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GROWTH HORMONE GENE POLYMORPHISM IN BOER GOAT

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BY

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CERTIFICATION

This project report entitled "Growth hormone gene polymorphism in Boer goat" is prepared by Magamaee A/P Malaswamy and submitted to the Faculty of Agriculture in fulfillment of the requirement of the course SHW 4999 (Final Year Project) for the award of the degree of Bachelor of Agriculture (Animal Science).

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LIST OF ABBREVIATIONS

µL	Microliter
µM	Micromole
bp	Base pair
℃	Degree Celsius
DNA	Deoxyribonucleic acid
EDTA	ethylenediaminetetraacetate
g	Gram
MADRI	Malaysian Agricultural Research and Development Institute
$MgCl_2$	Magnesium chloride
mL	Milliliter
mM	Millimole
NCBI	National Center for Biotechnology Information
ng	Nanogram
nm	Nanomole
PCR	Polymerase chain reaction
TBE	Tris borate EDTA
V	Voltage

ABSTRACT

GROWTH HORMONE GENE POLYMORPHISM IN BOER GOAT

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SUPERVISOR: PROF. DR. JOTHI MALAR PANANDAM

In meat goat industry, growth traits of goat are of primary concern during breeding due to its economic determinant value. Molecular genetic information may be applied with quantitative genetics approach to select parental stocks of high merit to be used in breeding programmers to achieve rapid genetic improvement of herds. The growth hormone (GH) gene plays an important role in the regulation of growth and enhancement of amino acid incorporation into muscle protein. Polymorphism of the GH gene and its association with birth chest girth and weaning weight has been reported. The objective of this study was to investigate the GH gene polymorphism in a Boer goat herd in Malaysia using restriction fragment length polymorphism (RFLP). DNA extracted from blood samples of 30 randomly selected Boer goats were amplified for a partial region of the GH gene using specific primers. The polymerase chain reaction (PCR) products were electrophoretically separated in 2% agarose gel for confirmation of amplification success. The PCR amplicons were then digested using the *Hae*III restriction enzyme. The RFLP products were subjected to electrophoresis using 2% agarose gel and the genotypes identified. The PCR amplified product for the partial region of the GH gene was approximately 422 bp (base pair). Two genotypes were identified for the GH locus in the Boer goat

population – AA and AB. The AA genotype showed two bands of approximately 366 bp and 56 bp. The AB genotype showed three bands which were approximately 422 bp, 366 bp and 56 bp. The frequency of the AB genotype was 0.9, while the frequency of the AA genotype was 0.1. The BB genotype was absent among the animals studied. Allele A was of higher frequency (0.55) compared to allele B (0.45). The population was not in Hardy-Weinberg equilibrium (HWE) for this locus ($P<0.05$). The herd exhibited heterozygosity excess. The Boer goat herd studied showed higher frequency of the AB genotype which has been reported to be associated with better growth performance than the other genotypes. This may be the result of the initial improvement strategies adopted during the development of the Boer goat. The deviation of the locus from HWE may be attributed to the selection and mating practices in the Boer goat herd studied. Future studies should include more Boer herds and investigate other regions of the GH gene. In addition, association analysis with growth traits should be conducted so that the molecular markers to be incorporated into the selection programmers may be identified and validated.

Keywords: Polymerase chain reaction (PCR), Restriction Fragments Length Polymorphism (RFLP), Haelll, Single nucleotide polymorphism

ABSTRAK

GROWTH HORMONE GENE POLYMORPHISM IN BOER GOAT

NAMA: MAGAMAEE A/P MALASWAMY (170251)

PENYELIA: PROF. DR. JOTHI MALAR PANANDAM

Dalam industri peternakan kambing pedaging, ciri tumbesaran kambing adalah pertimbangan utama dalam proses pembiakbakaan kerana penentu nilai ekonominya. Maklumat genetik molekul boleh digunakan bersama pendekatan genetik kuantitatif untuk memilih induk pembaka dengan merit genetik yang tinggi untuk program pembiakbakaan bagi mencapai peringkat genetik kerompok dengan cepat. Gen hormon tumbesaran (GH) memainkan peranan yang penting dalam mengawal selia tumbesaran dan peningkatan penggabungan asid amino dalam protein otot. Polimorfisme gen GH dan kaitannya dengan lilitan dada paada kelahiran dan berat badan semasa cerai susu telah dilaporkan. Objektif kajian ini adalah untuk mengkaji polimorfisme gen GH dalam satu kerompok kambing Boer di Malaysia dengan menggunakan *Restriction Fragment Length Polymorphism* (RFLP). DNA yang diekstrak daripada sampel darah 30 ekor kambing Boer yang dipilih secara rawak telah diamplifikasi bagi sebahagian gen GH menggunakan primers tertentu. Produk *polymerase chain reaction* (PCR) telah dipisahkan dengan elektroforesis dalam gel agarose 2% untuk pengesahan kejayaan amplifikasi. Amplikon PCR kemudiannya diurai dengan menggunakan enzim sekatan *Hae*III. Produk RFLP tertakluk kepada elektroforesis menggunakan gel agarose 2% dan genotip dikenal pasti. Produk PCR

