

A Review on the Role of Duckweed in Nutrient Reclamation and as a Source of Animal Feed

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ABSTRACT : The family of Lemnaceae colloquially known as duckweed contains the world's smallest species of flowering plants (macrophytes). Aquatic and free-floating, their most striking qualities are a capacity for explosive reproduction and an almost complete lack of fibrous material. They are widely used for reducing chemical loading in facultative sewage lagoons, but their greatest potential lies in their ability to produce large quantities of protein rich biomass, suitable for feeding to a wide range of animals, including fish, poultry and cattle. Despite these qualities 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. 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. (*Asian-Aust. J. Anim. Sci.* 2003. Vol 16, No. 2 : 297-305)

Key Words : Lemnaceae, Nutrient Uptake, Animal Feeding, Waste Treatment, Growth, Effluent

INTRODUCTION

Excreta is a normal, biological by-product of human and animal metabolism. However, western culture dictates that excreta is "dirty", and traditionally effluent has been "sent away" in pipes, barges, trucks and pits. As human populations have grown, and animal production intensified, this approach has caused two problems.

Firstly, society now produces such large amounts of effluent that in some parts of the world, riparian land and waterways are close to collapse under the massive nutrient load produced by both urban settlement and agriculture. Secondly, and at an increasing cost, massive quantities of nitrogen, phosphorous and clean water are lost in effluent.

Waste discharge from intensive animal operations is increasingly becoming the focus of public concern, and consequently the concern of legislators (Williams, 1995). Current methods of dealing with this "problem", such as anaerobic fermentation ponds, have received limited acceptance because of the disagreeable odours they emit. As an example, dairy farmers in Texas have been charged with polluting the water table due to the leaching of nitrate, ammonia and faecal coliforms from anaerobic effluent ponds (Ivins, 2001)

Anaerobic effluent ponds are known to be wasteful. It has been estimated that a dairy cow weighing 450 kg excretes the equivalent of 59 kg of nitrogen and 9 kg of phosphorus every year (Culley and Epps, 1973). Retrieval of 25% of these elements back into the production system could provide the equivalent of almost one ton of crude protein per year in a hundred-cow dairy.

Plants of the family Lemnaceae, known colloquially as

duckweed, have been used in Asian primary production systems for hundreds of years to produce animal feed (Leng, 1999). These small aquatic plants have also been identified by American researchers as being ideally suited to nutrient reclamation, and water cleansing (Culley and Epps, 1973; Hillman and Culley, 1978). What has followed is a substantial amount of inquiry into each of these three areas, using various duckweed species. Unfortunately, the vast majority of the research in the area has been fragmentary, looking at only one or two of the aspects of duckweed growth, physiology or nutritive value. Perhaps because of this, the potential for duckweed to play a major role in water cleansing and nutrient reclamation, whilst providing a source of animal feed, remains largely just potential.

This paper reviews current knowledge arising from a number of disciplines and endeavours to develop a framework incorporating the influences of species, climate, and environment, on growth and nutrient value in duckweed. The challenge for the future is to bring this fragmentary information together and bring to fruition the role that duckweed can have in water cleansing, nutrient recycling and animal feeding.

PROPERTIES OF DUCKWEED

The family Lemnaceae consists of two sub-families (Lemnoidea and Wolffioideae), with four genera (*Spirodella*, *Lemna*, *Wolffia* and *Wolffina*), encompassing at least 34 species (Landolt, 1986). All plants are tiny (0.4 to 15 mm) and identification is therefore difficult (Leng, 1999).

Duckweeds are monocotyledonous, floating plants, and are the world's smallest and simplest flowering plants (Hillman and Culley, 1978). Each plant consists of little more than two, poorly differentiated fronds, a combination

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of leaf and stem. The tissue is composed principally of chlorenchymatous cells, separated by large intercellular spaces that provide buoyancy. The upper epidermis is cutinized and sheds water. In *Lemna* and *Spirodella* the roots are believed to be adventitious, are only a small proportion of overall plant weight and lack root hairs. The other two genera lack roots. An important feature of their structure is the almost total lack of woody tissue.

Members of the *Lemnaceae* family are found almost world wide, being absent only in the Polar Regions and deserts. Distribution of species is however, far from uniform with the Americas having over 60% of recorded species, and Australia and Europe each having less than 30% of the total. Species recorded in Australia comprise *Spirodella polyrrhiza*; *S. punctata*; *Lemna disperma*; *L. trisulca*; *L. aequinoctialis*; *Wolffia australiana*; *W. angusta* (Landolt, 1986).

The habitat requirements of duckweed vary between species, but all share the need for sheltered still water. Depth of the plant mat is an important limitation to growth. A striking feature of duckweed species is their enormous reproductive capacity. Under favourable conditions they have been reported as doubling their biomass every 16 to 48 hours (Leng, 1999). The main form of reproduction is vegetative, through the production of "daughter" fronds that arise from one of two lateral pouches at the base of the frond. Whilst vegetative growth is usual, duckweed daughter fronds do not stay attached indefinitely, but rather break and form new colonies, only a few generations old. This novel facility has led to the suggestion that duckweed growth could be considered analogous to microbial growth (Hillman, 1961).

Individual fronds have a relatively short life span of 3 to 10 weeks when in the vegetative phase, depending on species, reproductive rate and photoperiod (Landolt, 1986). By this time, an original "mother" plant may have given rise to a clonal colony of tens of thousands of individual plants over more than 50 generations. There appears to be distinctive differences in longevity and mature size between generations (Landolt, 1986) that may be expressed as cyclicity in the growth pattern of a colony.

