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Possible participation of colloid antigen 2 and abhormone (IgG with hormone activity) for the etiology of Graves' disease

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ABSTRACT

The theory that antibody (Ab) directed against the TSH receptor (TSHR) (TSHRAb) is the causal factor of Graves' disease seems unlikely. Corticosteroids have not had a curative effect on the hyperthyroidism of Graves' disease despite their effectiveness for other autoimmune diseases. Two kinds of TSHRAb, thyroid-stimulating Ab (TSAb) and thyroid-blocking Ab (TBAb), are known as causal factors of hyperthyroidism and hypothyroidism, respectively. Previously, we reported that TSAb may be thyroid stimulating animal IgG-like hormone and TBAb may be the precursor of TSAb. In this paper we suggested that TBAb (precursor) converts to TSAb (active form) via the action of the protease, colloid antigen 2 (CA2). We speculate that the conversion of TBAb to TSAb is controlled by two factors: the protease and an anti-protease Ab. When anti-protease Ab levels are high, the patient exhibits hypothyroidism due to the increase in TBAb levels caused by neutralization of the protease. When anti-protease Ab levels are negative, the patient's hypothyroidism disappeared by the negative serum TBAb due to increased protease.

An immunoglobulin G (IgG) with enzyme activity is known as an abzyme, which may be an undeveloped form. IgG with hormone activity may be likewise called an abhormone, which could also be an undeveloped form. The tumor marker CEA is a known member of the IgG supergene family. Many ancestral versions of proteins may have been produced as an IgG form.

Possible participation of colloid antigen 2 and abhormone for the etiology of Graves' disease is suggested.

Introduction

Graves' disease is considered to be an autoimmune disease due to the associated high levels of anti-TSH receptor (TSHR) antibody (Ab), particularly thyroid-stimulating antibody (TSAb). In our previous paper, we described flaws in this autoimmune theory as an etiology of Graves' disease [1]. The main point is that corticosteroid treatment does not have curative effect on the hyperthyroidism and presence of TSAb in Graves' disease despite the positive effect of corticosteroid treatment for most autoimmune diseases. All known autoantibodies induce some tissue damage; in contrast, the TSAb that is characteristic of Graves' disease has a thyroid-stimulating action. Thus, TSAb exhibits an action dissimilar from all previously described autoantibodies.

In a previous manuscript, my group suggested that TSAb may be thyroid-stimulating animal IgG-like hormone and that thyroid-blocking antibody (TBAb) may be the precursor of TSAb [1]. In the present paper, I describe the additional supporting evidence for our previously proposed hypothesis on the etiology of Grave's disease.

Supporting evidence that TBAb is the precursor of TSAb

We proposed previously that TBAb may be the precursor of TSAb, based on the conversion phenomenon of TBAb to TSAb [1]. This conversion phenomenon was theorized because high cAMP production was observed when TBAb-IgG conjugated porcine thyroid cell was incubated with rabbit anti-human IgG, whereas no cAMP production was induced by its co-incubation with normal rabbit serum [2]. The mechanism of the conversion from TBAb to TSAb may be a protease (an enzyme converting TBAb to TSAb) on the cell because anti-protease Ab (human IgG) is neutralized by rabbit anti-human IgG (Fig. 1).

Furthermore, when TBAb-monoclonal Ab (Kl-70)-conjugated porcine thyroid cells were incubated with anti-human IgG, high cAMP production was also observed [3].

In a clinical studies, TBAb production is higher in hyperthyroid patients receiving isotope therapy than in those receiving anti-thyroid drug therapy [4–6]. The mechanism may be protease inactivation by the radiation damage to the protease in the thyroid gland.

