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3. APPLICATION AND ECONOMIC IMPORTANCE

3.1. GENERAL

Lemnaceae have a few characteristics which make a versatile application promising:

- simple culture conditions (aseptic conditions possible, small need of space)
- vegetative multiplication (genetically uniform)
- high multiplication rate
- high protein content
- easy harvest possibilities
- high need of nutrients
- pronounced ability to accumulate heavy metals
- only few diseases

Accordingly, the main applicability must be looked for in the following directions:

- test plant for phytophysiological experiments and for detection of toxicants
- source of protein (food for animals)
- removal of surplus nutrients and heavy metals from (waste) water
- production of energy and phytochemicals
- regulator in aquatic ecosystems

A comprehensive survey on the economic possibilities of Lemnaceae was done by RUSKIN and SHIPLEY (1976) in a wide-spread booklet on "Making aquatic weeds useful: some perspectives for developing countries" with an annex on duckweeds and their uses. Since then, many other compilations on the applicability of duckweeds have been presented: HILLMAN and CULLEY (1978a), STEPHENSON et al. (1980), EDWARDS (1980), CULLEY et al. (1981), AUGSTEN (1984a,b), BJOERNDahl (1984), HUBAC et al. (1984), PORATH et al. (1986). Earlier reviews can be found in HILLMAN (1961a) and SCHULZ (1962).

Lemnaceae are not always beneficial to man. In some cases when they cov-

er a water with a carpet centimeters deep, they may become a nuisance for motor-boating and fishing or for bathing. A typical action against duckweed invasion is described by HARGROVE (1976). The "Operation Duckweed" was performed with success in Alabama, USA, 70 ha of a water system was sprayed with 1 kg/ha diquat and 0.5 kg/ha copper. This action must be repeated at least once a year in order to keep the water free of a duckweed cover. Even if this method is effective and relatively inexpensive, it is ecologically not appropriate. The same is true for the frequent use of herbicides in fish ponds to achieve a better aeration of the water. The mass development of Lemnaceae certainly is a consequence of a high nutrient content in the water. Very often this is the result of industrial, agricultural or domestic pollution (especially with phosphorus and nitrogen). The high nutrient content should be counteracted by purification of the inlet water or by harvesting the Lemnaceae to lower the nutrient concentration in the water (see chapter 3.5).

3.2. BIOMASS, PRODUCTIVITY, ENERGY CONTENT AND NUTRITIVE VALUE

3.2.1. Productivity

The maximum growth rate in Lemnaceae is species and clone specific (LANDOLT 1957, REJMANKOVA 1975a, PORATH et al. 1979). The highest growth rate of Lemnaceae corresponds to about a doubling time of 24 hours. This was noted in L. aequinoctialis (LANDOLT 1957, CHANG et al. 1977, DATKO et al. 1980a), and in W. microscopica (VENKATARAMAN et al. 1970). This highest growth rate results in an increase of one gram per gram dry weight and day, or 64 grams per gram dry weight and week. In comparison, fast growing corn does not produce more than 2.3 g/g and week, according to HILLMAN and CULLEY (1978a). For S. polyrrhiza, the growth rate at high temperatures is not much below that of L. aequinoctialis. In this species, the dry weight amounts to at least 2 mg/cm^2 under optimal conditions. If we assume a doubling time of 24 hours and a completely covered water surface, the theoretical increase would be 20 g/m^2 and day or 73 t/ha and year, provided that the conditions are optimal during the whole year. This theoretical yield is never reached in nature, because temperature and nutrient condition are nowhere equally good at all seasons. However for shorter times, these values have been matched at least to 1/3-2/3 (LANDOLT 1957, SAHAI and SINHA 1970, PORATH and KOTON 1977, SAID et al. 1979, CORRADI et al. 1981, REJMANKOVA et al. 1983, MESTAYER et al. 1984, ORON et al. 1985, 1987, HENDERSON et al. 1984, and many others, see also table 3.1). Values of 1 to 15 g/m^2 and day have been achieved. Growth rate of Lemnaceae in axenic cultures is maximum at low frond densities (e.g. 10 g/m^2 dry weight) (REDDY and DEBUSK 1985a,b). The production of fixed carbon was calculated by WARD et al. (1963). The authors noted 7 mg carbon/g dry weight and hour in S. polyrrhiza under optimal nutrient conditions and at 12000 lux. WOHLER (1966) measured in pond water at 10000 lux $1.6 \text{ mg g}^{-1} \text{ h}^{-1}$ carbon in S. polyrrhiza, $1.4 \text{ mg g}^{-1} \text{ h}^{-1}$ in L. minor, $1.9 \text{ mg g}^{-1} \text{ h}^{-1}$ in L. trisulca and $2.7 \text{ mg g}^{-1} \text{ h}^{-1}$ in W. gladiata. The differences in the carbon content per dry weight are probably due to differences in assimilation area per dry weight for different species. FILBIN and HOUGH (1985) measured $2.5 \text{ mg g}^{-1} \text{ h}^{-1}$ carbon in L. minor during the growing season.

SAID et al. (1979) in Louisiana during summer time found 44 t duckweed

Table 3.1. Productivity of Lemnaceae in different regions of the world

* 0,65 t ha⁻¹ in January and 3.4 t ha⁻¹ in August

Region	dry weight in t ha ⁻¹ yr ⁻¹	authors
southern states of the USA	19.2 14.5 - 27 23.3* 13.5 16.1	STANLEY and MADEWELL (1975) MYERS (1977) CULLEY and MYERS (1980) DEBUSK et al. (1981) REDDY and DEBUSK (1985b)
Israel	39 10	HEPHER and PRUGININ (1979 cited from EDWARDS 1980) PORATH et al. (1979)
Egypt	10	EL-DIN (1982)
India	22	RAO et al. (1982)
northern Thailand	10.5	BHANTHUMNAVIN and McGARRY (1971)
Uzbekistan	7 - 15	TAUBAEV and ABDIEV (1973)
GDR	16 (calculated)	SCHULZ (1962)
CSSR	7.5 - 8	REJMANKOVA (1975b, 1979)

(dry weight) calculated per ha and year which is nearly 2/3 of the theoretical maximum. The actual productivity in nature during a whole year is 8-30% of the maximal value (table 3.1). A mixture of different Lemnaceae species might reach higher values because then differently adapted clones are available for different seasons and different layers of the water can be utilized. For a high productivity in nature, the Lemnaceae should cover the water completely to avoid growth of algae (DEBUSK et al. 1981).

In the first three regions of table 3.1, the productivity is reduced and in the last three regions it is stopped during the winter. In India and Thailand the temperatures are mostly optimal but during the rainy season growth is slowed down due to a lack of nutrients.

KOLES (1986) developed a model to predict growth rates of duckweeds under various environmental conditions and to predict changes in water nutrient conditions accompanying the growth of these plants.

3.2.2. Biomass

The biomass of Lemnaceae in nature is relatively low compared with other water plants and with terrestrial plants. This is due to the special growth habit: Lemnaceae only form rather thin layers of fronds and are composed of relatively light assimilating and aerenchymatic tissue. Highest values of the dry weight of a single frond amounted to 0.6 mg in S. polyrrhiza (LANDOLT 1957); the lowest weight of a frond was found in W. columbiana (c. 0.008 mg, measured as 0.15 mg fresh weight by ARMSTRONG 1982).

The following values of biomass (in g dry weight per m²) during the summer have been reported: 44-56 (MOORE 1962, 1965), 50-150 (IKUSIMA 1963b), up to 192 (JERVIS 1969), 70-180 (REJMANKOVA 1978, 1982), up to 280 (EWEL and ODUM 1978), up to 250 (HEJNY et al. 1981), 114 (SASTROUTOMO 1982), up to 180 (GHETTI et al. 1982), 20-184 (KUECHLER 1986), up to 220 (GEARHEART et al. 1986). In comparison, stands of Phragmites develop 1612 g/m² (KUECHLER 1986) or even up to 9860 g/m² (HEJNY et al. 1981) which is at least 40 times higher than the biomass of a duckweed cover. The biomass of Lemnaceae in waters which contain few nutrients is much lower. A Lemno-Utricularietum consists of 10 g/m² dry biomass (HEJNY et al. 1981), a "Lemnetum" 12 g/m² and 7 g/m² (KLOSE 1963 and VARFOLOMEEVA 1976, resp.). McLAY (1976) noted that S. punctata is able to develop a

Table 3.2. Number of fronds of Lemnaceae per m² water surface in nature

Species	number/m ²	author
<u>S. polyrrhiza</u>	10000-29000	KAUL and BAKAYA (1973)
<u>Lemna</u> sp.	40000 140000	KLOSE (1963) GHETTI et al. (1982)
<u>L. gibba</u>	16000-89000	KAUL and BAKAYA (1973)
<u>L. minor</u>	18000-84000	KAUL and BAKAYA (1973)
<u>L. trisulca</u>	10000-29000	KAUL and BAKAYA (1973)
<u>W. columbiana</u> , <u>W. borealis</u>	2000000	HICKS (1937)

2.5 times higher biomass than L. minor and a 17 times higher biomass than W. arrhiza.

Optimum stocking density for production was found to be 20 g (dry weight) /m² (DEBUSK et al. 1981) and 120 g/m² (PORATH et al. 1979), both for L. minor.

Numbers of fronds per m² were measured by different authors. The results have been put together in table 3.2. KLOSE (1963) counted fronds in Germany, GHETTI et al. (1982) in Italy, KAUL and BAKAYA (1973) in Kashmir (India) and HICKS (1937) in Indiana (USA).

