Reviewer's report

Title: Decreased expression of key tumour suppressor microRNAs is associated with lymph node metastases in triple negative breast cancer.

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Reviewer: Stefano Caramuta

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In this study, the authors evaluated the miRNA expression profiling in a set of primary TNBC, LN metastases and normal breast tissues. They identified a set of 71 miRNAs deregulated in primary TNBC as compared to normal tissue and 27 miRNAs associated with LN metastasis. The manuscript is well conducted; results and discussion are appropriate and give sufficient details.

Here are some major comments and suggestions that the authors should address before the manuscript could be accepted for publication:

1. The authors use two different reference genes (RNU44 and RNU49), however only one was shown in the results. Did the authors get same results (in term of miRNA relative expression) with the two reference genes? Did the authors try to use both genes as normalizer (e.g. geometric mean)?

2. To verify microarray results from the first comparison (IDC vs NAT), the authors chose 6 miRNAs: how were these miRNAs selected among the 71 deregulated miRNAs? Moreover, data for miR-205 are not reported in Fig.1B.

3. Among the 71 deregulated miRNAs, the authors claim that 5 miRNAs (miR-130a, miR-1280, miR-590-5p, miR-1308, miR-17*) were not previously reported to be implicated in breast cancer and might be associated with TNBC phenotype. It would be worth to investigate a bit more on these miRNAs and maybe include them for PCR validation.

Did author check possible pathways or target genes for these miRNAs? Did the authors try to perform a clustering analysis to check if these 5 miRNAs could be sufficient to distinguish IDC from NAT?

4. When the authors compare LN- vs NAT and LN+ vs NAT they find different miRNA profiling but also a set of 10 miRNAs which were commonly deregulated in both groups. However, none of the 5 miRNAs (which authors claim to be potentially associated with TNBC: miR-130a, miR-1280, miR-590-5p, miR-1308, miR-17*) were deregulated in both LN- and LN+ tumors. If they were associated with TNBC phenotype they should be commonly deregulated in LN- and LN+ tumors. It would be worth to discuss this issue.

5. How did the authors choose the 9 miRNAs (among the 27) for PCR validation?

6. Since the author mentioned the possible involvement of Dicer in the overall
down-regulation of miRNAs observed in LN+ tumors, it would be interesting to check at least the mRNA expression levels of Dicer in the tumors used in this study.

**Level of interest:** An article of importance in its field

**Quality of written English:** Acceptable

**Statistical review:** No, the manuscript does not need to be seen by a statistician.

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