Journal of the Hellenic Veterinary Medical Society

Vol 73, No 4 (2022)

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doi: 10.12681/jhvms.28470

To cite this article:

Şahin, A., Aksoy, Y., Yildirim, A., & Ulutaş, Z. (2023). Determination of Direct and Indirect Effects on Milk Yield of Anatolian Buffaloes Using Path Analysis. *Journal of the Hellenic Veterinary Medical Society*, *73*(4), 4935–4944. https://doi.org/10.12681/jhvms.28470

J HELLENIC VET MED SOC 2022, 73 (4): 4935-4944
ΠΕΚΕ 2022, 73 (4): 4935-4944

Research article
Ερευνητικό άρθρο

Determination of Direct and Indirect Effects on Milk Yield of Anatolian Buffaloes Using Path Analysis

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ABSTRACT: This study aimed to determine the direct and indirect effects of the independent variables presented by the lactation length (LL), age of calving (CAGE), and daily milk yield (DMY) on the dependent variable of lactation milk yield (LMY) in Anatolian buffaloes. In this study, 3761 LMY records of the 834 Anatolian buffaloes calving between 2012 and 2017 in Tokat province and around were used as the research material. In the study, the simple correlation coefficients between the dependent variable of LMY and independent variables were determined to be positive and significant (P<0.001). The research specified the direct determination coefficients of the independent variables of the LL, CAGE, and DMY on the dependent variable of LMY as 0.378, 0.004, and 0.350, respectively. The path coefficients related to LL, CAGE, and DMY were 0.615, 0.021, and 0.592. Since the DMY and LL examined in the study directly impacted the lactation milk yield, using these data as criteria in breeding programs conducted in the herds may increase the selection success.

Keywords: the dependent variable, the independent variable, the direct effect, the indirect effect

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Date of initial submission: 27-10-2021
Date of acceptance: 24-06-2022

INTRODUCTION

Quantitative characters are the features of economic importance in livestock breeding. Environmental factors influence the emergence of quantitative characters than qualitative characters. Before starting animal breeding studies, it is necessary to determine the environmental effects and their levels influencing the accentuated characteristics (Düzgüneş et al., 2012). For this purpose, the correlation coefficient, a value used to show the relationship between variables, is widely used (Supriatna et al., 2019).

The cause-effect relations are generally considered in the equations with more than one variable. When there is a functional relationship between the variables, the degree and functionality of the relationship are determined. While the direction and degree of the relationship between multiple variables are determined by performing correlation analysis, the relationship's structure can be detected using regression analysis (Bek, 1988; Norris et al., 2015). Since the relationship between two variables (or more) in the examination may depend on another variable, it is impossible to determine the highlighted yield(s) factors with a simple correlation coefficient. While the simple correlation determines the relationship between two variables, the partial correlation determines the relationship between two variables when the effect of one or more related random variables is eliminated.

Also, since the impact of environmental factors on the characteristics may occur indirectly or directly, the environmental factors affecting the output(s) directly or indirectly should be expressed in detail. Path coefficient explains the indirect and direct impacts of environmental factors affecting the yield(s). If there is causality between dependent and independent variables, in this case, path analysis is used to determine the size and significance of causal connections between variables (Vendruscolo et al., 2016; Tyasi et al., 2018). Path analysis is a multivariate statistical method used to explain independent variables' direct and indirect effects on the dependent variable through path diagrams (Düzgüneş et al., 2012; Tyasi et al., 2020; Mahapatra et al., 2020).

While parents of future generations are selected in herds in animal breeding studies, one or more productivity can be considered together as selection criteria. In such a case, determining whether other yields will cause a change in accentuated productivity(s) will affect the program achievement to be implemented. The efficiency of breeding activities can be increased by considering the relationships between outputs (Düzgüneş et al., 2012; Tyasi et al., 2020). Also, it is undeniable that environmental factors directly or indirectly influence on the mentioned productivity(s). In Turkey, no study has been encountered examining the direct and indirect effects of the environmental factors on the Anatolian buffaloes' milk yield using the path analysis. Therefore, this study used path analysis to reveal the direct and indirect impacts of the LL, DMY, and CAGE on LMY in Anatolian buffaloes.

