Geographical distribution of subclinical autoimmune thyroid disease in Britain: A study using highly sensitive direct assays for autoantibodies to thyroglobulin and thyroid peroxidase

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Abstract. In order to determine whether the geographical distribution of autoimmune thyroid disease in Britain is influenced by the pattern of iodine intake, the prevalence of subclinical disease (detectable antithyroid antibodies in biochemically euthyroid individuals) has been measured in female blood donors from seven towns in England and Wales previously characterised in terms of past and present iodine intake. Thyroglobulin antibody and thyroid peroxidase antibody were measured by highly sensitive assays which are based on the direct interaction between antibody and radiolabelled antigen. Excluding cases of overt thyroid disease (biochemically hypo- or hyperthyroid with thyroid antibodies), the overall prevalences of the antibodies in sera from the 698 female blood donors were 17.8% for thyroglobulin antibody and 17.8% for thyroid peroxidase antibody. Both antibodies were found in 12.3% of the female blood donors. In contrast, the prevalences of thyroglobulin antibody and thyroid peroxidase antibody were 41 and 43%, respectively, in the 117 female relatives of 18 probands with autoimmune thyroid disease, but the highest prevalences were observed in groups of women patients with Graves' disease (N=39) or Hashimoto's disease (N=39) (51, and 97% for thyroglobulin antibody, respectively, and 72 and 97% for thyroid peroxidase antibody, respectively). Antibody prevalence increased with age in the female blood donors rising from 10.6% at age 18-24 to 30.3% at age 55-64 for thyroglobulin antibody and from 14.9% at age 18-24 to 24.2% at age 55-64 for thyroid peroxidase antibody. Geographical differences in the prevalences of both antibodies were not significant and did not correlate with either the previous goitre prevalence or with current differences in iodine intake. Consequently, it seems unlikely that environmental factors play a major role in the development of subclinical autoimmune thyroid disease in the geographical areas studied.

Autoimmune thyroid disease encompasses a spectrum of different clinical conditions varying from overt hypo- or hyperthyroidism (Hashimoto's disease or Graves' disease) to subclinical disease in asymptomatic euthyroid or biochemically hypothyroid subjects. The link between these extremes is the presence of serum autoantibodies directed against thyroid peroxidase (TPO) and/or thyroglobulin (Tg) (reviewed in 1).

Clinical and epidemiological studies have suggested that dietary iodine could play a part in the aetiology of autoimmune thyroid disease (2). Thus the frequency of histological thyroiditis has been reported to increase after iodine prophylaxis (3), and the correction of iodine deficiency in iodine deficient populations appears to be followed by the development of thyroid autoantibodies (4). Furthermore, investigations of spontaneous and induced autoimmune thyroid disease in animals have shown that the prevalence of thyroiditis in susceptible strains of rats (5) or chickens (6) is affected by the iodine content of the diet.

In Britain endemic goitre used to be widespread. However, there has been a threefold increase in
dietary iodine intake over the past 30 years from 80 μg/day in 1952 to 255 μg/day in 1982 (7,8). Nevertheless, geographical variations in iodine intake still persist though the dietary sources of iodine are different (9,10). Variations in the prevalence of hyperthyroidism in Britain correlate with the previous goitre prevalence (11). In contrast, there are little data on whether variations in the prevalence of autoimmune thyroid disease exist and the extent to which they are influenced by the pattern of iodine intake. Consequently, we have investigated the geographical distribution of subclinical autoimmune thyroid disease by measuring the prevalence of Tg/TPO autoantibodies among female blood donors in seven locations in Britain previously characterised in terms of past and present iodine intake. The autoantibodies were detected using recently developed highly sensitive direct assays (12) and the prevalences and antibody levels were compared with those in patients with Hashimoto's disease and Graves' disease.

Subjects and Methods

Serum samples from approximately 100 consecutive female blood donors were obtained from each of seven towns in England and Wales: Aberdare, Chester, Middlesbrough, Newport (Gwent), Plymouth, Preston, and Southampton. The towns were selected to cover a wide geographical spread and a range of previous goitre prevalences. On the basis of a national survey of schoolchildren carried out in 1924 (13), previous goitre prevalences ranged from less than 0.1% in Middlesbrough to 17.4% in Preston. In addition, a further 100 samples were obtained from male donors in South Wales. Sera were obtained between the months of May and November. The sera were frozen (−20°C) within two days of collection and subsequently analysed for autoantibodies to Tg and TPO by direct assay (12). Briefly, sera diluted 1:20 (50-μl aliquots) were incubated with 50 μl of 125-I-labelled Tg or TPO (RSR Ltd, Pentwyn, Cardiff, UK) for 1 h at 37°C. Following the addition of protein A (Pansorbin, Calbiochem, La Jolla, CA), incubation at room temperature for 1 h and addition of 1 ml of assay diluent (150 mmol/l NaCl, 10 mmol/l TRIS-HCl, pH 7.5, 5 g/l bovine serum albumin, and 0.1% Tween 20), the bound and free antibody were separated by centrifugation and aspiration. Samples were classified as positive if Tg or TPO antibody levels were greater than 0.3 kU/l of the MRC anti-thyroglobulin antibody standard 65/93 or the anti-microsome serum standard 66/387, respectively. Details of the assay specificity and coefficients of variation are described elsewhere (12).

