Biodegradation of Polypropylene in Presence of Chromium Mediated by Stenotrophomonas sp. and Lysinibacillus sp. Isolated from Wetland Sediments

Biodegradation of polypropylene in presence of chromium mediated by *Stenotrophomonas* sp. and *Lysinibacillus* sp. isolated from wetland sediments

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Abstract

Plastics and heavy metals are deeply integrated into daily life, and their production and consumption contribute to waste generation and environmental contamination. In this study, native microorganisms from wetland sediments of the Paraná Delta, Argentina, were used to evaluate the simultaneous bioremediation of chromium and polypropylene. A total of 73 isolates were obtained from sediments contaminated with both heavy metals and microplastics using enrichment culture media. These isolates were characterized based on their metabolic properties through biochemical tests. Redundant isolates and those exhibiting signs of pathogenicity were discarded. From the remaining 51 isolates, a selection was made based on their biofilm-forming capacity, polypropylene biodegradation potential, and chromium resistance. These microorganisms were identified by sequencing the 16S rRNA region. The results showed that bacteria belonging to *Stenotrophomonas* sp. reduced the total polypropylene degradation time by more than 100-fold. Additionally, microbial activity led to detectable changes in the polypropylene surface and structure, as evidenced by modifications in surface morphology and FTIR spectra in the inoculated treatments. In the presence of 5 ppm Cr(VI), *Lysinibacillus* sp. achieved better results, reducing the degradation time by over 10-fold compared to the uninoculated treatment and demonstrating an enhanced ability to form biofilms under these conditions. At the same time, some removal of Cr(VI) (around 8%) was observed. On the other hand, *Stenotrophomonas* sp. reduced the degradation time 7-fold compared to the uninoculated treatment under these conditions. This is the first time that *Stenotrophomonas* sp. and *Lysinibacillus* sp. are reported to degrade polypropylene in the presence of other contaminants.

Keywords. bacteria; bioremediation; heavy metals; plastic; polypropylene; wetlands.

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Introduction

Continuous industrialization and globalization, coupled with the absence of effective waste management infrastructures, have progressively deteriorated natural environments by introducing components that compromise their holistic functioning. Plastics and heavy metals have multiple applications in daily life, whether for packaging, transportation, the elaboration of electronic devices, or textiles. However, their excessive use generates a high environmental impact [1]. In practice, landfilling is the most common method of final waste disposal; however, plastic can remain for many centuries in both terrestrial and aquatic environments, without some form of transformation, altering soil fertility and causing a detrimental effect on local inhabitants [2].

Unlike plastics, heavy metals cannot be degraded, and they persist in the environment. One of the most common heavy metals released to the environment due to anthropogenic activities is chromium (Cr) and its toxicological effect depends on its oxidation state. In the environment, Cr is found mainly in two forms: Cr(III), which is relatively harmless, and Cr(VI), which is considered a toxic species and highly soluble in water [3].

Meanwhile, the slow degradation rate of plastics under natural conditions, and the chemical nature of Cr, lead to the accumulation of these components in the environment. Additionally, there are studies that address the presence of both types of pollutants in the environment as combinations of microplastics and As, Ti, Ni or Cd [4]. Particularly, some studies also reported the presence of these pollutants simultaneously at different sites located in the wetland Ecoregion of Paraná Delta and Islands, Argentina [5, 6]. However, while the co-occurrence of microplastics with heavy metals has been documented, studies addressing the simultaneous presence of polypropylene (PP) and Cr in the environment remain scarce. This gap in knowledge is particularly relevant given that both pollutants are frequently detected in industrially impacted ecosystems. Given the affinity of heavy metals for plastic surfaces, it is likely that Cr ions interact with PP particles, potentially altering their environmental fate and bioavailability.

The presence of these contaminants in the environment represents a toxicological threat, and for that reason, much effort through the last decades has been invested in mitigating these effects. A promising alternative capable of contributing to the reduction of the environmental impact generated by microplastics and/or metals, as a consequence of anthropogenic activities, is bioremediation. Microorganisms contribute to bioremediation through specific biological processes, such as biotransformation and biodegradation, which modify or break down contaminants into less toxic forms [7]. The selection of an appropriate bioremediation strategy

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depends on factors such as the type of pollutant, environmental conditions, and the efficiency of microbial activity in the given context.

Biodegradation could potentially be applied to plastics/microplastics pollution. Indeed, sites contaminated with plastic harbor different potential polymer degraders. Plastic biodegradation occurs as a consequence of biofilm formation on the polymer surface. Biodeterioration and bio fragmentation result in destabilizing the carbon skeleton of the polymer, which promotes its breakdown into oligomers, dimers and monomers [8]. Therefore, carbon chain hydrolysis decreases the molecular weight of the polymer and, simultaneously, weakens the polymer's mechanical properties, making it more accessible for further microbial assimilation as a source of carbon and energy [9]. There are several studies dealing with the biodegradation of microplastics. A novel and recent publication collected several studies highlighting the fundamental role of microorganisms in transforming microplastics in the environment, while trying to unravel the complex and multifactorial area of study [10]. Another study reported the decomposition of polystyrene microplastics using a combination of three bacterial cultures: *Stenotrophomonas maltophilia*, *Bacillus velezensis* and *Acinetobacter radioresistens* [11].

On the other hand, most of the treatments used to transform Cr(VI) to its innocuous state involve high costs due to high reagents consumption and energy requirements and may end in an incomplete metal removal. Fortunately, as stated by several studies found in bibliography, Cr(VI) can be reduced to Cr(III) through direct or indirect microbiological reduction; representing an attractive alternative in bioremediation processes [12]. As an example, some bacterial isolates were found to be able to reduce 50% of the Cr(VI) to Cr(III) at an initial concentration of 100 mg/l [13]. Also, it was reported that the reduction of Cr(VI) to Cr(III) can be performed by *Agrobacterium* sp. and *Lysinibacillus* sp. [14].

Whereas some studies reported the co-occurrence of metals and microplastics, and how the latest could affect the fate of metals in the environment, there are scarce reports where simultaneous remediation is assessed, and they mainly focus on the interaction of microorganisms with heavy metals and organic compounds [15]. It has been reported that microorganisms isolated from plastic surfaces were shown to be resistant to the presence of heavy metals [16]. Another study indicated the influence of microplastics in the oxidation of Cr(III) and organic matter [17]. However, to the best of our knowledge, the biodegradation of plastics in the presence of heavy metals has not been systematically studied. This study aims to investigate how heavy metals influence the biodegradation process of plastic considering that the presence of these metals may alter microbial activity and the efficiency of degradation.

In the present study, bacterial species from contaminated wetlands were isolated, characterized, and identified to evaluate their potential for bioremediation through two distinct aspects: Cr(VI) removal and PP biodegradation. The study aimed to analyze surface changes in

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PP pellets induced by microbial activity and to measure the removal of Cr(VI). Both effects were investigated separately and then analyzed together in a joint assay.

Materials and methods

Environmental samples from wetlands

Sediment samples were taken from sites belonging to two wetlands of the Ecoregion of the Paraná Delta and Islands, Argentina: from the coastal wetland of the Río de la Plata, Buenos Aires (Vicente López (34°29'52.80"S 58°28'37.20"W), Puerto Madero (34°42'25.20"S 58°13'44.40"W) and Quilmes (34°37'04.80"S 58°20'45.60"W)) and from the wetland of the South Paraná River, Entre Ríos (33°29'32"S 58°43'44"W), all sites belonging to the Ecoregion of the Paraná Delta and Islands. All sampled sites are publicly accessible. Each sampling point consisted of a quadrat measuring 0.25 m on each side and 0.05 m in depth. The collected samples were stored in metal containers, transported to the laboratory for analysis, and subsequently maintained at 4°C until processing. It has been previously reported that sediment from Buenos Aires contains plastic particles [6]. The identification criteria for these particles were taken from the study proposed by Ng and Obbard [18]. On this basis, the presence of plastic fragments in sediment samples collected in Entre Ríos was analyzed using physical separation techniques by density. Optical visualization was performed under a magnification of 6.5X using the technique described by Lacava et al. [6].

Isolation of microorganisms from environmental samples

2 g of sediment were placed in 100 ml of liquid LB medium (NaCl 5 g/l, yeast extract 5 g/l, and tryptone 10 g/l) and incubated at 30°C and 200 rpm for 24 h for the isolation of microorganisms from environmental samples. After incubation, serial dilutions were performed in peptone water (NaCl 8.5 g/l, Peptone 1 g/l), and placed onto Petri dishes with agarized LB medium (LB with the addition of 15 g/l agar). The plates were incubated at 30 °C for 24-48 h. Each visually differentiated colony was placed on agarized LB medium and incubated at 30°C for 48 h to isolate the bacteria.

