INTERNATIONAL JOURNAL OF CONSERVATION SCIENCE
ISSN: 2067-533X Volume 8, Issue 4, October-December 2017: 715-722 www.ijcs.uaic.ro

DUCKWEED UTILIZATION FOR FRESH WATER CONSERVATION (MANAGEMENT) IN RECIRCULATED AQUACULTURE SYSTEMS

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Abstract

The paper aims at presenting key aspects of the use of duckweed in the industry applications and a concept for using duckweed bio filters in aquaculture systems. Duckweed is a plant adapted to aquatic systems and considered to be one of the smallest plants in the world, with a diameter of 1-15mm. Although it is a small plant, its properties make it suitable for industrial use (production of bio fuel, aquaponic, food source, waste water treatment). Several aspects of the duckweed crop management are mentioned in the paper. It has been shown that certain duckweed species can successfully lower the Chemical Oxygen Demand (COD), Biological Oxygen Demand (BOD), total nitrogen (TN), total phosphorus (TP) and orthophosphate (OP). The authors propose a new waste water treatment technology with duckweed tanks for recirculated aquaculture systems.

Keywords: *Duckweed; Ammonia; CBO_5; Wastewater treatment; Recirculated aquaculture systems (RAS)*

Introduction

Duckweed description

Duckweed (also known as Water lens and Bayroot) are aquatic plants which float on the surface of slow-moving or still bodies of water (both fresh and wetlands). Duckweed is as "bayroot", they arose from within the aroid family (Araceae) [1] and are classified as the subfamily Lemnoideae. Before the end of the XXth century, the duckweed is classified as a separate family, Lemnaceae. These aquatic plants are very simple, without stem or leaves obvious. The greater part of each plant is a small structure organized "thallus" or "frond". Only a few cells coarse, often with air pockets allow them to float on the surface of the water. Each plant, depending on species, can take a root or one or more roots simple [2].

Reproduction is mostly asexual and occurs from a meristem closed at the base of the branch. Occasionally, three tiny "flowers" consisting of the pistil and two stamens, by which it

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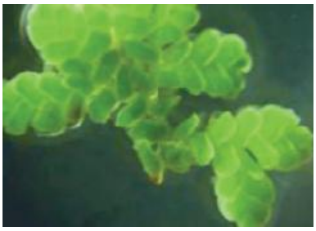
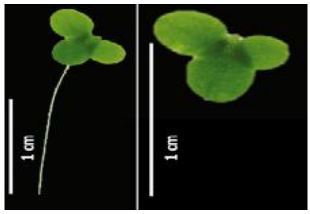
appears sexual reproduction. This "flower", as a pseudanthium, reduced inflorescence, with three flowers, which are distinctly either female or male. They are derived from the spadix in Araceae [3]. The evolution of the duckweed inflorescence remains ambiguous due to the reduction of evolutionary considerable of this plant. Duckweed genus *Wolffia* is the smallest flower known which measure about 0.3mm long [4]. The fruits that are produced by reproductions occasional are utricles, and seeds are produced in a bag with air that facilitates flotation. Some of the approximately 70 species of duckweed which exist around the world are presented in Figure 1.

Duckweed is adapted to aquatic habitats is among the smallest and simplest flowering plants, floating monocotyledons, with a size between 1-15mm. Roots grow up to 15cm, the tips are mostly round, and the sheath is not winged.


Azolla caroliniana [5]


Marsilea quadrifolia [6]



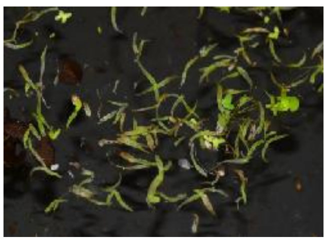
Lemna minor [7]


Lemna trisulca [8]


Salvinia natans [9]


Spirodela polyrhiza [10]


Wolffia arrhizal [11]


Wolfiella sp. [12]


Landoltia punctata [13]

Fig. 1. Varied species generally defined as duckweed

The tissue of duckweed contains twice the protein, fat, nitrogen, and phosphorus of other vascular plants because it is mostly made up of metabolically active cells with very little structural fiber. Nutrients are absorbed from each frond, not through a central root system, through the whole plant directly assimilating organic molecules such as simple carbohydrates and various amino acids. Most photosynthesis is devoted to the production of protein and nucleic acids, the entire body of the duckweed being composed of non-structural, metabolically active tissue, making them very high in nutritional value.

