March 2014, letter #5

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"Duckweed System Fertilization Q@A" with Dr. Louis Landesman

Journal of the First Year of GreenSun Products, LLC

New research publications

ISCDRA

"Duckweed System Fertilization Q@A" with Dr. Louis Landesman

Excerpts from the April 8, 2014 ILA Round Table online workshop series on duckweed production

Dr. Landesman is an international consultant, researcher, and lecturer specializing in aquaculture and fisheries. His expertise includes extensive research on CAFO wastewater remediation with duckweed. He is a founding member of the ILA.

We welcome Dr. Landesman today to take your questions on nutrient fertilization of duckweed production systems.

Q: What sort of nutrients can be used for duckweed production?

A: The beauty of duckweed is that you can use a wide variety of nutrient sources -manure, waste water treatment, fertile water from a lake or pond and chemical fertilizers. As long as the salinity is less than five parts per thousand, you are flexible as to what you can use/ another plus for duckweed are that it prefers ammonia as NH4+ to nitrates, making it unique among plants. Micronutrient requirements are similar to other plants. Most commercial fertilizers such as Miracle Gro already have micronutrients included. I use a good 20-20-20 commercial greenhouse fertilizer on occasion. Organic off-the-shelf fertilizer is good but can be more expensive.

Q: When just getting started in duckweed production, would you recommend using a chemical or animal-based fertilizer?

A: I would use chemical fertilizer to begin with and then gradually switch to organic. Organic fertilizers are more changeable and unpredictable.

Q: If you want to achieve the highest level of protein, what is the best level of nitrogen while minimizing costs of chemical fertilizer?

A: 30 per cent nitrogen when compared to phosphorus and potassium. You can go higher – as high as 50 per cent nitrogen. Ammonium nitrate works well.

Q: Given that a pH of 7 is optimum, what would you add to re-stabilize a pH that jumps because of algal respiration?

A: Add lots of bicarbonate in the form of sodium or calcium- either will increase hardness or stabilize the pH.

Q: Outdoor systems require more controls. What parameters do you have to monitor and *adjust?*

A: pH, hardness, alkalinity and temperature. In very hot and sunny seasons or climates you can partially shade the duckweed culture with netting without reducing growth.

Q: What animal fertilizers rank at the top for duckweed growth?

A: Cattle manure, pig manure, and horse manure, in that order. Chicken manure is the least effective.

Q: There has been talk of using brackish or saline water for duckweed production. What are your thoughts on this?

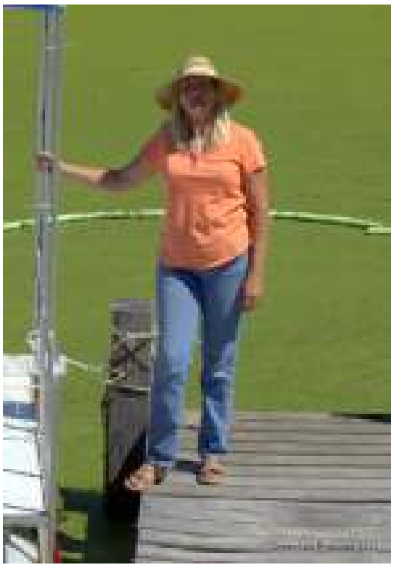
A: Avoid using brackish or saline water. Wolffia or water meal may be more tolerant of brackish water. The literature says 5 parts per thousand as the maximum salinity tolerated by duckweed.

Q: How do you approach using domestic wastewater for duckweed production in terms of nutrient loads?

A: If you plan to use wastewater, especially domestic wastewater, do a series of dilution trials first – i.e. 1:10 dilution with fresh water, 1:5, 1:1, etc.... This will give you optimal nutrient loads for your particular species and setting. Duckweed may not grow on full strength domestic wastewater. Concluding thoughts: Duckweed growing is still an art. You must practice growing it-there is no substitute for trial and error.

Dr. Landesman can be reached at llandesman@vsu.edu or through his website at http://www.duckweed49.com for more details.

Journal of the First Year of GreenSun Products, LLC

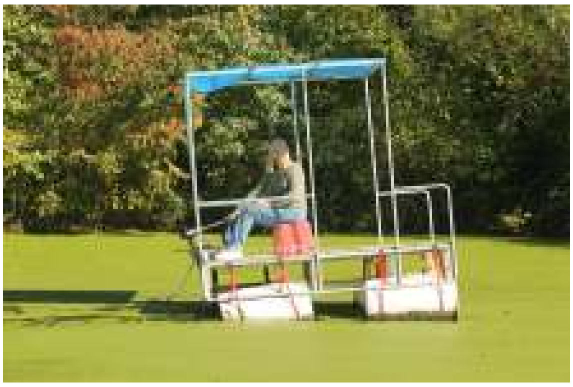


GreenSun is the first, if not one of the first, commercial duckweed farms in the United States. Founder, Tamra Fakhoorian shares what it takes to bootstrap a duckweed farm into existence.

