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### ESTIMATION OF GENETIC PARAMETERS AND BREEDING VALUES FOR DAILY MILK PRODUCTION OF DAIRY SIMMENTALS IN TERMS OF HEAT STRESS

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Variance components for daily milk production as well as breeding value of dairy Simmentals for daily milk production in terms of heat stress defined as different values of THI threshold values (72, 74, and 76) were estimated using 1,636,192 test-day records provided by the Croatian Agricultural Agency. Temperature-humidity index (THI) calculated from ambient temperature and relative humidity recorded in the barns on the milk recording day was used as the measure of heat stress. Bivariate estimation model accounted variances due to lactation stage, calving season, age at calving, breeding region, parity, permanent environment, interaction between hear and year, and individual animal. Obtained high genetic correlations between the daily milk production in normal and condition characterized by heat stress, as well as very high correlations between the EBVs in normal and heat stress conditions indicate animals' high level of acclimatization to the environment on dairy cattle farms. Since microclimate measurements were carried out only once at a milk recording day, these results should be taken with caution. Generally, further research with multiple daily measurements of the microclimate parameters in the facilities is necessary to provide a fully unambiguous conclusion.

*Keywords:* daily milk yield, heat stress, resistance, genetic parameters, Simmental breed.

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#### INTRODUCTION

Dairy farming and welfare is affected by various factors due to complex interactions between the animal and the environment with its different factors (LAMBERTZ et al., 2014). According to MOSELEY et al. (2012) mean annual temperatures are predicted to rise by 1°C for the period of 2021 to 2050 and 2.5°C for the period of 2071 to 2100 in Lower Saxony compared with the reference period of 1971 to 2000. With increasing of the adverse climate change, SEGNALINI et al. (2013) pointed out the necessity of adequate adaptation strategies in order to decrease negative effects of climate change on domestic animals. In intensive dairy breeding systems, the heat stress is a huge problem, because the high production makes cows more susceptible to heat stress (HANSEN, 2013). The level of milk production significantly alters the animal response to heat stress (WEST et al., 2003; COLLIER et al., 2006; GANTNER et al., 2011; DUNN et al., 2014), because the intensive genetic selection for higher milk production resulted in changes in the thermoregulation physiology of dairy cattle, which means that larger animals with higher production have larger gastrointestinal tracts enable them to digest more feed, but also creates more metabolic heat and reduces the animal's ability to regulate temperature at heat stress environment (KADZERE et al., 2002). Besides of the negative influence on the production level, the heat stress has also a negative influence on following traits: dry matter intake, milk composition, reproductive performances, somatic cell counts (SCC) and prevalence of mastitis (CASA and RAVELO, 2003; BOHMANOVA et al., 2007; COLLIER et al., 2012; HAMMAMI et al., 2013; SMITH et al., 2013; LAMBERTZ et al., 2014; GANTNER et al., 2017). Finally, accordingly to research of ST-PIERRE et al. (2003) heat stress induces considerable profit loss.

The temperature-humidity index (THI) is a commonly used indicator for determination of the heat stress influence on productivity of dairy cows (HUBBARD et al., 1999; GANTNER et al., 2011). THI incorporates the effects of ambient temperature as well as relative humidity, and according to previous research THI threshold value at which heat stress affects milk production and feed intake range from 68 to 72 (BOURAOUI et al., 2002; BERNABUCCI et al., 2010; COLLIER and HALL, 2012). There are numerous methods of minimizing the impact of heat stress on dairy cattle. These methods could be categorised as short-term and long-term methods. Short-term methods include optimization of feeding and application of different cooling systems in farm buildings, while long-term methods imply selection of dairy cattle for resistance to heat stress. Determination of the effect of heat stress on milk production and the estimation of genetic components of heat stress resistance were studied in previous research. MISZTAL (1999) proposed a model fitting a comfort zone, with no effect of temperature on production. SCHAEFFER (2004) emphasised that longitudinal data are commonly analysed with random regression models. DRUET et al. (2003), SILVESTRE et al. (2005) and BOHMANOVA et al. (2007) concluded that splines can be used to model (co)variances in test-day models. Furthermore, MISZTAL (2006) point out that the linear splines are easily interpretable, and have good numerical properties and local effects. SÁNCHEZ et al. (2009) used thermotolerance threshold and the subsequent slope of decay for each animal in the complex model. According to RAVAGNOLO and MISZTAL (2000) the additive genetic variability for heat stress tolerance are important for milk, fat and protein production of primiparous cows. Genetic evaluation for heat tolerance for Holstein cows were established BOHMANOVA et al. (2005), and they concluded that daughters of bulls with high genetic merit for heat tolerance had lower milk yields, higher contents of milk solids, more

