Diagnostic utility of thyroglobulin measurement in the fine needle aspirates from cervical lymph nodes: a case report

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SUMMARY: Diagnostic utility of thyroglobulin measurement in the fine needle aspirates from cervical lymph nodes: a case report.

Fine needle aspiration cytology (FNAC) is the more accurate diagnostic method for cervical lymph node (CLN) metastasis from differentiated thyroid cancers (DTC). However, FNAC diagnosis of cystic CLN is, in most cases, uninformative due to inadequate cellularity. Recently, thyroglobulin (Tg) detection in FNAC needle washout fluid has been shown to improve the diagnostic accuracy of FNAC, and its routine association with cytology is recommended. We here describe the case of a 20 yr old girl complaining of the recent appearance of palpable non-painful laterocervical nodes in the neck. Ultrasound examination revealed the presence of 3 cystic CLNs and 2 mixed thyroid nodules, with the larger one showing irregular margins. On the latter, and on 2 larger CLNs, FNAC was performed, and both Tg protein and mRNA were determined in the needle washout. The cytological analysis was not diagnostic for the two CLNs, while that of the thyroid nodule reported the presence of iodinated colloid groups of thyrocytes with normal morphology. Both CLNs showed, however, high levels of Tg protein and were positive for Tg mRNA, suggestive of metastatic DTC. Based on these findings, the FNAC analysis was performed on the second smaller thyroid nodule suggesting (Tir4) the presence of PTC. The patient was then subjected to total thyroidectomy with lymph nodes resection of the central and homolateral compartments. The histological diagnosis confirmed the presence of a PTC in the small nodule and metastatic lymph nodes.

In conclusion, this case confirms that the cytological diagnosis of cystic lymph nodes is challenging, and that the measurement of Tg protein and/or mRNA in the needle washout may overcome this limitation.

KEY WORDS: Thyroid cancer - Lymph node - Metastasis - Diagnosis - Thyroglobulin - Fine-needle aspiration cytology - Follow-up.

Introduction

Thyroid cancer represents the most frequent endocrine neoplasia, being the fifth most common tumor in women in the United States (1-4). The majority of thyroid carcinomas arise from the epithelial follicular thyroid cells and are represented by differentiated carcinomas (DTC), subdivided into papillary (PTC) and follicular (FTC) histotypes, while 1-2% of them are undifferentiated highly aggressive anaplastic carcinomas (ATC) (3-5). About 3-4% of thyroid cancers derives from the parafollicular C cells and is named medullary thyroid carcinoma (MTC) (3-6). The prognosis for thyroid cancer patients relies on clinical parameters, that is cancer type and stage of the disease at the time of diagnosis (2, 4, 7-11). The prognosis of DTC patients is favorable, with 10-year survival rate of about 90%, while that of ATC patients is very poor with 5-year survival below 5% (2, 4). The prognosis of MTC patients depends essentially on the state of the disease at diagnosis, with a 10-year survival rate of 90% when the disease is confined to the gland, of 70% when loco-regional metastases are present, and of 20% when distant metastases occur (2, 4).

Thyroid neoplasms manifest themselves as nodules and are very common, affecting 19% to 67% of the adult population (6, 7). Since only a minority of them (about 5%) harbors a malignant lesion the first aim of their clinical evaluation is to exclude malignancy (4, 12, 13). The diagnosis is based on patient’s clinical parameters, ultrasonography (US), scintigraphy, and fine needle aspir-
ration cytology (FNAC) (4, 12, 14-16). In addition to thyroid nodule, the accurate diagnosis of metastases to loco-regional lymph nodes is also crucial for the initial surgical approach as well as during patients’ follow-up (17-19). To date, FNAC has represented the main diagnostic tool for cervical lymph node (CLN) metastasis (20-23). However, inadequate cellularity, often associated with cystic lymph nodes, prevents diagnosis in about 20% of samples (24, 25).

Over the last years, different reports have suggested that measurement of thyroglobulin (Tg) protein and/or mRNA in the FNAC needle washout fluid may increase the diagnostic performance of FNAC in identifying CLN metastases from DTC (26-34). In line with these expectations, we here describe the case of a 20 yr old girl suffering from lymph node metastatic PTC, demonstrating the clinical utility of Tg protein and mRNA detection in the fine needle washout.

Case report

In December 2005 a 20 yr old girl was admitted to the Endocrinology Unit of the Umberto I General Hospital of Rome after complaining of the recent appearance of palpable, mobile and non-painful laterocervical nodes in the left side of the neck. The patient had normal thyroid function as revealed by normal TSH, FT$_4$, and FT$_3$, serum levels, with negative anti-TPO and anti-Tg antibodies. There was no familial history of thyroid cancer or previous exposure to either medical (radiation treatment to the head and neck region) or environmental (from fallout following nuclear power plant accidents) radiation.

