# 3. Floating aquatic macrophytes– Duckweeds

Duckweeds are small (1-15 cm) free-floating aquatic plants with worldwide distribution. They are monocotyledons belonging to the family *Lemnaceae* (which is derived from the Greek word '*Limne*', meaning pond) and are classified as higher plants or macrophytes, although they are often mistaken for algae and some taxonomists consider them as being members of the Araceae. Duckweeds serve as nutrient pumps, reduce eutrophication effects and provide oxygen from their photosynthesising activity. Duckweeds are often seen growing in thick blanket-like mats on still nutrient-rich fresh and slightly brackish waters. They do not survive in fast moving water (>0.3 m/sec) or water unsheltered from the wind. They grow at water temperatures between 6 and 33 °C (Leng, Stambolie and Bell, 1995).

#### 3.1 CLASSIFICATION

Duckweed consists of four genera: Lemna, Spirodela, Wolffia and Wolffiella. So far, 37 species belonging to the four genera have been identified from different parts of the world. Selected species are listed in Table 3.1. Taxonomically the family is complicated by clonal characteristics (Culley et al., 1981). The most commonly available species belong to the three genera Lemna, Spirodela and Wolffia. Illustrations of selected species of duckweeds are given in Figures 3.1 - 3.3. It is quite common for floating mats of duckweeds to consist of more than one species, e.g. Lemna and Wolffia.

Lemna is the largest genera of the family Lemnaceae. Lemna is among the most complex and confusing groups within the entire family. Landolt (1986) hypothesized that Lemna disperna and Lemna gibba are related as progenitor-derivative species and the former species differentiated from the latter one. Reduction of some structures such as frond size, number of nerves and the number of ovules in Lemna disperna, along with its narrower geographic distribution, support the hypothesis that it was derived from Lemna gibba or from a common ancestor. Lemna disperna has a chromosome number of 2n = 40, whereas the numbers 2n = 40, 50, 70 and 80 have been found in

TABLE 3.1

Classification of selected species of duckweeds

Lemna	Spirodela	Wolffia	Wolffiella
L. gibba	S. biperforata	W. arrhiza	W. caudate
L. disperna	S. intermedia	W. australiana	W. denticulata
L. gibba	S. oligorrhiza	W. columbiana	W. lingulata
L. japonica	S. polyrrhiza	W. microscopia	W. oblonga
L. minima	S. punctata	W. neglecta	W. rotunda
L. minor			
L. minuscula			
L. paucicostata			
L. perpusilla			
L. polyrrhiza			
L. turionifera			
L. trisulca			
L. valdiviana			

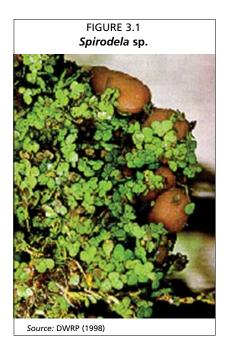
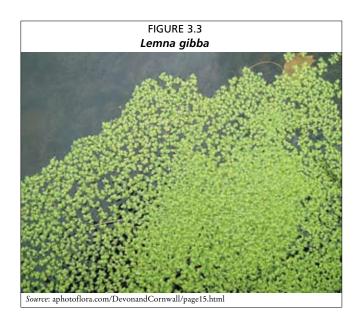


FIGURE 3.2
Common duckweed, Lemna minor grown in a pond (Phu Tho Province, Viet Nam)

Courtesy of M.G. Kibria



Lemna gibba (Crawford et al., 2005). The allozyme study supports the continued recognization of two species and is concordant with the hypothesis that the species are related as progenitor and derivative. The reduced morphology of Lemna disperna and the allozyme data indicate that this species originated via dispersal of Lemna gibba or of a common ancestor of the two species.

#### 3.2 CHARACTERISTICS

Duckweeds are adapted to a wide variety of geographic and climatic zones and are distributed throughout the world except in regions where temperature drops below 0 °C during part of the year. Most species are found in moderate climates of tropical and temperate zones. In deserts and extremely wet areas, duckweeds are rare. *Lemna* spp., for example are very rare in regions with high or very low precipitation and are not found in Greenland or the Aleutian Islands (Landolt, 2006). Although many species can survive extremes of temperature, they generally grow faster under warm and sunny conditions (Skillicorn, Spira and Journey, 1993). Most species show prolific growth in the tropics. Various microclimatic factors such as light intensity, salinity, regional temperature differences can influence the distribution of *Lemnaceae* species (Landolt, 1986). Birds and floods often disperse duckweeds to different geographic areas.

# 3.2.1 Reproduction

A duckweed plant consists of a single leaf or frond with one or more roots. Most species of duckweed multiply principally through vegetative propagation by the formation of daughter fronds from two pockets on each side of the narrow end of the frond (Gaigher and Short, 1986). Newly formed fronds remain attached to the mother frond during the initial growth phase and the plants therefore appear to consist of several fronds. Species of the genus Spirodela have the largest fronds, measuring as much as 20 mm across, while those Wolffia species are 2 mm or less in diameter. Lemna species are intermediate in size, being about 6-8 mm. An individual frond may produce as many as 20 daughter fronds during its lifetime, which lasts for a period of 10 days to several weeks. The daughter frond repeats the history of its mother frond. Some of the duckweed species, however, reproduce by producing unisexual and monoecious flowers and seeds. For example, *L. paucicostata* routinely flowers and seeds. However, the flowers are very small and rare in many species; male and female flowers are borne on the same plant. Each inflorescence generally consists of two male flowers and one female, but in Wolffia, there is one male and one female. The flowers are naked or surrounded by spathe. The fruit is a utricle and the seeds are smooth or ribbed. Vegetative reproduction is very rapid and is usually by the formation of buds of new fronds from the reproductive pouches (Guha, 1997).

Many species of duckweed survive at low temperatures by forming a special starchy 'survival' frond known as a turion. In cold weather, the turion is formed and sunk to the bottom of the pond where it remains dormant until warm water triggers resumption of normal growth. Several species survive at low temperatures without forming turions. During the winter season, the fronds are greatly reduced but remain at the surface. Occasionally, turion-like fronds will form, but the plants continue to slowly reproduce vegetatively. These plants are probably the best plants to utilize in a culture system, as restocking is virtually assured. *L. gibba*, *L. valdiviana*, *L. minor*, *L. trisulca* and *L. minuscula* are five duckweed species that frequently show some growth at cold temperatures.

## 3.2.2 Environmental requirements

A variety of environmental factors, such as water temperature, pH and nutrient concentration, control the growth and survivability of duckweeds. The other environmental factors that influence the growth rates of duckweed colonies are presence

of toxins in the water, crowding by overgrowth of the colony and competition from other plants for light and nutrients. However, the growth rate of duckweed is favoured by organic pollutants as well as inorganic nutrients (Guha, 1997). The effect of these various factors is summarised below.

## **Temperature**

Temperature tolerance and optima are dependent on species and possibly even on clones. Optimum temperature for maximum growth of most groups apparently lies between 17.5 and 30 °C (Culley et al., 1981; Gaigher and Short, 1986). Although some species can tolerate near freezing temperatures, growth rate declines at low temperature. Below 17 °C some duckweeds show a decreasing rate of growth (Culley et al., 1981). Most species seem to die if the water temperature rises above 35 °C. The effect of temperature on growth is affected by light intensity, i.e. as light increases, growth rates increase from 10 to 30 °C.

In Bangladesh, Khondker, Islam and Nahar (1993a) reported the temperature dependent growth of *S. polyrrhiza* with a maximum biomass of 76.4 g/m² recorded in the middle of February, after which the biomass depleted gradually with the rise in water temperature. The water temperature in the middle of February was about 19 °C. Islam and Khondker (1991) also obtained a high growth of *S. polyrrhiza* at a temperature of 16 °C. Furthermore, Khondker, Islam and Nahar (1993b) reported maximum growth of *S. polyrrhiza* at water temperatures of 22.2-22.5 °C in a growth study conducted in pond water. Khondker, Islam and Makhnun (1994) reported an inverse correlation between water temperature and the biomass of *L. perpusilla* when the water temperature varied between 15 and 28 °C. These authors also noted that the growth of this duckweed species ceased completely at 26 °C and above.

## pH

Duckweeds are generally considered to have a wide range of tolerance for pH. They survive well from pH 5 to 9, although some authors put their range between 3 and 10. However, pH tolerance limits of the various species differ. Stephenson *et al.* (1980) noted that duckweed display optimum growth in a medium of pH 5.0-7.0. Generally, duckweeds grow best over the pH 6.5 to 7.5 range. A doubling of biomass in 2 to 4 days has been demonstrated at pH levels between 7 and 8 (Culley *et al.*, 1981). Unionized ammonia is the preferred nitrogen substrate for duckweed. An alkaline pH shifts the ammonium-ammonia balance toward the un-ionized state and results in the liberation of free ammonia, which is toxic to duckweed at high concentrations (100 mg NH<sub>3</sub>/L).

