Purifying manure effluents with duckweed

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Samenvatting
Het doel van de studie was het uitvoeren van een korte literatuurinventarisatie over het zuiveren van mesteffluenten met behulp van eendenkroos, waarbij gekeken is naar kroossoorten, teelt, oogstmethode, gebruik en valorisatie van eendenkroos. De resultaten van de studie laten zien dat eendenkroos gebruikt kan worden om nutriënten terug te winnen uit mesteffluenten en dat het eendenkroos gebruikt kan worden als bron van veevoer, energie en ingrediënten.

Summary
The objective of this study was to perform a short literature survey to provide information about purifying manure effluents with duckweed with regard to varieties, cultivation, harvesting methods, utilization and valorisation of duckweed. The results of the study show that duckweed can be used to recuperate nutrients from manure effluents and that the concerning duckweed can be utilized as a source of feed, energy and ingredients.

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SUMMARY

DUCKOFARM is a sustainable livestock farming concept presently being developed by Cornelissen Consulting Services BV (CCS) an ProfiNutrients BV. In DUCKOFARM, animal manure is digested anaerobically, for generating renewable energy and releasing organically bound nutrients (N and P). Subsequently, the digestate is refined into N and P fertilizers (ammonium sulphate and struvite) by novel processes. The remaining effluent is fed into a duckweed pond, where the duckweed purifies the effluent by taking up leftover nutrients (N, P and others). Once harvested, duckweed substitutes for imported feed products like soybeans as a protein-rich animal fodder.

The objective of this study was to perform a literature survey to provide information about purifying manure effluents with duckweed with regard to varieties, harvesting methods, utilization and valorisation of the duckweed. Selected databases were consulted for an inventory of research papers about the use of duckweed for purifying manure effluents. Also a limited Internet search was part of the survey.

The results of the survey shows that duckweed can be used to recuperate nutrients from manure effluents and that the duckweed can be utilized as a source of feed, energy and ingredients.

The information in the literature about varieties and cultivation of duckweed grown on (diluted) manure effluents shows:

- Manure effluents have a too high concentration of nutrients to be used directly as a growth medium for duckweed. Therefore the manure effluents have to be diluted.
- In general the optimum level of nitrogen seems to be around 30 mg N/l, nevertheless maximum biomass yields have also been observed at levels of 72 mg N/l. The optimum level of nitrogen in the pond water is influenced by species and growing conditions (pH, temperature, etc.).
- Higher N content in the growth medium gives higher protein content, but there is an optimum level. The optimum level appears to be depended on the NH_4-N content, temperature and pH (influencing the toxicity of ammonia).
- The biomass yield and composition of the duckweed is depended on species, growing conditions and management of the duckweed pond.
- The root length of the duckweed is inversely related with the crude protein content of the duckweed.

The information in the survey about harvesting methods of duckweed shows:

- A couple of companies have developed a machine for harvesting of duckweed, but these were developed for waterways and not specifically for water ponds.
- On the basis of one publication it appears that harvesting less duckweed but more frequently favoured nutrient recovery and biomass production.

The information in the literature about utilization and valorisation of duckweed grown on (diluted) manure effluents shows:

- Duckweed can be used as a protein source for animal feed.
- The specific biogas production of duckweed is around 300 l CH_4/kg VS.
- By adjusting the growing conditions of duckweed the starch content of duckweed can be increased which could make the duckweed a suitable source of ethanol production.
- Duckweed can be used as a source of ingredients such as protein, cellulose and colouring agents.

Some papers showed that duckweed could be converted into a fuel source.

1 Introduction

1.1 Background

Responding to market pressures caused by on-going globalization, physical and operational sizes of EU farms are ever-increasing. Next to scale enlargement, the abolishment of EU milk quotas in 2015 will lead to greater numbers of cattle per unit land area, and hence in the Netherlands it will lead to larger manure surpluses. As a partial solution, the sustainable livestock farming concept DUCKOFARM is presently being developed by Cornelissen Consulting Services BV (CCS) and ProfiNutrients BV. In DUCKOFARM, animal manure is digested anaerobically, for generating renewable energy and releasing organically bound nutrients (N and P). Subsequently, the digestate is refined into N and P fertilizers (ammonium sulphate and struvite) by novel processes. The remaining liquid is fed into a duckweed pond, where the duckweed cleanses the water by taking up leftover nutrients (N, P and others). This step eliminates the need for complete nutrient removal during the manure refining, hence allowing use of lower-cost equipment. Once harvested, duckweed substitutes for imported soybeans as a protein-rich animal fodder. Economic feasibility of the first two steps of concept (Anaerobic digestion and manure refinery) was basically confirmed in a feasibility study (Kroes and De Jong, 2014). Additional confirmation of market prices and off-farm demand for the produced duckweed and fertilizer products are needed before upscaling of the concept. To this end, a project has been carried out to investigate the feasibility of a duckweed pond for polishing the effluent of the manure refining technology and utilization and valorisation of the duckweed. As part of this project, a short literature survey has been made about purifying manure effluents with duckweed.

1.2 Objective

The objective of this study was to perform a literature survey about purifying manure effluents with duckweed with regard to varieties, harvesting methods, utilization and valorisation of duckweed.

1.3 Approach

Selected databases have been consulted for an inventory of research papers about the use of duckweed for purifying manure effluents. Other effluents such as domestic wastewater or industrial wastewater were not part of this survey. There were no restrictions made regarding the date of publications. The following databases have been consulted with the following search queries:

- Scopus: (duckweed) AND (manure OR slurry OR dung OR muck OR effluent OR wastewater)
- WUR Manure Treatment Archive : (duckweed OR eendenkroos OR kroos)
- WUR Library Catalogue: (duckweed OR eendenkroos)

In addition a limited internet research has been carried out based on the same key words, but an extensive Internet search was not part of this literature review.

2 Varieties and cultivation

2.1 Properties of duckweed

Species
Duckweed is a generic term often used for the floating water plants duckweed and duckweed fern. According to Dutch species databases Soortenbank.nl and www.nederlandsesoorten.nl duckweed is classified as part of the family *Araceae* and duckweed fern is part of the family *Salviniaceae*. The genera of duckweed are: *Spirodela, Landoltia, Lemna, Wolffia and Wolffiella*. The genus of duckweed fern is: *Azolla*. The website Wayne's Word Lemnaceae On-Line (Armstrong, 2015) gives an extensive overview of the identification and information about duckweed species.

Composition
Depending on the growth circumstances, duckweed contains between 3-14% dry matter. Optimal growing conditions (high growth rates) results in relative low dry matter contents. Especially circumstances which lead to the forming of starch (such as high light intensity and high CO2-concentrations) results in greatly increased dry matter contents. The dry matter content is also depended on the species. The nutrient content in duckweed is depended on species and the nutrient concentrations in the water. Table 1 gives an overview of the nutrients content of several species and table 2 gives an overview of the organic contents of duckweed and duckweed fern (Otte, 2012).

