water

Article

Polyhydroxyalkanoates (PHAs) Production: A Feasible Economic Option for the Treatment of Sewage Sludge in Municipal Wastewater Treatment Plants?

Dafne Crutchik [1], Oscar Franchi [1], Luis Caminos [1], David Jeison [2], Marisol Belmonte [3], Alba Pedrouso [4], Angeles Val del Rio [4], Anuska Mosquera-Corral [4] and José Luis Campos [1,*]

[1] Faculty of Engineering and Sciences, Universidad Adolfo Ibáñez, Av. Padre Hurtado 750, 2520000 Viña del Mar, Chile; dafne.crutchik@uai.cl (D.C.); oscar.franchi@edu.uai.cl (O.F.); luis.caminos@uai.cl (L.C.)
[2] Escuela de Ingeniería Bioquímica, Pontificia Universidad Católica de Valparaíso, Av. Brasil 2085, 2362803 Valparaíso, Chile; david.jeison@pucv.cl
[3] Laboratorio de Biotecnología, Medio Ambiente e Ingeniería (LABMAI), Facultad de Ingeniería, Universidad de Playa Ancha, Avda. Leopoldo Carvallo 270, 2340000 Valparaíso, Chile; marisol.belmonte@upla.cl
[4] CRETUS Intitute, Department of Chemical Engineering, Universidade de Santiago de Compostela, E-15782 Santiago de Compostela, Spain; alba.pedrouso@usc.es (A.P.); mangeles.val@usc.es (A.V.d.R.); anuska.mosquera@usc.es (A.M.-C.)
* Correspondence: jluis.campos@uai.cl; Tel.: +56-(32)-250-3716

Received: 26 March 2020; Accepted: 12 April 2020; Published: 14 April 2020

Abstract: Sludge is a by-product of municipal wastewater treatment plants (WWTPs) and its management contributes significantly to the operating costs. Large WWTPs usually have anaerobic sludge digesters to valorize sludge as methane and to reduce its mass. However, the low methane market price opens the possibility for generating other high value-added products from the organic matter in sludge, such as polyhydroxyalkanoates (PHAs). In this work, the economic feasibility of retrofitting two types of WWTPs to convert them into biofactories of crude PHAs was studied. Two cases were analyzed: (a) a large WWTP with anaerobic sludge digestion; and (b) a small WWTP where sludge is only dewatered. In a two-stage PHA-production system (biomass enrichment plus PHAs accumulation), the minimum PHAs cost would be 1.26 and 2.26 US$/kg PHA-crude for the large and small WWTPs, respectively. In a single-stage process, where a fraction of the secondary sludge (25%) is directly used to accumulate PHAs, the production costs would decrease by around 15.9% (small WWTPs) and 19.0% (large WWTPs), since capital costs associated with bioreactors decrease. Sensitivity analysis showed that the PHA/COD (Chemical Oxygen Demand) yield is the most crucial parameter affecting the production costs. The energy, methane, and sludge management prices also have an essential effect on the production costs, and their effect depends on the WWTP's size.

Keywords: anaerobic digestion; bioplastics; economic analysis; methane; resource recovery; sewage sludge; WWTP size

1. Introduction

Wastewater treatments plants (WWTPs) produce large quantities of sewage sludge, which requires adequate and environmentally safe management and disposal. Sludge management is one of the most critical issues in WWTPs operation, and it can represent from 20% to 60% of the overall operating costs [1]. During the last few decades, sewage sludge management has moved from an approach involving only treatment and disposal, to its conversion into value-added products, such as bioenergy or biobased materials. The latter alternative has the potential to reduce the sludge quantity that needs to be finally disposed, and can decrease the overall operating costs [2]. In this context, anaerobic digestion is a very useful technology for sludge management, since it can convert the biodegradable organic carbon into methane containing biogas [2,3]. Even though anaerobic digestion is a mature and sustainable technology for sewage sludge valorization, recent studies indicate the convenience of exploring other technologies with the aim of producing higher-value end-products, such as polyhydroxyalkanoates (PHAs) [2,4,5]. PHAs are biodegradable polyesters synthesized by numerous bacteria that accumulate as intracellular carbon and energy reservoirs, under nutrient-limited growth conditions [5].

Research on the use of PHAs as raw material for the production of biobased biodegradable plastics (bioplastics) is receiving increasing attention. Bioplastics' physical and chemical properties are comparable to conventional petroleum-based materials. Additionally, they have the advantages of being producible from renewable resources and are completely biodegradable [2]. Then, bioplastics have great potential to increase the sustainability of the plastic industry, by mitigating environmental problems associated with this industry, such as the exploitation of nonrenewable fossil resources or degradation of the natural systems by plastics leaking [6].

However, at present, the replacement of petrochemical-based materials by PHAs has been hindered by their market prices, since the traditional methods for PHAs production are expensive. PHAs' market price is between 2.4 and 5.5 US$/kg PHAs, which is several times higher than petroleum-based plastics (1.2 US$/kg synthetic plastic) [7–9]. Traditional methods for PHAs production involve the use of pure cultures and expensive carbon sources such as noble oils, pure sugars, lipids, and animal or vegetable proteins. The cost of the carbon source contributes up to 30–50% of the overall PHAs' production costs. Therefore, the development of a cost-effective process for PHAs production requires the selection of a suitable carbon source [10]. Recent studies indicate that it is possible to reduce operational costs associated to the carbon source by using low-cost substrates, such as waste streams from WWTPs, agriculture, or the food industry [4,5].

