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ARTICLE

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Up-recycling oil produced water as the media-base for the production of xanthan gum

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Abstract

Produced water (PW) and crude glycerin (CG) are compounds overproduced by the oil and biodiesel industry and significant scientific efforts are being applied for properly recycling them. The aim of this research is to combine such industrial byproducts for sustaining the production of xanthan by *Xanthomonas campestris*. Xanthan yields and viscosity on distinct PW ratios (0, 10, 15, 25, 50, 100) and on 100% dialyzed PW (DPW) in shaker batch testing identified DPW treatment as the best approach for further bioreactor experiments. Such experiments showed a xanthan yield of 17.3 g/L within 54 h and a viscosity of 512 mPa s. Physical-chemical characterization (energy dispersive X-ray spectroscopy, scanning electron microscopy and Raman spectroscopy) showed similarities between the produced gum and the experimental control. This research shows a clear alternative for upcycling high salinity PW and CG for the generation of a valued bioproduct for the oil industry.

KEYWORDS
crude glycerin, produced water, reuse, stirred-tank bioreactor, xanthan gum

1 | INTRODUCTION

One of the main byproducts generated in the oil and gas industrial chain is an effluent known as produced water (PW).[1] PW is discharged into the environment, often after treatment, or recycled as additional injecting fluid in an attempt to extract more oil or gas from the rock formation. At some point, however, treatment is necessary in order to meet the specific environmental disposal demands of each country.[2]

PW has a high pollutant potential,[3] due to its composition that includes several contaminants in different concentrations, including inorganic ions (metals like Fe, Cr, Ba, Ni, Zn; oil and grease), organic acids (such as benzene, toluene, ethyl benzene, and xylene), and phenols, radioactive elements and production chemicals like emulsifiers, flocculants, anticorrosion agents and biocides.[1] Some works reported negative effects on aquatic life due the increase in ions present in the PW, affecting significantly survival rates of young fish and growth rates in fish and mussels[4]; Additionally, there is a concern about the accumulation of these compounds and even radioactive elements in the environment if PW is introduced without treatment, representing a potential health issue to humans, animals and plants surrounding oilfield areas.[5] Their wide composition and huge volume represents a challenge to the PW treatment, since one method suitable for one kind of PW may perhaps not be applied to another. Therefore, the development of a set of cost-effective technologies to treat PW is one of the major goals of this industry.[1]

The present work proposes an alternative biological treatment method to reuse saline PW to produce biomolecule xanthan gum, which due to its high adsorptive capability could diminishing the PW contaminants.

The type of process to be adopted for the treatment of PW depends on the compounds to be removed. In turn, these compounds

depend on the final destination to be adopted for the treated PW, which can be disposal, injection, or reuse. To define this destination, it is necessary to treat the PW using one or a set of treatment methods. Furthermore, the literature did not specify a method with a specific destination, choosing to rely on terms of cost-effectiveness and availability. Between them, there are physical methods (physical adsorption, sand filters, cyclones, evaporation, dissolved air precipitation, membrane treatment, C-TOUR), chemical methods (chemical precipitation, chemical oxidation, electrochemical process, photocatalytic treatment, Fenton process, ozone treatment, ionic liquids, demulsifiers), and biological methods (phytoremediation, microbial treatment, enzymes, aerated membrane biofilm reactors).[6] Between these methods, the membrane process has been widely used due to its high efficiency and greater feasibility in field operations when compared to methods such as anaerobic filtration or hydrocyclones that require a pretreatment step. Despite the capacitive deionization being a promising technology, characterized by its low operating cost, it still depends on the development of materials capable of storing large amounts of ions present in the PW. Moreover, the aerated membrane biofilm reactor method is an emerging technology and provides a 90% energy reduction for aeration, which is a traditional biological treatment step with higher energy consumption.[7,8] However, these technologies still need to reduce the costs of their implementation in order to increase their application in the treatment of large volumes of PW.

It is correct to assume that, in several cases, strict regulations for PW disposal have led to an increase in the development of new treatment technologies. Although biological treatment systems are also used for improving PW quality,[2,9] very few studies focus on the valorization of PW as potential culturing medium for the large-scale production of microbial bioproducts. For example, their use in biosurfactants production by bacteria,[10] phycocyanin through cyanobacteria,[11] biomass through microalgae[12–14] and a potential use as growth media to produce lipids and biofuels by microalgae[15,16] has been report. Such a use would result in the upcycling effect for this wastewater.

