

The nutritional value of rape seed, soya beans and sunflower seed after oil removing by hexane extraction for pigs and poultry is shown in Table 5

Table 5. The nutritional value of rape seed extract, soya bean meal and sunflower seed extract (oil removed by hexane extraction) for pigs and poultry (CVB, 2007).

	Unit	Rape seed meal (CP < 380)	Soybean meal	Sunflower seed meal (CF 200 – 240)
General chemical composition				
Dry matter	g/kg	873	873	890
Ash	g/kg	67	65	66
Crude protein	g/kg	335	464	313
Crude fat	g/kg	26	19	19
Crude fibre	g/kg	120	37	223
Starch	g/kg	61	---	37
Sugar	g/kg	90	107	53
NDF	g/kg	292	81	367
ADF	g/kg	194	48	267
ADL	g/kg	46	4	72
Ca	g/kg	7.4	2.7	3.5
P	g/kg	10.9	6.5	10.8
K	g/kg	12.5	22.4	15.1
Na	g/kg	0.3	0.2	0.2
Nutritional value for pigs				
NE	MJ	6.29	8.25	5.51
SID protein	g/kg	257 (77%)	431 (93%)	250 (80%)
SID Lysine	g/kg	13.3	25.5	8.3
SID Met+Cys	g/kg	11.0	11.5	9.9
SID Threonine	g/kg	9.9	15.1	8.7
SID Tryptophan	g/kg	3.0	5.2	3.0
SID Isoleucine	g/kg	9.4	18.7	10.3
Absorbable P	g/kg	2.9	2.5	1.6
Nutritional value for poultry				
ME Laying hens	MJ	7.09	9.22	6.04
ME Broilers	MJ	5.79	7.93	4.82
Dig. protein	g/kg	255 (76%)	404 (87%)	266 (85%)
Dig. Lysine	g/kg	14.7	25.3	8.6
Dig. Met+Cys	g/kg	11.5	11.4	10.1
Dig. Threonine	g/kg	11.5	15.4	9.7
Dig. Tryptophan	g/kg	3.5	5.4	3.0
Dig. Isoleucine	g/kg	10.5	18.8	11.3
Dig. Arginine	g/kg	18.4	31.0	23.1
Dig. Valine	g/kg	13.7	19.4	13.2
Absorbable P	g/kg	3.6	2.7	2.9

5.1.1 Rape seed protein concentrate

Rapeseed is commonly cultivated in Europe. Mainly rapeseed extract is used in pig diets. The protein content in rapeseed extract is about 335 g/kg and is lower than in soy bean meal. Moreover, the level of the majority of ileal digestible amino acids and the protein digestibility is lower in rapeseed extract than in soy bean meal (see Table 5). Rapeseed extract is mainly used in diets of sows and growing and finishing pigs and less in diets for piglets. Rapeseed contains erucic acid and glucosinolates, both antinutritional factors (ANF's). As a result of European breeding programs, the levels of erucic acid and of glucosinolates has decreased (Lamont and Lambrechts. 2005). Actually, the level of erucic acid is about 2% of total fat in rapeseed. The level of glucosinolates is about 8 µmol/g rapeseed (Lamont and Lambrechts, 2005). Rapeseed extract with a level of erucic acid lower than 2% and a level of glucosinolates lower than 25 µmol/g is called Canola (Canadian Oilseed, Low-Acid) or "raapzaadschroot dubbelnul" in Dutch (Lamont and Lambrechts, 2005). Preferably, canola extract is used in pig diets.

Since a few years a canola protein concentrate is produced by CanPro Ingredients Ltd in Canada. The concentrate contains about 60% protein and is specifically processed for use as a high-value protein rich ingredient in diets for production animals. Canola protein is extracted and fully denatured to render the protein insoluble. Soluble ANF's (like glucosinolates) are washed from the protein. Protein is 100% dephytinized with conversion of phytate phosphorus to available phosphorus (www.canproingredients.ca). Van der Peet-Schwering et al. (2011[a]) investigated the nutrient composition, digestibility and energy value of canola protein concentrate in group housed organically housed piglets (see Table 6). To compare these data with the data in Table 5, it should be taken in mind that digestibility coefficients in the CVB-table are based on digestibility trials with growing and finishing pigs. In general, digestibility coefficients are lower in piglets than in growing and finishing pigs (Noblet and Shi, 1994). When optimizing diets, the difference in digestibility coefficients between piglets and growing and finishing pigs should be taken into account.

Table 6. Nutrients (g/kg) and digestibility coefficients (%) of canola protein concentrate in group housed organically housed piglets (Van der Peet-Schwering et al., 2011[a]).

	Canola protein concentrate
Nutrients:	
Net energy (MJ/kg)	7.98
Crude protein	573
Ileal digestible lysine	18.79
Ileal digestible methionine + cysteine	15.46
Ileal digestible threonine	17.42
Ileal digestible tryptophan	5.27
Digestibility coefficient[1]:	
Crude protein	72.3
Lysine	66.4
Methionine + cysteine	70.3
Threonine	68.6
Tryptophan	65.1

[1] For crude protein the faecal digestibility coefficient is presented. For the amino acids the apparent ileal digestible coefficients are presented.

The level of ileal digestible lysine of canola protein concentrate is lower than that of soy bean meal (18.79 versus 25.5 g/kg). The level of ileal digestible methionine + cysteine, however, is higher (15.46 versus 11.5 g/kg). The levels of ileal digestible threonine and tryptophan are comparable in canola protein concentrate and soy bean meal.

The level of glucosinolates is low with 2,59 µmol/g protein concentrate (www.canproingredients.ca). Van der Peet-Schwering et al. (2011[b]) studied the effect of canola protein concentrate as main protein source in the diet compared to soybean expeller on performance of organically housed weaned piglets. The test diet contained 10% canola protein concentrate. Feed intake and daily gain were similar and feed conversion ratio was better in weaned piglets fed the diet with 10% canola protein concentrate compared to the control diet. Orr at al. (2002) compared the growth performance in weaned piglets fed diets with 9% soy protein concentrate, 6.8% menhaden fishmeal, 5% spray-dried

blood plasma or 11% canola protein concentrate. They found no differences in feed intake, daily gain and feed conversion ratio between the four dietary treatments.

There are no digestibility and performance data of canola protein concentrate available in poultry, but the good digestibility in piglets may indicate that this concentrate is a promising protein source for poultry as well.

In conclusion, canola protein concentrate can be used in pig diets at an inclusion level up to 10%. At this inclusion level the content of glucosinolates in the diet is below the advised maximum level of 3 mmol glucosinolates per kg diet (Lamont and Lambrechts, 2005).

5.1.2 Soybeans heat treated, soybeans extracted/expeller and soy protein concentrate

Soybean meal is one of the most important protein rich feedstuffs used in diets for pigs and poultry. Most of the soybeans used in the EU are imported. European soybeans are only available on a small scale (see 3.2.1). Soybean meal has a high protein level (approx. 440-540 g/kg). Moreover, the level of most ileal digestible amino acids and the protein digestibility are high (Table 5). The level of methionine + cysteine, however, is relatively low. To our knowledge, there are no data available about the nutritional value of European soybean meal for pigs and poultry. Moreover, performance trials in pigs or poultry with European soybean meal are not available. Consequently, it is not possible to establish whether the nutritional value of European soybean meal differs from that of imported soybean meal.

In conclusion, European soybean meal is an interesting protein rich feedstuff in diets for pigs and poultry provided that the nutritional value is similar to the nutritional value of the imported soybean meal.

5.1.3 Sunflower seed extract and concentrate

Sunflower seed extract
Sunflower seeds are amongst the world most important oilseed crops. In addition to the oil they contain about 20% protein. Currently, defatted sunflower meal is mainly used in diets of adult animals (dairy cows, growing-finishing pigs), and only at very limited levels in diets for young animals (piglets, broilers) and humans. Crude protein content of sunflower seed extract is about 310 g/kg, and protein digestibility is moderate to good (Table 1). Already in 1947, it was concluded that sunflower seed extract could be a useful feed ingredient for growing-finishing pigs, although proportionally replacement of soybean meal by sunflower seed extract reduced growth rate of the pigs due to a lysine deficiency (Krider et al., 1947). More recently, however, it was concluded that the performances of piglets and growing-finishing pigs were not affected when soybean meal was replaced by twice-dehulled sunflower meal (Cortamira et al., 2000). This substitution, however, needs the contribution of synthetic lysine and vegetable oil as sources of complementary nutrients to match the nutrient profile. The energy values of sunflower seed expeller and extract were assessed in adult Leghorn-type cockerels as 9.46 and 7.62 MJ/kg, respectively (San Juan and Villamide, 2000). High fibre sunflower cake (37% crude fibre) up to an inclusion level of 20% in the diet did not reduce broiler performance, but even exerted some positive effects on digestion and small intestinal health (e.g. counts of Clostridium spp.), although villus height was reduced with increasing dietary sunflower cake levels (Kalmendal et al., 2011). Moreover, fibre degrading enzyme supplementation can further improve nutritive value of high fibre broiler diets, as shown by improved growth rate and feed efficiency (Raza et al., 2009).

