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SWINE WASTEWATER AS MEDIUM FOR GROWING DUCKWEEDS TO NUTRIENT CYCLING AND VALUABLE BIOMASS CONVERSION

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ABSTRACT

Floating aquatic macrophytes, commonly known as "duckweeds" are the smallest monocot angiosperms and form dense populations with genetically uniform clones. The high level of nutrients, particularly nitrogen (N) and phosphorus (P), favors the development of these fast-growing plants with high biomass yield in aquatic ecosystems. In some Asian countries, they are vegetal protein source to several kinds of food for animals and humans. This study evaluated the yields (fresh and dry) and some nutritional properties from duckweeds biomass: *Landoltia punctata* and *Wolffia brasiliensis* grown (individually) for 20 days in a pilot-scale batch reactor using complex nutrient medium swine wastewater based. For acclimatization and density build-up, the biomass was maintained for 4 to 5 days in a restrict space (≈10% of the reactor area), followed by gradual expansion. After 20 days of cultivation, the total biomass was collected, drained and dried. The yields in fresh weight were to *Landoltia* of 7,000 and to *Wolffia* 4,300 kg.ha^{-1}; crude protein, 30 and 27%; carbohydrates, 35 and 33 %, respectively. Biomass production shows to be satisfactory, kinetic studies of growth and conversion of substrate into product will improve the yield and compositional selectivity of the biomass, in order to establish good management and cultivation practices.

Keywords: biotechnological process, macrophytes, vegetal biomass, nutrition.

1 INTRODUCTION

The demand for food and bio-based products limits the productivity growth and intensifies global competition for natural resources due to recent stricter environmental regulatory standards (Fritsche et al., 2021). Plants are important global sources of energy and food, taking into account that the world population is increasing rapidly. In aquatic ecosystems, macrophytes represent one of the main groups of photosynthetic organisms and primary producers. They play an essential role in nutrient cycling through significant accumulation in their submerged roots or leaves (Pott, 2000).

In recent years, plant-based proteins have attracted attention as an alternative source for daily consumption (Xu et al. 2020). Duckweeds are rich in protein, reaching up to 45% of the dry weight, increasing twice its biomass in 1 to 6 days, by almost exclusively vegetative reproduction (Armstrong; Thorne Society 1984). This plant group is characterized by the extremely small size belonging to Araceae family, which comprises 37 species of five different genera: *Spirodela*, *Landoltia*, *Lemna*, *Wolffiela* and *Wolffia*. The latter is the smallest angiosperm and the most evolved of the genera; it does not produce roots unlike other species (Fig.1), gets to a density of 1-2 million plants per square meter with their largest size reaching approximately 1.4 mm in their reproductive phase (Witty, 2008). In swine wastewater pond, the growth rate can reach 29 g m^{-2} day^{-1}, producing 106 ton ha^{-1} year^{-1}, in dry mass (Cheng et al., 2002). This amount is much higher than some crops, such as corn (7,84), wheat (3,15) and barley (3,70) ton ha^{-1} year^{-1} (Cui; Cheng 2015). In Southeast Asia, *Wolffia* is a natural food source called "water eggs" or "vegetable meat balls" (Appenroth et al., 2018). As an emerging crop, its rapid growth rates offer potential for cultivation in closed systems, besides the phytoremediating capacity. Thus, the present study evaluated the yields and some nutritional properties of *Landoltia punctata* and *Wolffia brasiliensis* biomass grown in "raceway" reactor (pilot scale) using swine wastewater as nutrition source.

2 MATERIAL & METHODS

The experiment was carried out at Embrapa Swine & Poultry Farm, Concórdia-SC, Brazil. Macrophytes were cultivated in a Raceway reactor (capacity of 12,000 L), installed in a greenhouse with humidity and temperature control. For the nutrient medium the adopted concentration was 60 and 30 mg L^{-1} of N-NH_3 for *Landoltia* and *Wolffia,* respectively. To start the cultivation the reactor was fulfilled with 3,000L of swine wastewater based complex medium (up to 15 cm reactor depth), followed by addition of one kilogram of fresh biomass to a restrict space (±10% surface area). The biomass was kept for 4-5 days in this condition for acclimatization and density build-up, with gradual expansion until complete coverage of the surface. At day 20, the total volume of *Wolfia* and *Landoltia* biomass were collected and weighed (fresh mass) and, after desiccating, the dry mass yield was evaluated. Nutritional and bacteriological parameters were determined to assess the quality of the produced biomass of both species as listed in the Table 1.

