

Peer Review File

Article information: <https://dx.doi.org/10.21037/cco-23-49>

Review Comments

I suggest to revise the English language. For example, the following sentence should be rephrased: “Aberrant expressions for abnormal cells exhibit two or more dots, while normal cells contain no to a single dot”

Answer: Thank you for the comment. We revised the manuscript by an English teacher. We also modified this part of the text because we understood that it was not clear for the readers:

“Translating the genetic information of “epigenetic imprinting biomarkers” detected by QCIGISH technology into practical words: **In normal somatic cells, maternal and paternal alleles of an imprinted gene are differentially methylated, thus one of allele is silenced and the other, activated. However, in the case of cancers, both alleles are expressed due to the activation of the imprinted gene. This is called “loss of imprinting” (18). So, the method discussed in the paper by Xu et al. analyses the non-coding intronic RNA aiming to visualize the transcription loci of the imprinted gene. For that, the QCIGISH method uses different colours to characterize the different structures of the nuclei: blue, red and brown. The different allelic expressions of the imprinted genes are quantified based on the transcription signals. Normal cells will show 1 or no colour, however, aberrant expressions will show more than one colour signal (19). Putting together, the principle of the QCIGISH methodology is to visualize, quantify and confirm pathologic allele expression of the investigated imprinted genes (10).”**

- I suggest to briefly discuss the main molecular markers, as TERT, BRAF, PAX8/PPAR γ , RAS, and RET/PTC, which can be considered in the management of patients with indeterminate FNA cytology, also citing recently published papers, such as 10.21037/gs.2017.11.07

Answer: we added more information about the main point mutations and gene fusions into the manuscript, and also cited the suggested paper. Please, find the changes highlighted in the text and here below:

The first molecular markers to gain prominence were point mutations such as BRAFV600E and RAS, and gene fusions like RET/PTC and PAX8/PPAR α , in addition to TERT mutations (6). It is known that in most thyroid cancers, mutations are mutually exclusive events, that is, only one of these mutations is found in each tumor (7). When these mutations are used as independent biomarkers, their sensitivity and specificity are too low to be clinically relevant, except for the presence of BRAF and TERT mutations as they are correlated with tumor malignancy (6,8). However, the combination of the analysis of these mutations in a panel has been shown to improve the sensitivity and specificity rates (6,8). Therefore, based on these data, the 7 genes panel was created, and it has inspired groups to create other types of panels that have begun to be commercialized.