Sunflower protein concentrate
By use of different extraction methods, it is technically possible to produce sunflower protein concentrates with a crude protein content of at least 75% (Salgado et al., 2011). Sunflower protein concentrates could be of interest in pig and poultry diets, especially in organic diets. Until now, however, no information is available regarding nutritional values of these concentrates for monogastrics. Moreover, costs of extraction methods have to be taken into account.

The potential application in feed can be significantly increased by reducing the levels of phenolics. Sunflower seed contains a high amount of phenolics, mainly consisting of chlorogenic acid (0.5 - 2.4% of dry matter) (Gonzalez-Perez et al., 2002). These compounds have the capability to interact with proteins, thereby decreasing the nutritional value for animal feed. Nevertheless, based on their homology with legume proteins, like soya, sunflower seeds have the potential to be high-value

ingredients. Rats that were fed increased amounts of chlorogenic acids (0, 0.056, and 0.28 mmol/g protein) showed increased excretion of faecal and urinary nitrogen. As a result, true nitrogen digestibility and net protein utilisation were adversely affected. Protein digestibility was decreased for lysine, tryptophan and sulphur containing amino acids (Rohn et al., 2006). Feeding full fat sunflower kernels (150 g/kg) to broilers resulted in shortening and thickening of the jejunal vili, hyperplasia and vacuolar degeneration of enterocytes, and hypertrophy and hyperplasia of goblet cells (Arija et al., 2000). These alterations could be the result of the presence of chlorogenic acid in full fat sunflower kernels, thereby explaining the reduced fat digestibility in previous experiments of the same authors (Arija et al., 1998). Despite these findings, no adverse effects of adding purified chlorogenic acid up to 6 g/kg diet on metabolizable energy value and digestibility of individual amino acids in broilers could be demonstrated (Trevino et al., 1998). In conclusion, more research is necessary to determine the effects of reducing the contents of chlorogenic acid in sunflower meals on digestibility and performance of broilers.

5.2 Legumes

The nutritional value of *Vicia faba*, lupines and peas is shown in Table 7.

Table 7. The nutritional value of Vicia faba, lupines and peas for pigs and poultry (CVB, 2007).

	Unit	Vicia faba, coloured	Vicia faba, white	Lupines, RE>335 g/kg	Peas
General chemical composition					
Dry matter	g/kg	863	872	888	867
Ash	g/kg	34	35	39	28
Crude protein	g/kg	251	275	372	211
Crude fat	g/kg	14	14	48	10
Crude fibre	g/kg	79	79	137	53
Starch	g/kg	326	338	21	387
Sugar	g/kg	28	39	49	43
NSP	g/kg	213	172	362	190
Ca	g/kg	1.0	1.8	2.4	1.0
P	g/kg	5.2	3.7	2.9	4.0
K	g/kg	12.2	13.3	8.5	10.0
Na	g/kg				
Nutritional value for pigs					
NE	MJ	8,33	8.89	8.40	9.46
SID protein	g/kg	199	227	263	180
SID Lysine	g/kg	12,6	15.1	15.5	11.8
SID Met+Cys	g/kg	2,9	4.2	6.8	3.4
SID Threonine	g/kg	6.3	7.5	10.7	5.4
SID Tryptophan	g/kg	1.4	1.8	2.5	1.2
SID Isoleucine	g/kg	7.9	9.4	12.9	6.6
Absorbable P	g/kg	1.9	1.4	1.5	1.8
Nutritional value for poultry					
ME Laying hens	MJ	10.37	11.15	8.50	11.36
ME Broilers	MJ	10.19	10.30	8.39	10.55
Dig. protein	g/kg	208	245	283	184
Dig. Lysine	g/kg	12.3	14.6	16.1	12.4
Dig. Met+Cys	g/kg	3.8	4.4	7.4	4.2
Dig. Threonine	g/kg	6.7	7.8	11.7	6.3
Dig. Tryptophan	g/kg	1.8	2.1	2.7	1.6
Dig. Isoleucine	g/kg	8.3	9.8	13.7	7.3
Dig. Arginine	g/kg	19.6	23.0	36.2	16.3
Dig. Valine	g/kg	8.9	10.5	13.1	8.4
Absorbable P	g/kg	2.3	1.6	1.4	1.7

5.2.1 Vicia faba

Vicia faba is well adapted to most climatic areas of Europe and is in Europe widely used for feed and food (Crépon et al., 2010). Based on flower colour, *Vicia faba* is grouped in white-flowering cultivars and in coloured-flowering cultivars. The content of ash, crude fat, crude fibre and starch are comparable in white-flowering and coloured-flowering cultivars (CVB, 2007). Protein content, protein digestibility, the content of ileal digestible amino acids and Net Energy, however, are higher in white-flowering cultivars than in coloured-flowering cultivars (CVB, 2007; Table 7). The content of protein and ileal digestible amino acids is lower in *Vicia faba* than in soy bean meal. Moreover, the ratio between ileal digestible methionine and ileal digestible lysine is lower in *Vicia faba* than in soy bean meal (Jongbloed et al., 2009). *Vicia faba* is mainly used in diets of sows and growing and finishing pigs and less in diets for piglets.

White-flowering *Vicia faba* has a low content of tannins and no condensed tannins and this allows a larger inclusion rate in animal diets for white-flowering cultivars than for coloured-flowered cultivars (Helsper et al., 2006). Makkar et al. (1997) compared the level of ANF's in six white-flowering and six coloured-flowering cultivars. The levels of total phenols, tannins and condensed tannins were 4.5 g/kg, 0.14 g/kg and 0 g/kg, respectively, in white-flowering cultivars and 20.1 g/kg, 14.1 g/kg and 26.2 g/kg, respectively, in coloured-flowering cultivars and hence lower in white-flowering than in coloured-flowering cultivars. TIA was low in both cultivars but higher in white-flowering cultivars than in coloured-flowering cultivars (3.05 versus 1.85 mg TIU/g dry matter). Low-tannin content generally results in higher protein and energy digestibility for monogastric animals (Crépon et al., 2010).
The ANF's vicine/convicine are specific for *Vicia faba* and they can disturb fat metabolism and fertility in laying hens (Helsper et al., 2006). In France, *Vicia faba* genotypes free of vicine/convicine, have been developed long ago (Duc *et al.*,1989). In organic poultry farming the cultivar Divine is the most popular *Vicia faba*, since this cultivar is almost free of convicine/vicine (Helsper et al., 2006). Convicine/vicine does not affect pig production (Grosjean et al., 2001). The development of *Vicia faba* cultivars with very low levels of vicine/convicine would be a real advantage in terms of nutritional performance in poultry diets (Crépon, 2010).

Van der Peet-Schwering et al. (2006) tested the effect on the performance of organically housed weaned piglets of 10, 20 and 30% *Vicia faba* (variety Aurelia, tannin-free) in the diet compared to a control diet containing 15% soybean expeller. They concluded that an inclusion level up to 20% tannin-free *Vicia faba* can be recommended in diets for weanling pigs. Higher levels decreased the performance of the piglets.

In a trail with organically housed weaned piglets, Van der Peet-Schwering et al. (2011[a]) investigated the nutrient composition, digestibility and energy value of *Vicia faba* protein concentrate.

Table 8. Nutrients (g/kg) and digestibility coefficients (%) of *Vicia faba* protein concentrate in group housed organically housed piglets (Van der Peet-Schwering et al., 2011[a]).

	Vicia faba protein concentrate
Nutrients:	
Net energy (MJ/kg)	9.55
Crude protein	390
Ileal digestible lysine	19.81
Ileal digestible methionine + cystine	4.07
Ileal digestible threonine	10.54
Ileal digestible tryptophan	2.09
Digestibility coefficient[1]:	
Crude protein	89.0
Lysine	81.2
Methionine + cystine	55.0
Threonine	77.5
Tryptophan	72.0

[1] For crude protein the faecal digestibility coefficient is presented. For the amino acids the apparent ileal digestible coefficients are presented.

The protein content in *Vicia faba* concentrate was lower than the protein content in soy bean meal (390 versus 464 g.kg). The level of ileal digestible lysine (19.8 versus 25.5 g/kg), methionine + cysteine (4.1 versus 11.5 g/kg), threonine (10.5 versus 15.1 g/kg) and tryptophan (2.1 versus 5.2 g/kg) were lower in *Vicia faba* protein concentrate than in soy bean meal. Performance trials with *Vicia faba* protein concentrate in diets for pigs and poultry were not found in literature.

In conclusion, *Vicia faba* with low tannin content can be used in piglet diets at an inclusion level up to about 20%. In poultry diets, it can be used at an inclusion level up to 20% in broiler diets (Farrell et al., 1999) and up to 30% in laying hen diets (Danner, 2003). It is recommended to use tannin-free varieties in diets for monogastrics and cultivars with very low levels of vicine/convicine in poultry diets.

5.2.2 Lupines

Lupines (white lupines, yellow lupines and blue lupines) are commonly cultivated in Europe and used in diets for pigs and poultry. The content of protein and of ileal digestible amino acids is lower in lupines than in soy bean meal. Moreover, the ratio between ileal digestible methionine and ileal digestible lysine is lower in lupines than in soy bean meal (Jongbloed et al., 2009).

