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Thyroid Function and Immune Profile in Rheumatoid Arthritis. A Controlled Study

A.P. ANDONOPOULOS, V. SIAMBI, M. MAKRI, M. CHRISTOFIDOU*, C. MARKOU, A.G. VAGENAKIS

Summary **The aim of our study was to determine the prevalence of thyroid dysfunction and autoimmune abnormalities in rheumatoid arthritis (RA) and to further investigate the possible association between D-penicillamine and autoimmune thyroiditis. For this purpose, one hundred and one unselected consecutive patients with RA and 70 age and sex matched controls were studied prospectively. Evaluation included a complete history and physical examination with special attention to symptoms suggestive of thyroid pathology, routine laboratory and serologic immune profile, plus determination of serum levels of thyroxine (T4), triiodothyronine (T3), thyroid stimulating hormone (TSH), antibodies to thyroid peroxidase (AbTPO) and TSH receptor antibodies (TRAB). Serum thyroxine binding globulin (TBG) was measured in all subjects with high thyroid hormone levels, whereas free T3 and T4 concentrations were determined in all individuals with abnormal T3, T4, TSH or TBG.**

Six patients with hyperhyroidism, 3 with hypothyroidism and 1 with the euthyroid hyperthyroxinemia (EH) syndrome were found, whereas four of the controls had hyperthyroidism. Thirteen patients and 6 controls had high AbTPO levels whereas no one had high TRAB. No association was detected between thyroid abnormalities and any serologic RA finding. Furthermore, no correlation between thyroid dysfunction and elevated AbTPO's was found.

A relatively high prevalence of thyroid dysfunction (9,9%) and subclinical autoimmune thyroiditis (12,9%), the latter indicated by elevated AbTPO's, was found in our RA patients. These figures were higher than those in the control group (5,7% and 8,6% respectively), but the difference did not reach statistical significance. Of further interest may be our finding that, despite anecdotal reports blaming D-penicillamine for cases of autoimmune thyroiditis, the incidence of the latter was similar among recipients and nonrecipients of the drug. Similarly, TRAB were not detected in any patient treated with D-penicillamine.

Key words **Thyroid Function, Antibodies to Thyroid Peroxidase, TSH Receptor Antibodies, Rheumatoid Arthritis, D-Penicillamine, Thyroiditis.**

INTRODUCTION

An association between autoimmune thyroiditis and rheumatoid arthritis has long been detected. The data mostly derive from a few studies (1-3), dating back to the early sixties, in which the incidence of RA in Hashimoto's thyroiditis or that of thyroiditis in RA is examined. However, the prevalence of thyroid dysfunction or that of thyroid autoimmunity in RA, using recently introduced sensitive assays, has not been determined. In a recent study an increased incidence of hypothyroidism in RA was found, but the population had not been tested for antithyroid antibodies (4). For this purpose, we undertook a prospective controlled study of a large unselected RA population, employing well accepted modern techniques, for the detection of both thyroid dysfunction as well as autoantibodies in the patients' sera, that would suggest an immune mediated thyroid disorder.

A further reason for the latter was to examine the possible role of D-penicillamine on the induction of autoimmune thyroiditis, indicated by elevated AbTPO's (5), because this drug has been anecdotally implicated, proba-

Department of Medicine and *Department of Microbiology, University of Patras School of Medicine, Patras, Greece.

bly within the spectrum of a variety of autoimmune syndromes it can cause (6). Furthermore, TSH receptor antibodies (TRAB) were sought, looking especially for involvement of D-penicillamine in such a process, probably through a mechanism similar to that, whereby this drug induces antibodies to acetylcholine receptor, resulting in myasthenia gravis (7,8).

