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Case Report

Algae Cultivation as Measure for the Sanitation of Organic Waste—A Case Study Based on the Alga *Galdieria sulphuraria* Grown on Food Waste Hydrolysate in a Continuous Flow Culture

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Abstract: Due to its growth under harsh acidic conditions, the microalga *Galdieria sulphuraria* may offer the opportunity to combine sanitation and the utilization of organic waste streams. To further deepen the knowledge of alternative waste treatment strategies that allow for holistic utilization, the control and removal of microbial contaminants via non-sterile heterotrophic *G. sulphuraria* on food waste hydrolysate were investigated in a continuous flow bioreactor culture. Furthermore, a substrate reservoir and harvested biomass were stored under non-sterile conditions over a period of 12 days. Despite the non-sterile conditions, the microbial load of the biomass could be kept under control. Neither the pathogen *Salmonella* sp. nor the coliform bacteria Escherichia coli could be found. Only nine counts per g of biomass were found for species belonging to *Enterococcus* spp., Enterobacteriacae, and moulds. Aerobic spore formers were counted with 2700 counts per g of biomass. Most of the aerobic mesophilic counts were formed by yeasts (1.5×10^6 vs. 1.3×10^6 counts per g biomass). The results revealed that, when using acidic growth conditions, contamination will not take over the culture; thus, the sterilization of waste materials can be skipped. It is assumed that such an approach can result in efficient processes for future waste-based bioeconomy strategies.

Keywords: bioeconomy; waste streams; bioprocess; sanitation; waste management

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1. Introduction

The increasing demand for food [1,2] and bioenergy [3] has led to an increase in waste streams that need to be managed in an efficient and sustainable way [4]. The proper management of organic wastes is a critical issue for society. The improper management of organic wastes, such as agricultural by-products, food waste, and manure, can result in environmental problems, including the release of greenhouse gases, soil, and water contamination [5,6], as well as pathogen proliferation [7].

The bioeconomy is a concept that refers to the production and use of renewable biological resources to replace fossil-based materials and energy. In this aspect, waste streams can be considered as valuable resources in the bioeconomy [8,9]. One approach to addressing the environmental issues associated with organic waste and allowing its utilization is through the use of heterotrophic algae-based bioprocesses for the conversion of organic wastes into valuable products, such as biofuels, chemicals, feed, and food, due to the ability of microalgae to make use of organic carbon and nitrogen compounds [10–13]. For example, *Galdieria sulphuraria* is a red alga that has been shown to be highly tolerant to extreme environmental conditions, such as high temperatures and low pHs [14,15]. The utilization of organic waste streams using *G. sulphuraria* typically involves the heterotrophic cultivation of the alga in a bioreactor, where the algae can use organic matter as a carbon, nitrogen, and phosphorus source for growth [16]. During this process, the organic matter

is converted into biomass, which can be harvested and converted into valuable products. *G. sulphuraria* is a promising candidate for the bioprocessing of wastes, as it can grow in harsh environments and utilize a wide range of carbon sources [17]. Furthermore, harsh conditions have been shown to suppress or even reduce the microbial load, including pathogens, and thus contribute not only to the sanitation of organic matter, but also to its utilization [18–21].

The use of *G. sulphuraria* offers a promising approach to the management of waste streams in the bioeconomy. By converting organic waste streams into valuable products, these bioprocesses can contribute to the development of a more sustainable and circular economy. Considering the capacity of *G. sulphuraria* to remove pollutants from municipal wastewater, further research should be undertaken to boost its contaminant removal ability, in order to utilize various waste streams [22]. For instance, more insight is needed into the applicability of *G. sulphuraria* for the sanitation of organic matter when the cultivation is carried out under continuous flow conditions with a non-sterile substrate. Therefore, the aim of this short report and communication was to cultivate *G. sulphuraria* under non-sterile conditions in the presence of hydrolysed food waste. Harvested biomass was stored at room temperature under non-sterile conditions and the microbial load was determined after 12 days of storage. It was expected that, when using acidic growth conditions, the sterilization of waste materials could be skipped and the implementation of a *G. sulphuraria*-based approach could result in efficient processes for future waste-based bioeconomy strategies.

