Open Access Research Journal of Life Sciences

Journals home page: https://oarjpublication/journals/oarjls/ ISSN: 2783-025X (Online)



(RESEARCH ARTICLE)

Check for updates

The valuable heterosis and genetic inheritance of live weight trait by growth hormone gene action in Ongole-grade cattle

Umar Paputungan *, Wapsiaty Utiah and Santie Turangan

Department of Animal Production, Faculty of Animal Sciences, Sam Ratulangi University, Manado 95115, Indonesia.

Open Access Research Journal of Life Sciences, 2023, 05(01), 019-027

Publication history: Received on 08 November 2022; revised on 04 January 2023; accepted on 06 January 2023

Article DOI: https://doi.org/10.53022/oarjls.2023.5.1.0080

Abstract

The crucial genetic factors of live weight inheritance including breeding value, dominance deviation, heritability and heterosis had not been studied in Ongole-grade cows. This study aims to identify the genotypic components including breeding value, dominance deviation, heritability and heterosis of live weight trait in Ongole-grade cows. A total of 78 blood samples from parental cows and 2 blood samples from parental Ongole-breed bulls were collected to be analyzed for presences of growth hormone (*GH*) genes. Presences of *GH* locus in blood samples were screened using PCR-RFLP method involving restricted enzyme Msp1 on agarose-gel (1.2%). Data were analyzed by statistical program in Excel XP. Results showed that the population mean (μ) of cow live weight was 385.98± 2.49 kg. Genetic component inheritance of homozygous genotypes of GH +/+ and *GH* -/- and heterozygous genotype of *GH* +/- were dominated by dominance gene action, rather than the additive gene action resulting the valuable heterosis for live weight trait of 3.2 percent. The heritability of dominance gene action (h_D^2) was 0.96 categorized as higher heritability, while heritability of additive gene action (h_A^2) was 0.04 categorized as low heritability under the equilibrium frequencies of both *GH* + (p = 0.48) and *GH* - (q = 0.52) genes without considering the nongenetic causes of environmental effect in the population of cow live weight.

Keywords: Genotypic component; Growth hormone gene; Live weight heterosis; Ongole breed

1. Introduction

Generally, the individual phenotype (P) is caused by its genotype (G), environmental effect (E) and both interaction (GE) that may now be written in common equation model: P = G + E + GE [1]. Genetically, the sample phenotype (P_{ij}) is caused by the average value of both homozygous phenotypes of P_{ii} and P_{ij} (denoted by m) and its genotypic value (V_{ij}) written in common equation model, P_{ij} = m + V_{ij} [1]. Furthermore, the population phenotype (P_{ij}) is caused by the population mean (μ), breeding value of the additive gene action (BV_{ij}) and the dominance gene action (D_{ij}) written in common equation model: P_{ij} = μ + BV_{ij} + D_{ij} without considering the nongenetic causes [1]. The parental genotype producing the highest average performance of the progeny was defined by an animal's breeding value. The function of the genotypic value was a difference in the phenotypes of animals in the single locus-example. Parents pass their genotype on only a random sample of one gene to each locus of the progeny. In the rural areas, the artificial insemination (AI) technical application was the most essential efforts enhancing higher reproductive performance at the same way of genetic improvement of the animals [2, 3].

A measure of the animal's expected progeny performance relative to the population mean is called breeding value. The progeny deviation itself represents the transmitting ability of the parent, which is one-half the breeding value [4]. This is the reason for doubling the progeny deviation due to containing only a sample one-half of the parent's genes. The breeding values are dependent on gene frequencies varying from population to population [1].

The breeder could rank the animals and cull those with the poorest evaluations while selecting those with the best evaluation as replacements. The proper application of heritability and relationships to weight records of the animal and

^{*} Corresponding author: Umar Paputungan

Copyright © 2023 Author(s) retain the copyright of this article. This article is published under the terms of the Creative Commons Attribution Liscense 4.0.

its relatives were required for an accurate evaluation [1]. The primary concern during breeding for its determinant economical value is the growth traits of animals in animal industry. Scientists are able to achieve more accurate and efficient selection goal by marker-assisted selection (MAS) under development of molecular biology and biotechnology. The initial and crucial step to establish a MAS system is validating the genetic markers of growth traits [5, 6].

An anabolic hormone synthesized and secreted by the somatotroph cells of the anterior lobe of the pituitary in a circadian and pulsatile manner is Growth hormone (GH) playing an important role in pubertal, prenatal and postnatal longitudinal growth and development, tissue growth, lactation, reproduction, as well as protein, lipid and carbohydrate metabolism [7]. The GH gene, with its functional and positional potential, has been widely used for marker in several livestock species, including the cattle such as *Bos taurus* and *Bos indicus* [8, 9].

The restriction fragment length polymorphisms (RFLP) of GH were associated with body weight in Grati dairy cows [10, 11]. In Ongole grade cattle, the studies of GH gene *MspI* locus have been reported [12]. Addition, this GH gene *MspI* locus has also reported in Brahman cattle [8], in Indian Zebu cattle [13] and in West coastal Sumatera cattle [14]. Their studies indicated that MspI +/+ and MspI +/- genotypes can be used as the candidate genes in cattle selection for breeding program. The difference between the genotypic value and the breeding value can be represented as the dominance deviation [1, 15, 16].