Table 1

Nutrient contents of three duckweed species in % of dry matter (Otte, 2012)

	N	P	K	Na	Ca	Mg	Mn	Fe	Zn
Spirodela	3.99	0.97	0.27	0.12	0.31	2.17	0.54	0.74	0.05
Lemna minor	8.74	0.83	1.53	0.02	0.18	1.92	0.03	0.06	0.05
Lemna trisulca	4.79	1.81	0.26	0.05	0.34	2.90	0.55	0.44	0.04

Table 2

Organic contents of duckweed and duckweed fern in % of dry matter (Otte, 2012)

Component	Duckweed (*Lemnaceae*)	Duckweed fern (*Azolla*)
Protein	6.8 – 45.0	13.0 – 23.4
Carbohydrate	14.1 – 43.6	6.4 – 61.0
Fat	1.8 – 9.2	4.4 – 6.3
Fiber	5.7 – 16.2	9.5 – 24.5
Ash	8.0 – 27.6	9.7 – 23.8

Balasubramanian and Kasturi Bai (1992) assessed the efficiency of common duckweed (*Lemna*) in harvesting the nutrients from biogas slurry effluent. The effluent originated from a biogas plant fed with cattle waste and was diluted 100 times to a 1% concentration level. The mean biomass yield was 1.06 g dry mass/m^2/day. The harvested duckweed had a dry matter content of 3.37%. The nutrient composition of the plant in percentage of dry matter was: 75.20% VS, 2.63% N_{kj}, 0.70% P, 2.82% K, 0.70% Na and 0.65% Ca.

Growth
The most commonly used method of measuring growth of duckweed is to count fronds, sometimes the increase in dry weight is used. Table 3 gives an overview of the maximum growth rates of some species found in the Netherlands at optimal temperature (25 – 28°C) (Otte, 2012).

Table 3

Growth rates of several species of duckweed per day at optimal temperature (Otte, 2012)

Species	Measured growth rate (min. – max.)
Lemna minor	0.16 – 0.36
Lemna gibba	0.12 – 0.33
Spirodela polyrhiza	0.21 – 0.39
Lemna trisulca	0.024
Not specified, field research	0.10 – 0.35

2.2 Performance of duckweed on manure effluents

Mestayer *et al*. (1984) investigated the growth of duckweed (*Spirodela punctata*) on cattle manure dilutions under different environmental conditions. In experiment 1 effects of different manure concentrations (2.5, 5.0 and 10.0 gram manure per litre tap water) and growth period lengths (1, 2, 3 or 4 days) were determined. The results showed that the daily growth rate and solar energy conversion efficiencies of duckweed were similar at all manure dilutions. In experiment 2 it was shown that duckweed was capable of night growth. The growth in terms of organic matter was the same or more during the night as during the day. The source of energy for night growth was not determined.

DeBusk *et al*. (1995) evaluated the effect of season and hydraulic retention time on the P uptake from dairy lagoon wastewater by duckweed (*Lemna obscura*) in Okeechobee, Florida, USA. The dairy lagoon wastewater (13.7 mg P/l and 62.9 mg TN/l) was diluted with well water on an one-to-one ratio. The maximum P uptake of 20 mg P/m^2/day was identical for both test periods in February and July. During July, the total P concentration was reduced from 7.3 to 2.4 mg/l at a 7-day HRT and during February, from 6.2 to less than 2.0 mg P/l at a 14-day HRT.

The 'Milieucoöperatie de Peel' in the Netherlands has tested in the late nineties a small duckweed pond for the treatment of separated liquid fraction of pig manure. The duckweed pond was covered with a plastic tunnel in which the carbon dioxide and ammonia rich air from beneath the slats in the stable was blown. The water evaporation was 1 cubic metre per m^2 per year after the first year of testing (Engelberts, 1997).

Anh and Preston (1997) carried out experiments to evaluate the use of diluted effluent from continuous flow biodigesters fed with cattle manure as a fertilizer for duckweed (*Lemna spp).* The results showed that when the influent concentrations of N was in the range of 10-30 mg/l, the fresh biomass was in the order of 100 g/m^2/day with 5-6% dry matter and 37-40% protein in the dry matter. The optimal harvest frequency was at a two-day interval.

Le (1998) carried out experiments to investigate the effect on yield and protein content of duckweed when biodigester effluent, pig and cow manure were used at different levels of N in the growth medium (10, 20 and 30 m N/l). The results showed that the manure supported slightly higher yields than the digester effluent, but the digester effluent supported higher protein content in the duckweed. The optimum level of nitrogen in the pond water was in the range of 20 to 30 mg/l. The root length of the duckweed was inversely related with protein content. Higher pH of the pond water in the range of pH 6.4 to 7.2 was associated with higher protein content in the duckweed.

Bergmann *et al*. (2000a) carried out a study to select superior duckweed (*Lemnacaea*) genotypes for the utilization of nutrients in animal wastes. A two-step protocol was used to select the promising duckweed geographic isolates to be grown on swine lagoon effluent from a collection of 41 geographic isolates which originated from the worldwide germplasm collection. The first step consisted of in vitro screening in jars (23^{o}C, 16-hour photoperiod) on a synthetic medium that approximated swine lagoon effluent based on a ranking of fresh weight gain, percent dry weight, protein content and total protein production. The results showed large differences among geographic isolates and that the variation among geographic isolates within species was as great as or greater than of species for all variables tested. The six most highly ranked and two other geographic isolates were used in the second step in which they were tested on swine lagoon effluent in a greenhouse. The characteristics of the used swine lagoon effluent are shown in table 4.

Table 4

Characteristics of swine lagoon effluent used to test eight geographic isolates of duckweed and used in a further study to test with three isolates with diluted effluent (Bergmann et al., 2000a+b)

N_{kj}	262.00 mg/l	OPO_4	50.57 mg/l	K	270.67 mg/l	Cu	0.28 mg/l
NH_4	172.00 mg/l	TOC	421.33 mg/l	Cl	149.67 mg/l	Zn	0.39 mg/l
NO_3	0.64 mg/l	COD	1286.67 mg/l	Na	82.70 mg/l	Fe	2.33 mg/l
NO_2	0.30 mg/l	TS	0.20%	Ca	61.67 mg/l	pH	7.84
P_{total}	87.53 mg/l	VS	42.41%	Mg	50.00 mg/l		

