

# Effect of Using Reproductive Technologies on Genetic Progress in Sistani Native Cattle of Iran: A Simulation Study

**Research Article** 

H. Faraji-Arough<sup>1\*</sup>, M. Rokouei<sup>2</sup>, A. Maghsoudi<sup>2</sup>, M. Mehri<sup>3</sup>, S. Ansari Mahyari<sup>4</sup> and A. Christian Sorensen<sup>5</sup>

<sup>1</sup> Research Center of Special Domestic Animals, University of Zabol, Zabol, Iran

Department of Animal Science and Bioinformatics, Faculty of Agriculture, University of Zabol, Zabol, Iran

Department of Animal Science, Faculty of Agriculture, University of Zabol, Zabol, Iran Department of Animal Science, College of Agriculture, Isfahan University of Technology, Isfahan, Iran

Department of Molecular Biology and Genetics, Center for Quantitative Genetics and Genomics, Blichers Allé 208830, Tjele, Denmark

Received on: 16 Jan 2018 Revised on: 5 Jul 2018 Accepted on: 31 Jul 2018 Online Published on: Mar 2019

\*Correspondence E-mail: hadifaraji@uoz.ac.ir

© 2010 Copyright by Islamic Azad University, Rasht Branch, Rasht, Iran Online version is available on: www.ijas.ir

### ABSTRACT

The effect of artificial insemination (AI), embryo transfer (ET) to bull dams (BD), and sexed semen (SS) to cow dams (CD) with and without controlling inbreeding were studied using stochastic simulation. Three levels of embryo transfer (no ET, ET on all BD, and ET on 20% of BD), five levels of sexed semen (no SS: control, SS-X on all CD, SS-X on 20% CD, SS-Y on all CD, and SS-Y on 20% CD), three levels of artificial insemination (no AI, AI on 50% cows, and AI on 90% cows), two levels of mating system (random and minimum consistory) were combined together to make 66 scenarios in which the combination of no ET, no SS, and no AI are assigned as a control. The simulated population consisted of 40 herds with 50 cows each was monitored for 30 years. The results showed that the use of AI, ET, and SS techniques increased the annual change of total merit index (TMI), inbreeding, and selection accuracy. Compared to control, the rate of annual change in TMI and inbreeding were increased as 41.95, 36.91 and 83.91%; and 192, 57 and 207%, for using of AI, ET and combination of SS + AI + ET, respectively. The minimum consistory mating decreased inbreeding, but not affected other parameters. The results suggested that using of ET on 20% BD, SS-Y for all CD, and 90% AI alone or in combination with each other along with minimum consistory mating might resulted in high genetic progress and low inbreeding rate. These technologies and inbreeding control strategies may increase the ratio of annual change of TMI to inbreeding.

**KEY WORDS** artificial insemination, embryo transfer, random mating, Sistani cattle.

### INTRODUCTION

Biotechnology as an efficient biological tool can be divided into two categories in animal breeding. First, reproduction techniques such as artificial insemination, embryo transfer and sex control. Second, molecular techniques that can be used to locate, identify, compare or manipulate genes through the application of DNA fingerprinting, markerassisted selection, and gene transfer (Bourdon, 1997). The reproduction technologies have main effect on the structure of breeding programs, genetic progress, and genetic trends in livestock production via increasing fertility that can be translated into the fewer parents for given number of progeny, and are useful tools to increase the effeciency of livestock reproduction and production (Ghavi Hossein-Zadeh, 2010; Kaya *et al.* 2018). This may increase the selection intensity and rate of inbreeding in the population.

The efficiency of reproduction technologies can be evaluated by the rate of genetic progress. However, different components of genetic progress, such as accuracy of prediction, generation interval, intensity of selection and genetic diversity may be variably affected by these technologies (Gengler and Druet, 2002). Identification of superior animals and their use in a population is the base of animal breeding programs, which is a two- stage operation. Genetic progress in cattle is determined by the merit males that used as sires in each generation, therefore, the selection of young bulls is an important stage in breeding programs (Andrabi and Moran, 2007).

Among reproduction technologies, artificial insemination was the first developed technology that is still used for genetic improvement in cattle. Two to 2.5 percent improvement in genetic progress was reported using of artificial insemination in dairy cattle (Van Vleck, 1981).

In this method, accuracy of selection and selection intensity would be high because of having more progeny from the superior sires and extensive use of these sires (Bourdon, 1997).

The introduction of sexed semen to commercial herds has caused to decrease the number of needed dams as replacement heifers resulting in increasing the selection intensity of dams and annual genetic gain for populations (Sørensen *et al.* 2011). The low pregnancy per artificial insemination and decreased *in vivo* embryo production could be one of the limitations using sexed semen than non-sexed semen (Filho *et al.* 2014; Seidel, 2014; Dahlen *et al.* 2014).

The researchers also used female reproduction technologies such as embryo transfer to accelerate the genetic gain in cattle breeding programs. The embryo transfer technology may increase the selection intensity on females resulting in reducing the optimal age for animal selection and the generation interval in a breeding program (Granleese *et al.* 2015). An increase in genetic progresses without increasing inbreeding has been reported in stochastic simulation by Bouquet *et al.* (2015). Use of embryo transfer can be valuable in livestock production when embryo production was obtained with low cost and surplus heifers sold from the herds (Kaniyamattam *et al.* 2017).

Sistani cow is native to the Sistan region with vast historical dimensions in the east of Iran and discovering the statue of a Sistani cow in Burnt City (Shahr-i sukhta) raised a theory that people respected domestic animals 5000 years ago (Mortazavi, 2011). Despite the importance of this ancient genetic source with notable resistance against pathogenic invaders and environmental parasites, the population of Sistani breed is at risk of being extinct due to climate changes and poor management of Iranian government. Sistani cows are classified in beef cattle category, which have a greater thermo-tolerability due to a better thermal regulatory response to higher ambient temperature than commercial breeds that can be used in traditional and natural insemination and embryo transfer programs especially in rough and tropical area (Barati *et al.* 2007).

The present study aimed to evaluate the some reproductive technologies, including artificial insemination, embryo transfer, and sexed semen on genetic progress in Sistani native cattle through the stochastic simulation approach.

# MATERIALS AND METHODS

In this study, the simulations were designed based on the breeding structure of Sistani beef cattle, in which the average of traits, reproductive information, and the range of animal age were implemented in the arrestee drug abuse monitoring (ADAM) program (Pedersen *et al.* 2009) with various strategies including 50 replications for a period of 30 years. The reproductive cycles and annual random culling for four selection paths were set at one year and 15%, respectively. Response of selection, rate of inbreeding, mean of generation intervals were measured for the population during the years 10 to 30, because it was assumed that equilibrium with respect to the Bulmer effect occurs at year 10 (Sørensen *et al.* 2011).

