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EVEDENCE THAT THYROID-STIMULATING IMMUNOGLOBULIN (TSI) OR TSH STIMULATES THYROGLOBULIN (Tg) RELEASE FROM HUMAN THYROID MONOLAYERED CELLS.Y.Fukue, H.Uchimura, H.Ikeda, T.Mitsuhashi, S.Okano*, Y.Kanaji* and F.Takaku ; The Third Department of Internal Medicine, Faculty of Medicine, University of Tokyo, and Kanaji Hospital* : Tokyo.

The purpose of this study was to show effects of TSI or TSH on Tg release from human thyroid cells during a long term culture. Thyroid tissues were obtained at surgery from Graves' patients. The tissue was minced and treated with enzymatic dispersion. Resulted cells ($1.5 \times 10^6$cells/dish)were cultured for a long time (up to 54 days) with or without TSI (0, 0.2, 0.4, 0.8mg/ml) or TSH (0.01, 0.1, 1, 10, 100mU/ml) using Ham's F-12 supplemented with 10% calf serum. Medium changes were performed every 3 days and Tg levels in the media were assayed by RIA.

Results.(1)Tg release was observed during the time of experimental periods in each scheduled conditions in culture.(2)Peaks of Tg release were observed at 15th days of culture in the presence of TSH and 18th days in culture with TSI, respectively, and after that the Tg levels were reduced gradually. (3)Tg release from the cells stimulated by both stimulators were in a dose dependent manner.

Conclusion. These data confirmed that TSI or TSH stimulates Tg release in human thyroid cells in vitro and suggest that circulating TSI in Graves' patients may involve in Tg secretion from the thyroid gland.

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AN IMPROVED ASSAY FOR THYROID STIMULATING ANTIBODIES USING CRUDE IMMUNOGLOBULIN FRACTIONS PRECIPITATED WITH POLYETHYLENE GLYCOL. K.Kasagi,J.Konishi,K.Arai,T.Misaki, T.Nakashima,K.Endo and K.Torizuka. Dept. of Nuclear Medicine, Kyoto University School of Medicine, Kyoto.

A simple, sensitive and practical assay for thyroid stimulating antibodies(TSAb) has been developed in which crude immunoglobulin fractions sedimented from serum with polyethylene glycol(PEG) were directly dissolved in the culture medium for cryopreserved porcine thyroid cells. The assay was sensitive enough to show 1.4-2.0 folds increase in cAMP released into Hank's medium without NaCl at 1μU/ml bovine TSH. TSAb activities in the fractions sedimented with PEG correlated well($r=0.955, n=20, p<0.001$) with those in the IgGs purified with Protein A, the former being more stimulatory than the latter. TSAb were detected in 41 (97.6%) out of 42 patients with untreated Graves' disease, 22(78.6%) out of 28 patients with euthyroid Graves' disease and 7 (25.9%) out of 27 patients with Hashimoto's thyroiditis. In patients with Hashimoto's thyroiditis, all hypothyroid patients whose serum TSH concentrations were higher than 94μU/ml gave positive results. Absorption of TSH with anti-TSH antibodies completely abolished the stimulatory effect on cAMP production. In Graves' disease treated with antithyroid drugs TSAb activities correlated slightly better with relapse or remission after discontinuation of the therapy than the results of T3 suppression test.

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CLINICAL SIGNIFICANCE OF ELEVATED SERUM NONSPECIFIC BINDING IN THE TBII ASSAY. T. AKAMIZU, H.ISHII, T. YOKOTA, H.NAKAMURA, T.MORI, and H.IMURA. Kyoto University Faculty of Medicine, Kyoto.

The existence of abnormal thyrotropin-binding immunoglobulins in Graves' patients has been reported, and the significance of nonspecific binding (NSB) measurment in the TSH binding inhibitor immunoglobulin (TBII) assay has been indicated. (Akamizu et al.: JCEM, vol.59, 240-245,1984). In 643 TBII assays NSBs were routinely measured using 1% Lubrol instead of TSH receptor. High NSB, which exceeded the average by 10% and mean +1s.d, was found in 76 cases and much more frequently seen in active Graves' than in inactive Graves' or other thyroid disorders. The correction of raw TBII activity by NSB resulted in elevation of TBII activity, and negative TBII activity turned out positive in about a half of the overall cases and 78% of active Graves' patients. Furthermore serial alterations of NSB and TBII through clinical course of 27 Graves' patients showed two patterns; parallel alterations in 16 cases and reciprocal in 8 cases. In one Graves' patient, a large dose predonisolone and plasmapheresis greatly changed the serum protein moieties, and NSB was found to be altered very closely with TBII. In conclusion, NSB in TBII assay was considered indispensable to improve the assay specificity.

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PAIRED DETERMINATION OF THYROID-STIMULATING AND TSH-BINDING INHIBITORY ACTIVITIES IN PATIENTS WITH GRAVES' DISEASE DURING ANTITHYROID DRUG TREATMENT. K.Kasagi,Y.Iida, J.Konishi,T.Misaki,T.Nakashima,K.Endo, K.Torizuka and *K.Kuma. Dept. of Nuclear Medicine, Kyoto University School of Medicine, Kyoto and *Kuma Hospital, Kobe.

Sequential changes in thyroid stimulating antibodies(TSAb) and TSH-binding inhibitor immunoglobulins(TBII) during antithyroid drug treatment were studied in 17 patients with Graves' disease. Before the treatment, TSAb and TBII were detected in 17(100.0%) and 13(76.5%) patients, respectively. There was a significant correlation between the initial TSAb and TBII activities($r=0.600, n=17, p<0.02$). At the end of the observation period ranging from 7 months to 6 years, 4 patients remained hyperthyroid, 3 were in remission and one developed hypothyroidism presumably due to blocking-type TBII. Average activities of both TSAb and TBII gradually decreased during the treatment. In 9 out of the 17 patients, there was a good correlation between the changes of TSAb and TBII. In 4 patients TSAb activity decreased with time whilst TBII activity was maintained or fluctuated. Analysis of the individual data in relation with the clinical course gave the following conclusion:The patient's clinical outcome could not be predicted from the initial values for either TSAb or TBII, but the maintenance of these activities during the treatment seemed to be associated with resistance to the therapy.

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