yang telah diamplifikasi bagi sebahagian gen GH tersebut adalah lebih kurang 422 bp (base pair). Dua genotip telah dikenal pasti untuk lokus GH dalam populasi kambing Boer tersebut - AA dan AB. Genotip AA menunjukkan dua jalur dengan lebih kurang 366 bp dan 56 bp. Genotip AB menunjukkan tiga jalur yang lebih kurang 422 bp, 366 bp dan 56 bp. Kekerapan genotip AB adalah 0.9, manakala kekerapan genotip AA adalah 0.1. Genotip BB tidak wujud dalam binatang yang dikaji. Alel A adalah dengan frekuensi lebih tinggi (0.55) berbanding dengan alel B (0.45). Populasi tersebut tidak berada dalam Keseimbangan Hardy-Weinberg (HWE) untuk lokus tersebut ($P<0.05$). Kerompok tersebut mempamerkan berlebihan heterozigositi. Kerompok kambing Boer tersebut menunjukkan frekuensi lebih tinggi genotip AB yang telah dilaporkan mempunyai hubungakait dengan prestasi pertumbuhan yang lebih baik berbanding dengan genotip lain. Ini mungkin hasil daripada strategi penambahbaikan awal yang digunapakai semasa pengwujudan kambing Boer. Penyelewengan lokus tersebut daripada HWE boleh dikaitkan dengan amalan pemilihan dan pembiakbakaan dalam kerompok kambing Boer yang dikaji. Kajian akan datang perlu menggunakan lebih banyak kerompok Boer dan mengkaji bahagian lain gen GH. Tambahannya analisis perkaitan ciri tumbesaran perlu dikendali supaya penanda molekul yang boleh digabungkan dalam program pemilihan dapat dikenal pasti dan divalidasi.

Keywords: Polymerase chain reaction (PCR), *Restriction Fragments Length Polymorphism* (RFLP), *Haelll, Single nucleotide polymorphism*

CHAPTER 1

INTRODUCTION

The breed of the goat is considered a primary and essential decision in any goat industry. In goat meat industry, growth traits are the primary concern during breeding due to their determinant of economic values (Blott *et al.*, 1998). The Boer is an improved breed developed from indigenous South African breeds with some infusion of European, Angora and Indian goat. This breed has become a popular meat breed worldwide, including Malaysia.

Molecular genetics information is a useful tool to be applied with quantitative genetics information to select parental stocks for breeding and genetic improvement. However, appropriate molecular markers for traits of interest must first be identified. Molecular markers that reveal polymorphism at DNA level are now playing a crucial role in animal genetics. Recently several potential candidate genes were recognized. The Growth hormone (*GH*) gene is considered as one of the most important candidate genes influencing economic traits in meat goats such as growth, nutrient utilization, milk composition, galactopoesis, aging, reproduction and metabolism (Bauman and McCutheon, 1986). Little is known about the GH alleles in the Boer goats in Malaysia.

Considering the importance of GH, the local goat herds should be evaluated for polymorphisms at the GH locus and the predominant alleles at this locus should be identified. Thereafter, studies on the association of the single nucleotide polymorphisms (SNPs) at different regions of the GH locus with growth traits may be evaluated.

1.1 Research problem

The importation of goat breed and chevon in large quantity would be detrimental to the nation's economy. In addition, there are many cases of inbreeding occurring which could lead to poor performance. Generally there is little knowledge and documentation on the genetics of goat breeds in Malaysia. Lack of selection based on genetic merit and inbreeding may affect the gene frequencies. The alleles at the GH locus in Boer goats in Malaysia are unknown. Low frequencies of the favorable GH alleles in a herd will decrease the herd performance. Structured breeding system with proper genetic evaluation of the breeding stock is very important to develop the goat industry to be cost effective, viable and sustainable. The Malaysia Boer goat if properly managed will have a very bright potential for contributing to the transformation of agriculture.

1.2 Research hypothesis

The Boer goat has been developed through breeding and selection for rapid growth, excellent meat quality, great adaption, resistance to disease and high kidding percentage (Greylin, 2000). Therefore, the Boer breed will have higher frequency of the GH alleles reported to be associated with good growth. The uncontrolled breeding and lack of emphasis on genetics of the goat herds may reduce the frequency of the desirable allele.

1.3 Objectives

The general objective was to determine whether Boer goat in Malaysia exhibit higher frequency of the GH allele reported to be associated with good growth.

The specific objectives were:

1. To identify the alleles present at GH locus in Boer goat using Restriction Fragment Length Polymorphism (PCR-RFLP) technique.
2. To determine the genotype and allele frequencies as well as the heterozygosity values for the *GH*.
3. To determine whether the GH locus is in Hardy-Weinberg equilibrium in the population evaluated.

1.4 Significance of study

Results from this study will identify whether the Boer goat population evaluated has the favorable allele at the *GH* locus. This information may be used to further develop and improve the breed genetically through marker assisted selection. This will in turn increase the productivity of the Boer goat. These activities will further support the development of the industry towards the prescribed goals as mapped out by the government and private sector as well as contribute to the nation is local meat production.

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