## Critical Evaluation of Plasma and Urinary Aldosterone Measurements by RIA for Clinical and Roution Purpose

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In order to evaluate a simple radioimmunoassay method for urinary and plasma aldosterone determination, critical comparison was made between a method with column chromatography (cyclohexan; benzen; methanol; 60:40:10) and that without purification. Using NEN antibody (aldosterone-18,21-dihemisuccinate) for the former method, plasma aldosterone was measured after CH<sub>2</sub>Cl<sub>2</sub> extraction following column chromatogrphic purification and urinary aldosterone was mesured after chromatographic purification of CH<sub>2</sub>Cl<sub>2</sub> extracted material from HCl hydrolysed urine specimen. For the latter method CIS kit (aldosterone-3-oxime antibody) was used as indicated in the instruction manual. Tritium labeled aldosterone was used as a tracer in both methods.

Satisfacory standard curves were obtained by both methods with  $B_0/T$  ratios of  $41.6\pm1.71\%$  (NEN) and  $43.4\pm2.81\%$  (CIS) respectively, showing rapid decline from 10 to 250 pg. Extraction ratios of plasma aldosterone were  $51.5\pm6.91\%$  by chromatographic method and  $84.06\pm6.12\%$  by CIS method. The latter extraction ratio was improved up to  $98.9\pm18.42\%$  by three times dilution of plasma using distilled water before

extraction. Urinary aldosterone was extracted almost completely by CIS method. However chromatographic method showed lower extraction ratio of  $50.59 \pm 10.41\%$ .

Specificity of antibodies compared using several steroids showed very high specificity in CIS antibody. Cross reactivity of cortisol was 0.01% by NEN antibody and  $10^{-5}\%$  by CIS antibody. Cortisone showed 0.12% cross reactivity by NEN antibody and 0.001% by CIS antibody.

Within-assay variations were 7.59% by NEN kits and 9.29% by CIS kits. Between-assay variations were 26.3% by NEN and 15.7% by CIC kits.

In case of urinary aldosterone determination, both method showed excellent correlation with correlation coefficient of 0.944 (p<0.001). (Y = 0.865X-0.488). Plasma aldosterone determinations were less well correlated (r=0.751, (p<0.05)) (Y=0.677X+0.737). Higher values were occasionally obtained by CIS method. (Y stands for NEN, and X for CIS)

However authors concluded the simple nonchromatographic radioimmunoassay of aldosterone could be used for routine examination.

## A Simplified Method Measuring Plasma Aldosterone by Radioimmunoassay Using 125-IAldosterone

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A radioimmunoassay method was developed to measure plasma aldosterone levels.

Antibody was produced in rabbits by injecting aldosterone-oxime coupled with porcine gamma globulin.

Plasma aldosterone was measured simultaneously by four method described below.

The first was a simple method of extraction using trichloracetic acid, the second a method using methanol, the third a direct method without extraction and the fourth a method using paper chromatography.

<sup>3</sup>H-aldosterone was used in the fourth method and <sup>125</sup>I-aldosterone in the others.