Author’s response to reviews

Title: The induction of activating transcription factor 3 (ATF3) contributes to anti-cancer activity of Abeliophyllum distichum Nakai in human colorectal cancer cells

Authors:

Gwang Hun Park (enter0230@hanmail.net)
Jae Ho Park (parkjh@jwu.ac.kr)
Hyun Ji Eo (ehj56@naver.com)
Hun Min Song (ou1@dreamwiz.com)
So Hee Woo (sososohee89@naver.com)
Mi Kyoung Kim (rlaalrud888@naver.com)
Jin Wook Lee (ghopur2@naver.com)
Man Hyo Lee (manhyo@hanmail.net)
Jeong Rak Lee (ljr21@nate.com)
Jin Suk Koo (kimkoo1114@andong.ac.kr)
Jin Boo Jeong (jjib0403@anu.ac.kr)

Version: 2
Date: 1 May 2014

Author’s response to reviews:

Dear Editors:

Please consider for publication in your esteemed journal our manuscript “The induction of activating transcription factor 3 (ATF3) contributes to anti-cancer activity of Abeliophyllum distichum Nakai in human colorectal cancer cells”

None of the material presented here has been published or under consideration elsewhere.

Main Point: Recently, Abeliophyllum distichum Nakai (A. distichum) has been reported to exert the inhibitory effect on angiotensin converting enzyme. However, no specific pharmacological effects from A. distichum have been described. We performed in vitro study to evaluate anti-cancer properties of A. distichum and then elucidate the potential mechanisms. Cell viability was measured by MTT assay. ATF3 expression level was evaluated by Western blot or RT-PCR and ATF3 transcriptional activity was determined using a dual-luciferase assay kit after the transfection of ATF3 promoter constructs. In addition, ATF3-dependent apoptosis was evaluated by Western blot after ATF3 knockdown using ATF3 siRNA. Exposure of ethyl acetate fraction from the parts of A. distichum including flower, leaf and branch to human colorectal cancer cells, breast cancer cells and hepatocellular carcinoma reduced the cell viability. The branch extracts from A. distichum (EAFAD-B) increased the expression of activating transcription factor 3 (ATF3) and promoter activity, indicating transcriptional activation of ATF3 gene by EAFAD-B. In addition, our data
showed that EAFAD-B-responsible sites might be between -147 and -85 region of the ATF3 promoter. EAFAD-B-induced ATF3 promoter activity was significantly decreased when the CREB site was deleted. However, the deletion of Ftz sites did not affect ATF3 promoter activity by EAFAD-B. We also observed that inhibition of p38MAPK and GSK3β attenuated EAFAD-B-mediated ATF3 promoter activation. Also, EAFAD-B contributes at least in part to increase of ATF3 accumulation. These findings suggest that the anti-cancer activity of EAFAD-B may be a result of ATF3 promoter activation and subsequent increase of ATF3 expression. We hope that the manuscript is acceptable. All authors listed in this manuscript have contributed to the work and agreed to submit the manuscript to BMC Complementary and Alternative Medicine. No part of the work has been published before. Thank you for your time and consideration. We look forward to a positive response from you.

Sincerely yours,