Korean J. Ginseng Sci.
Vol. 16, No. 1, 69~71(1992)

The Effects of Ginseng Components on Arachidonic Acid Metabolism

Chan-Woong Park
Department of Pharmacology, College of Medicine, Seoul National University, Seoul 110-460, Korea

A considerable amount of evidence has been accumulated which showed the beneficial effects of Korean Ginseng based on more scientific concepts and methodlogy. However, such multifarious pharmacological actions make it possible to suggest some common factors through which the effects of Ginseng might be mediated. Prostaglandins and oxygen free radical could be considered to be such common factors.

In this communication, I would like to present actions of two kinds of components of Korean Ginseng: one is a group of saponins which was found to have effects on prostaglandin synthesis and the other is maltol which was shown to be antioxidant and to protect the tissue damage induced by oxygen free radical.

The production of individual prostaglandins from the exogenous [^{3}H]AA by various enzyme sources such as RKM, BAM and HPH was determined. PGE_2 and $PGF_{2\alpha}$ production were highest by RKM compared with BAM or HPH. But TxB_2 production was highest by HPH and 6-keto-$PGF_{1\alpha}$ production was highest by BAM.

From this result different tissues were chosen to evaluate the production of PGs such as RKM for PGF_2 and $PGF_{2\alpha}$ and BAM for P.C. HPH for TxB_2. The amounts of total cyclooxygenase products produced by various enzyme sources did not show any significant changes in the presence of ginseng saponins. But each divergent prostaglandin productions was influencedd by ginseng saponins.

Both panaxadiol or panaxatriol increased the 6-keto-$PGF_{1\alpha}$ production and suppressed the $PFG_{2\alpha}$ and TxB_2 production but PGE_2 production was not influenced significantly. Ginsenoside Rb_2 also increased the production of 6k-$PGF_{1\alpha}$ and decreased the production of TxB_2 dose dependently but the productions of PGE_2 and $PGF_{1\alpha}$ were not significantly influenced by ginsenoside Rb_2 in concentration of 10 to 1000 μg/m*l*. This results are consistent with the results of panaxadiol & panaxatriol.

In the presence of ginsenoside Rc, the production of 6k-$PGF_{1\alpha}$ was increased but that of PGE_2 and PGF_2 were decreased dose-dependently. The results of PGE_2 is somewhat different from the result of panaxadiol, panaxatriol, & G-Rb_2.

However, the stimulation of P.C. production & the inhibition of TxB_2 production were consistent.

The effect of ginsenoside Re on divergent prostaglandin production was quite similar to that of ginsenoside Rb_2, the production of 6k-$PGF_{1\alpha}$ was increased and that of TxB_2 was decreased dose-dependently but the productions of PGE_2 and $PGF_{2\alpha}$ were not affected.

To determine effect of ginseng saponin on cyclooxygenase, the effect of indomethacin, C.O. inhibitor, or epinephrine, C.O. stimulator was examined using R.K.M. Comparing with the effect of indomethacin or epinephrine, the stimulatory or inhibitory effect of ginseng saponins on the production of PGE_2 was not significant. But a significant inhibitory effect on the $PGF_{2\alpha}$ was observed with the treatment of panaxadiol, panaxatriol, & G-Rb_2. From this result we could conclude the inhibitory effect of PGF_2 production was partly, if not mainly, due to the inhibition of C.O.

Further investigation on the mechanism of the inhibition of Tx synthesis was performed using imidazole, inhibitor of the Tx synthetase. The effect of ginseng saponins in a concentration of 500 μg/m*l* on the formation of TxB_2 was compared with that of imidazole in using HPH as enzyme source. In

general, ginseng saponins suppressed the production of TxB_2 and the potency of ginsenoside Rb in a concentration of 500 μg/ml was equivalent to that of imidazole in a concentrtions of 2 mM.

And ginseng saponins seems to potentiate the effect of imidazole additively. When HPH was treated with ginsenosides and imidazole together, the inhibitory effect on TxB_2 production was potentiated. Therefore, the inhibitory effect of ginseng saponins on the TxB_2 production may be mainly due to the inhibiton of the Tx synthetase.

The production on 6k-$PGF_{1\alpha}$ in the BAM was inhibitory by tranylcypomine dose dependently and the inhibitory effect of tranylcypromine on production of 6k-$PGF_{1\alpha}$ was nearly completely reversed by ginsenosides. This results suggest that ginsenoside stimulate the prostacyclin synthetase.