2. Material and Methods

2.1. Organic Materials and Galdieria sulphuraria

Food waste was collected from households and consisted of bread, meat, and noodles. The food waste was stored at −20 °C until used in the experiments.

G. sulphuraria strain 21.92 was purchased from the Culture Collection of Algae (SAG, University of Goettingen, Goettingen, Germany), maintained in 100 mL flasks containing 20 mL of a cyanidium medium consisting of 4 g L^{-1} of glucose, 1 g L^{-1} of $(NH_4)_2SO_4$, 0.02 g L^{-1} of K_2HPO_4, and 0.02 g L^{-1} of $MgSO_4 \times 7H_2O$ at pH 2 and 45 °C, and shaken at 130 rpm on an orbital shaker. Subcultivation occurred once per week by adding 50 µL of an algae suspension to 20 mL of fresh cyanidium medium.

2.2. Food Waste Hydrolysis

For the medium preparation, 985.3 g (dry matter) of bread and 201.8 g (dry matter) of noodles were suspended in 2.5 L of water and mixed with 20 mL of gluco-amylase (Glucoamylase AN, ASA Spezialenzyme GmbH, Wolfenbuettel, Germany). After hydrolysis for 24 h at pH 4.6 and 55 °C in a 5 L bioreactor (Sartorius GmbH, Goettingen, Germany), the hydrolysis was stopped, and the suspension was centrifuged for 10 min at 14,025× *g*. The supernatant was acidified to pH 2 and kept frozen at −20 °C until used in the experiments.

Furthermore, to produce a source of free amino nitrogen (FAN), 238.2 g (dry matter) of pig and chicken meat was ground and suspended in 0.5 L of water. The supernatant was acidified to pH 5.0, mixed with 10 mL of protease (Protease S-02, ASA Spezialenzyme GmbH), and hydrolysed for 24 h at 55 °C in a 1 L bioreactor (Eloferm GmbH, Berlin, Germany). After the hydrolysis, the suspension was centrifuged for 10 min at 14,025× *g*. The supernatant was filtered through a 5–8 µm filter, acidified to pH 2, and kept frozen at −20 °C until used in the experiments.

The digestion of the bread and noodle wastes resulted in a medium containing 162.8 g L^{-1} of glucose, while the digestion of the meat waste resulted in a medium containing 3.6 g L^{-1} of FAN. For the final preparation of the feed substrate, 460.5 mL of bread and noodles hydrolysate was mixed with 37.5 mL of meat hydrolysate and 1 L of water, as well as being supplemented with 0.75 g L^{-1} of KH_2PO_4. The final feed substrate contained 47.4 g L^{-1} of glucose, 0.09 g L^{-1} of FAN, and 0.75 g L^{-1} of KH_2PO_4. The preparation was repeated whenever feed substrate was needed.

2.3. Batch and Continuous Flow Cultures

Batch and continuous flow cultures of *G. sulphuraria* were carried out in a 5 L stirred and aerated Sartorius bioreactor at 45 °C and pH 2. The stirrer speed was adjusted between 200 and 400 rpm in order to maintain a dissolved oxygen concentration above 20%. The hydrolysates from the bread, noodles, and meat hydrolyses were mixed and supplemented with water and KH_2PO_4 to obtain a medium containing 52 g L^{-1} of glucose, 0.7 g L^{-1} of FAN, and 0.9 g L^{-1} of phosphate, respectively. The working volume was 1.5 L. Antifoam was added when necessary and the pH was regulated by automatic additions of 1 M of HCl or 1 M of NaOH. The inocula were grown for 4 days in 100 mL conical flasks containing 50 mL of medium, as described in Section 2.2. A 3% (*v*/*v*) inoculum was used. Continuous flow culture was carried out at a dilution rate of 0.38 day^{-1}. The dilution rate was controlled the by means of a peristaltic pump. The flow was started after a five-day-long batch phase before the FAN became limited. Neither the equipment nor the hydrolysate were sterilized. Neither the hydrolysate nor the harvested cells were stored under sterile or cooled conditions.

2.4. Microbial Analysis

A microbial analysis of the *G. sulphuraria* biomass was carried out, as described earlier in [16].