The use of primary sludge (PS) and waste activated sludge (WAS) as carbon sources for PHAs production has been demonstrated by several studies [11–13]. PS and WAS can be readily converted into volatile fatty acids (VFAs) during an acidogenic fermentation, which can be achieved by inhibiting the methanogenic step during the anaerobic digestion process. WAS can also be used as a PHAs-accumulating bacteria source, since many bacteria found naturally in WAS are able to accumulate PHAs. Indeed, PHAs content in WAS has been reported to range from 0.3 to 22.7 mg PHAs/g WAS [14–16]. Tian et al. [16] reached PHAs content of 51%, with a productivity rate of 2.19 g/(L·h) from WAS. In addition, the use of a mixed bacteria culture is cheaper than traditional PHAs production methods, since sterilization steps are not necessary [17]. Thus, WWTPs offer low-cost carbon sources and mixed bacterial cultures that have demonstrated potential for PHAs production. Therefore, integration of PHAs production and wastewater treatment may help to reduce PHAs prices to a range that could be competitive with the traditional raw materials for the plastics industry.

Several studies have investigated the valorization of sewage sludge by PHAs production [10–13]. However, there is a significant lack of knowledge about the economic balance of the implementation of this process in WWTPs. The potential implementation of a PHAs production process into existing WWTPs still needs to be economically evaluated, to ensure its cost-effective feasibility. The integration of a PHAs production process into a conventional WWTP diverts sewage sludge from existing process lines, affecting not only the obtained by-products (e.g., methane) but also the operating costs of the whole installation. For instance, re-routing sewage sludge for PHAs production can impact the methane production capacity, reducing the recovered energy from sewage sludge, and consequently affecting the revenues generated by selling the generated methane. However, PHAs seem to be a more valuable end-product than methane. The potential market value of PHAs is four times higher than that of methane: 1.3 US$/kg COD_{PHA} and 0.15–0.28 US$/kg COD_{CH4}, respectively [2,4].

This work was focused on the technoeconomic analysis of the implementation of PHAs production from sewage sludge in municipal WWTPs to generate a product with commercial value as raw material for the plastic industry. Two different case studies were evaluated concerning the WWTPs' size.

2. Materials and Methods

2.1. Case Studies Descriptions

Two differently-sized municipal WWTPs were studied in order to verify the influence of scale on the implementation of PHA-enriched biomass (PHA-crude). The first one is a municipal WWTP located in Santiago de Chile, treating an inlet flow rate of 777,600 m^3/d (3,110,400 of population equivalent, PE). This was considered as representative of a large-scale WWTP and hereafter will be referred to as a large WWTP. The large WWTP configuration includes primary settling followed by an activated sludge system, operated at a solid retention time (SRT) of 3 days (Figure 1a). PS and WAS are separately thickened and treated by eight mesophilic anaerobic digesters, operated at an SRT of 30 d, which generates 80,000 m^3 biogas/d (65% CH_4). Twenty-five percent of the produced methane is burnt to heat anaerobic digesters while the remaining CH_4 is sold. Digested sludge is dewatered using dehydrating centrifuges and 300 tons of dehydrated sludge (25% dry matter) are purged daily.

The second study case (small WWTP) is a fictitious WWTP with a capacity to treat the sewage from a population equivalent of 50,000 PE (12,500 m^3/d). Its configuration consists of an activated sludge unit operated at an SRT of 3 days in order to remove organic matter from wastewater. The generated dehydrated WAS is supposed to be disposed in landfill, without any valorization processes (Figure 1b).

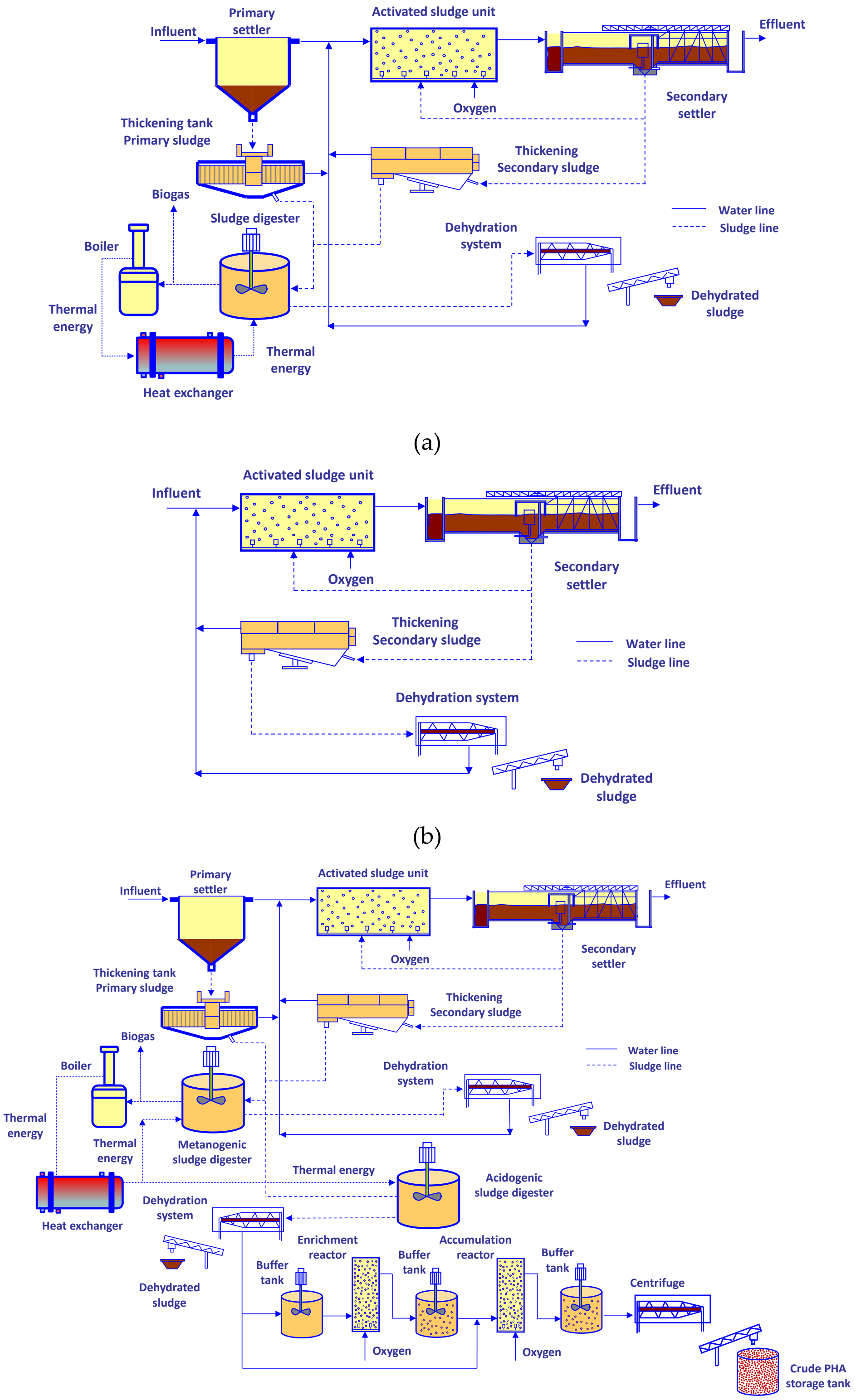
2.2. Proposed Scenarios for PHAs Production