MATERIAL AND METHODS

The study material consisted of 3761 lactation milk yield records of 834 Anatolian buffaloes, which calved between 2012-2017 in Tokat province. The data were obtained from the Association of Buffalo Breeders in Tokat.

While LMY was the dependent variable (Y), LL (X_1), CAGE (X_2), and DMY (X_3) were independent variables in the study. First of all, simple and partial correlation coefficients between dependent (Y) and independent variables (X_1, X_2, and X_3) were determined. The dependent (Y) and independent variables (X_1, X_2, and X_3) were standardized by performing regression analysis. The measurement units were "kg" for LMY and DMY, "days" for LL, and "months" for calving age. Direct and indirect effects between dependent variables and independent variables were determined by path analysis. SPSS statistical package program determined the direct impacts between variables.

Indirect and direct effects of independent variables on dependent variables are calculated with path analysis (Tyasi et al., 2017). The direct effect expressing the correlation coefficient between two variables is the direct effect of the independent variable on the dependent variable. Path coefficient is the relative amount of the variation displayed by a quantitative feature in a population resulting only from a constant variation factor. The path coefficient is denoted by the letter P and is calculated utilizing the following equation (Mendeş et al., 2005).

$$Pyx_i = b\frac{Sx_i}{S_y} \quad [1]$$

P_{yx_i} : the coefficient of path indicating the direct effect of the X independent variable on the Y dependent variable

b : the coefficient of partial regression

S_X : standard deviation of the X property

$$S_{x_i} = \sqrt{[\Sigma(x_i - \bar{x}_i)^2]}.\frac{1}{n} = \sqrt{\left[x_i^2 - \frac{(\Sigma x_i)^2}{n}\right].\frac{1}{n}} = \sqrt{S_{xx_i}} \quad [2]$$

P_y : standard deviation that occurs with the effect of all factors belonging to the Y property.

$$S_y = \sqrt{[(\Sigma (y - \bar{y})^2)]}.\frac{1}{n} = \sqrt{\left[\Sigma y^2 - \frac{(\Sigma y)^2}{n}\right].\frac{1}{n}} = \sqrt{S_{yy}} \quad [3]$$

There are four different relationships among the variables used in path analysis: direct, indirect, U, and S (Thesse effects were explained in Figure 3 and Figure 4). A path diagram can show these relationships using path coefficients (Mahapatra et al., 2020). One-way arrows are used in path diagrams. These straight arrows indicate relationships from each independent variable to the dependent variable. Two-way combining curved arrows between two variables represent correlations between variables not dependent on others in the system. Symbolic or numerical values of path coefficients are written on the diagram. In two-way curve arrows, the symbolic or numerical values of the simple correlation coefficients are written. In this study, direct and indirect effects are emphasized. The direct effect is the effect that a variable has on another variable.

The direct, indirect, U, and S effects between the variables are explained in Figures (Fig.1, Fig.2, Fig.3 and Fig.4).