A hypothyroid newborn baby positive for TBAb was reported to be

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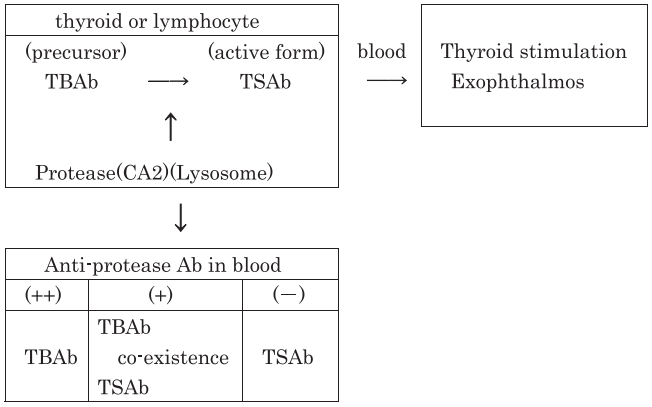

Fig. 1. Protease participating in the conversion of the precursor TBAb to active type TSAb.

converted to positive for TSAb in 2–4 months [7]. In contrast, there are no reported cases of TSAb-positive newborn babies converting to being TBAb-positive. This indicates that conversion from TBAb to TSAb occurs, but conversion from TSAb to TBAb does not occur, possibly due to a lack of new IgG synthesis in the few months directly after birth. Thus, TBAb (precursor) likely converts to TSAb (active form) via a protease.

If anti-protease Ab is produced in high levels in the blood, the protease activity is neutralized and TBAb does not convert to TSAb. In this case, hypothyroidism may occur due to the corresponding accumulation of TBAb. When anti-protease Ab is not present, hyperthyroidism may occur via the conversion of TBAb to TSAb. When anti-protease Ab levels are intermediate, hypothyroidism or hyperthyroidism may occur depending on the relative levels of TBAb or TSAb because of the co-existence of both TBAb and TSAb in the blood.

I propose that the protease that converts TBAb to TSAb may be colloid antigen 2 (CA2) [8–10]. The purification of CA2 was reported previously [11]. The characteristics of the protease and the anti-protease Ab in this system are shown in Table 1. The protease origin is the thyroid lysosome, it is probably pH 3.5 protease, and its MW is the sub-19S fraction. However, it is not conclusive that CA2 and pH 3.5 protease are the same.

The anti-protease Ab is a non-precipitating Ab, and it can be successfully detected via the fluorescent antibody method. This anti-protease Ab is positively detected mainly in Hashimoto disease and Graves' disease.

It is possible to explain that corticosteroids have no curative effect for TSAb-positive hyperthyroidism while having a prominent curative effect for TBAb-positive hypothyroidism because the inhibition of anti-protease Ab production by the corticosteroid treatment would lead to the disappearance of TBAb due to the resulting increased levels of protease [12,13] (Table 2).

From the evidence described above, this condition results from three kinds of proteins in the blood: TBAb (precursor of TSAb), the protease (CA2; enzyme that converts TBAb to TSAb), and the anti-protease Ab.

Table 1
Protease (colloid antigen 2; CA2) and Anti-Protease Ab.

Enzyme and Ab	Character	Classification
Protease	Origin	Thyroid lysosome
	Detection	Protease (pH3.5)
	MW	The sub-19S fraction*
Protease Ab	Specificity	No-precipitating antibody
	Detection	The fluorescent antibody method
	Positive disease	Hashimoto disease and Graves' disease

* No conclusive proof CA2 and protease (pH 3.5) are the same.

Table 2
Disappeared TBAb following corticosteroid administration in TBAb-positive hypothyroid patients.

Subject	Treatment	Possible serum level		
		Protease	Anti-protease Ab	TBAb
TBAb(+) hypothyroid patient	(−)	(−)	(+)	(+)
	Corticosteroid treatment	(+)	(−)	(−)

Abzymes, abhormones, and CEA

An IgG-type enzyme with enzyme activity is called an abzyme. VIP and DNAase are both known examples of abzymes [14,15] The amino acid structure of an abzyme is classified as an immature or undeveloped type. Primitive enzymes may have been abzymes, which have an IgG structure despite lacking antibody function, in contrast to most fully developed enzymes, which are not IgG type (non-IgG type).

TBAb may be the precursor of the primitive type thyroid stimulator with an IgG structure, and TSAb may be an active form of TBAb.