3 RESULTS & DISCUSSION

The yield obtained for *Landoltia* grown in a raceway reactor was 7% with 22 kg of fresh mass and 2 kg of dry mass, after culturing for 20 days (Figure 1) which amount is equivalent to 7,000 kg.ha^{-1} while *Wolfia* produced 13 kg of fresh mass and 840 g of dry

mass (4,333 kg.ha^{-1} f.m.) was achieved (Figure 2). Both in continuous cultivation in preliminary conditions. Comparatively, in the same

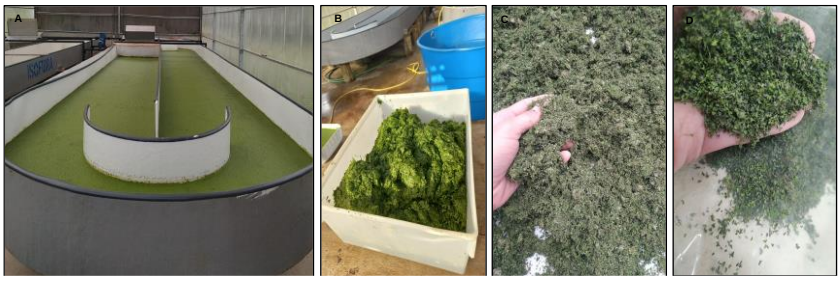

Figure 1 Growth of *Landoltia punctate* in a raceway reactor (A). Harvested fresh biomass (B), dried biomass in a greenhouse (C) and an oven-dried biomass (50º C) (D)

cultivation area, soybean productivity was 3,508 kg.ha^{-1} in 2023 and maize average productivity was 6, 057 kg.ha^{-1} in 2022-2023 (Conab 2023) suggesting macrophytes can have a higher productivity than soybean. It is noteworthy that after establishing ideal conditions, the accumulated fresh mass for *Wolffia* was 30 kg (1,95 kg.ha^{-1} f.m.) in three harvests during 15 days of cultivation.

Figure 3. Growth of *Wolffia brasiliensis* in a raceway reactor (A) fresh biomass collected (B) dry biomass after greenhouse environment (C) and dry biomass after dried in a stove at T= 50 ºC (D).

Figure 2 Growth of *Wolfia brasiliensis* in a raceway reactor (A). Harvested fresh biomass (B), dried biomass in a greenhouse (C) and an oven-dried biomass (50º C) (D)

Regarding the nutritional aspect, *Landoltia* and *Wolffia* showed good rates of carbohydrates and protein accumulation, as well as the energy expressed in the values of total digestible nutrients (Table 1). In turn, Buss (2015) found that *Wolffia brasiliensis* accumulated a higher carbohydrate content (41,48%) when grown in a natural lake compared to cultures in a wastewater decantation pond (27,04%) and a carbohydrate polishing pond (16,25%). In the latter media, considered impacted lakes, protein was in the range of 44%. In the present study, the crude protein content was lower than the obtained by Buss (2015), suggesting duckweeds respond adaptively to the nutrient concentration of the medium, mainly by the conversion of nitrogen into protein content.

Table 1 Centesimal and microbiological characterization of *Landoltia* and *Wolffia* dry biomass after growth in a pilot scale raceway reactor.

Parameters	Unit	Dry Mass	
		Landoltia	*Wolffia*
Total carbohydrates	%	35,00	33,00
Crude Fiber		27,89	34,46
Fiber in Neutral Detergent (FND)		20,63	24,57
Lignin		2,62	3,15
Lipids		1,30	1,40
Total Digestible Nutrients (TDN)		77,27	75,43
Crude Protein		29,87	27,39
Moisture (65 ºC)		<0,01	<0,01
Thermotolerant Coliforms	UFC 100 mL^{-1}	<1,0x10 ^{1}	<1,0x10 ^{1}
Salmonela sp.		absence	absence

Nutritional parameters showed differences on nutrient accumulation between the two species, with fibers and lignin especially higher in *Wolffia* and slightly lower in carbohydrates, nutrients and protein, compared to *Landoltia* (Table 1). Even devoid the roots, *Wolffia* showed a good absorption of nutrients from the wastewater. Improvement of the bioreactor structure to allow increment of oxygen and nutrients transference rate from medium will be implemented aiming a more nutrient rich biomass.

Regarding the bacteriological quality, no presence of thermotolerant coliforms (*E. coli*) and *Salmonella* bacteria was observed in both macrophytes biomasses (Table 1), indicating it has good sanitary quality.

4 CONCLUSION

The *Landoltia punctata* and *Wolffia brasiliensis* biomass can be scaled up without high investments, but for the yield enhancement the oxygen and nutrients transference rates of the cultivation medium must be adjusted. Therefore, a nutrient rich and safe green biomass for animal feed can be produced by promoting *on farm* nutrient cycling while using duckweeds as bioremediators of swine wastewater.

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