robust bodies, better udders, longer productive lives, and higher pregnancy rates than daughters of bulls with low genetic merit for heat tolerance. AQUILAR et al. (2009), using the test-day models that included a random regression on a function of THI, concluded that phenotypic variance and additive genetic effects for heat stress and yield traits increased greatly from the first to third parity, and consequently, later parity cows are expected to be much more susceptible to heat stress than are primiparous cows. BRUGEMANN et al. (2011) used a random regression models for genetic analyses of protein yield in dairy cows, and concluded that additive genetic variances for daily protein yield decreased with increasing degrees of heat stress and were lowest at the beginning of lactation and at extreme value of THI. CARABAÑO et al. (2014) established that Legendre polynomial model provided better explanation and fit than the splines model to describe changes in production and SCC along the range of studied temperatures at the phenotypic level as well as for the individual components. Since climate change and consequently heat stress becomes the reality of the world that we are living and producing, there is a necessity of adequate adaptation strategies aiming decrease of negative effects of climate change on domestic animals (SEGNALINI et al., 2013). Therefore, in order to apply long-term mitigation method, the objectives of this study were estimation of variance components for milk production as well as estimation of breeding value of dairy Simmentals in terms of heat stress defined as different values of THI threshold values (72, 74, and 76).

#### MATERIAL AND METHODS

Test-day records of dairy Simmental cows reared in Croatia were obtained from the Croatian Agricultural Agency. Test-day records were collected in the period from January 2005 to December 2013 during the regular milk recording (AT4 or BT4 method). At each recording, measuring and sampling of milk were performed during the evening or morning milking, and also, at each recording, ambient temperature and relative humidity in the barns were recorded. Furthermore, test-day records with lactation stage in (< 5 days and > 400 days), age at first calving in (< 21 and > 36 months), missing parity, missing breed, missing or nonsense daily milk traits (accordingly to ICAR standards, 2017), and missing or nonsense Ta and RH value were deleted from the dataset. After logical control dataset consisted of 1,636,192 test-day records from 117,659 Simmentals reared on 10,599 farms. Accordingly, to the parity, cows were divided into seven classes: 1., ..., 7. (animals in seventh and higher lactations). Furthermore, accordingly to location of farm, test day records were divided into 16 breeding regions. In accordance to the calving date, test day records were divided into four recording season: spring (March, April, and May), summer (June, July, and August) autumn (September, October, and November), and winter (December, January, and February). Based on measured microclimate parameters, temperature-humidity index (THI) was calculated using the equation by KIBLER (1964):

THI = 
$$1.8 \times Ta - (1 - RH) \times (Ta - 14.3) + 32$$

Where Ta presents the average temperature in degrees Celsius while RH is the relative humidity as a fraction of the unit.

A function (f) of THI was created as follows:  $f(THI) = \begin{cases} 0, & \text{if } THI \leq THI_{threshold} \\ 1, & \text{if } THI > THI_{threshold} \end{cases}$  where THI threshold was set to 72, 74, and 76.

