



HSP90AA1 Gene Polymorphism and its Relationship with Reproductive and Productive Traits in Holstein Cattle

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Abstract

THE *HSP90AA1* gene, as a member of heat shock protein family has a crucial role in cellular stress responses, particularly in thermotolerance and cattle fertility. Our study aims to determine the *HSP90AA1* gene polymorphism and its relationship with cattle fertility and production. A total number of 305 cows were categorized into 3 groups (cyclic, post-partum anestrus (PPA) from 6 to 12 months, and PPA>12 months). Data on reproduction and production were gathered from farm records, including age at first calving, calving intervals, calf birth weights, milk yields, and milking days. Genomic DNA extraction and PCR-SSCP were conducted to detect three fragments of *HSP90AA1* gene F1, F2, and F3 at 285, 355, and 280 bp, respectively. Three different patterns were observed in *HSP90AA1* F1, and F3. *HSP90AA1* F2 was monomorphic. Sequence analysis of *HSP90AA1* F1 showed 3 different genotypes (CC, AA, and AC) with single nucleotide polymorphisms (SNPs) (A/C) as a transverse single base substitution mutation located at 21:66942714 with ID rs207763220 as a synonymous mutation. While *HSP90AA1* F3 revealed 3 genotypes (TT, GG, and TG) with only one missense SNP located at 21: 66941688 (T/A/G) with ID rs210718366. In the *HSP90AA1* F1 gene, the CC genotype was linked to the lower calving interval and milk yield in cows with a PPA >12. In the *HSP90AA1* F3 gene, the GG genotype was related to lower days of milking and milk yield. Additionally, the TT genotype was correlated with lower age at first calving in the PPA > 12. Our findings suggest that genetic variations in the *HSP90AA1* gene may be used as a molecular marker for genetic selection to enhance reproductive and productive performance in Holstein cows, especially age at first calving, calving interval, and milk yield associated with post-partum anestrus.

Keywords: *HSP90AA1* gene, Polymorphism, SNPs, PPA, Fertility, cattle.

Introduction

The postpartum period is a critical phase for cattle reproductive performance and economic productivity, marked by transient sexual inactivity and incomplete ovarian maturation. The duration of this interval is influenced by multiple factors, necessitating the implementation of management strategies, hormonal interventions, and genetic approaches to expedite reproductive recovery. Selective breeding programs have traditionally aimed to enhance milk yield and improve functional traits.

The integration of measurable traits, including molecular markers associated with reproductive characteristics, has emerged as an advanced approach for improving fertility in cattle through genetic enhancement [1-3].

Thermal stressors significantly impact livestock activities, particularly when animals struggle to dissipate excess heat, leading to decreased production. In tropical, subtropical, and arid regions, high environmental temperatures and heat stress disrupt hormonal balance, particularly luteinizing

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hormone and progesterone, which are essential for successful reproduction. Consequently, excessive heat stress during the summer season has been associated with observable declines in milk production, reproductive performance, and overall fitness in cattle, ultimately affecting the livestock industry [4,5]. Different cattle breeds respond uniquely to heat stress, with Holstein Friesians being particularly vulnerable [6-8]. Animals express different genes from the heat shock proteins (HSPs) family to cope with various stressors. These proteins are essential for enhancing cattle resilience to thermal stress [9-11].

These highly conserved proteins serve as potential biomarkers for heat stress and are classified by their molecular weight into HSP40, HSP60, HSP70, HSP90, and HSP100. Among members of HSP family, HSP70 and HSP90 are the most prevalent in eukaryotic cells, providing protection against heat and other stressors [12]. HSP90 is crucial for cellular adaptation to thermal stress, constituting 1-2% of cellular proteins, which can increase to 4-6% under stress conditions. This highlights its essential role in cellular function. HSP90 exists in two isoforms: Hsp90 α (HSP90AA1), which is inducible, and Hsp90 β (HSP90AB1), which is constitutive. Both isoforms result from gene duplication. The HSP90 α gene is found on chromosome 21 including 12 exons and 11 introns [13, 14].

Heat shock protein family genes are typically highly conserved, yet studies have identified potential polymorphisms [15, 16]. Research has shown that these polymorphisms are associated with the ability to tolerate various stress conditions, particularly high temperatures, as gene expression tends to increase in response to heat [17]. Such polymorphisms, arising from several mutations, can significantly influence the structure and function of the resulting protein, leading to either beneficial or detrimental effects [18]. The understanding of these genetic variations is crucial for enhancing stress tolerance in cattle [19]. Polymorphisms linked to HSP have also been found in exons 3 and 8 of HSP90AA1 gene in Karan Fries and Sahiwal cattle [19-21].

