### UNIVERSITI TEKNOLOGI MARA

IDENTIFICATION and CHARACTERIZATION of EXOPOLYSACCHARIDE PRODUCED by Bifidobacterium pseudocatenulatum ATCC 27919 CULTIVATED in RICE WATER MEDIUM AGAINST COLON CANCER CELL

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**MSc** 

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### **AUTHOR'S DECLARATION**

I declare that the work in this thesis was carried out in accordance with the regulations of Universiti Teknologi MARA. It is original and is the results of my own work, unless otherwise indicated or acknowledged as referenced work. This thesis has not been submitted to any other academic institution or non-academic institution for any degree or qualification.

I, hereby, acknowledge that I have been supplied with the Academic Rules and Regulations for Postgraduate, Universiti Teknologi MARA, regulating the conduct of my study and research.

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#### **ABSTRACT**

Exopolysaccharides (EPS) produced by probiotics have generated increasing attention among researchers due to its beneficial effect as anti-cancer property. However, it has been hypothesized that different cultivation medium used during probiotic cultivation resulted in different carbon profiles of exopolysaccharides to leading to anti-cancer activity. Thus, in this study, rice water was used to cultivate Bifidobacteria pseudocatenulatum ATCC 27919 and commercial media was used as comparative medium. The objectives of this study were 1) To determine the cytotoxicity effect against Caco-2 cell line using EPS cultivated in rice water and aloe vera medium using MTS assay. 2) To identify the monosaccharide of EPS using thin-layer chromatography (TLC) cultivated in the selected medium 3) To characterize the structure of EPS using flourier-transform infrared (FT-IR), nuclear magnetic resonance (NMR) spectroscopy techniques and CHNS elemental analysis. Gel filtration method was used for EPS purification. Cytotoxicity experiments showed the percentage of the cell viability of EPS from rice water medium (52%) remarkably close to EPS from commercial medium (MRS) (38%) after 40 hrs of treatment. CHNS analysis was carried out to calculate the composition of carbon, hydrogen, nitrogen, sulphur. NMR analysis was done by dissolving EPS in D<sub>2</sub>O and conducted the entire 1D and 2D NMR procedure at 47 °C. Based on the monomer identifications using TLC, glucose was found to be the main repeating unit in this compound. FTIR, CHNS, and NMR analysis were proved the presence of the one monomer. Based on this study, the arrangement of the glucose repeating unit glycosidic linkage could be  $\alpha$ -(1 $\rightarrow$ 4) Glcp is the backbone of the repeating unit, and  $\alpha$ -(1 $\rightarrow$ 6) glycosidic link makes the branch for this structure. Cell viability testing performed in both rice water and commercial media did not show a significant difference (p>0.05) due to the same repeating unit structure for both EPS samples. Hence, this study manifests that EPS from rice water have the potential to be used in clone cancer therapy.

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