The composition of the bacterial cell wall was determined using the Gram staining test. Biochemical tests were performed on the Gram-negative bacteria group, which allowed the isolates to be categorized according to their metabolic properties: Triple Sugar Iron agar (TSI), Lysine Iron Agar (LIA) and Indole, Methyl Red, Voges-Proskauer and Citrate tests (IMVIC).

Inocula preparation

Inocula preparation of the isolates was carried out in LB medium at 30°C and 200 rpm for 24 h for all subsequent assays. The cultures were centrifuged at 4000 rpm for 10 minutes, and the pellet was resuspended in sterile distilled water. The optical density (OD) at 560 nm of the

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bacterial resuspended was measured to calculate the inoculum volume necessary to obtain the initial OD for each assay (initial OD of 0.2 for the biofilm formation tests and 0.1 for the PP degradation and growth in the presence of Cr assays, as detailed below).

Evaluation of the biofilm formation

To determine the ability of isolates to grow by forming biofilm, the crystal violet (CV) staining described by O'Toole [19] was performed. Inocula of each bacterial isolate was prepared as previously described. LB medium without NaCl was inoculated to obtain an initial OD of 0.2 and then 200 µl of each OD-standardized culture was placed in the wells of a sterile 96-well polypropylene plate. The plate was incubated at 30°C for 48 h. *Bacillus subtilis* was used as positive control due to its known ability to grow forming biofilm [20]. After incubation, the contents of each well were carefully removed and washed with distilled water. Then, 250 µl of a 0.1% w/v CV solution was added and the plate was incubated for 15 minutes at room temperature. After removing the CV solution, three washes with distilled water were performed, and the plate was left to dry for 1 h. Subsequently, 250 µl of ethanol was added to each well and incubated for 15 minutes at room temperature. Finally, the OD at 595 nm was measured. The experiment was conducted in three independent trials, with 5 replicates per treatment. The selection of the microorganisms with the best capacity to form biofilm was carried out according to the criteria described by Stepanović et al. [21].

Polypropylene degradation by the selected bacteria

Prior to examining the potential growth of microorganisms using PP as a carbon and energy source, the microorganisms were adapted to grow at decreasing concentrations of glucose as the carbon and energy source. For this purpose, inocula were prepared in LB medium, as previously described. Each bacterial resuspension was used to inoculate 10 ml of M9 medium with the addition of glucose 4 g/l. Incubation of the cultures was performed at 30°C and 200 rpm for 24 h. After incubation, the cultures were centrifuged, washed, and resuspended in distilled water, and this bacterial resuspension was used to inoculate the M9 medium with glucose 2 g/l. Incubation of the cultures was performed as previously described. Centrifugation and washing procedures were performed again after incubation, and the bacterial resuspension was used to inoculate M9 culture medium (without glucose addition) with 0.5 g of PP pellet (3 mm x 2 mm each), kindly provided by Arlanco S.R.L. (Ciudad Autónoma de Buenos Aires, Argentina). Prior to use, the PP was dried at 60°C for 5 days to determine its dry weight and then exposed to UV radiation for 2 hours to eliminate potential microbial contamination, with periodic repositioning to ensure uniform surface exposure. This pretreatment was performed prior to each trial in which the PP pellet was used, so hereafter, it will be referred to as UV-pretreated PP.

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After inoculation of the systems with PP as the only carbon and energy source, the cultures were incubated at room temperature and 160 rpm for 40 days, during which changes in the structure and surface of the plastics have been reported in previous studies [22]. At the initial time and every 10 days, viable microorganisms were counted, and the OD at 560 nm was measured. At the final time, the weight of the UV-pretreated PP dried for 5 days at 60°C was measured. Based on the UV-pretreated PP dry measurements at initial and final time, degradation parameters were determined following the Equations (1), (2), and (3) [23], where *Wo* is the initial weight (g), *Wf* is the final weight (g) of PP, *t* is time (days) and *K* is the first-order rate constant for PP degradation.

$$Weight\ loss\ (\%) = \frac{Wo}{Wf*100} \quad (1)$$

$$K = -\frac{1}{t} * ln\frac{Wo}{Wf} \quad (2)$$

$$t\ \frac{1}{2}(days) = \frac{ln\ 2}{K} \quad (3)$$

The value of the parameter *K* is characteristic of each bacterium and is indirectly proportional to the estimated degradation time, i.e. the higher the value of *K*, the shorter the average time necessary for the degradation of the plastic.

Furthermore, UV-pretreated PP was evaluated by Scanning Electron Microscopy (SEM) to compare the polymeric surface of the inoculated treatments and the positive and negative control. Prior to SEM analysis, samples were washed with 2% (v/v) aqueous sodium dodecyl sulfate (SDS) and with distilled water as described by Skariyachan et al. [24] to remove non-adherent biomass and biofilms, ensuring that the observed surface modifications were primarily due to polymer degradation rather than microbial residues. The PP samples were sputter coated with a silver layer under an atmosphere of approximately 7.75 x 10^{-2} torr in the Denton Vacuum equipment and then analyzed using a JEOL scanning electron microscope. Additionally, UV-pretreated PP after incubation was analyzed by Fourier Transform Infrared Spectroscopy (FTIR) to determine its composition at the molecular level. FTIR spectra were recorded using Nicolet iS50 FT-IR spectrometer with ATR (Thermo Scientific), at room temperature. The samples were examined in the range 4000-400 cm^{-1}.

Bacterial growth at different concentrations of Cr(VI)

Bacterial growth was evaluated in LB culture medium containing 0 ppm, 1 ppm, 3 ppm, and 5 ppm Cr(VI). The concentrations analyzed were selected based on previous reports of concentrations that can be found in the environment and considering that the allowable Cr(VI)

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concentration in industrial discharges is 2 ppm [25]. Being such a highly toxic metal, the permissible concentrations in drinking water are 0.1 ppm according to the United States Environmental Protection Agency (USEPA) and 0.05 ppm according to the World Health Organization (WHO) [26], and concentrations above 0.2 ppm of Cr(VI) are already considered toxic [25]. Therefore, the concentrations used in this study exceed these values. Each condition was prepared from a stock solution of 1000 ppm Cr(VI) prepared with $K_2Cr_2O_7$. Inocula were performed in liquid LB as previously described. The initial OD at 560 nm of the cultures was 0.1. Cultures were incubated at 30°C and 200 rpm for 24 h. OD measurements were recorded at the initial and final times. The experiment was conducted in three independent trials, with 3 replicates per treatment.

Evaluation of the removal of Cr(VI) mediated by microorganisms

The determination of Cr(VI) removal mediated by microorganisms was performed in batch cultures. Treatments were designed with 5 ppm Cr(VI) as initial concentration in liquid LB culture medium. Prior to inoculation, Cr(VI) measurements were carried out by a colorimetric method [27]. The inocula were prepared as previously described. Cultures were placed at 30°C and 200 rpm for 48 h. After the incubation period, cultures were centrifuged, and Cr(VI) was measured as described. The removal of Cr(VI) was estimated as the difference between the concentration at initial and final time. The experiment was conducted in three independent trials, with 3 replicates per treatment.

Bacterial culture in the presence of polypropylene and Cr(VI)

The procedure for the analysis of the interaction between the selected bacteria, PP and Cr, was similar to that previously described for the growth assay on PP. 5 ppm of Cr(VI) was added to each treatment containing 0.5 g of UV-pretreated PP pellet in M9 culture medium. The initial OD of the cultures was standardized to 0.1. The negative control consisted of treatments with uninoculated M9 culture medium, 0.5 g PP pellet, and 5 ppm Cr(VI). At the initial and final time (40 days), colony-forming units (CFU), Cr(VI) and dry weight of PP were checked as previously described. The experiment was conducted in three independent trials, with 3 replicates per treatment.

Additionally, the capacity of the studied bacteria to form biofilm was evaluated under previously described conditions in sterile 96-well polypropylene plates, both in the presence and absence of 5 ppm Cr(VI), to assess the potential influence of the metal on this type of growth. LB medium with and without the metal was used as controls. The experiment was conducted in three independent trials, with 5 replicates per treatment.

Identification of microorganisms by sequencing of the 16S rRNA gene

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Microorganisms were identified by sequencing of the nearly full-length 16S rRNA gene, performed by Macrogen Inc. (Seoul, South Korea). PCR amplification was carried out using the universal primers 27F (5'-AGA GTT TGA TCM TGG CTC AG-3') and 1492R (5'-TAC GGY TAC CTT GTT ACG ACT T-3'). The PCR products were purified and sequenced using the internal primers 785F (5'-GGA TTA GAT ACC ACC CTG GTA GTA-3') and 907R (5'-CCG TCA ATT ATT CMT TTR AGT TT-3'), resulting in nearly full-length sequences of approximately 1400 bp. Raw FASTA sequences were deposited in the Sequence Read Archive (SRA) database of the National Center for Biotechnology Information (NCBI). The identification of the microorganisms was carried out by comparison of homologous sequences using the BLASTn (Basic Local Alignment Search Tool) of NCBI. The List of Prokaryotic Names with Standing in Nomenclature (LPSN) was consulted for the genus and species level identification of the microorganism and its comparison with the reference strain. While it was previously accepted that a similarity percentage between the isolate and the type strain above 97% was sufficient to conclude that the microorganism belonged to the same genus and species, Chun et al. [28] proposed raising the cutoff to 98.7%. This criterion was used to identify the isolated microorganisms.