The results of the second step showed that none of the geographic isolates grew rapidly, but their non-quantified visual estimate response marked differently. *Lemna gibba* 8678 (India) appeared to tolerate the lagoon effluent better than the other geographic isolates and maintained healthy fronds that multiplied slowly. *Spirodela punctate* 7776 (Australia) remained relative healthy without noteworthy multiplication. *Lemna minor* 8627 (Denmark) exhibited about 50% survival of the fronds without noteworthy multiplication. The other geographic isolates did not tolerate swine lagoon effluent. These results indicate that selection from a genetically diverse duckweed collection for a given trait could afford significant improvement compared to duckweeds that happen to grow locally (Bergmann *et al*., 2000a). Bergmann *et al*. (2000b) tested these three geographic isolates further in a new study on diluted swine lagoon effluent since the full-strength lagoon effluent did not allow healthy, rapid growth. The results showed that *Lemna gibba* 8678 and *Lemna minor* 8627 gave greater total biomass production than the *Spirodela punctate* 7776 and that they should be grown on 50% swine lagoon effluent for effective treatment and also a healthy duckweed. The *Lemna minor* 8627 achieved on 50% swine lagoon effluent reduction rates of 83% N_{kj}, 100% NH_4, 49% TP, 31% OPO_4-P, 68% TOC, 21% K, 28% Cu and 67% Zn. The *Lemna minor* 8627 was further investigated in a study by Cheng *et al*. (2002). In this study the nutrient removal from swine lagoon effluent by *Lemna minor* 8627 was investigated in batch experiments at 4 dilutions (50%, 33%, 25%, and 20% strenght) in outdoor tanks under natural climate conditions of North Carolina, US. The nitrogen removal rate was highest at 50% dilution in both the spring and fall tests, respectively 1.24 and 2.11 g N_{kj}/m^2/day. Also the phosphorous removal rate was highest at 50% strength in the fall tests with 0.59 g P_{total}/m^2/day, but in the spring test the phosphorus removal rate at the 33% strength was slightly higher than at 50% strength, 0.29 vs 0.26 g TP/m^2/day. But the duckweed growth rate showed opposite results with the highest growth rates at the 20% strength both in the spring and fall tests with 28.5 g/m^2/day and 15.7 g/m^2/day. The results showed that the wastewater concentrations and seasonal climate conditions had direct impacts on the growth of the duckweed and nutrient removal in outdoor tanks.

Effluent collected from a displacement tank from a fixed dome bio-digester fermenting cow dung was used in a study to determine the biomass yield, nutritive value and efficiency of utilization of nutrient by different genera of duckweed, under the same nutritional and management conditions. Table 5 shows the results from this study (Chowdhury *et al*., 2000).

Table 5

Results of different species of duckweed grown in media (80 l water) fertilized once with 6.648 kg of anaerobically fermented cattle manure during a growing period of 34 days with 17 harvests (Chowdhury et al., 2000).

Parameter	*Lemna*	*Wolffia*	*Spirodela*	SED	Significance
Total N in medium (mg/l)	45.18	42.00	32.50	5.42	NS
Temperature	28.37	29.50	29.55	0.2189	NS
pH	7.92	8.20	8.18	0.0616	NS
Mean yield per harvest					
Biomass yield (kg/ha/d)	631	906	-	83.3	p<0.05
DM yield (kg/ha/d)	14.80	14.57	-	2.29	NS
CP yield (kg/ha/d)	4.83	4.32	-	0.79	NS
ADF yield (kg/ha/d)	1.83	4.17	-	-	-
Chemical composition					
Dry matter (g/100 g fresh)	6.14	4.25	8.50	0.684	p<0.05
Ash (g/100 g DM)	19.36	23.37	15.57	2.300	P<0.05
OM (g/100 g DM)	80.65	76.63	84.84	6.150	NS
CP (g/100 g DM)	34.06	31.25	28.75	4.581	NS
ADF (g/100 g DM)	12.39	28.59	19.47	-	-
Efficiency of utilization					
Dry matter (%)	137	132	1.5	7.9	p<0.01
Organic matter (%)	133	122	3.6	7.6	p<0.01
Crude protein (%)	314	273	1.5	72.3	p<0.01

The lack of mean yield of *Spirodela* is caused by cessation of growth of *Spirodela* within four days after startup (after two harvests) with no exact cause for cessation. But it might have been caused by liberation of gases from the anaerobic fermentation of the effluent which resulted in excessive froth on the surface of the media which resulted in floating of *Spirodela* inhibiting the nutrient uptake and/or the pH might have been too high and caused ammonia toxicity (Chowdhury *et al*., 2000).

Sultana *et al*. (2000) determined the effect of nutrient loading frequency on the biomass and nutrient yield of duckweed (*Lemna perpusilla*). Anaerobically fermented cattle manure effluent at two intervals: 1-day and 6-day interval. There were no significantly differences found in terms of biomass yield or nutrient composition (DM, OM, crude protein) of the duckweed.

Lampheuy *et al*. (2004) conducted an experiment to compare the growth response of duckweed (*Lemna minor*) at five levels of N from raw cow manure and effluent from a biodigester fed with the cow manure in plastic baskets. The water surface in each basket was 0.16 m^2 with 10 cm depth. The biomass yield increased with increasing N application and was higher for the biodigester effluent than for manure, while the root length decreased with increasing N application and was shorter in the biodigester effluent than with the cow manure, see table 6. The crude protein content increased with increasing N application, but the rate of increase was more marked with the biodigester effluent.

Table 6

Characteristics of duckweed grown on different sources and levels of N (Lampheuy et al., 2004).

N Level	Yield		Root length		Nitrogen		Dry Matter		Crude Protein	
(kg/ha)	(g/m^2/day)		(cm)		(%DM)		(%)		(%)	
	Effl.	Man.	Effl.	Man.	Effl.	Man.	Effl.	Man.	Effl.	Man.
0	12.2	7.8	2.09	2.21	2.66	2.71	4.99	5.62	16.7	16.9
50	44.4	28.6	2.05	2.68	3.95	3.35	5.85	4.90	24.7	21.0
100	60.2	44.7	1.27	2.31	4.87	3.46	5.40	6.00	30.5	21.6
150	65.6	54.4	0.91	1.96	5.32	3.49	5.83	5.98	33.2	21.8
200	63.6	62.2	0.94	1.58	5.53	4.26	5.21	6.34	34.5	26.6

Effl. = Effluent
Man. = Manure

Tu *et al*. (2012b) conducted experiments to determine the effect of different levels of cattle manure effluent from a biodigester on the yield and composition of duckweed and to determine the effect on the composition of duckweed after transferring the duckweed to plain water. The results showed there was no difference in dry matter content, but there were differences in chemical composition, biomass yield and the root length (table 7). After transferring the 12BE effluent to plain water the starch content increased after 5 days from 2.05 to 2.63% of DM.

Table 7

Effect of different levels of cattle manure effluent from a biodigester effluent on the composition and yield of duckweed (Tu et al., 2012b).