All three lupine varieties contain various types of alkaloids from which the quinolizidin alkaloids are the most relevant ANF (Helsper et al., 2006). Alkaloids have a bitter taste and this may result in inhibition of feed intake, but also neurophysiological effects, e.g. tremors, convulsions and pulmonary arrest have been described (Kingsbury, 1964; cit. by Helsper et al., 2006). Low-alkaloid varieties, also known as sweet lupines, are generally available. In contrast to *Vicia faba* and peas, lupines contain hardly any trypsin inhibitor activity and only low levels of saponins (Helsper et al., 2006).

Helsper et al. (2006) measured the level of alkaloids in five lupine cultivars (Table 9).

Table 9. Level of alkaloids (mg/kg dry matter) in five lupine species.

	Alkaloids (mg/kg DM)
Bora (blue)	680 ± 170
Wodjil (yellow)	80 ± 8
Amber (yellow)	n.d.
Dieta (white, UK)	850 ± 270
Dieta (white, The Netherlands)	510 ± 130

n.d. = not detectable

The yellow lupines contain very low levels of alkaloids. The blue and white lupines have much higher levels of alkaloids than the yellow species. The cultivars tested by Helsper et al. (2006) were low alkaloid cultivars, but they still contain alkaloids levels that make it necessary to limit the level of these low alkaloid cultivars in pig diets. Dunshea et al. (2001) concluded that in pigs, feed intake is reduced when the alkaloid level rises above 200 mg/kg feed. With concentrations in the low alkaloid lupine cultivars ranging from 510 to 850 mg/kg, maximum inclusion rates in pig diets should be less than 20 to 40%. Within the same cultivar however (see results of Dieta in Table 9), alkaloid level varied between production location. Therefore, for safety an inclusion rate in pig diets of less than 20 % should be used.

Van der Peet-Schwering et al. (2006) tested the effect on the performance of organically housed weaned piglets of 10, 20 and 30% white lupines (*Lupinus albus*, variety Dieta, low alkaloid content) in the diet compared to a control diet containing 15% soybean expeller. They concluded that an inclusion level of up to 10% low alkaloid lupines can be recommended in diets for weanling pigs. Higher levels decreased the feed intake and daily gain of the piglets. These results are in agreement with results of Van Barneveld (1999). In a review, he concluded that inclusion of Lupines albus in pig diets at levels above 100 g/kg diet results in a significant reduction in feed intake and daily gain. Ferguson et al. (2003) reported that increasing the dehulled lupine content of diets from 0 to 300 g/kg diet caused a linear decrease in feed intake and daily gain. They used a low alkaloid lupine and suggested that the decrease in feed intake and daily gain was caused by the level of raffinose + stachyose in the lupines. Raffinose and stachyose are carbohydrates that are not digested in the small testine but fermented by bacteria in the hindgut. During fermentation, gases are produced that cause flatulence and that may suppress the feed intake and daily gain of the piglets (Ferguson et al., 2003).

In conclusion, low alkaloid lupines can be used in pigs diets at an inclusion level up to 10%. In poultry diets, it can be used at an inclusion level up to 20% in laying hen diets, whereas it should kept below 10% in broiler diets.

5.2.3 Pea

White-flowering peas are commonly cultivated in Europe and used at a large scale in diets for pigs and poultry. The content of protein and of ileal digestible amino acids is lower in peas than in soy bean meal. Moreover, the ratio between ileal digestible methionine and ileal digestible lysine is lower in peas than in soy bean meal (Jongbloed et al., 2009). Peas are used in diets for both young and older animals. White-flowering varieties contain a low amount of tannins and are often low in trypsin inhibitor activity (TIA). Grey peas are high in TIA and therefore not suitable in animal diets (Helsper et al., 2006). White-flowering winter peas show fourfold higher TIA than spring types (Mariscal et al., 2002). The choice of the cultivar is therefore very important (Helsper et al., 2006). Wiseman et al. (2003) showed that digestibility of methionine and cysteine in broilers is higher in pea cultivars low in TIA than in cultivars high in TIA (Table 10). Comparable results have been found in a study with pigs (Grosjean et al., 2000).

Table 10. Apparent ileal digestibility of methionine and cysteine in peas with low and high trypsin inhibitor activity (TIA) according to Wiseman et al. (2003).

	Pea A5		Pea B5	
	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.

Phytate also is an important ANF in peas. The concentration is highly variable with the cultivar, differs between locations and depends on the maturity stage of the seed (Helsper et al., 2006). Phytate can bind metal ions and thus affect the uptake of iron and zinc. In addition, phytate can inhibit protein availablity (Frederikson *et al.*, 2001). Soaking of pea meal at 45 °C is very effective to decrease phytate levels (Fredrikson et al., 2001).

Pea protein concentrate
Since a few years, pea protein concentrates are available for use in pig and poultry diets. The concentrate contains about 80% protein and a low level of TIA of 2 to 2.5 TIU/mg (www.roquette.com). Van der Peet-Schwering et al. (2011[a]) investigated the nutrient composition, digestibility and energy value of LYSAMINE®GP pea protein concentrate in groups of organically housed piglets (Table 11). To compare these data with the data in Table 7, it should be taken in mind that digestibility coefficients in the CVB-table are based on digestibility trials with growing and finishing pigs. In general, digestibility coefficients are lower in piglets than in growing and finishing pigs (Noblet and Shi, 1994). By optimizing diets, the difference in digestibility coefficients between piglets and growing and finishing pigs should be taken into account.

Table 11. Nutrients (g/kg) and digestibility coefficients (%) of pea protein concentrate in groups of organically housed piglets (Van der Peet-Schwering et al., 2011[a]).

	Pea protein concentrate
Nutrients:	
Net energy (MJ/kg)	11.20
Crude protein	835
Ileal digestible lysine	55.45
Ileal digestible methionine + cysteine	11.89
Ileal digestible threonine	27.40
Ileal digestible tryptophan	6.25
Digestibility coefficient[1]:	
Crude protein	91.9
Lysine	90.9
Methionine + cysteine	66.8
Threonine	83.5
Tryptophan	77.1

[1] For crude protein the faecal digestibility coefficient is presented. For the amino acids the apparent ileal digestible coefficients are presented.

The level of ileal digestible lysine is higher in pea protein concentrate than in soy bean meal (55.5 versus 25.5 g/kg). The level of ileal digestible methionine + cysteine is similar in pea protein concentrate and soy bean meal (11.9 and 11.5 g/kg). The levels of ileal digestible threonine (27.4 versus 15.1 g/kg) and tryptophan (6.3 versus 5.2 g/kg) are higher in pea protein concentrate than in soy bean meal .

Van der Peet-Schwering et al. (2011[b]) tested the effect on performance of organically housed weaned piglets of pea protein concentrate as main protein source in the diet compared to soybean expeller. The diet contained 8.5% pea protein concentrate. Feed intake and daily gain were similar and feed conversion ratio was better in weaned piglets that were fed the diet with 8.5% pea protein concentrate compared to the control diet. Parera et al. (2010) studies the growth performance in weaned piglets fed a diet with 9.6% soy protein concentrate or 9.5% pea protein concentrate. The crude protein content in soy protein concentrate and pea protein concentrate were 52.4 and 46.5%, respectively. They found no differences in feed intake, daily gain and feed conversion ratio between the two treatments. Valencia et al. (2008) however found a reduced performance in piglets fed 10.5% pea protein concentrate compared to 12.2% soy bean meal. The pea protein concentrate contained 52.5% protein and had a quite high level of TIA of 4.9 TIU/mg. There is only scarce information available about the use of pea protein concentrate in diets for broilers.

In conclusion, pea protein concentrate can be used in piglet diets at an inclusion level up to at least 8%. Higher inclusion levels can be applied provided a low TIA content whereas the use of pea protein concentrate high in trypsin inhibitors should be limited in piglet diets.

5.2.4 Chickpeas

Based on seed colour and geographic distribution, the chickpea is grouped into two types: Kabuli of Mediterranean and Middle Eastern origin, and Desi of Indian origin. Kabuli cultivars are white to cream coloured, whereas the seeds of Desi cultivars are wrinkled at the beak with brown, light brown, fawn, yellow, orange, black or green colour (Bampidis and Christodoulou, 2011). The chemical composition is shown in Table 12.

Kabuli chickpeas seem to be more attractive for pig and poultry diets than Desi chickpeas because of its lower fibre content and higher energy value.. Chickpeas, however, contain a number of anti-nutritional factors, like protease inhibitors, amylase inhibitors, phytolectins, polyphenols, and oligosaccharides (Singh, 1988). The most important protease inhibitors are trypsin and chymotrypsin, but they are very thermo-labile, especially under moist heat conditions. Phytolectins are toxic factors that interact with glycoproteins on the surface of red blood cells, causing them to agglutinate. Phytolectins are also very sensible to heat treatment. Polyphenols decrease protein digestibility by making protein partly unavailable for digestive enzymes. Dehulling, soaking and cooking will destroy these polyphenolic compounds.

Table 12. Chemical composition of Kabuli and Desi chickpeas grains (Bampidis and Christodoulou, 2011).

