Basic reporting

Raw data for comparative analysis of co-expression of proteins (CTR vs CT, CTR vs PTEN, CT vs OPN) are lacking.

Experimental design

The rationale of investigating CTR expression in C cell-derived tumors that express the natural ligand (calcitonin), inferring an autocrine type of regulation, should be better explained and discussed. For instance, are there any information in the literature suggesting normal C cells themselves are regulated by CT? If such a mechanism is unknown it should be told so in the Introduction.

Method of data presentation on CTR expression in Table 2 should be explained. As judged from the Materials and Methods section it appears to be based on a scoring method combining IHC staining intensity and extent of staining. The general validity of this calculation may also be valuable to readers.

Validity of findings

The discordance of overall IHC staining intensity and expression of CTR at the cellular level (cells seem either strongly positive or negative), as presented in Table 1, should be commented on. Does this mean that faint/moderate immunostaining depends on fewer positive cells rather a lower expression level in the majority of cells? Figs. 3B and E argue in favor of the latter, so are these images indeed representative?

Imaging the expression of both CTR and CT in serial sections from the same tumor specimens is warranted to support the statement in Results of a statistically significant correlation with $p<0.001$. In fact, the same criticism accounts for CTR vs PTEN and CRT vs OPN. Cases (inter- or intratumoral) with differential expression pattern (CTRhigh but no CT/PTEN/OPN, or the reverse), if present, would also be of interest to report.

General comments

It is suggested that in their discussion of tumor differentation/dedifferentiation authors also consider the possibility of transient epithelial-mesenchymal transition (EMT), in which "differentiation genes" are repressed in locally invasive tumor cells (and circulating cancer cells) but re-expressed at metastatic sites. That this accounts for human MTC was recently reported (Johansson et al, Development 142: 3519-28, 2015). It is thus expected that CT and possibly also CTR are down-regulated in MTC cells that undergo EMT, which may explain some of the present findings.

Identification of high CRT mRNA levels in MTC cell lines in this study opens for functional studies with siRNA to eludicate a possible autocrine pathway. This may be mentioned in the paper.

Distinction of CT/CTC and CGRP/CGRP-receptors may be of interest to clarify to readers.