PW is not the only waste capable of supporting large scale production of microbial products. Another widely produced industrial byproduct is crude glycerin (CG). Its applications include animal feed, wastewater treatment, biogas production, and oil chemistry, among others.[17] Given the increase in industrial activities, the generation of CG has increased, leading to the search for new alternatives for their reuse.

Exopolysaccharides (EPS) are polysaccharides secreted by microorganisms and have special physicochemical properties (viscosity, mechanical stability, ability to form gels) and notable applications in the food and petroleum industries - xanthan gum,[18] cosmetics - FucoPol[19] and FucoGel,[20] pharmaceutical and biomedical - dextran,[21] cellulose, hyaluronic acid and levan,[22,23] packaging - pollulan.[24] Furthermore, a notable effort has been made in the prospection of new strains capable of producing new exopolysaccharides with properties applicable to biotechnology, such as metallic chelators, gelling agents[25] and cryoprotectants.[26]

The polysaccharide market is projected to reach over 22 billion dollars by 2030 and it is believed that the best opportunities and growth in production will be for the EPS from bacterial origin, followed by those of fungi, algae and plants.[27]

Distinct industrial activities require several microbial bioproducts. For instance, xanthan gum is globally used as a key stabilizer, emulsifier and thickener supplement in food, cosmetics and pharmaceutical products.[28] Furthermore, it has also been used by the oil industry during oil recovery strategies.[29]

The demand for bioproducts used during advanced oil recovery operations is expected to grow significantly with oil wells aging.[30–34] Thus, the global market of xanthan was estimated at $897 million and is projected to reach $1.5 billion by 2026,[35] due to the growing demand for gluten-free food and beverage additives as well as by-products for enhanced oil recovery operations.

Xanthomonas campestris can use a broad range of carbon sources to produce xanthan gum (starch, maltose, glucose, and sucrose),[36] glucose being a suitable substrate used to produce xanthan on an industrial scale.[37] However, the increasing demand for this biopolymer suggests that glucose will no longer be economically suitable as a raw substrate. Therefore, there is a need to use low-cost carbon sources to produce xanthan gum (such as saccharified starch or industrial wastes).[36]

We hypothesized that *X. campestris* can achieve high yields of xanthan gum production using oil PW and CG as a culturing medium.

The aim of this research is to test the use of PW and CG as main culturing media for the production of xanthan gum by *X. campestris*. This research shows the effect of using different PW concentrations and a dialysis PW pretreatment on xanthan gum yields. The product quality is evaluated by testing viscosity, energy dispersive X-ray spectroscopy (EDS), Raman spectroscopy and scanning electron microscopy (SEM) of the surface biofilm polymerization.

2 | MATERIALS AND METHODS

2.1 | Produced water sampling and chemical characterization

The PW used in this research was collected at a carbonate oil field in the primary stage of recovery. The PW and oil generated in the wellhead was first stored in a tank in order to allow water/oil phase separation. PW and oil were then transferred separately to their respective storage tanks. The PW was sampled at the opening valve during the procedure using borosilicate jars (Figure 1).

PW chemical characterization was carried out in order to determine chlorides compounds, pH, oxidation-reduction potential, total petroleum hydrocarbons (TPH), total dissolved solids, total alkalinity ($CaCO_3$), and sulfate and metallic concentrations. Such characterization was carried out by LEPETRO - Laboratório de Estudos do Petróleo, Bahia, Brazil. The lab reports the use of the appropriated methodology described by the Standard Methods for the Examination of Water and Wastewater[38] and the Selected Analytical Methods for Environmental Remediation and Recovery.[39]

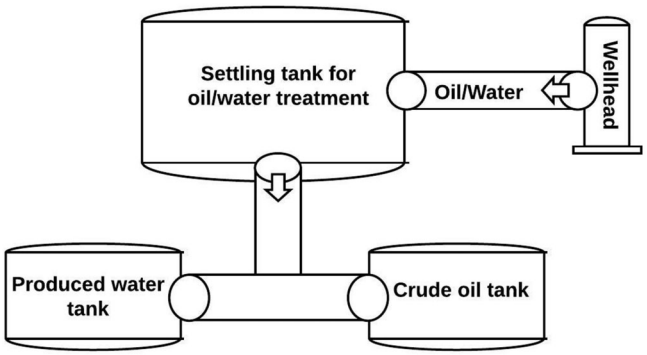

FIGURE 1 Diagram showing the position where produced water was collected

2.2 | Media preparation: control medium and produced water inorganic and organic supplementation

The control medium was prepared using the inorganic constituents of the "Mineral Salt Medium" (MSM)[40] supplemented with organics such as sucrose, CG and yeast extract. Distilled water was used for the preparation of the control medium. The MSM inorganic constituents are Na_2HPO_4 (2.7 g), KH_2PO_4 (1.0 g), $Ca(NO_3)_2.4H_2O$ (0.0713 g), $(NH_4)_2SO_4$ (1.0 g), $MgSO_4.7H_2O$ (0.2 g). Organic supplementation was carried out by adding yeast extract (0.5 g), Sucrose (25.0 g) and CG (20.0 ml). The pH was adjusted to 7.0 with 5 M NaOH and the final volume was 1.0 L.