The PHAs production plants were designed as a side-stream process of both WWTPs. The proposed scenarios involved the incorporation of specific equipment and, depending on the WWTP, the use of existing equipment and facilities in order to reduce the investment costs.

In the case of the large WWTP, methane production was partially replaced by PHAs production for sewage sludge valorization. The PHAs production plant included three biological steps: (i) acidogenic sludge fermentation for VFAs production, (ii) enrichment of biomass with PHAs-storing capacity, and (iii) PHAs accumulation. The VFA production took place in 6 of the 8 existing sludge anaerobic digesters. The obtained sludge fermentation liquid was distributed between the enrichment and accumulation stages. The remaining two anaerobic sludge digesters were used to produce methane that was burnt to heat acidogenic fermentation and methanogenic reactors, operated at mesophilic temperature. Enrichment and accumulation stages of PHA-accumulating biomass were developed in sequential batch reactors (SBRs). Also, storage tanks were required (i) to store the effluent from acidogenic fermentation reactors, (ii) to store the enriched biomass since enrichment and accumulation reactors were operated in a fed-batch mode, and (iii) to store produced PHAs-rich biomass. At the end of the accumulation stage, PHAs-rich biomass was dewatered by a dehydration centrifuge, reducing its volume. The dewatered PHAs-rich biomass was then transported to the storage hall for further commercialization. Other equipment such as pumps and a conveyor belt were also included for the proposed scenario.

As previously mentioned, the small WWTP was only conceived to remove organic matter from the wastewater using an activated sludge system. For this reason, the proposed scenario included the complete construction of the PHAs production plant, which means the implementation of primary settling in the waterline, generating PS, and all the required equipment for the PHAs production process. Like in the case of the large WWTP, VFAs production from sludge was developed in acidogenic fermentation reactors, and PHAs-rich biomass enrichment and later PHAs accumulation were performed in SBRs. Also, methane production from sludge was included in the proposed scenario to generate thermal energy for the acidogenic fermentation stage, and the required anaerobic digesters. Other equipment such as accumulation tanks, pumps, dehydration centrifuges, heat exchanger system, conveyor belt, and storage tank were also included. Figure 1c shows the process flow diagram for PHAs production integrated into each studied WWTP.

For the proposed scenarios, PHAs-rich biomass was the final product of the PHAs production system. The extraction of PHAs and their recovery were not considered in this work.



(a)

(b)

(c)

Figure 1. Schematic layout of (**a**) a large wastewater treatment plant (WWTP); (**b**) a small WWTP; and (**c**) a WWTP with integrated polyhydroxyalkanoates (PHAs) production.

2.3. Mass and Energy Balances

For both WWTP scales, mass and energy balances were performed, using Excel spreadsheets, according to the methodology proposed in [18]. Balances were performed considering traditional operating conditions of WWTPs (see Table 1). The total influent COD concentration was 500 mg/L (soluble biodegradable COD (S_S): 150 mg/L; soluble non-biodegradable COD (S_I): 50 mg/L; particulate biodegradable COD (X_S): 200 mg/L; particulate non-biodegradable COD (X_I): 100 mg/L). Mass and energy balances enabled determination of oxygen consumption, energy consumption, biogas production, sludge generation, PHAs production, and also sizing the required units. Operational conditions of sludge treatment units were considered as constant; i.e., no daily or seasonal variations occur during the WWTP operation.

Table 1. Summary of values used to perform both mass and energy balances for the different scenarios.