SUBJECTS AND METHODS

One hundred and one unselected, consecutive patients with rheumatoid arthritis, according to the 1987 ARA revised criteria (9), attending the outpatient clinic, were prospectively evaluated for evidence of thyroid disease, irespective of RA activity. Seventy, age and sex matched individuals with osteoarthritis, seen at the outpatient rheumatology clinic, but otherwise healthy, underwent the same clinical and laboratory evaluation and were used as controls. A complete history and physical were obtained, whereas symptoms and signs suggestive of thyroid dysfunction were particularly recorded. Besides routine laboratory, in the patients' and controls' sera levels of C reactive protein (CRP), rheumatoid factor (RF) and C3 and C4 complement components were determined by nephelometry (Beckman Array Protein System, USA), antinuclear antibodies by indirect immunofluorescence (Kallestad Quantafluor Fluorescent Autoantibody Test, Sanofi Diagnostics Pasteur Inc, USA) and antibodies to ds DNA and extractable nuclear antigens Ro(SSA), La(SSB), Sm and nRNP by ELISA (Shield Diagnostic Ltd, Dundee UK). Furthermore, in all sera levels of T4 were determined by fluorescence polarization immunoassay (Abbott IMX Autoanalyser, USA, normal range 4,5-12,0 micrograms/dL), T3 and TSH by microparticle enzyme immunoassay (Abbott IMX Autoanalyser, USA, normal ranges 0,8-2,0 ng/mL and 0,1-5,0 microunits/mL respectively), AbTPO's by radioimmunoassay (Sorin Biomedica, France, normal $<$ 10,0 units/mL) and TRAB by radioreceptor assay (Henning Berlin GMBH, Germany, normal $<$ 9,0 units/L). Finally TBG was measured in all sera, exhibiting high thyroid hormone levels, by radioimmunoassay (Incstar Corp, Stillwater, USA, normal range 13,0-30,0 micrograms/mL), whereas free T3 and T4 in all sera exhibiting abnormal T3, T4, TSH or TBG levels, by Amerlex immunoassay (Kodak Clinical Diagnostic, Bucks, UK, normal range for free T3 2,0-9,0 pmol/L and for free T4 9,0-24,0 pmol/L).

Elevation of serum TSH was diagnostic of hypothyroidism, overt or subclinical, whereas a level below 0,1 microunits/mL, by this very sensitive assay, was diagnostic of hyperthyroidism in our population, which consisted of non-critically ill patients. Elevated serum levels of thyroid hormones combined with a normal TSH and a high TBG suggested the euthyroid hyperthyroxinemia syndrome.

Administration of D-penicillamine, since it has been blamed for the induction of a variety of autoimmune syndromes (6), was recorded. The criteria of Vitali et al. were used for the detection of coexistent secondary Sjogren's syndrome (10).

For statistical analysis of the results the chi square test with Yates' correction and the Student t test were employed, where indicated.

RESULTS

Our 101 patients consisted of 29 males and 72 females with mean age 57,9 ± 13,5 years and mean duration of the disease 8,3 ± 7,7 years. The majority were of functional class II. Twenty-one males and 49 females with mean age 58,1 ± 10,8 years comprised our control population. At the time of the study, 31 of the patients had been taking D-penicillamine for a mean of 24,9 ± 16,2 months (range 8-72 months), at a dosage of 250-500 mg daily. Other second line drugs included dihydroxychloroquine in 34 patients, oral gold in 6 and methotrexate in 21, whereas 25 patients altogether were receiving 4 mg or less of methylprednisolone daily, as an adjunct to a major disease modifying medication. Rheumatoid factor was present in 45,0% of the patients, ANA's in 40,6% and anti-Ro(SSA) antibodies in 5,0%. Rheumatoid nodules were seen in 12 patients (11,9%) and secondary Sjögren's syndrome in 15 (14,9%).

Six patients (5,9%) with undetectable serum TSH, compatible with hyperthyroidism not previously diagnosed, were found. High levels of serum total thyroid hormones in three of them and of free fractions in all were also seen. Another three patients (3,0%) with elevated TSH levels, consistent with hypothyroidism, were detected.

Finally, there was one patient (1,0%) with elevated T4 and TBG but normal TSH and free T3 and T4 levels, suggestive of the EH syndrome. In general, all the aforementioned patients with thyroid dysfunction had mild or no symptoms or signs of overt thyroid disease. In the remaining patients serum T3, T4 and TSH levels were normal. The serum T3, in particular, was within normal range, suggesting that none of our patients was severely ill.

Regarding thyroid immune profile, 13 patients (12,9%), all female, had increased levels of AbTPO's (mean ± sd: 82,6 ± 83,5 u/mL), whereas the corresponding figure of the remaining 88 patients was 4,1 ± 1,0 u/mL. None of the patients had elevated TRAB, including those with hyperthyroidism.