According to RUSKIN and SHIPLEY (1976), a layer of L. minor may expand within 53 days from 6.4 cm² to 0.5 ha.

3.2.3. Energy content

The energy content of the Lemnaceae belong to the highest within water plants (STEUBING et al. 1980, see table 3.3). However, since the energy content is much dependent on the growth conditions, it is not clear if there are differences in the energy content between different species

Table 3.3. Energy content of different Lemnaceae

species	energy content in J/g	author
S. polyrrhiza	9660 15500-17090	SINGH and SHARMA (1975) SUTTON and ORNES (1977)
S. punctata	13880-17640 16840	SRIVASTAVA (1978) MESTAYER (1980)
L. minor	16920	MUZTAR et al. (1978a)
L. aequinoctialis	15360-16970	SRIVASTAVA (1978)
L. minuscula	15330	STEUBING et al. (1980)
Egeria densa	9310	STEUBING et al. (1980)
Juncus procerus	17380	STEUBING et al. (1980)

(table 3.3). SRIVASTAVA (1978) reports of somewhat higher energy content of dry weight during the cold season (average temperatures between 13°C and 19°C) in India than during the hot season. This is probably due to a higher starch content of the Lemnaceae frond at lower temperatures.

According to TRUAX et al. (1972), 4536 J/g of the energy content of the Lemnaceae are utilizable by animals. This corresponds to about 1/2-1/3 of the total energy content.

S. punctata has a mean solar energy conversion efficiency of 4.2% (highest value for a single sample up to 4.9%) of the photosynthetically active radiation (MESTAYER et al. 1984). Chapter 3.7.1 deals with the use of Lemnaceae as a source of energy.

3.2.4. Content of proteins and other nutritive substances

The high protein content of Lemnaceae has already been indicated in chapter 1.2.2. It reaches 15 to 45% of the dry weight. Starvation cultures sometimes contain only as little as 7% protein. After addition of an optimal amount of nutrients to the medium, the protein content is enhanced to 30% within two weeks (CULLEY et al. 1981). The lower limit of nitrogen content in the solution resulting in a high percentage of protein is 20-30 mg/l (FRYE and CULLEY 1980). The frond density in cultures of L. minor has no influence on the specific protein content nor on the cell wall components provided the nutrient supply is sufficient (TUCKER 1981).

Not only the quantity but also the quality of proteins makes the Lemnaceae economically interesting. With the exception of tryptophane and methionine, all essential amino acids used in human and animal food are satisfactorily present. Tryptophane is detected only in traces. Methionine content varies between 0.3% and 3% of the total protein, depending on investigated clone and author (PORATH et al. 1979, AMADO et al. 1980, RUSOFF et al. 1980 and further authors, see chapter 1.2.2). CHANG et al. (1977) analysed as much as 3.1-4.7% methionine, a value which would satisfy the requirements of the FAO. Possibly, the production of methionine can be improved by application of suitable nutrient solutions. Table 3.4 gives a survey of the protein content of Lemnaceae (according to AMADO et al. 1980) and several other plant and animal products.

The essential amino acid index (EAA) amounts to 76 for Lemnaceae (WUEST-

Table 3.4. Content of essential amino acids (given in % of total protein content) of Lemnaceae (mean of 94 investigated clones calculated from AMADO et al. 1980), rice, soybean, Chlorella, and egg compared to FAO reference pattern. The need of amino acids (in % of total feed) for young pigs and chickens is also given.

amino acids	amino acid content						
	in % of total protein						in % of total feed
	duckweed 1)	rice 2)	soybean 1)	Chlorella 1)	egg 3)	FAO 2)	
lysine	6.8	3.2	6.4	7.8	7.2	4.2	7
threonine	5.0	3.8	3.7	3.4	4.9	2.8	5
valine	6.6	6.2	4.9	5.8	7.3	4.2	5
methionine	1.0	3.4	1.4	2.0	4.1	2.2	5
leucine	9.6	8.2	7.4	4.0	9.2	4.8	6
isoleucine	4.8	5.2	4.7	3.6	8.0	4.2	5
phenylalanine	5.9	5.0	4.9	4.8	8.0	2.8	5
tryptophane	traces	1.3	1.3	1.5	1.2	1.4	1
arginine	6.7						2
histidine	2.0						2
							4

1 AMADO et al. 1980 2 RUSOFF et al. 1980 3 CHANG et al. 1977 4 MATSUMOTO 1981

LING and BOEHM 1980). In comparison, the EAA for eggs is reported as 97. The protein efficiency ratio for Lemnaceae fed to grass carp reached 2.36 (HAJRA and TRIPATHY 1985).

Considering the high productivity and the high protein content of Lemnaceae, the protein yield per area must be higher than that of any other crop plant. If we assume a maximum yield of 50 t dry weight per ha and year and a mean percentage of protein of 30% of the dry weight, we can expect a protein harvest of up to 15 t per ha and year. Accordingly, the results of ORON et al. (1987a) indicate that protein yield of duckweed grown in waste water systems may reach 12 t per ha and year. Soybean, belonging to crops richest in protein, yields maximum up to 0.7 t protein per ha and year which is about 1/20 of the potential yield of Lemnaceae. Even under suboptimal conditions, the Lemnaceae are able to produce a great amount of protein and also of carbohydrates. BHANTHUMNAVIN and MCGARRY (1971) give examples of yearly yields of different crop plants in northern Thailand of protein, carbohydrate and fat (table 3.5), compared with W. globosa (named as W. arrhiza).

SAID et al. (1979) estimate that the production of the same amount of protein needs a ten times greater cultivation area for soybeans than for Lemnaceae and a 2.5-9 times greater area for alfalfa. MATSUMOTO (1981) calculated the total digestible crude protein in S. polyrrhiza as 23.5% of the dry weight and the total digestible nutrients as 44.2%.

Table 3.5. Content of protein, carbohydrate and fat in different crop plants from northern Thailand (from BHANTHUMNAVIN and MCGARRY 1971)

	% dry weight	kg ha ⁻¹ yr ⁻¹				
	Wolffia	Wolffia	soya	nuts	rice	corn
protein	19.8	2080	303	229	71	179
carbohydrate	43.6	4589	255	164	849	1451
fat	5.0	533	158	397	4	87
fibres	13.3	1398	44	21	3	40
ash	18.3	1928	41	20	5	24

Vitamins A, B₁, B₂, B₆, C, E and PP are present in Lemnaceae (see chapter 1.2.10). Especially the content of vitamin E (0.02-0.04 mg per g fresh weight) and of vitamin PP (0.04-0.06 mg per g fresh weight) are remarkable (MUZAFFAROV et al. 1971).

3.3. CULTIVATION AND HARVEST

3.3.1. Cultivation

The cultivation methods are greatly dependent on the intended utilization of the Lemnaceae.

For physiological tests, aseptic cultures (as described in chapter 2.2.3) are necessary.

For cultivation in the field, the following guidelines may be useful:

- The cultivation area must be protected against wind in order to prevent an accumulation of the fronds in one corner; Lemnaceae should be able to cover the whole illuminated water surface evenly; larger areas should be divided into smaller ones; shores must preferably be steep to prevent the fronds from being washed ashore; water fluctuations should also be avoided.
- Since Lemnaceae are only able to utilize nutrients from the upper water layers, shallow waters with slow water circulation are best for growth; the optimal depth depends on the climatic conditions and on the intended utilization; in cool regions shallow water warm up faster but may not be favourable to survival in winter.
- Most Lemnaceae grow much slower at temperatures below 20°C; in regions where temperatures often remain under this point, species should be chosen which grow well in cooler waters: e.g. L. gibba, L. minor, L. trisulca, and S. punctata. CULLEY et al. (1978) recommend for Louisiana with a change of warm and fairly cool seasons a mixture of S. polyrrhiza, S. punctata, and L. gibba. Some species are not suited for warm regions (e.g. L. trisulca, L. minor), others do not grow in regions where temperatures often drop below 10°C (e.g. Wolffiella Welwitschii, W. neotropica, W. hyalina, W. arrhiza, L. aequinoctialis, S. polyrrhiza). Species which do well in warm regions are: S. polyrrhiza, L. aequinoctialis, W. microscopica, W. angusta, W. globosa.
- Lemnaceae only grow in waters relatively rich in nutrients; in waters where the conditions are not constantly optimal, a mixture of different duckweed species is preferable.
- The density of the Lemnaceae cover is important for maximum yield. MUZAFFAROV et al. (1971) received best yield with a density of 500 g fresh weight (corresponding to 25-30 g dry weight) per m². REJMANKOVA

(1978) and REJMANKOVA et al. (1983) noted 15-25 g dry weight per m^2 as optimal stocking density. DEBUSK et al. (1981), REDDY and DEBUSK (1985b) used the following operational plant densities in their experiments: 10-88 g m^{-2} for S. polyrrhiza and 10-120 g m^{-2} for Lemna sp. Growth rate was maximum at the lower plant densities used. Cultivation plants are described by REJMANKOVA (1978), SAID et al. (1979), CULLEY and MYERS (1980), and CORRADI et al. (1981).

