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Genetic diversity parameters of Baladi Black and New Zealand rabbits in comparison with their crossbred offspring

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Abstract

Genetic diversity in rabbit populations plays a critical role in breeding programs aimed at enhancing desirable traits. This study investigates allele diversity at microsatellite loci in Baladi Black and New Zealand White rabbit breeds, as well as their crossbred offspring. Genome scanning using 6 microsatellite markers was applied to genotype animals. The results revealed that allele counts in Baladi Black rabbits varied markedly among loci, ranging from 4 to 13 alleles per locus with an average of 7.33. Effective allele numbers (Ne) showed similar variability, ranging from 2.07 to 10.1. Contrasts between observed (No) and effective alleles (Ne) indicated varying degrees of genetic activity, with differences ranging from 0.7 to 3.9 across loci. New Zealand White rabbits displayed a range of 4 to 15 alleles per locus, with effective allele numbers from 2.81 to 13. Differences between No and Ne ranged from 0.9 to 2.9. Crossbred rabbits exhibited intermediate allele counts (average 5.66 alleles per locus) and effective allele numbers (ranging from 1.69 to 8.1), reflecting genetic contributions from both parental breeds. Heterozygosity analysis revealed moderate to high observed (Ho) and expected (He) heterozygosity levels across populations. Polymorphic Information Content (PIC) values indicated moderate to high levels of polymorphism, with Baladi Black and New Zealand White breeds averaging 0.720 and 0.758, respectively, and crossbreds averaging 0.641 across loci. These findings underscore the multi-allelic nature of microsatellite loci and highlight their utility in assessing and enhancing genetic diversity in rabbit populations, essential for sustainable breeding and conservation strategies.

Introduction

Rabbits are important part of the rich diversity in Egypt. There exist several native breeds of rabbits that have been adapted to in the harsh environmental conditions [1]. They have been valuable food sources and rich reservoirs of culture. Among these breeds are the Baladi Red, Baladi Black, and Gabali [2]. Unfortunately, the native populations of these animals have been decreasing over the past few decades. Two of the breeds (Baladi White and Giza White) have already gone into extinction, whereas for the rest, there is a looming threat of extinction [2]. Therefore, estimation of diversity parameters in those breeds is essential, where low level of genetic diversity indicates low biological survivability and environmental adaptation, which is required for sustained genetic improvement and stable inheritance of desirable traits [3].

The native Egyptian rabbit breeds possess some peculiar features which make them fit to live in the hot, dry climate of Egypt [4]. These also demonstrate very high disease resistance [5]. Conservation of such genetic diversity would not only save such breeds from total annihilation but also would be focused on preserving a rare pool of genetic resources that could be a trump card for future rabbit improvement ventures in disease resistance [6,5]

Native rabbit breeds of Egypt present a unique chapter in rabbit domestication history: chances are that they hold the key to understanding past rabbit breeding practices and breed evolution [7]. Nevertheless, those breeds are characterized by poor performance compared to commercial lines [8,9]. Genetic improvement of reproductive traits and prolificacy, particularly in local rabbit breeds, is a crucial step in improving their performances in general [10] .

One of the most successful approach for traditional rabbit breeding is crossbreeding. The inter-population mating result in offspring that often benefit from heterosis [11]. It results in faster growth, larger litter size, and better feed conversion [12].

Besides this aspect, crossbreeding involves the advantage of bringing genes for resistance to specific diseases from one breed into another, so as to have much healthier offspring [12,13]. Different studies have shown that microsatellites can help to evaluate crossbreeding and also to estimate heterozygosity and inbreeding coefficients, allowing for the identification of full sibling groups and maintaining genetic diversity within captive populations.

In this concern, microsatellite analysis provides advantages over traditional methods, offering high exclusion probabilities, requiring small sample sizes, and being applicable to animals of all ages [14] .

The current work therefore aimed at evaluating the genomic parameters of crossbred rabbits between a commercial line and a local breed in comparison to its parents.