One of the significant attributes of duckweed is its ability to be used as a source of proteinaceous food with a favourable profile of important amino acids (Rusoff et al., 1980) (Tables 1 and 2).

GROWTH CONDITIONS FOR DUCKWEED

The growth of *lemnaceae* may be nearly exponential, if carbon dioxide, light and nutrient supplies are satisfactory. Discussion in this review is limited to the three major plant macronutrients (nitrogen, phosphorus, potassium). Calcium and sulphur are not generally considered to be

Table 1. Proximate analysis of duckweeds (as percent of dry matter)

Species	Dry Matter	Crude Protein (N×6.25)	Fat Ether Extract	Crude Fibre	Ash
<i>L. gibba</i>	4.6	25.2	4.7	9.4	14.1
<i>S. punctata</i>	5.2	28.7	5.5	9.2	13.7
<i>S. polyrrhiza</i>	5.1	29.1	4.5	8.8	15.2
<i>W. columbiana</i>	4.8	36.5	6.6	11.0	17.1

Rusoff et al. (1980)

Table 2. Amino acid composition of several duckweed species (g/100 g)

Amino acid	<i>L. gibba</i>	<i>S. polyrrhiza</i>	<i>S. punctata</i>	<i>W. columbiana</i>
Alanine	4.59	4.48	4.79	3.75
Arginine	4.29	5.25	4.86	3.78
Aspartic	7.12	7.55	7.38	5.63
Glutamic	7.60	8.00	7.69	5.76
Glycine	3.79	3.95	3.93	3.04
Histadine	1.89	2.15	1.90	1.18
Isoleucine	3.87	3.75	3.76	3.06
Leucine	7.15	6.85	6.88	5.83
Lysine	4.13	4.30	4.26	3.37
Methionine	0.83	0.83	1.07	0.87
Phenylalanine	4.45	4.20	4.38	3.60
Proline	2.93	3.28	2.95	2.41
Serine	2.61	2.80	2.83	2.28
Threonine	3.20	3.45	3.31	2.55
Tyrosine	2.91	3.05	3.14	2.17
Valine	4.96	4.40	4.71	3.49

(Rusoff et al., 1980)

limiting to growth (Landolt, 1986), whereas nitrogen and phosphorus influence growth strongly and have an interactive effect.

Lemnaceae are able to absorb nitrogen as ammonium, nitrate, nitrite, urea and some amino acids, however the first two represent the main nitrogen source for most species. Minimum, optimal, and toxic levels of nitrogen vary greatly between species and geographic isolates and increasing light intensity is thought to elevate optimal nitrogen requirements for growth. Of the species studied, *L. miniscula* has the lowest (0.0016 mM/l) and an unclassified species of *Lemna* the highest (0.08 mM/l) minimum requirement for nitrogen (Landolt, 1986). Similarly, the maximum tolerated level of nitrogen varies from 30 mM/l (*L. miniscula*) to 450 mM/l for *L. aequinoctialis* (Landolt, 1986). The optimal recorded nitrogen requirement ranges from 0.01 mM/l for *W. colombiana*, up to 30 mM/l for *S. polyrrhiza* (Landolt, 1986).

Duckweed's requirement for phosphorus, is variable (0.003-1.75 mM/l) between species as is seen for nitrogen requirement, but appears unrelated to it (Landolt, 1986). Duckweed is reputedly able to accumulate up to 1.5% of its weight as phosphorus in nutrient rich waters (Leng, 1999). Between species differences are also evident for potassium, with requirements also being influenced by light intensity.

The pH of water exerts a profound effect on the growth of duckweed, influencing the uptake of nutrients and especially the nitrate:ammonium ratio and species availability (Caicedo et al., 2000). Both optimal and limiting values for growth vary widely, both between species and between colonies or isolates. Optimal growth occurs around neutral pH for both *Spirodella* and *Lemna* species, (Leng, 1999) and at around pH 5 for *Wolffia* (McLay, 1976). The lower limit of pH for growth in most species is disputed with Landolt (1986) suggesting a pH of 3 and Leng (1999) a pH of 5. The upper pH limit may be as high as 10.5. Landolt (1986) has also suggested that the presence of chelating agents have a major impact on the pH range at which these plants will grow.

The growth rate of duckweed is the result of many temperature dependent and interrelated reactions, with photosynthesis and respiration not having the same temperature curves. Most species however, appear to exhibit optimum growth between 20°C to 30°C (Landolt, 1986). The effect of light is positively reinforced by increases in temperature from 12°C up to 30°C at least, and most species maximise growth at around 9,000 lux (at 24°C) (Landolt, 1986, sourced from Docauer (1983)).

FACTORS AFFECTING GROWTH AND COMPOSITION OF DUCKWEED.

There is a great deal of literature published on actual and potential yields of duckweed (Culley and Epps, 1973; Hillman and Culley, 1978; Rusoff et al., 1980; Oran et al., 1987; Leng, 1999; Chowdhury et al., 2000). Unfortunately, there is little data available that records the interactions between genotype and environment. Many trials are based on short-term yields in small containers, with theoretical yields extrapolated to a per hectare per annum basis. Perhaps because of this, reported yields of duckweed vary widely. A summary of reported yields assembled by Leng (1999) show yields ranging from 2 to 183 t(DM)/ha/year. The extremely large range of recorded yields suggests that making estimates of productivity based on results from short trials in laboratory-scale vessels is of questionable value.