January 1, 2013 I officially announced to my family that I was starting my own duckweed production and processing company for pet food. Why pet food? Better margins than high volume animal feedstock. Plus, more fun!

February Established GreenSun Products as an LLC in the state of KY. Developed several pet food formulations with varying duckweed species that I had on hand, and began testing of same. Filed for provisional patents on formulations.

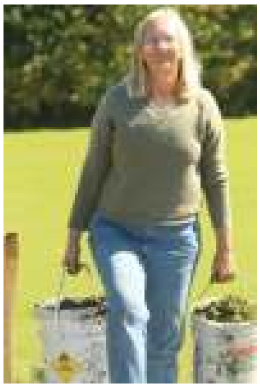
March Negotiated with owner of several unused catfish ponds for leasing for duckweed production. Built prototypes of various duckweed harvesting boats and harvesting systems. My goal was to build as sustainable a business as possible.



Early April Sourced duckweed seed stock locally for my first pond and began monitoring its growth habits. Worked with a neighboring farmer to dig a ninety foot test pond for doing controlled vs wild pond system comparisons. Designed a solar boat with aluminum framing tied to fifty-five gallon drums, and did the learning curve to come up with a solar panel charging system for the trolling motor.. "Yaught" was inaugurated on a windy,cold spring day on a five acre pond. Good thing it worked well, because I forgot to bring oars!

Mid-April Began seeding ponds and building pier on my primary pond. Had to get over fear of ticks, snakes, and poison ivy as all were present in great numbers at the pond sites.

May First harvests from both wild and controlled systems for open air solar drying tests showed that controlled pond growth was faster and responded better to organic and non-organic fertilizer than larger open wild ponds. Need to continue tests throughout the season though. Built several test solar hot air drying systems before settling on an indirect solar dryer with forced hot air from a solar tunnel- 20 ft. model. (Thanks to Paul Skillicorn for his great advice.) Tested lots of netting material in order to build thirty flexible yet rigid screens for drying duckweed on. My kitchen served as workshop. My kids simply got used to it. We also held our first open house for GreenSun Products, showcasing the control pond and several duckweed products. Duck hunters loved it. Local farmers were fascinated but needed financial proof of success. A news reporter came from Mayfield Messenger and did a nice story on GreenSun. I am now officially the "duckweed lady" in my hometown. Designed and ordered framework for a fifty foot long solar dryer tunnel.



June Began hand harvesting 300 pounds of duckweed a day for current 20 ft test solar dryer. (Ugh.. carrying all that duckweed in buckets up wooden steps every day!)

Friend, Ron Putt, an Auburn, Alabama engineer helped me design a washing station large enough to handle two tons of freshly harvested duckweed an hour. He is sourcing parts and will retrofit an existing conveyor system. Installed the fifty foot long solar dryer built

by Vegan Metal Fabricators, Sedalia, Kentucky. Meant to be temporary. Very effective! Solar chimney aides in moist air removal and large fans push 140 degree F air through all thirty flex shelves quite well.

July Began harvesting 800 lb batches of duckweed in just a few minutes via mechanical pumping (ahhh....SO much easier!). Duckweed is doubling between one and two days. My dryer can barely keep up with it.

Worked with Susan Metzger of Renaissance Marketing to understand customer needs. Began working with my graphics designer on packaging and labeling. Optimized a temporary washing and dewatering system. All water drained down to my primary pond. Dewatering system cuts drying time by 50%!!!

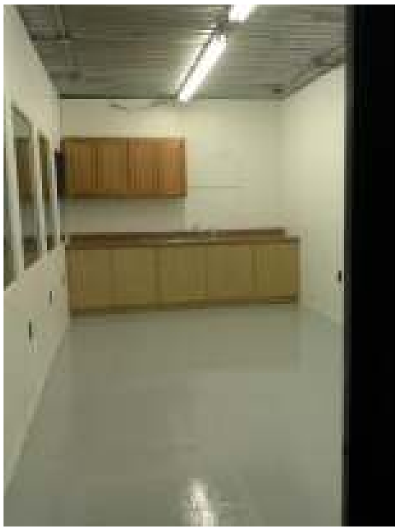
August Began pelletizing duckweed formulations. Built an outdoor washing station platform. Transported new duckweed washing station from Alabama, installed and did test runs. New conveyer belt material may or may not be the best choice due to volumes of water and duckweed being processed.