MATERIALS AND METHODS

Experimental design

Two rabbit breeds that differed in their genetic compositions were used in this experiment. The breeds were Baladi Black, which is a local breed in Egypt, and New Zealand White, which is an exotic commercial breed. The does and bucks of each breed were obtained and reared in a battery system housed in a naturally-ventilated conventional pen. The breeds were interbred for one generation, where one-way crossbreeding was performed between Baladi Black bucks and New Zealand White does. The bunnies remained with their mothers for a month for weaning and then were routinely raised and fed commercial diets.

DNA extraction

Blood samples (≈5ml) were collected from the ear vein of individuals of Black Baladi, New Zealand White, and crossed offspring (11 individuals/population) in sterilized tubes containing ethylene diamine tetra acetic acid (EDTA. All samples were immediately stored at -20°C.

Upon use, the samples were thawed and used for the genomic DNA extraction using the phenol/chloroform extraction procedure [15]. A blood sample (200µl) was put in a 1.5-ml Eppendorf tube and 20µl of proteinase K (10mg/ml) and 50µl of 10% sodium dodecyl sulfate (SDS) were added to the blood. The components were then thoroughly mixed by vortex and incubated in a water bath at 56°C for 120 minutes. Phenol/chloroform/isoamyl alcohol mixture was added to the digested blood at a ratio of 1:1, so 270µl of the mixture was added to the digested blood. The total volume was then mixed and centrifuged at 12000xg for 5 minutes. The upper phase (water phase) was transferred into another tube to repeat the last step. Then, 2.5 volumes of absolute ethanol and 1/10 volume of 3 M sodium acetate (pH = 5.2) were added, and mixed thoroughly. The mixture was incubated overnight at -20°C, and then centrifuged at 4°C at 12000xg for 15 minutes. The DNA pellet was collected, washed with ethanol 70%, dried and dissolved in 100µl TE buffer, and stored at -20°C until use.

Genotyping microsatellite markers

The individual samples were screened by six microsatellite primers, through the polymerase chain reaction (PCR) procedures. The molecular information of the different microsatellite primers is presented in Table (1). A total volume of 12.5 µl mixture was prepared for PCR, including 4.0 µl of DNA (75 ng), 1.0 µl of each of the forward and reverse primers (25 pmol), 6.0 µl master mix, and 0.5 µl PCR- grade water. The PCR reaction was performed using the thermal cycler (Techne, UK). The

amplification program included an initial denaturation step at94 ℃ for 5 min, and 35 Cycles of denaturation (94 ℃ /40 sec), annealing (60 ℃/45 sec), and extension (72 ℃/40 sec), followed by a final extension step at 72 ℃ for10 min.

Electrophoresis of DNA fragment

The PCR products were first separated by electrophoresis using the agarose gel electrophoresis. If DNA fragments were detected on agarose gel, the PCR products were then separated on 8% non–denaturing polyacrylamide gel by electrophoresis. The electrophoresis results were visualized and photographed using the WGD-30 WiseDoc Gel Documentation (Daihan Scientific, South Korea). The DNA images were analyzed for allele detection, volume, and length (bp) using the TotalLab software (Total LabLtd, UK).

Data analysis

The data generated from the microsatellite-genotyping of the rabbit breeds were used to calculate the number of alleles (*No*) per locus per breed and the observed heterozygosity (*Ho*) and effective number of alleles (*Ne*). The expected heterozygosity (*He*) within-breed was estimated according to Ott [16], and polymorphic information content (*PIC*) was estimated according to Botstein [17].

Results and Discussion

Allele diversity

Table (2) presents information on the microsatellite alleles detected at the different loci for the two breeds and their crossbred rabbits. For the Baladi Black breed, the loci varied in the observed number of alleles (No). The observed number of alleles were 4, 7, 13, 7, 8, and 5 for locus D5Utr4B, D5Utr4C, D5Utr4F, D7Utr4A, D7Utr4B, and D19Utr4B, respectively, with a mean of 7.33. The highest number of alleles was 13 alleles that were detected at locus D5Utr4f, and the lowest number of alleles was 4 at