Duckweed utilization

The plant duckweed can be exploited in several ways, since it can simultaneously clean the waste water, produce animal feed and used as bio fuel [14-23].

a. Duckweed utilization in aquaponics. The main advantages of using Duckweed in aquaponics [24, 25] are:

- some fish will eat duckweed, duckweed will repopulate faster than the fish can eat it;
- the dry duckweed biomass contains up to 40% protein, that can further be capitalized in feed applications.
- duckweed removes substantial amounts of nitrogen and phosphorus from water.

Main disadvantages of using Duckweed in aquaponics include:

- duckweed is about (86 - 97)% water [26] thus the protein content of the wet biomass is low and can reach 2%;
- high concentration of RNA and raphids present in duckweed as well accumulation of toxic chemicals in duckweed biomass may represent a problem in some feed formulations;
- duckweed grows faster than fish can eat it; the biomass can get out of the tank and can clog pipes and pumps.

b. Duckweed utilization as food. Duckweed protein content per dry biomass of duckweed ranges between 25% and 45% and in good growth conditions the biomass doubles every 36 hours. Especially tilapia (*Oreochromis niloticus*) enjoys eating it [27]. In aquaponics the great merit of duckweed is not that it contains high concentration of a single nutrient (proteins, starch, etc.), but that it is rich in numerous nutrients and a diet containing live duckweed may substantially lower the cost of fish feed. Fish (such as tilapia) may be fed a mixed diet that contains some nutrients in formula feeds and some nutrients (such as vitamins, minerals, proteins, fats, starch, etc.) can be provided via live duckweed [28-32].

To a whole meal with a residual moisture content of 10% duckweed dry can be stored without deterioration over time of at least five years without special precautions, if protected from sunlight and changes in humidity. The degradation of the pigments is done to direct exposure to sunlight and, therefore, reduce the nutritional value, but will not influence the protein content. Bags opaque plastic is recommended for long-term storage. Drying passive solar is made by spreading fresh material on the bare ground or on a pasture with grass, this being the simplest form of post-harvest and processing. However, exposure of fresh duckweed to ultraviolet light of the sun degrades the beta-carotene and other pigments, and reduces their concentrations.

c. Producing ethanol from duckweed [33]. Six-day old fresh duckweed, grown on nutrient deficient water, and the oven dried duckweed were utilized in a fermentation process [33]. The substrate load was 100 g DW/L, but this was only possible for the oven dried duckweed and not for the fresh duckweed biomass. Larger ethanol yield was obtained from fresh duckweed biomass relative to dried duckweed biomass regardless of the reduced amounts used (Fig. 2) [33]. The reason is unclear, but it was speculated that some starch is lost or becomes less available for hydrolysis in the dry duckweed.

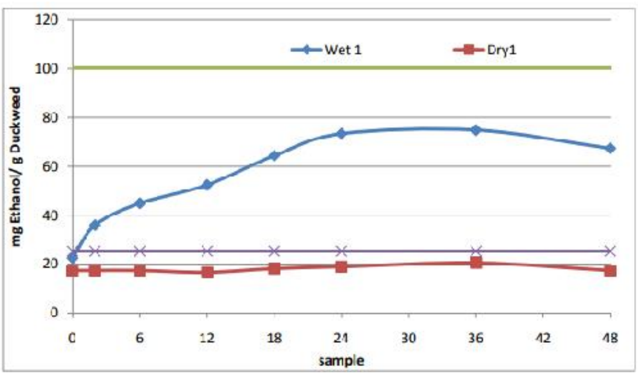


Fig. 2. Ethanol yield from fresh and dried ducked biomass [33]

d. Using duckweed to purify water.

Treating water is one of the most often proposed utilization for duckweed. Duckweed are yet sensitive and require controlled conditions to grow fast and remain healthy in the presence of pollutants. Maintaining a rich duckweed culture is difficult without sufficient illumination and nutrients. Moreover, duckweed biomass may accumulate toxins from water, rendering it unusable for feed applications.

