In the inhibition curve of CCK-PZ, minimum detectable concentration was about 200pg/m*l*, and measurable range was 200–3000pg/m*l* in this assay system.

We are now studing the measurement of CCK-PZ level in human serum.

Studies of Gastrointestinal Hormones by Radioimmunoassay
Report III Radioimmunoassay of Secretin

Akiko MORIYAMA, Hiroyasu MURAYAMA, Jujiro OKAMOTO and Yoji HARADA*
The Department of Nuclear Medicine, Cancer Center and the 4th Department of Internal Medicine, Tokyo Medical College*

We carried out and reported previously a series of fundamental studies on secretin RIA. Supplied with Secretin RIA Kit through the courtesy of Daiichi Radioisotope Laboratories, we conducted both fundamental and clinical investigations with this device. The results are presented in this paper.

In order to determine the optimal incubation time and temperature specimens were incubated at 4°C or room temperature for 1 to 5 days. A nearly identical calibration curve was obtained both from specimens incubated at 4°C for 3 to 5 days and from those incubated at room temperature for 2 to 4 days. Values for CV at different points on calibration curve ranged from 4.2 to 13%, 8% on the average, indicating an excellent reproducibility of calibration curve, with a significant difference in CV observed between levels of 0 and 50 pg/m*l*. The recovery test also yielded a satisfactory result with an average recovery rate of 101%. Mean CV for intra-assay of 11 serum specimens was 2.9% while a corresponding value for interassay was 8.1%. At low concentration levels of below 100 pg/m*l* CV was found at an average of 7.3%. From combined consideration of these figures and the reproducibility of calibration curve it seems possible to determine a secretin concentration level of down to 50 pg/m*l* with reasonable accuracy by this particular technique.

Average secretin levels in the fasting blood as measured by the radioimmunoassay method in a total of 40 subjects of 4 distinct categories, i.e. healthy state, gastric ulcer, doudenal ulcer and duodenal ulcer scar, were 113 pg/m*l*, 102 pg/m*l*, 117 pg/m*l* and 102 pg/m*l*, respectively, hence with no significant differences observed between these disease states. In a further study the blood level of this hormone was investigated for its eventual correlationship with gastrin levels in circulating blood. At the present stage, however, no definite correlation has been shown to exist.

Clinical Evaluation for Secretin Radioimmunoassay kit

Shigeki NAKATA*, Kunisaburo MORIDERA*, Keishiro INOUE*, Yutaka SEINO**, Kozaburo MORI**, Tomohiko TAMINATO**, Yuzuru KATO**, Shigeru MATSUKURA** and Hiroo IMURA***
**Hyogo Prefectural Tsukaguchi Hopsital*
***Third Division, Internal Medicine Kobe University School of Medicine*
****Second Division, Internal Medicine Kyoto University School of Medicine*

Sensitive and specific radioimmunoassay kit for secretin (Daiichi radioisotope laboratory Japan) was developed using antiserum raised in rabbit against synthetic secretin (synthesized by Yanaihara, Shizuoka pharmaceutical college Japan). Human insulin, pork monocompornent glucagon, C-peptide, human gastrin I, GIP, VIP, motilin and substance P did not cross-react with this antiserum. Labelled hormone was obtained by conventional chloramine-T method using [Tyr^1] secretin. Separation of bound label from free label was made by double antibody techni-

que. Minimal detectable quantity of this assay system was 50 pg/m*l* in plasma. Addition of secretin to two normal human plasma gave mean recovery rate 88.1±11.9 (±SD), 107.2±28.5 percent respectively. Four times of intra and inter-assay coeficient of variation of this assay system were 1.37% and 3.63% respectively. Fasting plasma secretin levels ranged mostly from 50 to 217 pg/m*l* in fasting healthy human subjects. Fasting levels of secretin in patients with diabetes mellitus, chronic liver disease, hyperthyroidism and hypothyroidism were not significantly different from those in normal subjects. In normal and diabetic subjects, the plasma secretin levels did not change significantly after ingestin of 50 gm glucose. Furthermore, plasma secretin levels unchanged following ingestion of a meal, although plasma gastrin levels increased significantly. These results suggest this secretin RIA kit is useful to evaluate the function of S cell.

Evaluation of AMICON MC-40 Ultrafiltration System for the Measurement of CEA by Z-gel Method

Daijiro TSUJINO, Yasuhito SASAKI, Reiko CHIDA, Hiromi SASAOKA, Satoru TAKAHASHI and Kazuhiko SOMEYA

The Third Department of Internal Medicine, St. Marianna University School of Medicine

Present method for the measurement of CEA by Roche kit consists of 3 steps, that is, deproteinization using perchloric acid, dialysis of samples and subsequent radioimmunoassay (RIA). The 2nd step is necessary for the removal of excess salts. It needs skilled and tedious technique and is time consuming as well, which makes the method unsuitable to be performed in clinical laboratories on rutine basis. The purpose of present study is to evaluate a simpler procedure using ultrafiltration in order to replace the dialysis.

AMICON MC-40 system was used for ultrafiltration. The apparatus consists of 4 units each of which contains 10 cells mounted with CM filters. Five m*l* of deproteinized samples were added by hand to the cells, to which 3M tris buffer, ammonium acetate buffer are successively added automatically. The ultrafiltration is performed under agitation, which takes 2 hours. Desalted samples are pipetted to test tubes, which are subjected to subsequent RIA.

Plasma CEA measured using AMICON MC-40 ultrafiltration system was evaluated in comparison with our previously reported data obtained by Z-gel method using dialysis.

Basic evluation of the ultrafiltration method included within assay error of 8–33% of coefficient of variance (C.V.) in the range of 0.6–10 ng/m*l*, between assay error of 8% (C.V.) in 8 measurements and recovery rate of 98–113% ($\overline{m}$ 107%). Plasma CEA levels in 19 patients measured by the two methods correlated well with regression equation of Y(Dialysis)=0.98 × (Ultrafiltration)−0.25 (r=0.93). Those reults agree well with the data obtained previously using dialysis.

Ultrafiltration method is suitable for routine clinical measurement of CEA, as it allows to complete the measurement during a working day, the major advantage over the former method which needs overnight dialysis changing water several times.