

January 28, 2003

Dr. Herbert Tabor, Editor, The Journal of Biological Chemistry, 9650 Rockville Pike, Bethesda, MD 20814-3997, U.S.A.

RE: JBC 275 (23):17223-17236 (2000)

Dear Dr. Tabor,

Sincerely yours,

This is to follow up on my mail of December 9 regarding the comments to our paper (JBC 275 (23):17223-17236, 2000). For your reference, I enclose copies of previous e-mails between us. As I reported in my previous mail, an ad hoc independent investigative panel was set up to examine the issue raised by an anonymous reader. The panel concluded that the duplication of the lane pointed by the reader did exist, but that this was caused by a mistake in making a composite figure by cut-andpaste processing of the original autoradiogram, rather than an intentional "fabrication". I understand that a copy of their report including the evidence has been sent to you. I enclose a copy, again, for your reference. According to their recommendation, I have asked two members of the laboratory who were not originally involved in the paper to reproduce the main results of the above manuscript, that is, those obtained by the pulldown experiments shown in Fig. 2 and 3. During this process, we found an incorrect description about the construction of an expression vector for ECT-N1 used in those experiments (please see an attached manuscript for correction). The two people performed experiments independently, and obtained essentially identical results, which correctly reproduced the results reported in the above paper (please see Fig. X and Y, which correspond to Fig. 2B and Fig. 3C of the above paper, respectively). Based on these findings, we have concluded that there has not been a systematic manipulation of experimental data, as was the original concern of the anonymous reader. As we have concluded that the results and conclusion of the above paper essentially holds true, we would therefore like to respectfully submit a correction of our paper regarding the pointed error in Fig. 4 and that in the primer sequence used in our vector construction, as attached. I sincerely apologize for the inconvenience we have caused and are truly thankful to the anonymous reader for enabling us to correct these errors.

(Influence
Shuh Narumiya
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I have read the above letter and concur in submission of the attached correction.

Kazuhiro Kimura Date Takahiro Tsuji /57/03

Takahiro Tsuji Date

Juha Jahoda /27/03

Yuka Takada Date Toru Miki Date

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Kazuhiro Kimura	Date		Takahiro Tsuji	Date
		.*	Combola.	1/27/03
Yuka Takada	Date		Toru Miki	Date

Enclosure: a set of copies of e-mail communication, a copy of the panel report and a CD containing files for correction text and a figure.

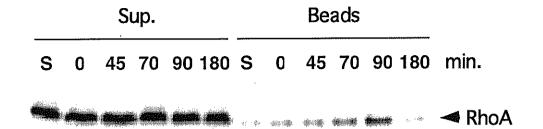


Figure X. Pull-down of GTP-Rho from lysates of HeLa cells collected at various phases of cell cycle. HeLa cells were arrested at S phase and M phase with thymidine and nocodazole. The cells arrested in the M phase were then released from the nocodazole arrest by wash, collected at 0, 45, 70, 90 and 180 min after the wash, nad subjected to the pull-down assay (conducted by S. Y.).

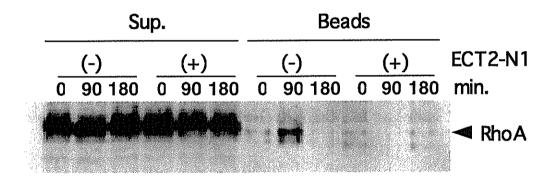


Figure Y. Effects of ECT2-N1 expression on GTP-Rho accumulation at 90 min after the nocodazole wash (conducted by S. Y.).

Corrections

Vol. 275 (2000) 17233-17236

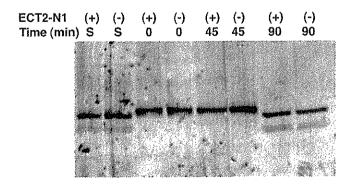
Accumulation of GTP-bound RhoA during cytokinesis and a critical role of ECT2 in this accumulation.

Kazuhiro Kimura, Takahiro Tsuji, Yuka Takada, Toru Miki, and Shuh Narumiya

Page 17233: The reverse primer sequence used in PCR for ECT-N1 was described incorrectly. The correct sequence and construction are as follows.

ECT2-N1 and ECT2-N2 were constructed as follows. The region encoding ECT2-N1(aa 5'-1-334) amplified polymerase chain reaction using was by CGGGATCCATGGCTGAAAATAGTGTA-3' forward primer and 5'as CGGAATTCCTATGATTTCTTGAGCTCAGG-3' reverse as a primer. After amplification, this cDNA fragment was digested with BamHI and EcoRI and subcloned into pBluescript SK(+). After sequencing, this clone was digested with BamHI and EcoRI and cloned into pCEV32F.

Page 17236, Fig. 4: One lane was incorrectly placed during making the composite figure. The original and correct autoradiogram is shown below. This change does not affect the conclusion of this experiment.



We regret that such an error was introduced during figure preparation, and thank an anonymous reader for pinpointing this issue.