

Five compounds were investigated for their protective activity against glutamate-induced neurotoxicity in HT22 cells. The relative neuroprotection of compounds is exhibited in Figure 3. Among them, epicatechin (2), nudiposide (3), lyoniside (4), and scopoletin (5) showed significant neuroprotective activities at concentrations ranging from 10, 50, and 100 μM in the dose-dependent manner. Four compounds increased HT22 cell density reduced by glutamate [Figure 4]. Neuroprotective activities of epicatechin, nudiposide, and lyoniside, scopoletin were firstly reported in this manuscript.

Scopoletin (5) (74.10% at 50 μM and 75.70% at 100 μM ; $P < 0.05$) showed the most potent activity against glutamate-induced neurotoxicity. The potency of scopoletin is similar with that of trolox, a positive control. It was reported that scopoletin increased rat retinal neuron cells at the high concentration and also showed anti-cholinesterase activity.^[30,31]

Therefore, scopoletin may be a good candidate of drug development for treatment of AD.

CONCLUSION

We isolated five compounds from *T. amurensis* and evaluated the neuroprotective effect of five compounds on glutamate-induced oxidative stress in HT22 cells in this study. Epicatechin, nudiposide, lyoniside and scopoletin significantly protected neuronal cells, and these results suggest that epicatechin, nudiposide, lyoniside and scopoletin are correlated with neuroprotective effect of *T. amurensis*.

Further study is required to understand the bio mechanism of neuroprotective effect of four compounds.

ACKNOWLEDGMENT

This study is supported by Kangwon National University.

REFERENCES

1. Swerdlow RH. Brain aging, Alzheimer's disease, and mitochondria. *Biochim Biophys Acta* 1978;23:229-33.
2. Swerdlow RH. Pathogenesis of Alzheimer's disease. *Clin Interv Aging* 2007;2:347-59.
3. Heo SJ, Cha SH, Kim KN, Lee SH, Ahn G, Kang DH, et al. Neuroprotective effect of phlorotannin isolated from *Ishige okamurae* against H_2O_2 -induced oxidative stress in murine hippocampal neuronal cells, HT22. *Appl Biochem Biotechnol* 2012;166:1520-32.
4. Butterfield DA, Howard B, Yatin S, Koppal T, Drake J, Hensley K, et al. Elevated oxidative stress in models of normal brain aging and Alzheimer's disease. *Life Sci* 1999;65:1883-92.
5. Abeti R, Duchon MR. Activation of PARP by oxidative stress