Material and Methods

Specific conditions for duckweed cultures

Duckweed plays a significant role in the removal of nitrogen and phosphorus from water. However, it needs some specific conditions:

a. Surface area and water depth. Surface area has to be maximized. Duckweed needs only a few inches high of water, but it needs as much space to spread. In direct sunlight is it recommended that the water is sufficiently deep, so it does not warm up too fast.

b. *Oxygen.* In swimming pool algae without aeration of the water will consume the oxygen inside water until it makes it anaerobic.

c. pH. Duckweed can survive at a pH ranging from 5 to 9, but grows best in the pH range from 6.5 to 7.5. When the pH is below 7.0, ammonia can be kept in the form of ionic (NH_4^+), which is actually the preferred form of nitrogen for *Lemna sp*. An alkaline pH changes the equilibrium ammonia ion ammonium to ammonia gas, which is toxic to duckweed.

d. Temperature. In the warm season duckweed grow faster. For faster growth, the temperature should be above 21.1°C [34]. Different duckweed species grow from polar regions to the Equator and are present at various altitudes. However, the temperature range for each species of duckweed is limited, so each temperature climate is populated by a different species.

e. Light. Duckweed prefers natural sunlight and commonly grows in open ponds or shallow wetlands exposed to sunlight [34]. In the containers covered, the plants grown can't lose water to evaporation, but the exposure of direct sun, inside containers or in greenhouses, the plant will overheat, it whitens and dies. Artificial illumination can also be utilized when growing duckweed, but it is expensive in terms of infrastructure, consumable materials and energy intensive.

f. Water movement. Duckweed does not like water moving at the surface, but only underneath. For this reason, it is often recommended to grow duckweed with surface barriers. Water in slow motion is preferred by duckweed however, yet fast growth can be observed in some areas with agitated water. To increase the process of asexual reproduction, it looks like the duckweed prefers to develop in water in which the phenomenon of shaking is low, as well as in the vicinity of the flows of the exhaust. (e.g. the duckweed will divide quicker).

g. Other parameters that influence the duckweed cultures are: salinity, ammonia, duckweed density, fertilizers, potassium, trace minerals and organic load.

The management of duckweed cultures

a. Crop density. In a culture the duckweed must remain at a reasonable density to divide fast and therefore it has to be harvested frequently. Even if crop density is a complete coverage, what is the optimal rate of growth; it still offers enough space for hosting a rapid growth of the colony. This is equivalent to one day of production of the crop which represents the rate of 0.5 to 1.5 tons of fresh duckweed per hectare [35, 36]. A dense crop cover will suppress nitrifying bacteria and will reduce dissolved oxygen in the water column. The process of denitrification improves through an increase in anaerobic bacteria and ensures a balance of nitrogen compared to ammonium vs. nitrate. When ammonium ions are assimilated by duckweed lowers the pH. The ability to form a mat over the surface of the water is one of the competitive advantages of duckweed.

b. Poly-culture or monoculture? Frequently, two or more duckweed species grow together naturally in aquatic systems, which increases the range of environmental conditions in which the crop has developed. Distinct species of duckweed have different growth optima, therefore seasonal variations may result in changes in species composition and dominance. It should be recognized that the reserve of seeds stored at different colonies of duckweed from the same species will be slightly different genetically and are likely to be adapted and separated at a lightweight set of environmental conditions.

c. Algae management. The primary competitors of duckweed for nutrients and fast growth are unicellular algae. Therefore, sufficiently dense biomass of duckweed must cover the water surface to suppress algae growth. This is an essential crop management technique because if they have the opportunity algae will grow faster than duckweed. The production of ammonia free of algae dominant that increase the pH of the environment is toxic to duckweed. Albeit mechanisms are yet unclear, some microscopic algae inhibit nutrient uptake and may thus reduce the growth of duckweed.

Results and Discussion

Duckweed utilization for wastewater treatment

Ammonium is the preferred form of nitrogen for duckweed species. The main source of ammonium for the colonies of duckweed in the wild is the decomposition of organic material by anaerobic bacteria existing. It was reported that duckweed plants will utilize most available ammonium before they begin assimilating nitrate, and grow faster in the presence of ammonium. In contrast, unicellular algae will prefer nitrate. This is interesting because it may offer a solution for water treatment in recirculated systems. Among all species of duckweed *Lemna gibba* and *Lemna minor* were found to be some of the most efficient in the treatment of wastewater, with the removal of nutrients, organic matter, soluble salts, heavy metals and in eliminating suspended solids, the abundance of algae and the presence of fecal coliforms.

In laboratory duckweed growth experiments, *N. Ozengin and A. Elmaci* [37] fed duckweed with municipal and industrial waste water at constant temperature. The water treatment efficiency of the duckweed bio-reactors (with regards to COD, total phosphorus (TP), total nitrogen (TN), and orthophosphate (OP) was evaluated by measuring influents and effluents. The removal efficiency was 73-84% for COD, 70-85% for TP, 83-87% for TN, and 83-95% for OP [37]. In an experiment by *M.W. Shammout and H. Zakaria* [38] duckweed (*Lemna sp.*) was used to upgrade the quality of waste water in a waste water treatment plant. Duckweed laboratory experiments on this wastewater have shown an average removal efficiency of 51% for BOD_5, 56% on nitrate, 48% on TN, 46% on organic nitrogen, 56% on phosphate, 50% on total phosphorus, 68% on total coliforms (TC), 75% on total viable count (TVC) and 69% on fecal coliforms (FC) [38].