For estimation of variance components for milk production as well as for estimation of breeding values of dairy Simmental cows in terms of heat stress following bivariate model was used:

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y_{iiklmnop} = \mu + b_1(d_i/305) + b_2(d_i/305)^2 + b_3 \ln(305/d_i) + b_4 \ln^2(305/d_i) + S_i + b_3 ac_k + b_6 ac_k^2 + R_l + P_{im} + Pe_n + Hy_o + a_p + e_{iiklmnop}
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where y_{ijklmnop} = estimated daily milk production at THI = 0 and THI = 1; \mu = intercept; b_1, b_2, b_3, b_4, b_5, b_6 = regression coefficients; d_i = days in milk (i = 5, ..., 400 day); S_j = fixed effect of calving season class j (j = 1/2005, 2/2005, ..., 4/2012); ac_k = fixed effect of age at calving as square regression; R_l = fixed effect of breeding region l (l = 01, ..., 16); P_{im} = fixed effect of parity m (m = 01, ..., 07; nested within the lactation curve); Pe_n - random permanent environmental effect for the cow; Hy_o - random additive genetic effect for the cow; e_{ijklmnop} = random residual effect.
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For the preparation and logical control of data SAS/STAT (SAS Institute Inc., 2000) was used. For estimation of variance components for milk production of dairy cows VCE-6, version 6.0.3-dev (KOVAČ *et al.*, 2012) was used. Furthermore, breeding values of dairy Simmental cows in terms of heat stress defined as different values of THI threshold values (72, 74, and 76) were estimated using same software.

#### **RESULTS AND DISCUSSION**

In this research the animal response to heat stress was analysed using bivariate model where the daily milk production in normal conditions (THI = 0) was taken as first variable while the daily milk production in heat stress environment (THI = 1) was taken as second variable. The results of estimation of variance components for daily milk production of dairy Simmentals, accordingly to the set threshold values (THI  $_{threshold}$  in 72, 74, or 76) are presented in Table 1. The variance for permanent environment when THI = 0 ranged from 3.627 kg² to 3.579 kg², while for THI = 1 ranged from 3.577 kg² to 3.670 kg² depending of THI  $_{threshold}$  value indicating that at higher THI values the effect of permanent environment was more pronounced. Similar trend of variance increase in normal condition regarding the THI  $_{threshold}$  value was also observed for variability due to herd-year interaction and residual variance. In terms of heat stress (THI = 1), regarding the set THI  $_{threshold}$  value, the decrease of residual variance was observed, while the heard-year variance increased. Estimated variance for animal was slightly higher in terms of heat stress as well as at higher values of set THI  $_{threshold}$  with exception when THI in 76.

AGUILAR *et al.* (2009) determined that the estimated additive genetic variance for milk, fat, and protein yields highly depends of THI, days in milk (DIM), parity, and statistical model used for the estimation. Similarly like in study of RAVAGNOLO and MISZTAL (2000), AGUILAR *et al.* (2009) find out that the function of total additive genetic variance and THI had a typical

quadratic shape. Furthermore, the genetic variance for heat tolerance significantly increased at the end of lactation and in higher parities. The influence of lactation stage on the additive genetic variance with the highest values determined at the end of lactation (DIM = 300) was also observed by BRUGEMANN *et al.* (2011). Same authors also determined the decrease of additive genetic variance and permanent environment variance for test-day protein yield in terms of increasing THI value (from 21 to 72).

Table 1. Estimated variances and covariance for milk production of dairy Simmental cows accordingly to set threshold values (THI threshold in 72, 74, or 76)

