

# Efek penambahan progesteron in vitro terhadap kemampuan kapasitasi dan ketahanan hidup spermatozoa = Effect of in vitro progesteron supplementation on sperm capacitation and survival

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## Abstrak

**LATAR BELAKANG :** Salah satu hambatan dalam program reproduksi berbantu adalah rendahnya viabilitas dan motilitas sel spermatozoa. Analisis proteomik menunjukkan adanya banyak protein yang diduga berperan dalam regulasi motilitas dan viabilitas spermatozoa antara lain progesteron. Penelitian ini bertujuan untuk menganalisis efek prosurvival progesteron terhadap spermatozoa melalui penekanan apoptosis.

**BAHAN DAN CARA KERJA :** Sampel spermatozoa dicuci dengan sentrifugasi gradient. Sampel spermatozoa di tambahkan progesteron dengan konsentrasi 0 kontrol , 250, 500, 750 dan 1000 ng/mL. Setelah perlakuan, sampel dilakukan pemeriksaan integritas membran dengan metode HOS dan pemeriksaan motilitas dengan Computer Assisted Sperm Analyzer CASA . Deteksi protein fosforilasi tirosin dan Akt serta aktivitas caspase dilakukan dengan metode western blot.

**HASIL :** Efek penambahan progesteron meningkatkan rerata motilitas spermatozoa namun berbeda tidak bermakna  $p>0.05$  . Integritas membran spermatozoa tidak berpengaruh pada pemberian progesteron. Analisis western blot menunjukkan peningkatan fosforilasi protein tirosin antara kelompok kontrol dan setelah diberikan progesteron  $p>0.05$  . Demikian halnya dengan hasil fosforilasi protein Akt juga mengalami peningkatan pada kelompok kontrol dan setelah diberikan progesteron berbagai dosis. Aktivitas caspase-3 mengalami penurunan bila dibandingkan antara kelompok kontrol dan setelah diberikan progesteron  $p$

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**BACKGROUND** One of the obstacles in assisted reproduction programs is the low viability and motility of spermatozoa cells. Proteomic analysis indicates that many proteins are thought to play a role in the regulation of motility and viability of spermatozoa, among others, progesterone. This study aims to analyze the prosurvival effect of progesterone against spermatozoa through apoptosis suppression.

**METHODS** Spermatozoa was washed with gradient centrifugation. Progesterone is added to each sample with a final concentration 0 control , 250, 500, 750 and 1000 ng mL. After the sample treatment was done, membrane integrity checking with hypoosmotic swelling test and motility examination with Computer Assisted Sperm Analyzer CASA . Detection of protein in the western blot will be done that recognizes the phosphorylation of tyrosine residues and Akt and caspase activity.

**RESULT** The effect of addition of progesterone increases sperm motility but not significantly different  $p 0.05$  . The integrity of the spermatozoa membrane is no effect in progesterone. Western blot analysis revealed an increase of tyrosine phosphorylation protein levels between control and after progesterone group  $p 0.05$  . Similarly, the results of Akt protein phosphorylation also increased in control and after progesterone group. Caspase 3 activity decreased when compared between control and after progesterone group  $p$