Mass Balances	
Unit Operation	**Values**
Primary settling	Solids removal efficiency: 60% Primary sludge concentration: 60 g VSS/L COD/VSS ratio of primary sludge: 1.8 g/g
Activated sludge process	Solids retention time (SRT): 3 d Biomass yield: 0.43 g VSS/g $COD_{consumed}$ [18] Decay coefficient: 0.24 d^{-1} [18] No biodegradable fraction of heterotrophic biomass (X_p): 0.15 [18] Solids concentration in the effluent: 20 mg VSS/L 1.42 g COD/g VSS for the biodegradable fraction of heterotrophic biomass [18] 1.55 g COD/g VSS for X_I COD fraction [19] 1.42 g COD/g VSS for X_p heterotrophic biomass fraction [18]
Thickening units	Primary sludge-thickened concentration: 60 g VSS/L WAS-thickened concentration: 40 g VSS/L
Anaerobic reactors	Specific methane production: 0.35 N m^3 CH_4/kg $COD_{degraded}$ All biodegradable COD is converted to methane or acetic acid Methane fraction in biogas: 65% Biomass yield: 0.08 g VSS/g $COD_{consumed}$ [18] Decay coefficient: 0.03 d^{-1} [18]
Sludge dewatering	Solids concentration at outlet: 250 g/L Solids capture efficiency: 96% TSS/VSS ratio: 0.75 g/g
PHAs production	Biomass yield: 0.50 g COD-biomass/g $COD_{consumed}$ (biomass enrichment stage) (Based on [20]) 1.67 g COD/g PHA [21] PHA yield: 0.6 g COD-PHA/g COD (PHA accumulation stage) (Based on [22]) Solids capture efficiency: 96% (PHA rich biomass dewatering)
Energy Balances	**Values**
Aeration	1 kW·h/kg O_2 [18]
Overall mixing and pumping	0.1 kW·h/$m^3_{influent}$ (used to calculate the current WWTP energy consumption) (Based on [23])
Thermal energy required for heating anaerobic systems	24% of CH_4 generated
Primary settling	0.0005 kW/m^3 [5]
Gravity belt thickener	0.23 kW/(m^3/h) [5]
Centrifuge	1.875 kW/(m^3/h) [5]
Thickener sludge tank	0.001 (kWh/m^3) [24]
Pumping energy required for biomass enrichment and PHA-accumulation bioreactors	Calculated based on the inlet flowrate, and considering the reactor height and a pump energy efficiency of 0.7.
Mixing anaerobic reactors	0.006 kW/m^3 [25]

2.4. Methodology for Economic Assessment

The economic analysis of PHAs production for the proposed scenarios was carried out considering the total capital costs, the increase of the operating and maintenance costs of the WWTP, the reduction of income derived from a decrease in the amount of commercialized methane (only in the case of the large WWTP) and the benefits obtained from the commercialization of PHA-rich biomass. The total capital costs include costs related to the purchase of equipment (bioreactors, buffer and storage tanks, dehydration centrifuges, pumps, conveyor belt, among others) and the required equipment for piping, instrumentation/electricity, engineering costs, civil works, and start-up of the PHAs production plant. Equipment investment costs were calculated based on the functions reported in studies found in the literature [26–29], updated using the Chemical Engineering Plant Cost Index (CEPCI). In the case of the small WWTP, the existing dewatering unit was considered to be able to manage both streams of primary and secondary sludge, since the overall production would notably decrease after the WWTP retrofitting. Moreover, for both large and small WWTPs, it was assumed that the installed blowers have sufficient overcapacity to supply oxygen to the biomass enrichment and PHAs accumulation reactors. Costs related to the required equipment for piping, instrumentation/electricity, engineering costs, civil works, and start-up of the PHAs production plant were estimated as 15%, 25%, 10%, 34%, and 12% of the equipment investment costs, respectively.

The operating and maintenance costs included the increase of energy consumption (due to oxygen requirements, mixing and pumping of anaerobic digesters and SBRs, and dehydration centrifuge operation), the increase of the amount of sludge to be disposed, the increase of the amount of reagents (polyelectrolytes) needed for the processes of thickening and dewatering of solids (considering a dose of 5 mg/kg TSS [18]), maintenance, insurance, and labor. The price of electricity was set at 0.109 US$/kWh [30] and sludge disposal and reagents costs were 40.4 and 2000 US$/ton. Maintenance and insurance costs were also taken as fixed percentages of the capital cost, 1% and 0.5%, respectively. The labor cost of operators was assumed to be 5.45 US$/person-hour.

The minimum cost of PHAs was estimated as the value that makes the net present value (NPV) zero (Equation (1)).

$$\mathrm{NPV} = \sum_{t=1}^{T} \frac{(B_t - C_t)\cdot(1+i)^t}{(1+r)^t} - \text{Total capital costs} \tag{1}$$

where B_t is the benefit from the PHAs sale; C_t is the sum of the operating costs and the loss of income coming from the reduction of the amount of sold methane (calculated taking into account a sale price of 0.137 US$/Nm^3); i is the inflation rate (3%); r is the interest rate (5%); and T is the payback time (20 years).

Uncertainty and variability can be present in the input variables used in economic analysis. Then, a sensitivity analysis was carried out. For this purpose, three economic parameters (price of energy, price of methane, and sludge management cost) and one intrinsic process parameter (COD-PHA/COD$_{\text{consumed}}$ yield coefficient) were considered. Values of the PHA/COD yield coefficient described in the literature for the PHAs-accumulation stage are generally between 0.50 and 0.70 g COD-PHA/g COD$_{\text{consumed}}$, and therefore, a range of ±15% was considered for each parameter.

3. Results and Discussion

3.1. Mass and Energy Balances

Plant mass and energy balances were calculated for proposed scenarios for both studied WWTP sizes. Results from the mass and energy balances related to net methane production, sludge generation, and energy consumption obtained for the large WWTP fit quite well to the actual data from the plant (Table 2). Then, it could be inferred that this methodology would also properly describe the operation of the small WWTP proposed in this work. The actual value of oxygen consumption was not available. However, considering the accuracy observed during the estimation of net methane production and sludge generation, the estimation for oxygen consumption may be considered correct according to the COD balance.