Numerous studies in cattle have recorded increased HSP90AA1 gene expression under heat stress [22-24]. For instance, Sahiwal cattle demonstrated higher expression in HSP90AA1 mRNA compared to Frieswal cattle, suggesting that

genetic factors influence HSP90AA1 expression and HS response [25]. HSP90AA1 gene in Deoni cattle has been identified as polymorphic, showing significant associations with reproductive and productive traits, such as calving interval and age at first calving [26-28]. The relationship between Hsp90AA1 gene variants and their impact on reproductive and productive traits in Egyptian Holstein cattle remains largely unexplored. To our knowledge, there has been limited research on HSP genes in livestock, particularly cattle, within Egypt. Therefore, our study aims to investigate SNP polymorphisms in the Hsp90AA1 gene and assess their effects on various phenotypic traits, including calving interval, age at first calving, calf birth weight, days of milking and milk yield in Holstein cows.

Material and Methods

Ethics statement

All procedures performed for cows were under the Faculty of Veterinary Medicine, Benha University ethical standards with a number of BUFVTM 14-09-24.

Animals and samples

A total number of 305 Holstein Friesian cows were subjected to rectal and ultrasonographic examinations using a B-mode scanner (Magic 2200, Eickemeyer Veterinary Equipment Inc., Germany) monthly to determine the animal's reproductive status. The cows were raised in stable weather conditions, with an average temperature of 91°F. They were housed in barns and had constant access to water and grass, in addition to being fed about 7.7 pounds of commercial concentrate each day. Cows were classified into three categories: cyclic, post-partum anestrus from 6 to 12 months (PPA 6-12), and more than 12 months (PPA>12). A comprehensive case history was recorded for all studied cows, including age at first calving, calving interval, calf birth weight, days of milking and milk yield. Blood samples were collected through the jugular vein in vacutainer tubes with EDTA then stored at -20 °C for genomic DNA isolation.

DNA isolation and PCR amplification

DNA was isolated from blood using genomic DNA isolation kit (Blood Cultured Cell, Taoyuan, Taiwan). DNA quantity and quality were determined by Nano-Drop 1000 spectrophotometer; Thermo Scientific, Wilmington, DE. PCR amplification was

carried out in Bio-Rad thermal cycler (T 100, USA) using three sets of primers (F: 5' - TGTCTTCAACTGAAAAATGCTGT -3' and R: 5' - CTGGACCAGAAACCCTGATGA -3') for fragment 1 of the *HSP90 AA1* gene, (F: 5' - ACGCCTTCCCCCATGTTTAG -3' and R: 5' - GCCCGGAGTCATTGTTAGGT -3') for fragment 2 of the *HSP90 AA1* gene, and (F: 5' - TCACCTAAGAAGCCAAAGTTTTAGA -3' and R: 5' - AGCTTTCAGAGCTGTTGAGGTA -3') for fragment 3 of the *HSP90 AA1* gene, which were designed using Primer3 Program version 4.0 (<https://primer3.ut.ee/>).

For PCR reaction, 2.5 µl of buffer 10x, 0.25 µl of 2.5 mM (dNTPs mixture), 0.125 µl of primer, 0.15 µl of Taq polymerase (5U/µl), 1.5 µl of 25 mM (MgCl₂), 17.85 µl of nuclease-free water, and 2.5 µl of DNA were all used to prepare the amplification processes in a 25 µl volume. For the *HSP90 AA1* gene fragments 1, 2, and 3, the PCR settings were one cycle at 95 °C for 4 minutes, followed by 33 cycles at 94 °C for 30 seconds, 62 °C for 30 seconds, and 72 °C for 30 seconds, followed by a final extension of 72 °C for 7 minutes to produce 285, 355, and 280 bp PCR products, respectively. PCR amplicons were electrophoresed in 1.8% agarose gels using 0.75 X TBE buffer, visualized, and photographed by Gel-Doc System (Bio-Rad, USA).

Single-strand conformation polymorphism (SSCP)

The 5.00 µl PCR products were mixed with 45 µl of denaturing solution (98% formamide; 20mM EDTA, pH 8.0; 0.05% bromophenol blue; 0.05% xylene cyanol and 28 µl ddH₂O). At this stage, the samples were denaturated by heating them to 95°C for 8 minutes, after which they were placed on ice for 5 minutes and loaded onto an 8% polyacrylamide gel in 0.75 x TBE buffer. This solution contains the following ingredients: 29:1 acrylamide: bis-acrylamide, 10 ml of 1x TBE buffer (Tris-base, Boric acid, Na₂EDTA), 2.5 ml of glycerol, 17.5 ml of deionized water, 40µl of TEMED (N, N, N', N'-Tetramethyl ethylene diamine), and 400 µl of APS solution (ammonium persulfate) [29-31]. Electrophoresis was done at 4°C, 130V, 60 mA for 9 hrs then 90V, 50 mA for 6 hrs, and finally 70V, 40 mA for 4 hrs. Ethidium bromide (0.5µg/ml) staining technique was utilized to visualize the separated DNA fragments with high sensitivity on polyacrylamide gels and then photographed by Gel-Doc System.