Late August Presented on GreenSun's progress and represented the ILA at the ICDRA conference at Rutgers University. Confirmed processing plant location after months of searching to that of a 22,000 sq. ft. warehouse in Mayfield, Kentucky. Washing belt on conveyor broke and I had to resort to washing by hand again- by the truckload!

October Began shutting down pond operations for winter and dismantled/reassembled solar dryer tunnel at new processing facility. Celebration time! We hosted GreenSun's first open house in conjunction with Mother Earth News. It was a crackers and wine event beside the ponds with interviews by a local newspaper and television station. The news coverage resulted in 422 hits on my blog, www.DuckweedFarming.com in one day! Several visits by state agriculture and legislative officials were very positive.

November Turned my energy to firing up utilities of the processing plant. Began remodel of an office space inside the warehouse for conversion to a wet laboratory.

December Built lab cabinetry, ran plumbing and sewer lines. Sourced wet chemistry equipment and chemicals. Designing new hybrid solar/gas assist dryer for spring production. Began the task of sourcing new funding for next phase of GreenSun. Refined my business plan and actually took a week-long break over New Years. Something about a road trip to help clarify my vision- works every time!



Note: While I admit to doing much of the actual elbow grease, I could not have done it without the wisdom and support of the following folks: Paul Skillicorn, Mark Johnson, Ron Putt, Susan Metzger, Gina McCord, Lewis Landesman, Ryan Integlia, Hamdi Shaar, and all ILA Round Table members. Many, many thanks to each of you.

Tamra can be reached at tamraf9@gmail.com or by phone at +1 270-356-0208, or via her website in http://www.DuckweedFarming.com

New research publications

The effect of aeration and effluent recycling on domestic wastewater treatment in a pilot-plant system of duckweed ponds

Ben-shalom, Miriam; Shandalov, Semion; Brenner, Asher; et al.

WATER SCIENCE AND TECHNOLOGY 69: 350-357 (2014)

Three pilot-scale duckweed pond (DP) wastewater treatment systems were designed and operated to examine the effect of aeration and effluent recycling on treatment efficiency. Each system consisted of two DPs in series fed by pre-settled domestic sewage. The first system (duckweed+conventional treatment) was 'natural' and included only duckweed plants. The second system (duckweed aeration) included aeration in the second pond. The third system (duckweed+aeration+circulation) included aeration in the second pond and effluent recycling from the second to the first pond. All three systems demonstrated similarly efficient removal of organic matter and nutrients. Supplemental aeration had no effect on either dissolved oxygen levels or on pollutant removal efficiencies. Although recycling had almost no influence on nutrient removal efficiencies, it had a positive impact on chemical oxygen demand and total suspended solids removals due to equalization of load and pH, which suppressed algae growth. Recycling also improved the appearance and growth rate of the duckweed plants, especially during heavy wastewater loads.

Bioaugmentation involving a bacterial consortium isolated from the rhizosphere of Spirodela polyrhiza for treating water contaminated with a mixture of four nitrophenol isomers

Kristanti, Risky Ayu; Toyama, Tadashi; Hadibarata, Tony; et al.

RSC ADVANCES 4: 1616-1621(2014)

A flask-scale laboratory study was performed to assess the bioaugmentation of water contaminated with a mixture of 2-nitrophenol, 3-nitrophenol, 4-nitrophenol and 2,4-dinitrophenol by using a bacteria consortium consisting of three nitrophenol-degrading bacteria strains (Pseudomonas sp. strain MFR-1, Pseudomonas sp. strain PFR-1 and Rhodococcus sp. strain DFR-1), reinoculated into the roots of Spirodela polyrhiza. The selected strains were colonized into the root at approximately 10(4) to 10(6) colony-forming units (CFU per plant). The high populations remained stable through five sequential two-days degradation cycles and complete nitrophenol removal was achieved within five-repeated cycles. Hence, inoculation of subjected degraders into the roots of aquatic plants is an effective treatment for nitrophenol-contaminated effluents or aquatic resources.

The Lemna Bioassay: Contemporary Issues as the Most Standardized Plant Bioassay for Aquatic Ecotoxicology

Mkandawire, Martin; Da Silva, Jaime A. Teixeira; Dudel, E. Gert

CRITICAL REVIEWS IN ENVIRONMENTAL SCIENCE AND TECHNOLOGY 44: 154-197 (2014)

The Lemna bioassay is one of the most standardized higher plant bioassays for assessing the impacts of contaminants in aquatic environments. The simple anatomy and ease with which Lemna sp. can be handled makes them ideal test organisms. They have been used to predict the cytotoxic, cytogenetic, and mutagenic effects of several chemical pollutants including, inter alia, polycyclic aromatic hydrocarbons, heavy metals, metalloids, organometallic compounds, pesticides, pharmaceuticals, radionuclides, and pharmaceuticals. However, there is still some contention as to the exact scope of application and definition of the Lemna bioassay regarding its accuracy of prediction and toxicity assessment. In this article, we review some critical issues on the development and effective use of the Lemna bioassay.