The medium containing PW (production medium) at different ratios was prepared similarly to the control medium but by varying the proportion of distilled water to PW (10, 15, 25, 50, and 100% of PW - identified as 10% PW, 15% PW, 25% PW, 50% PW, and 100% PW, respectively). The same approach was used for preparing the tests with 100% dialyzed PW (identified as DPW).

The PW dialysis pretreatment step was performed in order to remove the excess of salts present in the effluent as described by Pinheiro *et al.*[41] and the media was pasteurized at 65°C/30 min instead of undergoing the full autoclaving process.

To estimate the content of inorganic nitrogen (N) in the production medium, the total inorganic nitrogen was calculated according to the mass percentage of this element in the constituents $CaNO_3.4H_2O$ and $NH_4SO_4.7H_2O$.

2.3 | Microorganism and inoculum preparation

The procedure for preparing the microbial inoculum consisted of inoculating 500 μl of a cryopreserved strain in 20 ml of a YM medium[42] contained in a 150-ml Erlenmeyer flask, incubated in a shaker (model I26 Incubator Shaker Series rotational incubator, New Brunswick Scientific; Edison, NJ) at 28°C, 150 rpm. After 32 h of incubation, a 20 ml of a mid-exponential phase culture was transferred to a flask containing 180 ml of freshly prepared YM medium. After a further 16 h of incubation, the culture was transferred to the batch or bioreactor testing experiments with 10^7 cells/ml.

2.4 | Xanthan gum production in shake-flasks

Xanthan gum production experiments in shake flasks was carried out in 250 ml Erlenmeyer flasks containing 45 ml of previously prepared production medium and 5 ml of inoculum. The flasks were subjected to 250 rpm orbital shaking at a temperature of 28 °C (model I26 Incubator Shaker Series rotational incubator, New Brunswick Scientific; Edison, NJ) for 120 h. The inoculum volume was 5 ml (10% of the final volume). After production the xanthan gum was recovered as described by Pinheiro *et al.*[41]

2.5 | Xanthan gum production experiments in bioreactor

After the identification of the operating conditions in the orbital shaker experiments that resulted in higher viscosity xanthan solutions (control - distilled water and 100% dialyzed PW), two production tests were carried out within a 5-L bioreactor in order to evaluate the performance in a pre-pilot production scale. A SCR05 Duo (Allbiom, Cajuru, Brazil) bioreactor was used with a volume of 1.8 L of freshly prepared medium with either distilled water or 100% dialyzed PW. Inoculation was carried out as previously described and aeration was provided by an air pump (3.0 L/min) attached to a filter (0.22 μm). The adjustment of pH (7.0) was carried out by an automatic injection of NaOH 5 M. The pH was monitored using an InPro3100 sensor (Mettler Toledo, OH) and dissolved oxygen using a polarographic InPro 6800 sensor (Mettler Toledo, OH). Stirring was maintained at 300 rpm and a hot water jacket controlled the temperature at 28 ± 2°C.

Microbial growth was monitored using the most probable number - MPN[43] and the production of xanthan was monitored through ethanol extraction and gravimetric analysis. Briefly, triplicates of 100 μl were aliquoted in a serial dilution (10^{-1}) using 96-well autoclavable deep-well plates previously prepared with 900 μl of a sterile medium (YM) and supplemented with 0.1% iodonitrotetrazolium (Sigma-Aldrich, San Luis) as microbial growth indicator.[44]

Xanthan assessments were carried out in a 10 ml triplicate. The material was centrifuged to remove biomass and the supernatant was treated for xanthan precipitation by ethanol.

2.6 | Physical–chemical characterization of xanthan gum

The obtained xanthan powder was subjected to physical-chemical assessments such as viscosity measurements, elemental assessment, SEM, and Raman spectroscopy.

