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**TEKNOLOGI MARKA GEN α_1 -CASEIN SEBAGAI METODE
SELEKSI BIBIT UNGGUL KAMBING PERAH
PERANAKAN ETAWA (PE)**

Tahun ke 1 dari rencana 2 tahun

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SUMMARY

Technology of Marker α 1-Casein Gene for Selection Method in *Peranakan Etawah (PE)* Goats

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Peranakan Etawa (PE) goats are a local resource that has a high potential as a source of economic society. There are have a wide variety of farm value to people with a variety of purposes such as the maintenance of socio-economic, cultural and tourism. However until now, they have not been able to provide a significant economic contribution to the lives of the farmer and development of farm goat has a great opportunity to support the community's economy, because the management is still an conventional . Raising goats do not require large capital and its management is easy, can be done both women and children. They are a prolific, having more than one kid per birth and is a dual-purpose goat can produce meat and milk, which to this day still has a vast market share and market saturation has not occurred.

Goat milk production is still low, and also the availability of quality superior does of milk and meat production has not been there. To meet the demands of goat's milk in a relatively short time is needed based selection of genetic markers (Marker Assisted Selection / MAS). MAS is a selection method based on genetic markers that are supported by phenotypic data selection and breeding technology system that is efficient and accurate for developing breeds, which have several advantages including elements can be analyzed directly on the genetic (DNA) so they are not influenced by changes in the environment, MAS requires the candidate genes that have a strong influence as α 1-Casein gene, it is the major milk protein in the ruminants. The relation of α 1-casein gene with milk production has been reported in cows, sheep and goats. So it can be used as candidate genes for the production of goat milk.

The purpose of this study are 1) to design technology of selection method by analyze marker α 1-Casein gene was associated with milk production both in quality and quantity of goat 's milk ; 2) to genotyping PE goats by marker α 1-Casein gene to indentified superior genetic of PE goat which can be used to elite goats by farmer. And 3) International or national journal.

The benefits of the study are 1) . Science : the basis for the development of efficient and accurate selection based on gene markers for milk production and properties elite does of dairy goats produce both phenotypically and genetically; 2) . Applied : the results of this study can be applied by farmers and or groups and work closely with Universities to conduct elections goat that has the capability of high milk production 3) . Policy: The results of this study can help in the decision-making process by the government in the development of dairy goats which have the advantage of high milk production and quality.

The present study was conducted on a total of 54 PE lactation goats located in *Agus Farm* Bumiaji Malang. The data collected is 1). milk production during lactation for 3 months ; milk samples were collected immediately after milking for analysis of the components of milk every month ; BCS (according of Villaquiran et al., 2004) 4). Blood samples were taken from the jugular vein and collected in vacuainers containing K₃EDTA as an anticoagulant. Genomic DNA was isolated from blood using DNA extraction kit (**PROMEGA**) according to the manufacturer's instruction.

The PCR was carried out in a 10 μ L reaction mixture containing: 1 μ L genomic DNA, 2 μ L ddH₂O, 5 μ L PCR mix and 1 μ L each primer forward: F5' –CAT TCT TTA CTC CTG GGA AAG– 3' ; reverse: 5' –AGC ACT TTT GGG AAC AAT TTC-3 (Oligo™). The amplification protocol was used as follows: an initial cycle 94°C for 5 min, 35 cycles of steps containing denaturation 94°C for 30 sec , annealing 60° C for 30 sec , extension 72 ° C for 30 sec and the final extension 72 ° C for 10 minutes. Restriction enzyme digestion; A total volume of 5 μ L of each PCR product was digested with 1 μ L of F:5'---G/GATCC-3' dan R: 3'---CCTAG/G---5' Bam HI endonulcease 3 hours at 37°C. Digested products were analyzed by means of electrophoresis in PAGE 10% with silver stained.