Effect	Variance (THI – 0)	Variance (THI – 1)	Covariance					
THI threshold in 72								
Penv	$3.62684 \pm 0.023704 \text{ kg}^2$	$3.57655 \pm 0.054748 \text{ kg}^2$	$4748 \text{ kg}^2 \qquad \qquad 3.18274 \pm 0.029340 \text{ kg}^2$					
Hyear	$4.39042 \pm 0.057105 \ kg^2$	$5.35418 \pm 0.080783 \text{ kg}^2$	$4.22085 \pm 0.061951 \text{ kg}^2$					
Animal	$5.25454 \pm 0.056325 \text{ kg}^2$	$5.25809 \pm 0.078944 \text{ kg}^2$	$5.07305 \pm 0.058007 \ kg^2$					
Residual	$8.94054 \pm 0.012533 \text{ kg}^2$	$7.54261 \pm 0.038398 \text{ kg}^2$	-					
THI threshold in 74								
Penv	$3.59892 \pm 0.024255 \text{ kg}^2$	$3.61619 \pm 0.065184 \text{ kg}^2$	$3.18366 \pm 0.032497 \text{ kg}^2$					
Hyear	$4.39700 \pm 0.055753 \text{ kg}^2$	$5.50066 \pm 0.089483 \text{ kg}^2$	$4.25371 \pm 0.063407 \ kg^2$					
Animal	$5.23564 \pm 0.053151 \text{ kg}^2$	$5.25395 \pm 0.087959 \text{ kg}^2$	$5.09532 \pm 0.058297 \ kg^2$					
Residual	$8.96415 \pm 0.013173 \text{ kg}^2$	$7.40636 \pm 0.045108 \text{ kg}^2$	-					
THI threshold in 76								
Penv	$3.57874 \pm 0.022982 \text{ kg}^2$	$3.69503 \pm 0.082649 \text{ kg}^2$	$3.17386 \pm 0.036770 \text{ kg}^2$					
Hyear	$4.40475 \pm 0.053216 \ kg^2$	$5.58089 \pm 0.097791 \text{ kg}^2$	$4.25124 \pm 0.065262 \ kg^2$					
Animal	$5.22721 \pm 0.052722 \text{ kg}^2$	$5.17235 \pm 0.099102 \text{ kg}^2$	$5.10041 \pm 0.064147 \text{ kg}^2$					
Residual	$8.97953 \pm 0.013315 \text{ kg}^2$	$7.28479 \pm 0.052614 \text{ kg}^2$	-					

<sup>\*</sup>Penv - permanent environment; Hyear - herd-year interaction

Estimated variance ratios and correlations for daily milk production of dairy Simmental cows accordingly to set threshold values (THI  $_{\rm threshold}$  in 72, 74, or 76) are presented in the Table 2. The variability of daily milk production due to permanent environment ranged from 16.13-17.00% depending of environment characteristics. The effect of permanent environment was the most pronounced in the barns with THI > 76. The amount of variability of milk production due to herd-year interaction ranged between 19.77%-25.68% with the highest proportion of variability determined in environment characterized by heat stress and at highest THI  $_{\rm threshold}$  value. Finally, heritability for daily milk production in normal conditions (THI = 0) was around 23.50%, while in terms of heat stress heritability was higher and ranged between 23.78-24.20%.

Estimated genetic correlations was high and ranged, depending of THI threshold value, from 96.13% at the lowest THI (72) to 98.09% at the highest analysed THI (76) value. High values of estimated genetic correlations between the daily milk production in normal conditions and the daily milk production in conditions characterised with heat stress indicate that cows with high performance in normal condition will retain similar production level even in terms of heat stress.

SANCHEZ et al. (2009) determined relatively constant values of heritability for daily milk production as a function of THI (ranged from 60 to 90). AGUILAR et al. (2009) reported that

values of heritability for milk yield depends on lactation stage, parity and THI value with the highest values at the end of lactation, and in third parity, with slightly increase at higher THIs. RAVAGNOLO and MISZTAL (2000) determined a slightly increase of heritability for protein yield as well as slightly decrease of heritability for fat yield with increasing THI from 72 to 85. Contrary, BRUGEMANN *et al.* (2011) determined that the values of heritability for protein yield were highest at lower THI values (but with THI in interval 21 - 72) and at the end of lactation. Same authors emphasized that the effect of lactation stage on heritabilities was more pronounced that the effect of THI. AGUILAR *et al.* (2009) reported that the genetic correlations among parities for the effect of heat tolerance ranged from 0.56 to 0.79 indicated differences in heat tolerance due to parity.

