

## Evaluation of multiple stemness markers in normal thyroid and in thyroid carcinoma (TC) specimens

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The tumorigenic capacity in cancer, including TC, is confined in a small subpopulation of stem-like cells termed cancer stem cells (CSCs). To date, markers for thyroid CSCs remain to be confirmed. Thus, the identification of a specific antigen to distinguish thyroid CSCs from normal SCs represents a crucial step to develop targeted therapies. The aim of this study was to investigate the expression of stemness markers and to isolate putative SCs from both normal thyroid and TC.

Normal and neoplastic tissue samples were harvested from a cohort of 19 patients affected by TC, who underwent thyroidectomy. Fresh specimens were enzymatically digested. Cell suspensions were characterized by flow cytometry using anti-CD133, -CD45, -CD31, -CD44 and -CD105 antibodies. A portion of cells was suspended in SC medium (SCM) in order to obtain thyroid spheres.

CD133+ and CD105+ cell rates were significantly higher in TC than in normal thyroid tissue (2.5% vs. 0.1% and 3.9% vs. 0.5% respectively,  $p < 0.02$ ). No difference was found regarding CD44+. In TC both CD45+ and CD31+ cell rates were 1.3%, but 1.45% of CD133+ cells were also positive for CD45 and 0.8% for CD31. In normal tissues CD45+ and CD31+ cell rates were 0.5% and 0.03%, respectively, but none of CD133+ cells were positive for CD45 and CD31.

Putative SCs were isolated particularly from TC samples; they grew as both free-floating spheres and adherent cells. The re-evaluation of markers in putative SC isolated from 2 cases of normal tissue showed that cells expressed higher levels of CD133, CD105 and CD44 with respect to those obtained after tissue digestion.

These preliminary findings suggest the presence of a higher fraction of CD133+ and CD105+ cells in TC samples compared to normal tissues. The higher expression of the stemness markers in TC may be related to the better survival of cells obtained from the TC samples in the SCM. Further investigation is required to draw any conclusion, but our approach seems to be promising.

### Keywords

Stemness markers, normal thyroid, TC.