The value of the gene combination in the genotype is defined by the dominance deviation [1, 17]. The deviation of phenotype from the average of the two homozygous phenotypes is defined as the genotypic value [17]. The difference between breeding values is additive gene and representing the term of heritability for certain animal economical trait such as animal live weight. Heritability (h^2) is defined as the ratio of the additive genetic variance to the phenotypic variance. Thus, h^2 is the proportion of the total variance that due to differences among the breeding values of individuals in the population [1, 18, 19]. However, the breeding value of an individual local cow, referred to its additive genetic merit of live weight, has not been much studied. The objectives of this research were to identify the genotypic value, breeding value and dominance deviation of live weight from genotypic frequency of growth hormone (GH) restricted by *MspI* enzyme, and to define the heritability of live weight in Ongole-grade cow population in North Sulawesi province of Indonesia.

2. Material and methods

2.1. Animal Sample Collection

This research was carried out in the Sulawesi Province of Indonesia involving 78 cows as the parental generation at the all ages ranging 4 to 7 years old of Ongole Grade (OG) cows at Tumaratas Village as the artificial insemination (AI) service center of Minahasa regency, North Sulawesi province. All these parental cows were reared under private areas belong to farmers with the ancestor parental bulls of Ongole breed from "the artificial insemination bull germ plasma center" in Singosari, East Java province, Indonesia. All cows were born by mating using the artificial insemination generated from germ plasmas (semen) of the two Ongole bulls called "Kirsta" with genotype of Kr-GH^{+/+} and "Tunggul" with genotype of Tu-GH^{-/-}.

2.2. Analysis of the Genotypic Value



Figure 1 Samples of animal blood collected for DNA band analyses resulted in part of Ongole-breed bull called "Tunggul = Tu" with Growth Hormone (GH) genotype Tu-GH-/- and bull called "Krista = Kr" with GH genotype Kr-GH+/+, as well as other grade cow heterozygous codominant genotypes of GH+/- The extraction of DNA and genotyping for GH and allele identification were done using the protocols in DNA Laboratory [20, 21], as shown in Figure 1. The growth hormone locus genotype was performed using PCR-RFLP involving the restricted Msp1 enzyme produced by the Vivantis company which can be accessed through the <u>www.vivantechnologies.com</u> website (Product No. RE1302) and visualized at the standard of 1.2% agarose gel electrophoresis [20, 22].

1441							
	cccccacggg	caagaatgag	goccagcaga	aatcagtgag	tggcaacctc	ggaccgagga	
1501	gcaggggacc	tcc ttc atcc	taagtaggct	gccccagctc	ccgcaccggc	ctggggcggc	
1561	ctt ctc cccg	aggtggcgga	ggttgttgga	tggcagtgga	ggatgatggt	gggcggtggt	
1621	ggcaggaggt	cctcgggcag	aggccgacct	tgcagggctg	ccccagaccc	gcggcaccca	
1681	ccgaccaccc	acctgccagc	aggacttgga	gctgcttcgc	atc tcactgc	tcc tca tcca	
1741	gtcgtggctt	gggcccctgc	agttcctcag	cagagtette	accaacagct	tgg tgt ttgg	
exon	13. /g	ene-"GH1"	ę				
exon	13	Reve	rse				
intro	on 14 /g /g	/number=3 <u>1476</u> 1702 /gene="GH1" /number=3 1702 1964		Growth hormone locus Msp1 of cattle detected at intron number - 3, resulting in sequencing of base position to 1547			
intr	on 18	ene="GH1" umber=4 652137	(5' co 2.	egcac*egge	3') as seen i	n Figure	

Figure 2 Growth gene nucleotide sequences (Msp1) from primary GH5 Forward and GH6 Reverse marked cutting enzyme Msp1 (Retrieved on February 7 2022 from Cattle GenBank accession number: M57764) and the location of Msp1 growth hormone DNA Genome Nucleotide in the region structure of Exon and Intron 3 in cattle GenBank, Number M57764.1; http://www.ncbi.nlm.nih.gov)

Amplification of 327 bp fragments in intron 3 [23] was performed by PCR method using forward primer GH5: 5'-CCCACGGGCAAGAATGAGGC-3' and reverse primer GH6: 5'-TGAGGAACTGCAGGGGCCCA-3' [24] manufactured by the Laboratory of the Midland Certified Reagent Company Inc. Texas, *USA (Product Lot Number: 280511-03B)*". The PCR reaction was made through the application of 1x Taq pol 25µl from the master mix produced by the company "Axygen Biosciences, CA 94587, USA", (e-mail: support.axyprepkits@axygenbio.com).

