# Radioimmunoassay of Human Growth Hormone

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Radioimmunoassay of human growth hormon (HGH) was performed using the double antibody immunoprecipitation technique. Anti-HGH antisera were prepared in guinea pigs by repeated injections of HGH with complete Freund's adjuvant. HGH (Wilhelmi) was labeled with $^{131}I$ by the method of Hunter and Greenwood. The mixture of standard HGH or unknown sample, $^{131}I$-HGH and diluted antiserum was incubated for 3 days. The second antiserum (anti-guinea pig $\gamma$-globulin rabbit serum) was then added to the mixture. After being incubated further for 24 hrs, bound$^{131}I$-HGH was separated by centrifugation. Radioactivity of the precipitate was counted by the well-type scintillation counter. Log dose of the standard HGH (0.1～10 m$\mu$g/ml) showed negative linear relationship with per cent radioactivity in the precipitate. The minimal detectable value by this method was 0.1 m$\mu$g/ml. Recovery of added standrad HGH to plasma was approximately 100%, and reproducibility of the assay was satisfactory. Serial dilution of plasma obtained from an acromegalic patient gave a parallel assay curve with that of the standard HGH. Nonspecific inhibition of the antigen-antibody reaction was not observed in our experiments. These results indicate that this method is sensitive and reliable for the determination of plasma HGH.

Plasma HGH levels after overnight fasting and at resting state were $3.38 \pm 2.32$ m$\mu$g/ml (mean$\pm$S.D.) in normal subjects. In acromegalic patients, resting levels of plasma HGH exceeded 20 m$\mu$g/ml in most of the cases. Therefore, determination of the resting level of plasma HGH is useful for the diagnosis of acromegaly. In some non-acromegalic subjects, however, high resting levels of plasma HGH were observed, being probably due to the effect of exercise. These high resting HGH levels in non-acromegalic subjects are easily differentiated from those in acromegalic patients by the glucose suppression test. Oral administration of 50 mg of glucose suppresses plasma HGH in on-acromegalic subjects, whereas it does not suppress plasma HGH significantly in acromegaly. Cryohypophysectomy caused a marked decrease in plasma HGH, with significant improvement of clinical manifestations. Determination of plasma HGH seems to be a valuable mean to evaluate the effect of treatment in acromegaly.

Resting levels of plasma HGH tended to be low in patients with hypopituitarism, although there was an overlapping in plasma HGH levels between normal subjects and patients with hypopituitarism. Insulin induced hypoglycemia or intravenous infusion of argininine caused a significant rise in plasma HGH levels in normal subjects, whereas no significant change was observed in hypopituiary patients. These results suggest that these tests are useful for differentiating pituitary dwarfism from non-pituitary dwarfism, and for differentiating hypopituitarism from anorexia nervosa.

In patients with pheochromocytoma, resting levels of plasma HGH tended to be increased, with an exaggerated response to insulin induced hypoglycemia. Oral administration of glucose failed to decrease plasma HGH levels in most of the patients. Resting levels of plasma HGH seemed to be low in obese subjects, with diminished responses to all stimuli. Patients with hyperthyroidism showed a decreased response to insulin induced hypoglycemia. These abnormalities in HGH secretion in patients with pheochromocytoma and hyperthyroidism were significantly improved by effective treatments.