The antiserum (No. 30K unger) cross-reacts minimally with plasma obtained from a totally-pancreatectomized patients and with an acid-ethanol extract of dog intenstine which is supposed to have high concentration of glucagon-like immunoreactivity (GLI). Because of the easy fragility of circulating blood glucagon, blood samples was placed immediately after drawal in chilled tubes containing EDTA and 1000 U trasylol/ml of bolld and were centrifuged at 4°C.

In healthy normal males, plasma glucagon levels were 92.5±11 pg/ml at basal fasting state and exhibited biphasic rise in response to arginine infusion, with two peaks within 10 minutes and around 45 minutes after the start of infusion. When plasma glucagon was measured with an antiserum cross-reacting with GLI, increment over basal level following the infusion of arginine was almost identical to that measured by a pancreatic glucagon-specific antiserum, although absolute value was different. Radioimmunoassay of plasma glucagon using two antisera (specific and non specific) is a useful tool to investigate release mechanism of pancreatic glucagon and GLI.

## Analysis of ACTH Radioimmunoassay (RIA) Kit

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ACTH RIA Kit was manufactured recently by Radiochemical Centre in England and was supplied through Kaken Chemical Co., LTD to requestor. Using ACTH Kit, ACTH extraction process and RIA procedure are necessary for sensitive assay of ACTH. In extraction of ACTH, glass particles which is supplied from Kit adsorbes ACTH from plasma sample, and the ACTH is desorbed from the glass surface with aqueous acetone. Using $^{125}$I-ACTH, extraction percentage of $^{125}$I-ACTH from plasma to aqueous acetone was analysed in different amount of plasma, and decreased extraction percentage was noticed following the increased amount of plasma. No effect of incubation period of $^{125}$I-ACTH with plasma before extraction was noticed. In these extraction procedure, 2 ml of sample showed approximately 70% of extraction however 8 ml of sample showed 40% of extraction.

The standard curve of ACTH extracted from 5 ml of plasma with ACTH standard was higher in bound percentage as compared with one extracted from 2 ml plasma with ACTH standard. In these standard curve the lowest sensitivity of assay was less than 50 pg of ACTH.

In RIA procedure, the effect of incubation time and temperature on RIA was checked in different time and temperature, however indicated time or temperature in Kit was profitable for RIA against other incubation time or higher temperature. Decreased sensitivity or RIA standard curve was noticed in small amount of antiserum (0.05 ml). Clinically, insulin tolerance test was carried out with four normal volanteers and four patients of graves' disease. ACTH and Growth hormone increased in its plasma level following the decrease of blood glycose after insulin injection. The reprodusability of ACTH data is good in assay using Kit.