IJAS Iranian Journal of Applied Animal Science

Calculation of Inbreeding Depression Effects on Subclinical Mastitis Using Three Different Models

Research Article

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Received on: 28 Jul 2014
Revised on: 8 Dec 2014
Accepted on: 31 Jan 2015
Online Published on: Dec 2015

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ABSTRACT

Pedigree information of 3972 Holstein cows and bulls, from 1961 to 2008, was used to calculate inbreeding coefficients and their effects on production traits and subclinical mastitis of dairy farm cows in Azerbaijan province, Iran. Inbreeding was included in a linear mixed model as a class variable. Data were analyzed using the *PROC MIXED* of SAS by MIVQUE method. Results showed that average inbreeding coefficients were inferior in total population and inbred cows (0.8 and 1.3% in total population and inbred cows, respectively). Although the rate of inbreeding coefficients was low, inbreeding had decreasing effects ($P<0.01$) on milk, fat, and protein yields, while increasing effects ($P<0.01$) on prevalence of subclinical mastitis. Furthermore, animals with inbreeding coefficients between 0.59 to 1.11%showed a reduction in milk, fat, and protein yields by 2.4, 2 and 2%, respectively; whereas, animals with inbreeding coefficients between 1.11 and 1.75% showed a reduction in milk, fat, and protein yields by 2, 3.7 and 2.7%, respectively.

KEY WORDS Holstein cows, inbreeding depression, milk, prevalence, subclinical mastitis.

INTRODUCTION

Mastitis control is particularly important because of the high incidence of the disease and costs associated with the treatment. The prevalence of mastitis in the dairy cow population could be decreased by breeding cows with better resistance to udder diseases. Dutrring past decades, dairy cattle breeding focuses on improving production resultedin deteriorating of udder health (Nielsen, 2009). Inbreeding coefficient is the probability that two genes at any locus are identical by descent (Falconer *et al.* 1996). Using artificial reproductive technologies increased selection intensity by reducing the number of selected animals and therefore, reduced generation intervals by increasing the number of possible progeny (Pryce and Daetwyler, 2012). Increasing demand for artificial insemination causeda wide spread use of few elite sires. So, this event increased inbreeding and reduced genetic variability in livestock populations (Croquet *et al.* 2006; Panetto *et al.* 2010). The detrimental effects of inbreeding on economically important traits of dairy cattle have been determined in several studies. Inbreeding decreases milk production traits (Wiggans *et al.* 1995; Smith *et al.* 1998; Thompson *et al.* 2000) and increases somatic cell score (SCS) (Sorensen *et al.* 2006). Miglior *et al.* (1995) found a linear inbreeding depression in SCS by 10.5% of a phenotypic standard deviation per 0.1 increases in inbreeding coefficient. Smith *et al.* (1998) also showed a non-significant inbreeding depression in SCS. As an accurate estimation of inbreeding effects on mastitis prevalence and milk production traits is necessary for a proper breeding decision, this paper was designed to estimate the inbreeding depression effects associated with breeding decision of farmers. Accordingly, objectives of this study were 1) to assess inbreeding coefficients in Holstein cows and 2) to estimate the effects of inbreeding depression on milk production traits and subclinical mastitis prevalence based on three models presented in the paper.

MATERIALS AND METHODS

Pedigree

Pedigree information of 1114048 Holstein cows and bulls used in this study was collected by Animal Breeding Center (Karaj, Iran) from 1959 to 2010. In this work, Holstein cows that calved from 2006 to 2009 were also studied. From an original pedigree (n=1114048), the pedigree information related to 3972 Holstein cows, bulls and their calves that were born from 1961 to 2008 in dairy farms of Azerbaijan province was extracted by Pedig software (Boichard, 2007).

In the pedigree of Azerbaijan province, 82.17 and 17.82% of animals were female and male, respectively and are presented in Table 1.