Arsenic uptake by Lemna minor in hydroponic system

Goswami, Chandrima; Majumder, Arunabha; Misra, Amal Kanti; et al.

INTERNATIONAL JOURNAL OF PHYTOREMEDIATION 16: 1221-1227 (2014)

Arsenic is hazardous and causes several ill effects on human beings. Phytoremediation is the use of aquatic plants for the removal of toxic pollutants from external media. In the present research work, the removal efficiency as well as the arsenic uptake capacity of duckweed Lemna minor has been studied. Arsenic concentration in water samples and plant biomass were determined by AAS. The relative growth factor of Lemna minor was determined. The duckweed had potential to remove as well as uptake arsenic from the aqueous medium. Maximum removal of more than 70% arsenic was achieved at initial concentration of 0.5mg/l arsenic on 15th day of experimental period of 22 days. Removal percentage was found to decrease with the increase in initial concentration. From BCF value, Lemna minor was found to be a hyperaccumulator of arsenic at initial concentration of 0.5 mg/L, such that accumulation decreased with increase in initial arsenic concentration.

Bioremediation of an iron-rich mine effluent by Lemna minor

Teixeira, S.; Vieira, M. N.; Espinha Marques, J.; et al.

INTERNATIONAL JOURNAL OF PHYTOREMEDIATION 16: 1228-1240 (2014)

Contamination of water resources by mine effluents is a serious environmental problem. In a old coal mine, in the north of Portugal (SAo Pedro da Cova, Gondomar), forty years after the activity has ended, a neutral mine drainage, rich in iron (FE) it stills being produced and it is continuously released in local streams (Ribeiro de Murta e Rio Ferreira) and in surrounding lands.The species Lemna minor has been shown to be a good model for ecotoxicological studies and it also has the capacity to bioaccumulate metals.The work aimed test the potential of the species L. minor to remediate this mine effluent, through the bioaccumulation of Fe, under greenhouse experiments and, at the same time, evaluate the time required to the maximum removal of Fe. The results have shown that L. minor was able to grow and develop in the Fe-rich effluent and bioaccumulating this element. Throughout the 21days of testing it was found that there was a meaningful increase in the biomass of L. minor both in the contaminated and in the non-contaminated waters. It was also found that bioaccumulation of Fe (iron) occurred mainly during the first 7days of testing. It was found that L. minor has potential for the bioremediation of effluents rich in iron.

Cadmium removal by Lemna minor and Spirodela polyrhiza

Chaudhuri, Devaleena; Majumder, Arunabha; Misra, Amal K.; et al.

INTERNATIONAL JOURNAL OF PHYTOREMEDIATION 16: 1119-1132 (2014)

The present study investigates the ability of two genus of duckweed (Lemna minor and Spirodela polyrhiza) to phytoremediate cadmium from aqueous solution. Duckweed was exposed to six different cadmium concentrations, such as, 0.5, 1.0, 1.5, 2.0, 2.5, and 3.0mg/L and the experiment was continued for 22days. Water samples were collected periodically for estimation of residual cadmium content in aqueous solution. At the end of treatment period plant samples were collected and accumulated cadmium content was measured. Cadmium toxicity was observed through relative growth factor and changes in chlorophyll content. Experimental results showed that Lemna minor and Spirodela polyrhiza were capable of removing 42-78% and 52-75% cadmium from media depending upon initial cadmium concentrations. Cadmium was removed following pseudo second order kinetic model. Maximum cadmium accumulation in Lemna minor was 4734.56mg/kg at 2mg/L initial cadmium concentration and 7711.00mg/kg in Spirodela polyrhiza at 3mg/L initial cadmium concentration at the end of treatment period. Conversely in both cases maximum bio-concentration factor obtained at lowest initial cadmium concentrations, i.e., 0.5mg/L, were 3295.61 and 4752.00 for Lemna minor and Spirodela polyrhiza respectively. The present study revealed that both Lemna minor and Spirodela polyrhiza was potential cadmium accumulator.

Sustainable Removal of Nitrophenols by Rhizoremediation Using Four Strains of Bacteria and Giant Duckweed (*Spirodela polyrhiza*)

Kristanti, Risky Ayu; Toyama, Tadashi; Hadibarata, Tony; et al.