	Unit	Kabuli grain	Desi grain
Dry matter	g/kg	908	897
Ash	g/kg	34	34
Crude protein	g/kg	225	230
Crude fat	g/kg	62	48
Crude fibre	g/kg	47	88
NDF	g/kg	126	214
ADF	g/kg	54	132
Lignin	g/kg	13	12
Starch	g/kg	394	334
ME	MJ/kg dm	12.95	11.75
Lysine	g/kg	14.9	14.3
Methionine	g/kg	2.8	3.0
Cystine	g/kg	3.2	4.2
Threonine	g/kg	8.6	7.9
Tryptophan	g/kg	1.9	1.6
Valine	g/kg	9.8	9.0
Isoleucine	g/kg	9.1	8.9

In weaned piglets, the ileal apparent crude protein digestibility of a control diet based on casein, or a diet in which 50% of crude protein was supplied by white or black chickpea, was 0.912, 0.848, and 0.848, respectively (Salgado et al., 2001). In the chickpea diets, moderate villus atrophy and crypt hyperplasia were observed. Villus height at the ileum was smaller with the black compared to the white chickpea diet. The authors concluded that both white chickpea and black chickpea seem to be satisfactory protein and energy sources for the weaned piglet.

The inclusion of graded concentrations of chickpeas from 0 to 30% to diets of growing chickens decreased the apparent ileal digestibility of crude protein of the diets from 85.8 to 83.1% (Brenes et al., 2008). Extrusion of the chickpeas increased the crude protein digestibility, due to the inactivation of the trypsin, chymotrypsin, and α-amylase inhibitors. Apparent ileal digestibility of CP and apparent faecal digestibility of CF were improved (P<0.001) by extrusion. Increasing chickpea content in the diet did not affect weight gain, feed consumption and feed to gain ratio. Relative pancreas and liver weights, and relative lengths of duodenum, jejunum and ceca were increased in response to increasing chickpea concentration in the diet. Extrusion of chickpeas improved weight gain and reduced relative pancreas weight compared to birds fed raw chickpea-based diets. The authors concluded that the inclusion of chickpea up to 300 g/kg of chicken diets did not affect performance, but caused a negative effect on the relative weight of some digestive organs. Addition of 25% of chickpeas to a laying hen diet supported good performance, although there was concern about the increased weight of the pancreas (Perez-Maldonado et al., 1999). In broilers, diets with field peas and faba beans gave better growth rate and feed efficiency than those given sweet lupines and chick peas (Farrell et al., 1999). Similarly with findings in laying hens, the pancreas was significantly enlarged on the diets with chick peas. Steam pelleting of diets gave a consistent and positive response for weight gain and FCR. These authors recommend in broiler diets a maximal inclusion rate of 10% chick peas.

The threshold level of growing pigs (20 – 50 kg) to trypsin and chymotrypsin inhibitors was investigated by adding graded dietary levels of chickpeas (0, 250, 500, and 750 g/kg) up to 4.7 mg/g trypsin inhibitor levels and 4.5 mg/g chymotrypsin inhibitor levels (Batterham et al., 1993). Growth performance of the pigs fed the chickpea meals was similar to that of the pigs fed a control soya-bean meal diet. Inclusion levels of the chickpea meals did not affect organ weights. The results indicate that the growing pig can tolerate dietary levels of at least 4.7 and 4.5 mg of trypsin and chymotrypsin inhibitors, respectively per kg of diet.

In conclusion, raw chickpeas can be used in diets of broilers at inclusion levels up to 10% (Farrell et al., 1999) and in diets of laying hens and pigs up to 20% to support growth and egg production without detrimental effects on pigs and birds (Bampidis and Christodoulou, 2011). Higher inclusion levels can be used after removal of the anti-nutritional factors using heat treatments, which improves the nutritional value of chickpeas.

5.3 Leaf proteins

5.3.1 Grass protein and lucerne

Whole grass is already a common ingredient in organic pig husbandry. Organic pig farmers estimated that their gestating sows with access to a pasture received even up to 60% less feed than the usual amount (Bestman et al., 2001). Also 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 (Rivera-Ferre et al., 2007). Laying hens are able to consume considerable amounts of fresh grass, which might contribute for 12 – 13% of the total dry matter intake (Antell and Ciszuk, 2006).

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 wk later) stage, was 148 and 108 g/kg DM (Van Krimpen et al., 2006). Kemme et al. (2005) and Van Krimpen et al. (2006) investigated the digestibility and intake of different organic produced grass sources as well as lucerne hay in organic housed gestating sows (Table 13).

Table 13. Intake and digestibility of different organically produced grass sources and Lucerne hay in organic housed gestating sows (Kemme et al., 2005 ; Van Krimpen et al., 2006).

	Unit	Fresh grass (young)	Fresh grass (older)	Grass Silage	Grass Hay	Lucerne Hay
Dry matter content	(g/kg)	166	198	533	891	892
Protein content	(g/kg dm)	148	108	194	62	150
Protein digestibility	(%)	45.0	36.6	47.8	-26.7	38.5
Intake	Kg dm/sow/d	0.79	0.85	1.12	0.71	1.01
Dig. protein intake	g/sow/d	53	33	104	---	58

Protein digestibility of these grass sources ranged from a negative value in grass hay to 47.8% in grass silage (Kemme et al., 2005). Daily grass intake of the sows was 0.79 and 0.85 kg dm, for the young and older grass quality, respectively (Van Krimpen et al., 2006), thereby providing 53 and 33 g digestible protein per sow per day. Grass silage provided the highest amount of dig. protein (104 g/sow/d). The amount of dig. protein provided by lucerne hay was similar to that of fresh young grass (58 and 53 g/d).

Van der Peet-Schwering et al. (2010) observed that protein digestibility of grass silages in sows depended of grass yield/ha and ranged from 39.9% at a yield level of 5 ton dm/ha to 63.1% at 1.8 ton dm/ha. The protein content decreased and the fibre content increased with increasing grass yield/ha. More recently, it was found that gestating sows were able to consume even 1.5 – 1.6 kg DM grass silage per day (Bikker et al., 2011). DM content of these grass silages was 26%, which was significantly lower than in the study of Van Krimpen et al. (2006). Sows seem to prefer shortly chopped grass silage with a low dm content (Bikker et al., 2011). Intake of grass silages was found to vary largely between individual sows (Bikker and Wikselaar, 2012). Variation in silage intake levels might be reduced by mixing the grass silage with a high energy ingredient like wheat or corn cob mix, and by providing the opportunity for simultaneous silage intake for all sows. Organic housed growing finishing pigs were able to realize 6% of DM intake during the starter phase and 15% during the grower phase by consuming grass silage (Bikker and Binnendijk, 2012). Performance level of the grass silage treatment was reduced compared to the control treatment, e.g. due to overestimation of the digestibility and energy value of the grass silage.

Although amino acids must be absorbed in the small intestine to be used in protein metabolism, limited information is available on ileal digestibility of protein and amino acids in forages. From a study with growing finishing pigs, in which different forage meals (dried lucerne, white clover, and perennial ryegrass) as replacement for barley were compared with a barley-based control diet, it was concluded that, based on the amino acid appearance in the portal blood, the net absorption coefficients of the different amino acids did not change significantly among the different diets (Reverter et al., 2000). Total tract crude protein digestibility of different forage meals/grasses in growing finishing pigs, however, was rather low, varying between 71% (Lucerne) and 18% (perennial ryegrass) (Kemme et al., 2005 ; Lindberg and Andersson, 1998). Moreover, as a consequence of increased dietary fibre content, the crude protein digestibility will further decrease with increasing inclusion levels of forage meals in the ration. In addition, nutrient intake of pigs is affected by the dietary fibre content, resulting in reduced nutrient intake levels with increasing dietary fibre from forage meal (Lindberg and Andersson, 1998). Therefore, extraction of protein from grass, thereby separating proteins from fibres, might increase their applicability in pig and poultry diets (Chiesa and Gnansounou, 2011). These proteins can be valorised as alternatives to extracted soybeans (Van den Pol-Dasselaar et al., 2012). Until now, hardly any information regarding nutritional value of grass proteins for monogastrics is available. Growing finishing pigs fed a liquid grass extract (8 – 12% dry matter, 2 – 4% crude protein), thereby replacing 20% of a control diet, had a similar growth rate, feed conversion ratio and carcass weight compared to pigs fed the control diet (Maguire, 1971). Similar findings were reported by Houseman et al. (1976).

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 (Moughan and Rutherfurd, 2008). Moreover, further development of protein extraction techniques is necessary to increase protein yield and to make these techniques economically feasible (Van den Pol-Dasselaar et al., 2012).

It can be concluded that grass and lucerne hay to some extend can contribute to the protein supply of gestating sows and growing finishing pigs. The fibre content of these ingredients, however, will limit their use in monogastric diets. Biorefinery 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, no firm conclusion regarding their potential use in can monogastric diets be drawn.

5.3.2 Other leaf proteins

Information regarding application of European leaf proteins in pig and poultry diets is scarcely available. Therefore, some information about rats and non-EU leaf proteins is presented here. Already in 1965, digestibility of different leaf proteins, e.g. from rye, potato, pea, and red clover leafs, after freeze drying was determined in rats (Henry and Ford, 1965). Digestibility was reasonable, ranging from 70.6% in red clover leaf protein to 84.8% in rape leaf protein.

Nguyen et al. (2012) determined the ileal and total tract apparent digestibility of crude protein and amino acids in ensiled and dried cassava leaves (CL) and sweet potato vines (SPV) in growing (>60 kg BW) pigs. Ensiled material was mixed with rice bran (50 g/kg) and salt (5 g/kg) and thereafter stored in airtight plastic bags for two months. The dried material was sun dried for two to three days. The results of the chemical analysis of the leaf sources are shown in Table 14.