locus D5Utr4B. The effective number of alleles (Ne) also varied between different loci, with values of 2.07, 6.28, 10.1, 3.07, 7.2 and3.25 in D5Utr4B, D5Utr4C, D5Utr4F, D7Utr4A, D7Utr4B, and D19Utr4B, respectively, with a mean of 5.32. Allele frequencies across microsatellite loci were different due to the differences in the distribution of the allele frequency for each allele size among the breeds. The difference between No and Ne monitors the genetic activity of the population. The differences were 1.9, 0.7, 2.9, 3.9, 0.8 and 1.7 for primer D5Utr4B, D5Utr4C, D5Utr4F, D7Utr4A, D7Utr4B and D19Utr4B, respectively. The highest difference was estimated at 3.9 for prime D7Utr4A, and the lowest difference was 0.7 for D5Utr4C. The results indicate that the microsatellite loci were multi-allelic.

Table (2) also shows the information on alleles detected by different microsatellite primers in the New Zealand White breed. The results indicate that the microsatellite loci were multi-allelic. The allele number ranged from 4 to 15 loci, with a mean of 7.5 alleles/locus in D5Utr4B, D5Utr4C, D5Utr4f, D7Utr4A, D7Utr4B and D19Utr4B, respectively. The highest effective number of alleles was 13 alleles for prime D5Utr4F and the lowest was 2.81 in D5Utr4B.The difference between No and Ne were1.1, 1.7, 2, 0.92 0.9, 2.9 for D5Utr4B, D5Utr4C, D5Utr4f, D7Utr4A, D7Utr4B and D19Utr4B, respectively. The highest difference was estimated at 2.9 for prime D19UTR4B and the lowest was 0.9 for D7UTR4b in New Zealand rabbits.

The information on the recognized alleles in different microsatellite loci for crossbred rabbits is presented in Table (2). The total number of alleles detected in all loci was 34 alleles, with an average of 5.66 alleles/locus. The results indicate the multi-allelic nature of the microsatellite loci. In addition, the effective number of alleles varied between different loci, with values of 1.69, 3.65, 7.6, 2.5, 8.1and 2.74 in D5Utr4B, D5Utr4C, D5Utr4F, D7Utr4A, D7Utr4B, and D19Utr4B, respectively, with a mean of 4.38. The

difference between No and Ne was 1.3 for D5Utr4B ,1.3 for D5Utr4C ,2.3 for D5Utr4F, 0.5 for D7Utr4A ,1.9 for D7Utr4B and 0.6 for D19Utr4B.The highest difference was 2.3 for D5Utr4F and the lowest difference was estimated 0.6 for D19Utr4B.

Microsatellite markers are often recommending for genetic studies in animals due to their high polymorphism, which is repented by the number of alleles that is usually higher than the minimum of 5 alleles [18]. The effective number of alleles reflects the contribution of allele frequencies to genetic diversity, where a population with a lower effective number of alleles is usually more susceptible to genetic drift [19,20] . The effective number of alleles was previously reported to averaged 6.625 in seven breeds of rabbits and indicated that the gene polymorphisms and genetic diversity were abundant [21] . In Egypt, the number of detected alleles in five rabbit breeds varied among the different breeds and ranged between 2.00 in Baladi White and 2.71 in New Zealand rabbits [22]. Also, the number of observed alleles was reported to range between 3.64 and 10 with an average 6.125 in local Egyptian rabbits [6], Higher average number of alleles per locus (14.26) was previously estimated [23]. In the current study, the results of the number of alleles were aligned with that were previously reported [24.25]. However, the effective number of alleles was lesser in the crossbred rabbits than their parents, this may indicate loss of rare alleles due to crossbreeding.

Heterozygosity

The observed (*Ho*) and expected (*He*) heterozygosity at the microsatellite loci in Baladi Black rabbits are presented in (Table 3). The *Ho* in Baladi Black varied from low to high and ranged from 0.25 to 1.00, with an average of 0.76. In comparison, the *He* was moderate to fairly high and ranged from 0.52 to 0.90, with an average of 0.74.

The observed (*Ho*) and expected (*He*) heterozygosity at the microsatellite loci in New Zealand White rabbits is presented in (Table 3). The *Ho* in New Zealand White was low to high and ranged from 0.33 to 1.00 with an average of 0.74, whereas *He* varied from moderate to fairly-high, and ranged from 0.64 to 0.93 with an average of 0.77.