Table 2. Mass and energy balances of the large and small WWTPs.

Item	Actual	Calculated	Bioplastic Production	
			Enriched Sludge	Mixed Sludge
Large WWTP				
Net methane generation (m^3/d)	39,000	40,291	0	0
Sludge generation (kg/d)	300,000	287,824	282,180 (−2.0%)	270,738 (−5.9%)
O_2 consumption (kg/d)	---	79,843	131,905 (+65.2%)	127,655 (+59.9%)
Energy consumption (kWh/d)	157,300	157,603	210,224 (+33.4%)	205,973 (+30.7%)
PHAs production (kg/d)	0	0	16,342	15,067
Small WWTP				
Net methane generation (m^3/d)	---	0	0	0
Sludge generation (kg/d)	---	9431	4480 (−52.5%)	4085 (−56.7%)
O_2 consumption (kg/d)	---	2630	2565 (−2.5%)	2499 (−5.0%)
Energy consumption (kWh/d)	---	3880	3827 (−1.4%)	3761 (−3.1%)
PHAs production (kg/d)	---	0	254	234

In the case of the large WWTP, the change from methane generation to the production of bioplastics would cause an increase in energy consumption of 33.4%. This increase is mostly due to the aeration requirements for biomass enrichment and the PHAs-accumulation stages. The production of sludge would not be practically affected, since it is considered that all the biodegradable matter contained in the sludge, which initially was converted into methane, would be now degraded aerobically or converted into biomass and PHAs, which would be sold.

With respect to the small WWTP, the implementation of the sludge line would cause a notable decrease in sludge production (52.5%) while energy consumption would not be practically affected (decrease of 1.4%) since oxygen consumption remains almost constant. A priori, the incorporation of a primary settler could suggest a possible energy savings, since a fraction of the organic matter that enters to the WWTP would be derived from anaerobic reactors. However, it must be taken into account that this organic matter would not be converted into methane, but mainly aerobically degraded, and only a fraction would be converted into biomass and biopolymers.

When legal requirements for treated sewage disposal demands nitrogen and organic matter removal, the activated sludge units are generally operated at SRT around 15 d in order to promote nitrification and denitrification processes [21]. In this scenario, according to the mass balance, the biodegradable COD available in the WAS would decrease as bacteria endogenous respiration increases. This fact would cause a decrease of 22.3% in PHAs production. Therefore, one could expect that retrofitting a WWTP operated at low STRs would be more economically favorable than doing so with one operated at high SRTs.

3.2. Costs Associated with Bioplastics Production

Based on the mass balances, the size and the amount of required processing equipment for PHAs production was estimated for each WWTP. Capital costs were calculated and are presented in Table 3. If they are standardized considering the PHAs production rate, values of 8.1 and 41.1 US$/(kg PHA/year) can be obtained for the large and small WWTPs, respectively. The large difference between these values can be mainly attributed to the fact that the equipment requirement of the large WWTP is lower than the one of the small WWTP and, to a lesser extent, to a scale factor. Mudliar et al. [31] and Bengtsson et al. [21] calculated capital costs of 13 US$/(kg PHA/year) and 17 US$/(kg PHA/year) but, in both cases, they included PHAs extraction units. In the case of the large WWTP, the capital costs are mainly given by the SBR reactors for the biomass enrichment and subsequent accumulation of the bioplastics. In contrast, for the small WWTP, costs are distributed mainly between the SBR reactors (54.5%), the anaerobic reactors (16.9%), and the primary settling tank (10.8%). A previous economic evaluation of PHAs production from activated sludge also showed that bioreactor′ costs account for the major part of capital cost even when the PHAs purification stages are considered [31].

Table 3. Total capital costs (MUS$) and operating and maintenance costs (MUS$/year) estimated for PHAs production from sewage sludge for the proposed scenarios of a large WWTP and small WWTP.

Economic Item	Large WWTP	Small WWTP
A. Capital costs		
Buffer tanks	102	49
Biomass enrichment and PHAs-accumulation reactor	23,714	1006
Acidogenic fermentation reactors	-	197
Anaerobic digestion reactors	-	131
Primary settling tank	-	210
Thickening and dewatering units	275	109
Storage PHA tanks	414	7
Conveyor belt	30	23
Pumps	156	209
Heat exchanger	-	3
Boiler	-	3
Total	24,691	1947
B. Other capital costs		
Piping	3703	292
Instrumentation/electricity	6172	486
Detailed engineering costs	2469	195
Civil works	8394	662
Start-up of PHAs production plant	2963	234
Total	23,701	1869
Total capital costs (A + B) (MUS$)	48,392	3816
C. Operating and maintenance costs		
Equipment operation	2135	−73
Maintenance	247	19
Insurance	123	10
Operators	63	21
Total (MUS$/year)	2568	−23

Operating and maintenance costs were calculated from values obtained from mass and energy balances and the total capital cost. As can be seen in Table 3, in the case of the large WWTP, they increase mainly due to the energy consumption increase, attributed to the high oxygen requirement during biomass enrichment and the PHAs-accumulation stages. This result agrees with that obtained by [5] when studying different process configurations to integrate PHAs production in a WWTP that treated its sludge by means of anaerobic digestion. Nevertheless, the operating costs of the small WWTP decrease because of the high reduction of sludge generated, achieved when the treatment plant is retrofitted.