Table 1 Distribution of the 3972 Holsteincattle existingin the pedigree file

No. of females	3 264
No. of males	708
No. of animals with known sire, but unknown dam	74
No. of animals with known sire and dam	3 778
No. of animals with known dam, but unknown sire	42
No. of animals with unknown sire and dam	78
Total of animals	3 972

Inbreeding coefficient (F) of the population was calculated using the Pedig software by VanRaden's method, VanRaden (1992).

Performance data

Data related to production traits (milk, fat and protein yields, and also fat and protein concentrations), (n=3403) and California mastitis test (CMT) records (n=4439) were selected from 1572 Holstein cows calving between 2006 and 2009 in dairy farms of Azerbaijan province, Iran. These data were standardized based on twice-daily milking for 305 days.

In order to remove outliers, lactation lengths shorter than 100 days or longer than 400 days were removed. Afterwards, lactation lengths were grouped into 4 classes. Data related to milk yield was recorded as twice-daily milking for 305 days. Milk samples were also collected twice a month and analyzed for fat and true protein contents by Milkoscan (Foss Electric, Hillerød, Denmark; AOAC, 1996). Then, fat and protein yields were calculated by multiplying fat and true protein contents in milk weight.

Subclinical mastitis was diagnosed by California mastitis test (CMT); in this test, the nature of coagulation and viscosity of the mixture were indicators for presence and severity of the inflammation, respectively (Harmon, 1994; Rahman *et al.* 2009). CMT was performed twice a month in the morning before milking.

Parity was categorized as 1st, 2nd, 3rd, 4th and 5th-11th groups. Year of calving were from 2006 to 2009 and seasons of calving were spring (from the middle of March to the end of June), summer (from the end of June to the end of September), autumn (from the end of September to the end of December) and winter (from the end of December to the end of March).

For milk production, data which had higher or lower than three in standard deviation of the mean were considered as outliers and so, were removed. The final data set which was used in the analyses contained 2635 milk lactation records, in which 2354 records had CMT data information. Descriptive statistics on production traits are summarized in Table 2.

Designing three models for analyzing prevalence of subclinical mastitis

Prevalence of subclinical mastitis was analyzed by usingthree designed models (Figure 1):

Model 1) Udder Quarter model: study of subclinical mastitis in each udder quarter separately: in this model, the record of each quarter was considered separate. Therefore, each udder quarter was considered as a milk production unit.

Model 2) Mammary model: study of subclinical mastitis in cows in a way that the presence of mastitis record in any or all of the udders was coded as 1, while the absence of mastitis record in all of the udders was coded as 0.

Model 3) Episode model: subclinical mastitis as episode was calculated by sum of all udder quarters with CMT.

Therefore, the sum of CMT quarter scores of an animal was considered as its episode. In other words, CMT scores of all quarters including right rear, left rear, right front and left front were added up and considered as an animal's episode.

For example, the calculation of episode for an animal with the following udder quarter scores including: right rear= 0, left rear= 1, right front= 1 and left front quarter= 0, was counted by summing up the scores (0+1+1+0=2).

Analysis

Data were analyzed by Minimum Variance Quadratic Unbiased Estimation (MIVQUE) method using Rao (1970), Rao (1971a), Rao (1971b) and Rao (1977) notation and general formulation in MIXED model of SAS program (9.2), SAS (2005). Inbreeding coefficient was considered in the model as a classification variable: F= 0, $0 < F < 0.59$, $0.59 \leq F < 1.11$, $1.11 \leq F < 1.75$ and $1.75 \leq F$.

Table 2 Descriptive statistics for milk, and fat and protein yields, and fat and protein concentrations

Traits	No. of records	Mean	SD	Minimum	Maximum
Milk yield (kg)	2635	8315	2179	1913	14184
Fat yield (kg)	2629	235.6	67.3	64.2	431.3
Protein yield (kg)	2603	278.3	71.9	76.3	489.7
Fat concentration (%)	2618	2.8	0.4	1.6	4.1
Protein concentration (%)	2552	3.3	0.1	2.9	3.8

SD: standard deviation.