Table 2. Estimated ratios and correlations for milk production of dairy Simmental cows accordingly to set threshold values (THI threshold in 72, 74, or 76)

Effect	Ratio (THI – 0)	Ratio (THI – 1)	Correlation					
THI threshold in 72								
Penv	$0.16328 \pm 0.1117$ E-02	$0.16458 \pm 0.2509$ E-02	$0.88370 \pm 0.6348$ E-02					
Hyear	$0.19766 \pm 0.2286$ E-02	$0.24638 \pm 0.3164$ E-02	$0.87056 \pm 0.3506$ E-02					
Animal	$0.23656 \pm 0.2312$ E-02	$0.24196 \pm 0.3384$ E-02	$0.96513 \pm 0.3312$ E-02					
Residual	$0.40250 \pm 0.1189$ E-02	$0.34708 \pm 0.2144$ E-02						
THI threshold in 74								
Penv	$0.16215 \pm 0.1138$ E-02	$0.16605 \pm 0.2996$ E-02	$0.88250 \pm 0.7366$ E-02					
Hyear	$0.19810 \pm 0.2205$ E-02	$0.25259 \pm 0.3433$ E-02	$0.86493 \pm 0.3936$ E-02					
Animal	$0.23589 \pm 0.2190$ E-02	$0.24126 \pm 0.3719$ E-02	$0.97150 \pm 0.3927$ E-02					
Residual	$0.40387 \pm 0.1209$ E-02	$0.34010 \pm 0.2495$ E-02						
THI threshold in 76								
Penv	$0.16128 \pm 0.1073$ E-02	$0.17002 \pm 0.3690$ E-02	$0.87280 \pm 0.9648$ E-02					
Hyear	$0.19850 \pm 0.2114$ E-02	$0.25679 \pm 0.3768$ E-02	$0.85744 \pm 0.4453$ E-02					
Animal	$0.23556 \pm 0.2162$ E-02	$0.23799 \pm 0.4268$ E-02	$0.98090 \pm 0.5076$ E-02					
Residual	$0.40466 \pm 0.1188$ E-02	$0.33519 \pm 0.2930$ E-02	-					

 $<sup>*</sup>Penv-permanent\ environment;\ Hyear-herd-year\ interaction$ 

The variability of estimated breeding values (EBV) for daily milk production of dairy Simmental cows in normal and heat stress environment accordingly to set threshold values (THI threshold in 72, 74, or 76) is presented in the Table 3. Mean values of EBVs, regardless the environmental conditions, amounted around 100.5. Furthermore, mean value of EBVs for daily milk production in normal conditions was the highest at the lowest THI value, while in heat stress condition the highest EBVs was at highest THI value. The correlations between the EBVs in normal and heat stress conditions was high and ranged between 99.48-99.76% indicating strong link between cow's productivity in normal and heat stress environment.

AGUILAR et al. (2009) in the analysis of genetic components of heat stress for dairy cattle determined that the value of genetic variance for heat tolerance for milk yield depends of the used model (random regression model vs repeatability model). RAVAGNOLO et al. (2000) for the analysis of variance components of response used the so-called broken line model. This model is

defined by the thermoneutrality threshold and the slope of production decrease after defined threshold value. Furthermore, in some research (BRÜGEMANN et al., 2011; MENENDEX-BUXADERA et al., 2012; CARABAÑO et al., 2014) the variation in daily production due to heat stress was described as polynomial functions that enable more flexible approach comparable to broken line models. One of the main problems in the application of any evaluation model is how to combine (micro)climate variables (GAUGHAN et al., 2012) as well as the definition of the lag between the test-day and the date with measurements of (micro)climate (CARABAÑO et al., 2014). In continuation, the determination of the adequate selection criteria for each evaluation model is very important. In the broken line model, as selection criteria, both the thermotolerance threshold and the slope of animal's response could be used. In the case of higher-order polynomials used to describe the animal's reaction to heat stress, the slope of the individual polynomial curve under moderate or severe heat stress could be used as criteria for selection (CARABAÑO et al., 2014). Accordingly, to SÁNCHEZ et al. (2009) the practical application of model will show the animals with higher adaptation (those who have lower performance decrease as well as a later onset of environmental stress). Furthermore, CARABAÑO et al. (2014) reported the variability in genetic response and the reranking of animals accordingly to different temperatures indicating some interaction between the genotype and (micro) climate.