3.3.2. Harvest and processing

Pure cultures of Lemnaceae are relatively easy to harvest. The fronds can be skimmed off by some kind of net, or they can be collected at the outlet of the water by a grid. KOBAYASHI et al. (1977) developed an orifice type screen for harvesting Lemna in an irrigation channel. The highest yield is achieved if the close but not many-layered Lemnaceae cover is gathered at short intervals. According to SAID et al. (1979) and CULLEY and MYERS (1980), the daily harvest is more advantageous than the weekly removal (23.3 t dry weight per ha and year against 17.6 t). REJMANKOVA et al. (1983) developed best harvest strategy at 1 to 2 day intervals (800 g dry weight per m^2 in 90 days compared with 600 g if harvested every 14 days). Differently, RYTHER et al. (1980) and DEBUSK et al. (1981) did not get a higher production if harvested every day compared with every 5 to 10 days. In northern Thailand, W. globosa is gathered every 3 to 4 days, an interval which proved to be favourable after many years of experience (BHANTHUMNAVIN and MCGARRY 1971). The different results of different authors are certainly due to different techniques applied. In general a shorter harvest interval which is much better suited for an evenly optimal utilization of sunlight must result in more productivity.

Drying is necessary to store the Lemnaceae yield. According to SCHULZ (1962), Lemnaceae become dry on a sunny day in Central Europe within 10 hours if turned over several times. Without the periodic turning over the drying takes much longer than for making hay, and it is only possible if no rain or strong wind occurs (CULLEY and EPPS 1973). The drying of a 5 cm thick layer of S. punctata at 100°C takes about 10 hours (LAWSON et al. 1974). At this temperature, some of the proteins are lost. Also PORATH and KOTON (1977) point out the fact that the content

of digestible proteins and amino acids is diminished during heat-drying. If the water is pressed out mechanically, a 66-71% loss of proteins is recorded by LAWSON et al. (1974).

BALDWIN and MYERS (1980) describe harvest methods of Lemnaceae for feeding cattle. From the skimmer, the Lemnaceae reach a drop box. After partial dehydrating and chlorinating they are transported to storage, drying facilities, or feed bins. The treatment with hypochlorite is recommended to lower the content of bacteria (AMBORSKI and LARKIN 1980).

Little is known about the possibilities of silage of Lemnaceae. According to a survey of EDWARDS (1980), the silage of water plants (e.g. Eichhornia) did not prove to be good because the water content is too high. Therefore it is necessary either to pre-dry the material or to add some concentrated organic substances (e.g. carbohydrates). EVERSULL et al. (1980) succeeded however to silage Lemnaceae together with a high dry matter corn crop.

3.4. UTILIZATION AS FOOD PLANT

3.4.1. General remarks

As pointed out earlier, Lemnaceae are easily harvested, have extended growing and harvesting periods, high protein and low fibre and lignin content and only very few and not severe pests (CULLEY and EPPS 1973). Therefore, they are generally suited as a food plant. The application for human nutrition and as an animal feed has been known for a long time. Lemnaceae ("duckweed") are favoured as a fodder plant for fish and birds in many countries. They are on offer at markets, e.g. in Mexico for fowl (according to a personal communication from M. Seidl, Greifensee) or in Taiwan where they are sold at 1 \$ for 10 kg wet weight as foodstuff for fish and duck (T.P. CHEN in EDWARDS 1980). In eastern Asia (Northern Thailand, Burma, Laos) W. globosa (named as W. arrhiza is cultivated under the name khai-nam ("eggs of the water") for many generations and sold at markets (BHANTHUMNAVIN and MCGARRY 1971).

It is generally assumed that Lemnaceae will soon become more important as a crop plant (e.g. KRUGMANN-RANDOLF 1978). Extensive investigations are being made with Lemnaceae as protein suppliers in many regions of the world.

3.4.2. Human nutrition

The high content of protein, carbohydrate, and vitamins grant to Lemnaceae an outstanding nutritional value also for man. NAKAMURA (1960) mentioned the utilization of Wolffia as human food. In eastern Asia, W. globosa has been eaten by man for many generations (BHANTHUMNAVIN and MCGARRY 1971). The plants are cultivated in ponds of up to 100 m² area which are supplemented by rain water and shaded by bamboo. No artificial fertilizer is supplied. Every 3 to 4 days part of the Wolffia cover is harvested and eaten as a vegetable. The species flowers during the monsoon between August and October and is then considered not wholesome. Possibly, the growth is very slow during that time, due to the very diluted water. Cultivating of W. globosa in this way yields 2 t protein, 4.5 t carbohydrate, and 0.5 t fat per ha and year. NAKAMURA (1960) re-

ports that the taste of Wolffia is excellent and sweet, resembling that of cabbage.

It is rather astonishing that Lemnaceae which are so wide-spread have not been used as human food in other regions. R.A. ALBERTS (Springfield, Virginia, USA, in lit. 1979) assumes that the Mayas in Guatemala used Lemnaceae as foodstuff under the name of Xim Ha ("water corn"). He thinks that in expansive irrigation systems the Mayas were able to nourish the numerous people living in a relatively small area with the productive Lemnaceae.

The explanation that the use of Lemnaceae as a human food is restricted might be the following:

- 1) Lemnaceae and especially Spirodela and Lemna contain a great amount of oxalic acid, partly in solution and partly crystallized. They are therefore not very agreeable in taste (rather harsh). It might be possible to prepare the plants by some treatment to make them tasty and desirable. SUTTON (1981a) describes an edible L. gibba salad, and NEY (1960) states that L. minor is far superior in taste to Chlorella. It is evident that Wolffia and Wolffiella which have the oxalate in the free form are more suited for food than Spirodela and Lemna.
- 2) The harvested Lemnaceae are difficult to separate from other organisms such as snails, insects, worms, protozoa, algae and bacteria. There is also a certain danger of infection by pathogens when eating Lemnaceae that have been cultivated in waters polluted by waste water. AMBORSKI and LARKIN (1980) demonstrated that during the warm season in Louisiana one liter water of a pond covered with Lemnaceae and supplied with sewage of cattle contained 10^8 coliform bacteria (of which 10^6 were fecal), 10^4 fecal Streptococcae and up to 300 Salmonella and Shigella germs. Also pathogenic viruses could be detected. A treatment of 20 minutes with hypochlorite resulted in a marked reduction of the bacteria content. NGUYEN (1978) detected trematoda in Lemnaceae covered waters which were in connection with a pig-breeding plant. However the toxic blue alga Anabaena never showed up in waters with a Lemnaceae cover (KELLY 1980). Further investigations on safe cultivation and harvest of Lemnaceae are needed.

Another possibility to utilize Lemnaceae for human food is either via extraction of proteins (cf. RUSOFF et al. 1980) or in an indirect way via animal production. Certainly, this last possibility reduces the efficiency factor of the Lemnacean energy. CULLEY et al. (1981) cal-

culated a need of 50 m² pond area to nourish a family of 5 people via egg protein. To produce the 90 to 100 kg protein needed by the family each year, the egg producing chickens eat 40 kg of Lemnaceae protein and in addition 14 kg of animal protein (from worms and insects living in the soil of the surrounding area). The protein supply via fish still seems more favourable (see chapter 3.4.5.1).

3.4.3. Food for mammals

CULLEY et al. (1981) give a survey of all investigations made with Lemnaceae for use in farming.

3.4.3.1. Dairy heifers

RUSOFF et al. (1977, 1978) investigated the possibilities of using duckweed as a nutrient for Holstein cattle. The animals are able to take successfully more than 75% of fodder in form of Lemnaceae. The milk does not change in taste. Calves of 150 to 300 kg weight which have been fed with 67% duckweed (dry weight) and 33% silage of corn, showed an increase in weight of 0.95 kg per animal and day. In comparison, animals which received concentrated food and corn silage in addition to pasturing, only resulted in an increase of 0.5 kg per animal and day. According to CULLEY et al. (1981), 3.1 ha water area with Lemnaceae are sufficient to obtain enough protein for 100 dairy cattle. FRYE and CULLEY (1980) describe installations, functioning, and costs of a dairy farm in Louisiana based on a recycling principle and using Lemnaceae as main food for the animals.

3.4.3.2. Pigs

SCHULZ (1962) studied the nourishment of pigs with Lemnaceae. If fed with 75% grist of rye, wheat and barley, 5% of oat grist, 6% of wheat bran, 8% of fodder yeast, 5% of fish meal and 1% of minerals, the daily increase was still lower (455 g) than with 500 g fresh Lemnaceae fed in addition (546 g). In other experiments, the difference was still higher (up to 300 g per day). GALKINA et al. (1965) report a 14-20% weight increase of pigs in Russia if 1 kg fresh Lemnaceae was added to the normal

feed. HILLMAN and CULLEY (1978a) give some further examples.

3.4.3.3. Ram and sheep

TAUBAEV and ABDIEV (1973) noted 27% additional weight increase in ram and 14% in sheep if 0.5 kg Lemnaceae was added daily to the normal nutrient. PORATH et al. (1985) used duckweed as a substitute for animal protein rich feed in diets of young lambs and Awasi sheep.

3.4.3.4. Horses

STEWART (1972) mentions that horses feed on Lemnaceae in Dal Lake (Kashmir). However, no feeding under controlled and supervised conditions has been done so far.

3.4.3.5. Rabbits

MATSUMOTO (1981) made some investigations with rabbits. If he fed them with S. polyrrhiza alone, the increase was only 20-46% that with conventional fodder. Feeding a mixture of Spirodela with honey sugar resulted in an increase of 35-56%. Highest increase was reached if conventional fodder was supplemented with Spirodela.

3.4.3.6. Nutria and muskrat

Nutria (Myocastor coypus) and muskrat (Ondatra zibethicus) which are raised for fur, eat Lemnaceae in great amounts (WARKENTIN 1968, JACOBS 1947). For these animals, duckweeds are of great nutritional value and are recommended as a food in nutria farms. On account of the low content of fibre, indigestions may occasionally occur (SZUMAN and SKRZYDLEWSKI 1980).

3.4.3.7. Mice

WILKS (1962) demonstrated that mice can be sustained for indefinite periods on a strict diet of duckweeds without losing their normal physiological activity.