The investigation of the microbiological status of the produced *G. sulphuraria* biomass focused on a determination of aerobic, mesophilic microbes, yeasts/molds, entero-bacteria, enterococci, *Escherichia coli*, and *Salmonella* sp. according to § 64 of the German Food and Feed Code (LFGB) (Amtliche Sammlung von Untersuchungsverfahren—ASU). Finally, the test for the presence of aerobic spore formers was performed according to the protocol (ASU L 00.00-88/2: 2015-06).

2.5. Chemical Analysis

A biochemical analysis of the produced *G. sulphuraria* biomass and a chemical analysis of the cultivation medium were carried out as described in [16]. The biochemical composition of the *G. sulphuraria* biomass was analysed using near infrared spectroscopy (Unity Scientific GmbH, Weiler bei Bingen, Germany). The FAN and phosphate were determined following a modified EBC-ninhydrin method and via the generation of molybdenum blue, respectively. The glucose was determined using HPLC (Shimadzu: LC-10AD pump, SIL-10AD au-to-sampler, CTO-10AD oven, refractive index detector RID-20A, CBM-20A communication module).

3. Results and Discussion

3.1. Cultivation of Galdieria sulphuraria

The continuous flow cultivation of *G. sulphuraria* is shown in Figure 1. The initial concentrations of the glucose, FAN, and phosphate were 52 g L^{-1}, 0.7 g L^{-1}, and 0.9 g L^{-1}, respectively. The lag-phase lasted for 2 days and, afterwards, all the nutrients were consumed. After five days, before the FAN became limited, the flow with a dilution rate of 0.38 day^{-1} was started. The composition of the feed substrate was described in Section 2.3. After the flow was started, the concentrations of the glucose, FAN, and phosphate levelled off at around 31 g L^{-1}, 0.1 g L^{-1}, and 0.1 g L^{-1}, respectively, and remained constant until the end of the cultivation after 12 days.

Between days 2 and 8, the OD increased exponentially from 0.8 to 42.7 AU. Assuming that the increase in the OD reflected the increase in the biomass, the growth rate was 0.62 day^{-1}. The growth rate was thus faster than the dilution rate and, eventually, the biomass concentration increased, even though the flow was started after 5 days. Nevertheless, after 8 days, most likely the FAN and/or phosphate became limited for *G. sulphuraria* and the OD levelled off at around 55 AU. The glucose concentration was not reduced below 31 g L^{-1}, and thus most of the glucose was removed unused with the dilution (Figure 1).

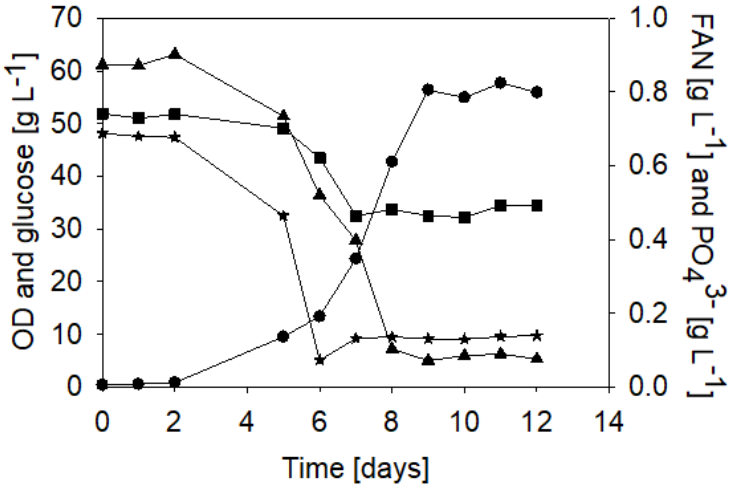

Figure 1. Development of optical density (OD, closed circle, left axis), glucose (closed square, left axis), free amino nitrogen (FAN, closed star, right axis), and phosphate (PO_4^{3-}, closed triangle, right axis) concentrations over time in the *Galdieria sulphuraria* continuous flow culture grown in food waste hydrolysate at a dilution rate of 0.38 day^{-1}.

Over the whole cultivation period of 12 days, 43.8 g (28.5 g L^{-1}) of biomass was produced. Based on *G. sulphuraria* biomass dry matter (*w*/*w*), the carbohydrate, protein, fat, and ash contents were 35.7%, 27.4%, 3.2%, and 4.9%, respectively, and thus may represent a promising source for proteins for various applications [23].