Significant variances in growth have been demonstrated between species and different geographic isolates of the same species (Bergman et al., 2000). A composite picture of yields of *L. gubba* on different media is shown in Figure 1.

These published results on actual and potential yield of duckweed indicate a general lack of agreement on the growth of these plants. There are a number of factors that may mediate these apparently conflicting results. Quite apart from procedural differences (such as different tank sizes, flow rate/retention times) there are numerous

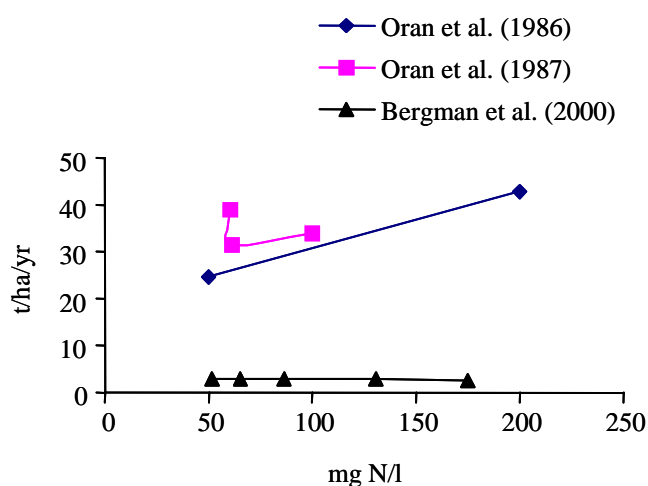


Figure 1. Extrapolated Yields of *L. gubba* grown on different [N]

physico-chemical differences that make establishment of equivalence, and thereby direct comparison difficult. Time of year (and hence ambient temperature and day length), latitude, and pH of growth media can all have a substantial influence on the physiology, and thus the growth of the plant.

There are many factors that influence growth, and the value of drawing comparisons between trials conducted without similar protocols and isolates, is also of limited value. Additionally, the levels of available nutrient, as well as species differences, can strongly influence both the quantity and quality of material produced. These differences may be interpreted in light of the existence of deficient, optimal and toxic levels for nutrients. Nitrogen in particular, whilst being an essential macronutrient, is toxic at high concentrations. Little interest has been shown in recent times in establishing an optimum nutrient range for growth of duckweed despite inconsistencies in published literature. Recent work (Bergman et al., 2000; Al-Nozaily, 2001) indicates that best growth is achieved where total nitrogen concentrations range from 10 to 40 mg N/l. However this conflicts with the work of Caicedo et al. (2000), who reported that growth rates of *S. polyrhiza* actually declined over a range of 3.5 to 100 mg N/l.

It has been demonstrated that lower (6 to 7) pH levels ameliorate the toxic effects of nitrogen (McLay, 1976; Caicedo et al., 2000) and Al-Nozaily (2000) has suggested that this may be because the low pH limits ionization of ammonia species, resulting in a low proportion of ammonia in solution.

The optimal nutrient profile for growth of duckweed doesn't necessarily produce the best quality of plant material in terms of protein content and digestibility. Leng (1999) has suggested that optimal protein content will be obtained where nitrogen is present at 60 mg N/l or greater.

Early field observations by Culley and Epps (1973) suggested that a strong positive relationship existed between high levels of dissolved nutrients and plant characteristics, especially protein and digestibility. Subsequently, several other researchers have reported positive relationships between nutrient concentrations and dry matter yield, crude protein and phosphorous content (Whitehead et al., 1987; Alaerts et al., 1996). In contrast, Bergman et al., (2000) found little difference in dry matter (DM) yield and no difference in protein content in *L. gibba* grown over a wide range of nutrient levels (52 to 176 mg N/l).

In practice, the depth of water required to grow duckweed will be determined by the purpose for which it is being grown, as well as management considerations (Leng, 1999). Ponds of less than 0.5 m depth may be subject to large diurnal temperature fluctuations. The greater the depth, the less likely it is that plants will have full access to nutrients in the water column. Recently it has been found that surface area, rather than depth, influences nitrogen removal in a duckweed lagoon (Al-Nozaily et al., 2000).

APPLICATIONS

The ability of duckweed to sequester nitrogen and phosphorus, and in so doing "cleanse" dirty water, has been widely discussed in the literature for nearly 30 years (Culley and Epps, 1973; Hillman and Culley, 1978; Oran et al., 1986; Landolt and Kandeler, 1987; Leng, 1999). Systems utilising various species of duckweed, either alone, or in combination with other plants, have been used to treat primary and secondary effluent in the U.S.A. (Zirschky and Reed, 1988), the Middle East (Oran et al., 1985) and the Indian subcontinent (Skillicorn et al., 1993; van der Steen et al., 1998).

Notwithstanding this reputation, some species and isolates are apparently quite sensitive to high levels of nitrogen and/or phosphorous (Bergman et al., 2000), and effluent with a high biological oxygen demand (BOD), such as abattoir waste, may kill the plants. Although duckweed has a reputation for absorbing large amounts of dissolved nitrogen, the degree of absorption appears to vary with concentration of nitrogen, time, species, and (at least in temperate zones) the season. There is also strong evidence that there is a symbiotic, or at least a synergistic relationship between duckweed and bacteria, both in the fixation of nitrogen (Duong and Tiedje, 1985), and the removal of Chemical Oxygen Demand (COD) (Korner et al., 1998) from water.