3.4.4. Bird food

It is well known that water fowl, especially ducks, feed on Lemnaceae (cf. JACOBS 1947, SCHULZ 1962, MACKENTHUN et al. 1964). A survey of the literature on experiments is given by CULLEY et al. (1981). In addition of small amounts of duckweed (2-5%) to the normal feed results in an additional daily weight increase of chickens by 10 to 32% (MUELLER and LAUTNER 1954, MUZAFFAROV et al. 1968, NAPHADE and MITHUJI 1969-1970, TRUAX et al. 1972, TAUBAEV and ABDIEV 1973). Other authors could not achieve an additional weight increase if the normal fodder was partly (2.5-25%) replaced by Lemnaceae. An addition of 50% Lemnaceae even gave negative results compared with controls (MUZTAR et al. 1976, 1977, JOHRI and SHARMA 1980). Positive results with layers were achieved if 25% of Lemnaceae was added (WILLIAMS 1978). SCHULZ (1962) reports on successful application of Lemnaceae in fattening of ducks. MUELLER and LAUTNER (1954) and NIKOLAEVA (1956) received additional weight gain (10-23%) in ducks if 2% or 37% of the foodstuff was replaced by duckweed. YAMANI et al. (1978) noted a favourable effect of Lemnaceae in protein nutrition of poultry. Cairina scutulata, the white winged wood duck, especially when young, prefers Lemnaceae (LUBBOCK 1975 from HUBAC et al. 1984). Successful effects of duckweed nutrition on ducks was also reported by GERGEL et al. (1985). Two phasianids (chukar and partridge) were fed effectively with duckweeds for 28 days (DEGEN 1987).

DYLIK et al. (1979) calculated for Anas platyrrhynchos an assimilation index of Lemnaceae food of 0.64 which is, compared with other crops, very high.

An advantage of Lemnaceae for poultry food is the high content of carotinoids, especially carotene and xanthophyll, which favours the colouring of fat and skin of the birds (MUZTAR et al. 1979). Also the egg yolk is more intensely coloured when the birds are fed with Lemna minor (GRAF 1987).

One ha water area is sufficient to raise 4000 to 7000 chickens and ducks during a vegetation period (HARVEY and FOX 1973). REJMANKOVA (1981) calculated an area of 1 ha Lemnaceae cover as sufficient to produce protein for 480 ducks during the warm season.

3.4.5. Foodstuff for fish and other cold-blooded animals

3.4.5.1. Fish

Lemnaceae are a very valuable source of food for many fish. On the other hand, a closed cover of Lemnaceae may prevent the supply of oxygen to the water, thus resulting in unfavourable conditions for fish sensitive to low oxygen content. Trout, for instance, are not suited for raising in Lemnaceae ponds (WRIGHT 1973).

A distinct positive effect on prospering of the grass-carp or white amur (Ctenopharyngodon idella) is reported by many authors (VERIGIN 1962, GALKINA et al. 1965, NIKOLSKIJ and VERIGIN 1966, FISCHER 1968, OPUSZYSKI 1972, EDWARDS 1974, PRISHCHEPOV 1974, VARGHESE et al. 1976, SUTTON 1976, VOVK 1976, PORATH and KOTON 1977, ROTTMANN 1977, SHIREMAN et al. 1977, 1978, BAUR and BUCK 1980, MACEINA and SHIREMAN 1980, HAJRA and TRIPATHY 1985). In cultures with a mixture of L. gibba and L. minor, the weight of the fish was tripled (from 100 g to 300 g per fish) within 50 days (PORATH and KOTON 1977). According to VAN DYKE and SUTTON (1977), the grass-carp is able to use 65% of the dry weight of Lemnaceae for food: 61% of the gross energy content, 70% of the rough protein, 72% of the organic cell content, 30% of the organic parts of the cell wall. The food conversion rate on a dry weight basis of grass-carp feeding on Lemnaceae amounts to 1.6 for a fish of 3 g and 2.7 for a fish of 63 g. No other foodstuff (catfish chow, rye grass, or a mixture of both) was nearly as efficient (SHIREMAN et al. 1978). SUTTON (1976) measured a food conversion rate between 1.1 and 5.3, BAUR and BUCK (1980) between 1.55 and 4.07, and HAJRA and TRIPATHY (1985) between 3.10 and 3.15. Further food conversion rates, however on a wet weight basis, are listed in SINGH and SINGH (1967), MICHEWICZ et al. (1972), TAL and ZIV (1978), HEPHER and PRUGININ (1979) and EDWARDS (1980).

If we assume in a closed grass-carp - Lemnaceae ecosystem a mean food conversion rate of 3.0 and a mean yearly production of 50 t/ha, the yield of grass-carp per ha and year can amount to 25 t. If in a less favourable case the yearly duckweed production is only 10 t per ha and the food conversion rate 5, the fish production still achieves 2 t per ha and year. Hybrids of grass-carp with other species of the same genus Ctenopharyngodon show similar positive results as grass-carp if fed with Lemnaceae (DUTHU and KILGEN 1975, THERIOT and SANDERS 1975, CASSANI 1981, CASSANI et al. 1982).

Channel catfish (Ictalurus punctatus) was successfully fed with up to 20% Lemnaceae (dry weight) (ROBINETTE et al. 1980). Other fish which can be partly nourished with Lemnaceae are: e.g. common carp (Cyprinus carpio) (HICKS 1937, PORATH and KOTON 1977, PANICKER et al. 1985), common mullet (Mugil cephalis) (CHABRECK cited in CULLEY et al. 1981), goldfish (Carassius auratus) (CULLEY et al. 1981). Species of the genus Tilapia (or Oreochromis) partly eat Lemnaceae (T. rendalli: MANN 1967) and partly not (T. mossambica, T. aurea: BAUR and BUCK 1980). However, HENDERSON et al. (1984) got positive results by feeding T. aurea additionally with Lemnaceae. T. rendalli is able to use 42-55% of the protein of S. polyrrhiza and 52-68% of the crude fibre (MANN 1967). KIM and KHANG (1982) received a 12% weight increase in red Tilapia fingerlings by supplying them additionally with Lemnaceae. According to HENDERSON et al. (1984) and HECKMANN et al. (1984) Tilapia grows rapidly in tanks containing duckweed only. They used water from zero discharge power plants to cultivate the Lemnaceae. Tilapia hybrid (Oreochromis niloticus x O. aureus) was fed by GAIGHER et al. (1984) with L. gibba in a recirculating unit. Intake rate was low and food conversion rate good (1:1), relative growth rate poor (0.67% of body mass per day). 65% of the duckweed consumed was assimilated and 26% converted to fish. EHRLICH (1966) proposed a polyculture of duckweed and Daphnia with the duckweed providing shade for Daphnia. These two organisms have a similar turnover rate and could be harvested as fish food simultaneously with the same equipment.

Often, Lemnaceae are not used as direct food for fish (and prawns) but via small organisms feeding on duckweed. The relations between Lemnaceae and other organisms are surveyed in volume 1, chapter 5.5.3 (LANDOLT 1986). In Bengal, Lemnaceae are cultured to increase the zooplankton on which carp feed. First, a phytoplankton bloom is established which disappears later when shaded by Lemnaceae. The Lemnaceae cover is subsequently removed in order to let the zooplankton which is nourished by the dying algae develop. The carp feed mostly on the zooplankton (ALIKUNHI et al. 1952). Also MALECHA et al. (1981) and HECKMANN et al. (1984) recommend the cultivation of Lemnaceae to increase the zooplankton for raising fish (e.g. carp or Tilapia) and prawns. In aquaculture of fish, L. gibba was shown to act as a biological ammonia stripper. A Lemnaceae mat was able to take up 80% of ammonia of a fish effluent in less than 48 hours (PORATH and POLLOCK 1982).

3.4.5.2. Crustaceae

L. minor is supposed to serve as a supplementary food source for the fresh water shrimp (Macrobrachium rosenbergii) (GODFRIAUX et al. 1975). HECKMANN et al. (1984) were able to raise the same prawn species in tanks containing duckweed only. Also MALECHA et al. (1981) report on Lemnaceae fed on by the shrimp. It is not investigated in detail if crayfish of the genera Oronectes, Procambarus and Cambarus feed partly or predominantly on Lemnaceae. However, it is known that Procambarus clarkii consumes fresh duckweed if offered (CULLEY et al. 1981). In Louisiana and California, crayfish are released in irrigated rice fields to keep weed (e.g. Lemnaceae) under control. After drainage of the water, the crayfish are collected and used as human food.

The ostracod Cypris spec. which is eaten extensively by fish feeds predominantly on Lemnaceae. Up to 90% of duckweed mat of 50 g fresh weight per m² has been eaten by Cypris within 96 hours at temperatures of 21-24°C (MANISSERY et al. 1981).

3.4.5.3. Turtles

The wide-spread aquarium turtle Pseudomys scripta feeds predominantly on Lemnaceae and Eichhornia. Therefore, commercial turtle farms cultivate Lemnaceae extensively as a foodstuff for the turtles (CHABRECK cited by CULLEY et al. 1981).

3.5. UTILIZATION IN WASTE WATER

3.5.1. General remarks

LUDWIG (1909) already pointed out the fact that Lemnaceae are able to tolerate a relatively high degree of pollution and that they are sometimes the only vegetation growing vigorously in dirty village pools and ponds. In contrast to many other water plants, Lemnaceae species (especially L. gibba) colonize polluted waters and are not impeded severely by a relatively high content of detergents (e.g. Lemnaceae tolerate more than 15 ppm tetrapropylene benzolsulphonate) (AGAMI et al. 1976). Even in waste waters with up to 500 mg COD (chemical oxygen demand) per liter, they still survive (GHETTI et al. 1982).