The results are : BCS affect milk production but no milk protein milk fat. Milk production (1072.09 ± 276.53 mlECM/ head /day); milk protein ($3.28 \pm 0.51\%$); milk fat ($5.77 \pm 1.38\%$); Lactose ($4.27 \pm 0.43\%$); SNF ($9.12 \pm 0.3\%$) and BCS (2.47 ± 0.31) of PE goats at the study.

Genotyping at the α_1 -Casein gene locus at the DNA level revealed the presence of two alleles, namely E and F and the degree of polymorphism and 78%. The PCR amplified product was observed as 900 bp. The PCR was digested with **Bam HI** were three genotypes; 420, 450, 480 and 500 bp band EE, 420 and 450; band EF 420, 450 and 480 ; band FF 480 and 500 bp were observed in the population.

The allelic frequencies E is 0.78 and F is 0.22 while the frequency of EE, EF and FF genotypes are 0.63; 0.30; and 0.07 respectively and the populations are in Hardy - Weinberg equilibrium .

The α_1 -casein genotype had a highly significant ($p < 0.01$) effect on milk production at three months of lactation and had not effect on milk protein and milk fat. EE genotype had higher milk production than EF and FF. The milk production was 1326.59 ± 372.41 ; $1017.28 \pm 306,41$ and 872.41 ± 150.78 mlECM/head/day in the EE, EF and FF genotype. The protein milk of EE, EF and FF genotype were 3.39 ± 0.55 ; 3.36 ± 0.6 and 3.11 ± 0.39 % respectively. The milk fat were $6.40 \pm 1.60\%$; $5.92 \pm 1.62\%$ and 4.99 ± 0.91 for EE, EF and FF genotype

The conclusion of this study is α_1 - casein genotyping observed were three variation genotype EE, EF and FF. The frequencies of E allele higher than F allele. The genotypes had highly significant effect on milk production and the superiority is EE genotype. Marker gen α_1 - casein can be used in selecting superior genetic structure for milk production in young female goats in shorter time than the conventional selection method. In addition α_1 - casein gene can be used genetic marker for breeding program to improve milk production for PE dairy goat.

The suggestion of this research is to improve the quality of PE goat needs to be done using a selection by marker genetic by α_1 - casein gene in order to obtain the superior goat on milk production. Importance of building a village breeding PE dairy goats at the provincial level in order to supply superior goat for milk production for farmers. The further studies to identified genotype kid (yearling) for breeding program and replacement stock of elite goats

Keyword: *Indonesian Goats, α_1 -casein, marker gene.*

RINGKASAN

TEKNOLOGI MARKA GEN α_1 -CASEIN SEBAGAI METODE SELEKSI BIBIT UNGGUL KAMBING PERAH PERANAKAN ETAWA (PE)

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Kambing Peranakan Etawah (PE) merupakan sumberdaya lokal yang memiliki potensi tinggi sebagai sumber ekonomi masyarakat. Kambing mempunyai berbagai variasi nilai bagi peternakan rakyat dengan berbagai tujuan pemeliharaan seperti social - ekonomi, kebudayaan dan pariwisata. Namun sampai saat ini pengeloannya masih bersifat sederhana sehingga belum mampu memberikan sumbangan ekonomi yang berarti bagi kehidupan peternakan rakyat. Pengembangan kambing PE berpeluang besar dalam mendukung perekonomian masyarakat, karena beternak kambing PE tidak membutuhkan modal besar dan pengelolannya mudah, bisa dilakukan baik wanita maupun anak-anak dan bersifat prolifrik yaitu mempunyai kemampuan beranak lebih dari satu ekor per kelahiran dan merupakan kambing dwi guna dapat menghasilkan daging dan susu, yang sampai saat ini mempunyai pangsa pasar yang masih luas dan belum terjadi kejenuhan dalam pasar.