Islam and Paul (1977) observed that *W. arrhiza* grew at a pH range of 5-10, although the optimum pH was found to be 7-8. In Bangladesh, *S. polyrrhiza* has been reported to grow best at a pH between 6.5 and 7.5 (Islam and Khondker, 1991). The range of pH for optimum growth of *S. polyrrhiza* reported in India was 6.8-8.5 (Kaul and Bakaya, 1976; Gopal and Chamanlal, 1991). Khondker, Islam and Makhnun (1994) reported the pH range of 6.9 and 7.8 to be suitable for the growth of *L. perpusilla*. Similarly, Van der Does and Klink (1991) observed pH of 7.36 in a lemnid habitat in the Netherlands supporting growth of *L. perpusilla*. A summary of minimum, maximum and optimum pH of various duckweed species is presented in Table 3.2.

## Conductivity

Electrolyte conductivity appears to have some effect on the growth of different species of duckweed. Zutshi and Vass (1973) found *L. gibba* and *L. minor* growing in stagnant waters rich in electrolyte ranging from 400-500 μS/cm. Gopal and Chamanlal (1991) reported the maximum biomass of *L. perpusilla* and *S. polyrrhiza* from roadside pools and ditches in India within a electrolyte conductivity range of 650-1 000 μS/cm. Khondker, Islam and Nahar (1993a) recorded the complete disappearance of

Duckweed species	Min	Max	Optimum	Reference
L. minor			6.1-6.7	Hicks (1932, cited by DWRP, 1997); McLay (1976)
L. perpusilla	3.2		6.9-7.8	Landolt and Kandeler (1987); Khondker, Islam and Makhnun (1994)
S. polyrrhiza	3.7		6.5-8.5	Gopal and Chamanlal (1991); Islam and Khondker (1991), Kaul and Bakaya (1976); Landolt and Kandeler (1987)
S. punctata			7.0	McLay (1976)
W. arrhiza	3.5	10.0	5-7.8	Hicks (1932, cited by DWRP, 1997); Islam and Paul (1977); Landolt and Kandeler (1987)
W. australiana			5.0	McLay (1976)
W. columbiana			6.4-7.0	Hicks (1932, cited by DWRP, 1997)

TABLE 3.2

Minimum, maximum and optimum pH of various duckweed species

Source: DWRP (1998)

S. polyrrhiza by the end of May when a sharp fall in conductivity and alkalinity was observed. The electrolyte conductivity of water supporting the growth of *L. perpusilla* in Bangladesh reported by Islam and Khondker (1991) and Khondker, Islam and Makhnun (1994) were 625 µS/cm and 200-890 µS/cm, respectively. High electrolyte conductivity (1 090 µS/cm) of water supporting the growth of *L. perpusilla* was also reported by Van der Does and Klink (1991) in a lemnid habitat in the Netherlands.

## Nitrogen

In general, temperature and sunlight control duckweed growth more than nutrient concentrations in the water. At high temperatures, duckweed can grow rapidly down to trace levels of phosphorus and nitrogen. The crude protein content of duckweed however, seems to increase to a maximum of ~40 percent DM over the range from trace ammonia concentrations to 7-12 mg N/L (Leng, Stambolie and Bell, 1995). Culley *et al.* (1981) reported that the TKN of water should not drop below 20-30 mg/l if the optimum production and a high crude protein content of duckweed are to be maintained.

Duckweeds prefer ammonia nitrogen (NH<sub>4</sub>-N) as a source of nitrogen and will remove ammonia preferentially, even in the presence of relatively high nitrate concentrations. Lüönd (1980) demonstrated that higher growth rates were attained when nitrogen was in the NH<sub>4</sub>-N rather than the NO<sub>3</sub>-N form. In organically enriched waters, nitrogen tends to be concentrated in the NH<sub>4</sub>-N rather than the NO<sub>3</sub>-N form at pH levels below 9 and plant growth is equally efficient in anaerobic and aerobic waters (Said *et al.*, 1979). In lagoons receiving organic animal wastes, the pH seldom exceeds 8, particularly with a full duckweed cover that suppresses phytoplankton growth (Culley *et al.*, 1978). The plants can tolerate very high ionized ammonia (NH<sub>4</sub>-N) concentrations but the effects of unionized ammonia (NH<sub>3</sub>-N) have not been clearly demonstrated. Urea is a suitable fertilizer and is rapidly converted to ammonia under normal conditions. According to the results of laboratory experiments, duckweed tolerates concentrations of elemental N as high as 375 mg/l (Rejmánková, 1979).

## Phosphorus and potassium

Phosphorus is essential for rapid growth and is a major limiting nutrient after nitrogen, although its quantitative requirement for maximum growth is generally low. Fast growing duckweed in nutrient rich water is a highly efficient sink for both phosphorus and potassium; little of each, however, is required for rapid growth. Saturation of phosphate uptake by duckweed occurs at available PO<sub>4</sub>-P concentrations of 4 to 8 mg/l. Duckweed growth is not particularly sensitive to potassium or phosphorus once an adequate threshold has been reached. Rejmánková (1979) reported good growth of duckweed within the P concentrations of 6 to 154 mg/l. Culley *et al.* (1978),

working in dairy waste lagoons, achieved doubled production from 2 to 4 days at P concentrations in excess of 35 mg/l. Reduced growth in some species occurs only after P values dropped below 0.017 mg/l (Lüönd, 1980). Khondker, Islam and Makhnun (1994) observed that both phosphate and silicate concentrations had significant positive correlation with the biomass of *L. perpusilla* in Bangladesh.

#### Other minerals

A range of other important mineral levels found in water supporting Lemnaceae is presented in Table 3.3. Although these minerals are essential for their survival, duckweed growth is not particularly sensitive to potassium or phosphorus once an adequate threshold has been reached.

TABLE 3.3
Range of important mineral contents (mg/l) of water supporting Lemnaceae

Parameter	Absolute range	Range of 95 percent of the samples
K	0.5 – 100	1.0 – 30
Ca	0.1 – 365	1.0 – 80
Mg	0.1– 230	0.5 – 50
Na	1.3 ->1 000	2.5 – 300
HCO₃	8 – 500	10.0 – 200
Cl	0.1– 4 650	1.0 – 2 000
S	0.03 – 350	1.0 – 200

Source: modified from Landolt (1986)

## In summary

Maximum, minimum and optimum requirements of some of the most important environmental parameters (temperature, pH, conductivity, nitrogen and phosphorus) are given in Table 3.4. It is apparent that the duckweeds are robust in terms of survival, but sensitive in terms of thriving. They have extreme range of tolerance for temperature, pH, conductivity, nitrogen and phosphorus with well-defined range of optimum requirement.

## 3.3 PRODUCTION

#### 3.3.1 Background information

Duckweed growth is largely a function of available nutrients, temperature, light, and degree of crowding. The highest growth rate reported for Lemnaceae under optimal laboratory conditions is about 0.66 generations per day, which corresponds to a doubling time of 16 hours (DWRP, 1997). Duckweeds generally double their mass in 16 hours to 2 days under optimal nutrient availability, sunlight, and water temperature. An individual plant, a small leaflet (frond), produces 10 to 20 daughter fronds during its lifetime, which lasts for a period of 10 days to several weeks. The daughter frond repeats the history of its mother frond. This results in an exponential growth, at least until the plants become crowded or run out of nutrients. Frequent periodic removal of the plants encourages continuation of the exponential growth.

TABLE 3.4

Summary of some important environmental requirements of duckweed

	<del>-</del>		
Environmental parameters	Minimum	Maximum	Optimum
Temperature (°C)	>0	35	15-30
рН	3.0	10.0	6.5-8.0
Conductivity (µS/cm)¹	200	1 090	n.s.
Nitrogen (mg/l NH₃-N)	Trace	375	7-12
Phosphorus (mg/l PO₄-P)	0.017	154	4-8

 $<sup>^{\</sup>mbox{\tiny 1}}\mbox{Conductivity}$  range found supporting growth of duckweed.

The individual clones of the same species may show quantitative variation in growth characteristics (Rejmánková, 1975; Porath, Hepher and Koton, 1979). In the Czech Republic, Rejmánková (1975, 1979) reported maximum dry matter yields of 3.14-3.54 g and 7.09 g/m²/day from unmanaged fish ponds and outdoor tanks respectively, when weekly harvesting was done. Rejmánková (1981) further reported that an estimated annual net dry matter production of 7.5-8.0 tonnes/ha could be obtained, provided nutrients and crowding were not limiting and harvesting was frequent.