Differences in methodology, scale, and the parameters, both recorded and measured, make direct comparisons between the many trials in published literature difficult. However most research indicates that duckweed removes

40 to 60% of nitrogen in solution over a 12 to 24 day period. Volatilization may account for a similar loss of nitrogen (Vermaat and Haniff, 1998), although recent work completed in Israel (Van der Steen et al., 1998), has suggested that direct duckweed absorption may account for less than 20% of nitrogen loss, and volatilization/denitrification may account for over 70%.

In a similar fashion, *lemnaceae* are generally able to absorb 30 to 50% of dissolved phosphorous, although one researcher (Alaerts et al., 1996) has claimed over 90% removal in a working, full scale system. Phosphorous uptake (as measured by tissue phosphorous) and crude protein, increased linearly with increases in nutrient concentration, up to approximately 1.5 g P/l, and increased in absolute terms, up to 2.1 g P/l (Sutton and Ornes, 1975). This was recorded in conjunction with a proportional rise in nitrogen concentration, thus the association between nitrogen and phosphorous concentrations was unclear.

COD is a measure that quantifies water quality as determined by dissolved oxygen. All research in the use of duckweed for improving effluent quality has determined significant but variable decreases in COD (Alaerts et al., 1996; Karpiscak et al., 1996; Bonomo et al., 1997; Vermaat and Haniff, 1998; van der Steen et al., 1999). However, a substantial decrease in COD would be expected in open ponds without the presence of duckweed (Al-Nozaily et al., 2000), so this improvement may not be attributable to the actions of duckweed.

Simplistically, the duckweed's environment is somewhat two-dimensional. In practice, this means that once the surface of a body of water is completely covered, the plant has limited further opportunities to grow. Thus, in situations where there are high nutrient levels, the clearance of dissolved nutrients is likely to be limited by harvesting rate. The work of Whitehead et al. (1987) confirms that at high average nutrient levels (short retention time), nitrogen and phosphorous removal is enhanced with increased cropping rate, whereas low nutrient concentrations favour low cropping rates. This latter state indicates that growth is limited by nutrient availability.

Degradation of bacterial pathogens is a complex process and a comprehensive discussion is beyond the scope of the current paper. However, two groups conducting specific investigations into this issue (Karpiscak et al., 1996; van der Steen et al., 1999) found that faecal coliforms decreased by 50 to 90% and that *Giardia* and *Cryptosporidium* fell by over 80% in eutrophic waters in which duckweed was grown.

ANIMAL NUTRITION TRIALS

As for many characteristics of the family, the reported energy value(s) of duckweed(s) vary significantly, ranging from 9.6 to 17.6 MJ/kg dry matter, (Landolt and Kandeler,

1987), indicating a moderate to high energy value, (on a dry matter basis). As discussed earlier, protein content and amino acid profiles vary, depending on nutrient media, species, and probably isolate or clone. Incident light and temperature affect growth rate, and thereby carbohydrate metabolism and composition. Most essential amino acids are available in abundance except for tryptophane, of which there are only traces, and methionine, which is present in small, but variable amounts (0.3 to 3% of total protein) (Rusoff et al., 1980). *Spirodella* and *Lemna* genera contain large amounts of oxalic acid, which may result in a disagreeable taste (Landolt and Kandeler, 1987)

An investigation into the protein quality of a number of aquatic plants, found that protein extracted from *L. minor* increased voluntary intake and weight gains in rats fed a diet high in wheat flour (Dewanji and Matai, 1996). The study also reported a phenolic content of 2.1% in the duckweed protein extract. A trial examining utilization of plant biomass proteins found that *L. minor* was well accepted by rats at up to 25% of total dietary intake (Phuc et al., 2001). Duckweed produced the best weight gains of the plants studied, although less than the control diet (12.2 g/day vs 15.2 g/day). Yet at 50% inclusion, appetite was suppressed, although the animals continued to gain weight, albeit at a lesser rate (10.8 g/day). The authors concluded that unspecified anti-nutritional factors arising from the plant's high mineral content were likely to be inhibiting digestion and metabolism.

Fishmeal is the major source of protein for farmed fish worldwide and is in limited supply (Refstie and Storebakken, 2001). With a high protein content and fairly full spectrum of amino acids, duckweed would appear to have the capacity to replace expensive fish and soya bean meal in aquaculture diet formulations. Sewage-grown lemnae has formed the basis for a large, ongoing commercial operation at Mirzapur in Bangladesh, and has yielded impressive results over several years (Skillicorn et al., 1993). However, the results of formal research trials have been more equivocal. Hassan and Edwards (1992) reported a linear increase in weight gain and improvements in food conversion efficiencies (FCE) into body weight gain, when they included *L. perpusilla* and *S. polyrrhiza* at up to 30 g DM/kg in the diet of Nile tilapia (*Oreochromis niloticus*). At higher levels, weight gain decreased, FCEs decreased, and there was significant increase in mortalities. Similarly, a recent study examining the culture of tilapia fed varying levels of *S. polyrrhiza* found that weight gain and food conversion ratios were unchanged from the control group at inclusion rates up to 30%, but that above this level growth rates were impaired (Fasakin et al., 1999).

