Reviewer's report

Title: Cyclin A1 promoter hypermethylation in human papillomavirus-associated cervical cancer

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Reviewer: Carsten Müller-Tidow

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General

This manuscript provides evidence that the endogenous cyclin A1 promoter is hypermethylated in HPV associated cervical cancer. Methylation is inversely correlated with mRNA expression. In addition, the number of patients with cyclin A1 promoter methylation increases with higher malignancy of the tumor. The authors conclude that cyclin A1 methylation can serve as a valid marker for progression in cervical cancer.

Minor Essential Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

There are several points that need to be revised:

1) The authors claim that cyclin A1 due to its inactivation by methylation acts as a tumor suppressor. However, the authors only demonstrate a correlation which does not establish any causative relationship. These sections need to be clarified.

2) Figure 1: The authors refer to HeLa (K) and HeLa (S) cells that differ in cyclin A1 methylation. The authors need to describe the orifin of these cells and the potential reasons for varying levels of cyclin A1 methylation in more detail. It would be preferable to demonstrate that HeLa (K) and HeLa (S) cells indeed show a common genetic background.

3) Figure 2: It is unclear how many PCR cycles were performed for the MSP and the USP reactions.

4) Figure 2B needs more explanation. Was densitometry used to quantitate the PCR bands? Inter- and Intra assay variability need to be explained in more detail? Do the error bars represent standard error or standard deviation? How many replicates were performed for individual experiments and how may independent experiments were performed?

5) The method used for methylation analysis is at best semiquantitative. It would have been preferable to use real-time PCR. At least, the authors should remove the statements about correlation and regression equation (page 9, first paragraph)

6) Figure 3: The normal tissue surrounding the tumor also shows cyclin A1 methylation but retain expression. This would either mean that methylation detection is sensitive (even when only the minority of cells is affected) or that expression and methylation do not always correlate with each other inversely. The authors should comment on this.

7) Several typos and spelling errors occur throughout the manuscript.

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What next?: Accept after minor essential revisions

Level of interest: An article of importance in its field

Quality of written English: Needs some language corrections before being published

Statistical review: No

Declaration of competing interests:
I declare that I have no competing interests