Culley and Myers (1980) and Said *et al.* (1979) working in the southern USA (9-10 months growing season) demonstrated that high nutrient lagoons and outdoor tanks (enriched with cattle manure) yielded the dry matter equivalent of about 15 g/m²/day (55 tonnes/ha/year) when regular daily harvesting was done to remove the excess. Said *et al.* (1979) reported an annual dry weight yield of 44 tonnes/ha or about 12 g/m²/day. Furthermore, Culley and Myers (1980) obtained an estimated average annual dry matter production of 23.3 tonnes/ha with daily harvesting ranging from 10 to 35 percent of the standing crop, depending on the season. In a sewage-fed culture system, the growth rate of *Azolla* spp., *Spirodela* spp. and *Wolffia* sp. were found to be 160, 350 and 280 g/m³/day, respectively (Reddy *et al.*, 2005).

Table 3.5 presents the yields of various duckweed species under different environmental conditions. The values varied widely, ranging from 9 to 38 tonnes (DM)/ha/year. This wide range of productivity may be attributed to differences in species, climatic conditions, nutrient supply and environmental conditions. Many of the reported high yields are based on extrapolated data obtained from short-term growth from small-scale experimental systems rather than potential long-term yields from commercial-sized systems. Edwards (1990) reported extrapolated yields of ~20 tonnes (DM)/ha/year of *Spirodela* from experiments that were carried out for periods of 1-3 months in septage-fed 200 m² ponds in Thailand; however, the yield declined to the equivalent of ~9 tonnes (DM)/ha/year over a 6 months period. Based on the available data and the foregoing discussion, it may therefore be concluded that an average annual yield of around 10-20 tonnes/DM/ha can be obtained from an aquatic environment where nutrients are generally not limiting and frequent harvesting is practised to avoid plant overcrowding.

#### 3.3.2 Duckweed farming

Duckweed farming is a continuous process requiring intensive management for optimum production. Daily attention and frequent harvesting are needed throughout the year to ensure optimum productivity. Duckweed can grow in water of any depth.

TABLE 3.5

Yields of various duckweed species under different environmental conditions

Species	Environmental condition	Yield (dry matter tonnes/ha/year)	Reference
L. minor	UASB effluent	10.7	Vroon and Weller (1995)
L. minor	Nutrient non-limiting water	16.1	Reddy and DeBusk (1984)
L. perpusilla	Septage-fed pond	11.2	Edwards, Pacharaprakiti and Yomjinda (1990)
L. perpusilla, S. polyrrhiza and W. arrhiza	Septage from septic tank	9.2-21.4	Edwards <i>et al.</i> (1992)
Lemna	Domestic wastewater	26.9	Zirschky and Reed (1988)
Lemna	Sugar mill effluent	32.1	Ogburn and Ogburn (1994)
<i>Lemna, Spirodela</i> and <i>Wolffia</i>	Domestic wastewater	13-38	Skillicorn, Spira and Journey (1993)
Lemna and Wolffia	Faecally polluted surface water	14-16	Edwards (1987)
S. polyrrhiza	Domestic wastewater	17-32	Alaerts, Mahbubar and Kelderman (1996)
S. polyrrhiza	Sewage effluent	14.6	Sutton and Ornes (1975)
S. polyrrhiza	Nutrient non-limiting water	11.3	Reddy and DeBusk (1985)

It will grow in as little as one centimetre of water. A pond depth of between 20 and 50 cm is generally recommended to reduce the potential sources of stress and to facilitate harvesting (Gaigher and Short, 1986). Duckweeds are prone to be blown into heaps by heavy winds or wave action. This allows light to penetrate the water column and would stimulate phytoplankton and algal growth. If the plants become piled up in deep layers, however, the lowest layer will be cut off from light and will eventually die (Skillicorn, Spira and Journey, 1993). Plants pushed from the water onto a bank will also dry out and die. Long narrow ponds that are sited perpendicular to the common wind are recommended. Dividing the pond into smaller segments by using bamboo can also mitigate the adverse effects of wind. An NGO called 'PRISM' applied a grid of bamboo poles (Figure 3.4) of approximately 5 x 5 m in large ponds and wide canals. This functioned satisfactorily for all conditions met up to that date in Bangladesh (DWRP, 1998). Lemma USA Inc. promotes floating barrier grids made of polyethylene that will reduce wind and wave action for its wastewater treatment plants. The sides of the ponds must preferably be vertical to prevent the plants from becoming stranded and at least 10 cm higher than the water level to accommodate heavy rains. The ponds must be fed with effluent through furrows rather than pipes because the latter tend to become clogged. Several inlets must be provided to spread the inflowing nutrients over the pond.

Since the growth of duckweed is dependent on water temperature, pH and nutrient concentration, these factors need to be balanced and maintained within reasonable limits for duckweed to thrive. The management strategies for duckweed culture should therefore focus on when to fertilize, harvest, and buffer; how much to fertilize and to harvest; and which nutrients to supply. Appropriate management should be aimed at maintaining a complete and dense cover of duckweed, low dissolved oxygen, and a pH of 6-8. A dense cover shuts out light and suppresses the growth of algae, which minimizes CO<sub>2</sub> production from algal respiration and prevents its elevating effect on pH.

Any waste organic material that is readily biodegradable and has a sufficiently high nutrient content could be used for duckweed cultivation. The most economical sources of such waste materials are all kinds of animal manure, kitchen wastes, wastes from a wide range of food processing plants, biogas effluents, and slaughterhouse wastes. Solid materials, such as manure from livestock, night soil from villages, or food processing

FIGURE 3.4

A slowly flowing wastewater treatment canal covered with duckweed and provided with a bamboo base to prevent the duckweed from floating along the stream (PRISM Experimental Project, Mirzapur, Bangladesh)



Source: DWRP (1998)

wastes, can also be mixed with water and added to ponds at suitable levels. All wastewater containing manure or night soil must undergo an initial treatment by holding it for a few days in an anaerobic pond, before using it to cultivate duckweed.

Sutton and Ornes (1975) and Said *et al.* (1979) demonstrated the necessity of periodic additions of nutrients to small duckweed culture systems receiving municipal or dairy cattle wastes. Within 1-3 weeks, there was a noticeable drop in N, P and K within the plants. There was a corresponding drop in crude protein as the plant nitrogen declined. In unmanaged ponds, where duckweeds are not routinely harvested, the plants quickly become crowded and those beneath the surface die back.

Due to the high nitrogen requirement of duckweed and the relatively rapid loss of nitrogen from aquatic system, this nutrient tends to be limiting in ponds fed with wastewater (Gaigher and Short, 1986). Studies at Louisiana State University have shown that the nitrogen conversion efficiency from agricultural waste to duckweed is only about 30 percent under normal field conditions (Culley et al., 1981). Large-scale duckweed production therefore requires the availability of relatively large quantities of organic waste. The addition of cheap inorganic nitrogen could also therefore improve the wastewater conversion efficiency. The other nutrients that are needed for optimum growth of duckweed are phosphorus, potassium and trace minerals.

#### Fertilization

Urea is a suitable fertilizer, containing approximately 45 percent nitrogen, and is rapidly converted to ammonia under normal conditions. Muriate of potash (MP) and triple superphosphate (TSP) are commercial sources of potassium and phosphorus that are widely available in most countries and have been used where duckweeds have been farmed. Duckweed growth is not particularly sensitive to potassium or phosphorus once an adequate threshold has been reached. A ratio of TSP to urea of 1:5 worked satisfactorily in an experimental duckweed production programme in Bangladesh (Skillicorn, Spira and Journey 1993). Similarly, a ratio for MP to urea of 1:5 was found to be satisfactory for good production in the same duckweed production programme in Bangladesh.

Nutrients are absorbed through all surfaces of the duckweed leaf (Leng, Stambolie and Bell, 1995). There are at least three methods of fertilizer application including broadcasting, dissolving in the water column of the plot, and spraying a fertilizer solution on the duckweed mat.

A fertilizer application matrix aiming to achieve variable daily production ranging from 500-1 000 kg of fresh duckweed per hectare was developed by PRISM in their experimental programme at Mirzapur, Bangladesh (Table 3.6). Furthermore, PRISM recommended daily fertilization rates for different types of duckweed (Table 3.7). The application rate varies from 21-28 kg/ha/day (amounting to >7 tonnes/ha/year) with an anticipated fresh biomass yield of 900-1 000 kg/ha/day. The daily fertilization rate for duckweed cultivation developed by the Bangladesh Fisheries Research Institute (BFRI) is presented in Table 3.8. The fertilizer schedules developed by PRISM and BFRI are very similar (Tables 3.7 and 3.8), except that BFRI recommended half the dosage of inorganic fertilizer when cow dung was used at the rate of 750 kg/ha/year.