WATER AIR AND SOIL POLLUTION 225 (4) Article Number: 1928 (2014)

We examined the effectiveness of rhizoaugmentation for treating water contaminated with the nitrophenols (NPs), 2-NP, 3-NP, 4-NP, and 2,4-dinitrophenol (2,4-DNP) using NP-degrading bacteria. We used 2-NP-degrading Pseudomonas sp. (strain ONR1), 3-NP-degrading Cupriavidus sp. (MFR2), 4-NP-degrading Rhodococcus sp. (PKR1), 2,4-DNP-degrading Rhodococcus sp. (DNR2), and giant duckweed (Spirodela polyrhiza). The four bacterial strains readily colonized Spirodela roots, as approximately 1x 10(5) colony-forming units [CFUs] plant(-1) to 10(6)-10(7) CFU plant(-1). The higher populations remained stable through five sequential 2-day degradation cycles and completely removed all four NPs within each cycle. The root-bacteria association also successfully treated wastewater effluent contaminated with NPs; 52-71 % of 2-NP and 100 % of 3-NP, 4-NP, and 2,4-DNP were removed within each of five 2-day cycles. These results demonstrate the potential of rhizoaugmentation to achieve efficient and sustainable treatment of NP-contaminated waters.

Reproducibility of effects of homeopathically potentised glibberellic acid on the growth of Lemna gibba L. in a randomised and blinded bioassay

Majewsky, Vera; Scherr, Claudia; Arlt, Sebastian Patrick; et al.

HOMEOPATHY 103: 113-126 (2014)

Background: Reproducibility of basic research investigations in homeopathy is challenging. This study investigated if formerly observed effects of homeopathically potentised gibberellic acid (GA(3)) on growth of duckweed (Lemna gibba L.) were reproducible.

Methods: Duckweed was grown in potencies (14x-30x) of GA(3) and one time succussed and unsuccussed water controls. Outcome parameter area-related growth rate was determined by a computerised image analysis system. Three series including five independent blinded and randomised potency experiments (PE) each were carried out. System stability was controlled by three series of five systematic negative control (SNC) experiments. Gibbosity (a specific growth state of L. gibba) was investigated as possibly essential factor for reactivity of L. gibba towards potentised GA(3) in one series of potency and SNC experiments, respectively.

Results: Only in the third series with gibbous L. gibba L. we observed a significant effect (p= 0.009, F-test) of the homeopathic treatment. However, growth rate increased in contrast to the former study, and most biologically active potency levels differed. Variability in PE was lower than in SNC experiments. The stability of the experimental system was verified by the SNC experiments.

Conclusions: Gibbosity seems to be a necessary condition for reactivity of L. gibba to potentised GA(3). Further still unknown conditions seem to govern effect direction and the pattern of active and inactive potency levels. When designing new reproducibility studies, the physiological state of the test organism must be considered. Variability might be an interesting parameter to investigate effects of homeopathic remedies in basic research.

Duckweed in bloom: the 2nd International Conference on Duckweed Research and Applications heralds the return of a plant model for plant biology

By: Lam, Eric; Appenroth, Klaus J.; Michael, Todd; et al.

PLANT MOLECULAR BIOLOGY 84: 737-742 (2014)

More than 50 participants from around the world congregated at Rutgers University for 4 days to discuss the latest advances in duckweed research and applications. Among other developments in the field, exciting new information related to duckweed including genome sequencing, improved genetic transformation, and the identification of a novel plant growth promoting substance from bacteria were reported.

The logistic growth of duckweed (Lemna minor) and kinetics of ammonium uptake

Zhang, Kun; Chen, You-Peng; Zhang, Ting-Ting; et al.

ENVIRONMENTAL TECHNOLOGY 35: 562-567 (2014)

Mathematical models have been developed to describe nitrogen uptake and duckweed growth experimentally to study the kinetics of ammonium uptake under various concentrations. The kinetics of duckweed ammonium uptake was investigated using the modified depletion method after plants were grown for two weeks at different ammonium concentrations (0.5-14 mg/L) in the culture medium. The maximum uptake rate and Michaelis-Menten constant for ammonium were estimated as 0.082 mg/(g fresh weighth) and 1.877 mg/L, respectively. Duckweed growth was assessed when supplied at different total nitrogen (TN) concentrations (1-5 mg/L) in the culture medium. The results showed that the intrinsic growth rate was from 0.22 to 0.26 d(-1), and TN concentrations had no significant influence on the duckweed growth rate.