Organic waste water (sewage from households and farms) is produced in the whole world in great quantities and becomes a great ecological problem. The biological purification and the recycling of nutrients (especially nitrogen and phosphorus) to plant proteins would be highly preferable to the present technical waste water treatments applied in the industrial states. Lemnaceae are well suited for biological treatment of waste water under certain conditions.

Lemnaceae have the following advantages for water purification:

- they are tolerant to high content of nutrients (S. polyrrhiza is still able to grow in solutions with 1 g nitrogen per liter and 1.5 g phosphorus per liter, according to EYSTER 1966)
- they are able to absorb and desintegrate toxic substances (cf. chapter 2.3.3.5.8) and to bring under control the content of some pathogens
- they are suited as food, energy producers and as manure
- they are able to accumulate heavy metals

Disadvantages of Lemnaceae are:

- they stop growth at low temperatures; outside the tropics and subtropics, the waste water plants have to be heated during the cold season
 - they live only in a thin layer on the surface of the water; large areas are needed to remove sufficient waste substances from the water
 - they accumulate heavy metals; it is not possible to use Lemnaceae for food or manure from waters containing heavy metals; waste water with heavy metals has to be purified separately from the normal waste water
- A survey of the possible utilization of different water plants to remove

nutrients and toxic substances is given by DINGES (1982).

3.5.2. Removal of nutrients

Numerous investigations have been made throughout the world in order to clean waste water and to recycle the nutrients. A bibliographic survey of macrophytes used in waste water treatment is given by BLAKE and DUBOIS (1979). Lemnaceae are shown to have the greatest capacity in assimilating the macroelements N, P, K, Ca, Na and Mg. Other studied macrophytes are less effective in this respect if the effluents are heavily loaded with nutrients (BLAKE and DUBOIS 1982).

In the CSSR according to KVET et al. (1979), Lemnaceae in basins of 0.5 m depth remove on an average 2 kg nitrogen per ha and day (calculated for the whole year). This corresponds to a 50% removal from water of 25 mg l⁻¹ nitrogen. If temperatures stay warm throughout the whole year, the effect is much higher. In waste water ponds of Louisiana covered with Lemnaceae CULLEY et al. (1978) observed a 20-40% lower nitrogen (predominantly ammonium) content than in ponds without Lemnaceae. Examples of the daily removal of elements are given in table 3.6.

CULLEY et al. (1981) calculated that on the average, a mixture of Lemnaceae could remove annually 1378 kg nitrogen, 347 kg phosphorus and 441 kg potassium from a ha water area in Louisiana. Further investigations with Lemnaceae alone or mixed with other water plants for purifi-

Tab. 3.6. Daily removal of N, P, and K by Lemnaceae during warm seasons

Region	species	daily removal in kg/ha			author
		N	P	K	
Louisiana	duckweeds	4.7	1.6	2.1	2
Florida	<i>S. polyrrhiza</i>		0.15		4
Italy	<i>L. gibba</i> and <i>L. minor</i>	4.15	0.97		1
CSSR	duckweeds	2.0			3

1 CORRADI et al. 1981

2 CULLEY et al. 1978, 1981, CULLEY and MYERS 1980

3 KVET et al. 1979

4 SUTTON and ORNES 1977

cation of sewage water were made by SUTHERLAND and BEVIS (1979) in Michigan, USA, WOLVERTON and McDONALD (1981) in Mississippi, USA, RYTHER et al. (1980), KNIGHT et al. (1985) and REDDY (1984b) in Florida, USA, CONN and LANGWORTHY (1984) and GEARHEART et al. (1984) in California, USA, COPELLI et al. (1982) in Italy, ORON et al. (1985) in Israel, RAKHIMOV and RAKHIMOVA (1983) in USSR, SMITH et al. (1983) in Australia, and MATSUMOTO (1981) in Japan. EDWARDS (1980) reports on waste water systems with Lemnaceae from Southeastern Asia.

In mixtures with other water plants, the removal of nutrients might be even better. KUMAR et al. (1983) propose a mixture of S. polyrrhiza and Azolla pinnata. A combined mat of Lemnaceae with Eichhornia removes 6 times more nitrogen and twice as much phosphorus from the water than Lemnaceae alone (EDWARDS 1980). This can be explained by the much bigger biomass and the deeper rooting of Eichhornia. According to MATSUMOTO (1981), S. polyrrhiza, L. aequinoctialis, and Eichhornia absorb about the same amount of N (86-91%), P (35-85%), and K (12-15%) each out of sewage water which still contained (after first purification) 12 mg N, 8 mg P and 14 mg K per liter. The removal of N and P from water decreases considerably as soon as the content of these nutrients falls below 4 mg/l (REJMANKOVA 1982). At these relatively low concentrations, other water plants are required to complete purification. A reduction of the phosphorus level down to 0.1 mg per liter as is proposed for strict water quality standards in the USA cannot be achieved with Lemnaceae alone (CULLEY et al. 1978). The efficiency of nutrient uptake varied within a day: it was highest between 9 a.m. and 3 p.m. (highest light intensity) and lowest between 9 p.m. and 5 a.m. (1/3 to 1/2 of the highest value) (MATSUMOTO 1981). MATSUMOTO aerated the waste water basins to increase the efficiency of the purifying system. In this way, the nutrient became evenly distributed throughout the water. MARTIN et al. (1978) report on a small waste water plant in southwestern France for tertiary (biological) treatment of the municipal sewage using Lemnaceae and Nasturtium. The water basins had a depth of 12 cm and the water was renewed every 5 days; the biomass was harvested twice a week. At the outlet, the waste water contained 13-48% of the original content of BOD (biochemical oxygen demand; on the average 6.9 mg/l), 18-27% COD (chemical oxygen demand; on the average 0.9 mg/l), 3% MBAS (methylene blue active substances; on the average 28.3 mg/l), 1.5-4.5% nitrogen and 3-7% phosphorus. A need of 650 m² water area per 100 inhabitants was calculated for purifi-

cation. With more elaborate installations, the area might be reduced to about 200 to 300 m² per 100 inhabitants. Further studies with biological waste water purification using, at least partly, Lemnaceae have been made in France by CHASSANY-DE CASABIANCA (1982a,b), SAUZE (1982) and CHASSANY-DE CASABIANCA and SAUZE (1981), in Mississippi, USA, by WOLVERTON (1979), in California, USA, by GOLUEKE (1979), in Israel by ORON et al. (1985), and in Western Australia by SMITH et al. (1983). DEGHI and EWEL (1984) simulated the effect of waste water application on phosphorus distribution in cypress domes of Florida. An addition of waste water at the rate of 2.5 cm per week resulted in a 200fold increase of the phosphorus content within the duckweed. ELLIS and DAVIS (1984) noted that the passage of municipal effluent through floating duckweed communities generally increases diversity of algal species and decreases phytoplankton biomass. Also HOSETTI and PATIL (1986) measured reduced algal activity in systems with L. minor. However, if algae are used to remove ammonia and phosphorus from waste water an addition of duckweeds is not advisable (KOLES et al. 1986). According to BURTON et al. (1978), the regeneration of eutrophic lakes by Lemnaceae is only to be recommended if the additional input of nutrients is relatively low. Duckweeds in a rice paddy field irrigated by secondary treated sewage water play a beneficial role in reducing the excessive nutrient supply and in purifying the sewage effluents (TATSUKAWA 1986).

Closed or semiclosed systems of waste water purification and utilization of the harvested Lemnaceae for food have been investigated many times and for different purposes. CHEN (cited in EDWARDS 1980) reports on a simple system of ponds in Taiwan. Sewage from households drains directly into the water system covered with Lemnaceae. The duckweeds are regularly collected and used as foodstuff for ducks and fish. In India NASKAR et al. (1986) established a semiclosed system with W. globosa (named as W. arrhiza). The duckweeds cover a basin with sewage effluent medium and serve as food for carp. 10358 kg fish per ha and year are produced. The conversion ratio W. globosa (wet weight) to fish amounted to 6:1. Cattle ponds are wide-spread in the USA. TRUAX et al. (1972), CULLEY and EPPS (1973), RUSKIN and SHIPLEY (1976), and KELLY et al. (1978) investigated closed systems of nutrient flow from dairy cattle into waste water, into cattle pond, into Lemnaceae cover and back to dairy cattle. According to CULLEY et al. (1978), Lemnaceae covering one ha of waste water are able to purify the sewage of 15.5 cows for nitrogen, 34 cows for phosphorus

and 8.8 cows for potassium. It is recommended to first collect the methane gas from the sewage, by fermentation, before transporting the water to the pond. The feces of 100 cows develop enough energy via the methane production to supply the whole dairy farm with operation energy (except transport energy) (HUFFMAN 1980, FRYE and CULLEY 1980, CULLEY et al. 1981). Pathogens (coliforms, streptococci, Salmonella, Shigella) in the water must be destroyed by hypochlorite (AMBORSKI and LARKIN 1980). In a similar way, the recycling of pig sewage over Lemnaceae is possible (CULLEY and EPPS 1973, STANLEY and MADEWELL 1975, GHETTI et al. 1982). Lemnaceae are also used in closed systems for pisciculture. It is possible to reduce the ammonium content in the water by cultivating duckweeds (KIM and KANG 1982, PORATH and POLLOCK 1982). According to the latter authors, the circulation of fish effluent through water containing a duckweed mat succeeds in an ammonia removal of 80% within 48 hours.

