

Comparison of Embryo Production Performance and Conception Rate after Embryo Transfer between Mongolian Cattle and Korean Native Cattle

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ABSTRACT

This study is to compare the effect of estrus synchronization and embryo transfer between Korean and Mongolian cattle. Embryos were collected from 9 donors housed in Asan city in South Chungcheong Province, South Korea. Embryos were collected 9 donors from Khushaat sum, Selenge province and Bayanchandmani sum, Tov province in Mongolia. Follicle Stimulating Hormone (FSH), Controlled Internal Drug Release (CIDR) and Prostaglandin (PG) were used for superovulation. Subsequently, Artificial Insemination (AI) was done for donor cow and embryo was collected after 7 and 8 days. Collected embryos were compared between Mongolian and Korean cattle. Finally, good quality and fresh embryos were transferred to 50 and 22 recipients of cows in Korea and Mongolia respectively. The findings show that Korean native cattle each donor cow produced on an average 16.9 embryos and, 10.9 embryos were found transferable. But in case of Mongolia the average production of embryos per donor cow was 8.6 embryos and, 6.2 embryos were found transferable. Embryo collection after 7 and 8 days was not difference in embryo production in Korea. But, in Mongolia embryo production after 8 days was found more efficient than the 7 days. Korean native recipient's cows (74.6%) and Mongolian recipient's cows (71.0%) respectively were found transferable ovarian stage. The result suggested that efficiency of embryo production from the superovulation method treated of Korean cow were higher than the Mongolian cow. The pregnancy rate of Korea native cattle was 72%, which was about 10% higher than that of Mongolia cattle.

(Key Words : Korean native cattle, Mongolian cattle, Pregnancy rate, Embryo Transfer (ET))

INTRODUCTION

Reproduction is one of the main factors in production efficiency of beef cattle. The largest loss of the potential calf crop occurs because cows fail to become pregnant due to anestrus and postpartum infertility (Short et al., 1990). Estrus synchronization has been studied for 40 years to control reproductive efficiency of beef cattle. Its purpose is to manipulate the estrous cycle of a herd to allow for timed artificial insemination (AI) and superovulation with subsequent embryo transfer into recipient cows at a predetermined time (Odde et al, 1990). The general aspects of embryo transfer indicate that the embryo transfer technology provides the opportunity to introduce desirable genetic

material into populations of livestock, while greatly reducing the risk for transmission of infectious diseases (Kim et al., 2013). The productivity of Mongolian cattle is low and so it's essential to make genetic progress and improvement of the cattle through selective breeding and crossbreeding with good feeding and pasture supply. To achieve desired goals, first we must learn from Korea how to improve the productivity of Korean native cattle. There is also computerized system for water and food supply and beef cattle growing conditions depends on hay and concentrates, water and mineral salts. In particular, the Embryo transfer was technical process by selecting breeding bulls and breeding cows with a high genetic traits embryo transfer to produce genetically superior offspring. These techniques are being used to improve livestock

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capacity. Therefore, this experiment was conducted to compare the embryo production performance and conception rate between Korean and Mongolian cattle when embryos are collected from donor cattle after superovulation induction and transferred to estrus synchronized recipients.

MATERIALS AND METHODS

1. Experiment Animals

We selected 9 Korean native cattle donor and 67 recipients cows from Asan city in South Chungcheong Province, South Korea. All donors are registered and genetic superiority guaranteed by Korea Animal Improvement Association. We randomly selected 9 donor and 31 recipients of Mongolian cattle from Khushaat sum, Selenge province and Bayanchandmani sum, Tov province in Mongolia. The recipients of Korean and Mongolian cattle with good corpus luteum and normal uterus, raising under normal nutritional conditions, were selected.

2. Superovulation treatment and artificial insemination (AI) of Donor Cow

Regardless of estrous cycle, Insert Controlled Internal Drug Release (CIDR; Hamilton, New Zealand.) to Donor cow and from 5th day after inserting CIDR, diminishing inject Follicle Stimulating Hormone (FSH; Kyoritsu Seiyaku Coporation,

Japan.) 36IU every 12 hours for 4 days. From the 3rd days after inject FSH, inject Dinoprost Tromethamine(PGF2 α ; LutalyseTM, Upjohn, U.S.A.) 30mg in the morning and 15mg in the noon. From the 4th days after inject FSH, remove the CIDR in the morning. Next day of remove the CIDR, inject Gonadorelin (GnRH; Dong-Bang, Korea.) 1000ug every 12 hours in the noon and did AI 3 times (Table 1).