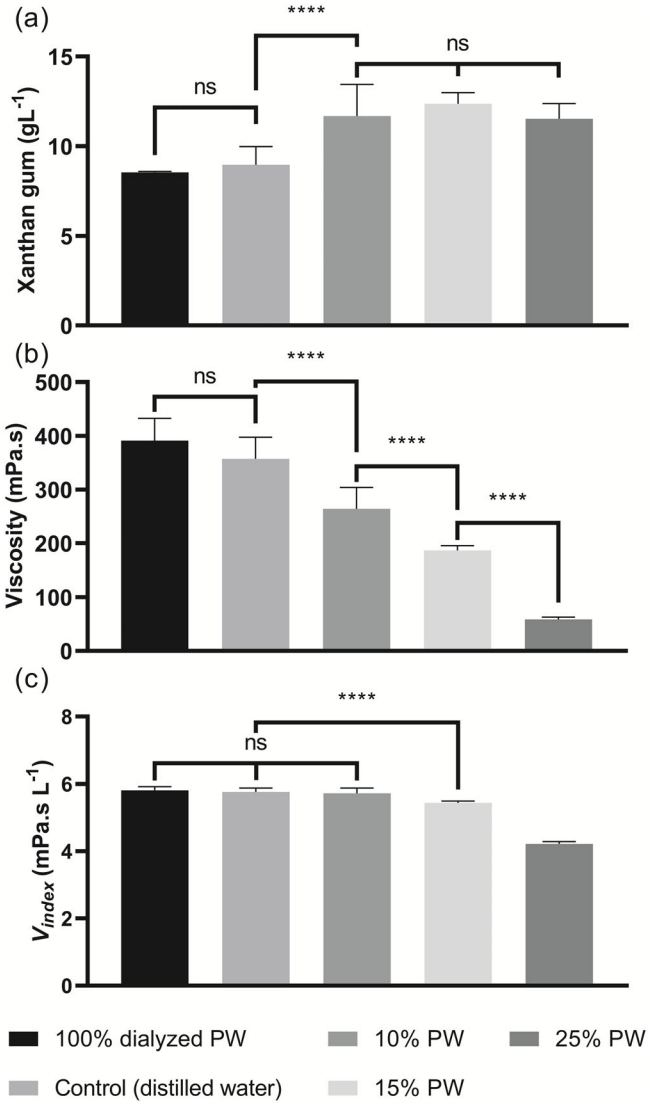

FIGURE 2 Xanthan gum production in batch mode shaker flasks. (a) Gravimetry; (b) Viscosity; (c) Vindex. Tests with 50% and 100% PW did not result in the production of xanthan. ****$P < 0.0001$

Viscosity was assessed using a Brookfield MVD-8 digital rotational viscometer (Marte Científica, Santa Rita do Sapucaí, Minas Gerais) with 1.0% polymer solution (wt/vol) at 25°C.

The elemental assessment was performed by energy dispersive X-ray spectroscopy (EDS) in a 20 mm^2 X-Max detector (Oxford Instruments, Oxfordshire, England). SEM was also used in order to investigate the details of the surface of xanthan biofilm produced with distinct PW concentrations onto a metallic surface. In this case, samples were covered in gold (10 min, 2 mbar) in the Desk V preparation system (Denton Vacuum, NJ) and observed by SEM using JSM-6610LV equipment (JEOL Brasil Instrumentos Científicos, Mirandópolis, São Paulo) at 10 kV and magnifications of 2000×.

This research proposes the calculation of a xanthan viscosity index in order to assist comparisons between different treatments (Equation 1).

$$V_{\text{index}} = \ln \frac{V \times P}{10} \quad (1)$$

where V is the viscosity in mPa s, P is the concentration of xanthan in g/L and 10 refers to the amount of xanthan used in the viscosity testing (g).

Statistical analysis of means, standard deviations, ANOVA and Tukey's multiple comparisons of triplicates of all variables were conducted using the GraphPad Prism 6 software (GraphPad Software, San Diego, CA).

A Raman spectrometer (model Cora 5700, Anton Paar GmbH, Graz, Austria) was used for the acquisition of Raman spectra as described by Sampaio *et al.*[45] A sample of pure xanthan gum from Sigma-Aldrich (G1253) was used as external standard.

3 | RESULTS AND DISCUSSION

3.1 | Xanthan gum production in shake-flasks

Tests using 10%, 15%, and 25% of PW diluted in distilled water showed a xanthan production of 11.6, 12.3, and 11.5 g/L, respectively (Figure 2a). There are no statistical differences between the total weights of xanthan recovered with these experiments. However, xanthan viscosity varied significantly in each treatment. Xanthan obtained with 25% of PW medium showed the lowest solution viscosity (58.74 mPa s) when re-suspended (Figure 2b), followed by 15% and 10% of PW (186.3 and 263.8 mPa s, respectively).