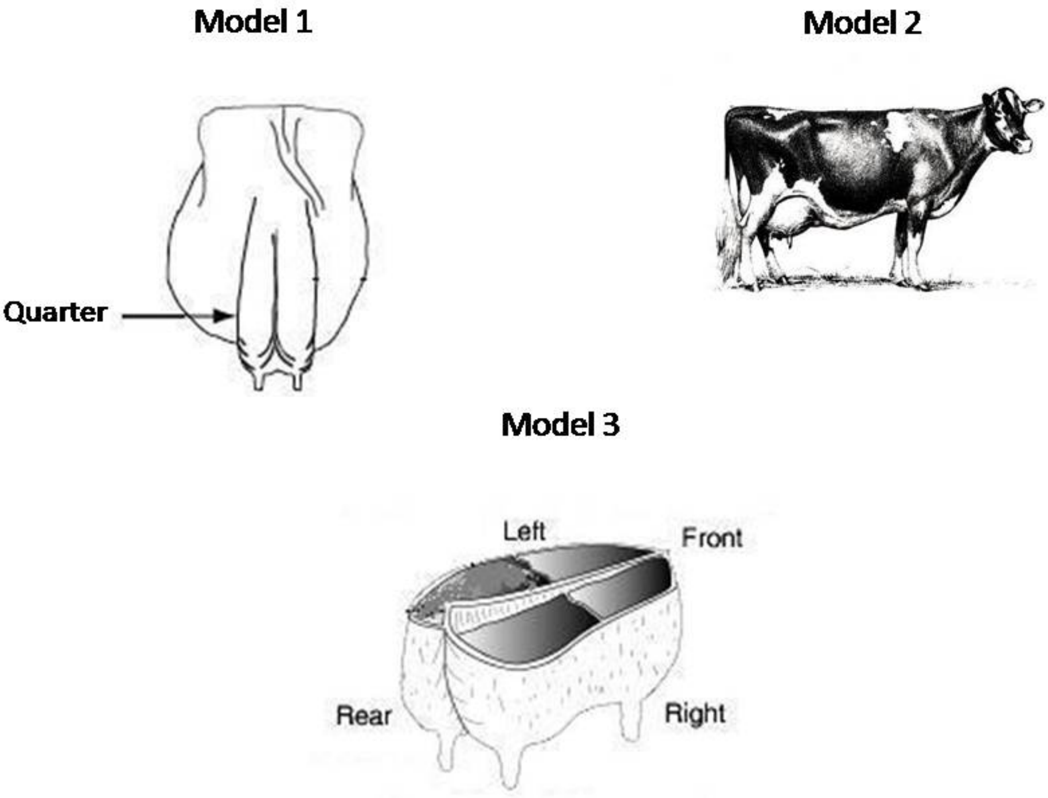

Figure 1 Schematic presentation of three models of California Mastitis Test scores: Model 1) Udder quarter model: in this model presence of subclinical mastitis in each udder quarter considered as a separate unit. Model 2) Mammary model: a study of subclinical mastitis in all udder quarters of a cow together. In this model (CMT=0, 1), healthy records coded as 0; while, the presence of at least one affected quarter of an animal with CMT coded as 1. Model 3) Episode model: the sum of CMT scores of all quarters belonging to each cow considered as its episode. (original diagram from http://agriculture.kzntl.gov.za/publications/production_guidelines/dairying_in_natal/dairy6_1.htm)

In this study, sire models were used. Furthermore, interactions between parity and inbreeding were considered in the model.

The mixed models were:

$Y_{1ijlmn} = \mu + YS_i + PAR_j + DIM_L + F_m + P_m + S_n + PAR_j \times F_m + exp_{ijlmn}$

$Y_{2ijmn} = \mu + YS_i + PAR_j + F_m + P_m + S_n + PAR_j \times F_m + exp_{ijmn}$

Where:

Y_{1ijlmn}: dependent variable for data including milk, fat and protein yields (kg) and fat and protein concentrations.

Y_{2ijmn}: dependent variable for subclinical mastitis observation in a cow.

μ: overall mean.

YS_i: fixed effect of year-season of calving.