3. Embryo collection and evaluation

Embryos were collected on the 6th day by a non surgical method after AI. Collected embryos were classified by manual of the international embryo transfer society (Stringfellow and Seidel, 1998). A group of transferable embryos were evaluated as code 1 (excellent or good) or code 2 (fair) and a group of non-transferable embryos were evaluated as code 3 (poor) or code 4 (dead or degenerated).

4. Embryo Transfer and Pregnancy Assessment

CIDR insert to Recipient cow. And same time, inject Estradiol Benzoate 1mg and Progesterone 50mg. From the 7th days after insert CIDR, inject Dinoprost Tromethamine 25mg in the morning. After 24 hours, remove the CIDR. From of 36 hours after remove CIDR, inject Gonadorelin 25ug. From the 7th days after inject Gonadorelin, transfer the Embryo. Rectal palpation was performed for pregnancy diagnosis 110 days after the embryo transfer.

Table 1. Experimental design for superovulation of the donor cow

Day 0	Insert Controlled Internal Drug Release(CIDR) Inject 1mg Estradiol Benzoate Inject 50mg Progesterone Inject
Day 5	AM : 6IU Follicle Stimulating Hormone(FSH) PM : 6IU Follicle Stimulating Hormone(FSH) Inject
Day 6	AM : 5IU Follicle Stimulating Hormone(FSH) PM : 5IU Follicle Stimulating Hormone(FSH) Inject
Day 7	AM : 4IU Follicle Stimulating Hormone(FSH) and 30mg Dinoprost Tromethamine PM : 4IU Follicle Stimulating Hormone(FSH) and 15mg Dinoprost Tromethamine Inject
Day 8	AM : 3IU Follicle Stimulating Hormone(FSH) and Controlled Internal Drug Release(CIDR) remove PM : 3IU Follicle Stimulating Hormone(FSH)
Day 9	PM : Artificial Insemination(AI), Inject Gonadorelin 1000 μ g
Day 10	AM : Artificial Insemination(AI) PM : Artificial Insemination(AI)
Day 16	Embryo collection, transfer.

Table 2. Experimental design for estrus synchronization of recipient cow

Day 0	Insert Controlled Internal Drug Release(CIDR) Inject 1mg Estradiol Benzoate Inject 50mg Progesterone
Day 7	AM : Inject 25mg Dinoprost Tromethamine
Day 8	AM : Controlled Internal Drug Release(CIDR) remove
Day 9	PM : Inject Gonadorelin 250 μ g
Day 16	Embryo Transfer(ET)

5. Statistics

The statistical significance of the data obtained in this study was analyzed by the *T*-test method using SAS package (version 9.1).

RESULTS

1. Embryo production of Korean and Mongolian Donor Cattle

Total collected embryo donor cow is 9 Korean cows and 5 Mongolian cows. Korean native 9 donors produced 152 embryos, each cow produced average 16.9 ± 6.6 number of embryos while on an average 10.9 ± 6.2 embryos were found transferable. Mongolian 5 donors produced 43 embryos, each cow produced average 8.6 ± 3.8 number of embryos and on an average 6.2 ± 1.8 embryos were found as transferable (Table 3). Most of the collected embryos from Mongolian and Korean cow were transferable. However, there was no significant difference between Korean native donor cows and Mongolian donor cows.

2. Embryo production according to flushing days after AI of Korean and Mongolian cattle

In case of Korean native cattle, we collected embryos from the 7 and 8 days after AI. After 7 days insemination, 112 embryos were collected from 6 donors and 65.2% (73/112) embryos were transferable. In group of embryo collection on

8 days after insemination, 40 embryos were collected from 3 donors and 62.5% (25/40) embryos were transferable. In case of Mongolian cattle 26 embryos were collected from 3 donors after 7 days insemination and 57.7% (15/26) embryos were transferable. In group of embryo collection on 8 days after insemination, 17 embryos collected from 2 donors and 94.1% (16/17) embryos were transferable (Table 4). These results indicated that although more embryo were collected on 7 days after insemination than 8 days and portion of compact morula stage embryo was higher on 7 days than 8 days, the Korean native cattle are no significant differences of embryo recovery and quality between collection of embryos on 7 and 8 days after AI. However, Mongolian cattle are significant difference ($p < 0.05$).