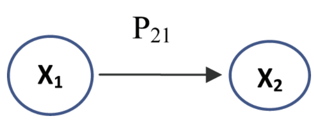

Fig. 1

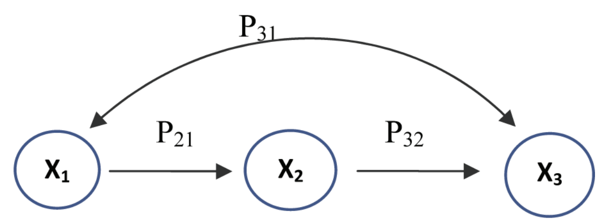

Fig. 2

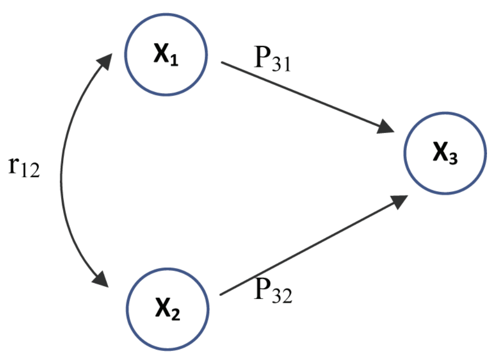

Fig. 4

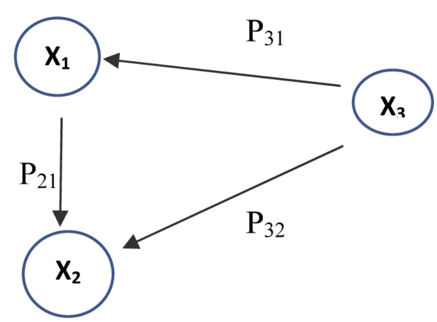

Fig. 3

İşci Güneri et al. (2015)

In Figure 1, the path coefficient P_{21}, which shows the direct effect of the first variable on the second variable, is equal to the correlation coefficient between the two variables [r_{12}=P_{21}].

Since indirect effects can emerge besides direct effects between the variables, the first variable's direct effect on the third variable (P_{31}) in Figure 2 is not equal to the correlation between variables as seen in the equation [$r_{12} = P_{21}$]. Therefore, the coefficient of correlation between the first and third variables equals to the sum of the direct and indirect effects. This effect is determined with the help of the following equation.

$$r_{31} = DE+IE=P_{31} +P_{21}.P_{32}$$

The effect that occurs when there is a mutual relationship between the cause variables is named the U effect (Fig. 3). The U effect was observed between the X_1 and X_3 variables (Fgure 4). Here, while a mutual interaction existed between the X_1 and X_2, the X_1 variable directly affected the X_3 variable. The path coefficient between the X_1 and X_3 constituted the direct effect of X_1 on X_3 (DE = P_{31}).

Multiplying "the correlation coefficient between the X_1 and X_2 variables" by "the path coefficient showing the direct effect of X_2 on X_3" yielded the U effect of the X_1 variable on X_3 via X_2. The sum of the "direct effect" and the "U effect" was equivalent to the correlation (r_{13}) between variables X_1 and X_3 (r_{13} = DE+UE=P_{31} +r_{12}.P_{32}). A similar situation occurred in the interaction between the variable X_2 and the variable X_3. (r_{23} = DE+UE=P_{32} + r_{12}.P_{31}).

The effect that occurs in the presence of a common cause variable affecting two variables is called the S effect (Fig. 4). The X_2 variable affected the X_3 variable in two ways. The first effect was the direct effect of the X_2 variable on the X_3 variable (P_{32}). The other effect was the S effect caused by the variable X_1, which was a common-cause variable affecting both X_2 and X_3 variables. The Path coefficient between the X_1 and the X_3 variables was equal to the direct effect of X_1 on X_3 (DE= P_{31}).

Multiplying "the Path coefficient showing the direct effect of X_1 on X_2" by "the path coefficient showing the direct effect of X_1 on X_3" and by "the path coefficient showing the direct effect of X_1 on X_2" yielded the S effect resulting from the impact of X_2 on X_3. (SE = P_{21} x P_{31}) The correlation coefficient between the X_2 and X_3 variables produced the sum of these effects. r_{23} = DE+SE=P_{32} +P_{21}.$P_{31.}$

The following equations, consisting of path and correlation coefficients, were created using the diagrams (Li, 1975). By utilizing these equations, direct and indirect effects are determined.

Correlation coefficients between LMY and LL, CAGE, and DMY can be distributed into direct and indirect effects as follows:

$r_{Y1} = P_{Y1} + r_{12}P_{Y2} + r_{13}P_{Y3}$

$r_{Y2} = P_{Y2} + r_{12}P_{Y1} + r_{23}P_{Y3}$ [4]

$r_{Y3} = P_{Y3} + r_{13}P_{Y1} + r_{23}P_{Y2}$

In equations, the P_{Yi} shows the path coefficient between the i^{th} independent variable and the dependent variable as Y (direct effect); the $r_{ij}P_{Yi}$ shows the effect of the i^{th} independent variable on the dependent variable Y through the j^{th} independent variable (indirect effect); the r_{Yi} shows the correlation coefficient between the Y and the i^{th} independent variable; the r_{ij} expresses the coefficient of correlation between independent variables. By specifying the direct and indirect effects, the correlation coefficient between Y and X_i is determined. In each equation with n independent variables, there are (n-1) indirect effects and one direct effect. The shares of direct and indirect effects between Y and X_i within the total correlation constitute the influence share.