It is challenging to achieve a real steady state using a complex medium. Food waste hydrolysates may contain compounds in addition to the measured glucose, FAN, and phosphate serving as nutrients; thus, a detailed quantification of all nutrients is hard to achieve. However, a steady state is necessary to obtain biomass with a stable biochemical composition, and fluctuations in nutrients can have a considerable impact [24]. The biomass produced in this study was, in terms of its biochemical composition, most likely stable for 3 days (Figure 1.)

3.2. Sanitation of Waste Streams

Even though the continuous flow cultivation was carried out in non-sterile conditions and neither the substrate nor the produced biomass were stored under sterile and cooled conditions, the microbial load of the biomass could be kept under control. Neither *Salmonella* sp. nor *Escherichia coli* could be found. Only nine counts were found for species belonging to *Enterococcus* spp., Enterobacteriacae, and moulds. Aerobic spore formers were counted with 2700 counts per g of biomass. Most of the aerobic mesophilic counts were formed by yeasts (1.5×10^6 to 1.3×10^6 per g biomass). The yeast formation could already be seen during the cultivation as precipitate in the storage tank.

In a previous study using phototrophic *G. sulphuraria* cultures and wastewater, bacteria were deactivated. The faecal coliform count was reduced to a non-detectable level, resulting in a log reduction factor of over 7, while the total bacteria population was reduced by a log reduction factor of 1.7 [21]. In accordance with the authors, *Enterococcus faecalis* and *E. coli* were reduced by log reduction factors of 3.8 and 5.4, respectively. Pathogens such as *Pseudomonas aeruginosa*, *Salmonella enterica*, and *Staphylococcus aureus* were absent. These reductions were predominantly caused by the low pH. However, in another study, a synergistic effect of pH, temperature, sunlight, dissolved oxygen, and algae might have been responsible for the inactivation observed [19]. Sunlight and dissolved oxygen are expected to form reactive oxygen species that act against bacteria. Similar results were found in another study, where the effluent showed high abundances of *Aeromonas* sp., *Clostridium* sp., *Legionella* sp., and *Streptococcus* sp. However, they were absent in the effluent of the *G. sulphuraria*, which makes the use of disinfection measures unnecessary [18].

A reduction in the microbial load of liquid waste streams using phototrophic *G. sulphuraria* cultivation was clearly shown, as outlined above. However, when using solid waste or residue streams, such as digestate, agricultural residues, and food waste, in a heterotrophic *G. sulphuraria* cultivation, the pressure from the microbial loads of these streams is different. Food waste, for instance, has high water and organic matter contents that can facilitate the spread of microbial pathogens [25]. Furthermore, food waste may undergo a spoilage process, and it was recently concluded that pathogens and mycotoxins accumulating during spoilage can present a health hazard [26]. A measure for reducing the microbial load of food waste is drying [27]. However, the drying of a large amount of food waste is energy intensive and results in a reduction and not a complete removal of microbial contaminants such as pathogens. This would still challenge the use of dried food waste in microbial utilization strategies.

In a previous study, it was shown that, during the hydrolysis of digestate and the cultivation of *G. sulphuraria*, the counts of aerobic, mesophilic organisms could be subsequently reduced by a log reduction factor of 3. The remaining microorganisms were almost exclusively spore-forming ones, which were reduced by a log reduction factor of 2 during cultivation under acidic conditions [20]. In this study using non-sterile food waste hydrolysate, only a few spore-forming microorganisms and no pathogens were found. The majority of the contaminants found were yeast cells. Yeasts are known to survive at a low pH; thus, this could be as serious threat for non-sterile *G. sulphuraria* cultivation. However, the growth of the yeast cells could be kept under control and *G. sulphuraria* was the dominant species. Nevertheless, the results shown here (where yeasts were dominant) and earlier [20] (where spores were dominant) indicate the importance of a hygenization process that works independently of the applied substrates. The acidic growth conditions did not diminish all the organisms and allowed for control of them. However, in every case, a further treatment is necessary to diminish all organisms.

