

Thesis Title	A Microtitre-plate Enzyme Immunoassay for Thyroxine in Dried Cord-blood Spot.
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ABSTRACT

Dried blood spots were collected on various filter papers, i.e. Whatman (W) 2, W 3mm, W 4, W BFC 180, Schleicher&Schuell(SS#)2992, SS#903 and serum paper for glucometre(GSP). The capacity of blood volume contained in 3 mm diameter blood spot on all filter papers except W 2 and W 4 was comparable. Absorption time, blood spot appearance and percentage of eluting recovery have been studied and compared with the approved filter paper widely used, SS#2992, SS#903. It was found that W 3mm gave somewhat similar results with those approved papers and was chosen to be used in comparison with SS#2992 throughout this study. It was shown that cord-blood spot could be kept as long as 15 days at room temperature and more than 1 month at 4°C or -20°C. Two assay techniques for thyroxine assay in dried cord-blood spots, modified conventional RIA and microtitre-plate EIA, have been proposed. Assay evaluation of these two techniques was found to be valid in that the percentages of recovery and precision assay were more than 90 % and less than 15 % respectively. There was a good correlation of thyroxine values between plasma RIA and cord-blood spot RIA on W 3mm and SS#2992, $r = 0.9527$ and $r = 0.9274$

respectively; $p < 0.0001$. Additionally, there was a good correlation of the paired plasma and cord-blood spots using RIA and microtitre-plate EIA with $r = 0.9726$; $p < 0.0001$ for W 3mm and $r = 0.9265$; $p < 0.0001$ for SS#2992. A good correlation of thyroxine levels in cord-blood spots using modified conventional RIA and microtitre-plate EIA between W 3mm and SS#2992 was also observed ($r = 0.8503$, $r = 0.9312$ respectively; $p < 0.0001$). The sensitivity assay of the proposed RIA was $1.43 \mu\text{g/dl}$ for W 3mm and $1.15 \mu\text{g/dl}$ for SS#2992. The sensitivity assay of the microtitre-plate EIA was $0.80 \mu\text{g/dl}$ for W 3mm and $1.25 \mu\text{g/dl}$ for SS#2992. The good parallelism was demonstrated in both proposed assay techniques. Whether these two proposed techniques could be practically applied for thyroxine assay in cord-blood spot as a mass screening test for neonatal hypothyroidism was discussed.