Produksi susu kambing PE saat ini masih rendah, dan juga ketersediaan bibit unggul produksi susu maupun produksi daging belum ada. Untuk memenuhi kebutuhan/permintaan susu kambing dalam waktu relatif cepat dibutuhkan seleksi berbasis marka genetik (Marker Assisted Selection/MAS).

MAS adalah metode seleksi berdasarkan marka genetik yang didukung oleh data fenotipik merupakan teknologi sistem seleksi dan pemuliaan ternak yang efisien serta akurat untuk mengembangkan bibit unggul, dimana memiliki beberapa keunggulan diantaranya bisa menganalisis langsung pada unsur genetik (DNA) ternak yang bersangkutan sehingga tidak dipengaruhi oleh perubahan lingkungan, informasi diperoleh dari individu yang bersangkutan, dan bukan informasi dari tetua, saudara atau keturunan seperti yang dilakukan pada teknik konvensional, dan dapat dilakukan pada ternak saat usia dini sehingga waktu yang dibutuhkan lebih pendek. MAS memerlukan gen kandidat yang mempunyai pengaruh cukup kuat seperti gen α_1 -casein, dimana α_1 -casein adalah komponen utama dari protein

Tujuan Penelitian ini adalah 1) mendesain suatu paket teknologi dalam menghasilkan kambing perah unggul melalui seleksi berbasis marka gen α_1 -Casein (2) Melakukan *genotyping* pada kambing PE berdasar marker gen α_1 -Casein dan kambing yang superior dapat digunakan sebagai sumber bibit pada industri kambing perah dan untuk memasok kebutuhan kambing bibit di peternakan rakyat. (3) Publikasi Nasional atau Internasional. Penelitian ini secara umum bermanfaat dalam mendukung pembentukan dan pengembangan kambing perah PE unggul produksi dan kualitas susu, sedangkan bagi Keilmuan : Hasil penelitian ini bermanfaat dalam pengembangan teknik seleksi yang efisien dan akurat berdasarkan marka genetik.;

Manfaat Terapan: Hasil penelitian ini dapat diterapkan oleh peternak bekerja sama dengan perguruan tinggi untuk melakukan seleksi berdasar gen penanda terhadap ternaknya yang memiliki potensi sebagai bibit unggul. Manfaat kebijakan Hasil penelitian ini dapat digunakan sebagai dasar kebijakan untuk membangun *breeding village* kambing perah PE unggul produksi dan kualitas susu, untuk penyediaan bibit unggul kambing PE bagi peternakan rakyat.

Materi yang digunakan adalah 54 ekor kambing perah PE laktasi yang berada di AGUS FARM Bumiaji Malang. Data yang dikoleksi adalah 1). produksi susu selama 3 bulan laktasi; sampel susu untuk analisis komponen susu setiap sebulan sekali; BCS (berdasar Villaquiran et al., 2004); 2) sampel darah kambing perah PE diambil dari *vena jugularis* dengan vacuoliner yang diberi K₃EDTA sebagai antikoagulant. DNA genom diisolasi menggunakan *Blood Genomic DNA Isolation Kit (PROMEGA)*

Prosedur PCR : 1 μ L DNA genom, 2 μ L ddH₂O, 5 μ L PCR mix and 1 μ L masing-masing primer (forward: F5' –CAT TCT TTA CTC CTG GGA AAG– 3' ; reverse: 5' –AGC ACT TTT GGG AAC AAT TTC-3 (Oligo™).) Program amplifikasi meliputi : predenaturasi 94 $^{\circ}$ C for 5 menit, dengan 35 siklus yang diulang antara lain denaturation 94 $^{\circ}$ C for 30 detik , annealing 60.0 $^{\circ}$ C 30 detikt , ekstension 72 $^{\circ}$ C for 30 detik dan ekstensi akhir 72 $^{\circ}$ C for 10 menit. Produk PCR didigesti dengan 1 μ L of Bam H1 endonulcease selama 3 jam pada suhu 37 $^{\circ}$ C. Produk digesti yang dihasilkan analisis dengan elektroforesis PAGE 10% dengan pewarnaan perak nitrat.