A phenomenon worthy of note is that Hassan and Edwards (1992) used duckweed of relatively poor quality (23% CP), and cited mortality rates in excess of 80% at the

highest feeding rates. In contrast a more recent study (Fasakin et al., 1999) used better quality material (cited 50% CP) and recorded no significant increase in mortality, compared to the control group, even with 100% substitution. Refstie and Storebakken (2001) have cited the sensitivity of fish to such factors as non-starch polysaccharides and phytates, and it may be, that as the protein component of plant material decreases, the exposure per unit DM to these anti-nutritional elements in the feed increase and thus elicit negative growth responses.

Certain amino acids are generally considered limiting in poultry nutrition (Nakaue and Arscott, 1991) and several studies have examined the effect of inclusion of duckweed in poultry diets, primarily as a substitute for protein meals (Haustein et al., 1990, 1992; O'Neill et al., 1996). Although there were significant differences between the control and treatment groups at high levels of substitution, the results of both studies by Haustein et al. (1992, 1995) demonstrated that *L. gibba* could replace more conventional protein sources at up to 15% total intake, maintain production characteristics and body condition, while improving protein content of the egg. This agreed closely with O'Neill et al. (1996) who found that feed intakes and production characteristics were unaffected by including up to 13% duckweed (*S. punctata*) in the diet of laying hens. Improvements in yolk pigmentation from the addition of duckweed have also been claimed (O'Neill et al., 1996). Men et al. (2001), has reported that substituting *L. minor ad libitum* for between 40 to 100% of soya bean meal and vitamin mineral pre-mixes in a diet fed to meat ducks maintained all carcass yield and quality characteristics.

The same positive effects cannot be claimed for feeding to juveniles. In a study examining the phenomena of compensatory growth in chicks, Haustein et al. (1992) found that both feed intake and growth rate was depressed (at all levels in one trial, and at amounts above 10% in a second). The degree of depression of intake was found to be in direct proportion to the rate of inclusion of *L. gibba* in the diet. It was suggested that duckweed might act in a similar manner to lucerne (*Medicago sativa*), although no evidence was advanced for this claim. Similarly, Leng (1999) has reported that feeding low levels (5 to 10%) of a comparatively poor quality (23%CP) duckweed led to pronounced decreases in the rate of weight gain in young pigs, although a second study (Leng, 1999) using low levels of supplementation combined with soya bean meal (SBM) tended to show no differences in the growth rate, compared to pigs fed an isonitrogenous SBM only diet.

The composition of ingested food is changed substantially by ruminal fermentation, and a system to recover nutrients (especially nitrogen and phosphorous) from cattle's manure using duckweed to reintroduce those nutrients back into the food chain, was propounded

over two decades ago (Hillman and Culley, 1978). That there has been little investigation into using these aquatic plants to feed ruminants may be due in part, to the large amount of material needed (Leng, 1999). Huque et al. (1996) has concluded that the daily intake of duckweed is well accepted by cattle at up to 10% of their live-weight. Further it was found that the feed was highly digestible, and the protein was highly ruminally degradable. Recent studies on fine wool Merino sheep have measured the effect of these different nitrogenous substrates on wool growth characteristics in Merino sheep, and found that duckweed compares favourably to other concentrates (cottonseed meal and urea) (Damry et al., 2001). These studies also demonstrated that duckweed was well accepted in both dried and fresh forms, had no negative effects on clean wool yield or wool fibre diameter and was superior to urea, and similar in quality to cottonseed meal. The authors suggested, on the basis of higher wool growth rates and lower ruminal ammonia concentrations post-feeding, that duckweed might prove to be a good source of rumen undegraded protein. This appears however, to be in conflict with some of the findings of Huque et al. (1996).

CONSTRAINTS AND CONTRAINDICATIONS

The ability of lemnae to selectively take up nitrogen and phosphorous, and concentrate them in its tissues is not limited to these macronutrients. Estimates of take up rates for a selection of elements, based on studies in a number of wastewater facilities have been published (Zirschky and Reed, 1988) (Table 3).

Rates of accumulation of heavy metals differ markedly between species, as does the relationship between concentration (in water) and take up - in some cases it is direct, in others reciprocal (Landolt, 1986). *L. minor* has been critically examined for its capacity to take up heavy metals, and has been found to be an efficient accumulator of cadmium in particular, but also of selenium and copper (Zayed et al., 1998).

The ability to concentrate trace elements gives duckweed the potential for use in bioremediation at mine sites and other contaminated areas. On the other hand, if used in a system for nutrient recovery in an animal production operation, care would need to be taken in isolate selection and/or feed analysis and/or animal sampling, to prevent the accretion of toxic levels of heavy metals within the system.

It is beyond the scope of this report to enter into a detailed analysis of the economics of duckweed production. However, aside from the initial infrastructure (i.e. pond earthworks), harvesting and drying are likely to be the only significant costs incurred, labour or otherwise. In Asia, where unit labour costs are significantly lower than in most

Table 3. Constituent uptake rates for lemnae

Constituent	Uptake rate (kg/ha/yr)	Constituent	Uptake rate (kg/ha/yr)
Aluminium	2,600	Manganese	90
Arsenic	6	Mercury	1
Calcium	6,000	Nitrogen	6,110
Chloride	1,000	Phosphorous	800
Chromium	6	Potassium	2,520
Copper	1	Sodium	390
Iron	800	Sulphur	650
Magnesium	800	Zinc	7

Adapted from: Zirschky and Reed (1988).

industrialised countries, manual harvesting with sieves is the norm (Leng, 1999). In Australia, it seems problematical to suggest that this could be undertaken in a profitable manner, without some degree of mechanisation. Automated harvesting systems do exist (Bell, 2001), and are operating in Australia on a limited basis, but neither their cost, nor performance has been reviewed.