Statistical analysis

Each parameter (PP pellet dry weight, Cr(VI) concentration, and CFU/ml) was analyzed separately using the Wilcoxon signed-rank test for paired samples to compare initial and final measurements. These measures were expressed as mean percent weight loss ± standard deviation (SD), percent Cr(VI) removal ± SD and CFU/ml ± SD, respectively. On the other hand, the differences in ΔOD between the different Cr(VI) concentrations evaluated in the assay *Bacterial growth at different concentrations of Cr(VI)* were analyzed by one-way ANOVA followed by Tukey's Multiple Comparison Test for each microorganism separately. Prior to performing ANOVA, data were tested for normality using the Shapiro-Wilk test and for homogeneity of variances using Levene's test. ANOVA was applied only when these assumptions were met. Statistical analysis of the biofilm formation capacity in the presence of 5 ppm Cr(VI) was performed by t-test between each culture in the presence and absence of metal. Each series of experiments was performed in triplicate or quintuplicate depending on the assay. The experiments were repeated separately to ensure reproducibility. The results obtained were considered statistically significant if p value < 0.05. All statistical analyses were performed using *Infostat* statistical software version 2019, FCE, National University of Córdoba.

Results

Plastic contamination in environmental samples

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The presence of plastic in sediment samples from Entre Ríos was qualitatively determined. The filter papers were visually inspected under magnification of 6.5X. Results are shown in Supplementary Information 1, where it can be observed that different forms of plastic were found: fibers, beads and sheets. The results of samples from Buenos Aires have been reported by Lacava et al. [6].

Isolation of microorganisms from environmental samples

From the enrichment of the environmental samples, 73 microorganisms were isolated and analyzed (Supplementary Information 2). Gram staining results determined that 14% were Gram-positive bacteria and 86% were Gram-negative. The number of isolates was reduced from 73 to 51 by discarding 11 microorganisms belonging to the same sample that showed equal results in the biochemical tests and 11 bacteria that presented positive lysine deaminase activity. The latter result is distinctive for strains of the genus *Proteus*, *Providencia,* and *Morganella* spp. Studies report that these genera can cause respiratory, gastrointestinal, or urinary infections in humans, so its use in subsequent assays was discontinued as they would represent a threat to human health despite their bioremediation potential. In this context, 51 microorganisms were selected to analyze their biofilm-forming capacity.

Evaluation of the biofilm formation of the selected microorganisms

The isolates were assessed in their capacity of forming biofilm by using CV as described in the methods section. Five bacteria showed OD values that were indicative of a high production of exopolysaccharides, in contrast to the rest of the bacteria assessed. The isolates that showed the highest biofilm formation capacity were selected for further testing: 34B (from Entre Ríos), PM6, PM7, VL5 and Q2 (from Buenos Aires).

Polypropylene degradation by the selected bacteria

a. Bacterial growth in the presence of polypropylene

To assess bacterial growth in the presence of PP, bacterial counts at the initial time and every ten days are shown in Fig. 1. In each treatment, no growth other than that of the inoculated microorganism was observed. Results showed the behavior of each microorganism using PP as the only carbon and energy source. Except for 34B, a relatively stable viable count value during the 40 days of the trial was maintained by all the bacteria analyzed. On average, the final bacterial count increased between 1 and 2 orders of magnitude compared to the initial value. In addition, no changes in OD values were observed at the different times analyzed.

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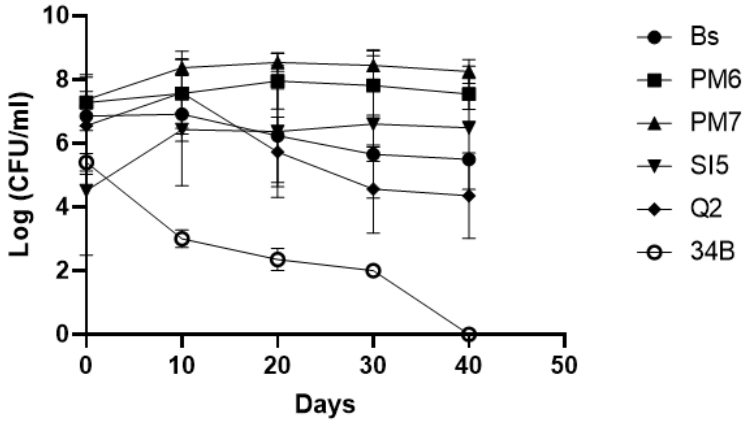

Fig. 1 Bacterial counts of PM6, PM7, VL5, 34B and Q2 and *Bacillus subtilis* (Bs, positive control) in treatments with UV-pretreated PP as the only carbon and energy source. The data are the means of three biological replicates, with the bars representing the standard errors of the means.

b. Quantitative analysis of changes in polypropylene

The results obtained from the experiment of the bacterial exposure to UV-pretreated PP and its biodegradation potential are shown in Table 1.

Table 1. UV-pretreated PP degradation parameters of PM6, PM7, Q2 and VL5, and of the positive (*B. subtilis*) and uninoculated controls.

Treatment	Weight loss (%)	K	Average total degradation time (days)
B. subtilis	1.120 ± 0.870	2.83 E-04	2449.28
PM6	2.015 ± 1.000	5.10 E-04	1359.11
PM7	1.680 ± 0.810	4.25 E-04	1630.93
Q2	0.540 ± 0.220	1.35 E-04	5134.42
VL5	0.080 ± 0.040	1.99 E-05	34831.51
Uninoculated	0.013 ± 0.023	3.33 E-06	208152.3

PM6 showed a significant decrease in the dry weight of UV-pretreated PP at the final time, resulting in a higher percentage of weight loss. PM7 did not show significant differences in weight measurements but had a weight loss percentage similar to PM6. Moreover, PM6 and PM7 showed a higher percentage of weight loss than *B. subtilis*. On the other hand, VL5 had a weight loss percentage similar to the uninoculated treatments. Q2 showed a lower average weight loss percentage compared to PM6 and PM7, but higher than VL5. Furthermore, it can be observed that the higher the percentage of dry weight loss correlates with a shorter time required for the

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total degradation of UV-pretreated PP compared to the uninoculated control. Particularly, in the treatments with PM6 and PM7, the degradation time was reduced by more than 100 times compared to the uninoculated treatments.

c. Qualitative analysis of changes in polypropylene

Scanning electron microscopy

Qualitative analysis of the UV-pretreated PP at final time by SEM allowed visualization of the plastic surface of both inoculated treatments and uninoculated control (Fig. 2). The treatment with 34B was not included in this analysis, since it did not show growth during the period studied.

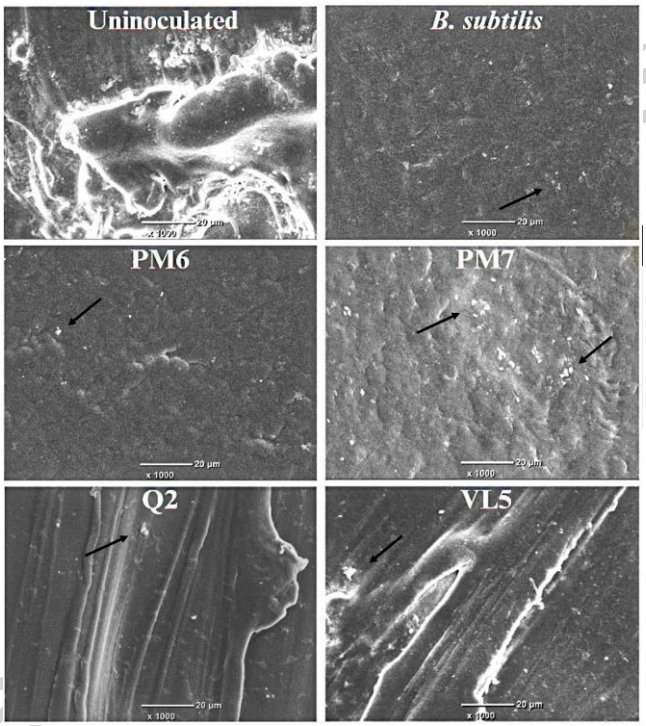

Fig. 2 Scanning Electron Microscopy (SEM) visualization of the UV-pretreated PP surface at final time (40 days) corresponding to the uninoculated PP, and the treatment inoculated with *B. subtilis*, PM6, PM7, Q2 and VL5. Bars represent 20 µm at 1000X magnification. Arrows indicate microorganisms attached to the surface.