Gene variations from the growth hormone locus to Msp1 in cattle can be detected at the position of intron 3 (Figure 2) with sequencing position 1547 based nucleotide sequencing from Cattle GenBank, accession number: M57764.1 [24]. The digested Msp1 of the PCR product produced the fragments of 104 bp and 224 bp for the Msp1⁺ (GH⁺) allele and 327 bp for the Msp⁻ (GH⁻) allele as shown in Table 1 with the band of fragment after Msp1 enzyme restriction as described on Figure 1. This enzyme can only recognize the location of the restrictios of the four nucleotide for the C¹CGG (Figure 2). The difference between the two allele fragments Msp1⁺ and Msp1⁻ was caused by the mutation of Cytosine (C) into Thymine (T) [25]. In contrast of *CRABP2* gene polymorphisms with growth traits in cattle breeds were found in intron 1 [26].

Table 1 Band of the fragment after Msp1 enzyme restriction

Length of DNA band (bp)	Identified allele	Genotype	
223	Normal allele (Men1 – CH+) *	Msp1 = GH+/+	
104	Normai anele (MSp1– GH+)		
327			
223	Msp1= GH+ and Msp1= GH-	Msp1 = GH+/-	
104			
323	Mutant allele (Msp1 = GH-) **	Msp1 = GH-/-	

*) Cut by Msp1 enzyme; **) Uncut by Msp1 enzyme.

The body live weights of animals (G0 and G1) records were determined by using a digital weighing scale when animals were standing [27, 28] prior to blood collection for DNA extraction of GH genes by electrophoresis method described in [20]. This study included the total of 78 cows with the numbers of animal genotypes of $GH^{+/+}$; $GH^{+/-}$ and $GH^{-/-}$ as shown in Table 2. The average of two homozygous phenotypes ($GH^{+/+} = P11$ and $GH^{-/-} = P_{22}$), denoted by *m*, was calculated by the equation [1, 29] as: $m = \frac{1}{2}$ ($P_{11} + P_{22}$).

The genotypic value, breeding value and dominance deviation for each genotype of the cows in this study were calculated with the formula [1] as follows: Genotypic value of the $GH^{+/+} = P_{11}$ (a) = $P_{11} - m$, Genotypic value of the $GH^{+/-} = P_{12}$ (d) = $P_{12} - m$ and Genotypic value of the $GH^{-/-} = P_{22}$ (– a) = $P_{22} - m$. Because *m* was defined as the phenotypic mean of both homozygous genotype animal groups, the genotypic values of each animal genotype of $GH^{+/+}$ (a), $GH^{+/-}$ (d) and $GH^{-/-}$ (– a) were found as presented in Table 2. These genotypic values were valuable in contribution for all phenotypic heterosis and genotypic trait parameters of breeding values, dominance gene action and heritability in the cow population.

2.3. Analysis of the Population Mean and Heterosis

In Hardy-Weinberg equilibrium for a population, the phenotypic population of the mean (μ) was defined[1, 29] as follows: $\mu = p^2 P_{11} + 2pqP_{12} + q^2P_{22}$. An alternative computing formula for obtaining the mean is based on substituting *m* + genotypic value for each phenotype. In equilibrium of a population, the mean is computed using Equation [1, 29] as follows: $\mu = p^2 (m + a) + 2pq (m + d) + q^2 (m - a)$, equal to *m* ($p^2 + 2pq + q^2$) + a ($p^2 - q^2$) + 2 pqd, equal to *m* + [a (p - q) + 2 pqd] (Table 3). The Standard Error of $\mu = \sigma / \sqrt{n}$ as described in [30].

The heterosis is also called hybrid vigour, the increase in such characteristics as size and growth rate of hybrid organism over of its parental yields [29]. Maximum heterosis is realized in the first cross of distinctly different parents. Heterosis and complementarity are powerful forces that combine to produce the total advantage of beef cattle crossbreeding [29, 31]. The Percent heterosis can be calculated (as: % heterosis = [(heterozygous phenotypic average – both homozygous phenotypic average] x 100 [31].

2.4. Analysis of the Breeding Value

The expected mean of the progeny of the homozygous male (μ_{11}) is the sum of the products of genotypic frequencies and corresponding phenotypic values, computed as: $\mu_{11} = P_{11} + P_{12} + P_{22}$, equal to p(m + a) + q(m + d) + 0(m - a), equal to m + pa + qd as described in [1, 29]. The breeding value of the homozygous male (BV₁₁) is twice the deviation of his progeny mean from the population mean and computed using equation as : BV₁₁ = 2 ($\mu_{11} - \mu$), equal to 2q[a + d(q - p)] as described in [1]. Likewise, computation using equation in [1]; the breeding values of the heterozygous male (BV₁₂) and homozygous male (BV₂₂) are as follows: BV₁₂ = 2 ($\mu_{12} - \mu$), equal to (q – p) [a + d (q – p)] and BV₂₂ = 2 ($\mu_{22} - \mu$), equal to – 2p[a + d (q – p)], as all presented in Table 3.

2.5. Analysis of the Dominance Gene Action

The difference between the genotypic value (V_{ij}) and the breeding value (BV_{ij}) for each genotype can be represented by the dominance deviation gene action (D_{ij}) using equation as: $V_{11} - BV_{11} = a - 2q[a + d (q - p)]$, equal to $a(p - q) + 2pqd - 2q^2d$; $V_{12} - BV_{12} = d - (q - p)[a + d(q - p)]$, equal to a(p - q) + 2pqd + 2pqd; and $V_{22} - BV_{22} = -a - (-2p[a + d(q - p)]$, equal to $a(p - q) + 2pqd - 2p^2d$ as described in [1, 29].

