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TSH-Receptor antibodies, HLA B8 and thyroid autoantibodies in patients with Graves' disease in therapeutically induced euthyroidism

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Summary. The prevalence of TSH-receptor antibodies and of thyroid autoantibodies was studied in 48 HLA-typed patients with Graves' disease, who were in an euthyroid state after antithyroid therapy with methimazole. TSH-receptor antibodies, which were found in 35% of the patients, did not correlate with the positivity of HLA B8. By contrast the persistence of thyroid microsomal antibodies was significantly associated with HLA B8.

Several studies have shown that Graves' disease is an HLA-associated autoimmune disorder[1-8] characterized by an interaction between the endocrine and the immune systems, which results in the formation of autoantibodies to TSH receptor. TSH-receptor antibodies, which are detectable in about two thirds (ranging from 50 to 70%) of untreated patients with Graves' disease[9-15], are assumed to mimic the stimulatory effect of TSH by binding to TSH-receptor and activating adenylate cyclase. Recently an association between HLA B8 positive thyrotoxic patients and prevalence[3,16] or persistence[8] of thyroid microsomal antibody formation was found. Furthermore, findings previously observed in patients with Graves' disease indirectly suggest a possible influence of HLA B8 on the persistence of thyroid stimulating antibodies[8]. Therefore we studied the prevalence of TSH-receptor antibodies and of thyroid autoantibodies in 48 HLA-typed patients with Graves' disease, who were in an euthyroid state after antithyroid therapy with methimazole.

Material and methods. 48 patients suffering from Graves' disease were studied; as a result of previous methimazole treatment all patients were in an euthyroid state at the time of investigation. For determination of TSH-receptor antibodies a modification[11] of a radioligand receptor assay described by Smith and Hall[17] was used. The assay was considered to be positive if the binding of 125 J-TSH to thyroid membrane was below the mean value minus the double SD in the presence of control IgG. Thyroid antibodies were measured by standard procedures with the tanned red-cell haemagglutination method for thyroglobulin and indirect immunofluorescence technique for microsomal antibodies. HLA A, B and C locus antigens were determined by the NIH microlymphocytotoxicity technique.

Results and discussion. HLA B8 was found in 15 of the 48 patients (31%) vs. in 81 of the 450 (18%) controls ($p < 0.03$), which confirms the previously reported association between Graves' disease and HLA antigens[1-8]. Detectable TSH-receptor antibodies were observed in 35% of the patients in therapeutically-induced euthyroidism, which is in accordance with data of Graves' patients treated by antithyroid drugs studied by other authors[12-14]. Interestingly, the incidence of TSH-receptor antibodies did not differ in B8 positive and B8 negative patients (table). Likewise, the prevalence of thyroglobulin antibodies was almost similar in the respective groups (table). By contrast the persistence of thyroid microsomal antibodies was significantly correlated with HLA B8, which is analogous to the islet-cell antibody persistence in insulin-dependent diabetes[18,19].

The present study demonstrates a relatively high incidence of TSH-receptor antibodies in patients with Graves' disease in euthyroidism after treatment with antithyroid drugs. Although the course of the disease seems to be genetically determined[6-8], persistence of TSH-receptor antibody production was not correlated with the HLA system. Interestingly, TSH receptor antibodies are also found in patients with Hashimoto's thyroiditis[15,20,21], ophthalmic Graves' dis-

TSH-Receptor antibodies, HLA B8 and thyroid autoantibodies in patients with Graves' disease in therapeutically induced euthyroidism

Thyroid autoantibodies	All thyrotoxic patients* (N = 48)	B8 positive thyrotoxic* patients (N = 15)	B8 negative thyrotoxic* patients (N = 33)
TSH-Receptor antibodies	N = 17 (35%)	N = 6 (40%)	N = 11 (33%)
Thyroid microsomal antibodies	N = 19 (40%)	N = 10 (67%)	N = 9 (27%)**
Thyroglobulin antibodies (> 1:250)	N = 10 (21%)	N = 4 (27%)	N = 6 (18%)

* All patients were euthyroid after treatment with methimazole. ** $p = 0.01$; χ^2-test.

ease[22] and nontoxic multinodular goitre[23]. Thus, TSH-receptor antibodies do not necessarily lead to thyroid cell stimulation in all cases. This finding seems paradoxical, but might be explained by complete target cell destruction in Hashimoto's thyroiditis and variations in affinity and specificity of thyroid receptor antibodies in ophthalmic Graves' disease[22] and nontoxic multinodular goitre[23]. Further studies, evaluating a possible influence of MHC genes (including Ia type alloantigens) on persisting TSH-receptor antibody production in those Graves' disease patients who tend to relapse frequently, seem to be essential.

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An immunohistochemical and ultrastructural comparison of the effects of 2-bromo-α-ergocryptine on intrasellar and transplanted rat pituitaries

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Summary. Following 2 weeks of administration of 2-bromo-α-ergocryptine, a marked decrease was observed in prolactin immunoreactivity of the grafted pituitaries, whereas no reduction was noted in the intrasellar pituitaries. No evidence of crinophagy was revealed by electron microscopy in prolactin cells of 2-bromo-α-ergocryptine-treated rats.

The drug, 2-bromo-α-ergocryptine (bromocryptine), a dopaminergic agonist is known to cause a reversible suppression of pituitary prolactin secretion[2-4]. Controversy still exists as to whether the action of the drug is mediated through the hypothalamus[5] or directly on the prolactin cells of the pituitary[6,7]. This study investigates the effects of bromocryptine on the immunohistochemical and ultrastructural features of prolactin cells in the intrasellar and ectopic pituitary transplanted beneath the renal capsule.

Materials and methods. 35 male R-Amsterdam rats, maintained on Purina Chow and tap water, ad libitum, were divided into 4 groups. Groups 1 and 2 contained nontransplanted animals. Groups 3 and 4 were comprised of hypophysectomized animals with portions of the anterior pituitary grafted under the renal capsule. Groups 1 and 3, consisting of 9 and 8 animals, respectively, served as controls and remained untreated. Groups 2 and 4, both with 9 animals each, were given bromocryptine (Parlodel®, Sandoz) in a dose of 0.5 mg/100 g b.wt, through a gastric tube, once a day for 2 weeks.

6 animals from groups 1, 2 and 4 as well as 5 animals from group 3 were decapitated and the intrasellar and grafted pituitaries were removed, fixed in formalin and embedded in paraffin for light microscopy. For the immunohistochemical demonstration of prolactin, the antiperoxidase technique was used as described previously[8]. The prolactin antibody was kindly donated by Dr A.F. Parlow, through the NIAMDD Rat Pituitary Hormone Distribution Program.

For electron microscopy, pieces of adenohypophyses were removed from 3 animals in each group, fixed in glutaraldehyde, postfixed in OsO_4 and embedded in Durcapan.

Results. Only the cells of the grafted pituitaries showed a marked decrease in immunoreactive prolactin content following bromocryptine administration (figure 1). No recognizable difference in ultrastructural morphology was noted in the intrasellar pituitaries of untreated and bromocryptine-treated rats. However, in the grafts of the hypophysectomized animals, treatment has resulted in involution of prolactin cells (figures 2 and 3).

Discussion. Previous investigations of the mechanism of action of bromocryptine in the rat have shown a rise of pituitary prolaction content with a parallel reduction of serum prolactin levels[9-11] and it was claimed that the discharge of prolactin and not its synthesis was inhibited by bromocryptine. Another study demonstrated that estrogen-stimulated pituitary prolactin secretion can be inhibited by bromocryptine[9]. More recent investigations found no sig-