TABLE 3.6

Dayly fertilizer application matrix for duck weed cultivation developed by PRISM in their experimental programme at Mirzapur, Bangladesh

Fertilizer application (kg/ha)	500	600	700	800	900	1 000
		Dai	ly production of	f fresh plants (kg	/ha)	
Urea	10.0	12.0	14.0	16.0	18.0	20.0
TSP	2.0	2.4	2.8	3.2	3.6	4.0
MP	2.0	2.4	2.8	3.2	3.6	4.0
Sea salt	4.5	5.4	6.3	7.2	8.1	9.0

Source: Skillicorn, Spira and Journey, (1993)

#### Seeding

Seeding is a highly important management measure since a full duckweed cover should be established before any algal bloom can start dominating the water body. The seed rate advised is 60 kg/100 m² for *Spirodela* spp. and *Wolffia* spp. and 40 kg/100 m² for *Lemna* spp. in order to obtain a dense cover in 3 days time (DWRP, 1998). From day four onwards daily harvesting can start.

#### Stress management

Stress management of the crop is necessary particularly during very hot and dry weather. 'Dunking' (dipping the duckweed below the water surface) once a day as a regular crop maintenance practice is recommended; this reduces the stress from overheating. Dunking consists of agitating the whole-cultivated area by hand until all plants have been physically immersed and wetted.

## Plant density and harvesting rate

The productivity of duckweed increases with increasing plant density up to a density where the plants completely cover the surface of the water, and then remains constant. In order to maintain good productivity and prevent competition by phytoplankton/suspended algae, the density must be maintained at this level or a slightly higher level. Competition between phytoplankton/suspended algae and duckweed is a potential constraint to the cultivation of the latter in nutrient-rich water. Phytoplankton smothers the roots of duckweed, which then turn yellow in colour, suffer a decline in growth rate, and eventually die. The development of an algal bloom can also reduce nutrient availability and thus eventually reduce the growth of duckweed.

An optimum standing crop density is a cover that is complete but which still provides enough space to accommodate rapid growth of the colony. In the PRISM experimental programme at Mirzapur, Bangladesh a base *Spirodela* density of 600 g/m² was shown to yield a daily incremental growth of 50 to 150 g/m²/day (Skillicorn, Spira and Journey, 1993). This is equivalent to a daily fresh (wet weight) crop production rate of 0.5 to 1.5 tonnes/ha. These authors recommended a plant density of 400 to 800 g/m² for optimum production. BFRI (1997) obtained duckweed production of 700-1 500 kg/ha/day at plant densities varying from 400-600 g/m² in their experimental programme at Mymensingh.

High-density populations contain a high ratio of old fronds, which can be detrimental in various ways. Duckweed should therefore be harvested frequently, preferably daily. The standing crop density, or the weight of fresh plant per square meter, will determine the amount and timing of harvests. Daily harvesting of the incremental growth of the duckweed plot - averaging approximately 100 g/m²/day is recommended (Skillicorn, Spira and Journey, 1993). Culley and Myers (1980) obtained an annual dry weight production of 23.31 tonnes/ha with daily harvesting ranging from 10 to 35 percent of the standing crop each day, depending on the season. Edwards (1990) recommended 25 percent harvesting of the duckweed biomass when duckweed growth completely covers the pond, with the remaining 75 percent left in the pond for further growth.

TABLE 3.7

Rates of fertilization application for duckweed cultivation techniques developed by PRISM

• •			•
Duckweed	Rate of ap	pplication (kg/ha/day)	
	Urea	TSP	MP
Spirodela	20	4	4
Wolffia	15	3	4
Lemna	15	3	3

Source: DWRP (1998)

TABLE 3.8

Rates of fertilization application for duckweed cultivation techniques developed by Bangladesh Fisheries Research Institute (BFRI)

Fertilizer combination	Rate	of application	n (kg/ha/da	y)
	Urea	TSP	MP	Cow dung
Inorganic fertilizer only	15-20	3-4	3-4	-
Combination of organic and Inorganic fertilizer	7.5	1.5	1.5	750

Source: BFRI (1997)

This author opined that this harvest could be made every 1-3 days, depending on the season.

## Duckweed in wastewater treatment

Ferdoushi et al. (2008) tested the efficacy of Lemna and Azolla as biofilters of nitrogen and phosphate in fish ponds in Bangladesh and found that they removed the excess amount of nutrients from the water body and maintained sustainable environmental conditions. Duckweeds have received much attention because of their potential to remove contaminants from wastewater (Leng, Stambolie and Bell, 1995). Duckweed wastewater treatment systems have been studied for dairy waste lagoons (Culley et al., 1981), raw domestic sewage (Oron, 1994; Skillicorn, Spira and Journey, 1993; Alaerts, Mahbubar and Kelderman, 1996), secondary effluent (Harvey and Fox, 1973), waste stabilization ponds (Wolverton, 1979) and fish culture systems (Porath and Pollock, 1982; Rakocy and Allison, 1981). The basic concept of a duckweed wastewater treatment system is to farm local duckweed on the wastewater requiring treatment. Duckweed has a high mineral absorption capacity and can tolerate high organic loading as well as high concentrations of micronutrients.

Duckweed wastewater treatment systems remove, by bioaccumulation, as much as 99 percent of the nutrients and dissolved solids contained in wastewater (Skillicorn, Spira and Journey, 1993). These substances are then removed permanently from the effluent stream following the harvesting of a proportion of the crop. The plants also reduce suspended solids and BOD by reduction of sunlight in lagoons. Duckweed systems distinguish themselves from other effluent wastewater treatment mechanisms in that they also produce a valuable, protein-rich biomass as a by-product.

Depending on the wastewater, the harvested crop may serve as an animal feed, a feed supplement supplying protein/energy and minerals, or a fertilizer. The question of toxic elements must be considered if certain types of waste material serve as the nutrient source for duckweed culture; for example, duckweed will absorb heavy metals and insecticides from the wastewater. It may, therefore, have to be decontaminated prior to feeding to animals if heavy metals are present in the water.

Landolt and Kandeler (1987) reported that of all aquatic plants, Lemnaceae have the greatest capacity in assimilating the macro-elements N, P, K, Ca, Na and Mg. Table 3.9 presents some data on daily nitrogen and phosphorus uptake efficiency by duckweed. The results from the various studies are not comparable because different species are used and different climatological and operational conditions were applied. Temperature may have a significant effect on nutrient uptake efficiency as has also been observed for other aquatic plants.

TABLE 3.9

Daily nitrogen and phosphorus uptake by duckweed

Region/Country	Species	Up	otake (g/m²)
		N	P
Italy	L. gibba/ L. minor	0.42	0.01
CSSR	Duckweed	0.20	-
USA	Lemna sp.	1.67	0.22
Louisiana, USA	Duckweed	0.47	0.16
India	Lemna sp.	0.50-0.59	0.14-0.30
Minnesota, USA	Lemna sp.	0.27	0.04
Florida, USA	S. polyrrhiza	-	0.015

Source: adapted from DWRP (1997)

Culley et al. (1981) made a comprehensive study on nutrient uptake from wastewater by a mixed culture of duckweed (Table 3.10). This shows that duckweeds are capable of removing considerable amounts of organic wastes from natural water. An annual nutrient removal capacity covered by *Lemnaceae* of 1 378 kg TKN, 345 kg P, and 441 kg K per hectare of water area was calculated.

Summarizing the results of PRISM Experimental Site at Mirzapur, Skillicorn, Spira and Journey (1993) reported that treating an average flow of 125 m³/day of hospital, school, and residential wastewater produced by a population of between 2 000 and 3 000 persons, the 0.6 ha duckweed treatment plant produces a final treated effluent that exceeds the highest quality standards mandated in the USA (Table 3.11). These authors also estimated that a typical duckweed wastewater treatment plant would yield a daily harvest of up to one ton of duckweed plants (wet weight) per hectare or 90 kg per hectare of dried, high protein duckweed meal each day.

TABLE 3.10

Mean annual dry duckweed yield and nutrient uptake by a mixed culture of duckweed¹ harvested daily² from a 25 m² lagoons³ receiving dairy cattle wastes⁴

	Duckweed yield		Duckweed⁵	
	(kg/m²)	TKN (g/m²)	P (g/m²)	K (g/m²)
Dec-Feb	0.195	11.5	2.9	3.7
Mar-May	0.576	34.2	8.5	10.9
Jun-Aug	1.020	60.2	15.1	19.3
Sep-Nov	0.540	31.9	8.0	10.2
Total (kg/ha)	23 310	1 378	345	441

<sup>&</sup>lt;sup>1</sup> S. polyrrhiza, S. punctata, L. gibba and W. columbiana in approximate equal amounts at Baton Rouge, Louisiana, USA (9-10 months growing season)

Source: modified from Culley et al. (1981)

TABLE 3.11

Quality of final treated effluent at Mirzapur Experimental Site on 23 March 1991

Treatment phase	BOD₅ (mg/l)	NH₃ (mg/l)	P (mg/l)	Turbidity (FTU¹)
Raw influent	120	39.40	1.90	113
Primary	60	32.20	2.00	85
Duckweed	1	0.03	0.03	10
US Summer Standards: Washington D.C. area	10	2.00	1.00	20

<sup>&</sup>lt;sup>1</sup>This turbidity unit standard is roughly equivalent to total suspended solids (TSS) times two

Source: Skillicorn, Spira and Journey (1993)

## 3.4 CHEMICAL COMPOSITION

Each frond of duckweed absorbs nutrients through the whole plant, not through a central root system, directly assimilating organic molecules such as simple carbohydrates and various amino acids. The entire body is composed of non-structural, metabolically active tissue; most photosynthesis is devoted to the production of protein and nucleic acids, making duckweeds very high in nutritional value. The nutritional content of duckweed is probably more dependent on the mineral concentrations of the growth medium than on the species or their geographic location. Water low in nutrients generally results in reduced nutritional content in duckweed. Crude fibre content is generally lower (varying between 7-10 percent) for duckweed grown in nutrient-rich water than that grown in nutrient-poor water (11-17 percent).

