

Growth inhibition of plant-pathogen bacteria by extracts from the therapeutic plant *Achillea millefolium* (Mil Folhas)

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Mil folhas (*Achillea millefolium* L.) is a therapeutic plant that presents anti-inflammatory and antimicrobial properties, and proteins and peptides could be involved in these antimicrobial mechanism(s). This work aims to determine the *in vitro* antimicrobial potential of proteic extracts of *mil folhas* leaves. Dried leaves were ground, macerated (Tris added of protease inhibitors), centrifuged and the supernatant named Soluble Extract (SE). The precipitate was extracted with LiCl, centrifuged and the supernatant was named Cell-Wall Extract (CWE). SE and CWE were separately submitted to ultra-filtration (30, 10, and 1kDa), resulting in three fractions with different MM ranges: superior to 30kDa (SE>30, CWE>30), 10-30kDa (SE10-30, CWE10-30), and 1-10kDa (SE1-10, CWE1-10). The six samples were fractionated with (NH₄)₂SO₄ (35-75% sat.), centrifuged and the precipitate desalted and concentrated. The antibacterial action of the fractions was evaluated in three concentrations against plant-pathogen bacteria. After 16h of cultivation, *Ralstonia solanacearum* growth inhibition by SE1-10 was around 37%, by SE10-30, 25%, and by SE>30, total inhibition. For *Clavibacter michiganensis* subsp. *michiganensis*, inhibition by SE1-10 was 32%; SE10-30, 32%; and SE>30, 62%. CWE1-10 was fractionated by DEAE Sepharose, the peaks were concentrated, desalted, and evaluated against five plant-pathogenic bacteria. After 12 hours of cultivation, the recovered cationic peak-1 promoted total growth inhibition of *R.solanacearum*. The anionic peak-7 inhibited *Erwinia carotovora* subsp. *carotovora* growth around 20%, and *Pseudomonas syringae* pv *tomato*, 25%. The identification of proteins and antimicrobial peptides may be a biotechnological strategy for the protection of crops of commercial importance.

Keywords: antimicrobial peptides, *Mil folhas*, therapeutic plant
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