Duckweed is between 92% and 96% water when harvested. Drying, especially to levels where it can be preserved, represents a major cost in terms of labour and/or energy. Whilst this may be overcome, the best solution seems to be to utilise it on site. *Lemna* species have been successfully fed in large quantities to fish (Skillicorn et al., 1993), waterfowl (Men et al., 2001), cattle (Huque et al., 1996) and sheep (Damry et al., 2001).

Although not pronounced in the tropics and sub-tropics, some species growing in temperate zones exhibit markedly seasonal growth patterns, based probably on fluctuations in temperature, light intensity and duration of sunlight (daylength) (Landolt, 1986). In high latitude environments, metabolic activity may cease altogether (Bonomo et al., 1997). Inability of a system to cope with a continuous supply of waste, may be a major impediment to the adoption of such technology in distinctly seasonal areas. Once again, the most positive option may be to select for cultivars that remain metabolically active at low temperatures.

Contamination of the food chain by engaging in "cannibalistic" animal production practices, has become an abiding concern of the public, health authorities, producers, and processors, since the outbreak of Bovine Spongiform Encephalopathy in the 1990s. Thus, the question of the possibility of introducing and or concentrating a prion-like agent or some other pathogen, needs to be addressed if this technology is to be widely adopted.

Plants do not absorb cellular organisms or large Dalton molecules, but duckweed has been noted as taking up short-chain peptides, so in theory, there may be some chance of transfer of sections of DNA, although this has not been previously raised in the literature. A far greater source of potential contamination is from the biofilm that clings to the

harvested plants. It appears that bacteriological activity on the surface of duckweed makes a critical contribution to both removal of COD (Korner et al., 1998) and nitrogen fixation (Duong and Tiedje, 1985). As might be expected, faecal coliforms, *Streptococcae*, *Salmonella* and *Shigella* may be found in abundance in waters constantly charged with faecal waste (Landolt and Kandeler, 1987).

It appears that duckweed plays a significant, though undefined role in reduction of microbiological contamination of wastewater (Karpiscak et al., 1996; van der Steen et al., 1999). In view of the interest in the microbiological status of wastewater, it was surprising that apparently no interest has been shown in determining whether or to what degree, pathological organisms attach to the plant itself. Similarly, there has been little interest in determining the transference of viable pathogens to animals in feeding trials, although what has been reported is positive. For example, in a trial examining laying performance in chickens, Haustein and co-workers (1980) took rectal swabs from chickens fed on sewage grown lemnae, and tested for the presence of *Vibrio*, *Aeromonas sp.*, *Campylobacter jejuni*, *Shigella* and *Salmonella*. They found no significant differences in the microbial populations of chickens fed sewerage-grown lemnae and the chickens fed commercial diets.

If duckweed, grown on wastewater, is to be considered as an animal feeding supplement in any meaningful way, investigation into the transference of pathogens will have to be completed and shown to be safe to contemporary community standards.

CONCLUSION

The family lemnae are a unique group of plants, with many striking features of great biological potential. As a group, they have an extremely small amount of structural tissue, and a mode of reproduction that allows them to increase their biomass at a much faster rate than any other macrophyte. Various species are distributed over most of the earth's surface, yet because of their unique ecological requirements, they rarely dominate their environment, and then only in response to unusual and generally man made environmental conditions.

Duckweed is able to accrete substantial amounts of macronutrients, and indeed its rate of growth is highly dependent on the appropriate amounts of these elements, along with a suitable pH, light and temperature regime. The mix of conditions that will promote growth is also highly dependent on species and even strain, and these differences have not been fully elucidated, especially under large scale or field conditions.

Various species are widely used in treating wastewater from sewerage plants, although the extent of their

contribution remains in some doubt. This is, in no small part, due to large differences in the conditions under which they are examined, as well as poorly understood and unreported species differences. It may be that their role in removing nitrogenous elements from water is subordinate to atmospheric volatilisation and cyanobacterial fixation, especially at high nutrient concentrations, but this is not certain. However, it is clear that a well-maintained cover of duckweed effectively suppresses algal growth, and its capacity for uptake of phosphorous has been demonstrated. As well as a reduction in mineral content, water so treated, has demonstrably fewer pathological bacteria and protozoa, although these are not totally eliminated.

The potential of these plants has been recognised by industrialised nations for some time, and limited animal feeding trials have been performed, the results giving some cause for optimism. The results from experimental feeding of a fairly wide range of monogastric species indicate that at low to moderate levels duckweed is well assimilated. It could provide a sustainable and cost effective alternative to expensive and potentially limited sources of high quality protein meals, such as fishmeal and soya bean meal. There is evidence however, that there is some type of anti-nutritional factor at play that limits intake and growth when fed at higher levels. Candidates may include organoleptic inhibitory factors such as oxalate, or compounds that interfere with digestion or metabolism such as phenolic compounds, tannins or saponins. Despite this being a well-documented problem, apparently no research has been undertaken to elucidate the mechanism(s) responsible.

Feeding trials involving ruminants are much less common, but show great promise. Untreated (wet) duckweed has been fed at high levels, and has been well accepted without any reported negative effects. *In sacco* studies suggest high degradability of protein and non-protein constituents. Finally there may be significant amounts of protein that escape rumen degradation—a possible bonus for wool production and liveweight gain.