### **Population structure**

The base population of each iteration was created according to the reproductive age of males and females. Sex and five age classes were considered for males and females, respectively. The simulated population size was 2000 cows that distributed among 40 herds with 50 heads. The selection paths for the simulated population were young bull (YB), active sire (AS), bull dam (BD) and cow dams (CD).

Every year, three AS were selected from across 20 YB that progeny tested within the simulated population. The best females across all ages and herds were as a BD. In this study, a truncation selection was used for selection of 50 BD every year, that is based on the total merit index. Bull dams and CD supplied as replacement heifers.

#### Selection

In the selection paths, selection of animals was performed as truncation selection and according to the total merit index. Total merit index consisted of birth weight (BW) and yearling weight (YW) traits. The equal economic value was used for all traits.

Selection of young bulls and active sires were performed when they were one year old, and five years old, respectively. The age range of two and eight years old was considered in order that dams became selection candidate. Selection of three selection paths, including young bulls, AS and BD were performed across the simulated population, whereas it was within individual herds for the CD. The selection criterion for all paths was estimated breeding value (EBV) that was predicted using best linear unbiased prediction (BLUP) by multivariate model:

 $yi = X_i b_i + Z_i a_i + e_i$ 

#### Where:

yi: vector of observations for  $i^{th}$  trait (birth weight and yearling weight).

 $b_i$ : vector of fixed effects for  $i^{th}$  trait, which include herd, year, and season. This effect was created in simulations by assigning animals randomly to one of the four seasons within herds and year (Sørensen *et al.* 2011).

a<sub>i</sub>: vector of additive genetic.

e<sub>i</sub>: residual effects.

Estimation of breeding value for traits was performed by DMU software (Madsen *et al.* 2006), which is linked with ADAM program.

The initial variance- covariance components for birth weight and yearling weight were based on values that estimated from Sistani data for these traits. The structure and size of simulated herds were similar to usual size and structure in the regions used for simulation of observations.

After estimation of breeding value for each two traits, the total merit index was calculated using the breeding value and economic weights of traits.

#### Scenarios

In order to answer the hypotheses, we investigated 66 scenarios combining the three reproductive technologies, including embryo transfer (ET), sexed semen, and artificial insemination (AI) as well as two levels of mating type (random and minimum consistory).

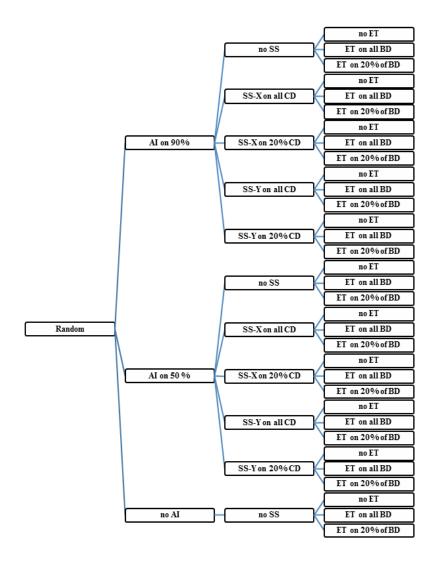


Figure 1 Diagram of combination reproductive technology levels for formation of scenarios in random mating

The combination of reproductive technology levels of random mating was shown in Figure 1 (33 scenarios for random, 33 for minimum consistory). Three levels of ET were No ET, use of ET on all BD, and use of ET on 20 % BD. Within each level of ET, fifth strategies of sexed semen were evaluated, including the use of conventional semen on CD, Y-semen on CD, X-semen on CD, and Ysemen and X-semen on 20% CD. The AI was used at three levels including no AI, AI on 50% cows, and AI on 90% cows.

The type of semen was conventional when the AI was not used. All BD had one calf per year when ET was not used. Four calves per year for BD was considered with using of ET (Galli *et al.* 2003). The accuracy of calves' sex for X-sorted semen was considered 90% (Seidel and Garner, 2002; Bodmer *et al.* 2005; Borchersen and Peacock, 2009), thus sex ratio (F/M) for conventional, Xsorted, and Y- sorted semen was considered as 50:50, 90:10 and 10:90, respectively.

#### Data analysis

The average true breeding values for total merit, birth weight (BW) and yearling weight (YW), coefficient of inbreeding, and the average generation interval for all calves born in the years of 10 to 30.were computed for all simulation iterations. The average annual change of total merit, inbreeding was calculated as regression of average total merit, inbreeding in any simulation year for the period of 30 years. The accuracy of the index was obtained from the correlation between the selection index and the aggregate genotype of selection candidates before selection (Pedersen et al. 2012). Both accuracy and the generation interval were averaged over the years and replicates (Buch et al. 2012). The selection intensity of selection paths was estimated as ratio of number of selection animal to candidate animal for any year and was averaged over years and replicates.

Statistical comparisons of genetic gain, inbreeding, generation interval, accuracy of selection in four paths between levels of factors, and between technologies with or without control inbreeding were performed using R-3.4.3 software (Team R Core, 2017).

### **RESULTS AND DISCUSSION**

The effect of ET on the annual change of TMI, BW, YW, generation interval, selection accuracy of all groups, and selection intensity for YB and BD were significant (Table 1). The using of embryo transfer increases the inbreeding, but it is not significant. The rate of increasing annual change of TMI, inbreeding, BW and YW and decreasing generation interval on 20% using ET rather than non-using

ET were 18.61, 6.84, 20.15, 17.79 and 0.27 %, respectively. The selection accuracy for YB and BD became higher when ET used for all BD and 20% BD. Maximum selection accuracy for all paths was achieved with the use of ET for 20% BD that result in a higher annual change of TMI in 20% using ET (Table 1).

Figures 2 and 3 showed the trend of TMI and inbreeding in a different pattern of ET indicating that no differences were observed between levels in first 10 years. After year 10, some differences TMI between levels were increasingly appeared by increasing year, in which the highest and lowest rates of TMI were achieved for ET on 20% BD and non-using of ET, respectively. Using the ET on all BD resulted in increasing the rate of inbreeding in all years (Figure 3).

In all years, the inbreeding difference between ET scenarios was higher in the case of ET on all BD than other options. Finally, the mean of inbreeding for ET on all BD and no ET reached to 0.27 and 0.25 in last year, respectively.

The using of SS increased annual change of TMI, in which the increasing rate of annual change of TMI for Ysemen was more than that for X-semen (Table 2). Maximum of annual change of TMI, BW, YW, and selection accuracy for all paths were observed for SS-Y on all CD with lowest inbreeding rate. The annual change for inbreeding was increased with the use of SS rather than no SS, except for SS-Y on CD. The significant increase in generation interval was observed in the case of SS-Y (Table 2).

Annual change of BW and YB had some differences among different levels of SS, so that the minimum annual change of YW and BW were observed in the cases that SS was not used for CD. Selection accuracy for all paths was highest in SS-Y for CD and it was lower for X-semen rather than no SS.