Capital costs could be drastically diminished if secondary sludge is directly used to accumulate PHAs from VFAs, which would only be generated from the primary sludge, avoiding the capital costs related to biomass enrichment [10]. In this sense, Werker et al. [32] tested the PHAs-accumulation potential of secondary sludge from several WWTPs. They found that secondary sludge was able to accumulate around 40% of PHA, a value that meets the minimum value required for a downstream economic recovery of the polymer. In this way, this operational strategy may achieve yields of 0.187 g PHA/g COD and 0.125 g VSS/g COD during the PHAs-accumulation stage [21]. Nevertheless, mass balances indicate that the use of the secondary sludge instead of enriched biomass to accumulate PHAs would imply a decrease of the g PHA/g VSS ratio from 61% to 16%, which makes polymer recovery unattractive. The sharp decrease of this ratio is caused by two main facts: (i) secondary sludge contains 31% of the biodegradable COD available to produce PHA, which is now not used for this purpose; and (ii) the high amount of VSS contained in the secondary sludge stream has a dilution effect on the PHAs generated. Even in the case of WWTPs operated at high SRTs (15 d), whose secondary sludge production is 56% of that of WWTPs operated at SRTs of 3 d, the estimated g PHA/g VSS ratio is 24%. Therefore, if this operational strategy was implemented, availability of biodegradable organic wastes would be necessary to feed into the anaerobic units and increase the PHAs production in order to achieve the desired g PHA/g VSS ratio of 40% [21].

The use of only a fraction of the WAS to accumulate PHAs while the PS and the remaining secondary sludge are used to produce VFAs is proposed as a novel strategy to reduce the capital costs and to foster the g PHA/g VSS ratio. Based on the material balance calculations, at least 75% of the secondary sludge should be derived from the anaerobic reactors to obtain a percentage of g PHA/g VSS greater than 40% at the end of the accumulation stage. Taking into account that secondary sludge accounts the 31% of the available biodegradable COD to produce PHAs, the application of this configuration would lead to a reduction of 7.8% in the PHAs production, which would be associated with the same decrease in O_2 consumption during PHAs production, compared to the use of enriched biomass (Table 2). As mentioned before, this operational strategy would enable a significant total capital cost drop (48.4% and 30.7% for the large and small WWTPs, respectively). This reduction is related to the fact that the biomass-enrichment stage is no longer necessary and, in the case of the small WWTP, there would also be a decrease in the costs associated with anaerobic equipment (10.2%), since the volume of solids to be managed is lower.

3.3. Economic Analysis

In order to determine the produced bioplastic cost, as well as the costs associated with their production, the income loss due to the methane production reduction was considered in the large WWTP case. Taking all these aspects into account, the minimum estimated production cost of the raw PHAs were 1.26 and 2.26 US$/kg PHA-crude for the large and small treatment plants, respectively. These results would indicate that retrofitting WWTPs would be more favorable when an anaerobic sludge treatment line is already implemented, probably due to the lower specific capital requirement. It should also be noted that a change in the production from methane towards bioplastics will cause the underuse of existing anaerobic sludge digesters, since the hydraulic retention time (HRT) necessary to produce the VFAs is much lower than that to produce methane [33]. Therefore, the process economy could be improved if a more efficient use of existing facilities is pursued, by providing an external organic waste to be converted in the anaerobic fermenters [21].

Minimum production costs of bioplastics were also calculated for a scenario where 25% of the secondary sludge is directly used to accumulate PHAs, in order to decrease the capital costs. Results show reductions of the production costs of 19.0% and 15.9% for the case of the large and small plants, respectively. This reduction is high in the case of the large WWTP, since the incidence of units related to biomass enrichment on the total capital costs is more significant than in the case of the small WWTP.

According to Bluemink et al. [10] the PHAs purification process would increase production costs by around 25%, so the minimum price of the processed product would be 1.58 and 2.83 US$/kg PHA-pure for the large and small treatment plants, respectively. Different bioplastic production costs from wastewater can be found in the literature ranging from 1.40–11.80 US$/kg PHA-pure [10,20,31,32], which can be associated to the scale and plant configuration proposed. This wide range of prices hinders the comparison of the obtained values with the previously reported ones. In any case, prices obtained in this work are lower than current market prices (4.9 US$/kg or higher [32]).

Sensitivity analysis shows that the PHA/COD yield coefficient is the parameter that most affects the production cost for the large and small WWTPs (Figure 2). This effect can be attributed to the fact that this parameter simultaneously affects both bioplastic production and energy consumption due to the increase of the aeration requirements, which also would explain why this parameter does not have a linear effect on the production costs [31]. In the case of the large WWTP, decreases of both energy and methane sale prices have a relatively strong positive effect on the bioplastic production cost, while sludge management prices have no influence. However, for the small WWTP, the increase of the sludge management price has a positive effect on the bioplastic production, but the energy price has no influence. The opposite results found for both cases can be explained by taking into account how the retrofitting of the WWTPs affects the fate of the incoming organic matter: conversion to methane, assimilation by biomass, or oxidation to CO_2.

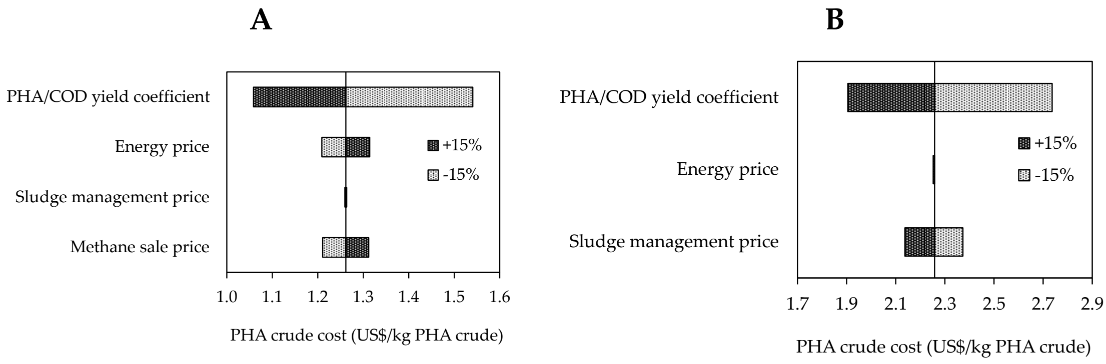


Figure 2. Sensitivity analysis for (**A**) a large WWTP and (**B**) small WWTP.