Compared with most plants, duckweed leaves have little fibre (5 percent in cultured plants) as they do not need to support upright structures (Leng, Stambolie and Bell, 1995). Crude fibre content was generally lower, varying between 7-10 percent, for duckweed grown in nutrient-rich water as opposed to 11-17 percent for duckweed

<sup>&</sup>lt;sup>2</sup>September-February: a mean of 105 of duckweed removed daily; March-August: a mean of 35 percent of duckweed removed daily

<sup>&</sup>lt;sup>3</sup> Trials were run in triplicate

<sup>&</sup>lt;sup>4</sup> Fresh manure loading in first stage lagoon provided an effluent to the test lagoons with 15-65 mg/l TKN; 18-28 mg/l phosphorus (P); 38-69 potassium (K); pH 7.6-7.9

<sup>&</sup>lt;sup>5</sup>TKN 5.9 percent of dry weight, P 1.48 percent and K 1.89 percent

grown in nutrient-poor water. In general, the ash content ranges between 12-18 percent (Leng, Stambolie and Bell, 1995).

Duckweeds are known to accumulate large amounts of minerals in their tissues. Skillicorn, Spira and Journey (1993) reported that fibre and ash contents are higher and protein content lower in duckweed colonies with slow growth. Duckweeds are rich source of nitrogen, phosphorous, potassium and calcium (Guha, 1997). The concentration of N and P in duckweed tissues depend on the amount of N and P in the water, up to a threshold concentration that has not been clearly defined. Above this threshold, there is little increase in the tissue. Culley et al. (1978) suggested that under lagoon conditions, 20-30 mg/l TKN might be required to maintain a crude protein level above 30 percent. The crude protein content of duckweeds grown on various nutrient solutions ranges from 7 to 45 percent of the plant dry weight, depending on the nitrogen availability (Culley et al., 1981). When conditions are good, duckweed contains considerable protein, fat, starch and minerals, which appear to be mobilized for biomass growth when nutrient concentrations fall below the critical levels for growth. Nutrient contents in duckweed may therefore vary according to the conditions in which it is grown. Slow growth, starvation and aging have been reported to result in protein levels as low as 7 percent DM (Landolt and Kandeler, 1987).

A summary of the nutritional composition of different species grown under different environmental conditions is presented in Table 3.12. Fresh duckweed contained about 91-95 percent water and the moisture content is apparently not influenced by the medium under which it was grown. Duckweed species grown under nutrient-poor water or under sub-optimum nutrient conditions have crude protein contents varying between 9-20 percent, while the level varied from 24-41 percent for duckweed species grown in nutrient-rich water. The crude protein content of duckweed seems to increase from trace ammonia concentrations to 7-12 mg N/L when crude protein reaches a maximum of about 40 percent (Leng, Stambolie and Bell, 1995). Similarly, the lipid content was lower (1.8-2.5 percent) in duckweed species grown in nutrient-poor water, while it generally varied between 3-7 percent for duckweed grown in nutrient-rich water. The medium in which duckweed was grown or the nutrient status of water did not influence the ash content of duckweed (Leng, Stambolie and Bell, 1995). Skillicorn, Spira and Journey (1993) reported that fibre and ash contents are higher and protein content lower in duckweed colonies with slow growth.

Studies by Porath, Hepher and Koton (1979) and Rusoff, Blakeney and Culley (1980) show clearly that the duckweed indeed has high quality protein. It has a better essential amino acid profile than most plant proteins and more closely resembles animal protein than any other plant proteins. According to Guha (1997), the protein of duckweeds is rich in certain amino acids that are often rather low in plant proteins. The nutritional value of Lemnaceae can be compared favourably with that of alfalfa in terms of lysine and arginine, two amino acids important in animal feeds. Duckweeds are rich in leucine, threonine, valine, isoleucine and phenylalanine and are low in methionine and tyrosine. Some information on the amino acid content of various aquatic macrophytes is contained in Annex 1. Annex 1 Table 3 shows mean values determined for amino acids in four species of duckweed. It is evident that the values for the essential amino acids compare favourably with the FAO reference pattern, with the exception of methionine. The levels of amino acids are very similar in the various species and all the essential amino acids were generally present.

Cultured duckweed has high concentrations of trace minerals and pigments, especially β-carotene and xanthophyll (Haustein *et al.*, 1988). Duckweeds store varying amounts of calcium as calcium oxalate crystals in the vacuoles. Calcium oxalate may be toxic in large doses and the amount should be reduced to make duckweeds more

<sup>1</sup> www.mobot.org/jwcross/duckweed/nutritional-composition.htm

Chemical analyses of various duckweed species grown under different environmental conditions **TABLE 3.12** 

									1-1-1	, , , , , , , , , , , , , , , , , , ,
Duckweed species	Aquatic environment	(percent)	Ď.	хішате соп	rroximate composition (percent Divi)	cent Divi)		(percent DM)	t DM)	Reletence
			8	Ш	Ash	ភ	NFE	Ca	4	
L. gibba, USA	Low nutrient lagoon <sup>2</sup>	n.s.	9.4	1.8	16.8	17.0	55.56	1.38	0.72	Culley <i>et al.</i> (1981)
L. minor, Bangladesh	Pond, nutrient status not specified	92.0	14.0	1.9	12.1	11.1	6.09	n.s.	n.s.	Zaher e <i>t al.</i> (1995)
L. minor, Bangladesh	Ditch, nutrient status not specified	93.8	20.3-23.5	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	Majid et al. (1992)
L. polyrrhiza, India, raw leaf meal	Freshwater	32.5	18.6	1.5	2.5	11.0	$66.4^{6}$	n.s.	n.s.	
L. polyrrhiza, fermented, India	:	n.s.	11.4	1.0	n.s.	7.5	n.s.	n.s.	n.s.	
S. polyrrhiza, USA	Low nutrient lagoon <sup>2</sup>	n.s.	13.1	2.5	13.3	16.1	$55.0^{6}$	1.21	0.56	Culley <i>et al.</i> (1981)
S. polyrrhiza, Bangladesh	Ditch, nutrient status not specified	95.0	17.3-28.4	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	Majid e <i>t al.</i> (1992)
S. punctata, USA	Low nutrient lagoon <sup>2</sup>	n.s.	10.6	2.3	14.1	11.3	61.76	0.98	0.61	Culley <i>et al.</i> (1981)
W. arrhiza, whole plant, Bangladesh	Ditch, nutrient status not specified	91.2	14.9	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	Majid e <i>t al.</i> (1992)
<i>L. gibba</i> , USA	High nutrient lagoon³	n.s.	36.3	6.3	15.5	10.1	31.86	1.81	2.60	Culley <i>et al.</i> (1981)
L. gibba, USA	Dairy cattle waste lagoon	n.s.	38.5	3.0	16.4	9.4	32.76	1.00	1.60	Hillman and Culley (1978)
L. minima, USA	Source not specified	n.s.	31.0	2.0	14.0	10.0	42.0	n.s.	n.s.	Shireman, Colle and Rottmann (1977)
L. perpusilla, Thailand	Septage-fed earthen pond	94.0-94.3	25.3-29.3	3.8-4.5	15.4-17.6	6.9-7.6	n.s.	n.s.	n.s.	Hassan and Edwards 1992)
S. oligorrhiza, USA	Dairy cattle waste lagoon	n.s.	37.8	3.8	12.0	7.3	39.16	1.30	1.50	Hillman and Culley (1978)
S. oligorrhiza, USA⁴	Treated wastewater effluent	n.s.	32.7	6.3	20.3	13.5	$27.2^{6}$	1.49	1.15	Culley and Epps (1973)
S. oligorrhiza, USA⁴	Untreated septic tank influent	n.s.	32.3	n.s.	n.s.	n.s.	n.s.	1.29	1.17	Culley and Epps (1973)
S. oligorrhiza, USA4.5	Anaerobic swine waste lagoon	n.s.	41.4	5.1	12.9	8.3	$32.3^6$	0.91	2.07	Culley and Epps (1973)
S. <i>polyrrhiza,</i> Thailand	Septage-fed earthen pond	91.0	23.8	3.8	18.3	11.7	42.46	n.s.	n.s.	Hassan and Edwards (1992)
S. polyrrhiza, USA	High nutrient lagoon³	n.s.	39.7	5.3	12.8	9.3	32.96	1.28	2.10	Culley e <i>t al.</i> (1981)
S. polyrrhiza, USA	Dairy cattle waste lagoon	n.s.	40.9	6.7	12.9	8.7	30.8	2.10	1.40	Hillman and Culley (1978)
S. punctata, USA	High nutrient lagoon³	n.s.	36.8	4.8	15.2	9.7	33.5	1.75	1.50	Culley <i>et al.</i> (1981)