Parameter	OBE	4BE	8BE	12BE	16BE	20BE	SEM	P_{value}
Levels of dilutions								
Biodigester effluent	0	4	8	12	16	20		
Fresh water (%)	100	96	92	88	84	80		
N-content pond								
N (mg/l)	0	24	48	72	96	120		
Mean chemical								
Dry matter (%)	5.00	4.99	5.04	5.01	5.02	5.02	0.017	0.335
OM (% of DM)	88.4[a]	87.2[b]	85.3[ab]	85.4[ab]	85.7[ab]	84.1[b]	0.227	0.028
N (% of DM)	3.74[a]	3.75[a]	4.47[ab]	4.89[b]	4.14[ab]	4.22[ab]	1.42	0.028
CP (% of DM)	23.4[a]	23.4[a]	28.0[ab]	30.6[b]	25.9[ab]	26.4[ab]	0.661	0.019
CF (% of DM)	10.9[a]	9.15[ab]	8.76[b]	7.41[bc]	7.34[bc]	5.66[c]	0.734	0.016
NDF (% of DM)	19.8[a]	18.3[ab]	18.1[ab]	16.0[ab]	14.0[ab]	13.1	0.457	<0.001
Mean biomass yield								
Fresh (g/m^2)	955[b]	1100[b]	1308[ab]	1468[a]	1428[a]	1253[ab]	94.9	0.018
DM (g/m^2)	47.8[b]	54.8[b]	66.0[ab]	73.5[a]	71.6[a]	62.8[ab]	4.7	0.018
DM (ton/ha/yr)	12.5	14.3	17.2	19.2	18.7	16.4		
Mean root length								
Root length (cm)	2.45	1.48	1.21	0.78	0.53	0.37	0.05	<0.001

abc Mean values without common letter differ at P<0.05

Xu *et al*. (2012) evaluated a pilot-scale duckweed pond at a commercial swine farm for the treatment of anaerobically digested swine wastewater. After filling the pond with well water, wastewater was intermittently added to maintain the NH_4-N concentration of pond at about 1.5 mmol/l. The performance of the duckweed (*Spirodela polyrrhiza)* in the pond was monitored during a 26 week period from May to November 2010. During this period, the production rate of protein was 2.68 g/m^2/day and of starch was 1.88 g/m^2/day. The removal rate of NH_4-N was 1.68 mg/m^2/day and of PO_4-P 0.28 mg /m^2/day.

Since little attention has been given into mixture of different duckweed species in treating wastewater, Zhao *et al*. (2014) researched the performance of local strains in mixture and monoculture in treating swine wastewater during a 12-day period. For the study, *Lemna punctate OT*, *Lemna minor OT* and their mixture were cultured in diluted swine wastewater with 51 mg NH_4-N/l and 12 mg PO_4-P/l. Furthermore, *S. polyrhiza C1*, *L. minor C2* and *L. punctata C3* and their different combinations were investigated under similar conditions, but with slightly modified concentrations of NH_4-N (31.6 mg/l) and (PO_4-P 2.4 mg/l). The results are shown in table 8.

Table 8

Nutrient content of duckweed in swine wastewater on day 12 (Zhao et al., 2014).

Strains	N (%)	P(%)	Starch (%)	CP (%)	Biomass (g/m2)
swine wastewater with 51 mg NH_4-N/l and 12 mg PO_4-P/l					
1) *L. punctate OT*	4.81	0.46	16.74	30.06	37.5
2) *L. minor OT*	5.15	0.81	9.37	32.19	31.6
3) Mixture 1+2	4.61	0.39	15.52	28.81	41.4
swine wastewater with 31.6 mg NH_4-N/l and 2.4 mg PO_4-P/l					
4) L. punctate C3	4.04	0.53	28.60	25.25	66.55
5) L. minor C2	4.35	1.25	13.42	27.19	55.69
6) S. polyrhiza C1	3.47	0.49	22.92	21.69	48.39
7) Mixture 4+5	4.19	0.23	24.80	26.19	77.87
8) Mixture 4+6	3.51	0.43	29.67	21.94	71.36
9) Mixture 5+6	4.25	1.02	13.35	25.56	73.00
10) Mixture 4+5+6	4.01	0.45	23.33	25.06	76.00

Adhikari *et al*. (2015) investigated the nutrient reduction of dairy wastewater by treatment in a hybrid wetland system. The hybrid system consisted of a primary surface flow wetlands with duckweed (*Lemna minor*) followed by three subsurface wetlands and then a secondary surface-flow wetland with pea gravel, bulrush and begger-tick. The dairy wastewater was strained of the large solid particles and diluted with tap water to three strengths of wastewater: 250 mg/l COD (low), 500 mg/l COD (medium) and 1500 mg/l COD (high). The results of the nutrient reduction are presented in table 9. The duckweed production was highest at an influent concentration of about 32 mg/l N_{total}.

Table 9

Influent and effluent concentrations of COD, N_{total}, P_{total} and E. coli in primary and secondary wetlands during low, medium and high COD concentrations (mean ± error) (Adhikari et al., 2015).

Strains	Loading	Primary wetlands		Secondary wetlands	
		Influent	**Effluent**	**Influent**	**Effluent**
	Low	268.8±44.1	130.5±2.0	86.2±5.7	89.5±3.0
COD (mg/l)	Medium	542.3±42.6	309.8±8.8	131.8±11.5	105.0±3.4
	High	1544.0±190.3	879.4±31.6	373.8±9.6	257.3±5.7
	Low	30.9±0.5	13.4±0.8	4.6±0.4	3.9±0.2
N_{total} (mg/l)	Medium	35.6±4.3	33.8±1.0	23.4±3.5	13.9±1.6
	High	118.9±3.0	98.0±1.7	63.8±1.7	45.8±0.9
	Low	2.9±0.5	2.1±0.2	2.4±0.5	2.3±0.3
P_{total} (mg/l)	Medium	1.6±0.9	2.7±1.0	1.7±1.5	1.3±0.5
	High	25.2±2.8	14.9±0.9	11.7±0.9	9.8±0.4
	Low	38.8±33.3	19.7±12.4	0.0±0.0	0.0±0.0
E. coli	Medium	0.0±0.0	0.0±0.0	0.0±0.0	0.0±0.0
	High	265.0±86.2	96.7±23.6	27.7±9.8	14.8±3.9

3 Harvesting methods

Hardly any references could be found about harvesting methods for duckweed from ponds or creeks. The few methods found are described in this chapter.

3.1 Harvesting equipment

Wheeled excavators

In 2007 a pilot study was carried out a to gain experience with the possibilities of collecting and processing duckweed into feed for cows (Holshof *et al.*, 2009) in which two different harvesting methods were used to harvest duckweed from creeks. At one location the duckweed was harvested by a wheeled excavator with a flat sieve pan and much attention was paid to get good quality duckweed (the harvesting spot in combination with the condition of the duckweed: smell and colour), see figure 1. The harvesting of duckweed with a flat sieve pan can be seen in a video online in the library of Wageningen UR (Anonymous, 2007).

Figure 1 *Harvesting duckweed from a creek with a flat sieve pan.*

At another location the duckweed was harvested by a wheeled excavator with a dredge which was covered with fine gauze, which was used to see whether the harvesting could be done more quickly, see figure 2.