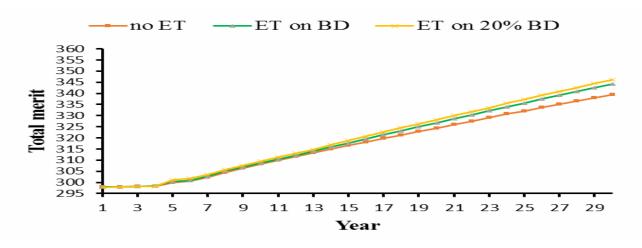
The total merit trend for different levels of the SS was shown in Figure 4. In all years, minor differences were observed among different levels of SS. Minimum and maximum total merit index were achieved for no SS and SS-Y on all CD, respectively. However, these differences between levels of SS became wide and visible in later years, except for SS-X and SS-Y on 20% CD. The use of SS-X in CD increased the inbreeding mean in all years and differences between SS-X for all and 20% CD, SS-Y for 20% CD with no SS scenarios were significant (Figure 5). In last year, the amount of difference inbreeding of no SS and SS-X for CD was around 0.7.

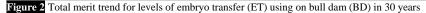
Effects of AI on the annual change to TMI, inbreeding, BW and YW and generation interval, selection accuracy, and intensity for selection groups were presented in Table 3.

T.	Embryo transfer					
Item	No ET	ET on BD	ET on 20% BD			
Annual change of TMI	1.499 <sup>b</sup>	1.725 <sup>a</sup>	$1.778^{a}$			
Annual change of inbreeding	0.906	0.970	0.968			
Generation interval	4.733 <sup>a</sup>	4.495 <sup>c</sup>	4.673 <sup>ab</sup>			
Annual change of BW	0.134 <sup>b</sup>	0.159ª	$0.161^{a}$			
Annual change of YW	1.377 <sup>b</sup>	1.569 <sup>a</sup>	$1.622^{a}$			
Selection accuracy of YB	0.205 <sup>b</sup>	0.228 <sup>a</sup>	$0.232^{a}$			
Selection accuracy of AS	$0.588^{\circ}$	0.638 <sup>b</sup>	$0.680^{a}$			
Selection accuracy of BD	0.267 <sup>b</sup>	0.283 <sup>a</sup>	$0.288^{a}$			
Selection accuracy of CD	$0.240^{b}$	$0.278^{a}$	$0.276^{a}$			
Selection intensity of YB	2.187 <sup>b</sup>	2.315 <sup>a</sup>	2.209 <sup>b</sup>			
Selection intensity of AS	2.654	2.654	2.654			
Selection intensity of BD	2.255°	2.322 <sup>b</sup>	$2.390^{a}$			
Selection intensity of CD	0.447	0.464	0.500			

 Table 1
 Annual changes to total merit index (TMI), inbreeding, birth weight (BW), yearling weight (YW), average generation interval and accuracy and intensity selection for young bull (YB), active sire (AS), bull dam (BD), cow dam (CD) in different levels of embryo transfer (ET)

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





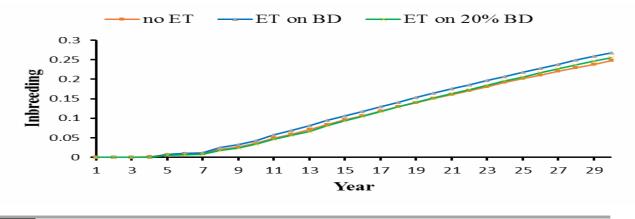


Figure 3 Inbreeding trend for levels of embryo transfer (ET) using on bull dam (BD) in 30 years

The use of AI up to 90% increased the annual change of TMI, inbreeding, annual change of BW and YB, generation interval and selection accuracy, in which the annual change to TMI and inbreeding rate were increased 24.93 and 125%, respectively. The generation interval was increased by inc-

reasing the rate of AI. Since the selection of AS for AI depends on progeny test of YB and the age of AS on AI ranged from five to 14 years, the superior active sire may be used for longer time and consequently leading to increasing generation interval.

Item	No SS	SS-X on all CD	SS-Y on all CD	SS-X on 20% CD	SS-Y on 20% CD
Annual change of TMI	1.501 <sup>c</sup>	1.516 <sup>c</sup>	2.031 <sup>a</sup>	1.639 <sup>b</sup>	1.649 <sup>b</sup>
Annual change of inbreeding	0.950 <sup>a</sup>	0.973 <sup>a</sup>	$0.847^{b}$	0.990 <sup>a</sup>	0.981 <sup>a</sup>
Generation interval	4.456 <sup>b</sup>	4.241 <sup>c</sup>	5.688 <sup>a</sup>	4.319 <sup>bc</sup>	4.464 <sup>b</sup>
Annual change of BW	0.136 <sup>d</sup>	$0.140^{d}$	0.181 <sup>a</sup>	0.149 <sup>bc</sup>	0.151 <sup>b</sup>
Annual change of YW	1.372 <sup>c</sup>	1.386 <sup>c</sup>	1.844 <sup>a</sup>	1.507 <sup>b</sup>	1.506 <sup>b</sup>
Selection accuracy of YB	0.203 <sup>bc</sup>	0.199 <sup>c</sup>	0.302 <sup>a</sup>	0.191°	0.212 <sup>b</sup>
Selection accuracy of AS	0.567 <sup>c</sup>	0.572 <sup>bc</sup>	0.796 <sup>a</sup>	0.619 <sup>b</sup>	0.623 <sup>b</sup>
Selection accuracy of BD	0.235 <sup>c</sup>	$0.188^{d}$	$0.486^{a}$	0.227 <sup>c</sup>	0.261 <sup>b</sup>
Selection accuracy of CD	0.232 <sup>b</sup>	0.192 <sup>c</sup>	$0.444^{a}$	0.229 <sup>b</sup>	0.226 <sup>bc</sup>
Selection intensity of YB	2.431 <sup>a</sup>	1.853 <sup>d</sup>	2.088 <sup>c</sup>	2.348 <sup>b</sup>	2.464 <sup>a</sup>
Selection intensity of AS	2.655ª	2.654 <sup>a</sup>	2.652 <sup>b</sup>	2.654 <sup>a</sup>	2.654 <sup>a</sup>
Selection intensity of BD	2.461 <sup>ab</sup>	2.521ª	1.731 <sup>d</sup>	2.445 <sup>b</sup>	$2.402^{bc}$
Selection intensity of CD	0.385 <sup>d</sup>	0.624 <sup>b</sup>	$0^{\rm f}$	0.764 <sup>a</sup>	0.579 <sup>bc</sup>

