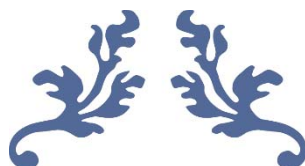


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**Bioprospecting: The quest for novel extracellular
polymers produced by soil-borne bacteria**



A thesis presented in partial fulfilment of the

requirements for the degree of

Master of Science

In

Microbiology

at Massey University, Palmerston North,

New Zealand

Jason Smith

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Dedication

This thesis is dedicated to my dad.

Vaughan Peter Francis Smith

13 July 1955 – 27 April 2002

Though our time together was short
you are never far from my mind nor my heart.

Abstract

Bacteria are ubiquitous in nature, and the surrounding environment. Bacterially produced extracellular polymers, and proteins are of particular value in the fields of medicine, food, science, and industry. Soil is an extremely rich source of bacteria with over 100 million per gram of soil, many of which produce extracellular polymers. Approximately 90% of soil-borne bacteria are yet to be cultured and classified. Here we employed an exploratory approach and culture based method for the isolation of soil-borne bacteria, and assessed their capability for extracellular polymer production. Bacteria that produced mucoid (of a mucous nature) colonies were selected for identification, imaging, and polymer production. Here we characterised three bacterial isolates that produced extracellular polymers, with a focus on one isolate that formed potentially novel proteinaceous cell surface appendages. These appendages have an unknown function, however, I suggest they may be important for bacterial communication, signalling, and nutrient transfer. They may also serve to increase the bacteria's surface area for nutrient adsorption without compromising structural integrity of the cell. The results from this study contribute to the scientific body of knowledge and provide avenues for further research into bacterial appendage formation.

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List of abbreviations

A full list of abbreviations used

°C	Degrees Celsius
AIA	Actinomycete isolation agar
AIB	Actinomycete isolation broth
APS	Ammonium persulfate
BCA	Bicinchoninic acid
BLAST	Basic local alignment search tool
BSA	Bovine serum albumin
bp(s)	Base pair(s)
DMSO	Dimethylsulfoxide
DNA	Deoxyribonucleic acid
DNase	Deoxyribonuclease
dNTPs	Deoxyribonucleotide triphosphates
EDTA	Ethylenediaminetetraacetic acid
EtOH	Ethanol
g	Gravity/gram
<i>gyrA</i>	DNA gyrase subunit A
k	Kilo/thousand
kb	Kilo base(s)
kDa	Kilo Daltons
LB	Luria-Bertani broth
LBA	Luria-Bertani agar
MLST	Multilocus sequence typing

OMF	Outer membrane fraction
<i>parC</i>	DNA topoisomerase IV subunit A
PCR	Polymerase chain reaction
Psi	Pounds per square inch
RNA	Ribonucleic acid
RNase	Ribonuclease
rRNA	Ribosomal ribonucleic acid
<i>rpoB</i>	DNA-directed RNA polymerase β -subunit
rpm	Revolutions per minute
SDS	Sodium dodecyl sulphate
SDS-PAGE	Sodium dodecyl sulphate polyacrylamide gel electrophoresis
SEM	Scanning Electron Microscopy
TBE	Tris-Borate-EDTA buffer
TEM	Transmission Electron Microscopy
TEMED	Tetramethylethylenediamine
Tris	Trishydroxymethylaminomethane
v/v	Volume per volume
WEF	Whole envelope fraction
w/v	Weight per volume

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