

#### **SDI Review Form 1.6**

Journal Name:	Journal of Pharmaceutical Research International
Manuscript Number:	Ms_JPRI_36743
Title of the Manuscript:	The expression patterns of APC2 and APC7 in newly diagnosed acute lymphoblastic leukemia
Type of the Article	

#### General guideline for Peer Review process:

This journal's peer review policy states that <u>NO</u> manuscript should be rejected only on the basis of '<u>lack of Novelty'</u>, provided the manuscript is scientifically robust and technically sound. To know the complete guideline for Peer Review process, reviewers are requested to visit this link:

(http://www.sciencedomain.org/page.php?id=sdi-general-editorial-policy#Peer-Review-Guideline)



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## PART 1: Review Comments

	Reviewer's comment	Author's comment (if agre highlight that part in the man his/her feedback here)
<u>Compulsory</u> REVISION comments	Major concern about the calculation of the fold induction of gene expression. The authors did not use the $2^{-\Delta\Delta Ct}$ method, instead they use the following formula: " $\Delta CT$ was calculated from CT, target genes-CT, ABL formula and $2^{-\Delta Ct, case}/2^{-\Delta Ct, control}$ was considered as gene expression fold changes" to calculate gene fold induction. The deduction of this formula was not described or mathematically proven by the authors or at least a reference should be added. The results should be re-calculated using the $2^{-\Delta\Delta Ct}$ method, where $\Delta Ct$ refers to the normalization of all genes and samples studied to the housekeeping gene and $\Delta\Delta Ct$ is the difference between $\Delta Ct$ values of ALL patients vs $\Delta Ct$ values of control samples. Statistical analysis – The number of independent experiments was not disclosed in the material and methods or results sections, or even in the figure legends. The data presented should be the result of at least three independent experiments to have any statistical	
	value/significance. The P values should be calculated from the averages of each independent experiment (for instance if 3 independent experiments were done, N=3) and the number of replicas should not be included in the calculation of P values. Figure legends are extremely incomplete, they only contain the figure title and lack	
	methodological detail, statistical information and explanation of data analysis. Ethical issue: they have been addressed by the authors. The work involves the collection of clinical samples, these were done with patients consent according to the Helsinki's Declaration.	
Minor REVISION comments	English grammar errors throughout the manuscript that need to be corrected. Table 2 – some of the values between brackets are missing the % symbol	
Optional/General comments	Standard deviations are quite significant particularly in the control samples which is a concern. Overall I feel that there are substantial issues with data analysis of the RT-qPCR data and I am unclear of the number of independent experiments done, I also think that the manuscript does not have enough data to be published. For this reason I would like to suggest additional experiments that would strengthen the manuscript: Knocking down or knockout the APC2 and APC7 genes in ALL cell lines should bring some more information to the role of these proteins in ALL, for instance by comparing cell proliferation/ apoptosis of KD versus WT cell lines. The manuscript would also greatly benefit from protein analysis – protein extraction followed by western blotting of clinical samples – to support the gene expression results.	

## **Reviewer Details:**

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