Therefore, the highest viscosity value was observed with the lowest PW ratio in the medium.

Xanthan conformational structure is dependent on the electrostatic forces of repulsion or attraction that can change with the presence of salts.[46–48] The higher the PW ratio in the culture medium, the higher the salts content and hence there is an increase in the electrostatic repulsion caused by the ions in the xanthan gum molecules. Consequently, the xanthan gum collapses to a small conformational structure, having a reduction in the interaction forces to other xanthan molecules and then reducing the viscosity of the solutions.[16–18] Additionally, the xanthan gum molecular weight is proportional to the viscosity of their solutions. Regarding this aspect, the literature reports the absence of changes in xanthan gum molecular weight by the salt content of their solutions.[49,50] Thus, it is possible that higher salinity because of higher PW ratios may have altered xanthan viscosity values. A change in the pyruvyl content of the xanthan gum was reported, which could increase the viscosity of the xanthan solutions.[15] Therefore, if the xanthan solutions are pre-treated with a dialysis process, they will reach a higher viscosity.

The average xanthan yields from a medium prepared with 100% dialyzed PW was of 8.55 g/L, but its viscosity after resuspension was 390.9 mPa s (Figure 2b). This is statistically similar to the averaged obtained with the control treatment (357.1 mPa s).

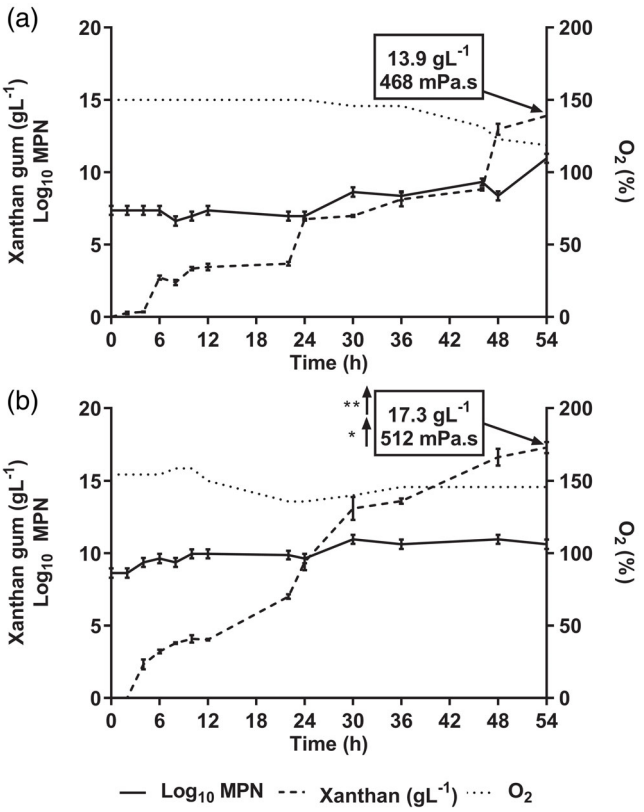

FIGURE 3 Xanthan production in the stirred-tank reactor. (a) Control (distilled water); (b) 100% dialyzed produced water. $^{*}P < 0.05$; $^{**}P < 0.01$

Xanthan gum is an important thickening agent for it shows a stable pseudoplastic behavior even at low concentrations and is inversely dependent on the shear stress.[51] However, the results suggest that quantification by weight is less important than viscosity assessments. Therefore, this research proposes the calculation of a viscosity index (Equation 1) for comparing the xanthan yields. Re-suspended xanthan from the 10%, 15%, 25% of PW experiments showed a viscosity index of 5.72; 5.44, and 4.21, respectively. The xanthan viscosity index figures for the product obtained with the control and DPW were 5.76 and 5.81, respectively (Figure 2c). Therefore, the xanthan produced with DPW can achieve the same viscosity observed with the control (distilled water).

It is important to highlight that this research shows significant yields of xanthan with low organic supplementation during a period of 120 h (11.5 and 8.55 g/L of xanthan with PW15% and DPW productions, respectively). Trindade *et al.*[52] reported lower yields (4.98, 4.55, and 4.07 g/L) when using CG (5.0%), sucrose (5.0%) or both synthetic supplementations (2.5% each). Similar result were reported elsewhere.[53] Higher production of xanthan is often associated with high carbon supplementations. Xanthan yields higher than 10 g/L may be achieved with the use of sugar cane molasses and yeast extract.[54] Very few reports show high yields with alternative medium made with effluent such as 8% cheese whey[55] or 17.5% of sugar beet molasses.[56]

3.2 | Xanthan gum production experiments in the bioreactor

The medium prepared with DPW showed the best result in the previous orbital shaker batch testing. Hence, this medium preparation was then selected for further testing of xanthan production within the bioreactor conditions. The results were compared with those of the control using a medium prepared with distilled water.