The current approaches to organic waste management, such as anaerobic digestion [28], composting [29], and vermicomposting [30], allow for a reduction in pathogens. Contrarily, incineration destroys pathogens [31] and antibiotic resistance genes [32]. For anaerobic digestion and a strongly contaminated feedstock, eliminations were achieved within 6.1 h, 5.5 h, and about 10 h for *Salmonella Senftenberg* W_{775}, *Enterococcus* spp., and *Ascaris suum*, respectively [28]. For vermicomposting, it can be shown that the homogenization of materials might be necessary before treatment to eliminate pathogens [30]. Temperature had a considerable effect on the reduction in *S. infantis* in a broiler litter during composition, and, in order to achieve a log reduction of 7, 1.5–1.5 days were needed at 60 °C, but 13.7–27.2 days were needed at 30 °C [33]. The elimination of pathogens such as *Salmonella enterica* was also achieved when hydrolysed waste streams were treated under acidic conditions for *G. sulphuraria* cultivations [20]. However, even though pathogens could be eliminated, there was still life in the form of remaining spore formers, which can grow-up again under favourable conditions. Nevertheless, except for incineration, no complete elimination of microorganisms can be achieved with the current approaches to waste management. Thus, the treatment of hydrolysed organic waste with *G. sulphuraria* and its acidic growth condition may represent an alternative.

4. Conclusions

Based on the results obtained in the present, as well as in previous studies, it could be shown that *G. sulphuraria* cultivation can be considered as measure for sanitizing not only liquid waste streams, but also hydrolysates from organic streams. The acidic growth conditions of *G. sulphuraria* prevented, on the one hand, the growth of microbial contaminations, and on the other hand, reduced the present microorganisms. However, it should be noted that microorganisms cannot be completely diminished and those organisms which form spores or are active under acidic conditions do survive, even over an extended treatment period (2700 counts per g of biomass). Most of the aerobic mesophilic counts were formed by yeasts (1.5×10^6 to 1.3×10^6 per g biomass). Yeast formation could already be seen

during the cultivation as precipitate in the storage tank. Nevertheless, the growth of the yeast cells could be kept under control. A cultivation of microalgae such as *G. sulphuraria* under acidic conditions paves the way towards an effective waste treatment, combining sanitation and utilization, as *G. sulphuraria* was, in all the reported studies, the dominant species, and expensive sterilization steps can be skipped.