The SEM analysis revealed that the PP from the uninoculated treatment exhibited greater surface roughness compared to the inoculated treatments. Interestingly, in the samples with higher weight loss (Table 1), the polymer surface appeared smoother, with fewer irregularities (as observed in PP inoculated with *B. subtilis*, PM6, and PM7). Conversely, the PP inoculated with Q2 and VL5, which exhibited lower biodegradation percentages (Table 1), presented a rougher

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surface compared to the previously mentioned strains, retaining the patterns observed in the uninoculated control but with a smoother appearance.

Prior to SEM analysis, samples were washed as previously described to remove non-adherent biomass and biofilms, ensuring that the observed surface modifications were primarily due to polymer degradation rather than microbial residues. Nevertheless, in some images of Fig. 2 microorganisms still adhered to the surface can be observed, marked with arrows.

FTIR spectroscopy

The FTIR spectroscopy analysis revealed specific changes in the chemical structure of PP in the samples treated with microorganisms (Fig. 3A). Compared to the uninoculated control, the inoculated samples (PM6, PM7, and Q2) showed noticeable variations in the spectral regions between 1000-1200 cm^{-1} and 2800-3000 cm^{-1}, corresponding to C-O and C-H stretching vibrations, respectively (Fig. 3B and 3C). In the 1000-1200 cm^{-1} region, PM7 and Q2 exhibited changes in peak intensity near 1100 and near 1160 cm^{-1}, with PM7 showing the most pronounced variations and PM6 showing a moderate variation with respect to the uninoculated control. In the 2800-3000 cm^{-1} region, more marked changes were observed in the peaks around 2840, 2920 and 2950 cm^{-1} for all three treatments, with PM7 showing the greatest extent of variation among the tested strains. These spectral modifications, indicated by arrows in Fig. 3, are consistent with the surface alterations observed in the SEM images (Fig. 2). The complete spectra for all analyzed isolates are available in Supplementary Information 3.

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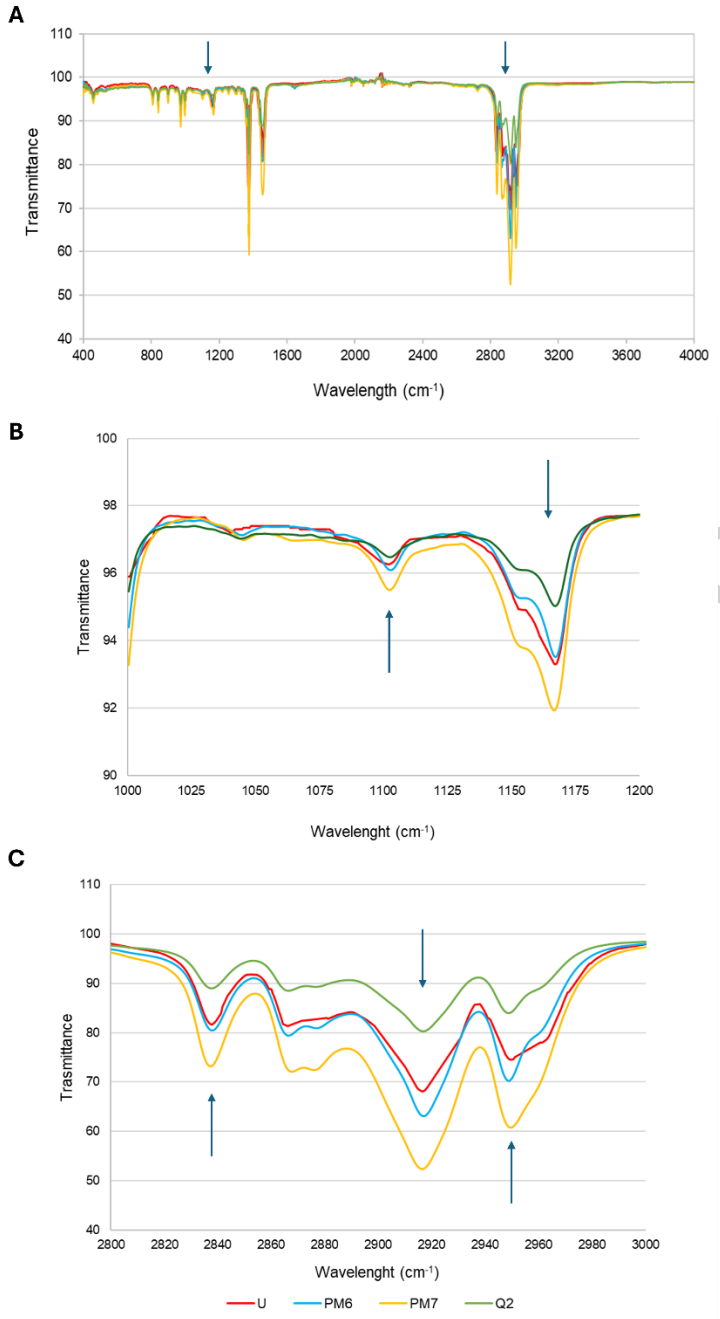

Fig. 3 FTIR analysis of UV-pretreated PP after 40 days of microbial treatment. Full FTIR spectra of PP after incubation with PM6, PM7, Q2, and the uninoculated control (U) (A). Spectral region between 1000-1200 cm^{-1} (B) and 2800-3000 cm^{-1} (C). Arrows indicate relevant regions with differences in inoculated and uninoculated spectra.

Bacterial growth at different concentrations of Cr(VI)

The study examined the effect of different concentrations of Cr(VI) on the growth of the 5 bacterial isolates that showed the highest biofilm formation capacity. Results are shown in Fig. 4. The data are the means of three biological replicates, with the bars representing the standard

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errors of the means. Statistical comparison of the measured variable (average Δ OD) was performed between the conditions evaluated for the same microorganism.

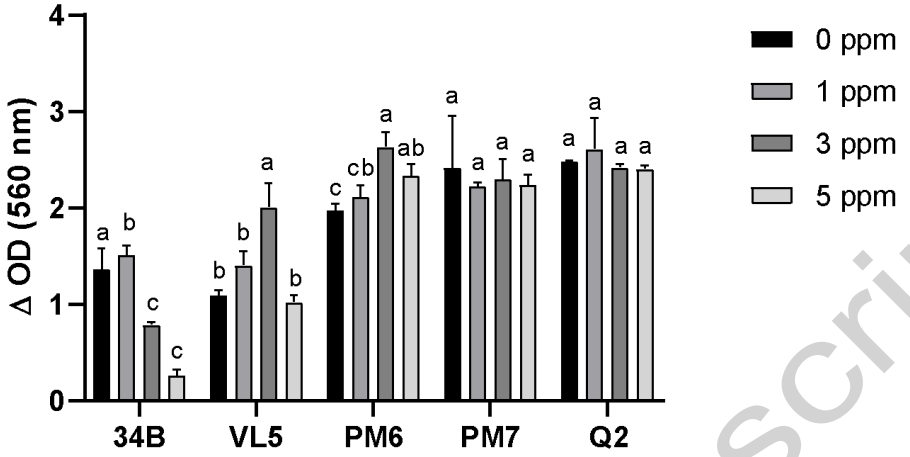

Fig. 4 Growth of 34B VL5, PM6, PM7 and Q2 in LB with 0, 1, 3 and 5 ppm Cr(VI) expressed as Δ OD. One-way ANOVA followed by Tukey's Multiple Comparison test were performed for each microorganism separately. Average OD values with different letters are significantly different ($p < 0.05$) in each treatment.

The results showed that 34B had a significant decrease in growth when exposed to higher concentrations of Cr(VI). Conversely, PM6 exhibited a significant increase in growth when exposed to 5 ppm of Cr(VI). For VL5, there was no significant difference in growth between the 0, 1, and 5 ppm conditions, but there was a significant increase in growth at 3 ppm. However, for PM7 and Q2, there were no significant changes in growth when exposed to any of the concentrations evaluated.

Selection and identification of microorganisms

Based on the results of the previous experiments, it was decided to use isolates PM6, PM7 and Q2 for further testing. These microorganisms displayed a higher percentage of PP weight loss and exhibited significant biomass generation when exposed to 5 ppm of Cr(VI), while isolate 34B did not tolerate the tested Cr(VI) concentrations and isolate VL5 was unable to grow when PP served as the sole carbon source.