The above equations show that the total correlation was divided into direct and indirect effects. The sum of the indirect effects having connections with variables and the direct effects of independent variables constitutes the correlation coefficient between the variables in question. Simple correlation coefficients between dependent and independent variables were determined and placed in the above equations [4]. By solving the equations with four unknowns, the path coefficients were obtained with the help of the equation [5].

$$\begin{pmatrix} r(X_1Y) \\ r(X_2Y) \\ r(X_3Y) \end{pmatrix} = \begin{pmatrix} 1 & r_{12} & r_{13} \\ r_{21} & 1 & r_{23} \\ r_{31} & r_{32} & 1 \end{pmatrix}^{-1} * \begin{pmatrix} P_{Y1} \\ P_{Y2} \\ P_{Y3} \end{pmatrix} \quad Z=X*Y^{-1} \ [5]$$

Equation [5] is formed by multiplying the path coefficients vector (Z), the inverse of the correlation matrix (Y) between the cause variables, and the vector (X) created from the correlations between the cause variables and the result variable.

RESULTS AND DISCUSSION

Direct effects between variables are obtained by performing correlation analysis. In a quantitative character, the path coefficient of the observed variable is expressed as the part of the observed variation sourcing from only one variable. Through path analysis, direct and indirect effects between dependent and independent variables can be revealed in detail, and these relationships can be shown in a path diagram. The path coefficient expresses the relationship of any quantitative character with its factors, and the determination coefficient is defined by squaring the path coefficient. (Düzgüneş et al., 2012; Mahapatra et al., 2020). In this study, direct and indirect effects of the LL (X_1), CAGE (X_2), and DMY (X_3) on LMY (Y) were determined by path analysis.

The simple and partial correlation coefficients between the LMY(Y) presented by the dependent variable in the study and the LL (X_1), the CAGE (X_2), and the average DMY (X_3) introduced by the independent variables are summarized in Table 1.

It was detected that simple and partial correlation coefficients between LMY (Y) and LL (X_1), CAGE (X_2), and DMY (X_3) were positive. The highest correlation was determined as 0.697 between LMY(Y) and LL (X_1), while the lowest positive correlation was determined as 0.014 between the CAGE (X_2) and LL

Table 1. Simple (lower diagonal), partial (upper diagonal) correlations and significance levels between variables in the study

	Y	X_1	X_2	X_3
Y	1	0.781**	0.043*	0.769**
X_1	0.697**	1	-0.025	-0.585**
X_2	0.053**	0.014	1	-0.008
X_3	0.616**	0.038*	0.039*	1

**p<0.001, *p<0.005 LMY (Y), LL (X_1), CAGE (X_2), and DMY (X_3)

(X_3). In the study, it was determined that the relationship between the LL (X_1) and DMY (X_3) is positive and significant ($p<0.005$). The relationship between LL and DMY was consistent with the value determined by Orhan and Kaşıkcı (2002) for Brown Cows but lower than the value for Holstein cows. Contrary to the research finding, Aytekin et al. (2016) found a negative correlation between LL and DMY in Holstein cows.

Simple correlations between LMY (Y) and LL (X_1), CAGE (X_2), and DMY (X_3) were determined as 0.697, 0.053, and 0.616. In this study, the relationship between LMY and LL was higher than the findings of Malhado et al. (2013), while the relationship between LMY and DMY was lower than the value determined by Şahin et al. (2013).

In the simple correlation analysis, the positive and significant correlation between LMY and DMY was still detected as positive and significant when removing the LL and CAGE effects.