- induced by β -amyloid: Implications for Alzheimer's disease. *Neurochem Res* 2012;37:2589-96.
6. Ames BN, Shigenaga MK, Hagen TM. Oxidants, antioxidants, and the degenerative diseases of aging. *Proc Natl Acad Sci U S A* 1993;90:7915-22.
7. Coyle JT, Puttfarcken P. Oxidative stress, glutamate, and neurodegenerative disorders. *Science* 1993;262:689-95.
8. Halliwell B. Reactive oxygen species and the central nervous system. *J Neurochem* 1992;59:1609-23.
9. Tan S, Sagara Y, Liu Y, Maher P, Schubert D. The regulation of reactive oxygen species production during programmed cell death. *J Cell Biol* 1998;141:1423-32.
10. Satoh T, Lipton SA. Redox regulation of neuronal survival mediated by electrophilic compounds. *Trends Neurosci* 2007;30:37-45.
11. Satoh T, Enokido Y, Kubo T, Yamada M, Hatanaka H. Oxygen toxicity induces apoptosis in neuronal cells. *Cell Mol Neurobiol* 1998;18:649-66.
12. Aruoma OI. Free radicals, oxidative stress and anti-oxidants in human health and disease. *J Am Oil Chem Soc* 1998;75:199-212.
13. Kornhuber J, Wiltfang J. The role of glutamate in dementia. *J Neural Transm* 1998;53:277-87.
14. Tan S, Wood M, Maher P. Oxidative stress induces a form of programmed cell death with characteristics of both apoptosis and necrosis in neuronal cells. *J Neurochem* 1998;71:95-105.
15. Tan S, Schubert D, Maher P. Oxytosis: A novel form of programmed cell death. *Curr Top Med Chem* 2001;1:497-506.
16. Fukui M, Song JH, Choi J, Choi HJ, Zhu BT. Mechanism of glutamate-induced neurotoxicity in HT22 mouse hippocampal cells. *Eur J Pharmacol* 2009;617:1-11.
17. Braun S, Liebetrau W, Berning B, Behl C. Dexamethasone-enhanced sensitivity of mouse hippocampal HT22 cells for oxidative stress is associated with the suppression of nuclear factor-kappaB. *Neurosci Lett* 2000;295:101-4.
18. Liu J, Li L, Suo WZ. HT22 hippocampal neuronal cell line possesses functional cholinergic properties. *Life Sci* 2009;84:267-71.
19. Cho N, Choi JH, Yang H, Jeong EJ, Lee KY, Kim YC, et al. Neuroprotective and anti-inflammatory effects of flavonoids isolated from *Rhus verniciflua* in neuronal HT22 and microglial BV2 cell lines. *Food Chem Toxicol* 2012;50:1940-5.
20. Fukui M, Zhu BT. Mitochondrial superoxide dismutase SOD2, but not cytosolic SOD1, plays a critical role in protection against glutamate-induced oxidative stress and cell death in HT22. *Free Radic Biol* 2012;48:821-30.
21. Kim KH, Moon E, Kim SY, Choi SU, Lee KR. Lignan constituents of *Tilia amurensis* and their biological evaluation on antitumor and anti-inflammatory activities. *Food Chem Toxicol* 2012;50:3680-6.
22. Choi JY, Seo CS, Zheng MS, Lee CS, Son JK. Topoisomerase I and II inhibitory constituents from the bark of *Tilia amurensis*. *Arch Pharm Res* 2008;31:1413-8.
23. Lee B, Weon JB, Yun BR, Lee J, Eom MR, Ma CJ. Simultaneous determination of four neuroprotective compounds of *Tilia amurensis* by high performance liquid chromatography coupled with diode array detector. *Pharmacogn Mag* 2014;10:195-9.
24. Weon JB, Lee B, Yun BR, Lee J, Ma CJ. Neuroprotective effects of 4,5-dimethoxyprocatechol isolated from *Cynanchum paniculatum* on HT22 cells. *Pharmacogn Mag* 2014;10:161-4.
25. Mehta BK, Sharma U, Agrawal S, Pandit V, Joshi N, Gupta M. Isolation and characterization of new compounds from seeds of *Nigella sativa*. *Med Chem Res* 2008;17:462-73.
26. Sudjaroen Y, Haubner R, Würtele G, Hull WE, Erben G,

- Spiegelhalder B, *et al.* Isolation and structure elucidation of phenolic antioxidants from Tamarind (*Tamarindus indica* L.) seeds and pericarp. *Food Chem Toxicol* 2005;43:1673-82.
27. Fuchino H, Satoh T, Tanaka N. Chemical evaluation of *Betula* species in Japan. I. Constituents of *Betula ermanii*. *Chem Pharm Bull* 1995;43:1937-42.
28. Lee MK, Sung SH, Lee HS, Cho JH, Kim YC. Lignan and neolignan glycosides from *Ulmus davidiana* var. japonica. *Arch Pharm Res* 2001;24:198-201.
29. Kim SS, Lee CK, Kang SS, Jung HA, Choi JS. Chlorogenic acid, an antioxidant principle from the aerial parts of artemisia iwayomogi that acts on 1,1-diphenyl-2-picrylhydrazyl radical. *Arch Pharm Res* 1997;20:148-54.
30. Orhan I, Tosun F, Sener B. Coumarin, anthroquinone and stilbene derivatives with anti-cholinesterase activity. *J Biosci* 2008;63:366-70.
31. Zhang Y, Sheng YM, Meng XL, Long Y. Effect of caffeic acid, seopletin and scutellarin on rat retinal neurons *in vitro*. *Zhongguo Zhong Yao Za Zhi* 2005;30:907-9.

Cite this article as: Lee B, Weon JB, Eom MR, Jung YS, Ma CJ. Neuroprotective compounds of *Tilia amurensis*. *Phcog Mag* 2015;11:303-7.

Source of Support: Nil, **Conflict of Interest:** None declared.