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References

1. Hemathilake, D.M.K.S.; Gunathilake, D.M.C.C. Chapter 31—Agricultural productivity and food supply to meet increased demands. In *Future Foods*; Bhat, R., Ed.; Academic Press: Cambridge, MA, USA, 2022; pp. 539–553.
2. Fernandez-Bayo, J.; Achmon, Y.; Guerrero, M.D.M.; Di Gioia, F. Editorial: Upcycling organic waste for the sustainable management of soilborne pests and pathogens in agri-food systems. *Front. Sustain. Food Syst.* **2022**, *6*, 1012789. [CrossRef]
3. Popp, J.; Kovács, S.; Oláh, J.; Divéki, Z.; Balázs, E. Bioeconomy: Biomass and biomass-based energy supply and demand. *New Biotechnol.* **2021**, *60*, 76–84. [CrossRef] [PubMed]
4. Ddiba, D.; Ekener, E.; Lindkvist, M.; Finnveden, G. Sustainability assessment of increased circularity of urban organic waste streams. *Sustain. Prod. Consum.* **2022**, *34*, 114–129. [CrossRef]
5. Yu, B.; Liu, X.; Ji, C.; Sun, H. Greenhouse gas mitigation strategies and decision support for the utilization of agricultural waste systems: A case study of Jiangxi Province, China. *Energy* **2023**, *265*, 126380. [CrossRef]
6. Budihardjo, M.A.; Humaira, N.G.; Ramadan, B.S.; Wahyuningrum, I.F.S.; Huboyo, H.S. Strategies to reduce greenhouse gas emissions from municipal solid waste management in Indonesia: The case of Semarang City. *Alex. Eng. J.* **2023**, *69*, 771–783. [CrossRef]
7. Zhan, J.; Han, Y.; Xu, S.; Wang, X.; Guo, X. Succession and change of potential pathogens in the co-composting of rural sewage sludge and food waste. *Waste Manag.* **2022**, *149*, 248–258. [CrossRef]
8. Tsui, T.-H.; van Loosdrecht, M.C.M.; Dai, Y.; Tong, Y.W. Machine learning and circular bioeconomy: Building new resource efficiency from diverse waste streams. *Bioresour. Technol.* **2023**, *369*, 128445. [CrossRef]
9. Mishra, K.; Siwal, S.S.; Nayaka, S.C.; Guan, Z.; Thakur, V.K. Waste-to-chemicals: Green solutions for bioeconomy markets. *Sci. Total Environ.* **2023**, *887*, 164006. [CrossRef]
10. Khan, S.; Das, P.; Thaher, M.I.; AbdulQuadir, M.; Mahata, C.; Al Jabri, H. Utilization of nitrogen-rich agricultural waste streams by microalgae for the production of protein and value-added compounds. *Curr. Opin. Green Sustain. Chem.* **2023**, *41*, 100797. [CrossRef]
11. Premaratne, M.; Nishshanka, G.K.S.H.; Anthonio, R.A.D.P.; Liyanaarachchi, V.C.; Thevarajah, B.; Nimarshana, P.H.V.; Malik, A.; Ariyadasa, T.U. Resource recovery from waste streams for production of microalgae biomass: A sustainable approach towards high-value biorefineries. *Bioresour. Technol. Rep.* **2022**, *18*, 101070. [CrossRef]
12. Rashid, N.; Selvaratnam, T.; Park, W.-K. Chapter 21—Resource Recovery From Waste Streams Using Microalgae: Opportunities and Threats. In *Microalgae Cultivation for Biofuels Production*; Yousuf, A., Ed.; Academic Press: Cambridge, MA, USA, 2020; pp. 337–351.
13. Pleissner, D.; Wimmer, R.; Eriksen, N.T. Quantification of amino acids in fermentation media by isocratic HPLC analysis of their α-hydroxy acid derivatives. *Anal. Chem.* **2011**, *83*, 175–181. [CrossRef] [PubMed]
14. Malavasi, V.; Soru, S.; Cao, G. Extremophile Microalgae: The potential for biotechnological application. *J. Phycol.* **2020**, *56*, 559–573. [CrossRef] [PubMed]
15. Varshney, P.; Mikulic, P.; Vonshak, A.; Beardall, J.; Wangikar, P.P. Extremophilic micro-algae and their potential contribution in biotechnology. *Bioresour. Technol.* **2015**, *184*, 363–372. [CrossRef] [PubMed]