The observed (*Ho*) and expected (*He*) heterozygosity at the microsatellite loci in Baladi Black is presented in (Table 3). The *Ho* in the crossbred rabbits varied from low to high and ranged from 0.00 to 1.00, with an average of 0.56. In comparison, the *He* was in general moderate to fairly high and ranged from 0.41 to 0.87, with an average of 0.68.

The average of Ho was previously reported to be lower than the He in four Egyptian rabbit breeds (Baladi Black, Gabali, Baladi Red, White Giza) and New Zealand White where Ho ranged from 0.477 in NZW to 0.581 in Giza White [26]. Also in NZW rabbits, the minimum expected heterozygosity was 0.008, the maximum value was 0.709, and the average value was 0.450 [27], Higher values for total population heterozygosity ranged between 0.340 and 0.878 with an average 0.705 in Angora rabbits [28] .

The values obtained for expected heterozygosity in the current study are comparable and consistent to those observed in Indian rabbits (0.842-0.849) [24]. in Egypt Moshtohor line rabbits (0.66-0.88) [29], but higher than that in Pygmy rabbits (0.54-0.60) [30]. indigenous Tunisian rabbit (0.39-0.58) [31], and in five Egyptian rabbit breeds (0.20-0.65) [32] , indicated that the gene polymorphisms and genetic diversity are abundant.

Moreover, the average expected heterozygosity for NZW rabbits was ranged between 0.33 at D19UTR4B locus to 0.935 at D7UTR4B locus, which are close to our results [21]. It worth mentioned that the Ho was less than He in both NZW and crossbred rabbits, which mainly indicate strong dominance relationships between loci in the

parental generation this also may be attributed to the loss of rare alleles due to crossbreeding. However, the unexpectedly high value of observed heterozygosity in Baladi Black rabbits can be explained by hidden population structure, where unknown breeding practices were applied for this population.

Polymorphic information content

The polymorphism measured as polymorphic information content (*PIC*) at different microsatellite loci is presented in Table (4). For the Baladi Black breed, The *PIC* in Baladi Black rabbits was in general highly moderate to high and ranged from 0.469 at the locus D5Utr4B to 0.890 at the locus D5Utr4F on chromosome 5, and it averaged 0.720 overall loci. In New Zealand White rabbits, *PIC* was moderate to high, with an average of 0.758 overall loci, and the highest value was 0.930 at the locus D5Utr4F on chromosome 5. The *PIC* in the crossbred population varied from low to high and ranged from 0.358 to 0.863, with an average of 0.641 overall loci. The polymorphism is significant in the adaptability of the living organisms to the local environment.

Polymorphic information content is a statistical measurement to assess the informativeness of genetic markers, and when values are ranged between 0.5 and 0.8, it indicates a highly informative marker, and suggests the marker can effectively distinguish between different genotypes [33]. The mean PIC value in 7 rabbit breeds was found to be ranging from 0.625 to 0.796 [22]. In NZW rabbits, the average PIC was 0.556 [21]. A wider range of PIC ranged between 0.083 and 0.936 was observed in Algerian rabbits [25].

Conclusion

This study employed microsatellite markers to analyze the genetic diversity of a hybrid population in comparison with its parental breeds (Baladi Black and New

Zealand White). The results revealed a high level of genetic diversity across all breeds. All loci examined were polymorphic, containing multiple alleles. The unique genetic makeup of Baladi black was distinct from imported breeds, and suggests a natural adaptation encoded in their genes.

Reference

Abou Khadiga, G., Y. M. K. Youssef, K. Saleh, R. Y. Nofal, and M. Baselga. 2010. Genetic trend in selection for litter weight in two maternal lines of rabbits in Egypt. World Rabbit Sci. 18:27–32 Available at https://polipapers.upv.es/index.php/wrs/article/view/674.

Khalil, M. H. 1999. Rabbit genetic resources of Egypt. Anim. Genet. Resour. Inf. 26:95–111 Available at https://www.cambridge.org/core/article/rabbit-genetic-resources-of-egypt/B928DB5AF743B6E015210601F8F3AB4E.