Effect of cold water extracts of Acacia modesta Wall. and Glycyrrhiza glabra Linn. on Tribolium castaneum and Lemna minor

Nazeefullah, Sayed; Dastagir, Ghulam; Ahmad, Bashir

PAKISTAN JOURNAL OF PHARMACEUTICAL SCIENCES 27: 217-222 (2014)

The aim of the present study was to introduce an alternative way for insects control through biodegradable plants materials. The different cold water extracts dilutions of Acacia modesta and Glycyrrhiza glabra were tested against Tribolium castaneum. The extracts dilutions of both plants caused mortality of the Tribolium castaneum. ANOVA revealed that dilutions and plants were highly significant. The interaction between plants and dilutions was also significant at $P < 0.05$. Phytotoxic activity showed that dilutions of

Acacia modesta and Glycyrrhiza glabra extracts significantly inhibited the growth of Lemna minor. ANOVA showed that dilutions of both plants extracts were significant at $P < 0.05$.

Synchronized urban wastewater treatment and biomass production using duckweed Lemna gibba L.

Verma, Rashmi; Suthar, Surindra

ECOLOGICAL ENGINEERING Volume: 64: 337-343 (2014)

This work compiles results of utilizing duckweed Lemna gibba L. in sewage wastewater treatment and converting wastewater nutrients into protein and carbohydrate rich weed biomass. A total of four different strengths of wastewater (100, 75, 50 and 25%) were used to build experimental setups and changes in chemical properties of wastewater were measured at the end. L. gibba L. caused a decrease in pH (9-13%), EC (13-26%), NO3--N (42-64%), SO4-2 (35-82%), total phosphorus (37-54%), totNa (44-75%), totK (45-76%) and totCa (51-72%). The biomass yield in L. gibba L. ranged between 132.62 and 200.95 g/m(2) day (fresh weight) or 23.87 and 36.17 g/m(2) day (dry weight); about 24.6-36.17% higher than initial level in all experimental setups. The weed biomass was 24.6-52.8% higher than initial level in all set-ups. The yield rate of protein and carbohydrate ranged 1.19-1.95 g/m(2) day (dry weight) and 22.72-35.58 g/m(2) day (dry weight), respectively in duckweed systems. The high carbohydrate and protein yield in L. gibba advocates the utility of the weed in animal feed and bioethanol production. Results suggested that L. gibba can be utilized effectively for designing of a synchronized wastewater treatment and biomass harvesting system.

Stimulation of nitrogen removal in the rhizosphere of aquatic duckweed by root exudate components

Lu, Yufang; Zhou, Yingru; Nakai, Satoshi; et al.

PLANTA 239: 591-603 (2014)

Plants can stimulate bacterial nitrogen (N) removal by secretion of root exudates that may serve as carbon sources as well as non-nutrient signals for denitrification. However, there is a lack of knowledge about the specific non-nutrient compounds involved in this stimulation. Here, we use a continuous root exudate-trapping system in two common aquatic duckweed species, Spirodela polyrrhiza (HZ1) and Lemna minor (WX3), under natural and aseptic conditions. An activity-guided bioassay using denitrifying bacterium Pseudomonas fluorescens showed that crude root exudates of the two species strongly enhanced the nitrogen-removal efficiency (NRE) of P. fluorescens ($P < 0.05$) under both conditions. Water-insoluble fractions (F) obtained under natural conditions stimulated NRE to a significant extent, promoting rates by about 30 %. Among acidic, neutral and basic fractions, a pronounced stimulatory effect was also observed for the neutral fractions from HZ1 and WX3 under both conditions, whereas the acidic fractions from WX3 displayed an inhibitory effect. Analysis of the active fractions using gas chromatography/mass spectrometry (GC/MS) revealed that duckweed released fatty acid methyl esters and fatty acid amides, specifically: methyl hexadecanoate, methyl (Z)-7-hexadecenoate, methyl dodecanoate, methyl-12-hydroxystearate, oleamide, and erucamide. Methyl (Z)-7-hexadecenoate and erucamide emerged as the effective N-removal stimulants (maximum stimulation of 25.9 and 33.4 %, respectively), while none of the other tested compounds showed stimulatory effects. These findings provide the first evidence for a function of fatty acid methyl esters and fatty acid amides in stimulating N removal of denitrifying bacteria, affording insight into the "crosstalk" between aquatic plants and bacteria in the rhizosphere.

Dual application of Duckweed and Azolla plants for wastewater treatment and renewable fuels and petrochemicals production

Muradov, Nazim; Taha, Mohamed; Miranda, Ana F.; et al.