Sequencing analysis

The PCR product was purified by QIAquick PCR purification kit from QIAGEN, to isolate DNA fragments from PCR reactions before genetic sequencing. The PCR products giving distinct SSCP band patterns were analyzed using direct sequencing by Macrogen Incorporation (Seoul, South Korea). Sequence data were examined, and the NCBI/BLAST/blastn suite was used for alignment. Codon Code Aligner and BioEdit software were used to further analyze the sequencing data in order to align it and find SNPs.

Statistical analysis

The Xlstat software, a general linear model, was used to evaluate the data as two-way analysis of variance [32]. The postpartum period and genotype were the main influences. The analyzed traits were age at first calving, calving interval, calf birth weight, days of milking and milk yield. This model was employed:

$$Y_{klm} = \mu + J_k + G_l + JG_{kl} + e_{klm}$$

Where:

Y_{klm} : The m^{th} observation of the l^{th} genotype within the k^{th} post-partum period.

μ : The total mean.

J_k : The effect of the k^{th} post-partum period.

G_l : The effect of the l^{th} genotype.

JG_{kl} : The interaction between the k^{th} post-partum period and the l^{th} genotype

e_{klm} : The random error.

Least square means (LSM) $\pm$ standard errors (SE) were used to report all data. When there was significance, mean values were evaluated by Duncan's multiple range test [33]. Five percent was chosen as the significance level.

Results

The polymorphism in sequences of *HSP 90AA1* gene fragments F1, F2, and F3 was determined in the genomic DNA of 305 cows using PCR-SSCP. The PCR amplification had been detected at 285, 355, and 280 bp, respectively (Fig. 1). The PCR-SSCP showed 3 different patterns in both *HSP 90AA1* F1 and F3 while F2 revealed a monomorphic pattern (Fig. 2). The sequence analysis of the different patterns of F1 confirmed our results and the sequence of the amplicon showed 3 genotypes (CC, AA, and AC) with a SNP as transverse single base substitution mutation located at 21:66942714 (A/C) with identification number (ID) rs207763220 as a synonymous mutation (Fig. 3, 4). While the F3

showed 3 genotypes (TT, GG, and TG) with only one missense SNP located at 21: 66941688 (T/A/G) with ID rs210718366 (Fig. 3, 5).

In all cow groups, including cyclic, PPA 6-12 months, and PPA >12 months, the CC genotype was the least frequent, occurring in only 14% of the animals. The AC genotype had a frequency of 46%, while the AA genotype was found in 40% of the samples in the *HSP90AA1* fragment 1. This indicates that allele A was more dominant than allele C (Table 1). In fragment 3, the frequencies for the genotypes were 51% for TT, 46% for TG, and 3% for GG, demonstrating that allele T was more dominant than allele G (Table 2). Additionally, the Holstein Friesian cattle populations were within equilibrium of Hardy-Weinberg, as determined by a chi-square test. The genotype and allele frequencies, along with the Hardy-Weinberg equilibrium findings, are presented in (Tables 1 and 2).

Concerning the studied reproductive and productive traits, calving interval, days of milking and milk yield were significantly increased in PPA > 12 months and PPA 6-12 months than cyclic animals (Tables 3 and 4). Moreover, age at first calving and calf birth weight significantly decreased at PPA > 12 months than cyclic animals. There was interaction between the post-partum anestrus and the genotypes.

In the *HSP90AA1* F1 gene, the CC genotype was linked to the short calving interval and lower milk yield in cows with a PPA >12 (Table 3). In the *HSP90AA1* F3 gene, the GG genotype was related to lower days of milking and milk yield. Additionally, the TT genotype was correlated with lower age at first calving in the PPA > 12 (Table 4).

Discussion

Bovines are particularly vulnerable to heat stress when raised in environments characterized by high humidity and temperatures. The physiological disruptions caused by heat stress can lead to significant declines in milk production, reproductive performance, and immune function, resulting in substantial economic losses and welfare challenges within the livestock sector [34-37]. Polymorphisms in the *HSP90AA1* gene are linked to thermotolerance and may serve as molecular markers to advance breeding strategies that enhance heat stress resilience in cattle [13, 19, 20, 24, 38, 39]. However, the genotyping of the *HSP90AA1* gene and the impact of its genetic variations on reproductive and productive traits remain inadequately explored. Our study aimed to determine SNP polymorphisms in the *HSP90AA1* gene and their association with calving interval, age at first calving, calf weight, milking duration and milk yield in Holstein cows.

In our study, two SNP loci were detected: A177C in the intron between exons 4 and 5, and T144G in the intron between exons 7 and 8. Hu et al. [39] discovered five SNPs in the *HSP90AA1* gene linked to heat tolerance in Holstein cows, which affects the binding activity of bta-miR-1224. While our study did not identify this specific SNP, the presence of other intronic variations suggests potential regulatory roles influencing productive and reproductive traits. The differences in SNP locations could be attributed to breed-specific genetic diversity or environmental selection pressures.

