Reviewer's report

Title: Claudin 1 expression in basal-like breast cancer is related to patient age

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Reviewer: En-Hua Wang

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Reviewer's report: Review Report for Manuscript entitled "Claudin 1 expression in basal-like breast cancer is related to patient age." The authors showed claudin 1 was related to patient age in basal-like breast cancer, and knocking-down of claudin 1 resulted in a significant decrease in migration rate. Moreover, knocking-down of claudin 1 altered the expression of some genes associated with EMT. The authors have already demonstrated that there is a positive association between claudin 1 expression and ER-ve breast cancers in reference 19. ER-ve, one of the criteria for BLBC, is usually found in postmenopause women. Therefore, the conclusion “Claudin 1 expression in basal-like breast cancer is related to patient age” is lack of innovation. Though it has merit, it is still not enough and not suitable for publication as its current version.

Major comments

Comment 1: The authors observed the expression of claudin 1 in membrane and cytoplasm and referred that "claudin 1 mislocalization was shown to increase the invasiveness of the cancer cells" in Discussion. However, the BT-20 cell line which chosen to perform the subsequent research only express claudin 1 in membrane (Figure 3A). It is confusing that inhibition of claudin 1 membrane expression alone could weaken the invasiveness of breast cancers. I am concerned whether it is suitable for choosing BT-20 cell line as the subject. It is equally possible that high expression of claudin 1 in BLBC is mainly localized in cytoplasm, while the membrane expression of claudin 1 was decreased indeed.

Comment 2: The authors should explain why was the cytoplasmic expression of claudin 1 increased slightly in Cldn1 KD group (Figure 3B). It is confusing that the localization of claudin 1 was altered after treatment with siRNA.

Comment 3: The authors should perform Invasion assay to detect the impact of claudin 1 on the invasiveness of cancer cells.

Comment 4: The authors provided some data to show that claudin 1 may be involved in regulating EMT and each discussion presented is intriguing, but overall the work falls short of demonstrating the mechanism by which claudin 1 regulates EMT.

Minor comments

Comment 1: The authors referred 151 breast cancer samples including 79 cases of BLBC and 72 cases of “non-basal” breast cancers in Methods, but only 144 cases (79 cases of BLBC and 65 cases of “non-basal” breast cancers) were analyzed for claudin 1 and 135 cases (73 cases of BLBC and 62 cases of
“non-basal” breast cancers) were analyzed for claudin 4 in Table 1. The authors should explain the inconsistency in these positions.

Comment 2: The authors referred 79 cases of invasive breast cancers were categorized as BLBC, but only 73 cases were analyzed for the correlation between claudin 1 and Node status, 76 cases were analyzed for the correlation between claudin 1 and tumor size in Table 2. For claudin 4, only 67 cases were analyzed with the Node status, 73 cases were analyzed with the patient age and tumor grade, and 71 cases were analyzed with the tumor size in Table 2. The authors should explain the inconsistency in these positions.

Comment 3:
Scale should be added in Figure 2 and 3C.

**Level of interest:** An article of limited interest

**Quality of written English:** Acceptable

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

**Declaration of competing interests:**

declare that I have no competing interests.