While some species are in common use in treatment plants, their adoption into the food production system in non-Asian countries seems distant. Several apparent reasons for this could, in large measure, be ameliorated by systematic research. The first could be said to be a lack of understanding and definition of performance differences and environmental needs of duckweed. Duckweed is essentially a wild plant, and much of the research to date has involved gathering wild populations and subjecting them to "performance trials". Under these circumstances, substantial variations in performance and frequent disappointment should almost be expected.

Accretion of heavy metals is an interesting feature of duckweed that bears further investigation, but one that could have severe human health implications, in situations

where nutrients are concentrated within a feeding system. How much of a problem this could be, is quite unappreciated. Another major source of concern, is the introduction or multiplication of pathogens, that may occur through feeding animals material grown on their own waste. The very limited research conducted 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.

Lemnaceae are indeed fascinating organisms, with the ability to perform functions outside the range of any crop currently available. However, to be useful in modern production systems work needs to be conducted to both define and refine their properties. Systematic investigation of growth and plant quality attributes in relation to environment, needs to be undertaken to establish the economic value of this crop for animal feeding. It seems doubtful whether the economic imperative exists, at present, to make this happen.

REFERENCES

- Alaerts, G. J., M. D. R. Mahbubar and Kelderman. 1996. Performance Analysis of a Full-Scale Duckweed-Covered Lagoon. *Wat. Res.* 30(4):843-52.
- Al-Nozaily, F., G. Alaerts and S. Veenstra. 2000. Performance of Duckweed-Covered Lagoons-II. Nitrogen and Phosphorous Balance and Plant Productivity. *Wat. Res.* 34(10):2734-41.
- Bell, R. E. 2001. "Bio-tech Waste Management." found at: <http://users.northnet.com.au/~bellbtwm>
- Bonomo, L., G. Pastorelli and N. Zambon. 1997. Advantages and Limitations of Duckweed-Based Wastewater Treatment Systems. *Wat. Sci. Technol.* 35(5):239-46.
- Bergman, B. A., J. Cheng, J. Classen and A. M. Stomp. 2000. In vitro selection of Duckweed Geographic Isolates for Potential Use in Swine Lagoon Effluent Renovation. *Bioresource Technol.* 73:13-20.
- Caicedo, J. R., N. P. Van Der Steen, O. Arce and H. J. Gijzen. 2000. Effect of Total Ammonia Nitrogen Concentration and pH on Growth Rates Of Duckweed (*Spirodella polyrrhiza*). *Wat. Res.* 34(15):3829-35.
- Chowdhury, S. A., N. Sultana, K. S. Huque and Q. M. E. Huque. 2000. Manure Based Production in Shallow Sink: Effect of Genera on Biomass & Nutrient Yield of Duckweed under the Same Nutritional Management Conditions. *Asian-Aus. J. Anim. Sci.* 13(5):686-693
- Culley, D. D. and E. A. Epps. 1973. Use of Duckweeds for waste Treatment and Animal Feed. *J. Water Pol. Cont. Fed.* 45:337-47.
- Damry, J. V. Nolan, R. E. Bell and E. S. Thomson. 2001. Duckweed as a Protein Source for Fine-Wool Merino Sheep: It's Edibility and Effects on Wool Yield and Characteristics. *Asian-Aust. J. Anim. Sci.* 14(4):507-514.
- Dewanji, A. and S. Matai. 1996. Nutritional Evaluation of Leaf Protein Extracted from three Aquatic Plants. *J. Agric. Food Chem.* 44(8):2162-66.
- Duong, T. P. and J. M. Tiedje. 1985. Nitrogen Fixation by Naturally Occurring Duckweed-Cyanobacterial Associations. *Can. J. Microbiol.* 31:327-30.
- Fasakin, E. A., A. M. Balogun and B. E. Fasuru. 1999. Use of Duckweed, *Spirodella polyrrhiza* L. Schleiden, as a Protein Feedstuff in Practical Diets for Tilapia, *Oreochromis niloticus* L. *Aquaculture Res.* 30:313-18.
- Hassan, M. S. and P. Edwards. 1992. Evaluation of Duckweed (*Lemna perpusilla* and *Spirodella polyrrhiza*) as Feed for Nile Tilapia (*Oreochromis niloticus*). *Aquaculture.* 104:315-26.
- Haustein, A. T., R. H. Gilman, P. Skillicorn, V. Vergara, V. Guevera and A. Gastanaduy. 1990. Duckweed, a Useful Strategy for Feeding Chickens: Performance of Layers fed with Sewage grown Lemnaceae species. *Poult. Sci.* 69:329-35.
- Hillman, W. S. 1961. The Lemnaceae or duckweeds: A review of the descriptive and experimental literature. *Botan. Rev.* 27:221-87.
- Hillman, W. S. and D. D. Culley. 1978. The Uses of Duckweed. *Amer. Scien.* 66:442-51.
- Huque, K. S., S. A. Chowdhury and S. S. Kibria. 1996. Study on the Potentiality of Duckweeds as a Feed for Cattle. *Asian-Aus. J. Anim. Sci.* 9(2):133-137.
- Ivins, M. 2001. Home, Home on the Latrine Time Aug:21.
- Karpiscak, M. M., C. P. Gerba, P. M. Watt, K. E. Foster and J. A. Falabi. 1996. Multi-Species Systems for Wastewater Quality Improvements and Habitat Enhancement. *Wat. Sci. Technol.* 33(10-11):231-36.
- Korner, S., G. B. Lyatuu and J. E. Vermaat. 1998. The Influence of L. Gibba on the Degradation of Organic Material in Duckweed-Covered Domestic Wastewater. *Water Res.* 32 (10):3092-98.
- Landolt, E. 1986. The Family of Lemnaceae - A Monographic Study vol 1. Part of the series: Biosystematic Investigations in the family of duckweeds (Lemnaceae), Vol 2 of 4. *Veroffentlichungen Des Geobotanischen (pub.)*
- Landolt, E. and R. Kandeler eds. 1987. The Family of Lemnaceae - A Monographic Study.. Vol 2. Part of the series: Biosystematic Investigations in the family of duckweeds (Lemnaceae), vol 3 of 4. *Veroffentlichungen Des Geobotanischen (pub.)*
- Leng, R. A. 1999. Duckweed, A Tiny Aquatic Plant with Enormous Potential for Agriculture and Environment. *FAO. Tran Phu Printing Co.*
- Men, B. X., B. Ogle and J. E. Lindberg. 2001. Use of Duckweed as a protein supplement for Growing Ducks. *Asian-Aust. J. Anim. Sci.* 14(12):1741-1746.
- McLay, C. L. 1976. The Effect of pH on the Population Growth of Three Species of Duckweed: *Spirodella oligorrhiza*, *Lemna minor* and *Wolffia Arrhiza*. *Freshwater Biol.* 6:125-36.
- Nakaue, H. S. and G. H. Arscott. 1991. Feeding Poultry" In: "Livestock Feeds and Feeding (Ed. D. C. Church). Prentice Hall International 3rd ed.
- O'Neill, P. J., J. V. Nolan and E. Thomson. 1996. Duckweed as an Alternative to Soyabean Meal in Diets for High-Producing Layers. *Proc. Poultry Foundation Meeting Sydney.*
- Oran, G., L. R. Wildschut and D. Porath. 1985. Waste Water Recycling by Duckweed for Protein Production and Effluent Renovation. *Water Sci. Technol.* 17:803-817.
- Oran, G., D. Porath and L. R. Wildschut. 1986. Wastewater Treatment & Renovation by Different Duckweed Species. *J. Environ. Enginrer.* 112(2):247-262.
- Oran, G., H. Jansen and D. Porath. 1987. Performance of the