PAR_j: fixed effect of parity.

DIM_L: days in milk.

F_m: fixed effect of inbreeding coefficient.
P_m: random permanent environmental effect.
S_n: random effect of sire.
$PAR_j \times F_m$: interaction among parity and inbreeding coefficient.
exp_{ijlmn}: experimental error for the i^{th}(1, 2, 3, 4) year-season of calving, j^{th}(1, 2, 3, 4, 5) parity, l^{th}(100 to 400 days) length of lactation, the n^{th}(1, 2, 3,…) sire and m^{th}(1, 2, 3…) animal (cow).

RESULTS AND DISCUSSION

The descriptive statistics of cow pedigree inbreeding coefficient showed that 64.03% of the cows in the data file were inbred. The maximum inbreeding coefficient in the data was 26% and the average inbreeding coefficient among the inbred cows was 1.3%. Total average inbreeding coefficient, including both inbred cows and none inbred ones, was 0.8%. When considering cows' inbreeding percentage, it was showed that the percentage of inbred cows increased more from 1996 to 2008 compared with that in 1985 to 1995. The reason for observing more inbreeding percentage from 1996 to 2008 could be because of uncontrolled inseminating of cows with semen from few excellent bulls. Average level of inbreeding coefficient increased slowly from 1985 to 2006 with an average increase of 0.2% per year. On the other hand, inbreeding coefficient of Holstein population decreased from 2006 to 2008. Because the rate of inbreeding estimated from 1961 to 1985 was observed to be zero, the percentage of inbred cows and average inbreeding coefficients were reported from 1985 to 2008. Distribution of cows by the classes of inbreeding coefficient showed that most of the inbred cows (25.4%) were in the class with inbreeding coefficients between $0.59 \leq F < 1.11\%$. Inbreeding coefficient of sires was also between zero and 25 percent.

The effects of inbreeding on milk production are presented in Tables 3 and 4. Milk, fat and protein yields decreased ($P<0.01$) with increasing inbreeding coefficient. When inbreeding coefficient was considered as a class variable, no significant differences ($P>0.05$) were observed in animals having low inbreeding coefficients between 0 and 0.59%, and also in this way of analysis the number of inbred animals turned to zero ($F=0$). On the other hand, by an increase in inbreeding coefficient from 0.59 to 1.11%, a considerable decline ($P<0.05$) was observed in milk (2.4%), fat (2%) and protein yields (2%). Also, inbreeding coefficients between 1.11 and 1.75% caused a reduction in milk, fat and protein yields by 2, 3.7 and 2.7%, respectively.

These results indicated that even small increases in inbreeding could have a negative effect on milk production in Holstein cows.

It was also showed that the effects of inbreeding on milk, protein, and fat yields varied ($P<0.01$) among different parities (Table 4). The intensity in negative effects of inbreeding on milk and protein yields was higher in multiparous animals (5≤). However, a reverse trend with fat yield was observed and it was showed that the negative effect of inbreeding on fat yield intensified in younger animals (first-parity animals). Similar to the present study, McParland *et al.* (2007) showed that inbreeding affected ($P<0.05$) protein yield and fat concentration and inbreeding effects were under the influence of parity. Therefore, the negative effect of inbreeding on protein yield was higher in older animals, while the negative effect of inbreeding on milk fat concentration was higher in younger animals (Mc Parland *et al.* 2007). In another study, Bezdicek *et al.* (2008) observed the negative effect of inbreeding on milk production of Holstein cattle population in the Czech Republic. In that study, the inbred cows with $F_x= 1.25\%$ showed -103.02 kg in milk yield, -0.003% in fat percentage, and -0.006% in protein percentage. On the other hand, Croquet *et al.* (2006) reported a negative quadratic effect of inbreeding on milk production. Thompson *et al.* (2000) found that the reduction in milk production per lactation for $F= 0.01$ was 3 kg, while the reduction in milk production per lactation for $F > 0.01$ was 35 kg. Also, for inbreeding coefficients between $F= 0.07$ and 0.10, a mass reduction in milk production (55 kg) was observed.