3. Comparison of recipient cow effect of ovary status between Korean and Mongolian cattle

Compared the effect of ovary from estrus synchronization with CIDR treated recipient cow and the findings are articulated in Table 5. Also, used same treatment for the estrus synchronization of Korean native cattle and Mongolian cattle. Total 67 numbers of Korean native recipients were treated; we found 50 number of recipients having transferable, Corpora Hemorrhagica 2 (CH2), Corpora Hemorrhagica 3 (CH3) - Corpora Luteum 3 (CL3) ovarian stage. Total 31 numbers of Mongolian recipients were treated; we found 22 number of recipients having transferable ovarian stage. We transferred good quality and fresh

Table 3. Comparison of embryo production performance between Korean and Mongolian donor cows

Cattle	No. of donors Flushed/Treated (%)	No. of embryos (mean \pm sd)			
		Total number of embryos collected	Average number of embryos collected	No. of transferable embryos	Average number of transferable embryos
Korean Native Cattle	9/9(100)	152	16.9 ± 6.6^a	98	10.9 ± 6.2^a
Mongolian Cattle	5/9(55.6)	43	8.6 ± 3.8^a	31	6.2 ± 1.8^a

within the same columns, values with superscripts differ no significantly ($p < 0.05$).

Table 4. Comparison of embryo production and quality by flushing day after artificial insemination (AI) between Korean and Mongolian cattle

Cattle	Flushing day	No. of donors	Total number of embryos collected	No. of embryos degenerated (%)	No. of transferable embryos (%)	Embryos developed to(%)			
						M ¹⁾	CM ²⁾	EB ³⁾	B ⁴⁾
Korean	7	6	112	39(34.8) ^a	73(65.2) ^a	–	53(47.3)	15(13.4)	5(4.5)
Native Cattle	8	3	40	15(37.5) ^a	25(62.5) ^a	–	6(15.0)	3(7.5)	16(40.0)
Mongolian	7	3	26	11(42.3) ^a	15(57.7) ^a	2(7.7)	11(42.3)	2(7.7)	–
Cattle	8	2	17	1(5.9) ^b	16(94.1) ^b	–	3(17.6)	7(41.2)	6(35.3)

¹⁾Morula ²⁾Compactmorula ³⁾EarlyBlastocyst ⁴⁾Blastocyst

within the same columns, values with superscripts differ significantly ($p<0.05$).

Table 5. Comparison of ovarian condition of recipient cows after synchronization treatment between Korean and Mongolian cattle

Cattle	Synchronization method	No. of recipients	Ovarian condition	
			Transferable(%)	Non-transferable(%)
Korean Native Cattle	CIDR+PG+GnRH	67	50(74.6) ^a	17(25.4) ^a
Mongolian Cattle		31	22(71.0) ^a	9(29.0) ^a

within the same columns, values with superscripts differ no significantly ($p<0.05$).

embryos to 50 Korean native recipient cattle and 22 Mongolian recipient cattle. The result suggested that efficiency of ovarian stage from CIDR, PG and GnRH treated estrus recipients of Korea native cattle and Mongolian cattle showed similar tendencies.

4. Comparison of conception rates embryos transfer to Korean and Mongolian cattles

The transferred good quality and fresh embryos to 50 Korean native recipient cattle and 22 Mongolian recipient cattle. Rectal palpation was performed for pregnancy diagnosis 60 days after the embryo transfer and conception rates were compared between two groups. There was no significant difference between Mongolian cattle and Korean native cattle, but the Mongolian cattle, the conception rate was 25% (1/4)

in dairy cows, 77.8% (7/9) in dairy heifers and 66.7% (6/9) in beef cows and for Korean native cattle, conception rate was 72.0% (36/50).

DISCUSSION

This is the first report to describe the comparison of superovulation and embryo transfer between Korean and Mongolian cattle. Our data showed that the synchronization method, total embryos, average embryo each cow, average transferable embryo each cow, total transferable %, degenerate embryos, conception rates did between the Korean and Mongolian cow. Successful embryo transfer is influenced by many donor factors, such as breed, age, parity, and reproductive

Table 6. Comparison of pregnant rates embryos transfer to Korean and Mongolian cattle

Cattle	Type of cattle	No. of cows		
		recipients	transferred(%)	pregnant(%)
Korean	Beef cow	67	50(74.6)	36(72.0)
	Total	67	50(74.6) ^a	36(72.0) ^a
Mongolian Cattle	Dairy cow	11	4(36.4)	1(25.0)
	Dairy heifer	10	9(90.0)	7(77.8)
	Beef cow	10	9(90.0)	6(66.7)
	Total	31	22(71.0) ^a	14(63.6) ^a

within the same columns, values with superscripts differ no significantly ($p<0.05$).