The raw sequences of 16S rRNA gene for these microorganisms were deposited in the SRA database under the Accession Numbers listed in Table 2, and the phylogenetic analyses are provided in Supplementary Information 4. A homologous sequence comparison analysis was conducted using the BLASTn database to determine the most probable taxonomic assignment for each bacterial species. Taking into account the criterion previously described, both PM6 and PM7 were identified as *Stenotrophomonas rhizophila*. However, the differences observed in the

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metabolic analysis described previously, as well as variations in behavior during the initial stages of the investigation, suggest that these isolates differ at the strain level. This is further supported by the differences in similarity percentages observed between the two isolates and the reference strain *S. rhizophila* DSM14405^T, as shown in Table 2. Based on these findings, the isolates will henceforth be referred to as *S. rhizophila*-PM6 and *S. rhizophila*-PM7 in subsequent assays.

On the other hand, Q2 showed a similarity percentage of 98.71% with the reference strain *Lysinibacillus macroides* LMG18474^T (Table 2). Therefore, it can be stated that Q2 belongs to the genus *Lysinibacillus*, but identification at species level cannot be determined with the data obtained. Henceforth, Q2 will be referred to as *Lysinibacillus*-Q2.

Table 2. Identification of the microorganisms PM6, PM7 and Q2 from the sequencing data of the 16S rRNA gene and comparison with databases.

Sample name	Sample Accession Number	Sequence length (bp)	Closest Related Type Strain	Type strain Accession Number	Similarity to Type Strain (%)
PM6	SRR3176 2988	1335	*Stenotrophomonas rhizophila* DSM 14405	AJ293463.1	99
PM7	SRR3176 8931	1476	*Stenotrophomonas rhizophila* DSM 14405	AJ293463.1	99.41
Q2	SRR3176 8972	1587	*Lysinibacillus macroides* LMG 18474	AJ628749.1	98.71

Evaluation of the removal of Cr(VI) mediated by microorganisms

Results from the analysis of Cr(VI) removed by *S. rhizophila*-PM6, *S. rhizophila*-PM7, and *Lysinibacillus*-Q2 are shown in Fig. 5A.

The uninoculated control showed no significant changes in the Cr(VI) concentration over time. However, the presence of bacteria showed a significant decrease in the Cr(VI) concentration. *S. rhizophila*-PM6 and *S. rhizophila*-PM7 achieved a reduction of 82 and 81% in the Cr(VI) concentration respectively, while *Lysinibacillus*-Q2 could reduce 88% of Cr(VI) concentration after 48 h of incubation.

Bacterial culture in the presence of polypropylene and Cr(VI)

a. Bacterial growth in the presence of polypropylene and Cr(VI)

After the 40-day assay, the bacterial count value decreased between 1 and 1.5 orders of magnitude compared to the initial value. *S. rhizophila*-PM6 and *S. rhizophila*-PM7 started with viable cell counts close to 8 Log(CFU/ml), while after 40 days they reached slightly above 6 Log(CFU/ml). In contrast, *Lysinibacillus*-Q2 began with values close to 6 Log(CFU/ml) and ended with values around 5 Log(CFU/ml). Although the decrease in bacterial count was

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significant, the CFU/ml value indicates that the bacteria were able to remain viable under these conditions in culture systems while subjected to the two stress factors (PP as the only carbon and energy source and Cr(VI)). Results are shown in Supplementary Information 5.

b. Evaluation of the removal of Cr(VI) in the presence of polypropylene

The capacity of Cr(VI) removal of the treatments in presence of PP are represented in Fig. 5B. Results indicated that the Cr(VI) at final time (48 h) did not significantly differ from the concentration at initial time in all the treatments analyzed.

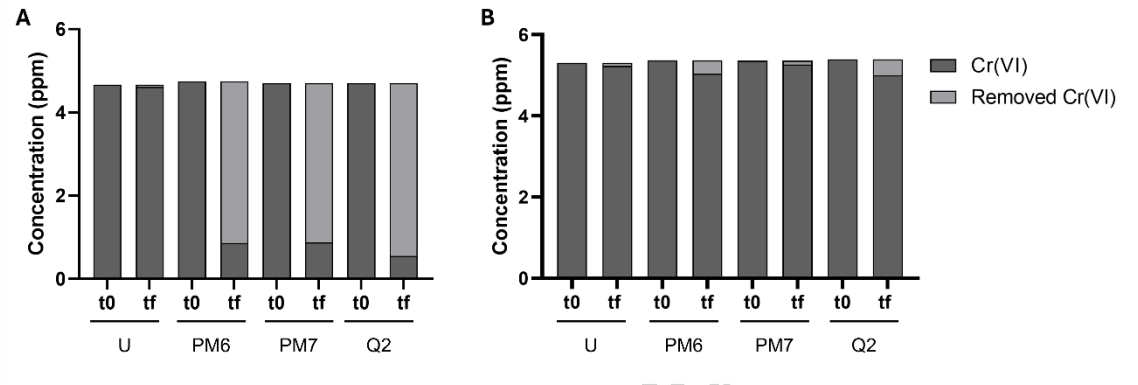

Fig. 5 Cr(VI) concentrations (ppm) at initial (t0) and final time (tf, 48 h), and Cr(VI) removed in the treatments with *S. rhizophila*-PM6 (PM6), *S. rhizophila*-PM7 (PM7), *Lysinibacillus*-Q2 (Q2) and the uninoculated control (U) in the absence (A) and presence (B) of UV-pretreated PP.

c. Quantitative analysis of the changes in the polypropylene in the presence of Cr(VI)

Table 3 indicates the weight loss percentages of the treatments after incubation with the selected microorganisms. The results showed that the UV-pretreated PP dry weight was significantly different between the initial and final time in treatments inoculated with microorganisms. Comparing the results of viable count and percentage weight loss, it was observed that the treatments inoculated with *S. rhizophila*-PM6 and *S. rhizophila*-PM7 had similar results. The average percentage of PP weight loss for *S. rhizophila*-PM6 and *S. rhizophila*-PM7 was lower compared to a previous test, while the treatment inoculated with *Lysinibacillus*-Q2 exhibited a higher value of PP weight loss in the presence of both pollutants.

Table 3. UV-pretreated PP degradation parameters of treatments inoculated with *S. rhizophila*-PM6, *S. rhizophila*-PM7, *Lysinibacillus*-Q2 and the uninoculated control in the presence of 5 ppm Cr(VI).

Treatment	Weight loss (%)	K	Average total degradation time (days)
***S. rhizophila*-PM6**	0.550 ± 0.186	1.38 E-04	5022.80
***S. rhizophila*-PM7**	0.523 ± 0.184	1.38 E-04	5022.80

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***Lysinibacillus*-Q2**	1.040 ± 0.100	2.62 E-04	2645.59
Uninoculated	0.085 ± 0.042	2.12 E-05	35695.62

A reduction in the degradation time of uninoculated plastic in the presence of Cr was observed when compared to its degradation time in the absence of the metal (Table 1).

d. Biofilm formation in the presence of 5 ppm Cr(VI)

The results regarding the biofilm formation capacity of *S. rhizophila*-PM6, *S. rhizophila*-PM7 and *Lysinibacillus*-Q2 in the presence and absence of 5 ppm Cr(VI) shown a statistically significant decrease ($p < 0.0001$) in biofilm formation for *S. rhizophila*-PM6 and *S. rhizophila*-PM7 in the presence of Cr(VI), indicating that the metal negatively impacts this type of growth. In contrast, *Lysinibacillus*-Q2 exhibited a statistically significant increase ($p < 0.0001$) in biofilm formation under the same conditions. Results are shown in Supplementary Information 6.

Discussion

The analysis of the environmental samples from Entre Ríos revealed the incidence of diverse forms of plastic present in coastal sediment. This is in agreement with the findings reported by Lacava et al. [6] for samples from Buenos Aires, and with other studies showing that plastics and microplastics are found in various ecosystems as a result of human activity [29].

It was possible to isolate morphologically distinct microorganisms from the sediment samples. The biochemical tests allowed us to discard some isolates that could be potentially pathogenic and therefore not applicable in biotechnological techniques. The capacity of biofilm formation is a crucial step for the subsequent biodegradation of plastics; thus its study allowed the selection of 5 potential plastic biodegrading microorganisms to carry out the subsequent biodegradation assays. Although it has been reported that some microorganisms have increased CFU/ml over time when using culture media with higher amounts of nutrients [30] and even presented variations in minimal media [23], the fact that the counts of the microorganisms under study in the analyzed treatments were able to remain constant is a promising result that indicates that these microorganisms would be using PP as a substrate. The potentially colonizable area by bacteria is the PP surface. Thus, bacteria growing in association with the polymeric material could explain the lack of variation in the OD values.

When analyzing the weight loss of PP in contact with the different microorganisms, it was observed that the isolates PM6 and PM7 presented the ability to use PP as the sole carbon source and the highest percentage of degradation. Although the percentage of degradation presented by Q2 was lower than PM6 and PM7, it was higher than the observed for VL5, and thus its study in subsequent trials was not discarded. The percentage of degradation observed for

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microorganisms Q2 and VL5 was not in agreement with their viable count in the presence of PP, where a higher viable count was observed in VL5 compared to Q2. However, the assays performed are not sufficient to explain these viable counts. Future analysis of protein expression and biodegradation-derived compound formation would be necessary to better understand these systems.

