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Xenotransplantation of bovine ovarian cortex tissue in SCID mice to study pre-antral follicular development: determination of the optimal graft site.

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Nowadays, increasing attention is given to strategies aiming to preserve or restore female reproduction. However, opportunities to study the dynamics of pre-antral folliculogenesis *in vitro* are not readily available because routine culture procedures for (ruminant) isolated follicles are still lacking. One of the current techniques to overcome this problem is xenografting bovine ovarian cortex tissue into immunodeficient mice [1]. Several transplantation sites have been described such as subcutaneously (SC), underneath the kidney capsule (SK), intramuscularly (IM) and underneath the peritoneum (SP). In the present study, our objective was to determine the optimal host type and graft location in order to maximize the success of graft retrieval, survival and follicular development. In total, 22 mice (12 conventional, 10 SCID -Severe Combined Immuno Deficient-) were used as graft recipients. All mice were anesthetized with an intraperitoneal injection of ketamine and xylazine and subsequently sterilized. Small pieces (maximum dimension of 9 mm^3) of adult bovine ovarian cortex, retrieved from slaughterhouse ovaries, were then grafted at 4 different sites: SC grafts were localized at the left hand side of the neck of the host, IM at the left hamstring (between the semimembranosus and semitendinosus muscle), SK under the left kidney capsule or SP, namely on the left and right hand side, retroperitoneal. Blood vessels were macroscopically localized and stimulated by curettage before the cortex piece was grafted. Fourteen days later, mice were euthanized and the graft was localized and retrieved (if possible) after which the presence of follicles was assessed by visualization following hematoxilin-eosin staining of histological slides. Data show that graft retrieval rates were highest when cortex fragments were grafted underneath the peritoneum (SP site). Although the extend of follicular presence and quality assessment of the detected follicles surely needs additional experiments, our data do not show a difference between SCID or a conventional mouse strain as optimal host type when it comes to graft retrieval rates and the determination of follicular activity. This was unexpected since conventional mice are supposed to reject the graft, but can probably be attributed to the fact that grafts are left in place for a limited time period of only fourteen days. Future research will focus on the vascular component in the success rate of the grafting procedure as well as on viability screening of the follicles by Neutral Red staining.

References

1. P. Bols, J. Aerts, A. Langbeen, I. Goovaerts, J. Leroy; Theriogenology, 73 (6), 740-747 (2010).