Several authors have reported that xanthan production is higher when nitrogen supplementation is low and oxygen concentration is high.[57–59] In this research, inorganic nitrogen supplementations (NO_3^- and NH_4^+) and aeration are similar to what is reported elsewhere.[42,60–62] Oxygen saturation within the culturing medium was always sustained above 100%. Using such an approach it was possible to increase xanthan yield to 17.3 g/L. This is 102% higher value than what was observed in the orbital shaker batch testing reported previously.

Xanthan production rates in rich synthetic media is reported to vary from 0.36 to 0.75 gL^{-1}/h.[63–65] This research reports a xanthan production rate of 0.34 and 0.23 gL^{-1}/h with a DPW-based medium and the control with distilled water within 48 h, respectively. A total xanthan production of 17.3 and 13.9 g/L was obtained during the whole 54-h incubation period for DPW and the control medium (Figure 3), respectively. Viscosity assessments showed a value of 512 and 467 mPa s for the xanthan produced with a DWP based medium and the control, respectively (Figure 3a,b).

Thus, it is clear that the organics present in the DPW assisted in the production of xanthan.

During bioreactor operation, medium viscosity increases due to xanthan accumulation resulting in oxygen depletion within the bioreactor. This is reported as the limiting factors affecting production.[61,62,65] In this research, however, oxygen levels were always above 100% saturation. Such high aeration and mixing were probably the operating variables responsible for a 102 and 55.1% increase in the total of xanthan obtained when using a DPW based medium and the control in the bioreactor compared to the results of the shaker experiments.

Correct organic supplementation is also important for increasing xanthan production. Peters *et al.*[62] reported a xanthan production of 18.9 g/L when using 5.5% of glucose. A similar production was observed with 4.0% sucrose.[42] In fact, larger xanthan yields (above 23 g/L) were only observed with the use of substrates with significant energetic value; such as 3% Ram horn hydrolysate (120 h of incubation) and continuously feeding the bioreactor with 5% glucose.[66] CG has also been used as carbon source for xanthan production. An average of 5.59 g/L of xanthan can be obtained after 120 h incubation period with 2% of CG.[67] This research reports a xanthan production of 17.3 g/L using 100% dialyzed PW supplemented with 2.5 and 2.0% of glucose and CG, respectively. The use of CG allowed a reduction in the glucose supplementation without decreasing xanthan production.

TABLE 1 Produced water physicochemical and metallic characterization

Characteristic	Result
pH (25°C)	6.50
Oxidation reduction potential – ORP Eh (mv)	189
Chloride (g/L)	71.4
Sodium (g/L)	13.9
Calcium (g/L)	1.64
Magnesium (g/L)	0.95
Salinity (g/L)	129
Total oils and greases (mg/L)	7.80
Total petroleum hydrocarbons – TPH (mg/L)	3.72
Unresolved complex mixture – UCM (mg/L)	0.97
Pristane (mg/L)	0.11
Phytane (mg/L)	138
Barium (mg/L)	0.95
Manganese (mg/L)	0.43

3.3 | Chemical characterization of the produced water

Chemical elements such as calcium and magnesium are important bacterial enzymatic cofactors (e.g., 8 and 129 mg/L) acting positively on supporting *Xanthomonas* spp. growth and the production of gum.[68–70] The PW chemical characterization (Table 1) showed the presence of high amounts of calcium and magnesium, respectively. Table 1 also shows the presence of manganese. This element in concentrations of up to 55 mg/L plays a significant role in the *Xanthomonas* spp. protection against oxidative stress.[71,72] This suggests a positive condition for this strain acclimation to toxic organics potentially present in the PW. Barium can affect microbial calcium and potassium uptake[73]; and it is only deleterious in concentrations above 15.68 g/L.[74] This is not the case with the PW used in this research (Table 1).