BIOTECHNOLOGY FOR BIOFUELS 7: Article Number: 30 (2014)

Background: Shortages in fresh water supplies today affects more than 1 billion people worldwide. Phytoremediation strategies, based on the abilities of aquatic plants to recycle nutrients offer an attractive solution for the bioremediation of water pollution and represents one of the most globally researched issues. The subsequent application of the biomass from the remediation for the production of fuels and petrochemicals offers an ecologically friendly and cost-effective solution for water pollution problems and production of value-added products.

Results: In this paper, the feasibility of the dual application of duckweed and azolla aquatic plants for wastewater treatment and production of renewable fuels and petrochemicals is explored. The differences in absorption rates of the key wastewater nutrients, ammonium and phosphorus by these aquatic macrophytes were used as the basis for optimization of the composition of wastewater effluents. Analysis of pyrolysis products showed that azolla and algae produce a similar range of bio-oils that contain a large spectrum of petrochemicals including straight-chain C10-C21 alkanes, which can be directly used as diesel fuel supplement, or a glycerin-free component of biodiesel. Pyrolysis of duckweed produces a different range of bio-oil components that can potentially be used for the production of "green" gasoline and diesel fuel using existing techniques, such as catalytic hydrodeoxygenation.

Conclusions: Differences in absorption rates of the key wastewater nutrients, ammonium and phosphorus by different aquatic macrophytes can be used for optimization of composition of wastewater effluents. The generated data suggest that the composition of the petrochemicals can be modified in a targeted fashion, not only by using different species, but also by changing the source plants' metabolic profile, by exposing them to different abiotic or biotic stresses. This study presents an attractive, ecologically friendly and cost-effective solution for efficient bio-filtration of swine wastewater and petrochemicals production from generated biomass.

Biologically Active Compounds from Lemna minor S. F. Gray

Vladimirova, I. N.; Georgiyants, V. A.

PHARMACEUTICAL CHEMISTRY JOURNAL 47: 599-601 (2014)

The chemical composition of biologically active compounds of the common duckweed (Lemna minor) was studied. A titrimetric method was used to assay the total iodine content, a spectrophotometric method was used for flavonoids, expressed as luteolin-7-glucoside; gas chromatography/mass spectrometry was used to identify 32 lipophilic substances of various chemical groups. Atomic emission spectrographic studies identified 14 macro and trace elements in the common duckweed. Studies of biologically active substances in the common duckweed are of interest for further investigation of its pharmacological properties with the aim of seeking new medicinal agents.

Different toxicity mechanisms between bare and polymer-coated copper oxide nanoparticles in Lemna gibba

Perreault, Francois; Popovic, Radovan; Dewez, David

ENVIRONMENTAL POLLUTION 185: 219-227 (2014)

In this report, we investigated how the presence of a polymer shell (poly(styrene-co-butyl acrylate) alters the toxicity of CuO NPs in Lemna gibba. Based on total Cu concentration, core-shell CuO NPs were 10 times more toxic than CuO NPs, inducing a 50% decrease of growth rate at 0.4 g l(-1) after 48-h of exposure while a concentration of 4.5 g l(-1) was required for CuO NPs for a similar effect. Toxicity of CuO NPs was mainly due to NPs solubilization in the media. Based on the accumulated copper content in the plants, core-shell CuO NPs induced 4 times more reactive oxygen species compared to CuO NPs

and copper sulfate, indicating that the presence of the polymer shell changed the toxic effect induced in L gibba. This effect could not be attributed to the polymer alone and reveals that surface modification may change the nature of NPs toxicity.

Second-generation bioethanol production from water hyacinth and duckweed in Izmir: A case study

Bayrakci, Asiye Gul; Kocar, Gunnur

RENEWABLE & SUSTAINABLE ENERGY REVIEWS 30: 306-316 (2014)

Water hyacinth and duckweed are both aquatic plants that live in clean water and wastewater. They have excellent filtration ability of nitrogen and supply oxygen to water while cleaning it up. The vegetative reproduction rates of these two plants are really high and it is about 8 days that of duplication time. Because of these features, water hyacinth and duckweed are useful for wastewater treatment. Also production of second-generation bioethanol from these plants is proper due to their cellulose ratios and carbohydrate potentials. In this study, the process of bioethanol production from water hyacinth and duckweed was studied by taking into account the potential of cultivation values and usability in wastewater treatment procedures in Izmir. Different microorganisms, yeast and bacteria (Saccharomyces cerevisiae, Pichia stipitis, and Clostridium thermocellum) have been considered to identify the best process of ethanol production, considering Turkey's policy regarding biofuels.

Uptake and toxicity of arsenic, copper, and silicon in Azolla caroliniana and Lemna minor

Rofkar, Jordan R.; Dwyer, Daryl F.; Bobak, Deanna M.