3.5.3. Removal of heavy metals and other toxicants

3.5.3.1. Heavy metals

In chapter 1.1 the ability of Lemnaceae to accumulate certain heavy metals is described. The relatively high tolerances to heavy metals are mentioned in chapter 2.3.3.4.6. These two characteristics of Lemnaceae make it possible to use the plants for removal of heavy metals from polluted waters. A survey of the suitability of different water plants in this respect is given by DINGES (1981, 1982). The accumulation factor* of Lemnaceae for certain heavy metals depends greatly on the concentration of the metals and of other metals in the water as well as on the species. Table 3.7 summarizes the results of different authors. The relatively high accumulation factor of SILVEY (1967) is due to the long stay of the fronds in the slowly flowing water of very low metal content. It seems that the concentration factor is much higher at very low concentrations than at medium and high concentrations. In medium concentrations of Pb (1 mM to 10 mM) the content of Pb within the Lemnaceae

* The accumulation factor is always given on a dry weight basis in this chapter. Original values on a wet weight basis have been multiplied by 20.

risers in direct proportion to the concentration in the water (see fig. 1.2). The same is true for Cd between the concentrations of 0.1 mM and 1 mM. Within these concentrations, the accumulation factor is similar (c. 1000). In contrast, the accumulation factor for Cu and Zn is lower at high concentrations (5 mM and 10 mM) than at lower ones (0.5 mM and 1 mM) (VAN DER WERFF 1981), VAN DER WERFF and PRUIT (1982). VAN DER WERFF (1981) points out the mutual effect of Zn and Cu. A reciprocal up-take stimulation of Cu and Ni was observed by HUTCHINSON and CZYRSKA (1975). Further interrelationship between different metals are dealt with in chapter 2.3.3.4.7. Differences for species have been noted by VAN DER WERFF (1981) and HUTCHINSON and CZYRSKA (1975). According to HUTCHINSON and CZYRSKA, L. valdiviana accumulates much more Cu than L. minor (accumulation factor 500-54000 compared with 80 to 8000). The accumulation factor of Cu and Cd is much higher, that of Pb is slightly higher and that of Zn is slightly lower in L. gibba than in S. polyrrhiza (HUTCHINSON and CZYRSKA 1975; see table 3.7). The high accumulation factors for Al and Mn (up to 660'000 and 850'000, respectively) are remarkable. RUSKIN and SHIPLEY (1976) report that L. minor and L. trisulca absorb 10 times more boron from waters in Michigan than all other float-

Table 3.7 (p. 396 and 397). Accumulation factors for different elements in Lemnaceae, calculated on a dry weight basis (data which were described on a fresh weight basis have been multiplied by 20)

Species

a Spirodela polyrrhiza	d Lemna minor	g Lemna perpusilla
b Spirodela punctata	e Lemna trisulca	h duckweed
c Lemna gibba	f Lemna aequinoctialis	

References

- | | |
|-------------------------------|---|
| 1 SILVEY 1967 | 13 ALLENBY 1967 |
| 2 MANGI et al. 1978 | 14 SAROSIEK and WOZAKOWSKA-NATKANIEC 1980 |
| 3 HONDA et al. 1971 | 15 SKLAR 1980 |
| 4 LEINERTE 1969 | 16 PIISPANEN and LAEHDESMAEKI 1983 |
| 5 HUTCHINSON and CZYRSKA 1975 | 17 SZABADOS et al. 1983 |
| 6 RODGERS et al. 1978 | 18 TRAPEZNIKOV and TRAPEZNIKOVA 1979 |
| 7 GUTHRIE and CHERRY 1979a | 19 NASU et al. 1985 |
| 8 CLARK et al. 1981 | 20 OZIMEK 1983 |
| 9 VAN DER WERFF 1981 | 21 MARCIULIONIENE 1980 |
| 10 KOVACS et al. 1984 | 22 VERMAAK et al. 1976 |
| 11 TRIDECH et al 1981 | 23 WAYMAN et al. 1977 |
| 12 ALLENBY 1981 | 24 WENTSEL and BERRY 1975 |

[illegible]

[illegible]

ing or submerged water plants. The same observation was made by TRIDECH et al. (1981) in Louisiana. Pistia was next to Lemna but accumulated only 1/3. The same authors stated that Lemnaceae grid the following percentage of metals out of a waste water system: 17.8% B, 70.5% Hg, 11% Se (beside 55% N and 17.9% P). GLANDON and McNABB (1978) measured up to 1500 ppm B in the dry weight of L. minor. According to FERNANDEZ et al. (1983), L. aequinoctialis is more efficient in absorbing Zn and Hg than Pistia but less efficient than Eichhornia. FERRARA et al. (1985) investigated the absorption of Cd and Zr by L. minor. Though Lemna is less effective than Elodea and Eichhornia in absorbing the two metals it is thought to be useful in water purification due to its rapid multiplication. GELLINI and PICCARDI (1981) noted that L. minor is able to reduce the Cu content of water from 5 mg/l to 1/4 within 48 hours. According to NASU (1983), L. aequinoctialis removes the following percentage of Cd within a week: 2% from a solution with 1 ppm Cd, 50% from a solution with 0.1 ppm, and 70% from a solution with 0.01 ppm. STAVES (1980) propose the use of a mixture of Lemnaceae (S. polyrrhiza, S. punctata, and L. gibba) in order to remove Cr from industrial waste water. However, the Cr concentrations should not exceed 10 ppm. STAVES and KNAUS (1985) discuss various types of biological treatment systems with duckweeds for the removal of Cr from waste water. At 0.1 ppm Cr S. polyrrhiza, S. punctata and L. gibba exhibit the greatest percentage of Cr removal (concentration factor up to 5700 for S. punctata). In waters with a thick Lemnaceae cover, Cr^{6+} is reduced to Cr^{3+} which then readily adsorbs to organic and inorganic surfaces. FARAGO and PARSONS (1985) mention the possibility of L. minor (as well as of Eichhornia) to recover Pt from polluted waters (without giving any details).

To remove the heavy metals from the water, the Lemnaceae cover must be taken away regularly. If this is not done, the amount of heavy metals (Fe, Zn, Cr, Cu, Ni, Pb) is finally immobilized in the sediments after decaying of the Lemnaceae fronds. Only Mn is released into the water during plant decomposition and returns to the cycling system (OZIMEK 1983).

3.5.3.2. Organic toxicants

Lemnaceae are especially efficient in removing polychlorinated biphenyls (PCB), which is a rather stable toxicant, from the water. They accumu-

late the substance 6 times more than Eichhornia and 70 times more than Scirpus. In a waste water system, they were able to take out 100% PCB (ENGLANDE and KAIGATE 1981, TRIDECH et al. 1981). Phenol is accumulated by duckweeds 5000 times* which is less than in most other water plants (TRIDECH et al. 1981).

MUIR et al. (1985) report a 2000- to 10000-fold accumulation of the insecticide deltamethrine by Lemnaceae from contaminated water. L. aequinoctialis bioaccumulates insecticides (e.g. DDT, endrin) from extremely low concentrations in the water. The absorbed insecticides remain in the tissue of the Lemnaceae (VROCHINSKI et al. 1970, 1971, DE LA CRUZ and YARBROUGH 1982). L. minor accumulates DDT up to c. 800 times and HCCH up to 1200 times (VROCHINSKI et al. 1970).

LOCKHART et al. (1983) measured the bioconcentration factors in L. minor for 10 herbicides. They varied between c. 80 (krenite) and 88000 (hexachlorobiphenyl). TCDD a highly toxic contaminant of the herbicide 2,4,5-T is accumulated in L. minor 20000 to 100000 times (concentration in the water 7.0-0.05 ppt) (ISENSEEE and JONES 1975a). A rapid uptake of two phosphate ester flame retardants (triphenyl phosphate and 2-ethylhexyldiphenyl phosphate) was observed by MUIR et al. (1982). The concentration factors amounted to c. 60000 for EHDPP and 42000 for TPP (10 hours after application) and 5700 and 3000, resp. (10 days after application).

* The accumulation factor is always given on a dry weight basis in this chapter. Original values on a wet weight basis have been multiplied by 20.

3.6. UTILIZATION AS TEST AND INDICATOR PLANT

The widespread use of Lemnaceae as test plants is due to the simple cultivation techniques, the need of little space, the fast growth rate and the genetical uniformity of its cultures which correspond to clones. Correspondingly, the literature is voluminous. The usefulness of Lemnaceae for investigation of physiological processes is dealt with in chapter 2. Lemnaceae are however not very suitable to show specific reactions towards different substances since only few distinguishing characteristics are present. In most studies, changes on growth rates are recorded. Also dry weight, size of frond, length of root, growth of root (LIEBERT 1986a), anthocyanin formation (reported as more sensitive than growth rate: EINHELLIG et al. 1985, LEATHER and EINHELLIG 1985, JUNG-NICKEL 1978, JUNG-NICKEL and AUGSTEN 1986), turion production and germination (JUNG-NICKEL 1978), gas exchange (MARTI et al. 1986) and CO₂ production (WUESTLING and BOEHM 1979) have been tested.