While heat and UV light are factors that can influence the initial degradation of plastic [31], it has been shown that these treatments generate improvements in the biodegradation process as they generate a surface more suitable for bacterial colonization and for the microbial enzymatic attack [32]. In addition, these factors can simulate the situation that plastics can go through when they are in the environment, due to exposure to sunlight, and it has been reported that many products made with plastic (especially those related to packaging), which can eventually be found as environmental contamination due to human activity, are subjected to this treatment [33]. It is well established that abiotic degradation, including UV-induced surface modifications, often precedes microbial biodegradation [32]. In this study, PP drying was performed to standardize dry weight measurements, while UV exposure was used to reduce potential microbial contamination on the pellet surface. However, it is known that UV treatment may have induced surface changes that could influence microbial attachment and degradation dynamics. While the results of this study indicate that the PP treatment ensured a uniform starting condition across all trials and did not induce degradation in the absence of inoculation, further studies would be needed to assess the specific physicochemical alterations caused by UV exposure and their potential impact on biodegradation.

The qualitative analysis of the UV-pretreated PP in terms of surface area and structure revealed that the uninoculated treatment presented a high roughness on the surface characteristic of the pellet. The images of UV-pretreated PP exposed to PM6 and PM7 showed smooth surfaces, in agreement with previous results, given that these microorganisms presented the highest percentage of degradation and count. The images obtained for Q2 and VL5 showed higher roughness compared to PM6 and PM7, but they also differed from the uninoculated control.

The observed surface smoothing in the SEM images after exposure with microorganisms may be attributed to multiple factors associated with microbial activity and enzymatic degradation. Previous studies have reported that rough surfaces facilitate initial microbial colonization by providing greater attachment points for biofilm formation [34]. In our study, the industrial plastic waste used as a substrate presented an irregular rough surface, which may have promoted early-stage microbial adhesion and activity. Furthermore, Priya et al. [35] demonstrated that an increased surface area enhances biodegradability, suggesting that the initial roughness of the plastic may have accelerated the degradation process. As biodegradation progresses, microbial enzymatic activity can alter the polymer structure, leading to surface modifications. After 40 days of incubation, a noticeable smoothing of the PP surface was observed in the inoculated treatments,

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in contrast with the initially rough morphology of the material. This phenomenon suggests a surface-level alteration potentially mediated by microbial activity. Although PP is a chemically inert polymer lacking hydrolysable ester bonds, and the tested strains were not characterized for specific enzymatic activities, biofilm formation and secretion of extracellular compounds could have contributed to the observed changes. Previous studies have shown that microbial colonization and biofilm formation on polyolefin surfaces may induce physicochemical alterations such as increased oxygen-containing functional groups and changes in hydrophobicity, even without direct enzymatic cleavage of the polymer backbone [24, 36]. Additionally, while enzymatic profiles of the isolates used in this study were not determined, previous research suggests that related strains of *Stenotrophomonas* and *Lysinibacillus* can produce oxidative enzymes potentially involved in surface modification processes. *Stenotrophomonas* sp. CFB-09 has been reported to produce extracellular laccases in agro-residues [37] and dioxygenases involved in aromatic compound degradation [38]. Likewise, *L. macroides* has demonstrated extracellular laccase production using bio-waste [39]. Although it remains to be determined whether *S. rhizophila* or the *Lysinibacillus* strain used in this study possess similar enzymatic capabilities, these findings support the plausibility of microbial enzymatic activity contributing to the surface smoothing observed.

The spectral changes observed in the FTIR analysis suggest that the tested microorganisms induced chemical modifications in the polypropylene matrix. The observed variations in peak intensities, particularly in regions associated with C-O and C-H bond vibrations, are consistent with structural alterations that may result from oxidation processes and polymer chain scission [40, 41]. These modifications support the hypothesis that microbial activity contributes to the degradation of PP by altering specific functional groups. The variation of absorption bands in the treated samples further supports the involvement of biochemical interactions between microbial metabolites and the polymer structure.

On the other hand, considering that the purpose of the present work was to analyze the biodegradation of PP in the presence of Cr, the survival of the isolates (PM6, PM7, Q2, VL5 and 34B) in the presence of Cr was assessed simultaneously. The results obtained showed that the growth of PM6, PM7 and Q2 was not affected by the metal concentrations used, while 34B and VL5 were significantly affected, with the response of VL5 being variable as a function of concentration.

All the results obtained regarding colonization and biodegradation of PP and Cr tolerance led to the conclusion that the most promising isolates were PM6, PM7 and Q2. Microorganism 34B was excluded from further trials because, despite being a good biofilm-forming microorganism, it was unable to survive using PP as the sole substrate and could not grow at high Cr concentrations. VL5 was also dismissed as it presented inconsistent results showing a great variability.

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The identification of the promising microorganisms showed that two of them were *Stenotrophomonas rhizophila*, while the other belonged to the *Lysinibacillus* genus, not being able to be identified to species level. Although there are some species of the genus *Stenotrophomonas* identified as pathogens in humans, comparative genomics studies have shown that *S. rhizophila* differ genetically from those species of the same genus [42], not being considered as a pathogenic microorganism. Both *Lysinibacillus* and *Stenotrophomonas* genus have been found to possess properties that make them suitable for applications in heavy metal bioremediation processes, particularly, in Cr(VI) reduction [14, 43]. There are also reports of biodegradation mediated by the bacterial genus identified in this work [30, 44]. Particularly, *Stenotrophomonas maltophilia* was found to degrade polyethylene by using it as a carbon source after degrading the polymers to simpler compounds [11]. Also, there is a report that indicated that bacteria from the genus *Lysinibacillus* sp. could degrade PP microplastics by the formation of a biofilm on the particles of the microplastics and by metabolic pathways which include enzymes like oxygenases, lipases or esterases [45]. A proteomic analysis could be useful in future studies to help establishing strain-specific degrading mechanisms.

In relation to the removal of Cr(VI) mediated by microorganisms, there are two possible responses of microorganisms to Cr exposure. The first possible response is that microorganisms invest a large amount of energy in actively transporting Cr into their cells, which slows down cell division but maintains a viable population. The other potential response is that microorganisms utilize energy for metabolic processes that increase the microbial population and directly contribute to the reduction of Cr(VI), leading to an increased rate of metal removal. The results obtained indicated that *S. rhizophila*-PM6, *S. rhizophila*-PM7 and *Lysinibacillus*-Q2 were not only able to enhance their population while exposed to increasing concentrations of Cr but were also involved in the absorption/adsorption of Cr or in the reduction of metal's toxicity to its environmentally safe form. Therefore, the presence of the heavy metal did not negatively impact cell division, as there was no significant decrease in average change in OD with 5 ppm Cr(VI) compared to the control cultures without the metal. This phenomenon was reported for bacteria of the genus *Lysinibacillus* which presented a high tolerance to Cr(VI), being able to grow in the presence of 250 ppm [46]. Moreover, in our study, the biomass produced by the bacteria successfully removed Cr(VI), either by absorbing or adsorbing the metal or by converting it to Cr(III). Based on the data obtained, it can be interpreted that the 3 bacteria had a high Cr(VI) detoxification capacity under the analyzed conditions. *Lysinibacillus* has been reported to exhibit high tolerance to metals. Specifically, in the case of Cr, *Lysinibacillus* HST-98 has been found to harbor genes related to Cr metabolism, including those encoding the chromate reductase enzyme, a chromate transporter protein, and other related genes [46]. This study successfully elucidates the metabolic steps involved in Cr processing by this bacterial genus. The process begins with the binding of Cr(VI) to proteins located on the cell surface, followed by the internalization of

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chromate ions, their reduction from Cr(VI) to Cr(III) within the cell, and the subsequent expulsion of Cr(III) from the cell. In the case of *S. rhizophila*, it has been reported that this species possesses several metal transporter proteins in its membrane, which are associated with its metal tolerance [47]. Gao et al. [43] investigated the reduction of Cr(VI) to Cr(III) by *S. rhizophila* DSM14405^T, demonstrating that this microorganism is capable of reducing Cr(VI) and accumulating Cr(III) intracellularly. Gene expression analysis in the presence of Cr revealed the upregulation of genes associated with Cr resistance, DNA repair, oxidative stress response, and central metabolism, among others. In addition, *S. maltophilia* has been demonstrated to reduce Cr(VI) to Cr(III). A protein similar to chromate reductases from other microorganisms has been identified in this species, and genomic analyses revealed the presence of the *chrR* gene, which is associated with this protein and plays a key role in metal reduction [48]. Considering all these, although the concentration of Cr(III) and the residual Cr in the bacterial biomass were not measured in this study, the microorganisms under investigation appear to be capable of removing Cr(VI) through uptake and subsequent reduction to Cr(III).