4. Conclusions

The retrofitting of WWTPs in order to generate crude PHAs is economically viable, even in the case of those WWTPs where no anaerobic reactors for treating sludge are available. In a two-stage PHAs production system like the one studied (biomass enrichment and PHAs accumulation), the minimum PHAs cost would be 1.26 and 2.26 US$/kg PHA-crude for the large and small WWTP, respectively.

Production costs could be decreased about 15.9–19.0% by using a fraction of the secondary sludge (25%) to accumulate PHAs directly. The PHA/COD yield coefficient is the parameter that most affects the production cost for large and small WWTPs. Energy, methane, and sludge management prices also have a significant effect on the production costs, which depends on the WWTP scale.

Author Contributions: Conceptualization, J.L.C. and A.M.-C.; methodology, L.C. and M.B.; writing-original draft, data curation, and formal analysis, D.C. and J.L.C.; writing-review and editing, O.F., D.J., and A.P.; supervision and validation, A.V.d.R. and A.M-C.; Funding acquisition, J.L.C. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the Chilean Government through the Projects FONDECYT 1200850 and CRHIAM Centre grant number ANID/FONDAP/15130015. This research was also supported by the Spanish Government (AEI) through the TREASURE project (CTQ2017-83225-C2-1-R). Authors from USC belong to the Galician Competitive Research Group ED431C 2017/29. All these programs are co-funded by FEDER (EU).

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Andreoli, C.V.; Sperling, M.; Fernandes, F. *Sludge Treatment and Disposal*; IWA Publishing: London, UK, 2007.
2. Alloul, A.; Ganigué, R.; Spiller, M.; Meerburg, F.; Cagnetta, C.; Rabaey, K.; Vlaeminck, S.E. Capture–ferment–upgrade: A three-step approach for the valorization of sewage organics as commodities. *Environ. Sci. Technol.* **2018**, *52*, 6729–6742. [CrossRef] [PubMed]
3. Amen, T.W.M.; Eljamal, O.; Khalil, A.M.E.; Matsunaga, N. Biochemical methane potential enhancement of domestic sludge digestion by adding pristine iron nanoparticles and iron nanoparticles coated zeolite compositions. *J. Environ. Chem. Eng.* **2017**, *5*, 5002–5013. [CrossRef]
4. Kleerebezem, R.; Joosse, B.; Rozendal, R.; Van Loosdrecht, M.C. Anaerobic digestion without biogas? *Rev. Environ. Sci. Bio.* **2015**, *14*, 787–801. [CrossRef]