The simple correlation analysis reveals that the positive and significant relationship between CAGE and LMY was still positive and significant when taking out the lactation duration and DMY effects. The positive correlation between LL and CAGE was negative when eliminating the effects of other variables. The positive and significant relationship between LL and DMY was negative and meaningful when removing the CAGE effect.

In their study, Orhan and Kaşıkcı (2002) found the correlation between CAGE and DMY for Holstein and Brown cows to be -0.066 and 0.144, respectively. However, the positive and significant relationship between DMY and CAGE was negative when eliminating the LL effect.

In the research, the direct determination coefficients were determined by taking the squares of the path coefficients. When the direct determination coefficients were examined, the LMY in Anatolian buffaloes was affected mainly by the LL (0.378), later by the DMY (0.350), and finally by the CAGE (0.0044), which was the lowest. The standardized multiple regression equation for LMY (Y), LL (X_1), CAGE (X_2), and DMY (X_3) of Anatolian buffaloes are given below;

$$Y_1 = 0.615\ X_1 + 0.021\ X_2 + 0.592\ X_3 \qquad R^2: 0.871$$

In this equation, where the partial regression coefficients explain the direct effects of each variable on the outcome variable, the value of coefficient (a) is zero since the coefficients are standardized. The coefficients in the equation express the direct effects of the independent variables on the dependent variable, that is, the path coefficients.

The determination coefficients (87.1%) for the LMY of the independent variables calculated in the study are significant. In this case, we can state that the independent variables examined have explained 87.1% of the variation observed in LMY. Standardized regression coefficients and significance levels between dependent and independent variables are presented in Table 2.

All the coefficients were found to be statistically significant. Since VIF values are below ten thresh-

Table 2. Standardized regression coefficients (b_1, b_2, b_3) and statistical levels.

Parameters	X_1	X_2	X_3
Cofficiens	0.615	0.021	0.592
Possibility	0.000	0.006	0.000
Tolerance	0.998	0.998	0.997
VIF value	1.002	1.002	1.003

independent (X_1, X_2, and X_3) variables, LMY (Y), LL (X_1), CAGE (X_2), and DMY (X_3)

olds, no multicollinearity was observed between variables (Table 2) (Önder and Abacı, 2015).

The direct and indirect effects of the independent variables of the LL (X_1), the CAGE (X_2), and the DMY (X_3) on the dependent variable of the LMY (Y) are seen in the path diagram in Figure 3.

The path analysis determined that the variables of the LL (X_1), DMY (X_3), and CAGE (X_2) were efficient on LMY (Y). The study findings were similar to Banık and Tomar's (2003) findings evaluating the Murrah buffaloes' yield records. In another study examining the indirect and direct effects of the LL, 305 DMY, age, service period, and DMY on lactation milk yield, the impacts of the LL, 305 DMY, and DMY on LMY were found significant (Orhan and Kaşıkçı, 2002). In a study using the Brown Swiss cows' records, the effects of LL and 305 DMY on LMY were determined by path analysis and, it was determined that the impact of LL and 305 DMY on the LMY were significant (Tahtalı et al., 2011).

The relationships between variables have been better revealed by showing the correlation coefficients and path coefficients between dependent and independent variables on the curves and arrows making up the diagram. In the study, the path coefficients related to direct and indirect effects between lactation milk yield, LL, CAGE, and DMY are given in Table 3.