 Table 2
 Annual changes to total merit index (TMI), inbreeding, birth weight (BW), yearling weight (YW), average generation interval, accuracy and intensity selection for young bull (YB), active sire (AS), bull dam (BD), cow dam (CD) in different levels of sexed semen (SS)

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

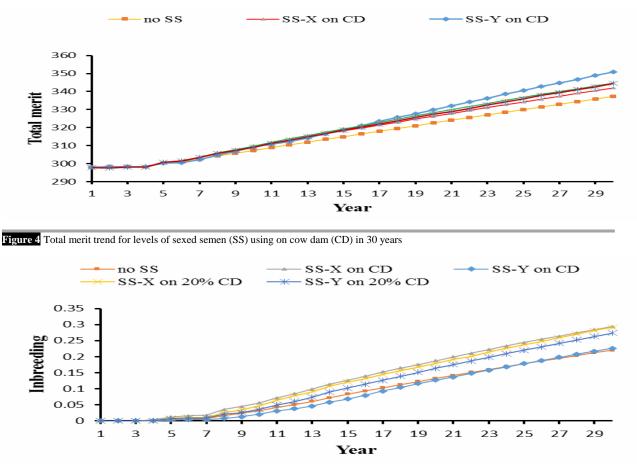


Figure 5 Inbreeding trend for levels of sexed semen (SS) using on cow dam (CD) in 30 years

The total merit index trend for no AI was lower than that in AI cases for all years (Figure 6). The difference between TMI in 90% AI and no AI increased by increasing year and using AI resulted in higher TMI than no AI group (Figure 7) as well as low inbreeding rate in no AI, in which the difference between no AI and 90% AI became 0.16 in the last year. Table 4 showed the effect of different reproductive technologies on annual changes to TMI, inbreeding, and average generation interval, either in combination or single scenarios. Using of AI and ET increased annual changes to TMI as 41.95 and 36.91%, respectively. However, the use of only ET was caused to lower the annual change of inbreeding rather than AI, in which annual change of

Inbreeding increases 192% and 57% for AI and ET, respectively (Table 4). Using of AI along with the SS and ET increased more annual change of TMI and inbreeding than non-using technologies (P<0.05). The effects of adding AI and SS along with ET on the annual change of TMI and inbreeding were significant (P<0.05). The use of reproductive technologies resulted in 83.91 and 207% more annual change of TMI and inbreeding, respectively, than non-using of those technologies (P<0.05).

As shown in Table 4, the using of AI, either only or in combination with other technologies, increased generation interval (P<0.05). Increment of annual change of BW and YW was observed using reproductive technologies either only or in combination. The selection accuracy for all paths was much more for technology-treated cases than non-treated ones (P<0.05). Differences between levels of selection accuracy were significant for YB and AS but not for BD and CD.

Annual change of TMI for random mating was higher than that for minimum consistory mating, but significant difference was not observed between two mating types (Table 5). Although minimum consistory mating had a low annual change of TMI, it was produced low inbreeding rate in which had 26% lower inbreeding rate than random mating. Since the annual change of TMI and inbreeding in random mating were much more than those in minimum consistory, the annual change of TMI to inbreeding ratio (G/F) as a criterion for comparison of two mating systems was 1.52 and 2.09 for random and minimum consistory mating, respectively.

This indicates that, minimum consistory mating was better than random mating. The effect of mating system on generation interval, annual change of BW and YB, and selection intensity of four paths was not significant. The higher selection accuracy for selection paths in random mating than minimum consistory mating might be due to the limitation of minimum consistory mating.

The annual changes to TMI, inbreeding, and G/F (annual changes to TMI/ inbreeding) ratio in different reproductive technologies for random and minimum consistory mating systems were presented in Table 6. The annual changes of TMI for random mating were more than those for minimum consistory mating in all reproductive technologies (exception of SSX and SSY for all CD), however, no significant differences were detected between two mating systems. In all reproductive technologies, minimum consistory mating system decreased the inbreeding (P<0.05) and G/F ratios for minimum consistory were more than those for random mating in all scenarios, along with a significant effect of mating system (P<0.05).

The main effect of mating system on different reproductive technologies was observed for annual change

of inbreeding (Table 7). The G/F ratio of the minimum consistory mating system was more than that for random mating, especially for SS + AI, ET + AI and ET + SS + AI scenarios. Moreover, the effect of mating system on G/F ratio was significant (P<0.05).

In this study, we examined the effect of reproductive technologies (i.e., ET, SS and AI) on genetic progress with or without control inbreeding. The results showed that using of ET for BD had a significant effect on the annual change of TMI, BW and YW but it was not significant for inbreeding. Increasing of selection intensity for YB and BD, selection accuracy for all selection paths and decreasing of generation interval could be caused of increasing genetic progress. Using of ET on 20% BD (the actual use of ET in modern dairy cattle breeding) increased annual change of TMI, BW and YW more than ET on all BD. The using of ET increased the number of YB candidates and forasmuch as the number of selected YB was same in all scenarios, thus the ratio of YB decreased and selection intensity of YB increased.

The using of ET had 36.91 and 57% more annual change of TMI and inbreeding rather than non-using ET, respectively (Table 4). With control inbreeding, the using of ET increased genetic progress while it produced low inbreeding (Table 6).

Pedersen *et al.* (2012) reported that ET reduced the number of selected animals as fewer BD were required, thus leading to increased selection intensity and, consequently, genetic gain. Increasing of inbreeding, accuracy and selection intensity using the ET were observed in other studies that agree with the result of this study (Pedersen *et al.* 2012). Improving reproduction rates through the multiple ovulation and embryo transfer can lead to an increase the selection response due to the increased selection intensity and decreasing generation interval (Villanueva *et al.* 1994). Nine and 26% increments for genetic progress and inbreeding as well as 3.9% decrement for generation interval using the ET were observed by Sørensen *et al.* (2011).

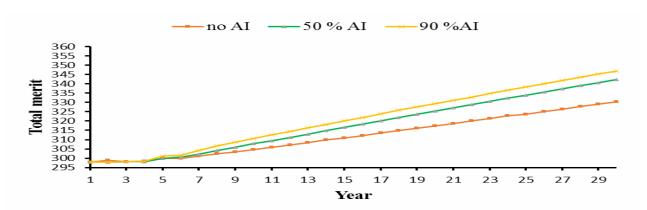
The ET considerably raised the genetic progress and inbreeding (Table 4) that was in line with others (Sørensen *et al.* 2011). The similar improvement in inbreeding and genetic progress was observed using the ET by Abdel-Azim and Schnell (2007), Ghavi Hossein-Zadeh (2010) and Granleese *et al.* (2015).

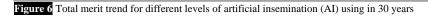
The use of embryo transfer could increase reproductive efficiency of the cow, therefore each of the cow can produced 100 calves rather than 5-7 calves in a lifetime. Increase of producing calves per cow increased selection intensity and accuracy of selection path and monitor genetic progress over time (Bouquet *et al.* 2015; Hansen and Siqueira, 2017; Kaya *et al.* 2018).