The main organic compounds found in PW are oils and greases and pristane and phytane that amounted to 150 mg/L. These compounds can be potential carbon sources for *Xanthomonas* spp.[75,76] In addition, *Xanthomonas* spp. has also been observed in oil contaminated environments.[77] Pristane and phytane are isoprenoids usually found in crude oil[78] and although more recalcitrant than other alkanes[79]; they can serve as carbon source and energy for microorganisms.[80]

Furthermore, some works reported strong evidence regarding the ability of *Xanthomonas* containing consortiums to catabolize these isoprenoids. For example, Hazaimeh *et al*.[81] showed the complete disappearance of phytane and pristane in crude oil after 6 weeks of a bioremediation experiment and Chalneau *et al*.[82] reported similar behavior in contaminated soil after 38 weeks. Despite the experiments of the present work being conducted in 5 days (shaker flask) and 54 h (bioreactor), it is believed that the high initial cell density, the control of temperature and the amendment of carbon sources (sucrose and CG) could enhance the catabolism of *X. campestris* population and consequently their ability to consume the residual hydrocarbons present in PW. This would explain the growth of *Xanthomonas* spp. observed in the bioreactor and higher yields of xanthan production when compared with the control (Figure 3).

3.4 | Physical–chemical characterization of xanthan gum

Microbial exopolysaccharides such as xanthan gum, undergo a change in their molecular conformation in the presence of salts. The molecule changes from a random spiral to a helix configuration. It is such a change that alters the rheological behavior of these hydrocolloids.[48,83] Furthermore, Cho *et al*.,[84] reported a significant reduction in rheological parameters when NaCl concentrations greater than 0.3% are present. In such a case, salt concentration shields the ionic groups in the xanthan gum side chains resulting in a reduction of its individual molecular volume.[85] Wyatt and Liberatore[86] define the phenomenon as a reduction of the hydrodynamic size and Meyer, Fuller, Clark, & Kulicke[87] as self-aggregation of the xanthan molecules. In both cases a reduction in xanthan viscosity is observed. This may explain the distinct results of viscosity obtained with increasing PW concentration ratios used in this research. Furthermore, as the dialysis can significantly remove salts from the PW; this pretreatment resulted in better quality of the xanthan production in regards to viscosity.

SEM imaging showed a smoother surface in the xanthan gum biofilm polymerization produced with 100% dialyzed produced water (DPW) and the control when compared with 10% PW (Figure 4a,b).

The xanthan gum mass fraction (wt%) determined by EDS showed a lower percentage of C and O, and an increase in Na, particularly as a result of growth on 10% PW treatment. The treatments with DPW and the control resulted in a gum with negligible wt% for Ca and Cl (Figure 4d,e). This was not observed with the product obtained with 10% PW treatment (Figure 4f). It was also observed that the presence of the metal Mo occurred only in the gum produced in PW treatments. Sodium molybdate is often used by the oil industry as an inhibitor of corrosion.[88–90] The presence of such an element in the xanthan gum may affect the lateral bonds of the heteropolymer and this would also explain its stability within the product.

Although the total C and O content was similar in the xanthan produced by control (distilled water) and DPW, their values were reduced by 16% in the xanthan samples obtained with 10% PW experiments (Figure 5). Salts and metals also varied in the three analyzed productions, showing a 4.6% and 10% higher value for DPW and PW than the control, respectively (Figure 5). This data suggests a strong capability of xanthan gum to adsorb the PW constituents, and therefore to treat it during the *X. campestris* cultivation.

Raman spectroscopy (Figure 6) showed a similarity in spectra between the commercial xanthan 99% (Sigma) and the polymers produced with different concentrations of PW and distilled water. Peaks