Though patients with a history of hypo- or hyperthyroidism are excluded from being blood donors, sera from a number of individuals were found to have abnormal thyroid function tests. Therefore, in order to eliminate bias arising from differences in the rates of diagnosis of thyroid disorders in the towns, all samples with detectable levels of TPO antibody were assayed for serum free thyroxine (Amerlex, Amersham International PLC, UK) and for thyrotropin (Amerlite, Amersham International PLC) to identify hitherto undiagnosed cases of hyper- or hypothyroidism. Donors identified as hyper- or hypothyroid on this basis were excluded from the subsequent analysis. In order to correct for differences in the age distribution of blood donors from the different towns, antibody prevalences were age-standardised using as a standard the rates for all the towns combined.

Sera were also obtained from groups of women patients attending general medical or endocrine outpatient clinics; 39 patients had Hashimoto's disease (diagnosed on the basis of raised TSH and/or low free T4 levels with thyroid autoantibodies) and 39 had newly diagnosed Graves' disease all of whom had measurable TSH receptor antibody (14).

Confidence intervals for the data were calculated by the normal approximation to the binomial, and significance tests carried out using the Chi-squared distribution.

Results

Of the 719 women blood donors tested, 144 (20.0%) had Tg antibody and 145 (20.2%) TPO antibody. After exclusion of 21 women with biochemical evidence of thyroid disease (2 hyperthyroid, 19 hypothyroid), 124 of the remaining 698 women had Tg antibody and 124 TPO antibody, a prevalence of 17.8% for both antibodies. Thirty-eight of the women (5.4%) had Tg antibody in the absence of TPO antibody and 38 women had TPO antibody alone. In the sample of sera from 100 male blood donors 12 were positive for Tg antibody and 10 were positive for TPO antibody.

In order to put the data obtained from blood donors into context, the prevalence of Tg and/or TPO antibody in the women blood donors was compared with the prevalence in patients with Graves' disease, Hashimoto's disease and 117 female relatives of 18 probands with autoimmune thyroid disease previously studied (15). As shown in Table 1, the prevalences of Tg and TPO antibody were lowest in the blood donors, more common among the families of probands with autoimmune thyroid disease, and highest in the groups with Graves' disease and Hashimoto's disease. The relative amounts of Tg antibody and
Comparison of the percentage prevalence of Tg antibody and TPO antibody in the 698 women blood donors with the prevalences in the female relatives of 18 probands with autoimmune thyroid disease, and groups of patients with Graves' disease or Hashimoto's disease. For each group the proportion of sera positive patients with high (>10 kU/l) Tg antibody or TPO antibody levels are shown in parentheses.

<table>
<thead>
<tr>
<th>Group studied (N)</th>
<th>Percent prevalence</th>
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<tbody>
<tr>
<td></td>
<td>Tg antibody</td>
</tr>
<tr>
<td>Blood donors (698)</td>
<td>17.8 (12.0)</td>
</tr>
<tr>
<td>Relatives of families with autoimmune thyroid disease (117)</td>
<td>40.9 (14.5)</td>
</tr>
<tr>
<td>Graves' disease (39)</td>
<td>51 (50)</td>
</tr>
<tr>
<td>Hashimoto's disease (39)</td>
<td>97 (60)</td>
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</tbody>
</table>

TPO antibody were also different in the groups studied. A smaller proportion of women blood donors had high levels (> 10 kU/l) Tg antibody than patients with Hashimoto's disease and Graves' disease (Table 1). Similarly, high levels of TPO antibody (> 10 kU/l) were present in less than 50% of the blood donors compared with high levels in 100% of the patients with Hashimoto's disease (Table 1).

Among the women blood donors, the age-specific prevalence rates of both antibodies increased with age (Fig. 1) rising from 10.6% in the youngest age group (18-24) to 30.3% in the 55-64 age group for Tg antibody and from 14.9% in the 18-24 age group to 24.2% at age 55-64 for TPO antibody. Similar age distribution patterns were observed for women with Tg antibody only, or TPO antibody only (data not shown).

