D_4. Measurement E (In Vitro Assay, C-Peptide)

Fundamental Studies on the Development of C-Peptide Radioimmunoassay Kit

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We carried out the fundamental studies of C-peptide Radioimmunoassay that recently reported with an aim to materialize "C-peptide kit" which can be usable as one of the pancrease function tests.

The antiserum was prepared by immunizing synthetic human proinsulin connecting peptide to rabbit, and ^{125}I-C-peptide was obtained by labelling ^{125}I to tyrosilated connecting peptide by Hunter & Greenwood's modified method.

As assay system, the double antibody method was carried out mainly, and on B, F separation the polyethylene glycol method was studied as well.

The usable antiserum at final dilution of 1/10,000 was obtained, and it hardly showed any cross reactions against other peptide hormones.

On B, F separation, the double antibody method was more suitable than the polyethylene glycol method. First incubation of 4°C, 48 hrs. was best in the double antibody method.

In our method, the recovery rate was good; 98.4−107.3%, the standard coincided well with the endogenous C-peptide, the serum of 100−300 μl seemed usable for assay with this method.

The variations of intraassay and interassay in C-peptide values of sera were small. Any remarkable variations were not detected in the control serum values we measured for 7 weeks using the kit (Storage at 4°C).

In glucose tolerance test, the curve of C-peptide and that of insulin were parallel in the case of normal blood.

From the above results, this method seems usable enough for C-peptide measurement in blood.

Analysis of C-Peptide Radioimmunoassay (RIA) Kit

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Human C-peptide RIA kit was manufactured recently by Daiichi RI Laboratories Ltd.

The antiserum did not cross-react with human insulin. The mean recovery was 111.7%. Intraassay variability was less than 4.2% (C.V.).

0.5 ng/ml of C-peptide in the smallest amount