Hasil penelitian: 1) BCS sangat nyata pengaruhnya terhadap produksi susu tetapi tidak berpengaruh terhadap protein dan lemak susu kambing saat laktasi. Rataan produksi susu kambing PE , (1072.09 \pm 276.53 mLECM/ ekor /hari); protein (3.28 \pm 0.51%); lemak (5.77 \pm 1.38%); Laktose (4.27 \pm 0.43%); SNF (9.12 \pm 0.3%) and BCS (2.47 \pm 0.31)

Genotyping gen α_1 -casein menghasilkan dua macam varian alel yaitu alel E dan F, sedangkan derajat polimorfisme 78 %. Produk amplifikasi yang dihasilkan 900 bp dan setelah dilakukan digesti menggunakan enzim Bam H1 t fragment yang dihasilkan diberi nama : untuk pita 420 and 450 bp genotip EE; untuk pola pita 420, 450 and 480 bp adalah genotip EF dan pola pita 480 and 500 bp adalah genotip FF, dengan rincian Kambing genotip EE (34 ekor); EF(16 ekor) dan BB (4 ekor). Frekuensi alel E(78%); alel F (22%), sedangkan frekuensi genotip EE (0,63); EF (0,30) dan FF (0,07) serta populasi berada dalam keseimbangan *Hardy-Weinberg*.

Hasil analisis statistik membuktikan bahwa genotip berpengaruh sangat nyata ($p < 0.01$) terhadap produksi Genotip EE menghasilkan produksi susu tertinggi dibandingkan genotip EF dan FF. Produksi susu kambing genotip EE, EF dan FF secara berurutan : 1326.59 \pm 372.41; 1017.28 \pm 306,41 and 872.41 \pm 150.78 mLECM/ekor/hari. Akan tetapi genotip tidak berpengaruh terhadap protein dan lemak susu. Protein susu kambing genotip EE, EF dan FF berurutan adalah : 3.39 \pm 0.55; 3.36 \pm 0.6 dan 3.11 \pm 0.39 %. Sedangkan lemak susu adalah 6.40 \pm 1.60%; 5.92 \pm 1.62% dan 4.99 \pm 0.91 untuk genotipe EE, EF dan FF.

Kesimpulan penelitian ini adalah *$\alpha 1$ -Casein-genotyping* menghasilkan tiga varian genotip yaitu EE, EF dan FF, dan terdapat hubungan yang sangat nyata antara genotip dengan produksi susu, kambing genotip EE merupakan kambing dengan produksi susu tertinggi dibanding kambing genotip EF dan FF, akan tetapi tidak terdapat hubungan antara genotip dengan protein dan lemak susu. Produksi susu kambing genotip EE, EF dan FF secara berurutan adalah $1326,59 \pm 372,41$; $1017,28 \pm 306,41$ dan $872,41 \pm 150,78$ mlECM.ekor/hari. Marka gen $\alpha 1$ -casein dapat digunakan sebagai metode seleksi bibit unggul produksi susu pada kambing perah PE dan alel E pada gen $\alpha 1$ -casein merupakan alel pembawa sifat produksi susu tinggi, dengan frekuensi 78%.

Saran : Genetik kambing PE perlu ditingkatkan dengan melakukan seleksi menggunakan marka gen $\alpha 1$ -casein sehingga diperoleh bibit-bibit kambing unggul produksi susu. 1). Membangun *breeding village* kambing perah PE, ditingkat propinsi guna penyediaan bibit kambing PE unggul produksi susu bagi peternak. 2). Untuk penelitian tahun ke dua melakukan identifikasi struktur DNA atau genotyping $\alpha 1$ -casein anak-anak kambing untuk tujuan seleksi dini.

Kata kunci : Kambing Peranakan Etawah (PE), $\alpha 1$ -casein, marker gen

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