Inbreeding increased ($P<0.01$) prevalence of subclinical mastitis based on all three models presented in this study (Table 5), while four classes of inbreeding ($0<F<0.59$, $0.59\leq F<1.11$, $1.11\leq F<1.75$ and $1.75\%\leq F$) did not show any differencesin the prevalence of subclinical mastitis based on the mentioned models ($P>0.05$). At a time inbreeding coefficient was considered as a class variable, a significant difference ($P<0.01$) in prevalence of subclinical mastitis was observed between non-inbred animals ($F=0$) and inbred animals. Furthermore, it was showed that the effect of inbreeding on the prevalence of subclinical mastitis was significantly different ($P<0.05$) for parities based on Episode model and Udder Quarter model (left rear quarter and right front quarter) (Table 6). The negative effect of inbreeding on the three presented models for subclinical mastitis was greater in multiparous animals than younger animals. SØrensen *et al.* (2006) found nonlinear effect of inbreeding on somatic cell score (SCS), indicating that higher levels of inbreeding had more unfavorable effect than lower levels of inbreeding. Then, lower levels of inbreeding (between 0 and 0.1) had lower prevalence of subclinical mastitis than higher levels of inbreeding. Croquet *et al.* (2006) and Mc Parland *et al.* (2007) found more intensified effects of inbreeding on prevalence of subclinical mastitis in older animals.

Table 3 Analysis of milk, fat and protein yield (kg), considering inbreeding (F) as a class variable

Items (%)	Milk yield (kg)	Fat yield (kg)	Protein yield (kg)
P-value	$P < 0.01$	$P < 0.01$	$P < 0.01$
$F = 0$	8683^{a}	246.9^{a}	289.4^{a}
$0 < F < 0.59$	8593^{a}	242.6^{ab}	287.4^{ab}
$0.59 \leq F < 1.11$	8382^{b}	238.6^{b}	281.8^{b}
$1.11 \leq F < 1.75$	8212^{c}	229.7^{c}	274.2^{c}
$1.75 \leq F$	8161^{c}	230.9^{c}	272.7^{c}

Table 4 Least square means for inbreeding depression: interaction between inbreeding^{a} and parity on milk, and fat and protein yields

Item	Parity	Milk yield	Fat yield	Protein yield
$0 < F < 0.59$	1	7569.3^{c}	193.1^{b}	286.6^{bc}
	2	8992.9^{a}	240.8^{a}	295.5^{b}
	3	8613.1^{b}	246.1^{a}	306.5^{a}
	4	8310.2^{b}	243.4^{a}	283.7^{c}
	5-11	7327.8^{c}	228.6^{b}	264.9^{d}
$0.59 \leq F < 1.11$	1	7467.9^{bc}	185.5^{c}	285.4^{b}
	2	9108.1^{a}	252.1^{a}	299.5^{a}
	3	7922.8^{b}	239.1^{b}	282.1^{b}
	4	8934.3^{a}	259.1^{a}	303.1^{a}
	5-11	7110.1^{c}	200.1^{c}	239.1^{c}
$1.11 \leq F < 1.75$	1	7708.1^{bc}	194.1^{b}	290.8^{a}
	2	8839.7^{a}	245.2^{a}	290.7^{a}
	3	8196.7^{b}	228.5^{b}	293.6^{a}
	4	7881.5^{b}	218.9^{b}	267.3^{b}
	5-11	6859.8^{c}	199.1^{b}	228.71^{c}
$1.75 \leq F$	1	7503.2^{c}	192.6^{c}	280.9^{abc}
	2	8544.1^{a}	233.9^{b}	280.3^{ac}
	3	7688.7^{c}	255.3^{bc}	279.8^{a}
	4	8445.1^{b}	251.8^{a}	281.3^{ab}
	5-11	7257.9^{c}	203.4^{c}	241.5^{d}

The means within the same column with at least one common letter, do not have significant difference ($P>0.05$).