The equations of calculating genotypic value, breeding value and dominance deviation were summarized in Table 3. The phenotype of an animal (P_{ij}) may now be written as $P_{ij} = m + [a(p - q) + 2pqd] + BV_{ij} + D_{ij}$. Because m + [a(p - q) + 2pqd] is μ , the phenotype is represented as $P_{ij} = \mu + BV_{ij} + D_{ij}$. Therefore, $P_{11} = \mu + BV_{11} + D_{11}$; $P_{12} = \mu + BV_{12} + D_{12}$ and $P_{22} = \mu + BV_{22} + D_{22}$ as described in [1].

2.6. Analysis of the Heritability

Heritability is an extremely important population parameter that is used both for the estimation of breeding values for quantitative characteristics and for predicting the response expected from various selection schemes [29]. The phenotypic variance (σ_P^2) is calculated by equation as: $\sigma_P^2 = 2pq [a + d(q - p)]^2 + (2pqd)^2$ as described in [1]. The additive genetic variance (σ_A^2) for a single locus, is calculated as: $\sigma_A^2 = 2pq [a + d(q - p)]^2$ as described in [1]. The dominance genetic variance (σ_D^2) for a single locus, is calculated as: $\sigma_D^2 = (2pqd)^2$ as described in [1, 29].

Heritability in the narrow sense (h²) is defined as the ratio of the additive genetic variance to the phenotypic variance as: $h^2 = \frac{\sigma_A^2}{\sigma_p^2}$ as described in [1, 29]. Furthermore, the heritability in the dominance genetic effect (h_D^2) is defined as the ratio of the dominance genetic variance to the phenotypic variance as: $h_D^2 = \frac{\sigma_D^2}{\sigma_D^2}$ as described in [29, 32].

3. Results

3.1. Genotypic Value of Animal Live Weight

The growth hormone (GH) genotypes using restricted enzyme of *Msp1* for 78 cows were applied in this study. The PCR-RFLP data were used in establishing the observed homozygous $GH^{+/+}$ genotype, heterozygous $GH^{+/-}$ genotype and homozygous $GH^{-/-}$ genotype (Table 1). The 78 genotyped parental cows showed that a total of 27 cows were detected to have homozygous genotype of the GH^{-/-} in GH locus, a total of 27 cows were detected to have heterozygous genotype of the $GH^{+/-}$ in GH locus, and a total of 24 cows were detected to have homozygous genotype of the $GH^{+/+}$ in GH locus (Table 2).

Table 2 Average of live weight and genotypic value for each genotype GH restriction enzyme Msp1 in Ongole-gradecows

Cow Geno- types	Cow numbers (n)	Genotype Frequen- cies	Gene Frequen- cies	Average of cow weight (kg) in the genotypic groups	Genotypic Value (Vij) in kg
GH +/+	24	p2	p = 0.48	P11= 381.34±21.63a	a = 1.315
GH +/-	27	2pq		P12= 392.06±22.82b	d = 12.035
GH-/-	27	q2	q = 0.52	P22= 378.71±21.37a	– a = – 1.315

^{a,b)} superscripts in the same column indicated the significantly difference (P<0.05). Genotypic values (V_{ij}) were derived from the Equation by Van Vleck et al (1987). Genotypic value (V₁₁) of $a = P_{11} - m$; genotypic value (V₁₂) of $d = P_{12} - m$; genotypic value (V₂₂) of $-a = P_{22} - m$. The value of $m = \frac{1}{2}$ (P₁₁ + P₂₂). Thus, the value of m = 380.025 kg. The number of cows and the average and standard deviation (σ) of the phenotypic live weight of cow population for this study were presented in Table 2.

3.2. Population Mean and Heterosis of Animal Live Weight

Table 3 The different genotypic values by cow measurement, population mean and their genotypic components predicted by the equations described by Van Vleck et al (1987)

Genotypic group (G _{ij}) of cows	Population mean (μ) by formula (kg) of cow live weight			Breeding Value (BV _{ij}) by Formula (kg) of cow live weight		Dominance Effect (D _{ij}) by Formula (kg) of cow live weight
GH +/+	$\mu = m + [a(p - q) + 2 pqd]$			BV ₁₁ = 2q [a + d(q – p)]		$D_{11} = -2q^2d$
GH +/-	$\mu = m + [a(p - q) + b(p - q)]$		+ 2 pqd]	BV ₁₂ = (q-p) [a+d (q-p)]		$D_{12} = 2pqd$
GH-/-	μ=	<i>m</i> + [a(p – q) ·	+ 2 pqd]	BV ₂₂ = – 2p [a+d (q–p)]		$D_{22} = -2p^2d$
Phenotypic values (P _{ij})	Gene Frequency		values of μ_{ij}		Values of BV _{ij}	Values of D _{ij}
P ₁₁ = 381.34	p ² p = 0,48		µ11 =385.98±4.41		BV ₁₁ = 1.87	D ₁₁ = - 6.51
P ₁₂ = 392.06	2pq		μ ₁₂ =385.9	98±4.39	BV ₁₂ = 0.07	D ₁₂ = 6.01
P ₂₂ = 378.71	q^2	q = 0,52	μ22 =385.9	98±4.10	BV ₂₂ = - 1.72	D ₂₂ = - 5.55
P _{ij Average} =	= 384.04	μ = 385.9		8± 2.49	BV _{ij Average} = 0.07	D _{ij Average} = - 2.01
The components of var	riance	$\sigma_P^2 = 3$	$\sigma_{P}^{2} = 37.7$		$\sigma_{A}^{2} = 1.61$	$\sigma_{D}^{2} = 36.09$
The values of heritability				$h_{A}^{2} = 0.04$		$h_D^2 = 0.96$
Phenotypic average of the homozygous genotypic cow groups (m) = 380.025 kg				Component of the heterosis = 12.035 kg		Percent of the heterosis = 3.2