Table 3 Annual changes to total merit index (TMI), inbreeding, birth weight (BW), yearling weight (YW), average generation interval, accuracy and	i
intensity selection for young bull (YB), active sire (AS), bull dam (BD), cow dam (CD) in different levels of artificial insemination (AI)	

Item	No AI	50% AI	90% AI
Annual change of TMI	1.452 <sup>c</sup>	1.736 <sup>b</sup>	1.814 <sup>a</sup>
Annual change of inbreeding	0.527 <sup>b</sup>	1.132 <sup>a</sup>	1.185 <sup>a</sup>
Generation interval	3.314 <sup>c</sup>	4.874 <sup>b</sup>	5.713ª
Annual change of BW	0.137 <sup>c</sup>	0.155 <sup>b</sup>	0.163ª
Annual change of YW	1.314 <sup>c</sup>	1.588 <sup>b</sup>	1.666 <sup>a</sup>
Selection accuracy of YB	0.206 <sup>b</sup>	0.238 <sup>a</sup>	0.220 <sup>b</sup>
Selection accuracy of AS	$0.617^{ab}$	$0.610^{b}$	$0.680^{a}$
Selection accuracy of BD	0.275	0.284	0.280
Selection accuracy of CD	0.255	0.263	0.276
Selection intensity of YB	2.239	2.234	2.238
Selection intensity of AS	2.531	2.531	2.531
Selection intensity of BD	2.319	2.313	2.313
Selection intensity of CD	0.466	0.459	0.484

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





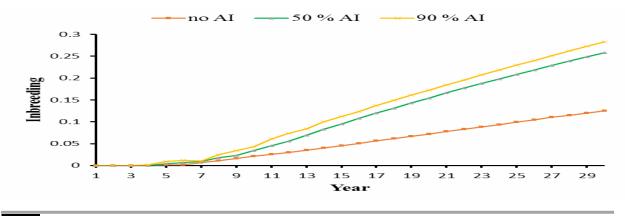


Figure 7 Inbreeding trend for different levels of artificial insemination (AI) using in 30 years

Different factors may be affected to lower outcomes of embryo transfer. These factors, including those animal related, environment, and management effects. For example, heat stress, the inefficiency of estrus detection in herds, are the main management problems that affected on embryo transfer efficiency. The downer cows are commonly high producing cows, which display shortened stress periods and less clearly behavioral signs of estrus (Walsh *et al.* 2011). Benefit of sexed semen on genetic progress depends on which part of dam population is inseminated with sexed semen and what type of semen is used for dams (Sørensen *et al.* 2011).

The use of Y-semen on all and 20% of CD was more effective than other types of semen. Maximum of annual change of TMI, BW, YW, generation interval, selection accuracy for all paths were observed when Y-semen was used in all CD. 

 Table 4
 Effect of different reproduction technology on annual changes to total merit index (TMI), inbreeding, birth weight (BW), yearling weight (YW), average generation interval, accuracy and intensity selection for young bull (YB), active sire (AS), bull dam (BD), cow dam (CD) in all scenarios

Itom				Scenario		
Item	NO	AI	ET	SS + AI	ET+AI	ET + SS + AI
Annual change of TMI	1.032 <sup>d</sup>	1.465 <sup>b</sup>	1.413 <sup>c</sup>	1.653 <sup>b</sup>	1.680 <sup>b</sup>	$1.898^{a}$
Annual change of inbreeding	$0.382^{d}$	1.114 <sup>a</sup>	0.602 <sup>c</sup>	1.131 <sup>ab</sup>	1.184 <sup>a</sup>	$1.172^{a}$
Generation interval	3.168 <sup>b</sup>	5.243 <sup>a</sup>	3.121 <sup>b</sup>	5.439 <sup>a</sup>	5.052 <sup>a</sup>	5.287 <sup>a</sup>
Annual change of BW	$0.096^{d}$	0.127 <sup>c</sup>	0.135 <sup>bc</sup>	0.146 <sup>b</sup>	0.151 <sup>b</sup>	0.171 <sup>a</sup>
Annual change of YW	$0.942^{d}$	1.355 <sup>bc</sup>	1.274 <sup>c</sup>	1.522 <sup>b</sup>	1.536 <sup>b</sup>	1.736 <sup>a</sup>
Selection accuracy of YB	0.150 <sup>b</sup>	0.195 <sup>b</sup>	$0.208^{ab}$	0.219 <sup>a</sup>	0.219 <sup>a</sup>	0.241 <sup>a</sup>
Selection accuracy of AS	$0.505^{\circ}$	0.535 <sup>c</sup>	$0.570^{\circ}$	0.612 <sup>b</sup>	0.598 <sup>b</sup>	$0.687^{a}$
Selection accuracy of BD	0.210	0.235	0.240	0.279	0.238	0.300
Selection accuracy of CD	0.190	0.220	0.237	0.252	0.245	0.291
Selection intensity of YB	2.421ª	2.420 <sup>a</sup>	2.439 <sup>a</sup>	2.122 <sup>b</sup>	2.436 <sup>a</sup>	$2.220^{a}$
Selection intensity of AS	2.899 <sup>a</sup>	2.532 <sup>b</sup>	$2.900^{a}$	2.532 <sup>b</sup>	2.532 <sup>b</sup>	2.531 <sup>b</sup>
Selection intensity of BD	$2.424^{a}$	$2.426^{a}$	$2.476^{a}$	2.170 <sup>b</sup>	$2.480^{a}$	2.329 <sup>a</sup>
Selection intensity of CD	0.368	0.373	0.390	0.466	0.394	0.506

AI: artificial insemination; ET: embryo transfer and SS: sexed semen.

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

 Table 5
 Annual changes to total merit index (TMI), inbreeding, birth weight (BW), yearling weight (YW), average generation interval, accuracy and intensity selection for young bull (YB), active sire (AS), bull dam (BD), cow dam (CD) in different mating system

T4	Mating system				
Item	Random	Minimum consistory			
Annual change of TMI	1.678	1.656			
Annual change of inbreeding	$1.104^{a}$	0.792 <sup>b</sup>			
Generation interval	4.624	4.644			
Annual change of BW	0.153	0.151			
Annual change of YW	1.535	1.510			
Selection accuracy of YB	$0.227^{a}$	0.216 <sup>b</sup>			
Selection accuracy of AS	0.640	0.630			
Selection accuracy of BD	0.285	0.274			
Selection accuracy of CD	0.264	0.265			
Selection intensity of YB	2.239	2.235			
Selection intensity of AS	2.654	2.654			
Selection intensity of BD	2.311	2.313			
Selection intensity of CD	0.455	0.484			

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

Table 6 Annual changes to total merit index (TMI), inbreeding, and G/F ratio in levels of different reproduction technologies with or without inbreeding control