Tests have been developed and used for the following substances:

- a) **heavy metals:** AUGSTEN (1983), FISCHER (1981), LIEBERT (1986), NASU and KUGIMOTO (1981), SZABADOS et al. (1983), WANG (1986a);
- b) **growth factors:**
 - ABA:** ENUKWESI and DUMBROFF (1978), GOLDBACH and MICHAEL (1976), JOHANSSON et al. (1982), LIEBERT (1980a), LIN and MATHES (1973), TAYLOR and DUMBROFF (1975), TILLBERG (1975), VAN STADEN and BORN-MANN (1970a);
 - kinetin:** LETHAM (1967);
 - different growth factors:** JUNG-NICKEL and AUGSTEN (1986), RANE and TUCKEY (1972), WUESTLING and BOEHM (1979);
- c) **herbicides:** AUGSTEN (1983), BAHADIR and PFISTER (1985), BIRMINGHAM and COLMAN (1983), DAMANAKIS (1970, 1972), DECLEIRE and DE CAT (1977), FISCHER (1981), LIU and CEDENO-MALDONADO (1979), O'BRIEN and PRENDEVILLE (1978), PESTEMER (1979), RICHARDSON (1985), ZAWADZKI (1975);
- d) **allelopathic substances:** EINHELLIG et al. (1985), LEATHER and EINHELLIG (1985), MARTI et al. (1986), SAGGESE et al. (1985);
- e) **drug substances:** SHIMOMURA et al. (1981, 1982);
- f) **further toxicants:** DAVIS (1981), KING and COLEY (1985), SLOOF and CANTON (1983), WALBRIDGE (1977), WARD et al. (1981);

g) **phosphate (and nitrate) pollution:** AEBLI (1986), FEKETE et al. (1976), JUNGNICHEL (1978), POTT (1981)

The following Lemnaceae species have been used as test organisms:

S. polyrrhiza: DAMANAKIS (1970, 1972), JUNGNICHEL (1978), JUNGNICHEL and AUGSTEN (1986), RICHARDSON (1985), WARD et al. (1981);

S. punctata: KLAINÉ (1985);

L. gibba: DAVIS (1981), KING and COLEY (1985), LIEBERT (1986a), TILLBERG (1975);

L. minor: AEBLI (1986), BAHADIR and PFISTER (1985), BLACKMAN (1952), BISHOP and PERRY (1981), EINHELLIG et al. (1985), FEKETE et al. (1976), FISCHER (1981), FROMM (1946, 1951), FUNDERBURN and LAWRENCE (1963), KING and COLEY (1985), LEATHER and EINHELLIG (1985), LOCKHART and BLOUW (1979), MARTI et al. (1986), O'BRIEN and PRENDEVILLE (1978), OFFORD (1946), RANE and TUCKEY (1972), ROULET (1975), SAMPFORD (1952), SIMON and BLACKMAN (1953), SLOOFF and CANTON (1983), SZABADOS et al. (1983), WALBRIDGE (1977), WANG (1986a), WARD et al. (1981);

L. obscura: EINHELLIG et al. (1985); LEATHER and EINHELLIG (1985);

L. aequinoctialis: KING and COLEY (1985), LIU and CEDENO-MALDONADO (1979), NASU and KUGIMOTO (1981), ROWE et al. (1982), SHIMOMURA et al. (1981, 1982);

W. arrhiza: AUGSTEN (1983), WUESTLING and BOEHM (1979).

Lemnaceae are instructive laboratory and experimental plants for school purposes (see RHODES 1968).

3.7. UTILIZATION FOR PRODUCTION OF ENERGY AND CHEMICAL COMPOUNDS

3.7.1. Energy production

WOLVERTON and McDONALD (1981) produced 0.14-0.22 m³ methane (CH₄) per kg dry weight of a mixture of water plants including Lemnaceae. SEKINE (1979) worked out a patent to obtain methane or ethanol from W. globosa (named as W. arrhiza) by fermentation with yeast. He got either 285 g ethanol per kg dry weight or 0.42 m³ methane per kg dry weight. He was able to cultivate the Wolffia on organic waste water treated first with photosynthetic bacteria, micro-algae and zooplankton before being inoculated with Wolffia. The author calculates that normal production of 1 liter ethanol in Japan amounts at present to 186 Yen. Production of ethanol via Wolffia is much cheaper (100 Yen per 1 liter). If all organic waste waters in Japan were used for Wolffia cultivation, a total production of 10⁶ t ethanol per year would be possible.

EL-HINNAWI (1983) proposes energy farms for fuel production in regions where duckweeds and water hyacinths are harvested. Also RAO (1984) suggests an aerobic digestion of the same water weeds for energy production. BAI (1985) investigated the biodegradability and methane production of cow-dung mixed with Lemna.

3.7.2. Production of pharmaceutical compounds

Lemnaceae have been known as medicinal plants for many centuries. DIOSCORIDES (first century) and GALEN (second century) used the name "phakos ho epi ton telmaton" or in latin "viparia" for L. minor. DIOSCORIDES and later M. ADANSON (18th century) and A. MORI (19th century) attributed the following pharmaceutical effects to Lemnaceae: soothing of freezing injuries, relief of aches of podagra and burns, healing of fractures (cited from BEAUVOIS 1816). Lemnaceae have also been used to cure hepatitis. Blood clots could be dispersed by applying L. trisulca dissolved in white wine. McCANN (1942) mentions that Lemnaceae are used in India to prevent blood loss during menstruation. A mixture of Lemnaceae with pepper is put on the eyes of unconscious typhus patients.

No positive effect of Lemnaceae against cough were observed by ANDRONOVA

(1972). In Canada, L. trisulca together with Stellaria media is applied by Iroquois Indians against swellings (ROUSSEAU 1945, ARNASON et al. 1981). According to WATT and BREYER-BRANDWIJK (1962), L. minor has been used in Africa as a remedy for dropsy and rheumatism. In China, it is used internally as a diuretic, antiscorbutic and antisphyllitic, and externally for eye diseases and carbuncles. The same authors report that Lemnaceae give negative antibacterial and antimalarial tests. On the other hand, STANGENBERG (1967) recorded a bacteriostatic effect of L. minor extracts against the gram-negative bacterium Sphaerotilus natans. No efficacy was observed against other bacteria, and not all clones of L. minor showed the same effect. Extracts of L. minor did not reveal any evident activity against Staphylococcus aureus, Mycobacterium smegmatis, Escherichia coli, Candida albicans and Fusarium roseum. However, Alternaria sp. was inhibited moderately by methanol extracts of L. minor (SU et al. 1973b). HILLMAN (in lit. 1979) noted an algicidal effect (against a unicellular chlorophyte) of W. globosa (originating from Thailand) in Petri dishes on nutrient agar.

YONG and THO (1976) measured 0.095 mg per g dry weight of a cardiac glycoside-like substance present in the aqueous extract of the fronds of L. aequinoctialis (named as L. minor). The substance was able to produce the same effect on the chicken heart as standard digoxin. The authors isolated the reducing sugar digitoxose which is typical of cardiac glycosides.

The toxicity of L. minor for white mice was investigated by ANDRONOVA (1972). The smallest toxic level was 577 mg per kg live weight and the smallest lethal dosis 735 mg per kg. In conclusion, there is not much known on pharmaceutically important substances produced by Lemnaceae.

3.7.3. Enzyme production

AUGSTEN (1984a) gives a survey of the possibilities of using Lemnaceae in biotechnology. Lemnaceae can be successfully applied in enzyme reactors for continuous catalysis of metabolic processes. The enzyme is stored immobile within the plant (especially at the surface). The high activity of the enzymes is preserved for a long time. The harvested plants are stabilized and conserved by lyophilization, by drying with acetone or by other methods. Fig. 3.1 shows the activity of some of the