US performed on the cervical area using the Aplio XV (Toshiba, Japan) system equipped with a linear transducer (PLT-805AT) revealed the presence of three enlarged cervical lymph nodes, two of which (lymph nodes A and B) are depicted in Figure 1. The latter showed a cystic appearance and diameter ratios comprised between 1.5 and 2.0. In addition, color-flow doppler analysis demonstrated in both an irregular vascularization (hilar and peripheral). Within the thyroid gland US evidenced two nodules (Figure 2: the first (nodule A of figure 2), mixed with regular margins and major diameter of 14.2 mm, was localized in the upper left lobe, while the second (nodule B of figure 2), mixed with irregular margins and major diameter of 22.1 mm, was localized in the inferior part of the left lobe.

Based on clinical and US examinations the patient was selected for FNAC and instructed not to take aspirin or any other anticoagulant in the 5 days prior to biopsy. The dominant thyroid nodule (nodule B of figure 2) and the two lymph nodes described in figure 1 were analyzed. To this end, a 25-gauge needle attached to 20 ml plastic syringe was used to aspirate nodes under US assistance. All aspirates were smeared directly on glass slides which were either hair-dried or wet-fixed using the Bio-Fix (Bio-Optica, Milan, Italy). Hair-dried slides were stained with the May Grunwald-Giemsa solution, while the wet-fixed slides were stained with the Papanicolau solution. The slides were then interpreted by a cytopathologist expert in thyroid cytology.

At the same time, the needle was washed with 1 ml of phosphate buffered saline by multiple pumping actions, and the suspension was centrifuged at 1200 rpm for 5 min. The supernatant was collected and analyzed for Tg protein by means of the immunoluminometric assay (ILMA) Tg-PluS (B.R.A.H.M.S., Hennigsdorf, Germany), while total RNA was extracted from the cell pellet and analyzed for Tg and calcitonin (CT) mRNA content as previously described (35, 36). The RT-PCR demonstrated the presence of Tg mRNA, but not CT mRNA, in the thyroid nodule (Figure 3). Similarly, the two lymph nodes analyzed were positive for Tg mRNA and negative for CT mRNA, suggesting the presence of metastases from DTC. These data were corroborated by the high level of Tg protein observed in both lymph nodes, that is 6620 ng/ml in lymph node A and 7480 ng/ml
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in lymph node B. However, the cytological diagnosis reported the presence of colloid and groups of thyrocytes with normal morphology in the thyroid nodule aspirated materials, while both lymph node FNACs were non diagnostic due to the presence of few lymphocytes and red blood cells.

Based on Tg mRNA and protein values found in the lymph nodes, it was decided to perform FNAC also on the second smaller thyroid nodule (nodule A), which suggested the presence of PTC. As a consequence, the patient was subjected to total thyroidectomy with lymphadenectomy of the central and homolateral cervical lymph node compartments. Histological analysis of the removed tissues (Figure 4) confirmed the presence of PTC in the apical portion of the left lobe, and a benign nodule in the inferior pole of the left lobe. In addition, three lymph nodes of the central compartment and three of the laterocervical compartment (the latter showed in figure 4) were metastatic.

Discussion

The accurate diagnosis of loco-regional lymph node metastasis is of primary importance for the initial surgical approach as well as for prognostic stratification and follow-up of thyroid cancer patients (17-19). FNAC represents the main diagnostic tool for the diagnosis of metastatic CLN from DTC, but the diagnosis may be challenging as lymph nodes could harbor metastases from a multiplicity of extra-thyroidal malignancies or be affected by several non-tumoral diseases (21-23). As a consequence, the accuracy of the technique, which was reported to vary from 73% to 94%, is highly dependent on the experience and ability of the cytopathologist (28-34). Different clinical reports demonstrated that Tg measurement in fine-needle washout may significantly improve the accuracy of FNAC in the evaluation of metastatic lymph nodes from DTC, and hence the routine association of Tg measurement with FNAC in the preoperative diagnosis of suspicious CLN is currently recommended (18, 19, 28-34). In this context, we here report our experience on a case of a 20 yr old girl suffering from metastatic PTC, for which Tg protein and mRNA detection in the needle washout of CLN provided the accurate diagnosis, making up for uninformative FNAC due to cystic CLN. It has to be mentioned, however, that in our experience, based on a case study of 33 CLN, we found that FNAC and Tg protein or mRNA determination in the needle washout showed a 90% overall agreement, and that the molecular diagnosis became clinically useful only in cases in which FNAC provides uninformative dia-
nagnosis, as in the present case report, or inconsistent diagnosis with regard to patient's biochemical and/or clinical parameters (unpublished observations). Based on such evidence, in our opinion FNAC remains the gold standard technique for the diagnosis of suspicious CLN due to its ability to efficiently diagnose metastases not only from thyroid cancers, but also from other primary tumors.

Conclusion

Thyroglobulin protein and/or mRNA determination in fine needle washout is a clinically useful method to improve the diagnostic accuracy of fine needle aspiration cytology of cystic cervical lymph nodes suspected to harbor a metastasis from differentiated thyroid cancers.

References