

Pemisahan P_4 (Progesteron) dari Serum Kuda Indonesia CBG4 Bunting 3,5 Bulan dengan *Sephadex G-75*

Separation of P_4 (Progesterone) from Serum of Three and A Half Months Pregnant Indonesian CBG4 Mares using *Sephadex G-75*

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Abstract

Equine Chorionic Gonadotropin (eCG) or Pregnant Mare Serum Gonadotropin (PMSG) is a gonadotropin hormone available in the market which is produced from pregnant Thoroughbred serum as the raw material. eCG is highly required to treat ovarian hypofunction cases (inactive ovaries/postpartum acyclicity) in cows. However, it is quite expensive and difficult to obtain in Indonesia. A previous research has proven that there was no difference in eCG level of three and a half months pregnant CBG4 mares (crossbred between Sandel and Thoroughbred horses up to the forth generation) to those of the Thoroughbred. Apart from containing eCG, pregnant mare serum also contains E_2 , P_4 (progesterone) and prolactin. *Sephadex G-75* is a polydextrose polymer product in the form of gel that is usually utilized to separate protein molecule of molecule mass range between 10,000–75,000 Dalton (10–75 kDa). The purpose of this research was to separate P_4 from serum of six, three and a half months pregnant CBG4 by means of gel filtration chromatography technique using *Sephadex G-75*. Separation was performed by pooling nine eluate fractions (fraction-3 until fraction-11). P_4 concentration of each fraction was measured by means of solid-phase radioimmunoassay (RIA). Research results indicated that the lowest P_4 concentration (ng/ml) was 1.34 ± 1.32 measured from the third eluate fraction which was significantly different to the eleventh eluate fraction i.e. 18.69 ± 2.99 . Besides, there was a tendency to the increasing concentrations of P_4 towards the last fraction of eluate. Through the separation method on the nine eluate fractions, later it will be possible to described the characteristics eluate fraction order for each protein component contained in the three and a half months pregnant CBG4 serum. To support this intention, it is recommend to continue a further research on attempts to separate eCG from three and a half months pregnant CBG4 serum.

Keywords: eCG, *Sephadex G-75*, progesterone, solid-phase RIA

Abstrak

eCG (equine Chorionic Gonadotropin) atau *PMSG* (Pregnant Mare Serum Gonadotropin) merupakan hormon gonadotropin yang beredar di pasaran yang saat ini masih diproduksi menggunakan bahan baku serum *Thoroughbred* bunting. *eCG* sangat dibutuhkan untuk mengobati kasus hipofungsi ovarium (inactive ovaries/post partum acyclicity) pada sapi, tetapi harganya mahal dan di Indonesia saat ini sulit memperolehnya. Sudah dibuktikan melalui penelitian sebelumnya bahwa kadar *eCG* pada kuda CBG4 (*cross bred* antara kuda Sandel dengan *Thoroughbred* hingga generasi keempat) tidak berbeda dengan *Thoroughbred* bunting 3,5 bulan. Serum kuda bunting di samping mengandung *eCG*, terdapat juga didalamnya E_2 , P_4 (progesteron) dan prolaktin. *Sephadex G-75* merupakan produk polimer polidekstran berbentuk gel yang biasa digunakan untuk memisahkan molekul protein dengan kisaran massa molekul antara 10.000–75.000 Dalton (10–75 kDa). Tujuan penelitian ini adalah memisahkan P_4 dari serum enam ekor CBG4 bunting 3,5 bulan dengan teknik kromatografi filtrasi gel menggunakan *sephadex G-75*. Model pemisahan dilakukan melalui penampungan sembilan fraksi eluat (fraksi ketiga sampai kesebelas). Selanjutnya, dengan teknik RIA fase padat akan ditentukan kadar P_4 tiap-tiap fraksi. Hasil penelitian menunjukkan bahwa kadar P_4 (ng/ml) terendah $1,34 \pm 1,32$ terdapat di dalam fraksi eluat ketiga yang berbeda secara bermakna dengan fraksi eluat kesebelas $18,69 \pm 2,99$. Di samping itu, terdapat kecenderungan bahwa kadar P_4 semakin

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meningkat seiring dengan semakin mengarahnya eluat ke fraksi akhir. Melalui model pemisahan atas kesembilan fraksi eluat tersebut nantinya digambarkan karakteristik kandungan urutan fraksi eluat untuk setiap komponen protein di dalam serum *CBG4* bunting 3,5 bulan. Untuk menunjang maksud tersebut, disarankan untuk meneruskan penelitian terhadap upaya pemisahan *eCG* dari serum *CBG4* bunting 3,5 bulan.

Kata kunci: eCG, sephadex G-75, P_4 , RIA fase padat