Finally, the study of treatments with both contaminants (PP and 5 ppm Cr(VI)) revealed that *S. rhizophila*-PM6, *S. rhizophila*-PM7 and *Lysinibacillus*-Q2 were able to survive obtaining high CFU/ml counts at 40 days post-inoculation, but that the conversion of Cr(VI) to Cr(III) was lower than that obtained when the microorganisms were grown in LB medium. In this context, it is important to consider that the growth conditions in these assays were different. While LB is a rich medium, the assay involving the two contaminants exposes the microorganism to two stress conditions: in addition to the presence of the heavy metal, microorganisms are growing in a minimal medium with no other carbon and energy source than PP. Consequently, its metabolism is likely focused primarily on survival under these conditions rather than on Cr reduction. Moreover, in the presence of the two stress factors, the biodegradation of PP by *S. rhizophila*-PM6 and *S. rhizophila*-PM7 was lower than in the absence of the heavy metal. This may be related to the decreased ability to grow as a biofilm in the presence of the metal, since, as mentioned earlier, surface colonization is a crucial step in the biodegradation process. It has been reported that some microorganisms reduce their ability to form biofilms in the presence of metals, due to the metal-induced stress, which alters metabolic pathways to enhance DNA repair mechanisms and oxidative stress responses [49]. On the contrary, *Lysinibacillus*-Q2 exhibited an increased tendency to biodegrade PP in the presence of Cr(VI) and higher metal reduction in the presence of both pollutants, being the one that best responded to a simultaneous bioremediation system. This result is consistent with the fact that this microorganism significantly increased biofilm formation in the presence of 5 ppm Cr(VI). Chien et al. [50] have studied the biofilm formation of *Pseudomonas* sp., finding that in the presence of the metal, there is an increase in exopolysaccharide production, which promotes this type of growth. It has also been reported that mutant strains of *Sinorhizobium meliloti* incapable of producing exopolysaccharides lost their

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ability to form biofilms in the presence of heavy metals, linking these two phenomena under the studied conditions [51]. Taking all this into account, it can be stated that the influence of the metal on biofilm formation capacity varies among different microorganisms. Therefore, studies on gene expression or exopolysaccharide production would be necessary to elucidate the interaction between the presence of the metal and this type of growth in each case.

When analyzing the degradation time of the PP, it was observed that the addition of the metal influenced this process in uninoculated treatments. Previous studies have reported that metals can accelerate the plastic degradation process through various mechanisms, depending on the type of polymer and metal involved. For instance, metals can catalyze the decomposition of hydroperoxides or interact directly with the plastic surface to generate free radicals, among other potential processes [52]. The PP degradation rate of *S. rhizophila*-PM6 and *S. rhizophila*-PM7 was slower in cultures containing both PP and Cr(VI) compared to the ones with only PP, results that are consistent with those previously described. Contrarily, *Lysinibacillus*-Q2 revealed a halving of the bioremediation time of the polymer under the evaluated conditions. This observation is in agreement with the previous results, and with reported results, where some microorganisms in the presence of heavy metals produce a greater amount of exopolysaccharides to protect themselves from its toxicity. Indeed, it has been reported that biofilm formation enhanced plastic biodegradation [53], a finding that aligns with what was observed for *Lysinibacillus*-Q2.

In further analysis, it is aimed to evaluate bacterial behavior under less stressful conditions and compare the results with and without the addition of nutrients, in order to adapt the culture conditions and eventually optimize the bioremediation processes for the degradation of plastics and the reduction of heavy metals.

The results in this study demonstrated the need for further research on the complete degradation of PP over an extended period of time to define the relationship between biodegradability and microbial activity.

Conclusions

This study focused on isolating bacteria from contaminated environmental samples to assess their ability to biodegrade PP and reduce Cr, both of which are contaminants found in the evaluated polluted sites. The samples exhibited a diverse range of microorganisms, which were characterized based on their morphology and metabolism. Biofilm formation was found to be an effective criterion for selecting microorganisms for further testing. Whereas bacteria isolated from Entre Ríos did not present an efficient UV-pretreated PP degradation and growth in the presence of Cr, three bacteria isolated from Buenos Aires samples (isolates PM6, PM7, and Q2) showed promising results in degrading UV-pretreated PP and removing Cr(VI), probably by the reduction to the less toxic Cr(III) form. PM6 and PM7 (both *Stenotrophomonas rhizophila*) were the

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microorganisms that showed the highest PP biodegradation and survival with plastic as the sole carbon source, while Q2 (*Lysinibacillus* sp.) was able to biodegrade more efficiently PP in the presence of 5 ppm Cr(VI). This could be attributed to the microorganism's enhanced biofilm formation in the presence of the metal, which represents a critical initial step in the biodegradation process. This study suggests that the isolated bacteria could potentially be used in future bioremediation processes to treat environments contaminated with PP and Cr, either alone or in combination. Thus, the design of bioremediation strategies now might make use of these environmentally friendly alternatives for the treatment of wastes generated as a consequence of anthropogenic activities.

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Statements & Declarations

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Competing Interests

The authors have no relevant financial or non-financial interests to disclose.

Author contributions

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by María Azul Denaro, Melisa Olivelli and Pamela Romina Bernabeu. The first draft of the manuscript was written by Pamela Romina Bernabeu and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Data availability

The datasets generated and analyzed during the current study are available from the corresponding author on reasonable request.

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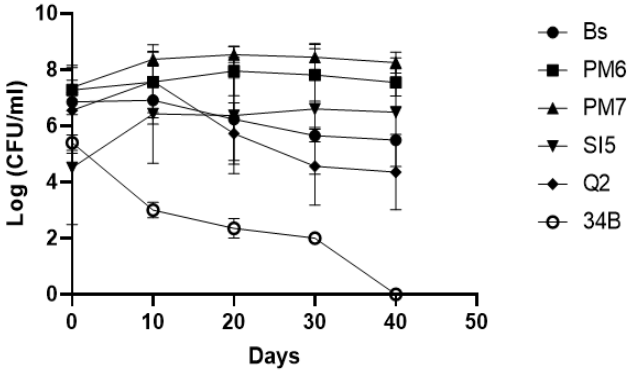

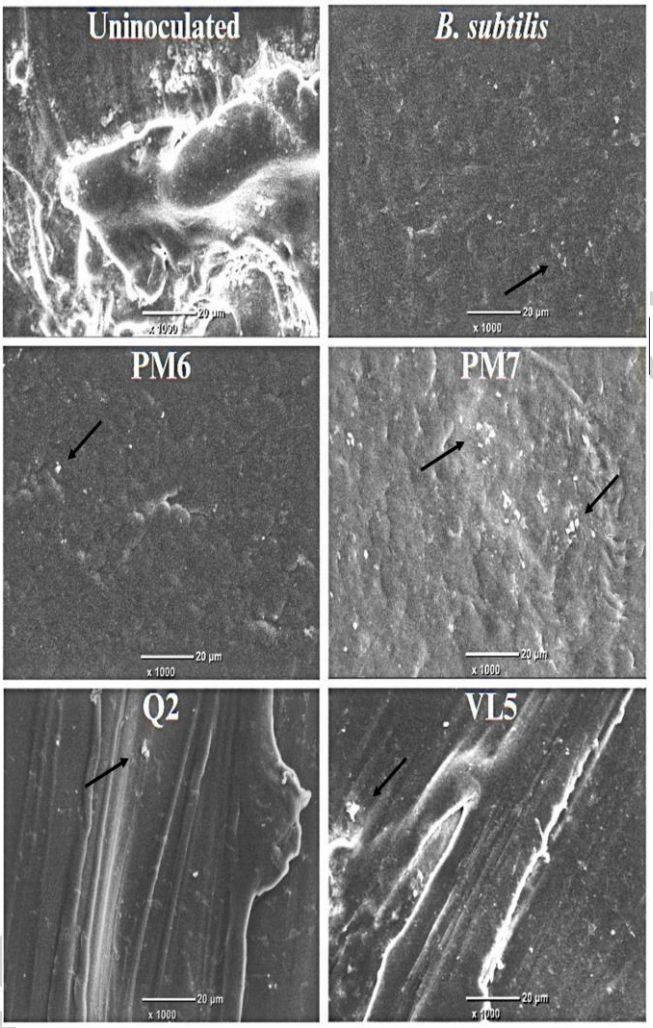

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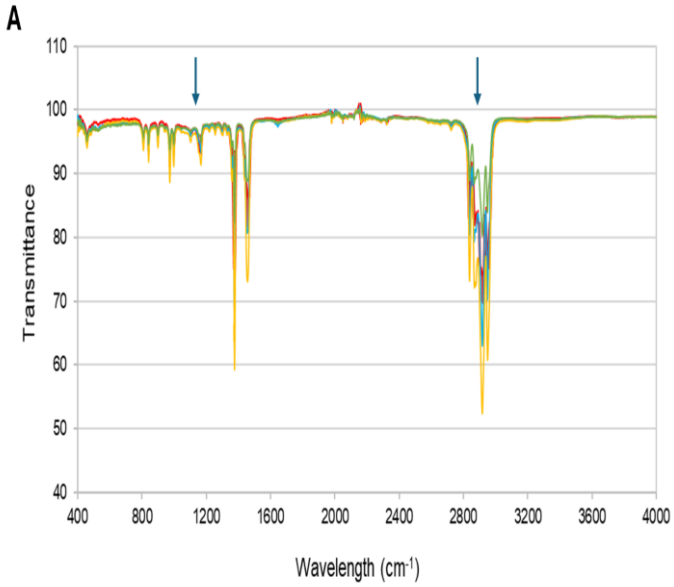