Tashnalaging		Annual change of TMI		Annual change of inbreeding		G/F	
Technologies	Levels	evels Random Minimum consistory Random Minimum consis				Random	Minimum consistory
	No	1.571	1.554	1.189 <sup>a</sup>	0.930 <sup>b</sup>	1.444	1.786
ET	All BD	1.794	1.781	1.307 <sup>a</sup>	$0.940^{b}$	1.411 <sup>b</sup>	1.931 <sup>a</sup>
	20% BD	1.859	1.823	$1.277^{a}$	0.965 <sup>b</sup>	1.496 <sup>b</sup>	1.923 <sup>a</sup>
	No	1.525	1.478	1.103 <sup>a</sup>	0.797 <sup>b</sup>	1.557 <sup>b</sup>	$2.008^{a}$
	SS-X on all CD	1.622	1.625	$1.352^{a}$	1.015 <sup>b</sup>	1.200 <sup>b</sup>	$1.608^{a}$
SS	SS-Y on all CD	2.132	2.145	1.213 <sup>a</sup>	0.902 <sup>b</sup>	$1.790^{b}$	2.393 <sup>a</sup>
	SS-X on 20% CD	1.758	1.733	1.347 <sup>a</sup>	1.054 <sup>b</sup>	1.323	1.642
	SS-Y on 20% CD	1.778	1.736	1.350 <sup>a</sup>	1.032 <sup>b</sup>	1.330	1.687
	No AI	1.294	1.278	0.586	0.472	2.258	2.725
AI	50%	1.748	1.723	1.309 <sup>a</sup>	0.955 <sup>b</sup>	1.348 <sup>b</sup>	$1.817^{a}$
	90%	1.824	1.804	1.340 <sup>a</sup>	1.030 <sup>b</sup>	1.391 <sup>b</sup>	1.774 <sup>a</sup>

ET: embryo transfer; SS: sexed semen; AI: artificial insemination and G/F: the ratio of annual change of TMI to annual change of inbreeding. The means within the same row with at least one common letter, do not have significant difference (P>0.05).

The use of X-semen increased genetic progress and inbreeding, but decreased generation interval. The beneficial effects of sexed semen (exception of SS-Y on all CD) on genetic progress could be explained by the increasing of selection intensity in CD. Increasing the annual genetic progress by 1.2 and 7.2% were reported using the sexed semen for best CD and all heifers (Sørensen *et al.* 2011). Moreover, Pedersen *et al.* (2012) reported that the use of sexed semen with genomic selection increased the genetic progress by 5.4 and 7.5% for

production and nuclear population, respectively. We found that the annual genetic change of TMI ranged from 0.99 to 35.3% for different patterns of sexed semen that was in line with the results of Sørensen *et al.* (2011) and Pedersen *et al.* (2012). Meanwhile, it has been reported that the inbreeding rate might be increased by 17% using the sexed semen for nuclear and production population (Sørensen *et al.* 2011; Pedersen *et al.* 2012) that was more than our finding in the present study. The higher genetic progress was observed for sexed semen in CD than in normal semen in which the average of inbreeding in the last year for sexed semen and normal semen was 0.0619 and 0.042, respectively (Abdel-Azim and Schnell, 2007).

The use of X-semen produced female progeny resulting in increasing the number of replacement heifers thus avoiding the cost of buying the replacement heifers in the herd and decreasing generation interval. On the other hand, the use of Y-semen may decrease the number of replacement heifers and necessitate the maintaining the cows in the herds for more years as well as increasing generation interval.

Sexing semen could be improved production efficiency through producing of the desired sex from genetically superior animals, and this affected on selection intensity and accuracy of young bull (YB) and cow dam (CD) (Boro et al. 2016; Fleming et al. 2018). The pregnancy rate per artificial insemination using sexed semen was lower than non sexed semen, because the lifespan of sexed semen reduced in the female reproductive tract, due to mitochondria modification and DNA fragmentation in sex sorting process by flow cytometry (Rath et al. 2013; Seidel, 2014). Despite of improvement of sex sorting processes, the use of sexed semen for artificial insemination has still a limitation to heifers due to their lower pregnancy rates, as well as high costs (De Jarnette et al. 2009; Fleming et al. 2018), however, embryo transferring embryos produced by sexed semen as in vivo could be a strategy for increasing pregnancy rates of sexed semen (Pellegrino et al. 2016).

In this study, minimum consistory mating was used for inbreeding control resulting in low inbreeding. It could be expected that genetic progress would increase due to the negative effects of inbreeding on variance in long-time.

Meuwissen (1997) suggested that non-random mating reduced the rate of inbreeding that may not be associated with maximum genetic progress necessarily. Low inbreeding due to the minimum consistory program was reported by other studies (Caballero *et al.* 1996; Sonesson and Meuwissen, 2000; Henryon *et al.* 2009; Nirea *et al.* 2012) that may be through two mechanisms, including the delay in starting of inbreeding by reducing the inbreeding in the next generation; and inducing low rate of inbreeding after starting (Caballero *et al.* 1996; Sonesson and Meuwissen, 2000). Inbreeding is an important issue arising from the application of reproductive technologies because of fewer active breeding animals used and new animals with high or even extreme inbreeding coefficient was produced (Gengler and Druet, 2002).

As found in this experiment, the annual change of TMI for ET was lower than that for random mating, however, the rate of inbreeding in minimum consistory mating was lower than that in random mating that was in agreement with others (Jeon *et al.* 1990; Ghavi Hossein-Zadeh, 2010).

Possibly, the most of superior animals do not allow for mating in controlled inbreeding, thus resulting in lower genetic progress than without control programs. Decreasing genetic diversity in the population is an important concern for any breeding strategy used. The reproductive technologies that evaluated in this study, increased inbreeding of the population and could be affected on genetic diversity. It was important with any technology used in a breeding scheme to prevent of decressing genetic diversity (Fleming *et al.* 2018). The using of minimum consistory mating has potential to monitor inbreeding.

Increase in genetic gain of Canadian dairy cattle by 46% compared to 20 years ago was reported by Van Doormaal et al. (2005). The increased accuracy and intensity of selection associated with the use of AI and genetic evaluation have also contributed to the rate of phenotypic and genetic progress. The heavy use of the best males resulted in a strong increase in inbreeding and a loss of genetic diversity. At a time, the annual increase in inbreeding trend in the Holsteins dairy cows of the USA was estimated to be near to 0.5% (Gengler and Druet, 2002). Artificial insemination was firstly developed in beef cattle and remained the most important tool for genetic improvement. The significant genetic improvement was obtained by genetic selection used by AI in beef through the mating of bulls to cows based on superior genetic values and then randomly mating the sons of them through AI to determine which bulls to use in the breeding program. In dairy cattle, the range of 0.5 to 2.5% increase rate of genetic improvement was reported (Van Vleck, 1981; Kinder et al. 2006). Increasing the production and reproduction efficiency due to using more superior bulls as an artificial insemination strategy for Brazilian beef cattle was reported by Ferraz et al. (2012) that was similar with the result of the present study. Artificial insemination can increase the genetic progress in several ways: first, it reduces the number of required bulls to breed the available cows and increase the selection intensity; second, the widespread use of tested older and progeny testing of younger bulls may affect the accurate estimation of breeding values (Van Vleck, 1981; Gengler and Druet, 2002; Kaya et al. 2018).