Abbaya et al. [40] identified six polymorphic sites (G136A, C89G, G136A, C89G, A86G, and A86G) in the Adamawa Gudali cattle, delineating four haplotypes with greater genetic variation among breeds (58.18%) than within breeds (41.82%). Similarly, Onasanya et al. [41] identified four SNPs (T116G, G220C, G346A, and G390A) in *HSP90* gene of Nigerian Zebu cattle, linking heterozygous genotypes within exon 3 to lower body temperature, rectal temperature, respiratory rate, and heat tolerance. While our study focused on Holstein cows and identified different SNPs, the presence of genetic variability across breeds aligns with previous findings, supporting the role of *HSP90AA1* polymorphisms in adaptive responses.

In terms of productive traits, our study found that the CC genotype of the *HSP90AA1* F1 gene was linked to lower milk yield in cows with a PPA >12, whereas the GG genotype of *HSP90AA1* F3 gene was linked with reduced milking days and milk yield in cyclic animals. This result is in partial agreement with Lamb et al. [42], who reported that a base change at G2033C in the *HSP90A1A* gene led to a glycine-to-alanine substitution, affecting milk yield and composition in Chinese Holstein cattle. However, Adzdzakiy et al. [43] found no association between *HSP90AB1* genotypes and milk production in Friesian Holstein populations, though the CC genotype exhibited superior milk production. The discrepancy may stem from differences in genetic markers studied, breed variations, or environmental influences affecting milk productivity.

Regarding reproductive traits, our study demonstrated that the CC genotype of the *HSP90AA1* F1 gene was linked to the shortest calving interval in cows with a PPA >12, and the TT genotype of the *HSP90AA1* F3 gene was associated with a lower age at first calving. [21, 28, 44] identified multiple SSCP genotypes within the *HSP90AA1* gene, suggesting genetic variation influences reproductive traits, including age at first calving and calving interval in Deoni cattle. Shergojry et al. [26] found that TG heterozygous cows had a higher age at first calving than

homozygous genotypes, but no significant differences in calving interval were observed. Our findings align with these studies in that *HSP90AA1* polymorphisms play a role in reproductive traits; however, breed differences may account for variations in specific SNP effects.

Additionally, Rosenkrans et al. [45] identified 11 SNPs in crossbred Brahman cows, including a deletion at position 895, which significantly impacted calving dates, with cows carrying the deletion calving 35 days later than those without it. The association of the AB genotype of the *HSP90AA1* gene with a longer service period suggests genetic factors influence reproductive performance. This supports the notion that while *HSP90AA1* may not directly affect physiological traits, it likely plays a crucial role in reproductive efficiency [21]. The complexity of genetic influences on dairy cattle performance highlights the necessity of considering both genetic variations and environmental conditions in future breeding programs.

Conclusion

In conclusion, the identification of SNP polymorphisms in the *HSP90AA1* gene correlated

with age at first calving, calving interval, and milk yield in cows with prolonged post-partum anestrus provides valuable genetic markers for selective breeding. These findings offer a foundation for improving reproductive efficiency and heat stress resilience in Holstein cattle, contributing to more sustainable and productive dairy farming.

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Funding statement

This study didn't receive any funding support.

Declaration of Conflict of Interest

The authors declare that there is no conflict of interest.

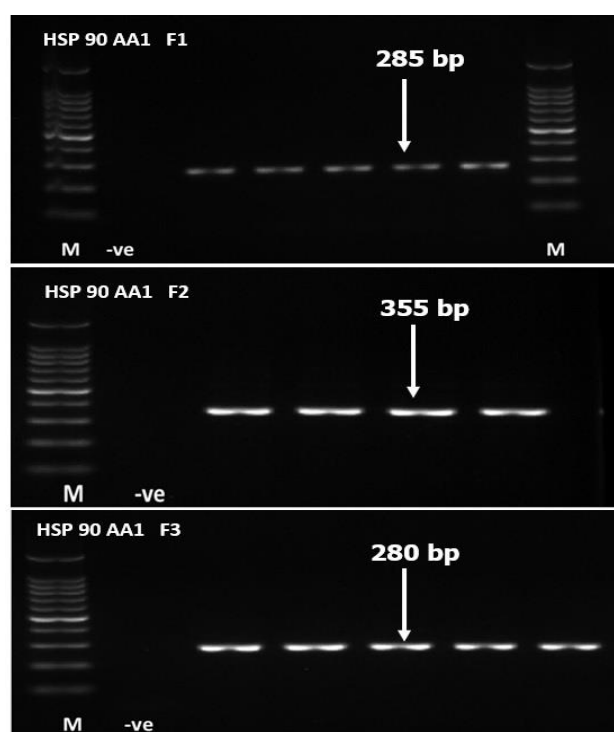


Fig. 1. Gel stained by ethidium bromide of PCR amplification for 3 fragments of *HSP90AA1* gene. PCR product of 285, 355, and 280 bp, -ve: control negative and M: 100 -bp ladder marker.