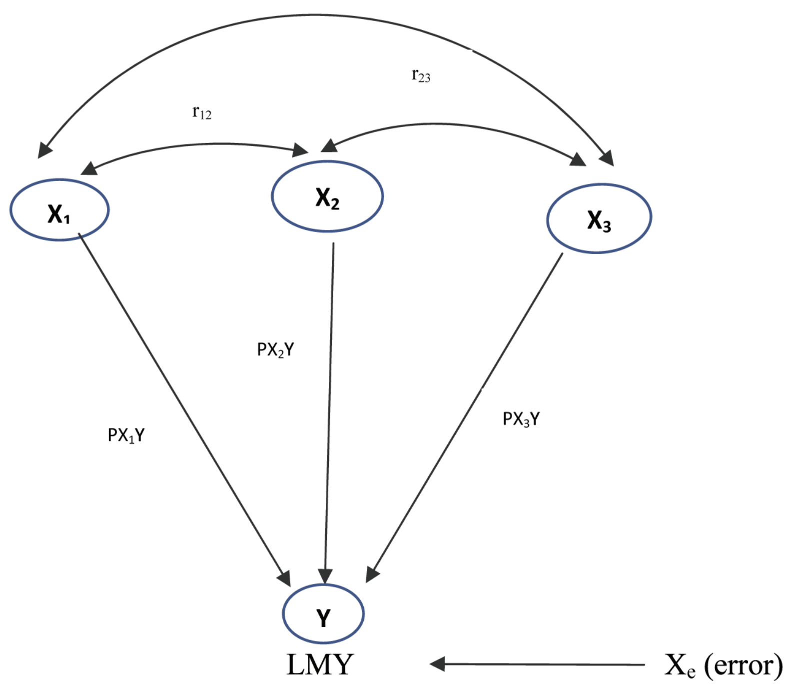

Figure 3. Path diagram for dependent (Y) and independent (X_1, X_2, and X_3) variables

Tablo 3. Path coefficients related to direct and indirect effects between LMY (Y), LL (X_1), CAGE (X_2) and DMY (X_3)

Direct effect		Indirect effect		r	CDD	P	EP (%)
				0.697	0.378	0.615	88.20
LL (X_1)	0.615	X_2	-0.000294				0.04
		X_3	0.08196				11.75
				0.053	0.00044	0.021	39.268
CAGE(X_2)	0.021	X_1	0.00039				0.729
		X_3	0.032088				60.002
				0.616	0.350	0.592	71.77
DMY(X_3)	0.592	X_1	0.12820				15.54
		X_2	0.1047				12.69

Coefficient of Direct Determination (CDD); EP: Effect Percentages (%), dependent (Y) and independent (X_1, X_2, and X_3) variables

While the DMY had the highest and most positive indirect effect on the LMY over the LL, the DMY effect on the LMY over the CAGE was negative.

The effect of the lactation duration on LMY over the CAGE was smaller than the indirect impact of LL on LMY over average daily milk yield.

The indirect effect of CAGE on LMY over average DMY was higher than the indirect effect of CAGE on LMY over the LL.

The indirect effect of DMY on LMY over the LL was higher than the indirect effect of DMY on LMY over the calving age.

The indirect effect of the LL on the LMY over the average DMY and the indirect influence of the DMY on the LMY over the CAGE were negative.

The path analysis determined that the LL was the variable with the highest direct effect on lactation milk yield, and later the DMY followed it. The calving age was the direct factor that had the most negligible impact on the LMY.

This study determined that the most significant factor affecting the LMY is the LL, and the average DMY followed it. As the LL and DMY increase, the LMY of buffaloes also increases. Besides, the fact that there was a positive and significant correlation between the LMY and the LL and the DMY in the study has been an indicator of this. Similarly, in Holstein cows (Aytekin et al., 2016), the correlations of LMY with LL and DMY were positive and significant. The research finding was compatible with this report. While the correlations of LMY with LL and DMY were 0.74 and 0.70 in Holsteins, it was 0.64 and 0.67 in Brown cows (Orhan and Kaşıkcı, 2002).

In the study, while the direct effects of DMY and LL on LMY were highly positive, the correlations between these variables were significant ($P<0.01$). When considering the model parameters, it can be stated that when the LL changes one unit, the LMY will change 0.615 units, when the average DMY changes one unit, the LMY will change 0.592 units, and when the CAGE changes one unit, the LMY will change 0.021 units.

The correlation coefficient between LMY and LL was significant (0.697) in the study. When this result was examined together with path coefficients, the direct effect of the LL on LMY was determined as 0.615. Since the correlation coefficient consists of direct and indirect impacts, 0.615 of the relationship between LMY and LL consisted of direct effects, and the other (0.082) consisted of indirect effects.