Table 14. Composition of ensiled and dried cassava leaves and sweet potato vines, as used in a digestibility experiment with growing pigs (Nguyen et al., 2012).

	Unit	Cassava leaf ensiled	Cassava leaf dried	Sweet potato vine ensiled	Sweet potato vine dried
Organic matter	g/kg DM	920	922	855	867
Crude protein	g/kg DM	242	299	197	229
Crude fat	g/kg DM	70	67	64	53
Crude fibre	g/kg DM	143	149	156	159
NDF	g/kg DM	365	369	415	412
Metabolizable energy	MJ/kg DM	10.8	10.6	9.5	9.5
Hydrogen Cyanide	Mg/kg DM	152	128	n.d.	n.d.

On a DM base, the leaf sources contain a substantial amount of crude protein. Cassava leaves contain considerable amounts of HCN, which might have anti-nutritional properties. The amino acid composition of the fresh, ensiled and dried cassava and sweet potato leaves is shown in Table 15.

Table 15. The amino acid composition of the fresh, ensiled and dried cassava and sweet potato leaves (g/kg DM) (Nguyen et al., 2012).

	Cassava leaves			Sweet potato vines		
	Fresh	Ensiled	Dried	Fresh	Ensiled	Dried
Crude protein	299	242	299	223	197	229
Lysine	13.0	11.3	12.6	9.7	8.3	8.3
Meth + Cyst.	6.2	4.8	6.3	4.5	4.2	4.3
Threonine	11.7	10.8	12.2	12.4	8.9	10.8
Arginine	17.2	14.4	16.8	14.7	14.0	14.0
Valine	12.7	11.7	12.3	12.8	9.5	12.0
Isoleucine	15.9	13.3	15.1	11.7	10.2	13.9

The first and second limiting amino acids were found to be methionine + cysteine and lysine. No indications of cyanide toxicity were observed. The ileal apparent digestibility coefficients for crude protein and the essential amino acids are presented in Table 16 and compared with the values of soybean meal (CVB, 2007).

Table 16. Ileal apparent digestibility coefficients (%) for crude protein and the essential amino acids of ensiled and dried cassava leaves and sweet potato vines, as used in a digestibility experiment with growing pigs (Nguyen et al., 2012) and compared with the values of soybean meal.

	Cassava leaf ensiled	Cassava leaf dried	Sweet potato vine ensiled	Sweet potato vine dried	Soybean meal
Crude protein	0.47	0.44	0.49	0.45	0.85
Lysine	0.70	0.68	0.72	0.67	0.89
Methionine + Cysteine	0.71	0.71	0.74	0.70	0.86
Threonine	0.66	0.63	0.75	0.71	0.84
Isoleucine	0.65	0.65	0.76	0.76	0.87
Valine	0.59	0.59	0.67	0.68	0.86

The amino acid digestibility of the ensiled sources were in most cases not different from the dried ingredients. Digestibility coefficients of crude protein and amino acids, however, were significantly lower compared to soybean meal.

From this experiment is was concluded that cassava leaves and sweat potato vines have the potential to improve protein and amino acid supply in diets for growing pigs, especially when combined with ingredients containing high concentrations of the first two limiting amino acids.

5.4 Aquatic proteins

5.4.1 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 (Alam et al., 1973 ; Gentile and Maloney, 1969 ; Pereira et al., 2000). 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 (Nichols and Wood, 1968 ; Schwartz et al., 1988). Intake of Chlorella extracts is associated with enhanced immune responses and antitumor effects (Tanaka et al., 1984).

Février and Sève (1975) incorporated dehydrated Spirulina (*Spirulina maxima*) in the diets of piglets, weaned already at 12 days of age. From 12 to 21 days of age, Spirulina was incorporated at a level of 12 % (25 % of total crude protein) in the ration, replacing dried skim milk. From 21 to 42 days of age, Spirulina was fed at a level of 8 % of the diet, replacing soybean meal. Although there was some reduction in digestibility of the diet when Spirulina was incorporated, growth was satisfactory and equivalent in all groups. The authors concluded that the metabolic utilisation of the fraction of absorbed feed was better for the Spirulina group than for the control group, notably during the period between 12 and 21 days, although the supply of lysine in the Spirulina group was 12 % lower.

Yap et al. (1982) replaced one-half of soybean meal (33 % of total dietary protein) in a corn-soybean meal/dried skim milk starter diet with algal proteins (*Spirulina maxima*, *Spirulina platensis*, and Chlorella sp.). The trial was performed with Yorkshire pigs weaned to a dry diet at 4 to 8 days of age. There was no significant difference between control and algal diets during the 15- and 26-day trial periods in growth, diarrhoea, loss of appetite, or toxicity. The researchers concluded that at least one-half of the protein supplied by soybean meal (one-third of the dietary protein) could be replaced by algal protein without adverse effects.

Grinstead et al. (2000) performed feeding experiments with dehydrated *Spirulina platensis* and weanling pigs (PIC, L326 X C22; initially 3.7 ± 0.85 kg and 11 - 12 days of age). From days 0 to 14 after weaning, pigs were fed a control diet or pelleted diets containing 0.2, 0.5, or 2 % *Spirulina platensis* replacing soybean meal on an equal lysine basis. With 2 % *Spirulina platensis*, only 3.2 to 3.4 % of total dietary lysine was replaced. No differences in pig performance, measured as average daily feed intake and gain, were observed during this interval. In contrast to pelleted diets, meal diets resulted in inconsistent responses to *Spirulina platensis*.

Algae meal from *Spirulina platensis* was evaluated as a poultry feed ingredient in an experiment with broilers (Gongnet et al., 2001). 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 by Novus (1992). 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 (Gongnet et al., 2001).

Daily supplementation of 2 g of a dried blue algae *Spirulina platensis* positively affected the reproductive properties of sows (Shimkus et al., 2009). The weight of a new-born piglet increased by 19.85% (P < 0.05), milk yield - by 11.23%, piglet weight on the 21st and 28th day of age by 17.12% (P < 0.05) and 16.56% (P < 0.05), respectively and liveability - by 10.1%. The amount of fat in the milk of sows increased by 0.33%, protein by 0.39% (P < 0.05) and lactose by 0.38% (P < 0.05).

Results available from literature suggest that algae can be considered as a useful protein source in monogastric diets. Further investigation, however, is needed concerning the composition, nutritive value and use of algae in animal nutrition (Christaki et al., 2010).

5.4.2 Duckweed

On a dry matter base, duckweed has a high protein content with a valuable amino acid composition (Rusoff et al., 1980). Chemical composition of different duckweed species is shown in Table 17. Protein content, however, is also affected by harvest date and location (Hoving et al., 2012).

Table 17. 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 18.

Table 18. Amino acid composition of several duckweed species (g/100 g protein) (Rusoff et al., 1980).

	L. Gibba	S. polyrhiza	S. punctate	W. columbiana
Lysine	4.1	4.3	4.3	3.4
Methionine	0.8	0.8	1.1	0.9
Threonine	3.2	3.5	3.3	2.6
Arginine	4.3	5.3	4.9	3.8
Valine	5.0	4.4	4.7	3.5
Isoleucine	3.9	3.8	3.8	3.1

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 (Haustein et al., 1990). 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 replacer 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 (Haustein et al., 1992).

Despite the nutritive value of duckweed, 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 before duckweed is widely accepted for nutrient reclamation and as a source of animal feed (Goopy and Murray, 2003). 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.

5.4.3 Seaweed

The moisture content of fresh seaweed (marine algae) is very high and might amount up to 94% of the biomass (Holdt and Kraan, 2011). The nutritional composition of seaweeds varies, depending on strain, season and area of production (Connan et al., 2004). 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) (Holdt and Kraan, 2011).

Carillo et al. (1992) provided some general nutritional characteristics of brown seaweed (Sargassum sinicola Setchel and Gardner), with and without washing with drinking water, after sun drying and grinding. The major components of those unwashed/washed seaweeds were 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 (29.01 and 24.68%), 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 (Whittemore and Percival, 1975). In this experiment, the digestibility of the residue of *Ascophyllum nodosum*, after extraction of alginate, was investigated in 40 kg pigs. The soluble energy sources were lost during the extraction process. The remaining carbohydrate sources in this seaweed extract were fucoidans and structural polysaccharides from the cell walls. The N fraction was largely insoluble, whereas the product might contain a low but significant level of protease inhibitor activity. The digestibility coefficients of the seaweed residue were 0.12 for energy and -0.25 for N, which was equivalent with a digestible energy value of 2.2 MJ/kg DM and a digestible crude protein value of -30 g/kg DM. The dietary inclusion level of the seaweed residue was 50%. Half of the pigs developed acute diarrhoea and refused to consume any more of the diet. Endogenous pig enzymes are not able to digest the nutrients that remained after extraction. The small amounts of digestible energy and nitrogen are mainly the result of microbial break down of the nutrients. The authors concluded that the seaweed residue in its present form is unsuitable as major dietary ingredient for the supply of energy or N to pigs. In line with these findings, 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 ($P < 0.05$).