INTERNATIONAL JOURNAL OF PHYTOREMEDIATION 16: 155-166 (2014)

Here we report on the analysis of two aquatic plant species, Azolla caroliniana and Lemna minor, with respect to tolerance and uptake of co-occurring arsenic, copper, and silicon for use in engineered wetlands. Plants were cultured in nutrient solution that was amended with arsenic (0 or 20M), copper (2 or 78M), and silicon (0 or 1.8mM) either singly or in combination. We hypothesized that arsenic and copper would negatively affect the uptake of metals, growth, and pigmentation and that silicon would mitigate those stresses. Tolerance was assessed by measuring growth of biomass and concentrations of chlorophyll and anthocyanins. Both plant species accumulated arsenic, copper, and

silicon; L. minor generally had higher levels on a per biomass basis. Arsenic negatively impacted A. caroliniana, causing a 30% decrease in biomass production and an increase in the concentration of anthocyanin. Copper negatively impacted L. minor, causing a 60% decrease in biomass production and a 45% decrease in chlorophyll content. Silicon augmented the impact of arsenic on biomass production in A. caroliniana but mitigated the effect of copper on L. minor. Our results suggest that mixtures of plant species may be needed to maximize uptake of multiple contaminants in engineered wetlands.

Isolation of Vibrio cholera El Tor Inaba From Lemna minor and Eichhornia crassipens Roots in Veracruz, Mexico

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Background: During epidemic periods, the strain Vibrio cholera El Tor has been isolated from the aquatic macrophyte roots of Eichhornia crassipens and Lemna minor, suggesting that aquatic plants could be environmental reservoirs through either a non-specific association or a commensalism relationship. Therefore, it is important to understand V. cholera reservoirs in order to establish prevention strategies against this pathogen.

Objectives: Our interest was to determine whether V. cholera could be isolated and typified from L. minor and E. crassipens roots.

Materials and Methods: From 2004 to 2005, plants were collected from various ecological niches and the roots were used to isolate V. cholera. Standard bacteriological, biochemical and serological tests were used for its typification.

Results: In five out of the nine ecological niches explored, we collected either L. minor or E. crassipens, as these specimens cohabited only in two niches. V. cholera was isolated from both L. minor and E. crassipens roots. The isolated V. cholera showed the same biochemical characteristics as the pure V. cholera strain which was used as a control. The isolated V. cholera corresponded to V. cholera O1 El Tor Inaba, which is the same serotype related to the last outbreak in Mexico.

Conclusions: For first time V. cholera El Tor Inaba has been isolated several years after the last emergence of cholera in Mexico. A viable and cultivable V. cholera strain, sourced from freshwater niches in E. crassipens and L. minor roots, suggests the importance of these plants as a permanent aquatic reservoir for these organisms. The monitoring of E. crassipens and L. minor is the responsibility of health institutions in order to evaluate the ongoing risks.

Morphological and biochemical responses of Lemna minor l. (common duckweed) to ciprofloxacin

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Active substances of pharmaceuticals occur not only in hospital waste or surface waters but also in rivers and seas in concentrations exceeding 1 mu g/l. As far as research is concerned, it is important to analyze the effect of pharmaceuticals on organisms other than their target organisms. It was the aim of this study to determine the phytotoxicity of increasing concentrations of soluble and insoluble ciprofloxacin (0; 0.0002; 0.0005; 0.0008; 0.001; 0.005; 0.01; 0.05; 0.1; 0.313; 0.63; 1.25; 2.5; 5; 10; 20; 40 mM) towards lesser duckweed. The phytotoxicity was evaluated on the basis of chosen morphological and biochemical features. In line with increasing concentrations, both soluble and insoluble ciprofloxacin present in the water caused reduction in yield, growth rate, amount of fresh mass, and chlorophyll content in duckweed tissues, with dry mass slightly growing. 1.25 mM of soluble ciprofloxacin was a lethal dose for the duckweed, since it hindered the growth rate and yield by 100%. Insoluble ciprofloxacin did not induce such a toxic effect even in its highest dose. The toxic effect of soluble ciprofloxacin also modified the content of soluble carbohydrates. For concentrations above 1.25 mM of soluble ciprofloxacin, the content of sucrose, D-chiro-inositol and monosaccharides dropped to zero. In 5 mM of soluble ciprofloxacin, acute dehydration of the tissues (the growth rate inhibited by more than 100%) led to a rapid increase in the content of myo-inositol. Yet, the content of soluble carbohydrates in the tissues of lesser duckweed was low and did not exceed 1% of fresh mass of the tissues.