Using a total of 78 samples of the cows with 24 cows of the $GH^{+/+}$ genotypes and 27 cows of the $GH^{+/-}$ genotypes in this study, the allele frequency of GH- $Msp1^+$ (p) was $75/_{156} = 0.48$. Because p = 0.48, the allele frequency of GH^- (q) was 0.52. Therefore, the population means of the cows (μ) using the previous equation was 385.98± 2.49 kg. This value indicated that live weight mean of the cow population in this study would be about 385.98± 2.49 kg (Table 3).

In order to obtain a stable natural equilibrium known as the ecological balance (The Hardy-Weinberg Principle) for each change of the animal generation, the animal mating system would be applied using all genotypes of cows with superior live weight of bulls of Krista (*Kr*-*GH*^{+/+}) and Tunggul (*Tu*-*GH*^{-/-}) to spread gene frequencies of *GH*⁺ (p = 0.50) and *GH*⁻ (q = 0.50) referring into a stable natural equilibrium known as the ecological balance (The Hardy-Weinberg Principle). If all animals with genotypes of *GH*^{-/-} were culled in the population, then the allele frequency of *GH*⁺ (p₁) existing in the population would be $p_1 = 1/(1+q) = 2/3$. In this way, the ratio of animal genotypes existing in the population consisted of only $1(GH^{+/+}) : 2(GH^{+/-})$; while $1(GH^{-/-})$ was culled without breeding in the population. Therefore, the survival genes existing were 4 (*GH*⁺) and 2(*GH*⁻). Consequently, allele proportion of *GH*⁺ on the next generation (p₁) would be 4/6 = 2/3, and allele proportion of *GH*⁻ (q₁) would be 2/6 = 1/3. In the same strategy, the ratio of animal genotypes existing in the population. Theselection differential was defined as the superiority of the selected parental cows over the population mean that would be reached the genetic development of animal population [33, 34].

3.3. Breeding Value, Dominance Deviation and Heterosis of Animal Live Weight

Breeding value was a function of gene frequency and genotype values. Gene frequency could differ from one generation to the next generation; likewise, the breeding value was depended on the gene frequency. Live weight of cows in this study (Table 2), the allele frequency of GH^+ was 0,48 resulted from allel of $p = \frac{75}{156}$. Thus, the genotype frequency of the animals in the population would be 0.23 for genotype $GH^{+/+}$, 0.50 for genotype of $GH^{+/-}$, and 0.27 for genotype of $GH^{-/-}$. Using the previous equation, the breeding value (BV₁₁) of homozygous genotype of $GH^{+/+}$ was 1.87 kg, the BV₁₂ of heterozygous genotype of $GH^{+/-}$ was 0.07 kg, and the BV₂₂ of homozygous genotype of $GH^{-/-}$ was – 1.72 kg (Table 3).

Furthermore, the dominance deviation (D₁₁) of homozygous genotype of $GH^{+/+}$ was – 6.51 kg, the D₁₂ of heterozygous genotype of $GH^{+/-}$ was 6.01 kg, and the D₂₂ of homozygous genotype of $GH^{-/-}$ was – 5.55 kg (Table 3). The breeding values and dominance deviation gene action were considered at the equilibrium under the existing of genotype frequency for phenotypic trait measurements to form population mean (μ) as shown in Table 3. The critical components were genetic development of local grade cattle breeds by choosing small proportion of 10% truncation point for intensification of selected elite cow groups among animal populations with the positive live weight gains [34].

3.4. Variance and Heritability of Animal Live Weight

The population mean is the phenotype average. The real observation varied in term of mean. Variation of observation for mean could be calculated in term of variance. In this study, the variance is denoted (6^2_p) to represent phenotype variance. The value of phenotype variance (6^2_p) in this study were 37.7 kg. The variance of additive genetic values (σ_A^2) was 1.61 kg. Therefore, the heritability (h_A^2) of animal live weight was 0.04, indicating low heritability of this animal live weight trait (Table 3). Moreover, the variance of dominance genetic values (σ_D^2) was 36.09 kg. Consequently, the heritability (h_D^2) of animal live weight was 0.96, indicating the highest heritability of this animal live weight trait without nongenetic causes of variation have been considered. This case agreed with the previous study in [9].