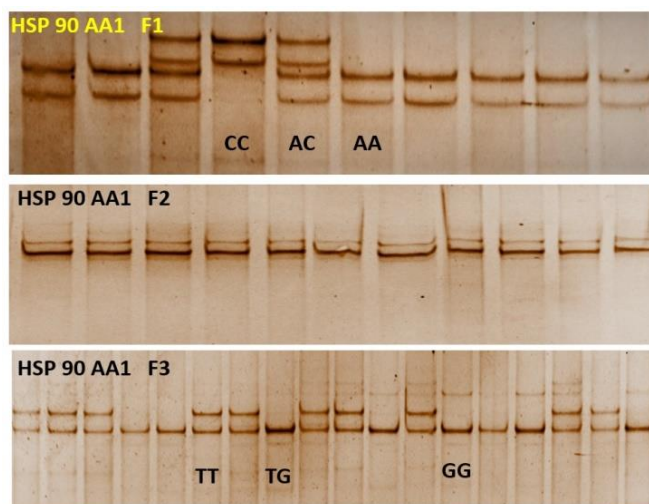


Fig. 2. Ethidium bromide-stained 11% polyacrylamide gel of HSP90 AA1 three primers PCR products of 3 different SSCP patterns in HSP90AA1 F1, monomorphic HSP90 AA1 F2, and 3 SSCP patterns in HSP 90 AA1 F3.

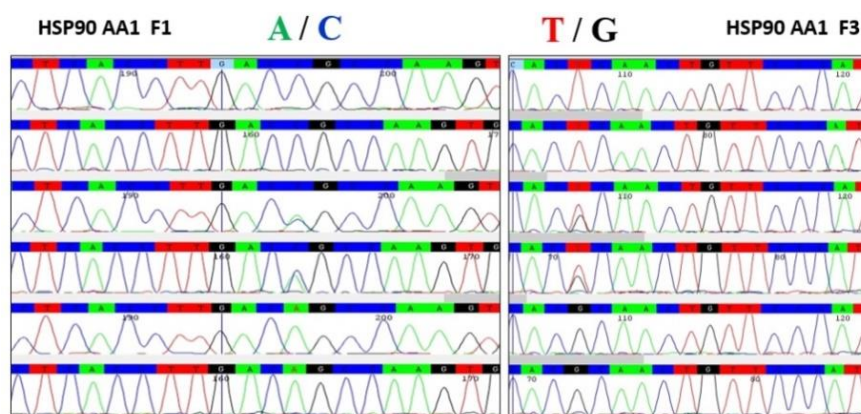


Fig. 3. Sequence analysis of the *HSP90 AA1* gene with 2 SNPs as transverse single base substitution mutation; A/C, T/G.

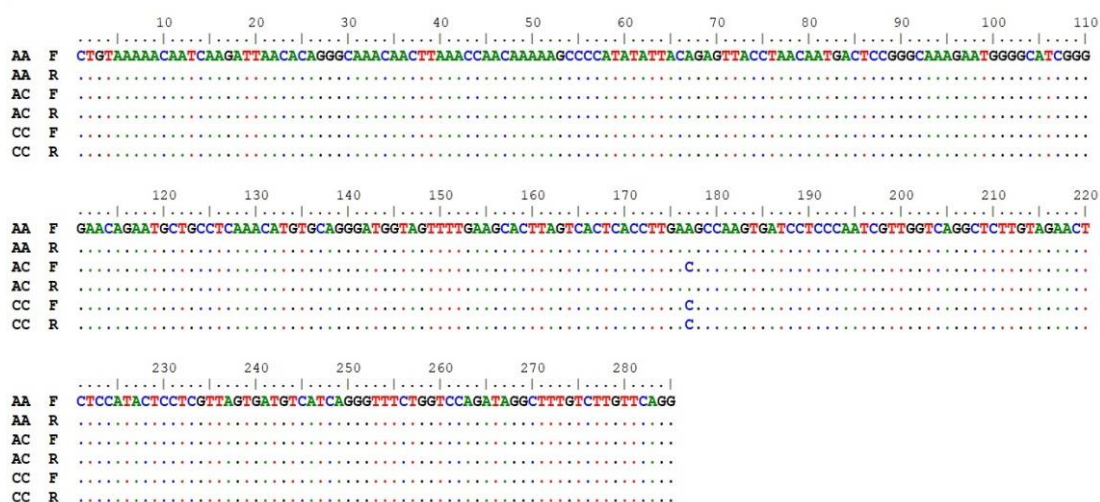


Fig. 4. *HSP 90 AA1* F1 gene sequence alignment of different genotypes.