The correlation between LMY and LL was 0.67 in Anatolian buffaloes (Öz et al., 2022), 0.56 in Nili Ravi buffaloes (Tamboli et al., 2021), 0.89 in crossbred buffaloes (Malhado et al., 2009), 0.39 and 0.71 in Murrah buffaloes (Suhail et al., 2009; Jakhar et al., 2017), and 0.72 in Murrah and Murrah crossbreeds (Rodrigues et al., 2010). The LMY and LL correlation of 0.697 in the study was higher than the values determined by Malhado et al. (2013) for Murrahs, Abo Gamil et al. (2017) for Egyptian buffaloes, Barros et al. (2016) for Murrah crossbreeds, and Tamboli et al. (2022) for Murrah crossbreeds, but it was compatible with the values determined by Rodrigues et al. (2010) for Murrah and Murrah crossbreeds and Kumar et al. (2022) for Murrah buffaloes. As is known, there is a linear relationship between LL and LMY. As the lactation period extends, the amount of milk obtained from this lactation also increases.

The direct effect of DMY on LMY was 0.592. The correlation between DMY and LMY was 0.616. It has been determined that 0.592 of the relationship between the variables in question consisted of indirect effects, and the remaining part (0.024) was indirect effects. Correlations between DMY and LMY were 0.689 (Aytekin et al., 2016) in Holstein cows. The research finding was compatible with this report.

The direct effect of CAGE on LMY was 0.021. The correlation coefficient between these two variables was 0.053 ($P<0,05$; $P<0,01$). Likewise, it can be stated that the 0.021 of the relationship between these two variables consisted of the direct effects and the other (0.032) consisted of the indirect effects.

The correlation between CAGE and LMY was positive and significant in the study. As is known, the sample size directly affects the significance test of the correlation coefficient. This situation results from the sample size. Previous studies reported the correlation between CAGE and LMY as negative in Holstein cows (Orhan and Kaşıkçı 2002) and Brown cows (Tahtalı et al., 2011), whereas a study found it as positive in Brown cattle (Orhan and Kaşıkçı 2002), contrarily.

The correlation between LL and CAGE in the study was consistent with the value determined by

Orhan and Kaşıkçı (2002) for Brown cows but higher than their measurement for Holstein cows; furthermore, higher than the value reported by another study for Brown cows (Tahtalı et al., 2011).

The indirect effect of CAGE on LMY over average DMY was higher than its direct effect. In this case, the CAGE indirectly affects the LMY, and this interaction is on the average daily milk yield.

The milk yield obtained from each buffalo is crucial from the perspective of buffalo breeders' socio-economic level and the country's economic situation. Copious milk production is a significant factor affecting the profitability of enterprises. Along with milk production, factors affecting milk production should be determined.

CONCLUSION

When more than one variable is considered in the study, the correlation coefficients may not express the relationship between the variables.

Here, it would be more beneficial to determine and evaluate the direct and indirect effects of the relations between the variables using the path analysis method.

The research findings, in which the relations between the variables are examined with comprehensive procedures, such as path analysis, will contribute to the preference of methods similar to "indirect selection" in the herd studied.

When one or more yields together are a criterion in deciding the parents of future generations in any herd, determining whether other yields -which are supposed to be effective on yield- will change the examined yield(s) will affect the success of the breeding program to be carried out in that herd.

In conclusion, in this study where the Anatolian buffaloes' yield records were evaluated, among the characteristics examined, especially considering that the LL had high shares of direct and indirect effects on lactation milk yield, and the partial correlation between LMY and LL and daily average milk yield, and the standardized regression coefficients as significant, it can be stated that considering these characteristics as selection criteria in breeding studies to be carried out in these herds will increase the success in the selection to be applied.

ACKNOWLEDGEMENT

Republic of Turkey Ministry of Food, Agriculture and Livestock, General Directorate of Agricultural Research and Policies (TAGEM/60MANDA2011-01) and Tokat Buffalo Breeders' Association supported this research. The authors were thankful the Association of Buffalo Breeders in Tokat for data. There is no conflict of interest between the authors.

CONFLICT OF INTEREST

None declared by the authors.

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