Short report

High CTG repeat number in nodular thyroid tissue from a myotonic dystrophy patient

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Abstract

CTG triplet expansion was studied in lymphocytes and thyroid tissue in a patient with myotonic dystrophy (DM) and associated thyroid nodular disease. An approximately 7 fold larger amplification was found in abnormal thyroid tissue compared to lymphocytes, suggesting that anomalies in the putative DM kinase gene might contribute to thyroid dysfunction.

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The recent characterisation of the genetic defect in myotonic dystrophy (DM) as the presence of an abnormal amplification of the CTG triplet in the 3' untranslated region of a putative protein kinase mRNA provided an explanation for various aspects of the disease.¹

Although the mechanisms leading to repeat expansion remain unclear, meiotic instability of repeat number has been extensively shown and provides an explanation for the phenomenon of anticipation. In addition, somatic tissue specific amplification has been detected in several tissues, most particularly in muscle, but also in liver, testes, and brain, well known targets of the disease, where an increased level of expansion compared to that in lymphocytes has been found.¹² Interestingly, somatic heterogeneity of CTG expansion was not detected in two fetuses.³ Although triplet amplification is not systematically correlated with disease severity, it presumably plays a significant role in tissue dysfunction. The presence of somatic mosaicism raises the question as to whether tissue specific amplification might account for some of the widely known clinical variations of the disorder, as has been described recently in muscle.²

We had the opportunity to study a female patient with myotonic dystrophy and associated thyroid disease in whom a larger expansion of the CTG repeat was found in abnormal nodular thyroid tissue, compared to lymphocytes, providing further evidence for genetic mosaicism. The patient, aged 34 years, was diagnosed as having DM after giving birth to a child with congenital DM. Her myotonic symptoms were very mild and there was no evidence of cataract or frontal balding; she was intellectually normal. She had undergone two partial thy-

roidectomies for cold nodules, nine and 17 years respectively before the recent discovery of another thyroid nodule for which she sought medical attention. On examination, a 3 cm diameter mass was felt at the top of the left thyroid lobe. A left thyroidectomy was performed and showed multinodular goitrous lesions, with no histological evidence of malignancy. A fragment of nodular thyroid tissue was taken for DNA extraction and analysis.

Southern blot analysis of CTG expansion was carried out using probe p25B1.4 (kindly given by K Johnson). After *EcoRI* digestion, the probe indicated the presence of a 23.1 kb fragment in thyroid tissue, compared to a 13.6 kb fragment in lymphocytes. A 12.0 kb fragment corresponding to the normal allele was detected both in lymphocytes and in thyroid tissue, so that CTG expansion in thyroid tissue (23.1 kb minus 12.0 kb) was about seven times greater than that in lymphocytes (13.6 kb minus 12.0 kb); similar results were obtained after *BglII* and *BamHI* digestion. Comparable analyses were performed on lymphocyte DNA from other family members. A minimal but significant expansion of 200 bp, with a fragment of 12.2 kb (normal fragment at 12.0 kb), was found in the asymptomatic patient's father; a sister had a fragment of 12.4 kb, corresponding to an expansion of 400 bp, and a band of 15.8 kb (expansion of 3800 bp) was found in the patient's son. This progressive increase of triplet expansion in three successive generations, correlated with increased severity of symptoms, provides a further example of anticipation.

The observation of a larger amplification of CTG triplets in thyroid tissue than in lymphocytes has not been reported previously and raises intriguing questions. First, it provides further evidence for somatic mosaicism of triplet number and for the instability of triplets during the mitotic as well as the meiotic process. Such a tissue specific expansion of triplet number was recently shown in several tissues, including the liver, testes, and brain, but has not been described before in thyroid tissue.² This tissue heterogeneity was not present in fetuses with congenital DM,³ suggesting that the potential for mosaicism is greatest in adult onset patients. In order to assess whether triplet expansion in the DM kinase gene might be associated with the development of goitre in

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DNA fragment sizes (in kb) in nodular thyroid tissue (T) and lymphocytes (L) of DM patient, and in a control subject (C). Right lane: lambda HindIII molecular weight markers. Southern blot analysis carried out with probe p25B1.4, after EcoRI digestion. Two alleles of 10.4 and 12.0 kb are found in the normal population.

conditions other than myotonic dystrophy, analyses of CTG triplet expansion were carried out on DNA extracted from blood lymphocytes

and normal and nodular thyroid tissue in five patients with cold thyroid nodules. In all samples, the fragments were within the normal range, suggesting that this genetic anomaly does not play a significant role in the development of thyroid nodules in the general population. Although the occurrence of thyroid disease in DM is not generally appreciated, an incidence of thyroid dysfunction as high as 15 to 20% has been clearly documented.⁴

The observation of an enormous triplet expansion in abnormal thyroid tissue versus peripheral blood suggests that repeat amplification in thyroid tissue contributes to its dysfunction. It could be interesting to assess whether somatic mutations of the DM kinase gene might be associated with the development of goitre in conditions other than myotonic dystrophy.

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- 2 Anvret M, Ahlberg G, Grandell U, *et al.* Larger expansions of the CTG repeat in muscle compared to lymphocytes from patients with myotonic dystrophy. *Hum Molec Genet* 1993;2:1397-400.
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- 4 Fukazawa H, Sakurada T, Yoshida K, *et al.* Thyroid function in patients with myotonic dystrophy. *Clin Endocrinol* 1990;32:485-90.