It can be concluded that only a limited number of experiments has studied the nutritional value of seaweed for monogastrics. The general conclusion of these studies is that seaweed in its present form is unsuitable as energy and protein source for pigs and 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.

5.5 (Pseudo) cereals

5.5.1 Oat proteins

Oats are distinct among cereals due to their considerably higher protein concentration (Klose and Arendt, 2012). According to the CVB Table (2007) crude protein content of oat, wheat, barley and maize is 131, 111, 104 and 82 g/kg, respectively. At the same time oats possess a protein quality of high nutritional value and a special protein composition. Most cereals have a high percentage of prolamins, which usually constitute most of the storage proteins, but the major oat storage proteins belong to the salt-water soluble globulin fraction (CVB, 2007). Amino acid composition of oats is superior to that of other cereals due to the higher amount of limiting amino acids like lysine and threonine.

Further concentration of oat protein might be helpful, especially for application in organic diets. In literature, however, only one study was found that determined the nutritional value of oat protein concentrate in monogastrics (Delisle et al., 1991). The tested oat protein concentrate was prepared by alkali extraction, resulting in a crude protein content of 65.9%. In this study, oat protein concentrate was compared with skimmed milk powder. The amino acid composition of these sources is shown in Table 19.

Table 19. Amino acid composition (% of crude protein) of skimmed milk powder and oat protein concentrate as determined by Delisle et al. (1991).

	Skimmed milk powder	Oat protein concentrate
Crude protein	35.3%	65.9
Essential amino acids		
Arginine	3.4	8.5
Histidine	2.3	2.3
Isoleucine	3.9	3.4
Leucine	8.3	6.5
Lysine	7.0	3.4
Methionine	2.3	1.6
Phenylalanine	4.4	5.4
Threonine	4.1	3.1
Valine	5.2	4.5
Nonessential amino acids		
Alanine	2.8	4.0
Aspartic acid	6.5	6.8
Cystine	0.5	1.8
Glutamic acid	18.6	17.8
Glycine	1.8	3.7
Serine	5.2	4.4
Tyrosine	4.4	3.8

Two experiments were conducted with very young male pigs, from 24 to 72h after birth to 4 wk of age. A control milk replacer diet, in which 100% of the dietary protein originated from low-heat skimmed milk powder, was compared with the other diets in which either 25 or 50% of the milk protein was replaced by oat protein concentrate. Dry matter intake was not affected by dietary treatments while body weight gain declined and feed to gain ratio increased significantly when oat protein concentrate replaced 50% of the milk proteins. Digestibility of dry mater and nitrogen was similar for both the oat and milk protein treatments, but amino acid profiles differed between treatments. The authors concluded that oat protein concentrate can replace up to 50% of milk proteins in a milk replacer diet without apparent detrimental effect on dry matter intake and N digestibility, although growth rate of piglets will be reduced.

5.5.2 *Quinoa*

Due to its relatively high protein content of 12 to 18% (Balkema-Boomstra, 2004), its low level of anti-nutritional factors and its favourable amino acid profile, quinoa might be an interesting protein crop for monogastrics (Helsper et al., 2006). In a study with weaned piglets, however, crude protein content and *in vitro* crude protein digestibility of quinoa were found to be rather low (Van der Peet-Schwering et al., 2006). Addition of this quinoa quality to piglet diets (20, 40 and 60%), thereby replacing, wheat, barley and soybean expeller, resulted in reduced growth and increased feed conversion ratio, compared to piglets fed the control diet. These adverse results might be explained by reduced values of some ileal digestible amino acids in the diet (lysine, methionine, isoleucine, phenylalanine, aspartic acid, glutamine and proline), compared to the control diet. The composition of diets in this experiment was based on the assumption that the amino acid composition and digestibility of quinoa was similar to that of wheat. The results of this study, however, indicate that the level and ileal digestibility values of amino acids in quinoa were overestimated (Van der Peet-Schwering et al., 2006). An experiment of Diaz et al. (1995) showed that an inclusion level of 5% quinoa at the expense of cereals did not affect performance of weaned piglets, whereas feed conversion ratio deteriorated if 10% quinoa was added to the diet.
In an *in vitro* experiment, extraction of saponin increased the protein efficiency ratio of quinoa proteins and did not affect the biological value (Chauhan et al., 1999 ; Ruales and Nair, 1994). These results suggested that saponin removal from quinoa will increase the potential for this crop in monogastric nutrition. On the other hand, saponin could have functional properties, as activating the immune system of weaned piglets (Ilsley et al., 2005), and increasing the length of villi in the small intestine (Alfaro et al., 2007). These effects, however, could not be confirmed in a study in which saponin rich quinoa hulls were fed to weaned piglets (Carlson et al., 2012).
Although quinoa might have some promising nutritional properties, current knowledge of the nutritive value is not sufficient for accurate inclusion of this ingredient to diets of monogastrics.

5.6 Conclusions

Proteins in (defatted) oil seeds are very useful for application in pig and poultry diets, and the use of soybean, rape seed, and sunflower seed meal in these diets already is widespread. These protein sources are well known in terms of chemical composition and digestibility coefficients. It is assumed that the nutritional characteristics of European cultivated soybean meal will be similar to the ones cultivated in South America, but until now this is not proven. Less information is available with respect to concentrates of these protein sources. Experimental results indicated that rape seed (canola) protein concentrate can be used up to 10% in piglet diets.
Legumes, e.g. Vicia faba, lupines and peas, and chickpeas, can significantly contribute to the protein supply of pigs and poultry, although their anti-nutritional factors have to be taken into account. Results in piglets showed that the digestibility coefficients of pea protein concentrate were similar or even better than whole peas. Moreover, piglets that were fed a diet supplemented with pea protein concentrate performed equally well to piglets fed a soybean based control diet. Based on these results, and considering that the production process of protein concentrates from legumes is sustainable and already commercially available, it can be concluded that these concentrates represent a promising category of European produced high quality protein, especially for application in organic diets.
Results of organic housed sows and growing finishing pigs showed that grass proteins might be a useful protein source for pigs. The biorefinery techniques for separating proteins from leaves are currently not applied on a practical scale. Moreover, until now the nutritional value of these protein sources for pigs and poultry is not known.
Some aquatic proteins, e.g. micro algae and duckweed, might be valuable protein sources for pigs and poultry, whereas intact seaweed seems to be less suitable as animal feed ingredient, but information is scarcely available. Besides the necessary developments regarding protein extraction from these sources, more research is required to investigate the nutritional characteristics of these ingredients, including questions with respect to cell wall degradation, feed safety, and legislation.
Oat protein has a good nutritional value for monogastrics and can be used as high quality protein in diets for young piglets. Although quinoa might have some promising nutritional properties, current knowledge is not sufficient for accurate supplementation of this ingredient in diets of pigs and poultry.

6 Environmental issues

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. This chapter focusses specifically on the emission of greenhouse gasses (GHG) and N-requirement of the selected protein sources.

Feed production, including crop cultivation, feed processing and transport are responsible for about 54 – 73% of total GHG emissions per kg of pig (Basset-Mens and Van Der Werf, 2005). Therefore, 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 (Meul et al., 2012). GHG emission is expressed in g CO_2-equivalents per kg of crop yield, also known as carbon footprint (CFP).

Besides 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 (Meul et al., 2012). 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. Vellinga et al. (in preparation) elaborate the idea of Audsley et al. (2010) that human consumption is the driver and that all agricultural production systems are connected. This is especially the case for market oriented agriculture and to a lesser extent for non-commercial agriculture. From this point of view all land use change emissions related to conversion in agricultural land (direct and indirect) should be related to the agricultural land itself and all agricultural land is part of the global land use change emissions. Land Use relates to the changes in carbon stocks within one land use type.

Table 20 provides the values for carbon footprint, LuLuc and N-requirement. N-requirement is the amount of N-input per hectare via manure or fertilizer divided by the yield per hectare.

Table 20. Footprint values of the selected ingredients, derived from the CFP calculation tool 'Feedprint' (Vellinga, in preparation).

	Carbon footprint (g CO_2-eq/kg)[1]	LuLuc (g CO_2-eq/kg)	N-requirement (g N input/kg yield)[2]
Oil seeds			
Soybean meal (South America)	578 (67)	412	17.1
Rapeseed meal (Europe)	521 (28)	162	68.9
Sunflower meal (Europe)	474 (15)	396	14.5
Grain legumes			
Pea (Europe)	673 (0)	711	12.7
Vicia faba (Europe)	466 (0)	377	12.3
Lupine (Europe)	806 (0)	1052	9.6
Forage legumes			
Lucerne, artificially dried (Europe)	2190 (1195)	424	3.0

[1])Value between brackets is the CO_2-equivalent necessary for processing; it is assumed that this value will be significantly increased by upgrading the protein content to a concentrate level.
[2])This value refers to the primary product (e.g. soybeans instead of soybean meal).

Based on these values, own calculations showed that the CFP value of a standard grower pig diet amounted 491 CO_2-eq/kg. Excluding soybean meal from the optimization resulted in a increased CFP value of 518 CO_2-eq/kg, whereas a minimal CFP value (430 CO_2-eq/kg) was realized with a high (15%) soybean meal content.