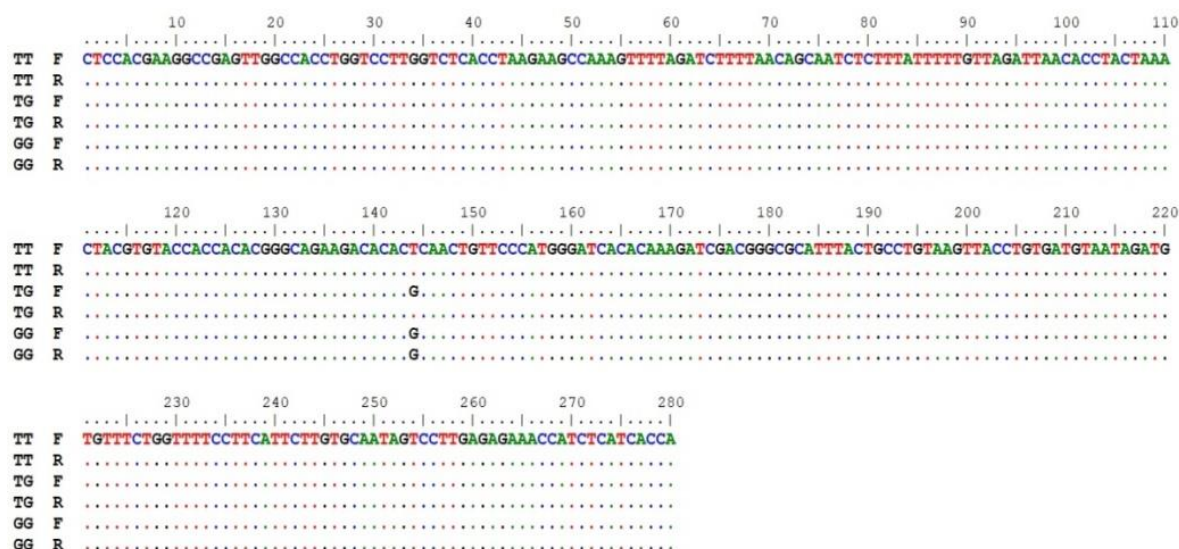


Fig. 5. *HSP 90 AA1 F3* gene sequence alignment of different genotypes.

TABLE 1. Genotypes and alleles frequency of *HSP 90 AA1 F1* gene in cattle

Status	No	Observed Genotype			Expected Genotype			Allele Frequencies			Hardy-Weinberg equilibrium
		AA	AC	CC	AA	AC	CC	A	C	χ^2 test	
Cyclic	150	0.40	0.40	0.20	0.36	0.48	0.16	0.60	0.40	16.94	0.0001
PPA 6-12	90	0.39	0.50	0.11	0.40	0.47	0.13	0.64	0.36	31.25	0.0001
PPA >12	65	0.38	0.54	0.08	0.42	0.45	0.13	0.65	0.35	38.83	0.0001
Total	305	0.40	0.46	0.14	0.39	0.47	0.14	0.62	0.38	7.13	0.0076

TABLE 2. Genotypes and alleles frequency of *HSP 90 AA1 F3* gene in cattle

Status	No.	Observed Genotyp			Expected Genotype			Allele Frequencies		Hardy-Weinberg equilibrium	
		TT	TG	GG	TT	TG	GG	T	G	χ^2 test	P value
Cyclic	150	0.53	0.40	0.07	0.52	0.40	0.08	0.73	0.27	17.26	0.0001
PPA 6-12	90	0.50	0.50	0.00	0.56	0.37	0.07	0.75	0.25	33.55	0.0001
PPA >12	65	0.46	0.54	0.00	0.52	0.40	0.08	0.73	0.27	39.96	0.001
Total	305	0.51	0.46	0.03	0.53	0.39	0.08	0.74	0.26	2.42	0.1198

TABLE 3. Least square means and SE of studied traits in relation to *HSP 90 AA1 F1* gene