Table 5 Analysis of prevalence of subclinical mastitis while considering inbreeding (F) as a class variable for the three different models: Model 1: Udder quarter model; Model II: Mammary model and Model III: Episode model

Items	Model I				Model II	Model III
	Right rear	Left rear	Right front	Left front	Mammary	Episode
P-value	$P < 0.01$	$P < 0.01$	$P < 0.01$	$P < 0.01$	$P < 0.01$	$P < 0.01$
$F = 0$	0.0^{b}	0.09^{b}	0.16^{b}	0.36^{b}	0.08^{b}	0.50^{b}
$0 < F < 0.59$	0.13^{a}	0.34^{a}	0.59^{a}	0.78	0.21	1.83^{a}
$0.59 \leq F < 1.11$	0.0^{a}	0.14^{a}	0.28^{a}	0.65 b^{a}	0.17^{a}	1.03^{a}
$1.11 \leq F < 1.75$	0.13^{a}	0.36^{a}	0.42^{a}	0.76^{a}	0.20^{a}	1.67^{a}
$1.75 \leq F$	0.17^{a}	0.34^{a}	0.48^{a}	0.76^{a}	0.24^{a}	1.74^{a}

The means within the same column with at least one common letter, do not have significant difference ($P>0.05$).

In another study, Van Tassell *et al.* (2000) studied multiple lactations in cow and showed weaker decreasing effect of inbreeding on SCS, in a way that for each 0.01 increase in inbreeding, the regression estimate was 0.0037. Mrode *et al.* (2004) also reported lower decreasing effect of inbreeding on SCS in UK Holsteins.

Table 6 The effect of interaction between inbreeding and parity on prevalence of subclinical mastitis in left rear quarter, right front quarter, and Episode

Items	Parity	Left rear quarter	Right front quarter	Episode
$0 < F < 0.59$	1	-0.24^{c}	0.15^{d}	-0.38^{c}
	2	0.19^{c}	0.47^{cd}	1.41^{c}
	3	0.33^{bc}	0.85^{b}	2.37^{b}
	4	0.49^{b}	0.66^{bc}	2.60^{b}
	5-11	1.03^{a}	1.11^{a}	3.78^{a}
$0.59 \leq F < 1.11$	1	-0.32^{a}	-0.13^{b}	-0.74^{c}
	2	0.36^{a}	0.27^{b}	1.31^{b}
	3	0.38^{a}	0.59^{a}	2.03^{a}
	4	0.25^{a}	0.35^{b}	1.41^{ab}
	5-11	0.13^{a}	0.43^{b}	1.73^{b}
$1.11 \leq F < 1.75$	1	-0.37^{c}	-0.22^{b}	-0.86^{d}
	2	0.16^{c}	0.41^{b}	1.14^{c}
	3	0.66^{b}	0.81^{a}	3.06^{a}
	4	0.91	0.87^{a}	3.68^{a}
	5-11	0.49^{b}	0.39^{b}	1.95^{b}
$1.75 \leq F$	1	0.03^{b}	0.02^{b}	0.09^{c}
	2	0.40^{b}	0.45^{b}	1.65^{b}
	3	0.31^{b}	0.68^{a}	2.35^{ab}
	4	0.52^{ab}	0.66^{ab}	2.71^{a}
	5-11	0.66^{a}	0.66^{ab}	2.62^{a}

The means within the same column with at least one common letter, do not have significant difference (P>0.05).

In this study, each 0.03 increase in inbreeding coefficient had 1.8% phenotypic standard deviation, and then for each 0.01 increase in inbreeding, the regression estimate was 0.0039 (Mrode *et al*. 2004).

CONCLUSION

Analysis results showed that milk, fat and protein yields, and also subclinical mastitis were under the negative influences of inbreeding. It was also showed that even low levels of increase in inbreeding coefficient had a negative effect on udder health and milk production traits in Holstein cows.

ACKNOWLEDGEMENT

We would like to express our gratitude to Azarnegin Dairy Herd and Iranian Animal Breeding Center for their assistance throughout the whole research period, and for allowing us to run the present study.

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