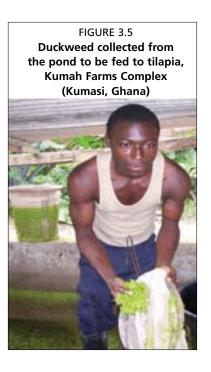
<sup>1</sup> CP = crude protein; EE = ether extract; CF = crude fibre; NFE = nitrogen free extract; Ca = calcium; P = phosphorus
<sup>2</sup> Low nutrient lagoon contained less than 5 mg/l TKN
<sup>3</sup> High nutrient lagoon contained more than 30 mg/l TKN

<sup>4</sup> Proximate composition and mineral content values corrected to dry basis <sup>5</sup> Mean of eight values sampled over five months period <sup>6</sup> Adjusted or calculated; not as cited in original publication

nutritious and digestible (Franceschi, 1989). The metabolic precursor of oxalate is L-ascorbic acid (vitamin C). A study with water lettuce (*Pistia stratiotes*) (see section 5) indicates that L-ascorbate and oxalate are synthesized within the crystal idioblast cells (Kostman *et al.*, 2001).

# 3.5 USE AS AQUAFEED

Because of its attractive nutritional qualities and the relative ease of production, a significant number of studies have been carried on the potential utilization of duckweed biomass as fish feed (Shireman, Colle and Rottmann 1977, 1978; Hillman and Culley, 1978; Stephensen et al., 1980; Gaigher, Porath and Granoth, 1984; Naskar et al., 1986; Hassan and Edwards, 1992). Available literature indicates that duckweeds are fed to fish in fresh form as a sole feed or in combination with other feed ingredients (Figure 3.5). Duckweeds are also fed as a dried meal ingredient in pelleted diets. Intensive fish production with duckweed as a predominant feed constituent has been reported by a number of authors (Hepher and Pruginin, 1979; Robinette, Brunson and Day, 1980; Culley et al., 1981; Landolt and Kandeler, 1987, Skillicorn, Spira and Journey, 1993). Research studies on the use of duckweed as fish feed have been carried out under laboratory as well as under field conditions. Successful results have also been obtained on the on-farm utilization of duckweed as fish.



## 3.5.1 Laboratory studies

Successful feeding trials for grass carp with duckweed have been carried out since the early 1960s. Studies on the consumption of duckweed by aquatic animals have generally been confined to this species, although more recently feeding trials have also been carried out with others, including common carp, catfish, Indian major carps and tilapia.

Results on the use of duckweed as a feed for grass carp are generally very positive (Galkina, Abdullaev and Zacharova, 1965; Nikolskij and Verigin, 1966; Fischer, 1968, 1970; Edwards, 1974; Porath and Koton, 1977; Shireman, Colle and Rottmann, 1977, 1978; Baur and Buck, 1980; Hajra and Tripathi, 1985). Galkina Abdullaev and Zacharova (1965) reported that the grass carp showed more rapid growth when using duckweed than other feed materials. Porath and Koton (1977) noted that the weight of grass carp could be tripled (from 100 g to 300 g) within 50 days when feeding a mixture of *L. gibba* and *L. minor*.

Fresh duckweeds have also been efficiently utilized by common carp, catfish, Indian major carps and tilapia (Hepher and Pruginin, 1979; Robinette, Brunson and Day, 1980; Stephensen *et al.*, 1980; Gaigher, Porath and Granoth, 1984; Naskar *et al.*, 1986; Hassan and Edwards, 1992).

Summary results of selected growth studies carried out on the use of fresh and dried duckweed as feed for different fish species are presented in Tables 3.13 and 3.14. Fresh and dried duckweed were fed to grass carp, Nile tilapia, common carp, Indian major carps (rohu and mrigal), silver carp, Java barb, hybrid grass carp and hybrid tilapia. The duckweed species evaluated were *L. gibba*, *L. perpusilla*, *L. minima*, *L. minor*, *Wolffia columbiana* and *W. arrhiza*. Fresh duckweeds were fed as a sole feed whereas dried duckweed meal was incorporated by partially replacing other conventional feed ingredients in pelleted diets. Feeding trials were conducted for varying periods, ranging from 60 to 155 days. Fish were fed *ad libitum* or at restricted level. In some studies,

TABLE 3.13
Performance of fish fed fresh duckweed

Duckweed/ Fish species	Rearing system	Rearing days	Control diet	Feeding rate (percent BW/day)	(percent y)	Fish	SGR (percent)	SGR as percent	FCR (DM basis²)	Reference
				Fresh weight	Dry weight	(b)		of control		
L. gibba/ tilapia hybrid'	Recirculatory tanks	68			>1.0	2.66	0.67		1.0	Gaigher, Porath and Granoth (1984)
L. gibbal hybrid grass carp	Static water outdoor concrete tank	09	Catfish pellet (32 percent protein)	Ad libitum		1 015	0.21	56	6.69	Cassani, Caton and Hansen (1982)
<i>L. perpusillal</i> Nile tilapia	Static water outdoor concrete tank	70			2.5	26.3	0.97		2.2	Hassan and Edwards (1992)
<i>L. perpusillal</i> Nile tilapia	Static water outdoor concrete tank	70			3.0	43.7	1.34		1.6	Hassan and Edwards (1992)
L. minimalgrass carp	Flow-through circular fibre glass tank	88		Ad libitum		2.7	3.74		1.72-1.97	Shireman, Colle and Rottmann (1977)
L. minima/grass carp	Flow-through circular fibre glass tank	89	Catfish chow (32 percent	Ad libitum		2.8	3.88	212	1.6	Shireman, Colle and Rottmann. (1978)
S. polyrrhizal Nile tilapia	Static water outdoor concrete tank	70	protein)		2.5	25.6 27.9	0.59		3.1	Hassan and Edwards (1992)
W. columbiana/ hybrid grass carp	Static water outdoor concrete tank	09	Catfish pellet (32 percent protein)	Ad libitum		1 033	0.51	135	3.76	Cassani, Caton and Hansen (1982)
W. arrhiza/ grass carp	Cement cistern	133		Ad libitum		5.5	3.50			Naskar et al. (1986)
W. arrhiza/ silver carp	Cement cistern	133		Ad libitum		15.5	2.33			Naskar <i>et al.</i> (1986)
W. arrhizal common carp	Cement cistern	155		Ad libitum		15.0	1.90			Naskar <i>et al.</i> (1986)
W. arrhizal Java barb	Cement cistern	120		Ad libitum		9.5	2.49			Naskar <i>et al.</i> (1986)
<i>W. arrhizal</i> rohu	Cement cistern	155		Ad libitum		2.0	2.87			Naskar e <i>t al.</i> (1986)
<i>W. arrhiza</i> / mrigal	Cement cistern	155		Ad libitum		0.9	2.54			Naskar <i>et al.</i> (1986)

<sup>1</sup> O. niloticus X O. aureus <sup>2</sup> Dry matter basis

TABLE 3.14
Performance of fish fed pelleted feeds containing dried duckweed

	L 50:	5)	6							
Duckweed/ Fish species	Rearing system	Rearing days	Rearing Control diet days	Composition of test diet	Inclusion level (percent)	Fish size (g)	SGR (percent)	SGR as percent of control	FCR	FCR Reference
<i>L. minorl</i> Nile tilapia	Static water glass aquaria	70	Fishmeal: sesame oil cake: black gram bran: rice bran (27:25:19:25)	Duckweed meal incorporated by partial replacement of black gram and rice bran	13.5	2.55	2.13	86		2.1 Zaher e <i>t al.</i> (1995)
<i>L. minorl</i> Common carp	Static water outdoor cement	140	Rice bran: groundnut cake (60:40) (17.5 percent protein)	Duckweed meal, groundnut cake, rice bran and ragi flour (40:20:20:20) (21.3 percent protein)	40	3.0	2.29	94	3.1	Devaraj, Krishna and Keshavappa (1981)

the performances of fish fed duckweed were compared with control diets, although in many of these studies no control diet was used for comparison.

