

ABSTRACT - PLANT PHYSIOLOGY

EVALUATION OF SAPONIN EXTRACTION PROCEDURES FOR QUILLAJA BRASILIENSIS CELL CULTURES

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Saponins from leaves of *Quillaja brasiliensis* (currently *Q. lancifolia*) show pronounced immunostimulatory activity in experimental vaccines, comparable to that of the commercial product Quil-A®. We have recently established plant cell cultures of *Q. brasiliensis* as a potential source of bioactive saponins. Typical *Quillaja* saponins were detected in these cultures by thin layer chromatography and LC-MS 2, which also displayed immunomodulatory action. Fast, reproducible and low cost saponin extraction methods are essential to achieve higher yields of metabolites and maximize the use of cell biomass. In order to address this problem, freeze-dried cell cultures were extracted in water at 1:100g/mL using different procedures: stirring, stirring + sonication, stirring + partitioning with ethyl acetate at 1:6 ratio, and microwave heating for 90s. After extract lyophilization, yield of total saponins was spectrophotometrically determined using the vanillin-sulfuric acid test and standard curves of genuine immunoadjuvant saponin fraction QB-90 purified from *Q. brasiliensis* leaves. Higher saponin amounts were obtained with microwave extraction and partitioning with ethyl acetate. Analyses of detailed composition and bioactivity

of the extracted saponins, along with extraction experiments with leaves and barks of cultivated ministumps of *Q. brasiliensis*, are ongoing.