Figure 2 *Harvesting duckweed from a creek with a dredge covered with fine gauze.*

Both harvesting methods had advantages and disadvantages. The first method relatively time consuming, but the harvested product was relatively clean and did not need post-treatment. The second method was twice as fast, but the product contained much undesired material. This was partly due to the location (waste such as bottles, plastic and the like) and partly to the method (more subaquatic plants and more sludge). The authors concluded that harvesting of duckweed in creeks has to be further optimised in order to enable large-scale duckweed harvesting (Holshof *et al*., 2009).

Automatic solar-powered conveyor belt
Bom Aqua BV (www.bom-aqua.nl) has developed an automatic solar-powered conveyor belt (Krooswiel) for the harvesting of duckweed. By this method, in principle without manpower, duckweed is harvested by a paddle wheel powered by solar energy, see figure 3.

Figure 3 *Harvesting duckweed from a creek with an automatic solar-powered conveyor belt.*

The conveyor belt method disturbs the water surface considerably less than the wheeled excavator method in which the duckweed disperses quickly, so the excavator has to be moved often. A prerequisite is, however, that the equipment is placed favourably with respect to the direction of the water current and placed at the location where the duckweed is accumulated, for example, at a pumping station or a sluice (Holshof *et al*., 2009).

Duckweed Guzzler with belt
Bom Aqua BV (www.bom-aqua.nl) has developed the Duckweed Guzzler for the removal of duckweed in shallow waterways. The Duckweed Guzzler makes use of present water current and accelerates this current with the Duckweed Guzzler, see figure 4. By using a Duckweed Band the duckweed can be removed from the water system. The target price for the Duckweed Guzzler is circa €5.000 and combined with a Duckweed Belt €20.000. A video of the operation of the Duckweed Guzzler and Belt can be seen online at the website of Bom Aqua BV (Anonymous, 2015).


Figure 4 *Harvesting duckweed with the Duckweed Guzzler and Belt (Bom Aqua B.V.).*

Aquatic weed harvesters
Aquatic weed harvesters are designed for the removal of excessive aquatic plant material and other vegetation in waterways, see figure 5. These work boats are not designed to specifically remove duckweed, but might be altered in such a way that they can be used in duckweed ponds.

Figure 5 Harvesting water plants with an aquatic weed harvester.

Colubris

Colubris Technologies (www.colubris-technologies.nl) has developed an installation for harvesting of duckweed by sucking the duckweed into a special basket from which the duckweed is pumped to the bank of the waterway, see figure 6.

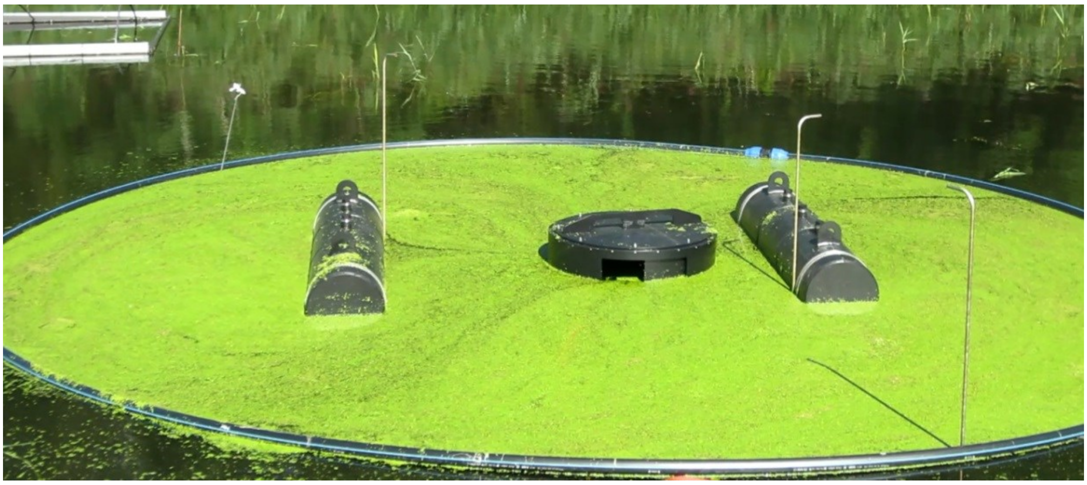

Figure 6 *Colubris duckweed harvester*

3.2 Harvest regime

Xu and Shen (2011) have carried out research into the effects of harvest regime and water depth on the duckweed growth and nutrient removal from swine wastewater by a *Spirodela oligorrhiza* system. In the study the following four harvest regimes were examined in tote boxes (26 x 36.5 x 15.5 cm) at a water depth of 10 cm:

- Harvesting 20% of duckweed twice per week,
- Harvesting 40% of duckweed once per week,
- Harvesting 60% of duckweed once every 2 weeks, and
- Harvesting 80% of duckweed once every four weeks.

The results showed that harvesting less duckweed biomass at a shorter time interval resulted in a higher biomass density. Although this caused some crowding, the nutrient recovery and biomass production was favoured. The regime of harvesting 20% of duckweed twice per week resulted in the highest specific growth rate which led to a duckweed production of 20.0 g fresh biomass/m^2/day and a removal of 83.7% of the nitrogen and 89.4% of the phosphorous. The study on water depth was carried out using cylindrical plastic water drums (d=40 cm) at a depth of 10, 20 or 40 cm with an initial concentration of ±67 mg N_{total}/l and ±18 mg P_{total}/l. The experiment showed that a deeper system resulted in a better nutrient removal in terms of total amounts, although it required a longer residence time for nutrient concentrations to decrease. At a depth of 40 cm, the removal rates for nitrogen and phosphorous were more than 3 times as high as those obtained at a depth of 10 cm. Furthermore, the biomass production and protein content of the duckweed increased at higher surface loading of nutrients.

4 Utilization and valorisation

4.1 Animal Feed

Goopy and Murray (2003) have reviewed the role of duckweed as a source of animal feed. The results from feeding experiments with a fairly wide range of monogastric animals indicate that at low to moderate levels (5 up to 25%) duckweed is well assimilated. When fed at higher levels (>25%) there is some evidence that some type of anti-nutritional factors limits intake and growth. Feeding experiments with ruminants are less common, but untreated (wet) duckweed has been fed at high levels (up to 10% live-weight of cattle) without negative effects. There are concerns about sequestration of heavy metals and the possible transference of pathogens through feeding animals with plant material which has grown on their own waste. The very limited research in this area suggests that this may not be a problem, but there is potential for great harm, and so the issue needs to be resolved.

Nutrient content

Culley and Epps (1973) determined the nutritive value of Dotted Duckweed (*Spirodela oligorrhiza*, which recently became known as *Spirodela punctata* or as *Landoltia punctate*) grown on an anaerobic swine waste lagoon at different dates during the fall and winter of 1970/71. The results are shown in table 10. Compared to the nutritive value of duckweed from natural waters the crude protein and fat content were higher, but the fibre content was lower.

Table 10

Chemical composition of dried Dotted Duckweed grown in an anaerobic swine waste lagoon (Culley and Epps, 1973).