The highest value of heritability (h_D^2) in this study was indicating the highest roles of the heterozygous genotype effect of $GH^{+/-}$ contributing markedly higher average body weights of the animals showing hybrid vigor or heterosis, as presented in Table 3. This case revealed the potential contribution of the growth hormone (*GH*) for body weight inheritance of the animals due to a co-dominance action of both additive gene action resulting a dominance deviation effect of the *GH*⁺ and *GH*⁻ (Table 3). The dominance deviation effect is the ability of a dominant gene to express itself in a phenotypic trait, when the gene is paired with another (recessive) gene that would have expressed itself in a different way as shown in the result of the electrophoresis analysis (Figure 1) visualizing the three genotypic bands of the growth hormone (*GH*) genes [20, 35, 36, 37].

4. Discussion

In this study, live weight was affected by the genotype at the locus of *GH*. The animal population was considered at the equilibrium with existing of gene frequency and phenotypic measurements as shown in Table 3. The *GH* $^+$ represented allele affecting animal live weight. Cow genotype represented each animal phenotype performance measured in the kg

unit of cow body weight. Genotype value was defined as deviation of phenotype from the mean of both homozygous phenotypes of P_{11} and P_{22} [29].

The heterosis by mating of bull called Krista with the genotype analysis using restricted enzyme of Msp1 as genotype Kr- $GH^{+/+}$ and bull called Tunggul with the genotype analysis using restricted enzyme of Msp1 as genotype Tu- $GH^{-/-}$ could be applied by mating system for the heterozygous genotypes of cows with the heterosis of 12.035 kg or 3.2 percent in this this generation with the equilibrium gene frequency from GH- $Msp1^+$ (p) of 0.42 and q of 0.52 in the population mean as shown in Table 3. The BNT statistical test showed that the heterosis of the population based on the heterozygous genotypic component of $GH^{+/-}$ prominent production of live weight could increase significantly (P<0.01) of 3.2 percent heterosis compared with weight average value (m) in both homozygous genotypic group of $GH^{+/+}$ and $GH^{-/-}$ in the Ongole Grade cow population.

The phenotype values of P₁₁, P₁₂, and P₂₂ were the averages of phenotype values of live weight as shown in the Table 3. These values indicated that the trait of live weight was more dominated by dominance gene action rather than the additive gene action in the homozygous genotypes of both $GH^{+/+}$ and $GH^{-/-}$ as indicated by the highest live weight values by the heterozygous genotype of $GH^{+/-}$ with 3.2 percentages of heterosis (12.035 kg). Breeding value of an individual is referred to its additive genetic merit [1, 38]. The difference between breeding values is additive and representing in term of heritability for certain animal economical trait such as animal live weight [1, 39]. The breeding values of cows in this study varied from – 1.72 to 1.87 kg, while those in temperate beef cows varied from –15.0 to 22.0 kg [4]. These values indicated that the Ongole breed cows and the temperate beef cows were included into various breeding values. The various breeding values would give chance for valuable genetic heterosis in productivity development by cross mating system and grading up program using the artificial insemination within local beef breeds in the Eastern part of Indonesia including North Sulawesi Province as the location of this study.

Based on the genotypic analysis (Figure 1) as described in Table 1 and the different values by cow measurement (Table 2 and Table 3), both alleles of GH^+ and GH^- could be categorized as the codominant alleles indicated by the three different genotypic component groups of $GH^{+/+}$, $GH^{-/-}$ and $GH^{+/-}$ in animals. Codominance of heterozygous $GH^{+/-}$ genotype was almost the same as the incomplete dominance, because there is no dominant allele. In contrast to incomplete dominance, the phenotypic performance trait of the cow population with the heterozygous $GH^{+/-}$ codominant genotype of live weight of this study was differently prominent in heterosis than those in the incomplete dominance as the average performance of both homozygous genotypes ($GH^{+/+}$ and $GH^{-/-}$) in animal live weight. If both alleles are codominant, then at the time they were heterozygous, their traits would appear in a different superior performance as hybrid vigor or heterosis of the offspring generations [25, 31].

5. Conclusion

Growth hormone (*GH*) genes defined by *Msp1*-enzyme restriction at intron 3 could be included into the potential DNA marker of *GH*⁺ and *GH*⁻ genes contributing the heterozygous codominant *GH*^{+/-} genotypic component in Ongole-grade cattle. The population mean (μ) of cow live weight was 385.98± 2.49 kg. The genotypic analysis showed that *GH*^{+/-} genotype performed the codominant prominent live weight compared with those of both homozygous genotypes of *GH*^{+/+} and *GH*^{-/-} resulting the valuable heterosis (hybrid vigor) for live weight trait of 3.2 percent or 12.035 kg. This investigation displayed those potential genes in regenerating the outstanding phenotypic performance of cattle live weight due to this heterozygous codominance effect of *GH*^{+/-} genotypic component. The heritability of additive gene action (h_A^2) was 0.04, while that of dominance gene action effect (h_D^2) was 0.96, included as the highest heritability under the equilibrium frequencies of both *GH*⁺ (p=0.48) and *GH*⁻ (q=0.52) genes without considering nongenetic causes of environmental effect in this cow population.