	Age at 1st calving (month)	Calving Interval (days)	Calf Birth Weight (Kg)	Milk Yield (Kg)	Days of Milking (days)
Animal Status					
Cyclic	36.86 ± 0.396 ^a	420.76 ± 7.786 ^c	29.33 ± 0.374 ^a	2438.76 ± 70.22 ^c	287.65 ± 7.08 ^b
PPA 6-12	34.55 ± 0.512 ^b	471.33 ± 10.051 ^b	29.75 ± 0.483 ^a	2702.79 ± 90.65 ^b	308.81 ± 9.14 ^b
PPA>12	31.61 ± 0.602 ^c	596.92 ± 11.828 ^a	27.02 ± 0.569 ^b	3494.80 ± 106.67 ^a	382.58 ± 10.75 ^a
Probability	0.0001	0.0001	0.001	0.0001	0.0001
Genotype <i>HSP 90 AA1 F1</i>					
AA	32.70 ± 0.51 ^b	528.00 ± 9.65 ^a	27.97 ± 0.51 ^b	3006.60 ± 89.82 ^a	329.95 ± 8.96 ^a
AC	34.10 ± 0.39 ^a	507.58 ± 7.48 ^a	29.56 ± 0.40 ^a	3091.91 ± 69.69 ^a	340.7 ± 6.95 ^a
CC	33.23 ± 0.87 ^{ab}	423.27 ± 16.47 ^b	28.37 ± 0.88 ^{ab}	2351.1 ± 153.34 ^b	323.1 ± 15.29 ^a
Probability	0.101	0.0001	0.045	0.0001	0.448
Animal Status * Genotype <i>HSP 90 AA1 F1</i>					
Cyclic*AA	37.20 ± 0.58 ^a	401.50 ± 10.94 ^f	28.28 ± 0.58 ^{bcd}	2143.10 ± 101.85 ^d	249.82 ± 10.16 ^c
Cyclic*AC	36.10 ± 0.58 ^{ab}	426.75 ± 10.94 ^{ef}	31.01 ± 0.58 ^{ab}	2805.30 ± 101.85 ^b	314.25 ± 10.16 ^b
Cyclic*CC	38.20 ± 0.89 ^a	434.80 ± 16.95 ^{def}	27.90 ± 0.90 ^{bcd}	2206.23 ± 157.79 ^{cd}	309.26 ± 15.74 ^b
PPA 6-12*AA	33.33 ± 0.82 ^{bc}	496.16 ± 15.47 ^c	28.04 ± 0.82 ^{bcd}	2944.58 ± 144.04 ^b	360.20 ± 14.37 ^{ab}
PPA 6-12*AC	33.62 ± 0.71 ^{bc}	478.00 ± 13.40 ^{cd}	31.80 ± 0.72 ^a	2754.38 ± 124.74 ^{bc}	300.65 ± 12.44 ^{bc}
PPA 6-12*CC	36.50 ± 1.42 ^{ab}	470.00 ± 26.80 ^{cde}	27.22 ± 1.43 ^{cd}	2707.00 ± 249.49 ^{bcd}	305.97 ± 24.89 ^b
PPA>12*AA	27.66 ± 1.16 ^d	686.33 ± 21.88 ^a	27.60 ± 1.17 ^{bcd}	3932.16 ± 203.71 ^a	379.83 ± 20.32 ^a
PPA>12*AC	32.57 ± 0.76 ^c	618.00 ± 14.32 ^b	25.78 ± 0.76 ^d	3716.07 ± 133.36 ^a	407.40 ± 13.30 ^a
PPA>12*CC	25.00 ± 2.00 ^d	365.00 ± 37.90 ^f	30.00 ± 2.03 ^{abc}	2140.00 ± 352.83 ^d	354.00 ± 35.20 ^{ab}
Probability	0.0001	0.0001	0.0001	0.0001	0.0001

S.O.V source of variation ** a...f Means within trait, followed by different superscripts, differ significantly (Duncan, 1955*).

TABLE 4. Least square means and SE of studied traits in relation to *HSP 90 AA1 F3* gene.

	Age at 1st calving (month)	Calving Interval (days)	Calf Birth Weight (Kg)	Milk Yield (Kg)	Days of Milking (days)
Animal Status					
Cyclic	36.86 ± 0.396 ^a	420.76 ± 7.786 ^c	29.33 ± 0.374 ^a	2438.76 ± 70.22 ^c	287.65 ± 7.08 ^b
PPA 6-12	34.55 ± 0.512 ^b	471.33 ± 10.051 ^b	29.75 ± 0.483 ^a	2702.79 ± 90.65 ^b	308.81 ± 9.14 ^b
PPA>12	31.61 ± 0.602 ^c	596.92 ± 11.828 ^a	27.02 ± 0.569 ^b	3494.80 ± 106.67 ^a	382.58 ± 10.75 ^a
Probability	< 0.0001	< 0.0001	0.001	< 0.0001	< 0.0001
Genotype <i>HSP 90 AA1 F3</i>					
TT	35.33 ± 0.45 ^a	457.07 ± 10.11 ^a	28.16 ± 0.41 ^{ab}	2626.88 ± 77.56 ^b	314.47 ± 7.46 ^a
TG	34.44 ± 0.45 ^a	491.51 ± 10.11 ^a	29.91 ± 0.41 ^a	3026.33 ± 77.56 ^a	334.37 ± 7.46 ^a
GG	32.00 ± 1.65 ^a	496.00 ± 37.16 ^a	27.00 ± 1.52 ^b	1420.00 ± 284.98 ^c	160.00 ± 27.42 ^b
Probability	0.09	0.048	0.045	0.0001	0.0001
Animal Status * Genotype <i>HSP 90 AA1 F3</i>					
Cyclic*TT	38.20 ± 0.50 ^a	400.00 ± 10.54 ^c	28.32 ± 0.52 ^b	2260.52 ± 92.45 ^c	281.61 ± 8.93 ^d
Cyclic*TG	36.08 ± 0.56 ^b	426.75 ± 11.79 ^c	31.01 ± 0.58 ^a	2805.30 ± 103.36 ^b	314.25 ± 9.99 ^c
Cyclic*GG	32.00 ± 1.38 ^c	496.00 ± 28.88 ^b	27.00 ± 1.43 ^{bc}	1420.00 ± 253.18 ^d	160.0 ± 24.47 ^e
PPA 6-12*TT	34.12 ± 0.69 ^c	489.62 ± 14.44 ^b	27.83 ± 0.71 ^{bc}	2885.18 ± 126.59 ^b	346.64 ± 12.23 ^b
PPA 6-12*TG	33.62 ± 0.69 ^c	478.00 ± 14.44 ^b	31.88 ± 0.72 ^a	2754.38 ± 126.60 ^b	300.65 ± 12.23 ^{cd}
PPA>12*TT	27.00 ± 0.98 ^d	606.00 ± 20.42 ^a	28.20 ± 1.0 ^{bc}	3484.12 ± 179.02 ^a	373.37 ± 17.30 ^{ab}
PPA>12*TG	32.57 ± 0.74 ^c	618.00 ± 15.43 ^a	25.78 ± 0.76 ^c	3716.07 ± 135.33 ^a	407.40 ± 13.08 ^a
Probability	0.0001	0.0001	0.0001	0.0001	0.0001