Date	CP	Fat	Fibre	Ash	Ca	K	P	Mg	Fe	Mn	Water	Cu	Zn
18/9/70	37.9	5.8	8.9	11.7	-	-	1.40	-	-	-	8.7	-	-
8/10/70	35.8	3.4	8.6	13.7	0.94	2.26	1.32	0.25	0.41	0.15	10.0	15	124
20/10/70	40.6	3.9	7.1	13.1	0.97	2.41	1.64	0.27	0.50	0.11	9.3	15	128
1/11/70	40.9	4.2	7.6	12.3	0.93	2.56	1.67	0.24	0.50	0.18	8.4	10	104
4/12/70	36.6	4.3	7.2	12.7	0.83	1.70	2.84	-	-	-	8.8	-	-
25/1/71	36.1	5.4	7.6	10.8	0.69	2.76	1.39	-	-	-	9.9	-	-
14/2/71	37.0	5.2	7.3	9.9	0.76	1.39	2.84	-	-	-	7.3	-	-
25/2/71	37.0	6.4	6.4	10.0	0.70	1.50	2.00	-	-	-	9.0	-	-

Russof *et al*. (1980) investigated the chemical and the amino acid composition of four species of duckweed grown on anaerobic dairy waste lagoons. The results of the study are presented in tables 11 and 12.

Table 11

Chemical composition of four duckweed species grown on anaerobic dairy waste lagoons (Russof et al., 1980).

	L. gibba	*S.polyrhiza*	*S.punctata*	*Wolfia*
Dry matter (%)	4.6	5.2	5.1	4.8
Chemical composition (g/kg DM)				
Ash	14.1	15.2	13.7	17.1
Crude protein	25.2	29.1	28.7	36.5
Fat	4.7	4.5	5.5	6.6
Crude fibre	9.4	8.8	9.2	11.0

Table 12

Amino acid composition (in g/100 g protein) of four duckweed species grown on anaerobic dairy waste lagoons (Russof et al., 1980).

	L. gibba	***S.polyrhiza***	***S.punctata***	***Wolfia***
Cysteine	<0.05	<0.05	<0.05	<0.05
Aspartic	7.12	7.55	7.38	5.63
Threonine	3.20	3.45	3.31	2.55
Serine	2.61	2.80	2.83	2.28
Glutamic	7.60	8.00	7.69	5.76
Proline	2.93	3.28	2.95	2.41
Glycine	3.79	3.95	3.93	3.04
Alanine	4.59	4.48	4.79	3.75
Valine	4.96	4.40	4.71	3.49
Methionine	0.83	0.83	1.07	0.87
Isoleucine	3.87	3.75	3.76	3.06
Leucine	7.15	6.85	6.88	5.83
Tyrosine	2.91	3.05	3.14	2.17
Phenylamine	4.45	4.20	4.38	3.60
Histidine	1.89	2.15	1.90	1.18
Lysine	4.13	4.30	4.26	3.37
Arginine	4.29	5.25	4.86	3.78
True protein	66.32	38.29	67.80	52.77

N.B. Any thryptophan present would have been destroyed by the acid hydrolysis.

Cheng and Stomp (2009) reviewed the potential use of duckweed grown on diluted anaerobically treated swine wastewater as a protein source for animal feed. Depending on the species and strain within species a protein content ranging from 15 to 45% dry weight can be achieved. A comparison was made between three duckweed species and protein from grains, legumes, milk-casein and the amino acid requirement of laying hens, see table 13. They conclude that overall the biological value of duckweed total proteins would be less than that of soybean meal. However, animal feeding trials with duckweed have shown promising results.

Table 13

Amino acid composition of bulk protein (g/100 g protein) of duckweed species, grains, legumes, casein and recommended levels of essential amino acids for chicken feed (Cheng and Stomp, 2009).

Amino Acid	*Lemna gibba*	*Spirodela punctata*	*Spirodela polyrrhiza*	Green Gras	Soybean Meal	Peanut	Rice	Corn Gluten Meal	Casein	Levels Chicken Feed
Leu	7.15	6.88	6.85	10	8.0	6.7	8.2	15.3	10.0	7.5
Ile	3.87	3.76	3.75	5	6.0	4.6	5.2	4.9	7.5	5.0
Val	4.96	4.71	4.40	5	5.3	4.4	6.2	5.1	7.7	5.0
Met	0.83	1.07	0.83	2.5	1.7	1.0	3	2.35	3.5	2.0
Cys	NA	NA	NA	2.0	1.9	1.6	1.3	1.65	0.4	3.6
Phe	4.45	4.38	4.20	5-6	5.3	5.1	5.0	5.6	6.3	4.4
Tyr	2.91	3.14	3.05	5.0	4.0	4.4	5.7	2.3	6.4	6.4
Lys	4.13	4.26	4.30	5.5	6.8	3.0	3.2	1.85	8.5	4.0
Thr	3.20	3.31	3.45	5.4	3.9	1.6	3.8	3.0	4.5	3.5
Trp	NA	NA	NA	2.2	1.4	1.0	1.3	0.5	1.3	1.0
His	1.89	1.90	2.15	2.0	2.9	2.1	1.7	2.1	3.2	1.9
Arg	4.29	4.86	5.25	7.0	7.3	11.3	7.2	3.25	4.2	5.0
Ser	2.61	2.83	2.80	5	4.2	NA	NA	NA	3.3	NA
Pro	2.93	2.95	3.28	NA	5.0	NA	NA	NA	13.1	NA
Gly	3.79	3.93	3.95	NA	NA	5.0	NA	NA	2.1	NA
Glu	7.60	7.69	8.00	11.5	18.4	17.7	NA	NA	23.0	NA
Asp	7.12	7.38	7.55	5.3	NA	NA	NA	NA	7.0	NA

Chicken feed

Anderson *et al*. (2011) evaluated duckweed grown on effluent from anaerobic digestion of swine wastewater as a protein source in laying hen feed. The dried duckweed had a dry matter content of 97.83%, a crude protein content of 29.05%, a fat content of 5.02% and an acid detergent fibre content of 25.08%. The laying hen diet contained 12.6% duckweed. The duckweed diet had no impact on hen performance compared to the control diet. The duckweed diet resulted in a significant increase in B-grade eggs of 2%. However there was no difference in the nutrient composition of the eggs except for Omega 3 levels which were 0.06% higher ($P<0.001$).

Duck feed

Tu *et al*. (2012a) evaluated the use of duckweed as a replacement for soybean meal in a diet on growth performance and carcass traits of growing Muscovy ducks and to determine the apparent nutrient digestibility and N-retention. Two kinds of duckweeds were used in the study: 1) high protein duckweed cultivated in ponds supplied with biodigester effluent, and 2) low protein duckweed collected from natural ponds. In the experiment three diets were tested: a control diet including rice bran and soybean meal, rice bran with high protein duckweed and rice bran with low protein duckweed. All three diets contained the same level of crude protein of 15% in DM. The live weight gain was highest in the high protein duckweed diet (28.7 g/day) and lowest on the low protein duckweed diet (22.3 g/day), while the control diet showed an intermediate gain (25.4 g/day). No difference in carcass traits were observed with exception of the gizzards, which were the lowest for the control diet. The apparent digestibility and N-retention of the different diets are presented in table 14.