An innovative technology, using duckweed, for wastewater treatment within RAS

The authors propose a new wastewater treatment technology, with duckweed tanks, for recirculated aquaculture systems (RAS) (Fig. 3). In common RAS 90% of the water is recirculated, thus 10% fresh water is replaced each day. In the purification system proposed here, the influent enters directly in the fish pond. From this basin, polluted water is discharged to a primary decanter to remove solid suspensions, after which the water is pumped into a duckweed purification step. Duckweed pools are a new concept designed for minimal footprint. These ponds can be aerated to ensure both the oxygen demand for plants and the reduction of CBO_5. Depending on the location, artificial lighting systems can be provided, especially for the bottom-level basins of the tower configuration. The first Duckweed basin in which the waste water enters is located at a higher level, after which water flows gravitationally into the lower basins. It is mainly in these pools that the reduction of CBO_5 and ammonium is achieved. It is further proposed to place an anoxic basin to reduce the amount of nitrates and nitrites.

The biological treatment step is followed by another step of removing the solid suspensions. Before re-entering the treated water into the fish pond, disinfect it with a disinfection system with UV lamps. The sludge is discharged from the treatment basins. For efficient water purification, the excess duckweed has to be removed. Within the purification scheme provided in Figure 3 can still insert another float with dissolved air flotation.

The advantage of this technology is that it introduces air in the form of nano-bubbles, which increases the efficiency of oxygen transfer from air into water and helps the process of water purification. Further, by flotation, it is possible to remove "light" solid suspensions which have a density close to that of water and which do not settle or settle very hard.

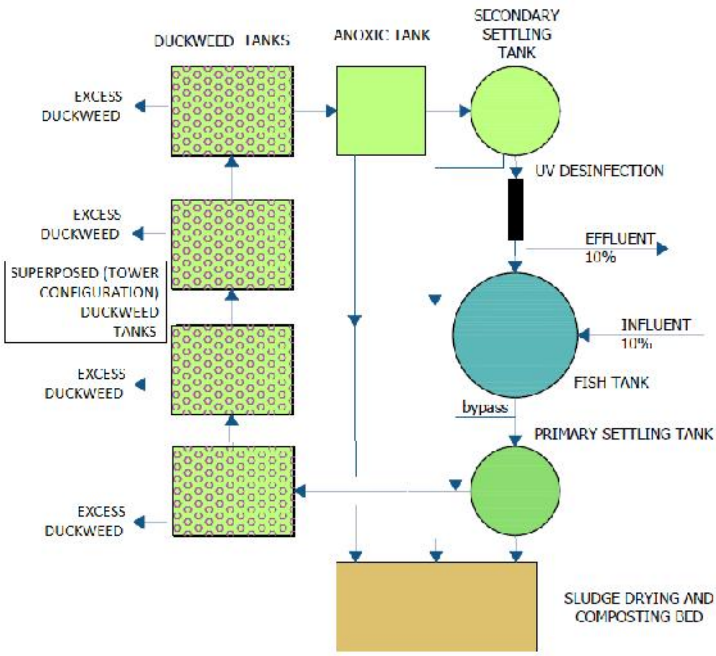


Fig. 3. Duckweed utilization in RAS wastewater treatment plants

Conclusions

Duckweed-based waste water treatment plants are relatively simple as both investment and operation/maintenance. They do not require expensive technologies or equipment. Therefore, this technology can be considered to be cost-effective and Eco-friendly for waste water treatment in RAS. Furthermore, a supplementary utilization of the excess ducked biomass will add benefits to end-users.

Acknowledgments

This work was supported by a grant of the Romanian National Authority for Scientific Research and Innovation, CCCDI – UEFISCDI, project number COFUND-WATER WORKS ERANET 2015-ABAWARE (1), within PNCDI III.

This work was also supported by a grant of the Romanian National Authority for Scientific Research and Innovation, CNCS/CCCDI – UEFISCDI, project number PN-III-P2-2.1-PTE-2016-0183 (APIFLOT-II), within PNCDI III.

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Received: February 10, 2017
Accepted: November 28, 2017