There was no correlation between thyroid dysfunction and immune abnormalities, since only one patient out of the 10 with thyroid dysfunction, who had hypothy-

Table I: *Comparison of patients and controls regarding thyroid dysfunction and immune abnormalities.*

	Numbers of individuals (and percentages) with					
	High AbTPO's	High TRAB	Hyperthyroidism	Hypothyroidism	EH syndrome	Overall thyroid dysfunction
Patients	13 (12,9%)	0 (0,0%)	6 (5,9%)	3 (3,0%)	1 (1,0%)	10 (9,9%)
Controls	6 (8,6%)	0 (0,0%)	4 (5,7%)	0 (0,0%)	0 (0,0%)	4 (5,7%)

roidism, had elevated AbTPO's. Furthermore, no association was detected between either thyroid dysfunction or immune disorder and any clinical or serologic feature of the underlying rheumatoid disease. However, the patients with thyroid dysfunction, but not those with thyroid autoimmunity, had significantly shorter duration of their rheumatoid disease compared with the rest (5,5 ± 4,8 years vs 8,7 ± 8,2 years, $p < 0,035$).

Three patients of those 13 with elevated AbTPO's had been on D-penicillamine and one of these three had hypothyroidism (the above already mentioned). The incidence of elevated AbTPO's, suggestive of subclinical autoimmune thyroiditis (5), was not statistically different between recipients (3 of 31: 9,7%) and nonrecipients (10 of 70: 14,3%) of D-penicillamine (p=NS). As a matter of fact, according to these figures, autoimmune thyroiditis was less common in the patients treated with D-penicillamine.

Four of the 70 control individuals (5,7%) had undetectable serum TSH levels, suggestive of hyperthyroidism, subclinical in all, and one of them had elevated AbTPO's. Six of the controls, all female, (8,1%) had elevated AbTPO'5s (mean ± sd: 463,7 ± 173,8 u/mL) whereas in the remaining 64 the serum concentration of the antibody was 3,9 ± 1,3 u/mL. TRAB serum levels were normal in all the control persons. Again, no correlation between thyroid dysfunction and immune profile abnormalities was detected in our controls.

When patients and controls were compared, no statistically significant differences were found, regarding either thyroid dysfunction or immune abnormalities (Table I).

Finally, all patients and controls with hyperthyroidism were subjected to ultrasound and photoscanning evaluation of the thyroid gland, which revealed a multinodular goiter in all. The EH syndrome, seen in one patient, was thought to be secondary to the underlying RA or to an inherited increase of serum TBG, since no other acquired cause for the elevated TBG concentration could be identified (11).

DISCUSSION

The first reports of an association between autoimmune thyroiditis and connective tissue disease such as rheumatoid arthritis, Sjögren's syndrome and systemic lupus erythematosus (SLE) appeared in the early sixties (1-3). Briefly, the literature available from that time can be summarized as follows: In 1961 W. Hijmans et al. (1) investigating autoantibody production in SLE, RA and Hashimoto's thyroiditis (HT), found that the incidence of thyroid antibodies in RA was not significantly higher than in controls. In the same year, W.W. Buchanan et al. (2) found the prevalence of RA in HT significantly higher than in controls, whereas that of thyroid antibodies significantly higher in RA patients than in controls. In 1963 K.L. Becker et al. (3) found a 4,0% incidence of RA in HT. Later, in 1973 K. Whaley et al. (12) reported an 18,3% prevalence of thyroglobulin antibodies in patients with RA and Sjögren's syndrome. For some time since those days, although technology had been greatly improved, an attempt to reassess the relationship between RA and thyroid disease has not been made, although two studies have appeared relatively recently dealing with the relationship of SLE and thyroid disease (13,14). It was only very recently, in 1993, when a controlled study examined thyroid status in RA (4) and found an excess of thyroid dysfunction, mostly hypothyroidism. However, the immune thyroid profile was not evaluated at all in that study.

We therefore aimed at assessing first the thyroid functional status in RA, and second the thyroid immune profile in this disease, using sensitive radioimmunoassays, not available at the time of the initial descriptions of the aforementioned association or not included in the relatively recent considerations of the subject.

