

## Microsatellite Analysis of the Mafriwal Dairy Cattle of Malaysia

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### Introduction

The genetic variability of a population or breed is an important element in deciding on breeding and genetic conservation strategies. Molecular markers have been widely used to access genetic variability since they screen a wide region of the genome. RFLP and microsatellites markers have been used to explain bovine domestication and migration patterns (Loftus *et al.*, 1994; Bradley *et al.*, 1994) and to characterise cattle populations (MacHugh *et al.*, 1997; Kemeses *et al.*, 1999).

The Mafriwal dairy cattle of Malaysia is a tropicalised Sahiwal-Friesian crossbred with 50-75% Friesian genes. It has been developed by the Department of Veterinary Services Malaysia as a high milk producing breed type, adapted to the hot and humid local environment and resistant to local diseases. Since the Mafriwal is a new breed type, still under development and with varying Friesian gene content, its gene pool is subjected to changes. There has been no study on its genetic (molecular) variability. It is essential that the genetic variation that exists in the Mafriwal be evaluated, monitored and utilized. In addition, to accelerate the genetic improvement of Mafriwal modern techniques of selection, such as marker assisted selection, may be adopted if markers linked to economically important quantitative trait loci are identified.

The objectives of this study are to evaluate genetic variability of the Mafriwal dairy cattle of Malaysia using microsatellites, and to identify microsatellite markers that show association with important economic traits.

### Materials and Methods

Blood was randomly sampled from 40 Mafriwal cows at Institut Haiwan Kluang, Johor, of the Department of Veterinary Services Malaysia. The DNA was extracted from the whole blood and analysed for 32 microsatellite loci selected from the cattle genome database (MARC, 2003).

The Mafriwal cows available at the Institute were also ranked based on their milk production (average daily milk yield, total milk yield, lactation length and dry period). Blood samples were collected from 60 animals classified as the highest (30 animals) and lowest (30 animals) milk producers. DNA was extracted from the blood and analysed for 10 polymorphic microsatellite loci, which have been suggested to be associated with milk production. In an attempt to investigate the associations between the microsatellite alleles and the milk production traits, the average daily milk yield, total milk yield, lactation length and dry period of the two groups were compared with respect to presence or absence of alleles which were predominant in one of the groups.

### Results and Discussion

The microsatellite analysis generated 30 polymorphic loci (TGLA 53 and TGLA116 were monomorphic). The number of alleles per locus ranged from 4 (BM1329) to 8 (BM2639). The mean observed allele number was 6.23 and the effective mean allele number was 5.02. The number of genotypes per locus ranged from 8 (BMS1716) to 19 (BM6425) with a mean 6.23 and mode 7. The genotypic frequencies ranged between 0.03 – 0.33. The observed heterozygosity per locus ranged from 0.38 (MB002) to 0.78 (BRN); the mean was 0.55 and the modes 0.42 and 0.55. The mean overall heterozygosity was 0.79. The large number of polymorphic loci with high allele number and heterozygosity indicates high genetic variability in the Mafriwal. This may be partly attributed to Mafriwal being a relatively new breed type and comprising of animals with varying Friesian gene content. The mean number of alleles per locus is similar to that reported by Bishop *et al.* (1994), 6.8 alleles per locus from a survey of 468 microsatellite markers in 206 *Bos taurus*/*Bos indicus* animals. This suggests that microsatellite loci in cattle show high variability, which in turn may be attributed to the fact that microsatellites may be situated at the non coding regions of the genome and, therefore, are unaffected by the selection influencing functional genes. Significant ( $P < 0.05$ ) deviations from Hardy-Weinberg equilibrium were observed for all the polymorphic loci. This is as expected as animals from other breeding farms are often introduced into this herd and, since this herd serves as a nucleus herd the animals are subjected to selection.

High and low milk producers did not vary in the number of alleles per locus or heterozygosity. There was no allele totally absent in one of the groups but in high frequency in the other that could be associated with one of the milk production locus. High producers with allele A180 at HUV177 and low producers with allele D244 at MB002 had significantly ( $P < 0.05$ ) higher total milk yield than their group members that lacked these alleles. When the data of both groups were pooled, the animals with the absence of allele E146 at UWCA26 and J198 at BM1329 had significantly ( $P < 0.05$ ) higher total milk yield and average daily milk yield. The lack of clear association between the microsatellite markers and milk production traits may be due to the small sample size used in the investigation and the small number of loci investigated. Furthermore, the large

number of alleles detected per locus made individual allele frequencies too low to observe any association. The association suggested by some of the alleles is confined to one group or the pooled data and is not observed for the individual groups. This too may be due to the low allele frequencies for the markers concerned.

### Conclusions

The Mafriwal dairy cattle at Institut Haiwan Kluang shows high variability with respect to microsatellite loci. Since the Mafriwal is still in the development stage, its genetic variability is expected to change and should be monitored. There was a lack of clear association between the microsatellite markers and milk production traits. The possible association suggested by some of the markers must be further investigated using more animals and pedigree data before any definite conclusions may be made.

### Benefits from the study

This is the first molecular evaluation of a Mafriwal population. It will serve as a reference for subsequent monitoring of the changes in the genetic variability of this and similar populations.

Some microsatellite markers have been identified in this study that may be further investigated for association with milk production traits. If the association is confirmed, they may be used in marker assisted selection which would accelerate genetic improvement of the Mafriwal.

### Patent(s), if applicable :

Nil

### Stage of Commercialization, if applicable:

Nil

### Project Publications in Refereed Journals:

Nil

### Project Publications in Conference Proceedings

1. Kalaiselvi P, Panandam JM, Tan SG and Yusoff K. 2002. Association between microsatellite markers and milk production traits in Mafriwal dairy cattle. Proceedings of the 12<sup>th</sup> Scientific Meeting and 13<sup>th</sup> Annual General Meeting of the Malaysian Society for Molecular Biology and Biotechnology, 2002; p 39 (abstract).

### Graduate Research

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Name of Graduate	Research Topic	Field of Expertise	Degree Awarded	Graduation Year
Kalaiselvi Palani	Genetic characterisation of the Mafriwal dairy cattle of Malaysia using quantitative and molecular methods	Animal Genetics	Ph.D (working towards)	Anticipated 2004

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