

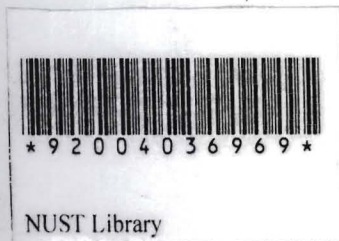


**NATIONAL UNIVERSITY OF SCIENCE AND TECHNOLOGY**

**Faculty of Applied Sciences**

**Department of Applied Biology and Biochemistry**

**Characterisation of *Fasciola gigantica* isolates from cattle from south-western  
Zimbabwe and Botswana using RAPD-PCR and ARDRA**



**EPHRAIM CHAUKE**

**STUDENT NUMBER: N0114399X**

LIBRARY NATIONAL UNIVERSITY OF SCIENCE AND TECHNOLOGY P.O. BOX 216 BULAWAYO ZIMBABWE		
DATE	ACCESSION	CLASS No.
24/10/14	14/593	

**A thesis submitted in partial fulfilment of the requirements for a Master of Science degree in Applied**

**Microbiology and Biotechnology**

**2013**

## Abstract

The study sought to characterize *Fasciola gigantica* isolates from cattle in different localities using ARDRA and RAPD-PCR. Adult flukes morphologically identified as *F. gigantica* were collected from slaughtered infected animals during meat hygiene inspections. DNA was extracted from single flukes (the conical anterior end of the worms) and subjected to ARDRA and RAPD-PCR analysis. Internal Transcribed Spacers (ITS) of nuclear ribosomal DNA (rDNA) were amplified and restriction analysis was done using the enzymes *AluI* and *HaeIII*. *Fasciola gigantica* ITS region was amplified successfully for all samples and a band of 1000bp was observed in all cases. All the isolates used in this study did not show any genetic variations in rDNA-ITS as all the isolates produced the same pattern after ARDRA. In the RAPD-PCR analysis, DNA templates were amplified by the polymerase chain reaction, using 10 random oligonucleotide primers. Depending upon the *Fasciola gigantica* isolate-primer combination, 1-13 DNA fragments in the range of 75-2000bp were amplified. It was observed that all the 10 oligonucleotide primers directing amplification of DNA were of potential interest in the generation of polymorphic DNA. The percentage polymorphic loci ranged from 33.33-100%. Polymorphic bands were scored and used to calculate Nei's 1978 genetic identity and genetic distance. Genetic identity values ranged between 0.5429, isolate 6 from Gwanda and isolate 13 from Matopo and 0.9333, isolate 5 and 6 from Gwanda. Genetic distance values ranged between 0.0690, isolate 5 and 6 from Gwanda and 0.6109, isolate 6 from Gwanda and isolate 13 from Matopo. The mean Nei's (1973) gene diversity was 0.2839. The study showed the variability of *Fasciola gigantica* isolates, using RAPD markers. No Polymorphism was seen in the *Fasciola gigantica* isolates after ARDRA, indicating the highly conserved nature of this region.