Table 14

Apparent total tract digestibility of dietary components and N-retention in ducks fed fed high (HPDW) or low (LPDW) protein duckweed (Tu et al., 2012a).

	Control	HPDW	LPDW	SEM	P_{value}
Apparent digestibility. %					
DM	74.6[a]	76.0[a]	72.5[b]	0.622	0.009
Organic matter	77.0[a]	79.1[a]	76.0[b]	0.521	0.007
NDF	39.8[a]	48.2[b]	46.7[b]	1.264	0.002
Crude fibre	40.2	40.7	39.0	1.247	0.629
N intake, g/day	2.19[a]	2.36[b]	2.09[c]	0.055	0.020
N retention, g/day	1.75[b]	1.94[a]	1.66[b]	0.060	0.027

abc Mean values in the same row without common letter differ at P<0.05

Pig feed

Domínguez *et al*. (1996) carried out experiments to determine *in vitro* N digestibility and the ileal digestibility of nutrients by pigs fed a low fibre sugar cane molasses-soybean meal diet containing supplements of high fibre and N content. One of the fibre sources used was duckweed (*Lemna minor*), collected from a secondary lagoon containing effluents from pig stables. The estimates of the protein digestibility in duckweed for pigs are for *in vitro* 674±78 g/kg and *in vivo* 560±69 g/kg. The composition of diets and ileal digestibility of nutrients and energy in pigs fed duckweed are presented in table 15.

Table 15

Composition of diets and ileal digestibility of nutrients and energy in pigs fed duckweed (Domínguez et al., 1996).

	0	100	200	SE	P_{value}
Composition					
Ash (g/kg DM)	121	151	177		
N (g/kg DM)	21.4	23.6	26.0		
Crude fibre (g/kg	25.5	41.4	56.4		
NDF (g/kg DM)	42.9	77.2	111		
Energy (MJ/kg)	14.6	14.6	14.6		
Digestibility					
DM (g/kg)	800a	782a	734	±4.1	0.05
Organic matter	819	792	769	±11.0	
N (g/kg)	719	662	648	±14.2	
Crude fibre (g/kg)	133	203	304	±111	
NDF (g/kg)	341	423	488	±80	
Energy (J/KJ)	809a	798a	754b	±12.0	0.05

ab Values within a row with the same superscript were not significantly different (P<0.05)

Du (1998) evaluated duckweed (*Lemna minor*) as a protein supplement in diets for growing-fattening pigs fed on local resources at small-holder farm level in Vietnam. On all five farms the pigs supplemented with duckweed consumed more dry matter and grew 35% faster (552 gram/day versus 404 gram/day). The stimulating effect on weight gain might be partially explained by the additional intake of protein.

Technology developed by Parabel Inc. is capable of extraction of crude protein (CP) and amino acids (AA) from duckweed (*Lemna*) which results in a lemna protein concentrate (LPC) that contains approximately 68% CP. Rojas *et al*. (2014) carried out experiments to determine the concentration of digestible energy (DE) and metabolizable energy (ME), the standardized total tract digestibility (STTD) of P, and the standardized ileal digestibility (SID) of CP an AA in LPC. The DE of the LPC was 4342 kcal/kg DM and the ME was 3804 kcal/kg DM. The STTD of P of the LPC was 72.8% and the SID of CP was 78.12%, while the SID of all AA 80.25% was.

Cattle feed
Holshof *et al*. (2009) evaluated duckweed grown in creeks as feed source for dairy cattle on two commercial dairy farms. There were no problems encountered during the processing of duckweed into standard concentrate with 7% duckweed, a protein-rich concentrate with 25% duckweed or concentrate with 96% duckweed and 4% molasses for dairy cattle. Only a qualitative assessment was made on the uptake of the duckweed concentrate by the dairy cattle. All three kinds of duckweed concentrates were well eaten by the dairy cows.

4.2 Biogas production

Clark and Hillman (1996) investigated the enhancement of biogas production in laboratory-scale anaerobic digesters using duckweed as a coproduct to the digestion of poultry manure. The authors state that iron has been identified as being the most important micronutrient to anaerobic bacteria, due its redox properties and the role which it plays in energy metabolism. Duckweed has a high iron content of 21000 mg/kg dry weight. Fresh poultry manure diluted to 5% total solids content was used as the feedstock for the anaerobic digesters. Duckweed replaced 14.5% of the diluted poultry manure during codigestion in the batch experiments. Despite the total biogas production per digester the specific biogas production was comparable with 0.295 l/g VS for the poultry manure and 0.281 g/l VS of the co-digested poultry manure. According to the authors this indicates that the effect of the duckweed is to increase the rate of decomposition, but not the total amount of biogas production. And the effect was identified as being due to its high iron content. Subsequent semi-continuous digesting experiments supported this conclusion.

Banning (2011) has performed batch test to determine the biogas production of Lesser Duckweed (*Lemna minor*) and Duckweed Fern under mesophilic conditions. The anaerobic digestibility of both duckweeds were comparable. After shredding the biogas production of both duckweeds was around 300 l/kg VS and the biogas production of non-shredded duckweed was less. The addition of enzymes or pasteurization had no effect.

Henderson *et al*. (2012) performed a study into the effects of the use of duckweed as a coproduct in anaerobic digestion of dairy manure. A substantial increase of 1.4±0.2 to 1.9±0.1 times greater methane production was observed from co-digestion 2% dry mass duckweed with dairy manure at 35°C, yielding an additional 0.08 ±0.01 ml methane/ml waste/day. Also potential relationships between macro- and micronutrient concentrations in duckweed tissues and methane production were examined. Variations in these nutrient concentrations could account for up to 85% of the observed variability.

Cu *et al*. (2015) assessed the biochemical methane potential of Common Duckweed (*Spirodela polyrrhiza*) and measured a methane production of 340 l_n CH_4/kg VS.

4.3 Ethanol production

Duckweed can be an alternative starch source for ethanol production. Cheng and Stomp (2009) discussed the potential use of duckweed biomass for the production of ethanol. The duckweed starch content can be manipulated by adjusting growth conditions (e.g. pH, nutrient content, etc.) that affect proliferation. They argue that *Spirodela polyrrhiza* has shown a great potential for starch production. In a laboratory test they transferred *Spirodela polyrrhiza* from a nutrient-rich solution to tap water for 5 days and reached a starch content of 45.8% (dry based). Enzymatic hydrolysis of the duckweed yielded a hydrolysate of 509 mg reducing sugars per gram of dry duckweed. Fermentation of this hydrolysate using yeast gave an ethanol yield of 258 mg per gram of dry duckweed which indicates that duckweed can be readily fermented into ethanol in appreciable amounts.