Growth responses of different fish species fed various species of fresh duckweed were variable. However, the general trend was that the grass carp performed better than Nile tilapia and other species and the performances of duckweed as whole feed were better than control diet (Table 3.13). Similarly, duckweed meal incorporated in pelleted diets at 13.5 and 40 percent showed similar growth responses compared to the growth responses of fish fed control diets (Table 3.14). The SGRs obtained for grass carp fed fresh duckweed as whole feed varied between 1.2 and 3.9 while the SGR values for Nile tilapia were between 0.6 and 1.4. Fasakin, Balogum and Fasuru (1999) reported that duckweed meal (*Spirodela polyrrhiza*) can form up to 30 percent of the total diet of Nile tilapia without significant effect on performance, compared to a control without duckweed. However, inclusion levels above this level progressively decreased fish performance.

Duckweed are generally the preferred macrophytes for most of the herbivorous fish, although several authors reported that submerged macrophytes such as oxygen weed (*Hydrilla*) and water velvet (*Najas*) are more preferred than others. The preference of duckweed to other aquatic plants has been reported for grass carp and other fish species in several observations (Opuszynsky, 1972; Duthu and Kilgen, 1975; Rifai, 1979; Cassani, 1981; Cassani and Caton, 1983). Information on whether fish prefer any particular duckweed species over others is lacking.

Ad libitum feeding of fresh duckweed is mostly used for herbivorous fish. Limited numbers of investigations have been carried out to optimize the feeding or consumption rate of duckweed but most were carried out for grass carp and Nile tilapia. Nikolskij and Verigin (1966) reported grass carp consumed fresh duckweed equal to their body weight over a 24 hour period. Baur and Buck (1980) reported that grass carp consumed from 85 percent to 238 percent of their body weight/day (BW/day) on a mixed diet of *Lemna, Spirodela* and *Wolffia* spp. Shireman, Colle and Rottmann (1977) recorded consumption rates varying from 7.2-7.4 percent BW/day on a dry weight basis (DW) for grass carp while fresh duckweed (L. minima) was fed ad libitum. Since duckweed contains about 92 percent moisture, the dry weight feeding rates given above are equivalent to about 90-92 percent BW/day on a fresh weight basis. Shireman, Colle and Rottmann (1978) fed fresh *L. minima ad libitum* to grass carp and recorded daily mean consumption rates of 7.6 percent and 4.3 percent BW/day DW for 2.8 and 62.8 g sized fish respectively. Similar size-dependent feeding rates were reported by Hassan and Edwards (1992) for Nile tilapia. These authors studied the effect of feeding rate of L. perpusilla on the survival, growth and food conversion rate of Nile tilapia and recorded that the optimal daily feeding rates of Lemna were 5, 4 and 3 percent BW/day DW for fish of 25 to 44 g, 45 to 74 g and 75 to 100 g, respectively. Hassan and Edwards (1992) concluded that duckweed should be fed to tilapia according to its consumption rate, in order to avoid creating adverse water conditions, and that the feeding rate should be decreased as the fish grow larger.

A study by Effiong, Sanni and Sogbesan (2009) also indicated that the inclusion of duckweed meal in fish feeds could improve its binding potential and water stability.

## 3.5.2 Field studies and on-farm utilization

Several field studies and reports about the on-farm utilization of duckweed as feed for various fish species exist (e.g. Edwards, 1980, 1987; Edwards, Pacharaprakiti and Yomjinda, 1990; Skillicorn, Spira and Journey 1993; DWRP, 1998).

Edwards (1987) reported the on-farm utilization of duckweed in China and Taiwan Province of China. This author reported that the duckweeds *L. minor*, *S. polyrrhiza* and *W. arrhiza* are cultivated in small shallow ponds (similar to that illustrated in Figure 3.6) fertilized with manure (livestock or human) and fed to grass carp fry

and fingerlings in nursery areas. Initially the fry are fed the smaller Wolffia, but when they reach 6 to 7 cm in length they are fed the larger Lemna and Spirodela. In Taiwan Province of China a mixture of Lemna and Wolffia is cultivated in shallow earthen ponds fed with faecally-polluted surface water for use as fish feed.

Skillicorn, Spira and Journey (1993) described the 'duckweed-fed carp polyculture system' developed in the PRISM experimental farm at Mirzapur, Bangladesh. The duckweed (*Lemna*, *Spirodela and Wolffia*) carp polyculture model has an 18-month cycle. Fingerlings were introduced in August and

FIGURE 3.6
Two boys collecting duckweed from a village pond
(Jessore, Bangladesh)



September and harvesting began in March and continued for approximately one year. A second 18-month cycle began the following year and continued concurrently for six months. After the initial six months, the model allowed year-round harvesting. Biweekly harvesting was the preferred pattern, following a simple protocol to take the largest fish (75 to 100 percentile) and the smallest (0 to 25 percentile) in each species. The rationale was the assumption that the largest fish will exhibit a declining growth rate and that the small fish are simply poor performers.

The production rates achieved in this programme suggested that one hectare of duckweed production can support two hectares of carp polyculture. Empirical results suggested that a polyculture stocked at about 30 000 fish/ha may be fed as much duckweed as they will eat daily, regardless of the season. Fish were fed duckweed throughout the day. Freshly harvested duckweed was brought in baskets to the pond and distributed evenly among several 'feeding rings or squares' (Figure 3.7) consisting of 4 m² open-bottom enclosures. Feeding rings provide access by the fish to the duckweed and prevent it from dispersing over the pond surface. The feeding ring can be a

floating enclosure anchored near the shore. Six feeding rings/ha were installed in the Mirzapur experimental site and appeared to provide sufficient access to food for all fish. Figures 3.8 and 3.9 show the transport and utilization of duckweed in fish culture.

In the Mirzapur experimental ponds, grass carp was the primary consumer of duckweed in the polyculture. However, both catla and common carp also competed aggressively for available duckweed feed and consumed it directly.

FIGURE 3.7

Duckweed cultivated in an undrainable pond (Mymensingh, Bangladesh)



FIGURE 3.8

A duckweed collector carrying a bag of duckweed in a rickshaw van. These professional duckweed collectors collect duckweed from various derelict ponds and sell them to fish farmers (Jessore, Bangladesh)

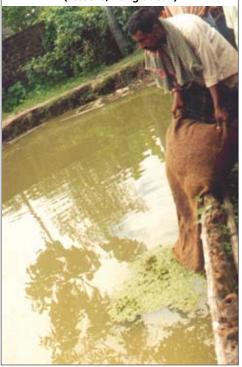


Top-feeders directly absorb about percent of duckweed nutrients in their digestive systems. Their faeces contain the balance of the original duckweed nutrients and furnish relatively high quantity detritus to the bottom-feeders. A duckweed-fed fish pond thus appears to provide a complete, balanced diet for those carp that consume it directly, while the faeces of duckweedfeeding species, which are consumed directly by detritus feeders or

indirectly through fertilization of plankton and other natural food organisms, provide adequate food for the remaining bottom and mid-feeding carp varieties. The fertilization of a duckweed-fed fish culture is therefore indirect and gradual, resulting from bacterial decomposition of fish faeces, dead algae, and other fermenting organic material.

FIGURE 3.9

A farmer is releasing a bag of duckweed to his carp pond. These duckweeds are generally purchased from a group of professional duckweed collectors (Jessore, Bangladesh)



Skillicorn, Spira and Journey (1993) reported that the first annual cycle of carp production produced slightly more than 10 tonnes/ha/year. However, these authors opined that a yield of between 10 to 15 tonnes/ha/year appears to be sustainable before biological constraints become limiting factors.

DWRP (1998) reported further follow-up of the duckweed-fed carp polyculture system developed by PRISM in Bangladesh. This report included the results of the demonstration farms as well as the results of the farmers' ponds. The duckweed-fed carp polyculture system practised by PRISM had two distinct differences from the model described by Skillicorn, Spira and Journey (1993). Apparently, PRISM included Nile tilapia with the traditional six-species carp culture system and provided other supplemental feed along with the duckweed. The farmers that adopted duckweedbased aquaculture produced an average of 3.6 tonnes/ ha/year in comparison to the national average fish production of 2.1 tonnes/ha/year in Bangladesh at that time. PRISM itself achieved a production level of 11 tonnes/ha/year in 1993 and 16 tonnes/ha/year in 1996 in its demonstration farm.

The results of the duckweed-based carp polyculture of PRISM are presented in Tables 3.16, 3.17 and 3.18. Table 3.15 presents fish stocking and harvesting data in 1994. Tilapia were not stocked but multiplied on their

own. Data for feed application, fish yield and food conversion ratio in farmers' ponds and in demonstration farms of PRISM are presented in Tables 3.16 and 3.17. The specific influence of duckweed feeding on yield and food conversion was obscured because other supplemental food was also added to the fish ponds. In neither demonstration ponds nor farmers' ponds could any example be found of pure duckweed feeding.