Badr, O. A. M., I. I. S. El-Shawaf, M. H. A. Khalil, M. H. Refaat, and S. I. A. Ramadan. 2019. Molecular genetic diversity and conservation priorities of Egyptian rabbit breeds. World Rabbit Sci. 27:135–141.

Sabry, M. I. E., M. M. Zaki, F. A. Elgohary, and M. M. Helal. 2021. Sustainable Rabbit Production under the Global Warming Conditions in Southern Mediterranean Region. World's Vet. J. 11:543–548

Setiaji, A., D. A. Lestari, N. S. Pandupuspitasari, I. Agusetyaningsih, and F. A. Khan. 2023. Genetic characteristics of complete mtDNA genome sequence of Indonesian local rabbit (Oryctolagus cuniculus). J. Genet. Eng. Biotechnol. 21:96.

Abdel-Kafy, E. S. M., S. S. El-Din Ahmed, A. El-Keredy, N. I. Ali, S. Ramadan, and A. Farid. 2018. Genetic and phenotypic characterization of the native rabbits in

Middle Egypt. Vet. World 11:1120–1126 Available at https://pubmed.ncbi.nlm.nih.gov/30250372.

Khalil, M. H., and G. Bolet. 2010. Sustainable rabbit breeding and genetic improvement programs achieved in developing countries.Page 962 in In Proceedings of the World Congress on Genetics Applied to Livestock Production. Leipzig, Germany

Helal, M. M. 2019. Association between growth hormone receptor gene polymorphism and body weight in growing rabbits. Adv. Anim. Vet. Sci. 7:994–998.

Safaa, H. M., M. Ragab, M. Ahmed, B. El-Gammal, and M. Helal. 2023. Influence of polymorphisms in candidate genes on carcass and meat quality traits in rabbits. PLoS One 18:e0294051.

Helal, M., J. Sameh, S. Gharib, R. M. Merghany, M. Bozhilova-Sakova, and M. Ragab. 2024. Candidate genes associated with reproductive traits in rabbits. Trop. Anim. Health Prod. 56:94.

KHALIL M.H., , AFIFI E.A., YOUSSEF Y.M.K, and KHADR A.F. 2010. Heterosis, Maternal and Direct Genetic Effects for Litter Performance and Reproductive Intervals in Rabbit Crosses. World Rabbit Sci. 3 Available at http://dx.doi.org/10.4995/wrs.1995.247

Ragab, M., M. Baselga, A. G. EL Nagar, C. Mínguez, and J. Pablo Sánchez. 2021. Genetic analysis of functional longevity in a complete diallel crossing experiment involving four maternal rabbit lines. J. Anim. Breed. Genet. 138:474–481

Ragab, M., I. Elkhaiat, H. Younis, M. Ahmed, and M. Helal. 2022. Genotype by heat

conditions interaction effects on growth and litter traits in rabbits. Front. Vet. Sci. 9 Available at https://www.frontiersin.org/articles/10.3389/fvets.2022.1018625.

Canales Vergara, A. M., V. Landi, J. V. Delgado Bermejo, A. M. Martínez, P. Cervantes Acosta, A. Pons Barros, D. Bigi, P. Sponenberg, M. Helal, M. H. Banabazi, and M. E. Camacho Vallejo. 2020. Design and development of a multiplex microsatellite panel for the genetic characterisation and diversity assessment of domestic turkey (Meleagris gallopavo gallopavo). Ital. J. Anim. Sci. 19:392–398.

Sambrook, J., E. F. Fritsch, T. Maniatis, and Sambrook-Fritsch-Maniatis... 1989. Molecular Cloning: A Laboratory Manual;[prepared for Use in the CSH Courses on the Molecular Cloning of Eukaryotic Genes]. Cold Spring Harbor Laboratory Press.

Ott, J. 1992. Strategies for characterizing highly polymorphic markers in human gene mapping. Am. J. Hum. Genet. 51:283–290.

Botstein, D., R. L. White, M. Skolnick, and R. W. Davis. 1980. Construction of a genetic linkage map in man using restriction fragment length polymorphisms. Am. J. Hum. Genet. 32:314–331.