history. In addition, FSH preparation and superovulation protocol, climate, nutrition, and other management factors can influence superovulation outcome (Massey and Oden, 1984; Crister et al, 1980; Darrow et al, 1982; Gordon et al, 1987; Greve et al, 1979). Our result that the findings show that Korean native cattle each donor cow produced on an average 16.9 embryos and, 10.9 embryos were found transferable. However, the case of Mongolia average production of embryos per donor cow was 8.6 embryos and, 6.2 embryos were found transferable. Korean native recipient's cows (74.6%) and Mongolian recipient's cows (71.0%) respectively were found transferable ovarian stage. The efficiency of embryo production from CIDR, PG and GnRH treated of Korean cow was higher than the Mongolian cow but it was not significantly. Requirements for order to increase the pregnancy rate during embryo transfer, first, there are embryo status such as the number of embryos to be transplanted, development date of embryo, development stage, quality of embryo, second, there is transplant technology, third, there are three main factors such as the use management of recipient and condition of uterus and corpus luteum, calving number and the transplant site (Hasler, 2001; Misra et al, 1999; Numabe et al, 2000). However, there is no difference in pregnancy rate by estrus synchronization using PGF2 α (Tervit et al, 1980; Sreenan, 1975; Wright, 1981; Coleman et al, 1987). As reported above, the same results were obtained in this experiment.

In other words, there was no difference in in vivo embryo production, embryo transfer and pregnancy rate of embryo transfer processing between Korean native cattle and Mongolian cattle. It needs more investigation to conclude as the sample size was small. It was confirmed that this estrus synchronization and embryo transplantation technology can be applied as an important technology for cattle reproduction in Mongolian livestock industry.

ACKNOWLEDGEMENTS

This study was carried out by the support of Korea International Cooperation Agency (KOICA) and we would like to thank Asan City for cooperation.

REFERENCES

- Coleman DA, Dailey RA, Leffel RE and Baker RD. 1987. Estrous synchronization and establishment of pregnancy in bovine embryo transfer recipients. *J. Dairy Sci.* 70:858-866.
- Criste JK, Rowe RF, Delcampo MR and Ginther OJ. 1980. Embryo transfer in cattle: Factors affecting superovulatory response, number of transferable embryos and length of post-treatment estrus cycles. *Theriogenology* 13:397-406.
- Darrow MD, Linder GM and Goemann GG. 1982. Superovulation and fertility in lactation and dry dairy cows. *Theriogenology* 17:84(Abstract).
- Hasler JF. 2001. Factors affecting frozen and fresh embryo transfer pregnancy rates in cattle. *Theriogenology* 56:1401-1415.
- Kim NH, Jeon SH, Jung KS, Choi JW, Heo YT, Xu YN. 2013. Optimization of In Vivo Embryo Production and Pregnancy following Embryo Transfer in Hanwoo Cattle. *J. Emb. Trans.* 28:307-314.
- Massey JM and Oden AJ. 1984. No seasonal effect on embryo donor performance in the southwest region of the USA. *Theriogenology* 21:196-217.
- Misra Sk, Mutha Rao M, Jairaj R, Ranga Reddy NS and Pant HC 1999. Factors affecting pregnancy rate following nonsurgical embryo transfer in buffalo(*pubalis bubalis*): A retrospective study. *Theriogenology* 52:1-10.
- Numabe T, Oikawa T, Kikuchi T and Horiuchi T. 2000. Birth weight and birth rate of heavy calves conceived by transfer of in vitro or in vivo produced bovine embryos. *Anim. Reprod. Sci.* 64:13-20.
- Numabe T, Oikawa T, Kikuchi T and Horiuchi T. 2000. Production efficiency of Japanese Black calves by transfer of produced in vitro. *Theriogenology* 54:1409-1420.
- Odde KG. 1990. A review of synchronization of estrus in postpartum cattle. *J Anim Sci.* 68(3):817-830.
- Short RE, Bellows RA, Staigmiller RB, Berardinelli JG, Custer EE. 1990. Physiological mechanisms controlling anestrus and infertility in postpartum beef cattle. *J Anim Sci.* 68(3):799-816.
- Sreenan JM. 1975. Successful non-surgical transfer of fertilized cow eggs. *Vet. Rec.* 96:490-49.
- Stringfellow DA and Seidel SM. 1998. Manual of the International Embryo Transfer Society. 3rd ed International Embryo Transfer Society Inc Illinois. 165-170.

Tervit HR, Coper MW, Good PG and Haszad GM. 1980. Non-surgical embryo transfer in cattle. *Theriogenology* 13:63-71.

Wright JM. 1981. Non-surgical transfer in cattle: embryo-recipient interaction. *Theriogenology* 15:43-46.

Received December 08 2016, Revised March 08, 2017,
Accepted October 16, 2017