Table 3. Variability of estimated breeding values (EBV) for daily milk production of dairy Simmental cows in terms of heat stress accordingly to set threshold values (THI threshold in 72, 74, or 76)

THI	EBV (THI – 0)				EBV (THI – 1)						
	mean	SD	CV	MIN	MAX	mean	SD	CV	MIN	MAX	r
72 100.486	13.32	12.26	26 52 127	147.98	100 422	13.32	12.27	52.100	147.98	0.9947	
	100.486	5	13.26	52.127	1	100.423	6	13.27	52.106	0	7
74 100.482	13.32	13.26	52.122	147.98	100.446	13.32	12.26	52.107	147.98	0.9959	
	100.462	5	13.20	13.20 32.122	1	100.446	3	13.26	32.107	0	1
76 100	100.481	13.32	13.26	52.126	147.98 1	100.451	13.32	13.26	52.116	147.98	0.9976
	100.461	4	15.20	32.120			3			0	2

<sup>\*</sup>r - correlation between the EBV in normal (THI – 0) and heat stress environment (THI – 1); all correlation coefficients were statistically highly significant (p < 0.001)

In order to simplify the practical selection of animals for resistance to heat stress, in this research, daily milk production was analysed as bivariate variable, in the normal conditions (under the set THI threshold value), and in terms of heat stress (above the set THI threshold value). Estimated very high genetic correlations between the daily milk production in normal and heat stress condition, as well as very high correlations between the EBVs in normal and heat stress conditions show that high-performance cows in the normal condition will maintain a similar level of production even in terms of heat stress. Given the obtained results, good acclimatization of dairy Simmental cows to environmental conditions on dairy cattle farms could be assumed.

#### **CONCLUSIONS**

Estimated high genetic correlations between the daily milk production in normal and condition characterized by heat stress, as well as very high correlations between the EBVs in

normal and heat stress conditions. These results should be taken with caution, since microclimate measurements were carried out only once at a milk recording day, therefore the question is whether they could be relevant as such. Generally, further research with multiple daily measurements of the microclimate parameters in the facilities is necessary to provide a fully unambiguous conclusion.

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## PROCENA GENETSKIH PARAMETARA I OPLEMENJIVAČKIH VREDNOSTI ZA DNEVNU PROIZVODNJU MLEKA SIMENTALSKE RASE U USLOVIMA TOPLOTNOG STRESA

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#### Izvod

Komponente varijanse za dnevnu proizvodnju mleka kao i oplemenjivačka vrednost mlečnih krava simentalske rase za dnevnu proizvodnju mleka u uslovima toplotnog stresa definisanoj pri različitim vrednosti THI-a (72, 74 i 76) procenjene su upotrebom 1.636.192 zapisa na kontrolni dan. Temperaturno-humidni indeks (THI) izračunat na osnovu temperature okoline i relativne vlage izmerene u stajama na kontrolni dan, korišćen je kao mera toplotnog stresa. Bivarijatni model procene uvažio je varijance usled stadijuma laktacije, sezone telenja, dobu na telenju, regije uzgoja, pariteta, permanentne spoljašnje sredine, interakcije između stada i godine te pojedine životinje. Dobijena visoka genetska korelacija između dnevne proizvodnje mleka u normalnim i uslovima toplotnog stresa, kao i vrlo visoka korelacija između procenjenih oplemenjivačkoh vrednosti (EBV) u normalnim i toplotno stresnim uslovima, ukazuju na visok nivo aklimatizacije životinja na spoljašnju sredinu na farmama mlečnih goveda. Budući da su merenja mikroklime sprovedena samo jednom na kontrolni dan, ove je rezultate potrebno uzimati s oprezom. Nadalje, potrebna su dalja istraživanja s višestrukim dnevnim merenjima mikroklimatskih parametara u objektima kako bi se dobio potpuno nedvosmislen zaključak.

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