El-Aksher, S. H., H. S. Sherif, M. H. Khalil, H. A. S. El-Garhy, and S. Ramadan. 2017. Molecular analysis of a new synthetic rabbit line and their parental populations using microsatellite and SNP markers. Gene Reports 8:17–23

Hardy, O. J., N. Charbonnel, H. Fréville, and M. Heuertz. 2003. Microsatellite allele sizes: a simple test to assess their significance on genetic differentiation. Genetics 163:1467–1482

Helal, M., and E. El-Gendy. 2019. Estimation of Genetic Differences Between Rabbit Breeds Using Microsatellite Loci on Chromosomes 5, 7 and 19. Egypt. Poult. Sci. J. 39:731–740.

Tian-Wen, W., X. Gui-Jiang, P. Yu-Lai, X. Xi-Ping, L. Bi-Chun, and W. Xin-Sheng. 2010. Study on Gennetic Diversity of 7 Rabbit Populations Evidenced by Microsatellite Makers. J. Anim. Vet. Adv. 9:359–365 Available at http://dx.doi.org/10.3923/javaa.2010.359.365.

El-Gendy, E., Y. Hafez, A. M. Mohamed, M. M. Hela, and M. Abd Elrahman. 2014. Genome analysis of local and exotic rabbit breeds in Egypt. African J. Biol. Sci. 10:101–110.

Boukabene Fouzi, K., Homrani, and A. Ammam. 2018. Population structure and genetic diversity using microsatellite markers of four Algerian rabbit populations precludes hybridization with foreign breeds. South Asian J. Exp. Biol. 7:191–200 Available at http://dx.doi.org/10.38150/sajeb.7(5).p191-200.

Kannegundla, U., S. S. Reddy, P. Amareswari, M. G. Prakash, and M. Mahender. 2018. Genetic Diversity and Phylogenetic Relationship Analysis of two Rabbit Breeds by Microsatellite Markers. J. Anim. Res. 8:289–296.

Bouhali, A., A. Homrani, N. Ferrand, S. Lopes, and A. M. Emam. 2023. Assessment of genetic diversity among native Algerian rabbit populations using microsatellite markers. Arch. Anim. Breed. 66:207–215.

Grimal, A., H. M. Safaa, and G. M. K. Mehaisen. 2012. Phylogenetic Relationship Among Four Egyptian and One Spanish Rabbit Populations Based on.Pages 177–181 in Proc.: 10th World Rabbit Congress, 3-6 September, 2012, Sharm El-Sheikh, Egypt

Li, J., B. Zhao, Y. Chen, B. Zhao, N. Yang, S. Hu, J. Shen, and X. Wu. 2020. A Genetic Evaluation System for New Zealand White Rabbit Germplasm Resources Based on SSR Markers. Anim. an open access J. from MDPI 10:1258 Available at https://pubmed.ncbi.nlm.nih.gov/32722175.

Xin-Sheng, W., W. Tian-Wen, Z. Hui-Ling, C. Guang-Long, X. Qi, C. Jin-Hua, Z. Xiu-Bai, and C. Guo-Hong. 2008. Correlation Analysis of Wool Yield in Wan Line Angora Rabbits Using Microsatellite DNA Markers. J. Biol. Sci. 8:679–682 Available at http://dx.doi.org/10.3923/jbs.2008.679.682.

El-Aksher, S. H., H. S. Sherif, M. H. Khalil, H. A. S. El-Garhy, and S. Ramadan. 2016. Comparative genetic analysis among Moshtohor line rabbits and their parental lines using microsatellite markers.Pages 5–9 in 3rd International Conference on Biotechnology Applications in Agriculture (ICBAA), Benha University.

Thimmayya, A. C., and S. W. Buskirk. 2012. Genetic connectivity and diversity of pygmy rabbits (*Brachylagus idahoensis*) in southern Wyoming. J. Mammal. 93:29–37 Available at http://dx.doi.org/10.1644/11-mamm-a-045.1.

Ben Larbi, M., M. San-Cristobal, C. Chantry-Darmon, and G. Bolet. 2014. Population structure in Tunisian indigenous rabbit ascertained using molecular information. World Rabbit Sci. 22:223–230 Available at http://dx.doi.org/10.4995/wrs.2014.1468.