Our data showed a higher overall prevalence of thyroid dysfunction among RA patients (9,9%) than in controls (5,7%), but the difference did not reach statistical significance. Furthermore, this dysfunction was not related to any serologic marker of autoimmune thyroiditis in both patients and controls. This suggests that the functional disorder was not of autoimmune origin, which was rather unexpected in the RA group. The relatively high prevalence of thyroid dysfunction in the RA patients (9,9% versus 5,7% in the controls), although not clinically obvious, is worth noting and clinicians should become sensitized to that. In particular, hypothyroidism was detected in 3 patients versus none among the controls, and this difference, although not statistically

significant, is compatible with the results of a recent, already mentioned study (4). Hyperthyroidism, due to multinodular goiter, was equally common among our patients and controls. This was not of autoimmune aetiology, as judged by the morphology of the gland, the paucity of ABTPO's and the absence of TRAB. Furthermore, it is known that multimodular goiter is the prevailing type of hyperthyroidism in this age group.

Thyroid autoimmunity, as expressed by the presence of antibodies to thyroid peroxidase, was found in 12,9% of the patients vs 8,6% in the controls. The difference, although it did not reach the level of statistical significance, suggests an increased tendency of patients with RA to develop thyroid autoimmunity. This is compatible with the results of the initial studies (1,2,12), although the latter had not employed AbTPO's. These antibodies have been considered markers for autoimmune thyroiditis (5), and even a pathogenic role has been attributed to them (15), although the latter has been recently disputed (16). Thyroid function was normal in all patients and controls with elevated AbTPO's, except for one individual in each group. It has been postulated, however, that such individuals have silent autoimmune thyroiditis and it is worth following them for development of overt disease (5).

In the present study, we did not test our population for the presence of antithyroglobulin antibodies (anti-TG's). This because AbTPO's have been proven much more sensitive than anti-TG's and furthermore, when both are present, the titers of AbTPO's are higher (17,18). It is very rare to find anti-TG's alone in the absence of AbTPO's, whereas the opposite is common (17-19), so that it has been recommended that, for clinical purposes, measurement of AbTPO's is more than adequate (18).

Of interest, in our study, was the unique presence of AbTPO's only in female individuals, both among patients and controls. This, however, was rather expected, since females significantly outnumbered males in both groups, and furthermore since it is known that autoimmune thyroiditis affects females at least five times more frequently than males (18).

We cannot provide an adequate explanation for the significant difference observed between the levels of elevated AbTPO's in the controls compared to the patients. Probably, steroid consumption by the patients at the time of the study or previously might have contributed to a decrease in their AbTPO serum levels.

Finally, another finding from this study was the absence of correlation between D-penicillamine administration and the presence of TRAB or AbTPO's in the sera of the study individuals. D-penicillamine is known to cause a variety of autoimmune syndromes, including myasthenia gravis, myositis, pemphigus, SLE like and Goodpasture's syndrome (6-8,20-23). One, therefore, could probably expect induction of TRAB and possible Graves' hyperthyroidism (24) by D-penicillamine in our patients who received the drug, through a mechanism similar to that responsible for the production of anti-acetylcholine receptor antibodies resulting in myasthenia gravis (25). However, no increase in TRAB title was observed in any of the study individuals. On the other hand, autoimmune thyroiditis has been mentioned in an extensive review concerning chemistry and clinical use of the drug, in 1986 (6); however, no references for this particular side effect were provided. A meticulous search of the literature by us identified only a short case report in French, by F. Delrieu et al. (26). It is unclear from this report, though, whether D-penicillamine was responsible for the thyroiditis of the patient, who had rheumatoid arthritis and myasthenia gravis as well. It is more likely that thyroiditis had happened within the context of RA and not as a result of D-penicillamine administration. In the present study no evidence for D-penicillamine causing autoimmune thyroiditis was found, since there was no statistically significant difference, regarding elevated AbTPO's, between recipients and nonrecipients of the drug. Indeed, AbTPO's were more common in those RA patients who did not receive D-penicillamine. This, in combination with the absence of convincing evidence from the literature, suggests that autoimmune thyroiditis should be excluded from the list of side effects of D-penicillamine.

In conclusion, the present study suggests that thyroid dysfunction and autoimmunity, not necessarily related to each other or to D-penicillamine, are relatively common and run a subclinical course in rheumatoid arthritis. Thyroid function at least should be checked in this disease. Whether the same could be suggested for thyroid immune profile is a matter of debate, depending probably on the elucidation of the physical history of the presence of thyroid autoantibodies and its consequences (5).

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Correspondence to: A. P. ANDONOPOULOS, MD, FACP
Associate Professor of Medicine/Rheumatology, Department of Medicine, University of Patras School of Medicine, Patras Regional University Hospital, 265 00 Rio, Patras, GREECE.