Ekspresi dan regulasi gen defensin beta 20 pada epididimis mencit = Expression of defensin beta 20 and its regulation in mouse epididymis

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Abstrak

ABSTRAK
Proses pematangan spermatozoa terjadi karena adanya interaksi antara protein dengan membran plasma spermatozoa. Walaupun proses pematangan spermatozoa ini sangat penting, namun gen yang berperan dalam sekresi protein di epididimis ini masih banyak yang belum dikarakterisasi. Gen-gen yang berperan dalam proses pematangan spermatozoa umumnya merupakan protein sekretorik, terekspresi pada segmen spesifik, diregulasi androgen, faktor testikular dan perkembangan postnatal. Pada penelitian sebelumnya diketahui bahwa b-defensin merupakan gen yang banyak terekspresi di organ reproduksi pria dan memiliki peran dalam pertahanan tubuh dan pematangan spermatozoa. Penelitian ini dilakukan untuk mengkarakterisasi ekspresi gen Defb20 untuk mengetahui perannya dalam proses pematangan spermatozoa. Studi in silico dilakukan untuk prediksi struktur gen, signal peptide dan domain fungsional. Quantitative real-time PCR digunakan untuk mengukur ekspresi gen Defb20 pada analisis sebaran jaringan, regulasi androgen dan faktor testikular serta postnatal developmen. Hasil penelitian mendapatkan bahwa sekuen Defb20 mengandung domain penting seperti N-myristoilation dan beberapa situs phosporilasi protein kinase yang mungkin berperan dalam mekanisme interaksi protein dengan membran plasma. Sekuen asam amino Defb20 mengandung signal peptides, mengindikasikan protein yang disekresikan dan terlibat dalam proses pematangan spermatozoa. -defensins 20 (Defb20) terekspresi spesifik di epididimis dengan ekspresi tertinggi terdapat pada kaput epididimis. Defb20 diregulasi oleh androgen yang ditunjukkan dengan adanya penurunan ekspresi Defb20 paska dilakukan gonadektomi dan kondisi ini dapat diperbaiki dengan pemberian hormon pengganti. Defb20 juga diregulasi oleh faktor testikular, yang dibuktikan dari menurunnya ekspresi Defb20 setelah ligasi pada duktus eferen (efferent duct ligation (EDL)). Defb20 mulai terekspresi pada hari ke-21 setelah lahir yang mengindikasikan gen Defb20 terekspresipada suatu periode perkembangan epididimis. Berdasarkan hasil penelitian disimpulkan bahwa Defb20 memiliki karakteristik ekspresi : mengandung signal peptide yang mengarahkan sintesis protein pada jalur sekretorik, spesifik terekspresi di epididimis, diregulasi androgen dan faktor testikular serta mulai terekspresi pada masa pubertas hingga dewasa
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ABSTRACT
Epididymal sperm maturation occurs via interactions between sperm and proteins secreted by the epididymal epithelium. Although this is an important process, the genes that encode secreted proteins remain largely uncharacterized. The genes that play a role in sperm maturation process has character, among others; is a secretory protein, expressed specifically in the epididymis, regulated by androgen, testicular factor, and postnatal development. Previous studies showed that family of-defensins preferentially eaxpressed in male reproductive tracts and play an important role in both innate immunity and sperm fertility. This study aimed to characterize Defb20 to understand its role in sperm maturation. This study

using in silico analyses and quantitative real-time PCR (qRT-PCR). In silico analyses were performed to predict gene structure, signal peptides and functional domains. Defb20 expression in various tissues, after gonadectomy, efferent duct ligation and postnatal development were measured using quantitative real-time RT-PCR. Defb20 sequence contains important domains such as N-myristoilation and kinase binding sites which are putatively involved in the protein activation and protein-plasma membrane interaction. The amino acid sequence of Defb20 contains signal peptides, indicating characteristic of secretory proteins involved in the sperm maturation. β-defensins 20 (Defb20) was expressed exclusively in the epididymis with the highest expression in the caput region. Defb20 was regulated by androgen showing down-regulation after gonadectomy and the expression was recovered after testosterone replacement. However, Defb20 was also regulated by testicular factors in which the expression was down-regulated after efferent duct ligation (EDL). The dependency on the androgen was further confirmed by postnatal expression analysis in which Defb20 begin to express at day21 postnatal indicating specific stage of expression after initial development of the epididymis. In conclusion, Defb20 have a potential to be involved in the epididymal sperm maturation process. Defb20 has characteristic expression; has a signal peptide sequence that directs synthesis in the secretory pathway, specifically expressed in the epididymis, androgen and testicular factors regulated, and expressed in puberty to adulthood