We reported the occurrence of an immunological cross-reaction between CEA (carcinoembryonic antigen; MW = 180,000) and α1-acid glycoprotein (MW = 60,000) [16]. CEA is known as a member of the IgG supergene family [17–19] and the serum CEA concentration in normal subjects is extremely low (5 ng/ml). Based on this example, I speculate that many ancient or ancestral proteins may have been produced as an IgG type and that serum concentrations of these ancestral versions are extremely low (Table 3).

The participation of non- antibody IgG in the etiology of Graves' disease

Back in 1956 when LATS was originally detected and found to have an IgG structure, IgG structures were thought to be possessed by only antibodies. Thus, the existence of an antigen corresponding to this IgG was suggested, and the theory of TRAb (TSH receptor Ab) has gone unchallenged until now.

When an IgG capable of hydrolyzing VIP was found, the existence of enzymes with an IgG-type structure, called abzymes, was proposed. Unlike antibodies, abzymes were not thought to have corresponding antigens. When TSAb (LATS) was first discovered, the possibility that it was a hormone with an IgG structure was not considered. TSAb was assumed to be an antibody despite the lack of curative effect by corticosteroids on TSAb-positive Graves' disease. I propose that the correct nomenclature for TSAb should be abhormone (i.e., a hormone with an IgG structure), as described previously [1]. The etiology of Graves' disease may begin the protein synthesis of TBAb, which has an IgG structure, as its precursor form. TBAb then converts to TSAb (active form) via the action of a protease (CA2) in the thyroid lysozome.

Further study is important to resolve the etiology of Graves' disease.

Discussion

Many findings on TSAb and TBAb remain unexplained by the

Table 3
Possible functions of IgG type proteins.

Classification	Structure	Protein
Abzyme	IgG type	VIP DNA ase
Abhormone	IgG type	TSAb TBAb
Tumor marker	IgG supergene family	CEA

current theory concerning the etiology of Graves' disease.

1) When porcine thyroid cells were incubated with 5% PEG (polyethyleneglycol) and TSAb, cAMP production increased about 2–3 fold (many cases) and 5–10 fold (few cases), in contrast with the lack of cAMP production induced by incubation with 5% PEG and bTSH [20].
2) The lack of increase in TBII (TSH-binding inhibitory immunoglobulin) activity by TSAb in a prolonged (> 1h) incubation suggests that the reaction of TSAb with TSHR is not an antigen–Ab reaction but rather a hormone–receptor reaction [1]. In contrast, receptor binding increases following a prolonged incubation in an acetylcholine RAb assay kit, because this is an antigen-antibody reaction [1].
3) In the acetylcholine RAb assay using ^{125}I-bungarotoxine, the receptor (R) and anti-R Ab (RAb) are co-precipitated (co-precipitation type) [22,21]. Similar co-precipitation type antibody was detected in the insulin RAb assay [23]. In contrast, TSAb displaces the ^{125}I-TSH binding to TSHR (displacement type). Thus, TSAb binding to TSHR has the characteristics of hormone binding, similar to TSH.
4) The physiological serum concentration of TSAb-IgG is 1 μg/ml, but those of the anti-thyroglobulin Ab and anti-TPO Ab can each be up to 1 mg/ml. Thus, TSAb comprises < 1/1000 of the total IgG [24]. The extremely low concentration of TSAb is similar to that of TSH. Thus, the possibility that TSAb does not act as an antibody remains viable.
5) Immunological similarity was found between animal TSH and TSAb. An IgG antibody that was cross-reactive with animal TSH (e.g., dog, bovine, rat, rabbit, and whale) but not with human TSH was observed in the serum of TSAb-positive Graves' patients [25]. This kind antibody is found in < 1% of Graves' patients. However, it suggests the existence of an antibody with immunological similarity to animal TSH in Graves' disease.
6) It is well known that anti-thyroid drug (ATD) has an inhibitory effect on T4 synthesis through the reducing action of thyroid peroxidase (TPO). However, the existence of other mechanisms is unclear. We previously found that ATD inhibited TSAb binding by the formation of S-H from S-S in the TSHR via a reducing action [26]. This is a novel mechanism of ATD.

Conflict of interest

No conflict of interest exists for the author.

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