ABSTRAK

PENGARUH UKURAN SAMPEL JARINGAN OVARIUM TERHADAP REKRUTMEN FOLIKEL, VASKULARISASI, DAN EKSPRESI PROTEIN PTEN PASCATRANSPLANTASI PADA *CHORIONIC ALLANTOIC MEMBRANE* (CAM)

Latar belakang: Simpan beku dan transplantasi ovarium merupakan salah satu pilihan preservasi fertilitas yang sedang dikembangkan di seluruh dunia. Optimasi prosedur transplantasi jaringan ovarium perlu dilakukan sebelum aplikasi pada sampel manusia dan klinis. Ukuran jaringan transplan merupakan faktor penting yang dapat mempengaruhi terjadinya *burn-out* folikel yang dapat mempengaruhi terjadinya cedera iskemik pada transplan. Oleh karena itu, penelitian ini berfokus dalam evaluasi 3 faktor yang dapat menentukan keberhasilan transplan yaitu rekrutmen folikel, vaskularisasi dan PTEN pada masing-masing ukuran jaringan transplan.

Metode penelitian: Sampel jaringan ovarium kambing *Capra Hircus* dikelompokkan menjadi kelompok pre transplan dan transplan pada media *chorionic allantoic membrane* (CAM) telur ayam, masing-masing dikelompokkan lagi menjadi ukuran 2,5x2,5x1 mm; 5x5x1 mm dan 10x10x1 mm. Parameter luaran yang dinilai adalah jumlah folikel, vaskularisasi serta ekspresi protein PTEN.

Hasil penelitian: Total sampel potongan jaringan ovarium dari keseluruhan adalah 36 sampel (18 untuk pre-transplan dan 18 untuk transplan). Terdapat total 414 folikel, dengan rincian 275 folikel ditemukan pada sampel pre transplan dan 139 folikel ditemukan pada sampel transplan. Total jumlah folikel pada sampel transplan (30,14 $\pm$ 7,71) menunjukkan penurunan yang signifikan jika dibandingkan dengan sampel pre transplan (42,86 $\pm$ 4,29; P <0,05), dimana pada sampel ukuran 10x10x1 mm terdapat penurunan jumlah folikel paling bermakna pasca transplan (P <0,05). Terkait vaskularisasi, terdapat peningkatan bermakna total jumlah vasa pada ukuran 2,5x2,5x1 mm di sampel transplan dibandingkan sampel pre transplan (9,54 $\pm$ 3,2; 15,46 $\pm$ 1,85; P <0,05). Tidak didapatkan peningkatan jumlah vasa yang bermakna pada ukuran 5x5x1 mm dan 10x10x1 mm. Sedangkan pada sampel transplan berukuran 10x10x1 mm, melalui pengamatan ekspresi PTEN terdapat penurunan ekspresi protein PTEN (6,63 $\pm$ 3,36) dibandingkan dengan sampel pre transplan (11,80 $\pm$ 0,42; P <0,05).

Kesimpulan: Pada sampel jaringan ovarium berukuran 2,5x2,5x1 mm didapatkan jumlah vaskularisasi yang paling banyak diikuti dengan penurunan jumlah folikel yang paling sedikit pascatransplantasi di media CAM jika dibandingkan dengan ukuran sampel lainnya.

Kata kunci: preservasi fertilitas, transplantasi ovarium, CAM, *burn-out* folikel, vaskularisasi, PTEN

ABSTRACT

OVARIAN TISSUE SAMPLE SIZE AFFECTS FOLLICULAR RECRUITMENT, VASCULARIZATION, AND PTEN PROTEIN EXPRESSION AFTER TRANSPLANTATION INTO *CHORIONIC ALLANTOIC MEMBRANE* (CAM)

Background: Ovarian tissue cryopreservation and transplantation is an alternative fertility preservation method that has been developed worldwide. This method should be optimized before being applied in human samples and clinical settings. The size of transplant tissue plays a major role in follicle burn-out which then affects post-transplantation ischemia injury. Therefore, this study aimed to evaluate 3 factors that contribute to the transplantation's success, i.e., the size of ovarian tissue, ischemia injury and PTEN expression on each size of the transplant tissue.

Methods: Ovarian tissue samples from goat (*Capra hircus*) were divided into pre transplantation group and transplantation group on chorionic allantoic membrane (CAM) 5-days-old fertilized eggs, each then were cut into 2.5x2.5x1 mm; 5x5x1 mm and 10x10x1 mm sizes. The outcome parameters were assessed for total number of follicles, vascularization, and PTEN protein expressions.

Results: Thirty-six total ovarian tissue strips were divided equally into pre-transplantation and transplantation samples. There was a total of 414 follicles; 275 follicles in the pre-transplantation samples and 139 follicles in the transplantation samples. Compared to the pre-transplantation samples (42.86 $\pm$ 4.29), the total number of follicles in the transplantation samples (30.14 $\pm$ 7.71; $P < 0.05$) was significantly lower, where 10x10x1 mm samples showed the most significant decrease ($P < 0.05$). As for vascularization, the total number of vessels in 2.5x2.5x1 mm transplantation samples was significantly higher compared to the pre-transplantation samples (9.54 $\pm$ 3.2; 15.46 $\pm$ 1.85; $P < 0.05$). There was no significant increase in 5x5x1 mm and 10x10x1 mm samples. Meanwhile, PTEN protein expressions in the 10x10x1 mm transplantation samples (6.63 $\pm$ 3.36) were significantly lower than the pre-transplantation samples (11.80 $\pm$ 0.42; $P < 0.05$).

Conclusions: Ovarian tissue samples, size 2.5x2.5x1 mm, is the most vascularized and shows the least decrease of total follicles number post-transplantation on CAM media compared to the other sample sizes.

Keywords: fertility preservation, ovarian transplantation, follicle burn-out, CAM, vascularization, PTEN.