5. Morgan-Sagastume, F.; Heimersson, S.; Laera, G.; Werker, A.; Svanström, M. Techno-environmental assessment of integrating polyhydroxyalkanoate (PHA) production with services of municipal wastewater treatment. *J. Clean. Prod.* **2016**, *137*, 1368–1381. [CrossRef]
6. World Economic Forum, Ellen MacArthur Foundation and McKinsey & Company, The New Plastics Economy—Rethinking the Future of Plastics. 2016. Available online: https://www.ellenmacarthurfoundation.org/assets/downloads/EllenMacArthurFoundation_TheNewPlasticsEconomy_Pages.pdf (accessed on 23 February 2019).
7. Gholami, A.; Mohkam, M.; Rasoul-Amini, S.; Ghasemi, Y. Industrial production of polyhydroxyalkanoates by bacteria: Opportunities and challenges. *Minerva Biotecnol.* **2016**, *28*, 59–74.
8. Chanprateep, S. Current trends in biodegradable polyhydroxyalkanoates. *J. Biosci. Bioeng.* **2010**, *110*, 621–632. [CrossRef]
9. Castilho, L.R.; Mitchell, D.A.; Freire, D.M. Production of polyhydroxyalkanoates (PHAs) from waste materials and by-products by submerged and solid-state fermentation. *Bioresour. Technol.* **2009**, *100*, 5996–6009. [CrossRef]
10. Bluemink, E.; van Nieuwenhuijzen, A.F.; Wypkema, E.; Uijterlinde, C.A. Bio-plastic (poly-hydroxy-alkanoate) production from municipal sewage sludge in the Netherlands: A technology push or a demand driven process? *Water Sci. Technol.* **2016**, *74*, 353–358. [CrossRef]
11. Raheem, A.; Sikarwar, V.S.; He, J.; Dastyar, W.; Dionysiou, D.D.; Wang, W.; Zhao, M. Opportunities and challenges in sustainable treatment and resource reuse of sewage sludge: A review. *Chem. Eng. J.* **2018**, *337*, 616–641. [CrossRef]
12. Frison, N.; Katsou, E.; Malamis, S.; Oehmen, A.; Fatone, F. Development of a novel process integrating the treatment of sludge reject water and the production of polyhydroxyalkanoates (PHAs). *Environ. Sci. Technol.* **2015**, *49*, 10877–10885. [CrossRef]
13. Tyagi, V.K.; Lo, S.L. Sludge: A waste or renewable source for energy and resources recovery? *Renew. Sustain. Energy Rev.* **2013**, *25*, 708–728. [CrossRef]
14. Salehizadeh, H.; Van Loosdrecht, M.C.M. Production of polyhydroxyalkanoates by mixed culture: Recent trends and biotechnological importance. *Biotechnol. Adv.* **2004**, *22*, 261–279. [CrossRef] [PubMed]
15. Baetens, D.; Aurola, A.-M.; Foglia, A.; Dionisi, D.; van Loosdrecht, M.C.M. Gas chromatographic analysis of polyhydroxybutyrate in activated sludge: A round-robin test. *Water Sci. Technol.* **2002**, *46*, 357–361. [CrossRef] [PubMed]
16. Tian, P.; Shang, L.; Ren, H.; Mi, Y.; Fan, D.; Jiang, M. Biosynthesis of polyhydroxyalkanoates: Current research and development. *Afr. J. Biotechnol.* **2009**, *8*, 709–714.
17. Albuquerque, M.G.E.; Eiroa, M.; Torres, C.; Nunes, B.R.; Reis, M.A.M. Strategies for the development of a side stream process for polyhydroxyalkanoate (PHA) production from sugar cane molasses. *J. Biotechnol.* **2007**, *130*, 411–421. [CrossRef]
18. Tchobanoglous, G.; Stensel, H.D.; Tsuchihashi, R.; Burton, F.L.; Abu-Orf, M.; Bowden, G.; Pfrang, W.; Metcalf & Eddy Inc. *Wastewater Engineering: Treatment & Resource Recovery*, 5th ed.; McGraw Hill: New York, NY, USA, 2014.
19. Takács, I. Experiments in Activated Sludge Modelling. Ph.D. Thesis, Ghent University, Gent, Belgium, 2008.
20. Fernández-Dacosta, C.; Posada, J.A.; Kleerebezem, R.; Cuellar, M.C.; Ramirez, A. Microbial community-based polyhydroxyalkanoates (PHAs) production from wastewater: Techno-economic analysis and ex-ante environmental assessment. *Bioresour. Technol.* **2015**, *185*, 368–377. [CrossRef]
21. Bengtsson, S.; Werker, A.; Visser, C.; Korving, L. *PHARIO: Stepping Stone to a Sustainable Value Chain for PHA Bioplastic Using Municipal Activated Sludge*; Stichting Toegepast Onderzoek Waterbeheer: Amersfoort, The Netherlands, 2017; pp. 1–93.
22. Conca, V.; da Ros, C.; Valentino, F.; Eusebi, A.L.; Frison, N.; Fatone, F. Long-term validation of polyhydroxyalkanoates production potential from the sidestream of municipal wastewater treatment plant at pilot scale. *Chem. Eng. J.* **2020**, *390*, 124627. [CrossRef]
23. Siegrist, H.; Salzgeber, D.; Eugster, J.; Joss, A. Anammox brings WWTP closer to energy autarky due to increased biogas production and reduced aeration energy for N-removal. *Water Sci. Technol.* **2008**, *57*, 383–388. [CrossRef]
24. Wang, Q.; Sun, J.; Zhang, C.; Xie, G.J.; Zhou, X.; Qian, J.; Wang, D. Polyhydroxyalkanoates in waste activated sludge enhances anaerobic methane production through improving biochemical methane potential instead of hydrolysis rate. *Sci. Rep.* **2016**, *6*, 19713. [CrossRef]

25. Bolzonella, D.; Innocenti, L.; Cecchi, F. Biological nutrient removal wastewater treatments and sewage sludge anaerobic mesophilic digestion performances. *Water Sci. Technol.* **2002**, *46*, 199–208. [CrossRef]
26. Remosa Tarifas 2014. Available online: https://es.scribd.com/document/289436343/REMOSA-tarifas-2014 (accessed on 23 February 2019).
27. Paredes, C. Cambio Tecnológico y Disponibilidad de Gas Natural Como Opción Para Reducir Emisiones de MP2,5 en el Concepción Metropolitano. Master's Thesis, Universidad de Concepción, Concepción, Chile, 2013.
28. Informe Final Licitación Pública N° 1588-151-LE09. Consultoría de Apoyo a los Procesos de Normas Ambientales en Sistemas Hídricos: Estimación de Costos de Abatimiento de Contaminantes en Residuos Líquidos. Available online: http://metadatos.mma.gob.cl/sinia/articles-50002_recurso_2.pdf (accessed on 23 February 2019).
29. Couper, J.R.; Penney, W.R.; Fair, J.R. *Chemical Process Equipment-Selection and Design*, 2nd ed.; Gulf Professional Publishing: Houston, TX, USA, 2009; pp. 1–819. [CrossRef]
30. ENEL. Available online: https://www.enel.cl/es/clientes/informacion-util/tarifas-y-reglamentos/tarifas.html (accessed on 23 February 2019).
31. Mudliar, S.N.; Vaidya, A.N.; Suresh, K.M.; Dahikar, S.; Chakrabarti, T. Techno-economic evaluation of PHB production from activated sludge. *Clean Technol. Environ. Policy* **2008**, *10*, 255–262. [CrossRef]
32. Werker, A.; Bengtsson, S.; Korving, L.; Hjort, M.; Anterrieu, S.; Alexandersson, T.; Morgan-Sagastume, F. Consistent production of high quality PHA using activated sludge harvested from full scale municipal wastewater treatment–PHARIO. *Water Sci. Technol.* **2018**, *78*, 2256–2269. [CrossRef] [PubMed]
33. Van, D.P.; Fujiwara, T.; Leu Tho, B.; Toan, S.; Phu, P.; Hoang Minh, G.; Minh, G.H. A review of anaerobic digestion systems for biodegradable waste: Configurations, operating parameters, and current trends. *Environ. Eng. Res.* **2020**, *25*, 1–17. [CrossRef]