Table 3.17 presents data collected from demonstration farm production in Mirzapur over a period of three years. The results from fish ponds fed duckweed from organic wastewater plants have been kept separate from those fed on duckweed grown on chemical fertilizers. Whether there has been any difference in nutritive value between the duckweed from these different treatments could not be checked. What is interesting is to compare the difference in the ratio of duckweed to the other supplemental feed that was being applied. At first the information in Table 3.17 suggests that higher proportions of duckweed influence the conversion rate adversely. This conclusion may not be true, however, since the total amount of food applied in the ponds treated with waste-grown duckweed was clearly too high. Since the ponds did not show any increased production with a high rate of feeding, it must be assumed that they were at their carrying capacity most of the time and that all the extra food offered was apparently wasted. This seems to imply that the sustainable level of fish production from a duckweed-based polyculture lies around 10-15 tonnes/ha/year.

TABLE 3.15

The ratio of fish species stocked and harvested by PRISM in 1994

Species	Stocking rate (percent)	Harvest rate (percent)
Tilapia	0	38.8
Catla	20	6.7
Rohu	20	9.7
Mrigal	20	9.3
Silver carp	15	24.3
Grass carp	20	7.3
Common carp	5	3.3
Other	0	0.6

TABLE 3.16

Feed application, fish yield and food conversion ratio in farmers' ponds in two locations in Bangladesh during 1995-96

Location	Oil cake (kg/ha DM)	Wheat bran (kg/ha DM)	Duckweed (kg/ha DM)	Total (kg/ha DM)	Fish yield (kg/ha)	FCR
Tangail	2 742	1 441	1 526	5 833	3 290	2.1
Manikganj	2 556	1 854	1 465	5 874	5 007	1.5

Source: DWRP (1998)

TABLE 3.17

Feed application, fish yield and food conversion ratio in demonstration ponds at Mirzapur Experimental Site, Bangladesh during 1993-95

Nutrient source of duckweed	Season	Oilcake (kg/ha DM)	Wheat bran (kg/ha DM)	Duckweed (kg/ha DM)	Total (kg/ha DM)	percent duckweed used in feed	Fish yield (kg/ha)	FCR
Chemical	1993	8 504	1 065	6 662	16 231	41	13 430	1.2
	1994	11 722	507	5 902	18 131	33	15 080	1.2
	1995	12 107	122	5 810	18 039	32	11 520	1.6
Wastewater	1994	9 810	-	19 840	29 650	67	10 580	2.8
	1995	18 307	-	23 300	41 607	56	12 620	3.3

Source: DWRP (1998)

The food conversion values obtained when various duckweed species were fed to different fish species are presented in Table 3.18. Duckweeds were fed mostly in the fresh form and most of the values available are for grass carp and Nile tilapia. The

values are variable, but the available data does not indicate if the variability was due to the fish species or to the duckweed species used. Generally, most FCRs are between 2.0 and 3.0, although an FCR of 1.0 was reported for hybrid tilapia and a very high FCR (6.7) for hybrid grass carp when both were fed *L. gibba*. This latter result was probably due to size of the fish (>1.0 kg) used in the feeding trial. Shireman, Colle and Rottmann (1978) reported an FCR of 1.6 for 2.8 g grass carp when fed fresh *L. minima* but the FCR was 2.7 for 63 g fish. Hassan and Edwards (1992) reported that food conversion was significantly affected by the feeding rate. For example, the FCR was 3.1 when *S. polyrrhiza* was fed to Nile tilapia at a feeding rate of 2.5 percent BW/day whereas it was 5.9 at a feeding rate of 5.0 percent BW/day. Similarly, FCR increased from 2.2 to 3.7 with an increase in feeding rate from 2.5 to 5.0 percent BW/day for Nile tilapia when fed *L. perpusilla*.

Generally, the FCR values reported for duckweed-based polyculture of carps in Bangladesh (Tables 3.16 and 3.17) were very good, being between 1.5 and 2.1 for farmer's ponds and 1.2 to 1.6 for demonstration ponds. However, it must be pointed out that duckweed was not used as the sole feed in these ponds, which were usually fertilized in addition to the use of oilcake, rice bran and wheat bran as supplemental feeds. Low FCRs for duckweed may be expected, since these plants have relatively low fibre and high protein contents (Table 3.12) and a good amino acid profile (Annex 1 Table 3). Although it is difficult to generalize from the available data, an FCR value of 2.5 may be a reasonable expectation for grass carp and Nile tilapia based.

TABLE 3.18

Food conversion ratios of duckweed to fish

Duckweed	Fish species	Fish size (g)	Food conv ratio (F		Reference
		-	DM <sup>2</sup>	FW <sup>2</sup>	
L. gibba, fresh	Tilapia hybrid	2.7	1.0		Gaigher, Porath and Granoth (1984)
L. gibba, fresh	Hybrid grass carp	1 015	6.7		Cassani, Caton and Hansen (1982)
L. perpusilla, fresh	Nile tilapia	8-10	3.7	60.6	Edwards, Pacharaprakiti and Yomjinda (1990)
L. perpusilla, fresh	Nile tilapia	26-27	2.2-3.7		Hassan and Edwards (1992)
L. perpusilla, fresh	Nile tilapia	40-44	1.6-1.9		Hassan and Edwards (1992)
L. minima, fresh	Grass carp	2.7	1.7-2.0		Shireman, Colle and Rottmann (1977)
L. minima, fresh	Grass carp	2.8	1.6		Shireman, Colle and Rottmann (1978)
L. minima, fresh	Grass carp	63	2.7		Shireman, Colle and Rottmann. (1978)
L. minor, fresh	Nile tilapia	n.s.		33	Rifai (1979)
L. minor, dried	Nile tilapia	2.5	2.1		Zaher et al. (1995)
L. minor, dried	Common carp	3.0	3.1		Devaraj, Krishna and Keshavappa (1981)
Lemna sp., fresh	Grass carp	n.s.		37	Hepher and Pruginin (1979)
S. polyrrhiza, fresh	Nile tilapia	26-28	3.1-5.9		Hassan and Edwards (1992)
S. polyrrhiza, dried, 30 percent inclusion	All-male tilapia	13.9	2.0		Fasakin, Balogun and Fasuru (1999)
W. arrhiza, fresh	Six carp species <sup>1</sup>	5.0-15.5	5.6	78.8	Naskar <i>et al.</i> (1986)
W. columbiana, fresh	Hybrid grass carp	1 033	3.8		Cassani, Caton and Hansen (1982)
Mixture of <i>Lemna,</i> <i>Spirodela</i> and <i>Wolffia</i>	Grass carp	n.s.	1.6-4.1		Baur and Buck (1980)

<sup>&</sup>lt;sup>1</sup> Polyculture of six carp species (grass carp, silver carp, common carp, Java barb, rohu and mrigal)

<sup>&</sup>lt;sup>2</sup> FW = fresh weight basis; DM = dry matter basis

Digestibility coefficients of *Lemna*, *Spirodela* and *Wolffia* fed to grass carp, tilapia and rohu are presented in Table 3.19. Considering the importance of duckweed as fish feed, it is surprising to note that only a few studies have been carried out to investigate its digestibility for fish. Van Dyke and Sutton (1977) were probably the first to investigate the digestibility of duckweed (mixture of *L. minor* and *L. gibba*) in detail for grass carp. These authors estimated the true dry matter digestibility of duckweed to be 65 percent, while the apparent digestibility was 53 percent for dry matter, 80 percent for crude protein, 58 percent for organic matter, 26 percent for ash and 61 percent for gross energy. The dry matter digestibility of *L. gibba*, *S. polyrrhiza* and *W. arrhiza* for grass carp found by other authors (Table 3.19) varied between 67-82 percent, while the dry matter digestibility of *L. gibba* for hybrid tilapia was reported to be 65 percent. Grass carp passes its food rapidly through a short, unspecialized gut and the fish probably does not produce cellulase (Van Dyke and Sutton, 1977); it is therefore unrealistic to expect that more than 50-60 percent of the feed consumed would actually be digested.

TABLE 3.19

Digestibility of duckweed for selected fish species

Duckweed	Fish species	Fish size (g)			Digestib (percer	•			Reference
			DM	CP	EE	NFE	CF	GE	
L. gibba	Hybrid tilapia²	2.7	65	86					Gaigher, Porath and Granoth (1984)
L. gibba	Grass carp		82						Lin and Chen (1983, cited by Wee, 1991)
L. gibba and L. minor (1:1)	Grass carp	320	53	80				61	Van Dyke and Sutton (1977)
S. polyrrhiza	Grass carp		75						Lin and Chen (1983, cited by Wee, 1991)
W. arrhiza	Grass carp		67						Lin and Chen (1983, cited by Wee, 1991)
W. arrhiza	Rohu	3.6		91.5	93.5	81.2		84.4	Ray and Das (1994)

<sup>&</sup>lt;sup>1</sup> DM = dry matter; CP = crude protein; EE = ether extract; CF = crude fibre; NFE = nitrogen free extract; GE = gross energy

<sup>&</sup>lt;sup>2</sup> O. niloticus X O. aureus