CFP values are currently lacking for leaf proteins, aquatic proteins and cereal concentrates. Because drying and oil extraction processes are not relevant in grain legumes, CO_2 equivalents for processing for these crops were set at 0.

Artificially drying of ingredients, e.g. lucerne or leaf proteins, significantly increases CFP value. Grain legumes have a rather low N-requirement, whereas N-requirement of rapeseed meal is very high. On the other hand, grain legumes have relatively high CFP- and LuLuc values. From the limited number of available footprint data, it can be concluded that none of the ingredients has a good score for all sustainability criteria. More work has to make a well-considered balance of these criteria.

7 Considerations and inventory of desired actions

7.1 Summary of protein source characteristics

In Table 21, 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.

Table 21. Characteristics of the 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]
Reference							
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	-	?	?	?	?	?	+

[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
     +   = yes, under the condition that the starting material is of organic origin. Oil seed concentrates are only applicable in organic diets if hexane is not used for fat extraction.

7.2 Oil seeds

Within the category of oil seeds, European produced soybean meal seems to be the most promising alternative for the ones imported from South America. Nutritional value and especially protein digestibility is very good. Protein yield is already reasonable, but should be further increased to make this crop attractive for the farmer. To realize this, varieties have to be selected with an ultra-short growth season. Experts expect that it takes at least 10 years to realize a crop yield of 5 tons/ha, which is equivalent with 1750 kg protein/ha. The cultivation of rape seed and sunflowers is already common practice in Europe, but protein yield and nutritional value are lower compared to soybean meal. Increasing the areas of these protein crops in Europe, however, might help to reduce the amount of imported soybean products in short-term. Protein meals, derived via hexane extraction, are not allowed for use in organic diets. However, meals prepared by expelling to remove the fat are allowed. Hence, on the short term this type of rapeseed and sunflower meal might be an alternative for non-European soybean meal.

7.3 Grain and forage legumes

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 reasonable, but should be further improved to make it a real alternative for the farmer. Because of the sensitivity for pathogens and pests, however, a rotation period of at least 5 years has to be taken into account. The nutritional value of pea is very good. For producing legume concentrates, a sustainable process (wind sifting) is available. Based on a limited number of animal experiments, it can be concluded that legume concentrates are very useful ingredients for application in (organic) pig and poultry diets, thereby providing a real solution for the 100% organic ingredient requirement. Until now, however, there is no production of protein concentrate from organic produced peas.
Forage legumes , e.g. artificially dried lucerne, have a reasonable protein yield/ha, although nutritional value for the animal is rather low, and drying of the fresh product requires a lot of energy, resulting in a high CFP.

7.4 Leaf proteins

Because of the high fibre content, the nutritional value for pigs and poultry is low. Currently, however, a lot of experimental work is being conducted to develop biorefinery of leaf proteins. Separating proteins from fibres will increase the nutritional value of these products. However, the economics of the process involved is still hampering commercialization. Therefore, on short-term leaf proteins will not provide a solution for reducing soybean imports but they have the potential to become interesting.

7.5 Aquatic proteins

Production of aquatic proteins might be promising because of the low level of land use and the potential protein yield per hectare, as estimated on the basis of small scale pilots. Less information, however, is available with respect to the nutritional value of e.g. algae and duckweed, whereas some authors concluded that intact seaweed is unsuitable for monogastrics. High amounts of energy are required for drying the protein sources and the biorefinery to produce in produce protein fractions in addition to other compounds, is still in its infancy. Therefore, it is not expected that these protein sources contribute to reduction of soybean imports in short-term. Nevertheless, there certainly is a long-term perspective for these protein sources in maintaining food security for the increased human population in the world.

7.6 (Pseudo-) cereals

Although the protein contents of oat and quinoa compared to other cereals are relatively high, the protein yields, particularly for quinoa, are rather low. Oat protein contains relatively high amounts of favourable globulins. Oat protein concentrate was found to be a high value protein source for young pigs. Little information is available regarding the nutritional value of quinoa protein. Improving crop yields and more nutritional research is necessary before these products might contribute to a lower soybean import.

7.7 Land use

The European protein production can be increased by 1) increasing the yield/ha of current protein crops, 2) replacing crops with a low protein content by crops with a high protein content, and 3) by using fallow land for cultivation of protein crops. The first option is already discussed during this report. Regarding the second option, protein crops have to compete with cereals and root crops (potato, sugar beet) in Europe. Currently, we are more than self-sufficient on the EU-level for potatoes, thereby exporting about 1 million ton of seed potatoes and serious amounts of pommes frites. Moreover, the EU is self-sufficient for sugar beets as well, but experts expect a drop of 30% in production after the soon expected release of the fixed (high) price for sugar.
Regarding the third option, it is estimated that 1.8 million ha of fallow land is available in the Danube region in the South-East part of Europe (Bittner, 2012). The estimated minimum potential for cultivation of protein crops in Europe is 2.4 million ha. Leafs and aquatic protein sources are not in direct competition with the land use of other crops.

7.8 Economic aspects of European cultivated protein sources

Gross margins (€/ha) of protein crop cultivation in West-Europe are low compared with wheat cultivation, and therefore not very attractive for the farmers in this area. Differences between both type of crops, however, are low or absent in the southern and eastern part of Europe (Kamp et al., 2010). The main reason for the often higher gross margin of wheat cultivation is related to the difference in yield/ha. To solve this gap, some options are available:

- Increasing the price of protein crops
- Increasing the yield of protein crops
- Decreasing the costs of protein crop cultivation.

During the last decade, the Chinese imports of soybean meal shows an increasing trend. Moreover, the production of GMO-free soybean meal is decreasing, whereas the demand of this product in Europe is increasing. These trends might increase the price of soybean meal, which makes the cultivation of European protein crops in term more attractive.
Besides, feed producers in Europe are showing an increasing demand for RTRS-soy (Round Table on Responsible Soy). Producers and processors of soybean meal have to fulfil requirements regarding labor conditions, land rights, use of chemicals, water management, and land clearing (Van der Bijl, 2012). It is not expected, however, that the RTRS-requirements will negatively affect the soybean yield, nor will increase the price of this crop (Kamp et al., 2010).

As mentioned earlier in this report, breeding is an essential tool to increase the yield of protein crops. This, however, requires a lot of efforts from breeding companies and research institutes. Improving the management conditions might help to increase the yield as well. The main challenge is to provide improved plant protection against different types of pests that are persistent in the soil. Available tools are plot selection, use of pesticides and herbicides, and breeding to improve pest resistance (Kamp et al., 2010).

Due to the low level of N fertilizing in legumes, the cultivation costs of these protein crops are relatively low. A increased price of fertilizers, therefore results in a smaller cap in gross margin between these crops and cereals.
Feed producers optimize their diets on the basis of least cost price formulation. In this calculation process, the price of feed ingredients relative to their nutritional values are determining the inclusion

level in the diet. In most of the pig and poultry diets, energy is the most expensive part. Therefore, protein sources that still contain a considerable amount of energy have a higher nutritional value and feeding price, which increase the attractiveness to be included in the diet. As an example, the energy and crude protein values of rapeseed meal and soybean meal relative to wheat are shown in Table 22.

Table 22. Energy and crude protein values of rapeseed meal and soybean meal relative to wheat.

	Energy content	Crude protein content
Wheat	100%	100%
Rapeseed meal	64%	342%
Soybean meal	84%	423%

Although rapeseed meal contains 3.4 times more crude protein than wheat, the relative energy value is only 64%. Soybean meal even contains 4.2 times more crude protein, whereas the energy value is only 16% less compared to wheat. This partly explains the use of soybean meal as major protein source in pig and poultry diets.

8 Recommendations for further research

Plant breeding aspects
Although soybeans might be an interesting protein crop, the yield per hectare in Europe should be increased by further breeding programs to make this cultivation less attractive for farmers in Europe. Also oats needs intensive breeding input to increase yields per hectare.
Large scale cultivation, processing and feasibility of aquatic proteins for application as feed still needs more research.

Plant processing aspects
Enhancing plant protein content by processing still needs further developments. This is particularly relevant for processing steps to enhance the protein content of the aquatic resources algae and duckweed. Moreover, more knowledge should be gathered regarding the sustainability aspects involved in this processing and regarding the division among the various (by)products obtained by this processing.

Nutritional aspects
It is assumed that the nutritional characteristics of European cultivated soybean meal will be similar to the ones cultivated in South America, but until now this is not proven. More research is desired to determine the nutritional value of concentrates of the different oil seeds.
Although quinoa, leaf proteins and some aquatic proteins, e.g. micro algae and duckweed, might be valuable protein sources for pigs and poultry, the nutritional value of these protein sources for pigs and poultry is not known. Besides the necessary developments regarding protein extraction from these sources, more research is required to investigate the nutritional characteristics of these ingredients, including questions with respect to cell wall degradation, feed safety, and legislation.