Compliance with ethical standards

Acknowledgments

The financial support of the Sam Ratulangi University through their Research Partnership Program is gratefully acknowledged. The authors also acknowledge Mr. Jan Kuhu for the assistance in animal data collection at the artificial insemination service center at Tumaratas village, West Langowan district of Minahasa regency, and Mr. Rizky for his assistance in animal data collection at Sangkub district of Bolmut regency, North Sulawesi province of Indonesia.

Disclosure of conflict of interest

The authors declared no conflict of interest.

Statement of informed consent

Informed consent was obtained from all individual participants included in the study.

References

- [1] Van Vleck LD., Pollak EJ and Oltnacu EAB. Genetics for the Animal Science. W.H. Freeman and Company, New York. 1987, Pp:181-197.
- [2] Thundathil JC, Dance AL and Kastelic JP. Fertility management of bulls to improve beef cattle productivity. Theriogenology. 2016, 86 (1): 397-405.
- [3] Chawala AR, Banos G, Komwihangilo DM, Peters A and Chagunda MGG. Phenotypic and genetic parameters for selected production and reproduction traits of Mpwapwa cattle in low input production systems. South Af. J. Anim. Sci. 2017, 47(3): 307-319.
- [4] Legates JE and Warwick EJ. Breeding and Improvement of Farm Animals. McGraw-Hill Publishing Company. New York. 1990.
- [5] Maki-Tanila A and Hill WG. Influence of gene interaction on complex trait variation with multilocus models. Genet. 2014, 198, 355–367.
- [6] Wulandari A, Nurgiartiningsih VMA, Kuswati K, Susilorini TE and Partogi PA. Kinship of several Indonesian local cattle by using DNA mitochondrial COI. Int. Res. J. Adv. Eng. Sci. 2019, 4 (3): 165-167.
- [7] Bycroft C, Freeman C, Petkova D, Band G, Elliott LT, Sharp K, Motyer A, Vukcevic D, Delaneau O and Connell JO. The UK Biobank resource with deep phenotyping and genomic data. Nature. 2018, 56 (2): 203–209.
- [8] Beauchemin VR, Thomas MG, Franke DE and Silver GA. Evolution of DNA polymorphisms involving growth hormone relative to growth and carcass characteristics in Brahman steers. Genet. and Mol. Res. 2006, 5 (3):438-447.
- [9] Hou K, Burch KS, Majumdar A, Shi H, Mancuso N, Sankararaman S and Pasaniuc B. Accurate estimation of snpheritability from biobank-scale data irrespective of genetic architecture. Nat. Genet. 2019, 51, 1244–1251.
- [10] Maylinda S. Genetic polymorphism of growth hormone locus and its association with body weight in Grati dairy cows. Int. J. Biotech. and Mol. Biol. Res. 2011, 2 (7):117-120.
- [11] Cano G, Blanco M, Casasus I, Cortes-Lacruz X and Villalba D. Comparison of B-splines and non-linear functions to describe growth patterns and predict mature weight of female beef cattle. Anim. Prod. Sci. 2016, 56: 2161-2161.
- [12] Sutarno A, Junaidi J and Tappa B. The MspI polymorphism at locus 2 of growth hormone gene in Ongole-grade cattle and its effect on daily weight gain. Biodiversity, 2005, 6 (No. 2): 77-81
- [13] Sodhi M, Mukesh M, Prakash B, Misha BP, Sobti RC and Singh KP. Msp1 allelic pattern of bovine gene in Indian zebu cattle (Bos indicus) breeds. Biochem. Genet. 2007, 45 (12):145-153.
- [14] Jakaria D, Duryadi D, Noor RR, Tappa B and Martojo H. Evaluation of growth hormone (GH) genetic diversity in West Sumatra coastal cattle using PCR-RFLP markers. Livestock Media. 2007, 30 (No.1):1-10
- [15] Jakaria R, Noor RR, Martojo H, Duryadi D and Tappa B. Identification of growth hormone (Gh) gene MspI and AluI loci polymorphism in beef cattle. Faculty of Animal Science, Bogor Agricultural University. The 1st International Seminar on Animal Industry. 2009, p.42-47.
- [16] Zhou X. A unified framework for variance component estimation with summary statistics in genome-wide association studies. Ann. Appl. Stat. 2017, 11: 2027–2051.
- [17] Golan D, Lander ES and Rosset S. Measuring missing heritability: inferring the contribution of common variants. Proc. Natl. Acad. Sci. USA. 2014, 111, 5272– 5281.
- [18] Visscher PM, Hemani G, Vinkhuyzen AAE, Chen GB, Lee SH, Wray NR, Goddard ME and Yang J. Statistical power to detect genetic (co) variance of complex traits using snp data in unrelated samples. Genet. 2014, 10, 42-69.
- [19] Evans LM, Tahmasbi R, Vrieze SL, Abecasis GR, Gazal S, Bjelland DW, de Candia TR, Goddard ME and Neale N. Comparison of methods that use whole genome data to estimate the heritability and genetic architecture of complex traits. Nat. Genet. 2018, 50, 737–745.

