P181

Induction of Apoptosis by Ethyl Acetate Extracts of *Hizikia fusiforme* in Human Leukemic U937 Cells is Mediated with Modulation of Bcl-2 family, Activation of Caspases and Inactivation of AKT and JNK

Tae Yeon Kim[1], Hye-Rin Lee[1], Min-Jeong Seo[2], Hak-Seob Lim[2], Woo-Hong Joo[3], Kang Byoung Won[5], Young-Hyun Choi[4] and Yong-Kee Jeong[1,2]*

[1]*Department of Biotechnology, Dong-A University, Busan 604-714, Korea*
[2]*Bioinstitute, MILLENNIUM PROMISE CO., LTD., Gijang-gun, Busan 619-962, Korea*
[3]*Department of Biology, Changwon National University, Changwon City, 641-773 Korea*
[4]*Callegy of Oriental Medicine, Dong-Eui University, Busan, 614-052*
[5]*BK21 Center for Silver Bioindustrialization, Dong-A, University, Busan, Korea*

Brown algae, *Hizikia fusiforme* extracts have recently been found to have antioxidant and anti-tumor activities. In this study, we examined the antitumor effect of ethyl acetate extracts of *Hizikia fusiform* (EHF) on human leukemic U937 cells. MTT assays showed that the EHF significantly inhibited the cell viability and proliferation of U937 cells in a time- and dose dependent manner. Specifically, these EHF induced apoptosis of U937 cells as evaluated by detection of hypodiploid cells using flow cytometry and observation of condensed and fragmented nuclei in EHF-treated cells. Treatment of EHF in human leukemic U937 cells decreased the levels of antiapoptotic proteins including IAP family proteins (cIAP and XIAP) and Bcl-2 members (Bcl-2 and Bcl-XL), and enhanced the levels of proapoptotic proteins such as tBid and Bax. EHF induced the proteolytic activation of caspase 3, caspase 8, caspase 9, PARP (poly (ADP-ribose) polymerase) and PLC-γ1. EHF treatment resulted in down-regulation of phospho-AKT and phospho-JNK expressions. Pretreatment with LY294002, a specific inhibitor of Akt, and SP600125, a specific inhibitor of JNK, significantly increased EHF induced apoptosis. Taken together, these findings suggest that ethyl acetate extracts of *Hizikia fusiform* (EHF)-induced inhibition of human cancer cell proliferation is associated with the induction of apoptotic cell death via modulations of Bcl-2 members and IAP family proteins, activation of caspases, inhibition of AKT and JNK activation. And ethyl acetate extracts of *Hizikia fusiform* (EHF) may have therapeutic potential in human cancer.

Key words: *Hizikia fusiform*, apoptosis, U937, AKT, JNK

P182

20S Proteasome Alpha 5 Subunit, *OgPAE1*, Derived from Wild Rice (*Oryza grandiglumis*) Conferred *Botrytis cinerea* Resistance

Eun-Hee Jeon, Jung-Hun Pak, Mi-Jin Kim, Hye-Jeong Kim, Ki-Jung Lee, Young-Soo Chung* and Kyung-Ho Kang[1]

Dept. of Genetic Engineering, Dong-A University, Busan, Korea
[1]*National Crop Experiment Station, Suwon, Korea*

Oryza grandiglumis(CCDD, 2n=48), one of the wild rice species, has been known to possess fungal resistance against sheath blight, rice blast, bacterial leaf blight and insect resistance against brown plant hopper(*Nilaparvata lugens*). *OgPAE1* contains 714bp of nucleotides and 237 amino acids. The highest homology was observed with *OsPAE1* mRNA for alpha 5 subunit of 20S proteasome from *Oryza sativa*(japonica cultivar-group). The 20S proteasome is the proteolytic complex that is involved in removing abnormal proteins and other various biological functions. The *OgPAE1* gene was induced by wounding, yeast extract, JA and SA, protein phosphatase inhibitors CN as well as EN. To identify *in vivo* function of gene *OgPAE1*, the gene was transformed into *Arabidopsis thaliana* and high concentration of *Botrytis cinerea* (5×10^5/mL) was inoculated on the *OgPAE1* transgenic plants. The growth of *Botrytis cinerea* was suppressed in *OgPAE1* transgenic plants but most of control plants destroyed completely. Average necrosis sizes of transgenic plants were 3.8 through 6.4 mm^2, whereas that of control was 11.2 mm^2. Our results suggest that *OgPAE1* may contribute to partial resistance against fungal diseases, even though clear in vivo function of the gene has not been reported yet from any plant species.

Key words: *Oryza grandiglumis*, 20S proteasome, *Botrytis cinerea*