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Appendices

Annex 1   Long list of potential protein rich ingredients for regional production

	Regional (N/W Europe)[1]	Regional (S/E Europe)[1]	Suitable for young animals[2]	Suitable for adult animals[2]	Suitable for organic feeds[2]	Further processing necessary	Sustain-ability	Obstruc--tions	Availability after 2020[3]	Already common practice	Suitable for short list
Category high crude protein content (> 25%), high protein digestibility (>75%), high methionine content relative to crude protein (> 1.6%)											
Fish meal	-	-	++	+	+/-	No	?	no	-/-	Yes	No
Milk powder, skimmed	+	+	++	+	+/-	No	?	no	-/-	Yes	No
Sesame seed extr./expeller	-	+	+	+	++	No	?	no	+	No	No
Maize gluten meal	+	+	++	+	++	No	?	no	?	Yes	No
Potato protein	+	+	++	+	-	No	?	no	+	Yes	No
Sunflower seed extr./expeller	+	+	+	+	+	No	?	no	+	Yes	No
Sunflow. protein concentrate	+	+	+	+	++	Yes	?	no	+	No	Yes
Rape seed prot. concentrate	+	+	+	+	++	Yes	?	no	+	No	Yes
Rape seed extr./expeller	+	+	+	+	+	No	?	no	+	Yes	No
Larvae meal/other insects	+	+	?	?	?	Yes	?	Yes	+	No	Yes
Mussel meal	+	+	?	?	?	Yes	?	Yes	-/-	No	No
Mushrooms	+	+	+	+	+	No	?	no	-	No	No
Category high crude protein content (>25%), high protein digestibility (> 75%), low methionine content relative to crude protein (<1.6%)											
Wheat gluten meal	+	+	++	+	+	No	?	no	+	Yes	No
Sorghum gluten meal	-	-	+	+	+	No	?	no	+	No	No
Whey powder	+	+	++	+	+	No	?	no	-/-	Yes	No
Brewer's yeast dehydrated	+	+	+	+	+	No	?	no	+	Yes	No
Soya beans extr./expeller	-	+	+	+	+	No	?	no	+	Yes	Yes
Soya beans prot. concentrate	-	+	+	+	+	Yes	?	no	+	Yes	Yes
Soya beans heat treated	-	+	+	+	+	No	?	no	+	Yes	Yes
Processed animal protein	+	+	+	+	-	No	?	Yes	+	Yes	No
Ground nut expeller dehulled	-	+	-	+	+	No	?	no	+	Yes	No
Blood meal spray dried	+	+	++	+	+	No	?	no	+	Yes	No
Blood plasma	+	+	++	+	+	No	?	no	+	Yes	No
Lupines	+	+	+/-	+	+	No	?	no	+	No	Yes

[1]) - = Cultivation not perspective in this region; + = cultivation perspective in this region.
[2]) - = Not suitable as feed ingredient for this category; +/- = moderate suitable; + = suitable; ++ = very suitable; ? suitability unknown
[3]) -/- = not available for feed after 2020; - = probably available after 2020; +/- = supposed to be available after 2020; + = available after 2020;   availability unknown

	Regional (N/W Europe)[1]	Regional (S/E Europe)[1]	Suitable for young animals[2]	Suitable for adult animals[2]	Suitable for organic feeds[2]	Further processing necessary	Sustain-ability	Obstruc--tions	Availability after 2020[3]	Already common practice	Suitable for short list
Feather meal hydrolyzed	+	+	-	+	n.a.	No	?	Yes.	+	No	No
Pea protein concentrate	+	+	+	+	+	Yes	?	no	+	No	Yes
Lentils	+	+	?	+	+	No	?	no	+	No	No
Chick peas	+	+	?	+	+	No	?	no	+	No	Yes
Category low crude protein content (<25%), high protein digestibility (>75%), high methionine content relative to crude protein (>1.6%)											
Millet	-	-	?	+	+	No	?	no	+	No	No
Maize	+	+	+	+	+	No	?	no	+	Yes	No
Corn Cob Mix	+	+	+	+	+	No	?	no	+	Yes	No
Maize feed flour	+	+	+	+	+	No	?	no	+	Yes	No
Rice hulled/dehulled	-	-	?	+	+	No	?	no	+	No	No
Rice protein concentrate	-	-	?	+	+	Yes	?	no	+	Yes	No
Maize feed meal	+	+	+	+	+	No	?	no	+	Yes	No
Maize grits	+	+	?	+	+	No	?	no	+	Yes	No
Rape seed	+	+	+/-	+	+	No	?	no	+	Yes	No
Quinoa	+	+	+/-	+	+	No	?	no	+	No	Yes
Category high crude protein content (>25%), moderate protein digestibility (<75%), low methionine content relative to crude protein (<1.6%)											
Linseed extracted/expeller	+	+	+/-	+	+	No	?	no	+	Yes	No
Cotton seed dehulled	-	-	-	+	+	No	?	no	+		No
Vicia Faba	+	+	+/-	+	+	No	?	no	+	No	Yes
Vicia Faba prot. concentrate	+	+	+/-	+	+	Yes	?	no	+	No	Yes
Category low protein content (<25%), high protein digestibility (> 75%), low methionine content relative to crude protein (<1.6%)											
Oats	+	+	+	+	+	No	?	no	+	+/-	Yes
Other cereals (e.g. wheat, barley, rye)	+	+	+/-	+	+	No	?	no	+	+	No
Brewer's grain	+	+	+/-	+	-	No	?	no	+	Yes	No
Peas	+	+	+	+	+	No	?	no	+	Yes	No
Category other ingredients											
Casein	+	+	++	+	-	No	?	no	-/-	Yes	No
Collagen and gelatin	+	+	-	-	-	No	?	Yes	-/-	No	No
Keratin	+	+	-	?	-	Yes	?	Yes	+	No	No
Algae	+	+	?	?	?	Yes	?	no	+	No	Yes
Seaweed	+	+	?	?	?	Yes	?	no	+	No	Yes
Leaf protein grass (rubisco)	+	+	?	+	+	Yes	?	no	+	No	Yes

[1]) - = Cultivation not perspective in this region; + = cultivation perspective in this region.
[2]) - = Not suitable as feed ingredient for this category; +/- = moderate suitable; + = suitable; ++ = very suitable; ? suitability unknown
[3]) -/- = not available for feed after 2020; - = probably available after 2020; +/- = supposed to be available after 2020; + = available after 2020;   availability unknown

	Regional (N/W Europe)[1]	Regional (S/E Europe)[1]	Suitable for young animals[2]	Suitable for adult animals[2]	Suitable for organic feeds[2]	Further processing necessary	Sustain-ability	Obstruc--tions	Availability after 2020[3]	Already common practice	Suitable for short list
Other leaf protein (spinach, sugar beet)	+	+	?	+	+	Yes	?	no	-/-	No	Yes
Leaf protein Jatropha (rub.)	-	-	?	+	+	Yes	?	no	+	No	No
Co-products bio ethanol pr. (only second/third generation products)	+	+	?	+	?	No	?	no	+/-	No	Yes
Olive protein	-	+	?	+	+	Yes	?	no	+	No	No
Crambe protein	+/-	+/-	?	+	+	Yes	?	no	+	No	No
Luzern	+	+	-/+	+	+	No	?	no	+	Yes	Yes
Coffee pulp	-	-	?	?	?	No	?	no	+	No	No
Duckweed	+	+	?	?	+	Yes	?	no	+	No	Yes
Shrimp meal	+	+	?	+	+	No	?	no	+	No	No
Hemp	+	+	?	+	+	No	?	no	+	No	No

[1]) - = Cultivation not perspective in this region; + = cultivation perspective in this region.
[2]) - = Not suitable as feed ingredient for this category; +/- = moderate suitable; + = suitable; ++ = very suitable; ? suitability unknown
[3]) -/- = not available for feed after 2020; - = probably available after 2020; +/- = supposed to be available after 2020; + = available after 2020;   availability unknown

Specific remarks

- **Fish meal:** production is not very common in Europe. This protein source will be appropriate for human consumption in the future.
- **Sunflower protein concentrate:** be aware of high content of polyphenols.
- **Insects:** on short term, cost price is a point of concern.
- **Mussel meal:** in Sweden, this source was considered as perspective full (PhD thesis ….)
- **Mushrooms:** See study Petra Becker about single cell proteins.
- **Sorghum:** climate conditions in Europe are not appropriate for cultivation.
- **Soya beans and co-products:** new cold tolerance varieties should be developed.
- **Horse beans:** unknown whether this plant can be cultivated in Europe (Ingrid).
- **Lupines:** due to low yields, cultivation of this crop is not attractive.
- **Pea protein concentrate:** separation in starch and protein provides two interesting ingredients.
- **Lentils:** due to low yields, cultivation of this crop is not attractive. Moreover, protein is appropriate for human consumption.
- **Chick peas:** in Russia, cultivation of chick peas will significantly increase in the coming years.
- **Millet:** because of the small size of the seed, large harvest losses occur.
- **Quinoa:** this crop seems promising, although crop yield and nutritional value for piglets failed.
- **Lin seed extract/expeller:** this protein source is a co-product from linen production, which is nearly disappeared in Europe.
- **Vicia Faba:** at the moment, crop yields are too low, whereas this crop is sensitive to pathogens. Attention should be given to improvement of this crop.
- **Oat protein:** this crop fits very well in European conditions. Functional properties of oats are investigated in some current projects.
- **Keratin:** isolating of protein from this matrix is difficult.
- **Olive protein:** olive cultivation is concentrated in a small specific region. Transferring this cultivation to other seems not perspective full.
- **Crambe:** improvement of crop yield is necessary.
- **Shrimp:** isolating protein from chitin matrix is difficult.