Fig. 2 shows the geographical variation in the prevalence of Tg/TPO antibodies among the women blood donors. The towns have been divided into two groups according to their previous goitre prevalences in a national survey in 1924 (13); goitre prevalences of greater than 5% being observed in Chester, Preston, Aberdare and Plymouth and less than 5% in the remaining three towns. The age-standardised prevalences of Tg antibody ranged from 12% in Newport to 22% in Plymouth and for TPO antibody from 15% in Newport to 23% in Southampton. Although prevalences of both Tg and TPO antibodies were somewhat lower in Newport and Middlesbrough, there was no evidence of a statistically significant geographical variation (Tg antibody $\chi^2=4.43$, 6 degrees of freedom (df), $p>0.1$; TPO antibody $\chi^2=3.63$, 6 df, $p>0.1$) and in particular no association with the previous goitre prevalences. The percentage of cases with high Tg antibody or TPO antibody titres (>10 kU/l) did not vary significantly between the towns (data not shown).

Previously published data on current iodine intakes were available for five of the towns (10). These data, based on measurements of urinary iodide excretion in samples of between 52 and 74 women aged 25-64 randomly selected from General Practitioners' lists in each town (obtained in February when iodine intakes in the UK are highest and May when intakes are lowest), also showed no correlation with the prevalences of Tg and TPO antibodies (Table 2).
Fig. 2.
Geographical variations in the prevalence (mean and 95% confidence intervals) of thyroglobulin (Tg) antibody and thyroid peroxidase (TPO) antibody in seven towns in Britain which have been grouped according to whether the previous goitre prevalences were high (Chester, Preston, Aberdare and Plymouth) or low (Southampton, Newport, Middlesbrough).

Table 2.
Comparison of the average urinary iodide excretion (µg/g creatinine) in winter (February) and summer (May) in casual specimens from women aged 25-64 in five of the towns (ranked in order of the winter iodide excretion) with the prevalences of Tg antibody and TPO antibody.

<table>
<thead>
<tr>
<th>Town</th>
<th>Urinary iodide excretion (µg/g creatinine)</th>
<th>Percent prevalence of antibody</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Winter</td>
<td>Summer</td>
</tr>
<tr>
<td>Southampton</td>
<td>126</td>
<td>78</td>
</tr>
<tr>
<td>Plymouth</td>
<td>124</td>
<td>88</td>
</tr>
<tr>
<td>Middlesbrough</td>
<td>106</td>
<td>66</td>
</tr>
<tr>
<td>Preston</td>
<td>102</td>
<td>97</td>
</tr>
<tr>
<td>Chester</td>
<td>102</td>
<td>76</td>
</tr>
</tbody>
</table>

Spearman rank correlation coefficient between winter urinary iodide excretion and Tg antibody, 0.39 p>0.1 and TPO antibody, 0.67 p>0.1
Discussion

Tg or TPO autoantibodies were detectable in 18% of an apparently healthy group of women blood donors compared with 43% of the female relatives of patients with autoimmune thyroid disease and over 70% of patients with overt autoimmune thyroid disease. Titres of autoantibodies were lowest in healthy blood donors or the relatives of autoimmune thyroid disease probands and were highest in Hashimoto's disease in accordance with the evidence for a role of these autoantibodies in thyroid damage (1). As in previous studies (16), the prevalences of both antibodies were much higher in women than in men.

The prevalence increased with age, approaching 25% for TPO antibodies and 30% for Tg antibodies by the sixth decade of life. These values are higher than previously reported prevalences which range from 8 to 26% in women compared with 3 to 6% of men (16), but our observations are supported by the histological evidence of thyroiditis (more than 10 foci per square cm of thyroid tissue) in 22% of adult women and 6% of men (17). The increasing prevalence of thyroid autoantibodies with age has been documented in other studies. In a survey of Australian women, Hawkins et al. (18) reported a 7.5% prevalence of thyroid microsomal antibody at age 20-30 which increased to 15% in 50-60 year-old women. In the Whickham study in North-East England, thyroid microsomal antibodies were detectable in 7.9% of young women (aged 18-24) rising to 13.7% at age 45-54 years (19). However, in these earlier studies, antibodies were measured by agglutination or immunofluorescence techniques, which are less sensitive than the new direct assay methods we have used. In contrast to these earlier studies (19), our investigations showed the prevalences of Tg and TPO antibodies to be almost identical, although small similarly sized subgroups had either one (but not both) antibodies.

We found no evidence of significant geographical variations in the prevalence or the levels of TPO and/or Tg autoantibodies. Furthermore, there was no association between the prevalence or the level of TPO and/or Tg autoantibodies and the patterns of iodine intake in Britain. The lack of geographical variation in the prevalence of thyroid autoantibodies contrasts with the fivefold differences observed in the incidence of hyperthyroidism in Britain (11). This is consistent with our hypothesis that genetic predisposition is of major importance in the tendency to produce thyroid autoantibodies (14), whereas environmental factors have a major role in the development of clinically recognisable autoimmune thyroid disease.

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References


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