Xu *et al*. (2011) evaluated duckweed (*Spirodela polyrrhiza)* from a pond at a commercial swine farm treating anaerobically digested swine wastewater as a source for the production of ethanol. The biomass production rate of the duckweed in the pond was 12.4 g dry biomass/m^2/day. By transferring the harvested duckweed to tanks with well water the starch content of the duckweed increased by 64.9% and had a final starch content of 31%. After enzymatic hydrolysis and yeast fermentation, the overall starch conversion rate of 94.7% was achieved.

Ge *et al*. (2012) investigated the growth and starch accumulation of duckweed (*Lemna minor*) grown in a swine wastewater lagoon and explored to subsequent processing of the duckweed into ethanol. The growth rate was 3.5 g/m^2/day. The accumulation of starch (10-36% w/w) was triggered by nutrient starvation of growing in dark condition with the addition of glucose. Enzyms could release up to 96.2% (w/w) of glucose. The fermenting by two yeast strains of the hydrolysates generated a high ethanol yield of 0.485 g/g (glucose).

4.4 Ingredients

ABC Kroos (www.abc-kroos.nl) processes duckweed for the production of protein, cellulose for use in fibres and colouring agents (chlorophyll, carotene and xanthophyll). The waste material after processing can be used for biogas production (Anonymous, 2012). A patented process is being used to isolate the protein from the duckweed while containing the important functional aspects of the protein for the food industry.

4.5 Fuel production

Baliban *et al*. (2013) have made a optimization-based framework for the process synthesis and simultaneous heat, power, and water integration of a duckweed to gasoline, diesel, and kerosene refinery. Through the development of a model all of the units of the refinery were optimized. Four case studies were used to demonstrate the capability of the process synthesis framework and determine the process design that had the lowest overall cost. Depending on the case study the price of crude oil for which duckweed BTL refinery was competitive was between $69 and $105 per barrel.

Xu and Deshusses (2015) demonstrated the proof-of-concept of conversion of duckweed (*Spirodela polyrrhiza)* to biohydrogen. Duckweed was harvested from a swine wastewater treatment system and subjected to different pretreatments followed by fermentation. The mild acidic thermal pretreatment (1% H_2SO_4 and 85°C for 1 h) showed to be more effective than either thermal or alkaline thermal pretreatments. After a 7-day fermentation period of the acid-pretreated duckweed, the biohydrogen production was up to 75 ml H_2 per gram dry biomass.

5 Discussion

A disadvantage in growing duckweed on effluents from animal manure is the fact that the effluent needs to be rather clean or has to be diluted for optimal growth. Leeflang (2008) argues that water pennywort (*Hydrocotyle ranunculoides*) is better at purifying waste water than duckweed, since it needs 'severely' organic polluted water in order to grow and has also a high protein content just as duckweed which makes it suitable as a feed source and replacement for soy. Furthermore is stated that water pennywort isn't bothered by wind influences and that it is easier to harvest than duckweed. Also other water plants could be an alternative. Reddy and De Busk (1985) made an evaluation of different types of aquatic plants (water hyacinth, water lettuce, pennywort, duckweed and egeria) into removing N and P from simulated wastewater. The results showed that depending on the circumstances different aquatic plants gave the highest removal rates. It was also shown that other water plants can outperform duckweed and therefore might be better suited for purifying high strength effluents from manure treatment systems than duckweed. A hybrid system could also be a possibility. For this hybrid system two possibilities exists: 1) a water pond filled with for example pennywort to lower the initial mineral concentrations in the pond water followed by a second water pond with duckweed for polishing, and 2) a water pond with a mix of water plants, for example water hyacinth, and duckweed, but the question is whether or not the species could successfully grow together in one pond.

According to the results of the research by Mohedano *et al*. (2012), next to the nitrogen removal by the duckweed, a duckweed pond could also remove nitrogen by converting it into nitrogen gas by nitrification and denitrification. They evaluated a full-scale swine waste treatment system that consisted of an anaerobic digester, a storage pond and two duckweed ponds in series. A nitrogen mass balance was calculated where total nitrogen removal was obtained by the sum of duckweed biomass removal, ammonia volatilization, nitrogen sedimentation and nitrification-denitrification process. According to the authors, the ammonia volatilization and sedimentation were negligible due the low pH levels and lack of sludge formation. Therefore they concluded, based on the yield and analysis of the duckweed, that 28% of the nitrogen removal in the first duckweed pond was due to the biomass absorption of the duckweed and 72% of the nitrogen removal was performed by nitrification and denitrification. In the second duckweed pond the nitrogen removal was 96% performed by duckweed and 4% by denitrification.

6 Conclusions and recommendations

6.1 Conclusions

The results of the literature survey show that duckweed can be used to recuperate nutrients from manure effluents and that the duckweed can be utilized as a source of feed, energy and ingredients.

The information in the literature about varieties and cultivation of duckweed grown on (diluted) manure effluents shows:

- Manure effluents have a too high concentration of nutrients to be used directly as a growth medium for duckweed. Therefore the manure effluents have to be diluted.
- In general the optimum level of nitrogen seems to be around 30 mg N/l, nevertheless maximum biomass yields have also been observed at levels of 72 mg N/l. The optimum level of nitrogen in the pond water is influenced by species and growing conditions (pH, temperature, etc.).
- Higher N content in the growth medium gives higher protein content, but there is an optimum level. The optimum level appears to be depended on the NH_4-N content, temperature and pH (influencing the toxicity of ammonia).
- The biomass yield and composition of the duckweed is depended on species, growing conditions and management of the duckweed pond.
- The root length of the duckweed is inversely related with the crude protein content of the duckweed.

The information in the survey about harvesting methods of duckweed shows:

- A couple of companies have developed a machine for harvesting of duckweed, but these were developed for waterways and not specifically for water ponds.
- On the basis of one publication it appears that harvesting less duckweed but more frequently favoured nutrient recovery and biomass production.

The information in the literature about utilization and valorisation of duckweed grown on (diluted) manure effluents shows:

- Duckweed can be used as a protein source for animal feed.
- The specific biogas production of duckweed is around 300 l CH_4/kg VS.
- By adjusting the growing conditions of duckweed the starch content of duckweed can be increased which could make the duckweed a suitable source of ethanol production.
- Duckweed can be used as a source of ingredients such as protein, cellulose and colouring agents.
- Some papers showed that duckweed could be converted into a fuel source.

6.2 Recommendations

No papers were found on the results of a year-round production of duckweed in a pond with manure effluents as the source of nutrients for growing the duckweed. A study of a year-round system will give better insight into the yields, composition during the year and possibilities of growing duckweed for puryfing manure effluents under North-West European conditions.

Research and development into the management of duckweed grown on manure effluents is necessary in order to optimize the production of duckweed and make it more feasible. Aspect such as method of supplying the manure effluent (e.g. times and amount per day), harvesting regime (e.g. amount and frequency), control of pests and diseases and also issues regarding feed safety needs further investigation.

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