	Annual cha	Annual change of TMI		Annual change of inbreeding		G/F	
Technologies	Random	Minimum consistory	Random	Minimum consistory	Random	Minimum consistory	
NO	1.020	1.044	0.399	0.366	2.556	2.857	
AI	1.488	1.442	1.248 <sup>a</sup>	$0.980^{b}$	1.197	1.480	
ET	1.431	1.395	0.679	0.525	2.108	2.659	
SS + AI	1.661	1.646	1.273 <sup>a</sup>	$0.989^{b}$	1.367 <sup>b</sup>	1.730 <sup>a</sup>	
ET + AI	1.716	1.645	1.419 <sup>a</sup>	0.949 <sup>b</sup>	1.211 <sup>b</sup>	1.735 <sup>a</sup>	
ET + SS + AI	1.904	1.892	1.337 <sup>a</sup>	1.007 <sup>b</sup>	1.433 <sup>b</sup>	$1.884^{a}$	

Table 7 Effect of different reproduction technologies on annual changes to total merit index (TMI), inbreeding, and G/F ratio with or without inbreeding control

NO: not using from three technologies; ET: embryo transfer; SS: sexed semen; AI: artificial insemination and G/F: the ratio of annual change of TMI to annual change of inbreeding.

In addition, the accuracy of expected progeny differences (EPDs) of young sires with a large number of offspring (typical of "proven" AI sires) is higher than that of sires with no progeny (i.e., typical of most natural service sires) (Harris and Newman, 1994). Access to EPDs with high accuracy is one of the primary advantages of the use of such semen. The confidence of realizing phenotypic characteristics of offspring produced from proven AI sires with high accuracy service sire (Pruzzo *et al.* 2003; Dahlen *et al.* 2014).

The prediction of sire breeding value with high accuracy could be affected on the profitability and improved the overall genetic gain in beef operations (Harris and Newman, 1994; Betz, 2007; Dehlen *et al.* 2014). The profitability of AI was generally higher than the ET system because of the high cost of the ET system (Kaniyamattam *et al.* 2017). The using of the large number of sires for artificial insemination in the population is a strategy to prevent inbreeding rate without strongly reducing genetic gain (Bouquet *et al.* 2015).

# CONCLUSION

Application of assisted reproductive technologies either simply or in combination with each other can enhance the annual change of TMI without considering inbreeding. The use of ET for 20% BD, Y-semen for 20% CD, and AI for 90% cows could optimize the productivity through beneficial effects on annual change of TMI and reducing the cost of animal managements in the farm. Using the studied technologies in combination with each other under controlled conditions decreased the annual change of inbreeding and increased G/F ratio.

# ACKNOWLEDGEMENT

The authors are grateful from Dr Anderson Christian Sorensen and the authors of the ADAM program for their collaboration and guidance for this preform this research. Also, thanks from research center of special domestic animals, University of Zabol, Zabol, Iran for supporting this research.

# REFERENCES

- Abdel-Azim G. and Schnell S. (2007). Genetic impacts of using female-sorted semen in commercial and nucleus herds. J. Dairy Sci. 90, 1554-1563.
- Andrabi S. and Moran C. (2007). Selection of dairy cow bulls for artificial insemination. *Int. J. Agric. Biol.* **9**, 175-178.
- Barati F., Niasari- Naslaji A., Bolourchi M., Razavi K., Naghzali E. and Sarhaddi F. (2007). Pregnancy rates of frozen embryos recovered during winter and summer in Sistani cows. *Iranian J. Vet. Res.* 8, 151-154.
- Betz G.C.M. (2007). Using the rate of genetic change and the population structure of cattle to better target genetic progress.
  Pp. 103-109 in Proc. 39<sup>th</sup> Beef Improv. Federation Symp., Fort Collins, United states.
- Bodmer M., Janett F., Hässig M., Den Daas N., Reichert P. and Thun R. (2005). Fertility in heifers and cows after low dose insemination with sex-sorted and non-sorted sperm under field conditions. *Theriogenology*. 64, 1647-1655.
- Borchersen S. and Peacock M. (2009). Danish AI field data with sexed semen. *Theriogenology*. **71**, 59-63.
- Boro P., Naha B.C., Madkar A. and Prakash C. (2016). Sexing of semen in bulls: A mini review. *Int. J. Appl. Res.* **2**, 460-462.
- Bouquet A., Sørensen A.C. and Juga, J. (2015). Genomic selection strategies to optimize the use of multiple ovulation and embryo transfer schemes in dairy cattle breeding programs. *Livest. Sci.* **174**, 18-25.
- Bourdon R.M. (1997). Understanding animal breeding. Prentice Hall Englewood Cliffs, New Jersey, United States.
- Buch L.H., Sørensen M.K., Berg P., Pedersen L.D. and Sørensen A.C. (2012). Genomic selection strategies in dairy cattle: Strong positive interaction between use of genotypic information and intensive use of young bulls on genetic gain. J. Anim. Breed. Genet. 129(2), 138-151.
- Caballero A., Santiago E. and Toro M. (1996). Systems of mating to reduce inbreeding in selected populations. *Anim. Sci.* **62**, 431-442.
- Dahlen C., Jamie L. and Lamb G.C. (2014). Impacts of reproductive technologies on beef production in the United States. Pp. 97-114 in Current and Future Reproductive Technologies and World Food Production. G.C. Lamb, N. DiLorenzo, Eds., Springer, New York.