- Duckweed Species *Lemna gibba* on Municipal Wastewater for Effluent Renovation and Protein Production. *Biotech. Bioengin.* 29:258-268.
- Ouazzani, N., K. Bouhoum, L. Mandi, L. Bouarab, K. Habbari, F. Rafiq, B. Picot, J. Bontoux and J. Schwartzbrod. 1995. Wastewater Treatment by Stabilization Pond: Marakesh Experiment. *Water Sci. Technol.* 31(12):75-80.
- Phuc, B. H. N., J. E. Lindberg, B. Ogle and S. Thomke. 2001. Determination of the Nutritive Value of Tropical Biomass Products as Dietary Ingredients for Monogastrics Using Rats: 1. Comparison of Eight Forage Species at Two Levels of Inclusion in Relation to a Casein Diet. *Asian-Aust. J. Anim. Sci.* 14(7):986-93.
- Refstie, S. and T. Storebakken. 2001. Vegetable protein sources for Carnivorous fish: potential and challenges. *Recent Advances in Animal Nutrition in Australia.* 13:195-203.
- Rusoff, L. L., E. W. Blakney and D. D. Culley. 1980. Duckweeds (Lemnaceae Family): A Potential Source of Protein and Amino Acids. *J. Agric. Food Chem.* 28:848-50.
- Skillicorn, P., W. Spira and W. Journey. 1993. Duckweed Aquaculture-a New Aquatic Farming System for Developing Countries. The World Bank Washington DC.
- Sutton, D. L. and W. H. Ornes. 1975. Phosphorus removal from Static Sewage Effluent using Duckweed. *J. Environ. Qual.* 4(3):367-70.
- Van der Steen, P., A. Brenner and G. Oron. 1998. An Integrated Duckweed and Algae Pond System for Nitrogen Removal and Renovation. *Wat. Sci. Technol.* 38(1):335-343.
- Van der Steen, P., A. Brenner, J. Van Buuren and G. Oron. 1999. Post-Treatment of USAB Reactor Effluent in an Integrated Duckweed and Stabilization Pond System. *Wat. Res.* 33(3): 615-620.
- Vermaat, J. E. and M. K. Haniff. 1998. Performance of Common Duckweed Species (Lemnaceae) and the water Fern *Azolla Filiculoides* on Different Types of Waste Water. *Wat. Res.* 32(9):2569-2576.
- Whitehead, A. J., K. V. Lo and N. R. Bulley. 1987. The Effect of Hydraulic Retention Time and Duckweed Cropping Rate on Nutrient Removal from Dairy Barn Wastewater. In: *Aquatic Plants for Water Treatment and Resource Recovery* (Ed. K. R. Reddy and W. H. Smith). Magnolia Publishing Inc.
- Williams, P. E. V. 1995. Animal production and European Production Problems. *Anim. Feed Sci. Technol.* 53:135-44.
- Zayed, A., S. Gowthaman and N. Terry. 1998. Phytoaccumulation of Trace Elements by Wetland Plants. I: Duckweed. *J. Envir. Qual.* 27(3):715-721.
- Zirschky, J and S. C. Reed. 1988. The Use of Duckweed for Wastewater Treatment. *J. Water Poll. Cont. Fed* 60(7):1253-57.