S.O.V source of variation ** a...d Means within trait, followed by different superscripts, differ significantly (Duncan, 1955*).

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تعدد أشكال جين بروتين الصدمة الحراري AA190 وعلاقته بالصفات التناسلية والإنتاجية في أبقار هولشتاين

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المخلص

يعتبر جين HSP90AA1 عضواً في عائلة بروتينات الصدمة الحرارية وله دوراً حاسماً في استجابات الإجهاد الخلوية، وخاصة في تحمل الحرارة وخصوبة الماشية. تهدف دراستنا إلى تحديد تعدد أشكال جين HSP90AA1 وعلاقته بخصوبة وإنتاج الأبقار. تم تصنيف إجمالي عدد 305 بقرة إلى 3 مجموعات حسب نشاط المبايض بعد الولادة (حيوانات ذات دورة شبق منتظمة، حيوانات ذات دورة شبق متأخرة من 6 إلى 12 شهراً، وأخري ذات دورة شبق متأخرة لأكثر من 12 شهراً). تم جمع البيانات المتعلقة بالتكاثر والإنتاج من سجلات المزرعة، بما في ذلك العمر عند الولادة الأولى، وفترات ما بين الولادات، وأوزان العجول عند الولادة، وإنتاج الحليب، وأيام الحلب. تم استخلاص الحمض النووي الجينومي وتحليل PCR-SSCP للكشف عن ثلاث قطع من جين HSP 90AA1 وهما F1, F2, F3 و F3 عند 285, 355, و 280 زوجاً قاعدياً على التوالي. لوحظت ثلاثة أنماط مختلفة في القطعتين HSP90AA1 F1 و HSP90AA1 F2 بينما كانت القطعة HSP90AA1 F3 أحادية الشكل. أظهر تحليل تسلسل HSP90AA1 F1 ثلاثة أنماط جينية مختلفة AC, AA, CC مع وجود طفرة استبدال قاعدة واحدة عرضية (A/C) تقع عند 21:66942714 معرفة باسم rs207763220 كطفرة مترادفة. بينما أظهر HSP90AA1 F3 عن 3 أنماط جينية TT و GG و TG مع وجود طفرة استبدال تقع عند 21:66941688 (T/A/G) معرفة باسم rs210718366 ارتبط النمط الجيني CC في جين HSP90AA1 F1 بانخفاض فترة ما بين الولادة وإنتاج الحليب في الأبقار ذات نشاط المبيض المتأخر لأكثر من 12 شهراً بعد الولادة. كما ارتبط النمط الجيني GG في جين HSP90AA1 F3 بانخفاض أيام الحليب وإنتاجيته. بالإضافة إلى ذلك، ارتبط النمط الجيني TT بانخفاض العمر عند الولادة الأولى في الأبقار ذات نشاط المبيض المتأخر أكثر من 12 شهراً بعد الولادة. و تشير نتائجنا إلى أن الاختلافات الجينية في جين HSP90AA1 يمكن استخدامها كعلامة جزيئية للاختيار الجيني لتعزيز الأداء التناسلي والإنتاجي في أبقار الهولشتاين، وخاصة العمر عند الولادة الأولى، وفترة ما بين الولادات وإنتاج الحليب المرتبط بفترة ما بعد الولادة.

الكلمات الدالة: جين بروتين الصدمة الحراري AA1 90 ، النمط الجيني، تعدد أشكال النيوكليوتيدات المفردة، فترة ما بعد الولادة، الخصوبة و الأبقار.