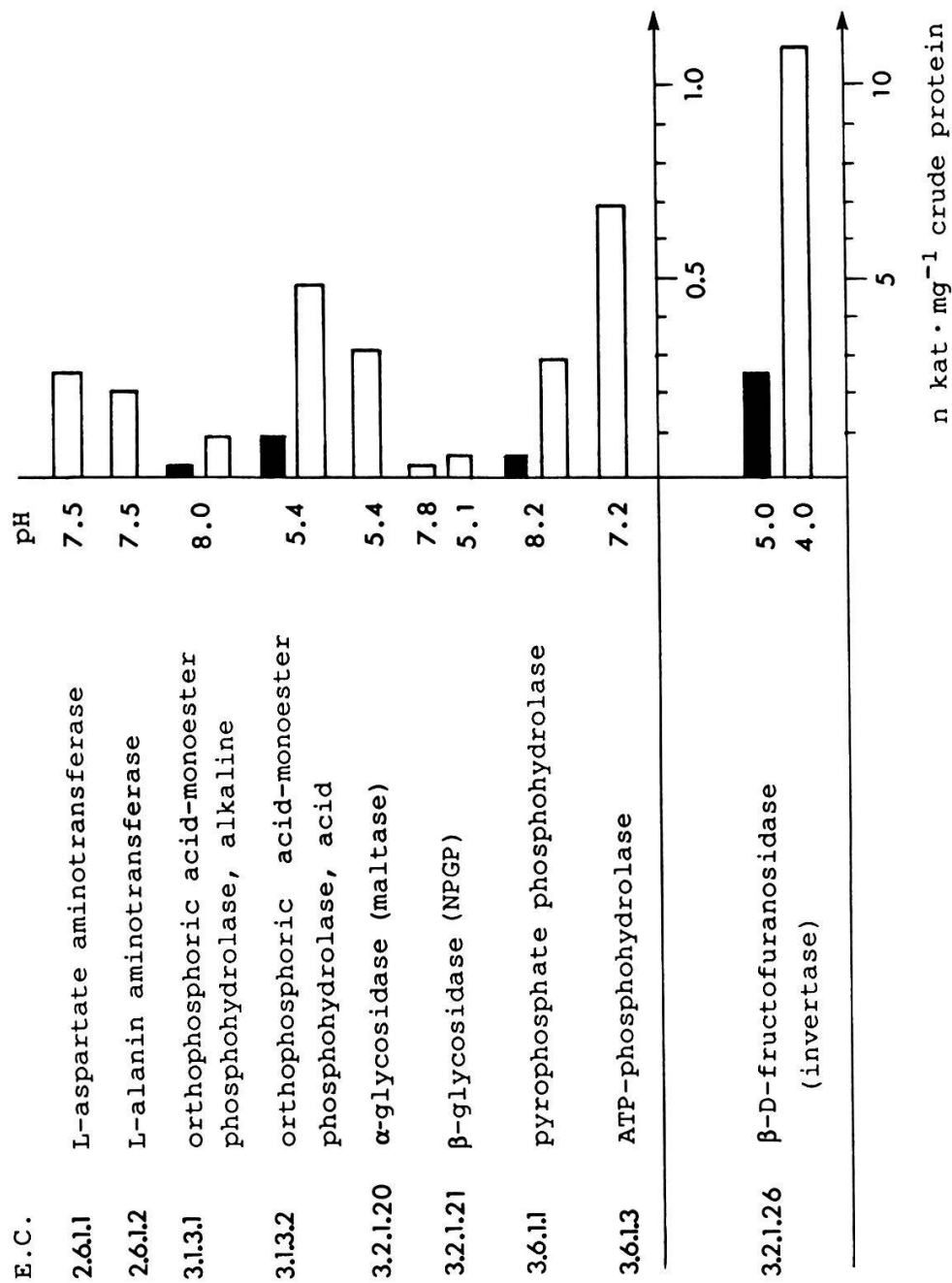


Fig. 3.1. Activity of some enzymes of *Wolffia arrhiza* in a reactor after having cultured the plants in a normal medium (■) or with limited phosphorus (□) (after AUGSTEN 1984a).

enzymes of W. arrhiza in a reactor used in Jena, GDR. Also turions of S. polyrrhiza have been investigated. TLOMAC et al. (1984) cultivated Lemnaceae to transform testosterone to androstenedione and 5 α -androstane-3, 17-dione.

3.8. SPECIAL UTILIZATIONS

3.8.1. Cosmonautic plant

Since Lemnaceae are effective oxygen producers and CO₂ removers, some investigations have been performed to check the suitability as cosmonautic plants. EYSTER (1966) studied S. polyrrhiza in respect to a possible use in space flight. S. polyrrhiza was also investigated under simulated space-laboratory conditions. Early turion development was negatively affected by these conditions (KUTLJACHMEDOW et al. 1978). MILLER (1978) investigated the effect of vibrations (100 Gc, 0.6 mm) and ionizing irradiation (X-rays, 1-3 grad) on S. polyrrhiza. Vibrations affect the growth rate negatively. If applied together with irradiation, they increase the radiosensitivity of the plants. NEY (1960) and WILKS (1962) studied the conditions for the successful cultivation of water plants in spaceships. The efficiency of a duckweed system proved to be superior to one of algae. To produce enough oxygen for one person, about 2.5 m² of cultivation area are necessary. Photosynthetic and respiratory gas exchange of S. polyrrhiza was not affected by exposure to near weightlessness over a period of 230 hours (WARD et al. 1970).

3.8.2. Manure

In regions with mass development of Lemnaceae, the duckweed cover is distributed onto the fields and gardens as manure (e.g. ALIKUNHI et al. 1952). Already 1854 WELWITSCH reported (in herb.) that W. arrhiza is used as manure in Angola. The cultivation of Lemnaceae for manure in China is described by TAI-HSING et al. (1975). In southern and eastern China, Lemnaceae are cultivated in ponds, channels, rice fields and other waters and harvested every 4 to 5 days to fertilize the fields (MERIAUX 1978, McCALLA and PLUCKNETT 1981). In Mexico, the agricultural system of chinampa generates from aquatic vegetation containing mats of Lemnaceae mixed with other water plant. The detritus of these mats raises the soil level and further sedimentation accumulates by addition-

al duckweed cover and mud brought from the neighbouring channels. Eventually, a soil with excellent texture, and good water and cation exchange capacity develops and offers the possibility of harvesting four crops a year of vegetables or corn (LOT et al. 1979).

In the dry state, Lemnaceae contain 2-7% N, 0.5-3% P, and 2-5% K (CULLEY et al. 1978, cf. chapter 1.1). Mixed stands with Azolla are especially suited for manure production. Azolla has a symbiotic interrelation with the blue algae Anabaena azollae which is able to fix nitrogen from the air. PARK and YATAZAWA (1979) and ZUBERER (1981, 1982) demonstrated that Lemnaceae also live together with some nitrogen fixing heterotroph bacteria (e.g. Klebsiella) and Cyanophyta (e.g. Calothrix, Microchaete; Anabaena was only found in L. gibba). Up to 10^5 individual organisms per g dry weight could be counted. They cannot supply more than 15-20% of the nitrogen needed by Lemnaceae (ZUBERER 1982). The nitrogen fixation in Azolla seems to be more efficient. SUTTON (1981b) states that up to 50% of the nitrogen need of rice in rice fields can be supplied by Azolla. According to EL-DIN (1982) a L. gibba association is able to fix 60 kg nitrogen per ha in 100 days which amounts to about 1/7 of the value of an Azolla cover. In Egypt the total N accumulation by L. gibba is 119- 140 kg per ha and year.

3.8.3. Reduction of water loss in arid regions

Lemnaceae are possibly the only water plants that evapotranspire less than the same area of open water evaporates. The ratio evapotranspiration/evaporation amount to c. 0.9 for a L. obscura cover (named as L. minor) in southern USA (DEBUSK 1980, RYTHER et al. 1980, DEBUSK et al. 1983). BOYD (1975) noted a difference between different species. He measured a ratio of 0.9 for W. columbiana, and 0.85 for S. polyrrhiza. SEYBOLD (1930, cited from DEBUSK 1983) observed a lower ratio with greater wind velocities. Similarly, ORON et al. (1985) measured, in Israel, a distinctly lower ratio if the evaporation rate is higher than 4.5 mm per day: 0.75 for S. polyrrhiza and 0.7 for L. gibba. Most other water plants (e.g. Typha, Cyperus, Eichhornia) have a much greater evapotranspiration than the evaporation. Only in Pistia, Trapa, and Ipomoea it is not significantly higher than the evaporation (BREZNY et al. 1973). The water loss of ponds covered with Eichhornia turned out to be

3 to 5 times greater than in free surface ponds (ORON et al. 1985). The relatively low evapotranspiration of the Lemnaceae is due to the small frond surface in comparison with the water surface and with the surface of Eichhornia.

In conclusion, a Lemnaceae cover on the surface of water reservoirs and ponds in climatically dry regions, in order to prevent great water loss, may be advisable if it does not interfere with other purposes.

3.8.4. Reduction of mosquito breeding

A survey of possible use of Lemnaceae in fighting mosquito breeding is given by JENKINS (1964). In waters with a complete Lemnaceae cover JOHNSON (1902) observed no mosquito breeding whereas in open spaces, he was able to detect larvae of Culex and Anopheles. According to ANCONA (1930), a closed cover of Lemnaceae mixed with Azolla prevented the development of some mosquito larvae. On the other hand, DYAS and KNAB (cited in MATHESON and HINMAN 1929) state that one of the most abundant breeding grounds of Culex salinarius was a large marsh completely covered by Lemna. BENTLEY (1910) reports from India that a cover of Lemna or Azolla is of no value in preventing the presence of mosquito larvae. However, a layer of Wolffia (probably W. globosa named as W. arrhiza) keeps the water free of larvae of Anopheles, Culex and Stegomyia. Larvae which were placed in water with a Wolffia cover died within several hours. Most authors observed a positive effect of Lemnaceae cover in preventing the breeding of mosquitoes (e.g. ADIE 1904, HILDEBRAND 1925, MATHESON and HINMAN 1929, HESS and HALL 1945, LAIRD 1956, JENKINS 1964, SMITH and ENNS 1967, SJOGREN 1968, KERBABAIEV et al. 1985). CULLEY and EPPS (1973) state that Spirodela hinders Anopheles mosquitoes from laying eggs. In addition, the larvae do not get enough oxygen below a duckweed cover. Moreover, Lemna covered ponds harbour numerous predatory insects which attack mosquito larvae. BRADLEY (1932) lists three main factors resulting in a decrease of Anopheles breeding in waters completely covered by Lemnaceae:

- 1) larval food supply is low due to a poor development of phytoplankton;
- 2) larvae cannot break the surface cover for respiration;
- 3) larvae are not very effectively hidden from their enemies in the water below the Lemnacean cover.

FURLOW and HAYS (1972) report a decrease of Culicidae (Diptera) larvae below a S. punctata cover. If the cover was completely closed, no mosquito breeding occurred. In general, Anopheles species and Uranotaenia sapphirina reacted much more sensitively than Culex erraticus. Probably, the continuous surface mat represented a barrier to female oviposition. ANGERILLI (1980) investigated the influence of extracts of different water plants (included L. minor) on the development of the mosquito Aedes aegypti. An extract of L. minor had a toxic effect on the larvae. Into the vessel with the extract, only about 5% of the eggs were laid compared with the number in distilled water. JUDD and BORDEN (1980) demonstrated that aqueous and methanolic extracts of L. minor prevent the oviposition of Aedes aegypti but not of Culex pipiens at concentrations of 1000 and 10000 ppm. At concentrations of 1, 10, and 100 ppm, no effect showed up. In an outdoor experiment, in basins with L. minor, ANGERILLI and BEIRNE (1980) observed distinctly less eggs (c. 1/2) and larvae (c. 1/8) than in basins without Lemnaceae. This effect was studied in Culex inornata and Culex pipiens.