Rabie, T. S. K. M. 2020. Assessment of Genetic Variability and Population Structure of Five Rabbit Breeds by Microsatellites Markers Associated with Genes. J. World's Poult. Res. 10:125–139 Available at http://dx.doi.org/10.36380/jwpr.2020.17.

Serrote, C. M. L., L. R. S. Reiniger, K. B. Silva, S. M. dos Santos Rabaiolli, and C. M. Stefanel. 2020. Determining the polymorphism information content of a molecular marker. Gene 726:144175.

Table (1): The molecular information of microsatellite primers.

Locus	Allele size (bp)		Sequence	GC (%)
D5UTR4B	188-194	F R	5′CAGCGGTAAGAGTGAGAAAC 3′ 5′TCCCCCATAACAAAAGAGG 3′	50.0 47.3
D5UTR4C	278-270	F R	5′ GCTCTTGGCTCCTGGTTTC 3′ 5′ AGAGTTCTCCGTCCCTGATGG 3′	57.8 57.1
D5UTR4F	214-244	F R	5′ CCAGCTGGTAATAGTAGAGA 3′ 5′ AAGGCATTTGTGGAGTGAA 3′	45.0 42.1
D7UTR4A	225-237	F R	5′ TGCTAATGTGCCCAGAAAGGTA 3′ 5′ GGCATCCCAAAAGGCAGTAT 3′	45.4 50.0
D7UTR4B	153-163	F R	5′ TAGGCATTTAGGGAGTGAAC 3′ 5′ GGAGGGGGATGGTAGAG 3′	45.0 64.7
D19UTR4B	237-239	F R	5′ TGTATGTGGGTGTGGGTGTAGAG 3′ 5′ TACTGTTGCTTGCTGGGATTTTTA 3′	52.3 37.5

Table 2: The allele diversity in different microsatellite loci

	Baladi Black			***New Zealand White***			***Crossbred***		
locus	N_o	N_e	diff	N_o	N_e	Diff	N_o	N_e	Diff
D5Utr4B	4	2.1	1.9	4	2.8	1.1	3	1.7	1.3
D5Utr4C	7	6.3	0.7	5	3.3	1.7	5	3.7	1.3
D5Utr4F	13	10	2.9	15	13	2	10	7.6	2.3
D7Utr4A	7	3.1	3.9	6	5.1	0.9	3	2.5	0.5
D7Utr4B	8	7.2	0.8	7	6.1	0.9	10	8.1	1.9
D19Utr4B	5	3.3	1.7	8	5.1	2.9	3	2.7	0.6
Mean	7.3	5.3	2	7.5	5.9	1.6	5.7	4.4	1.3
SE	1.3	1.3	0.5	1.6	1.5	0.3	1.4	0.5	0.1

N_o and N_e indicates number of alleles and effective number of alleles, respectively.

Table 3: The observed heterozygote (H_o) and expected heterozygote (H_e) at different microsatellite loci

Locus	***Baladi Black***		***New Zealand White***		***Crossbred***	
	H_o	H_e	H_o	H_e	H_o	H_e
D5Utr4B	1	0.5	0.3	1	0.4	0.4
D5Utr4C	1	0.8	0.6	1	0.6	0.7
D5Utr4f	1	0.9	1	1	0.6	0.9
D7Utr4A	0	0.7	0.9	1	0	0.6
D7Utr4b	1	0.9	1	1	1	0.9
D19R4B	1	0.7	0.7	1	0.7	0.6
Mean	1	0.7	0.7	1	0.6	0.7
SE	0	0.1	0.1	0	0.1	0.1

Table 4: polymorphic information content (*PIC*) at different microsatellite loci

locus	***Genotype***		
	Baladi Black	***New Zealand White***	***Crossbred***
D5Utr4B	0.469	0.595	0.358
D5Utr4C	0.820	0.654	0.679
D5Utr4F	0.890	0.930	0.855
D7Utr4A	0.653	0.774	0.529
D7Utr4B	0.845	0.816	0.863
D19Utr4B	0.640	0.778	0.563
Mean	0.720	0.758	0.641
SE	0.065	0.048	0.080