P-2 Establishment of Bovine Ovum Bank: I. Full Term Development of Vitrified Hanwoo (Korean Cattle) In Vitro Matured Oocytes by Minimum Volume Cooling (MVC) Method

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Objective: This study was to test whether Hanwoo *in vitro* matured oocytes can be successfully cryopreserved by a new vitrification procedure using MVC method.

Materials and Methods: For the vitrification, oocytes were pretreated in 10% ethylene glycol (EG10) for $5 \sim 10$ min, exposed in EG30 for 30 sec, each oocytes were individually put on the inner wall of 0.25 ml straw, and then straws were directly plunged into LN₂. Thawing was taken by 4-step procedures [1.0 Msucrose (MS), 0.5 MS, 0.25 MS, and 0.125 MS] at 37 $^{\circ}$ C.

Results: In vitro developmental capacity (survival, cleavage (≥2-cell) and blastocyst rates) in vitrified group was no significant difference compared to that in other treatment groups (exposed; 100.0, 74.4, 32.3% and control; 100.0, 78.3, 36.3%); high mean percentage of oocytes (91.2%) was survived, 69.4% of them were cleaved and 27.9% of cleaved embryos were developed to blastocyst. Especially, after transfer of in vitro developed embryos in vitrified group, four of six recipient animals were found to pregnant and three of them were ongoing pregnant by manual palpation at 250 days after transfer. Among them, two healthy female calves (23 kg and 25 kg) were born.

Conclusion: This result demonstrates that MVC method is very appropriate freezing method for the Hanwoo *in vitro* matured oocytes and that ovum bank can be maintained efficiently by MVC cryopreservation method.

P-3 인간 포배기 배아의 유리화 동결·융해 및 임신·분만 2례

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인간 수정란의 배양 기술 발달은 인간 포배기 배아의 획득율을 증가시키고 있다. 지금까지 인간 포배기 배아의 동결은 glycerol을 동결 보호제로한 slow freezing 방법이 주를 이루어왔으나, 유리화 동결법의 장점과 그 발달로 인해 동결 방법에 변화가 있어 왔다. 이에 본원에서 Human IVF-ET 후 잉여배아의 동결에 독성이 낮으며, 높은 생존율이 보고된 ethylene glycol을 기본 동결 보호제로한 EFS 40 (40% ethylene glycol, 18% ficoll, 0.3 M sucrose)과 electron microscope grid를 이용하여 유리화 동결법을 시