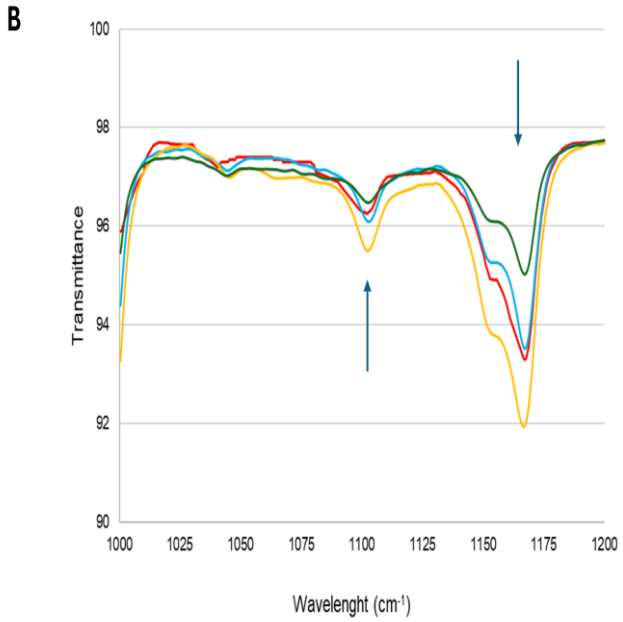

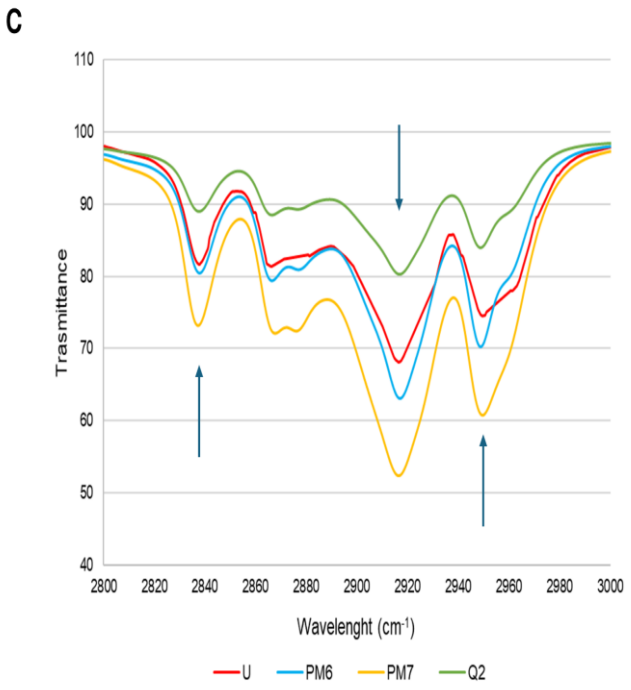

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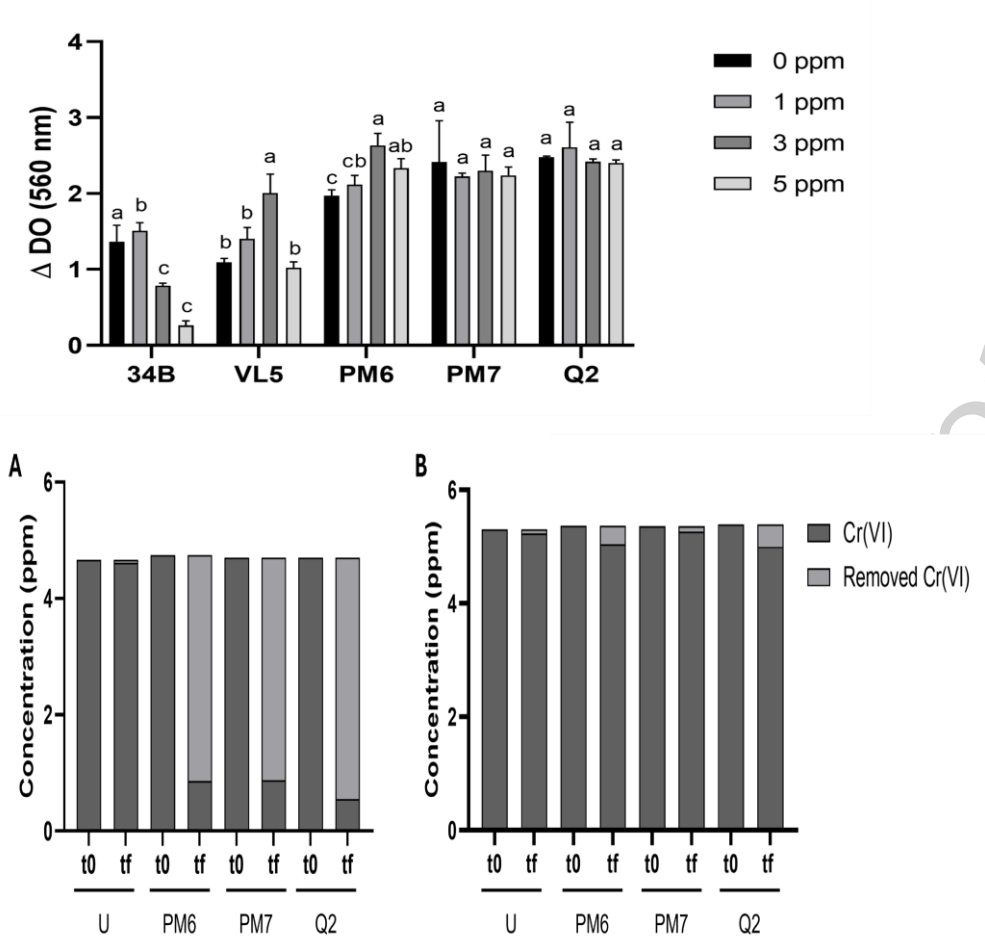

Table 1. PP degradation parameters of PM6, PM7, Q2 and VL5, and of the positive (*B. subtilis*) and uninoculated controls.

Treatment	Weight loss (%)	K	Average total degradation time (days)
B. subtilis	1.120 ± 0.870	2.83 E-04	2449.28
PM6	2.015 ± 1.000	5.10 E-04	1359.11
PM7	1.680 ± 0.810	4.25 E-04	1630.93
Q2	0.540 ± 0.220	1.35 E-04	5134.42
VL5	0.080 ± 0.040	1.99 E-05	34831.51
Uninoculated	0.013 ± 0.023	3.33 E-06	208152.3

Table 2. Identification of the microorganisms PM6, PM7 and Q2 from the partial sequencing data of the 16S rRNA gene and comparison with databases.

Sample name	Sample Accession Number	Sequence length (bp)	Closest Related Type Strain	Type strain Accession Number	Similarity to Type Strain (%)

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PM6	SRR3176 2988	1335	*Stenotrophomonas rhizophila* DSM 14405	AJ293463.1	99
PM7	SRR3176 8931	1476	*Stenotrophomonas rhizophila* DSM 14405	AJ293463.1	99.41
Q2	SRR3176 8972	1587	*Lysinibacillus macroides* LMG 18474	AJ628749.1	98.71

Table 3 PP degradation parameters of treatments inoculated with *S. rhizophila*-PM6, *S. rhizophila*-PM7, *Lysinibacillus*-Q2 and the uninoculated control in the presence of 5 ppm Cr(VI).

Treatment	Weight loss (%)	K	Average total degradation time (days)
***S. rhizophila*-PM6**	0.550 ± 0.186	1.38 E-04	5022.80
***S. rhizophila*-PM7**	0.523 ± 0.184	1.38 E-04	5022.80
***Lysinibacillus*-Q2**	1.040 ± 0.100	2.62 E-04	2645.59
Uninoculated	0.085 ± 0.042	2.12 E-05	35695.62

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