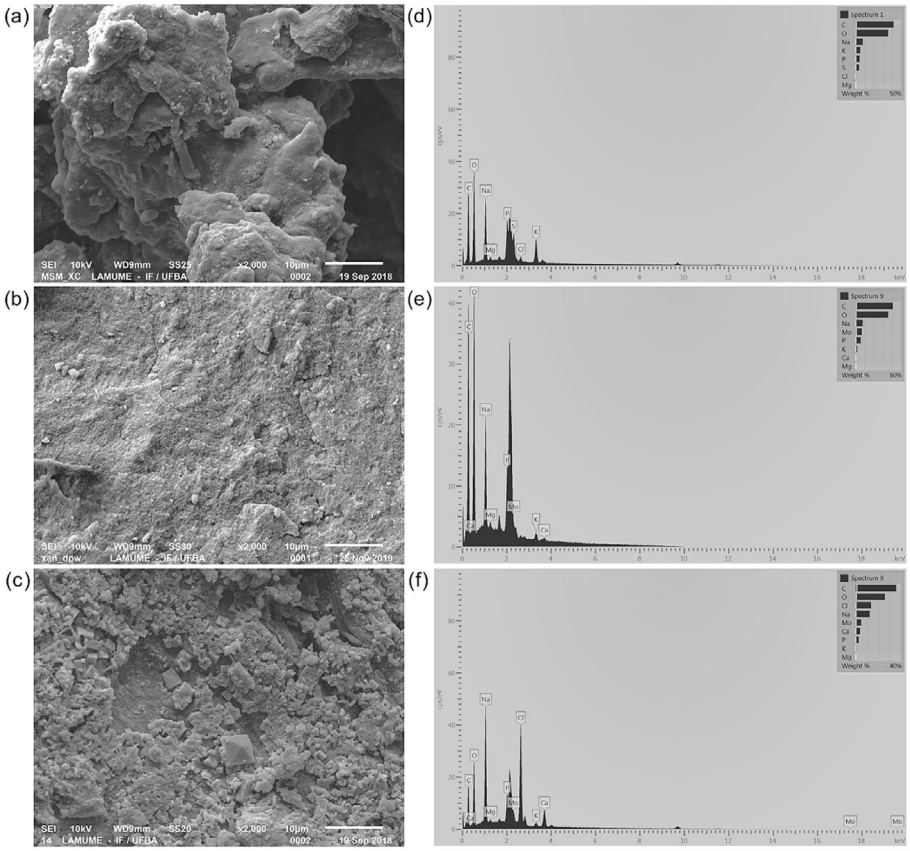

FIGURE 4 Scanning electron microscopy at 10 kV and magnifications of 2000× and Energy dispersive X-ray spectroscopy (EDS) of xanthan gum samples produced in shaker flasks. (a,d) Control: distilled water; (b,e) 100% dialyzed produced water; (c,f) 10% PW

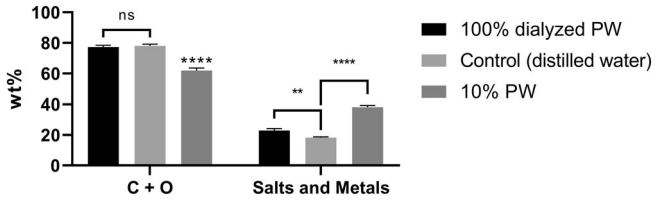

FIGURE 5 Energy dispersive X-ray spectroscopy (EDS) values for the total content of C, O, salts and metals in the xanthan obtained with 100% dialyzed produced water, control (distilled water) and 10% PW. ** $P < 0.01$; **** $P < 0.0001$

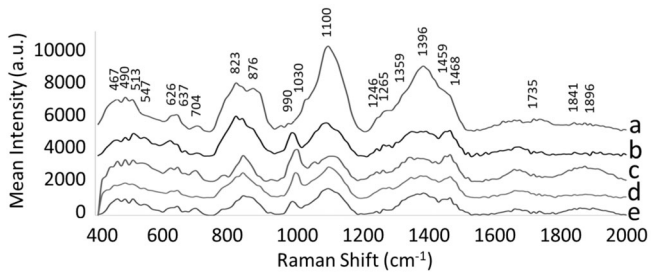

FIGURE 6 Raman spectroscopy (1064 nm) with dislocated spectra. (a) Commercial xanthan gum; (b) Control (distilled water); (c) 25% PW; (d) 10% PW; (e) 100% dialyzed produced water

attributed to mannose (513; 637; 823; 876; 1459 cm^{-1}), peaks attributed to glucose (547; 1140 cm^{-1}) and peaks attributed to mannose and glucuronic acid (1030 cm^{-1}) were easily observed.[41]

4 | CONCLUSION

This research showed that it is feasible to produce 17.3 g/L of xanthan within 54 h using 100% dialyzed PW supplemented with low concentrations of CG and sucrose (2.0% and 2.5%, respectively). The presence of salts may be a significant variable affecting *Xanthomonas* spp. growth on PW. Recycling PW and CG as a medium for xanthan production shows significant economic potential and environmental sustainability as a waste management strategy for the oil industry.

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CONFLICT OF INTEREST

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AUTHOR CONTRIBUTION

Igor Carvalho Fontes Sampaio: methodology, investigation, formal analysis, writing - original draft preparation. Pedro Jorge Louro Crugeira: investigation. Joalene de Azevedo Santos Ferreira: investigation. Jacson Nunes dos Santos: investigation. Josilene Borges Torres Lima Matos: writing - review & editing. Antônio Luiz Barbosa Pinheiro: writing - review & editing. Fabio Alexandre Chinalia: writing - review & editing, approval of the submitted and final versions. Paulo Fernando de Almeida: writing - review & editing.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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