- De Jarnette J.M., Nebel R.L. and Marshall C.E. (2009). Evaluating the success of sex-sorted semen in 374 US dairy herds from on farm records. *Theriogenology*. **71**, 49-58.
- Ferraz J., Eler J. and Rezende F. (2012). Impact of using artificial insemination on the multiplication of high genetic merit beef cattle in Brazil. *Anim. Reprod.* **9**, 133-138.
- Filho M.F., Nichi M., Soares J.G., Vieira L.M., Melo L.F., Ojeda A., Campos Filho E.P., Gameiro A.H., Sartori R. and Baruselli P.S. (2014). Sex-sorted sperm for artificial insemination and embryo transfer programs in cattle. *Anim. Reprod.* **11**, 217-224.
- Fleming A., Abdalla E.A., Maltecca C. and Baes C.F. (2018). Invited review: Reproductive and genomic technologies to optimize breeding strategies for genetic progress in dairy cattle. Arch. Anim. Breed. 61, 43-57.
- Galli C., Duchi R., Crotti G., Turini P., Ponderato N., Colleoni S., Lagutina I. and Lazzari G. (2003). Bovine embryo technologies. *Theriogenology*. 59, 599-616.
- Gengler N. and Druet T. (2002). Impact of biotechnology on animal breeding and genetic progress. Pp. 33-45 in Biotechnology in Animal Husbandry. R. Renaville and A. Burny, Eds., Springer, Dordrecht, The Netherlands.
- Ghavi Hossein-Zadeh N.G. (2010). Evaluation of the genetic trend of milk yield in the multiple ovulation and embryo transfer populations of dairy cows, using stochastic simulation. *Compt. Rend. Biol.* **333**, 710-715.
- Granleese T., Clark S.A., Swan A.A. and Van der Werf J.H. (2015). Increased genetic gains in sheep, beef and dairy breeding programs from using female reproductive technologies combined with optimal contribution selection and genomic breeding values. *Genet. Sel. Evol.* 47, 1-13.
- Hansen P.J. and Siqueira L.G.B. (2017). Postnatal consequences of assisted reproductive technologies in cattle. Pp. 490- 496 in Proc. 31<sup>st</sup> Ann. Meet. Brazilian Emb. Technol. Soc., Cabo de Santo Agostinho, Pernambuco, Brazil.
- Harris D.L. and Newman S. (1994). Breeding for profit: Synergism between genetic improvement and livestock production (a review). J. Anim. Sci. 72, 2178-2200.
- Henryon M., Sørensen A. and Berg P. (2009). Mating animals by minimising the covariance between ancestral contributions generates less inbreeding without compromising genetic gain in breeding schemes with truncation selection. *Anim.* **3**, 1339-1346.
- Jeon G., Mao I., Jensen J. and Ferris T. (1990). Stochastic modeling of multiple ovulation and embryo transfer breeding schemes in small closed dairy cattle populations. *J. Dairy Sci.* 73, 1938-1944.
- Kaniyamattam K., Block J., Hansen P.J. and De Vries A. (2017). Comparison between an exclusive *in vitro*-produced embryo transfer system and artificial insemination for genetic, technical, and financial herd performance. *J. Dairy. Sci.* 100, 5729-5745.
- Kaya A., Gunes E. and Memili E. (2018). Application of reproductive biotechnologies for sustainable production of livestock in Turkey. *Turkish. J. Vet. Anim. Sci.* 42, 143-151.
- Kinder J., Osborne J., Davis M. and Day M. (2006). Impact of reproductive technologies on improved genetics in beef cattle.

Pp. 141-146 in Australian Beef - the Leader Conf., Armidale, Australia.

- Madsen P., Sørensen P., Su G., Damgaard L.H., Thomsen H. and Labouriau R. (2006). DMU-a package for analyzing multivariate mixed models. Pp. 11-27 in Proc. 8<sup>th</sup> World Congr. Genet. Appl. Livest. Prod., Belo Horizonte, Brazil.
- Meuwissen T. (1997). Maximizing the response of selection with a predefined rate of inbreeding. *J. Anim. Sci.* **75**, 934-940.
- Mortazavi M. (2011). From ancient to modern urbanization: intermediary function of an urban society. *Int. J. Hist. Archaeol.* **15**, 126-137.
- Nirea K.G., Sonesson A.K., Woolliams J.A. and Meuwissen T. (2012). Effect of non-random mating on genomic and BLUP selection schemes. *Genet. Sel. Evol.* 44(11), 1-7.
- Pedersen L., Sørensen A., Henryon M., Ansari-Mahyari S. and Berg P. (2009). ADAM: A computer program to simulate selective breeding schemes for animals. *Livest. Sci.* 121, 343-344.
- Pedersen L.D., Kargo M., Berg P., Voergaard J., Buch L.H. and Sørensen A.C. (2012). Genomic selection strategies in dairy cattle breeding programmes: sexed semen cannot replace multiple ovulation and embryo transfer as superior reproductive technology. J. Anim. Breed. Genet. 129, 152-163.
- Pellegrino C.A.G., Morotti F., Untura R.M., Pontes J.H.F., Pellegrino M.F.O., Campolina J.P., Seneda M.M., Barbosa F.A. and Henry M. (2016). Use of sexed sorted semen for fixed-time artificial insemination or fixed-time embryo transfer of *in vitro*-produced embryos in cattle. *Theriogenology.* 86, 888-893.
- Pruzzo L., Cantet R. and Fioretti C. (2003). Risk-adjusted expected return for selection decisions. J. Anim. Sci. 81, 2984-2988.
- Rath D., Barcikowski S., De Graaf S., Garrels W., Grossfeld R., Klein S., Knabe W., Knorr C., Kues W., Meyer H., Michl J., Moench-Tegeder G., Rehbock C., Taylor U. and Washausen S. (2013). Sex selection of sperm in farm animals: status report and developmental prospects. *Reproduction*. **145**, 15-30.
- Seidel G.E. and Garner D.L. (2002). Current status of sexing mammalian spermatozoa. *Reproduction*. **124**, 733-743.
- Seidel G.E. (2014). Update on sexed semen technology in cattle. *Animal.* **8(1)**, 160-164.
- Sonesson A.K. and Meuwissen T.H. (2000). Mating schemes for optimum contribution selection with constrained rates of inbreeding. *Genet. Sel. Evol.* 32, 231-248.
- Sørensen M.K., Voergaard J., Pedersen L.D., Berg P. and Sørensen A.C. (2011). Genetic gain in dairy cattle populations is increased using sexed semen in commercial herds. *J. Anim. Breed. Genet.* **128**, 267-275.
- Team R Core. (2017). R: A Language and Environment for Statistical Computing. Available at: https://www.R-project.org.
- Van Doormaal B., Miglior F., Kistemaker G. and Brand P. (2005). Genetic diversification of the Holstein breed in Canada and internationally. *Interbull Bulletin.* **33**, 93-97.
- Van Vleck L. (1981). Potential genetic impact of artificial insemination, sex selection, embryo transfer, cloning, and

selfing in dairy cattle. Pp. 221-242 in New technologies in Animal Breeding. B.G. Brackett, G.E. Seidel and S.M. Seidel, Eds., Academic Press, London, United Kingdom.

- Villanueva B., Woolliams J. and Simm G. (1994). Strategies for controlling rates of inbreeding in MOET nucleus schemes for beef cattle. *Genet. Sel. Evol.* 84, 177-184.
- Walsh S., Williams E. and Evans A. (2011). A review of the causes of poor fertility in high milk producing dairy cows. *Anim. Reprod. Sci.* 123, 127-138.