

**Characterisation and antimicrobial activity of *Pediococcus* species
isolated from South African cheese**

By

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Declaration

I declare that this dissertation that I hereby submit for the degree of MSc at the University of Pretoria is my own work and has not been previously been submitted by me for a degree at any other University or institution of higher education.

Dedication

This dissertation is dedicated to my brother Gilbert Shamiso Gurira, my grandmother Mrs Grace Mukundidza and my parents Mr and Mrs Gurira.

Acknowledgement

Dear God, *I would like to thank you for bringing me to this stage in my life. You have upheld me with your righteous right hand and lead me through the path of my educational career. You gave me strength when I was weak, courage when I was despondent and to this day I would wish to thank you. Amen.*

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Abstract

Characterisation and antimicrobial activity of *Pediococcus* species isolated from South African cheese.

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Pediococci are Gram-positive, non-spore forming lactic acid bacteria, LAB, that are commonly used in the industrial fermentation of vegetables and meat. On the other hand species of *pediococci* may constitute part of the secondary microflora as non-starter lactic acid bacteria, NSLAB, responsible for cheese ripening. This study was conducted to determine and isolate *pediococci* from South African farm-style cheese and to morphologically and physiologically characterise the *pediococci* isolates to species level. Strains of *Pediococcus* species isolated from South African farm-style cheese were evaluated for the production of antimicrobial peptides, pediocins, and their effect against food pathogens *Bacillus cereus* ATCC 1178 and *Listeria monocytogenes* ATCC 7644.

Eight farm-style cheeses namely pasteurised young Gouda (PYG), pasteurised matured Gouda (PMG) as well as pasteurised matured Parmesan (PMP); unpasteurised or raw milk aged Bouquet (RAB), Barbond (RMB), aged Gouda (RAG) and matured Gouda (RMG) as well as goat cheese Gouda (RGG) were chosen for the isolation of *Pediococcus* species. LAB were cultivated on MRS agar where logarithmic counts of LAB ranged from 6.90 cfu/g to 9.40 cfu/g. Microscopic examination of selected colonies identified 110 (18 %) of 606 isolates as Gram-positive, catalase negative presumptive *pediococci* occurring in pairs, clusters and tetrads. Presumptive *pediococci* were distributed among five of the eight cheeses namely PYG, PMG, RAB, RAG and RMG in numbers of 33, 21, 28, 12 and 16 respectively. Physiological characterisation of presumptive *pediococci* isolates was determined under specific growth parameters, temperature, pH and salt (w/v). These isolates were characterised as *P. acidilactici*, forty-nine, and *P. pentosaceus*, sixty-one, isolates.

For the antimicrobial assay three techniques namely, the agar disc, spot and overlay methods were evaluated. Comparisons on the sensitivity or susceptibility of two *Lactococcus* strains, *L. diacetylactis* NCDO 176 and *L. lactis* NCDO 605, to crude extract from *P. acidilactici* ST1 was assessed. The agar disc assay technique produced more reliable results compared to the other techniques and *L. diacetylactis* NCDO 176 was more susceptible to crude pediocin extract produced from *P. acidilactici* ST1. A total of fifty-two strains (47 %) from both species, twenty-seven (24 %) *P. acidilactici* and twenty-five (23 %) *P. pentosaceus*, exerted antagonism against *L. diacetylactis* NCDO 176 through the action of pediocins. Among these strains thirteen (13 %) of both species, seven (6%) *P. acidilactici* and six (7 %,) *P. pentosaceus* inhibited *B. cereus* ATCC 1178 while a total of thirty-seven strains (33 %) of both species where, seventeen (15 %) *P. acidilactici* and twenty (18 %) *P. pentosaceus* showed inhibition against *L. monocytogenes* ATCC 7644. Among these strains inhibition of food pathogens was variable against *L. monocytogenes* ATCC 7644 and low against *B. cereus* ATCC 1178. Comparison of the antimicrobial activity of the two *Pediococcus* species showed similarity in the inhibition pattern, however, more strains of *P. pentosaceus* exerted antagonism against *L. lactis* NCDO 176 and *L. monocytogenes* ATCC 7644 compared to *P. acidilactici*.

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