- [20] Paputungan U, Hakim L, Ciptadi G and Lapian HFN. The allele frequencies of growth hormone gene on the parental and progeny of Ongole-crossbred cattle population in the North Sulawesi of Indonesia using PCR-RFLP. Journal of Evolution and Biological Research. 2012, 4 (3):52-58.
- [21] Maharani D, Amrullah AHK, Widayati DT, Sumadi S, Fathoni A and Khusnudin M. Predicting the age and weight at puberty of Ongole grade cattle and using nonlinear mathematical model in Kebumen Farmer Association. J. Indonesian Trop. Anim. Agric. 2017, 42, 233-239.
- [22] Sulandari S and Zein MSA. Protocols in DNA Laboratory, Center of Biology Research. Laboratory Guideline Book. The Indonesian Institute of Sciences. 2003, Pp.23-45.
- [23] Dybus A. Associations of growth hormone (GH) and prolactin (PRL) genes polymorphism with milk production traits in Polish Black-and-White cattle. Anim. Sci. Papers Reports. 2002, 20(4):203-212.
- [24] Gordon DF, Quick DP, Ewin CR, Donelson JE and Maurer RR. Nucleotide sequences of the bovine growth hormone chromosomal gene. Mol. Cell Endocrinol. 1983, 33:81-95.
- [25] Rifa'i M. Genetika Rekombinasi dan Populasi. Edisi Pertama. Penerbit Galaxy Science, Malang, 65145. ISBN: 978-602-97628. 2010, 1-5.
- [26] Yi-Fan W, Zheng L, Niu H, Zhang GL, Zhang GM, Ma YL, Tian YR, Liu YR, Yang P, Yang DY, Lei CZ, Dang RH, Qi XL, Chen H, Huang BZ and Huang YZ. Exploring genotype-phenotype relationships of the CRABP2 gene on growth traits in beef cattle, Anim. Biotechnol. 2018, 31:1, 42-51.
- [27] Pradana IYW, Sampurna IP and Suatha IK. The growth of body weight dimensions of Bali calves. Bul. Vet. Udayana. 2014, 6 (1): 81-85.
- [28] Ratnasari D, Atabany A, Purwanto BP and Salman IB. Growth patterns of Holstein Friesian dairy cow (FH) from birth to first child based on mathematical analysis of the Gompertz model. Bullet. Anim. Sci. 2019, 43 (3): 184-187.
- [29] Pazokitoroudi A, Chiu AM, Burch KS, Pasaniuc B and Sankararaman S. Quantifying the contribution of dominance deviation effects to complex trait variation in biobank-scale data. The Am. J Human Genet. 2021, 108, 799–808.
- [30] Byrkit DR. Statistics Today: A Comprehensive Introduction. The Benyamin/ Cummings Publishing Company, Inc. Menlo Park, California. 1987.
- [31] Spangler ML. The value of heterosis in cow herds: Lessons from the past that apply to today. Proceedings, The Range Beef Cow Symposium XX December 11, 12 and 13, 2007 Fort Collins, Colorado. 2007.
- [32] Zhu Z, Anna AB, Vinkhuyzen AE, Hemani G, Lee SH, Nolte IM, van Vliet-Ostaptchouk JV, Snieder H, Study TLLC, Esko T, Milani L, Mägi R, Metspalu A, Hill WG, Weir BS, Goddard ME, Visscher PM and Yang J. Dominance Genetic Variation Contributes Little to the Missing Heritability for Human Complex Traits. Am. J. Hum. Genet. 2015, 96: 377–385.
- [33] Thekkoot D. Selection Intensity and Genetic Improvement. The University of Alberta and Genesus Inc. Animal breeding aims to improve livestock population by utilizing the genetic differences among individuals. 2017.
- [34] Paputungan U, Hendrik MJ and Siswosubroto SE. Comparison of the favorable gain values of genetic improvement among Indonesian grade cow breeds selected for agrotechnopark intensification. J. Indonesian Trop. Anim. Agric. 2021, 46 (2):106-113.
- [35] Tutkun M. Growth curve prediction of Holstein-Fresian bulls using different non-linear model functions. Appl. Ecol. Environ. Res. 2019, 17 (2): 4409- 4416.
- [36] Selvaggi M, Laudadio V, D'Alessandro AG, Dario C and Tufarelli V. Comparison on accuracy of different nonlinear models in predicting growth of Podolica bulls. Anim. Sci. J. 2017, 88 (8): 1128-1133.
- [37] Ashwini1 JP, Sanjay P, Amipara GJ, Lunagariya PM, Parmar DJ and Rank DN. Prediction of Body Weight based on Body Measurements in Crossbred Cattle. Int. J. Curr. Microbiol. App. Sci. 2019, 8 (3): 1597-1611.
- [38] Vitezica ZG, Varona L and Legarra A. On the additive and dominant variance and covariance of individuals within the genomic selection scope. Genet. 2013, 195, 1223–1230.
- [39] Gazal S, Loh PR, Finucane HK, Ganna A, Schoech A, Sunyaev S and Price AL. Functional architecture of lowfrequency variants highlights strength of negative selection across coding and non-coding annotations. Nat. Genet. 2018, 50: 1600–1607.