

# Effect of TiO<sub>2</sub> nanoparticles and extrusion process on the physicochemical properties of biodegradable and active cassava starch nanocomposites

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## 1. Antibacterial activity

Antibacterial effect was evaluated following the agar diffusion method in Muller-Hinton agar [1]. Briefly, *Escherichia coli* DH5 $\alpha$  (*E. coli*) cultivated in LB medium at 37°C for 24 h were diluted to a final concentration of 10<sup>8</sup> CFU/mL. Then, 200  $\mu$ L of the resulting bacterial suspension were evenly inoculated on agar plates containing Muller-Hinton agar. Circular samples of 2 cm diameter of each material were sterilized under UV radiation for 1 h and placed in the inoculated agar. Photographic records of the agar plates were taken after 24 h incubation at 37 °C.

Figure S1 shows the plates containing bacteria and the S80, S80-TiO<sub>2</sub>NP and S120-TiO<sub>2</sub>NP materials after 24 h incubation at 37°C. As can be seen in the picture, no inhibition halo was observed in any of the studied films. Similar results were reported for nanocomposites containing non diffusive antimicrobial nanoparticles in textiles [2] and in biodegradable polymers [3]. Other authors reported antimicrobial activity of TiO<sub>2</sub> nanoparticles incorporated inside a polymer matrix [4–6], however, in these researches higher TiO<sub>2</sub> concentrations were employed. Besides, in those works, the films were illuminated with UV light. In the presence of UV light, TiO<sub>2</sub> generates highly oxidative reactive species that could kill bacteria or inhibit their growth [7]. Taking into consideration that the final disposition after usage of the films present in this study is composting, bacterial activity should not be examined under UV radiation, since the films would not be exposed to light. As a low concentration of TiO<sub>2</sub> was employed and samples were not illuminated under UV light, it was expected that the materials didn't present antimicrobial activity.

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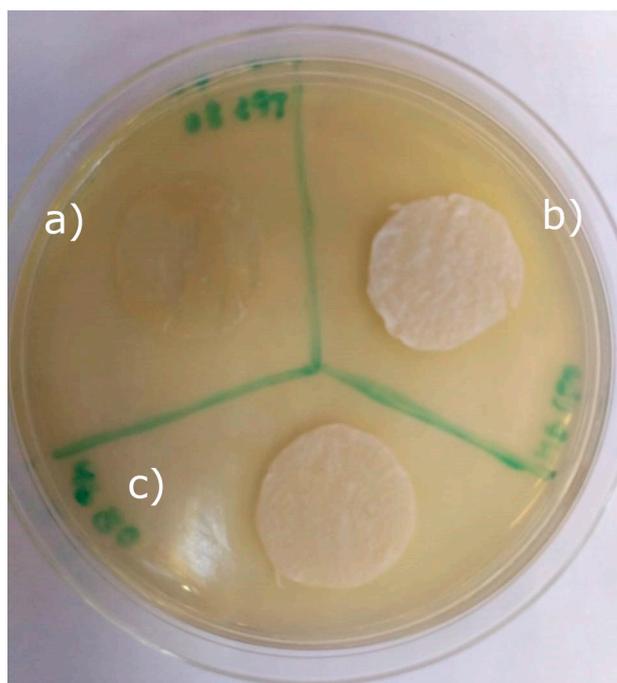
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**Figure S1:** Antibacterial activity of (a)  $S_{120}$ , (b)  $S_{120}$ -TiO<sub>2</sub>NP and (c)  $S_{80}$ -TiO<sub>2</sub>NP against *E. coli* studied by the agar diffusion method.

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