16. Pleissner, D.; Schönfelder, S.; Händel, N.; Dalichow, J.; Ettinger, J.; Kvangarsnes, K.; Dauksas, E.; Rustad, T.; Cropotova, J. Heterotrophic growth of *Galdieria sulphuraria* on residues from aquaculture and fish processing industries. *Bioresour. Technol.* **2023**, *384*, 129281. [CrossRef] [PubMed]
17. Čížková, M.; Vítová, M.; Zachleder, V. The red microalga *Galdieria* as a promising organism for applications in biotechnology. In *Microalgae—From Physiology to Application*; IntechOpen: London, UK, 2019. [CrossRef]
18. Delanka-Pedige, H.M.K.; Munasinghe-Arachchige, S.P.; Cornelius, J.; Henkanatte-Gedera, S.M.; Tchinda, D.; Zhang, Y.; Nirmalakhandan, N. Pathogen reduction in an algal-based wastewater treatment system employing *Galdieria sulphuraria*. *Algal Res.* **2019**, *39*, 101423. [CrossRef]
19. Munasinghe-Arachchige, S.P.; Delanka-Pedige, H.M.K.; Henkanatte-Gedera, S.M.; Tchinda, D.; Zhang, Y.; Nirmalakhandan, N. Factors contributing to bacteria inactivation in the *Galdieria sulphuraria*-based wastewater treatment system. *Algal Res.* **2019**, *38*, 101392. [CrossRef]
20. Pleissner, D.; Händel, N. Reduction of the microbial load of digestate by the cultivation of *Galdieria sulphuraria* under acidic conditions. *Waste Biomass Valorization* **2023**, *14*, 2621–2627. [CrossRef]
21. Tchinda, D.; Henkanatte-Gedera, S.M.; Abeysiriwardana-Arachchige, I.S.A.; Delanka-Pedige, H.M.K.; Munasinghe-Arachchige, S.P.; Zhang, Y.; Nirmalakhandan, N. Single-step treatment of primary effluent by *Galdieria sulphuraria*: Removal of biochemical oxygen demand, nutrients, and pathogens. *Algal Res.* **2019**, *42*, 101578. [CrossRef]
22. di Cicco, M.R.; Iovinella, M.; Palmieri, M.; Lubritto, C.; Ciniglia, C. Extremophilic Microalgae *Galdieria* Gen. for Urban Wastewater Treatment: Current State, the Case of "POWER" System, and Future Prospects. *Plants* **2021**, *10*, 2343. [CrossRef]
23. Montenegro-Herrera, C.A.; Vera-López Portillo, F.; Hernández-Chávez, G.T.; Martinez, A. Single-cell protein production potential with the extremophilic red microalgae *Galdieria sulphuraria*: Growth and biochemical characterization. *J. Appl. Phycol.* **2022**, *34*, 1341–1352. [CrossRef]
24. Korozi, E.; Tsagou, V.; Kefalogianni, I.; Markou, G.; Antonopoulos, D.; Chakalis, L.; Kotzamanis, Y.; Chatzipavlidis, I. Continuous culture of *Auxenochlorella protothecoides* on biodiesel derived glycerol under mixotrophic and heterotrophic conditions: Growth parameters and biochemical composition. *Microorganisms* **2022**, *10*, 541. [CrossRef] [PubMed]
25. Hu, X.; Zhang, M.; Yu, J.; Zhang, G. Food waste management in China: Status, problems and solutions. *Acta Ecol. Sin.* **2012**, *32*, 4575–4584.
26. Wu, S.; Xu, S.; Chen, X.; Sun, H.; Hu, M.; Bai, Z.; Zhuang, G.; Zhuang, X. Bacterial communities changes during food waste spoilage. *Sci. Rep.* **2018**, *8*, 8220. [CrossRef] [PubMed]
27. Pinto, R.B.; Oduro-Kwarteng, S.; Hamidu, J.A.; Essandoh, H.M.K. Sensitivity of nutritional and microbial content of food wastes to drying technologies. *Sci. Afr.* **2022**, *16*, e01130. [CrossRef]
28. Seruga, P.; Krzywonos, M.; Paluszak, Z.; Urbanowska, A.; Pawlak-Kruczek, H.; Niedźwiecki, Ł.; Pińkowska, H. Pathogen reduction potential in anaerobic digestion of organic fraction of municipal solid waste and food waste. *Molecules* **2020**, *25*, 275. [CrossRef]
29. Böhm, R. Chapter Nine—Pathogenic agents. In *Waste Management Series*; Diaz, L.F., de Bertoldi, M., Bidlingmaier, W., Stentiford, E., Eds.; Elsevier: Amsterdam, The Netherlands, 2007; Volume 8, pp. 177–200.
30. Soobhany, N.; Mohee, R.; Garg, V.K. Inactivation of bacterial pathogenic load in compost against vermicompost of organic solid waste aiming to achieve sanitation goals: A review. *Waste Manag.* **2017**, *64*, 51–62. [CrossRef]
31. Sharma, R.; Sharma, M.; Sharma, R.; Sharma, V. The impact of incinerators on human health and environment. *Rev. Environ. Health* **2013**, *28*, 67–72. [CrossRef]
32. Chi, T.; Zhang, A.; Zhang, X.; Li, A.-D.; Zhang, H.; Zhao, Z. Characteristics of the antibiotic resistance genes in the soil of medical waste disposal sites. *Sci. Total Environ.* **2020**, *730*, 139042. [CrossRef]
33. Avidov, R.; Varma, V.S.; Saadi, I.; Hanan, A.; Lublin, A.; Saldinger, S.S.; Chen, Y.; Laor, Y. Factors influencing the persistence of *Salmonella infantis* in broiler litter during composting and stabilization processes and following soil incorporation. *Front. Sustain. Food Syst.* **2021**, *5*, 645721. [CrossRef]