Sodium arachidonate induced platelet aggretation was significantly inhibited by all the ginseng saponins tested except ginsenoside Rh_1 but ADP induced platelet aggregation was not affected. While indomethacin inhibited the both of sodium arachidonate or ADP induced platelet aggregation, imidazole only inhibited sodium arachidonate induced aggregation.

In the mean time, around 10 kinds of phenolic acids are reported to be contained in Ginseng, among them, maltol and salicylic acid which have antioxidant effect, and trans-p-coumaric acid which is not an antioxidant, were subjected to study the prostaglandin synthesis.

Total prostaglandin formation by various enzyme sources such as RKM, BAM, HPH were not affected by maltol and salicylic acid but significantly inhibited by p-coumaric acid. p-coumaric acid also inhibit the formation of $PGF_{2\alpha}$, TxB_2, and 6k-$PGF_{1\alpha}$ in HPH reaction system.

Platelet aggregation induced by U46619 was significantly inhibited by p-coumaric acid but the effects of maltol and salicylic acid on platelet aggregation were not different from that of control.

In conclusion, these findings suggest that both of Ginseng saponins and phenolic acids seem to play a role in the regulation of the arachidonic acid metabolism and phenolic acids may act on cycloxygenase directly while Ginseng saponins probably affect the divergent synthetic pathway of prostaglandins from endoperoxide.

Second part will be effect of maltol on oxygen free radical induced tissue damage. Maltol is one of the component of ginseng which is isolated by Prof. Han and reported to be antioxidant by observing its inhibitory effect on the lipid peroxidation in mice induced by ethanol intoxication and quenching activity for DPPH radicals.

Therefore we would like to confirm the antioxidant effect of maltol the topic to be discussed here.

1. Confirmation of its antioxidant action
2. Its potency as an antioxidant
3. Its protective action against oxidation damages at cellular and organ level as well as *in vivo*
4. Mechanism of its antioxidant action

To confirm its antioxidant action *in vitro*.

We used two kinds of reaction system to induce lipid peroxidation Fe-Ascorate system and Cu-H_2O_2 system as non-enzymatic reaction and Fe-/ADP-NADPH system and paraquat-NADPH system as enzymatic reaction. Lipid peroxidation was determined by measuring two products MDA and hydrocarbons.

Maltol inhibits the lipid peroxidation of hepatic microsomes induced by various reaction systems, MDA level and ethane produced by Fe/ascorbate reaction system. The antioxidant potency of maltol was about 1/50 compared with p-phenylemediamine, butylated hydroxyaminol, and butylated hydroxytoluene. The antioxidant potency of maltol in various reaction system was compared with p-PDA, BHA or BHT.

The reciprocal of IC_{50} of matol was taken as 1. Other known antioxidant show roughly around 50.

The action of maltol was tested at two kinds of cellular level.

1. Paraquat induced toxicity in isolated hepacyte
2. Adriamycin induced toxicity in isolated cardiomyocyte

Paraquat is reduced first at microsome by NADPH dependent reductase to paraquat radical and

this paraquat radical react with oxygen to produce superoxide radical and this radical is responsible for cellular damage. Paraquat itself did not show any effect on heptocyte. But when treated with BCNU, which is a glutathion reductase inhibitor, Paraquat induce cell death dose-dependently.

Here we observed cell death as toxic effect.

Maltol inhibit the paraquat-induced cell death and LDH leakage as well as MDA production. The effect of 2 mM maltol was comparable with that of 75 μM tocopherol.

To confirm that maltol can be effective on oxygen radical mediated cytotoxicity, effect of maltol was compared in paraquat induced cell toxicity and MPTP induced toxicoty. The paraquat produces hydrogen peroxide but MPTP did not produce H_2O_2. Maltol or tocopherol did not protect the cell death induced by MPTP. This drug, MPTP did not produce oxygen radical. This means that maltol shows protective effect probably because it reacts with oxygen radical.

Adriamycin treated cardiomyocyte produce the MPA. Adriamycin is also known as a redox cycling compound. And maltol inhibit the adriamycin induced MPA production dose-dependently.

The protective effect at organ level was tested using oxygen paradox, also called reperfusion damage in Langendorff preparation. Isolated heart was initially perfused with normoxic K-H solution and then perfused with hypoxic cardioplegic solution for 90 min. After that heart was perfused with oxygenated cardioplegic solution for 20 min. During reoxygenated perfusion we determined the level of CPK, LDH and MDA from perfusate. And these level of CPK, LDH and MDA were reduced by treatment with various known radical scavengers SOD, Catalase, DMSO.

Thus we can confirm that the oxygen paradox in ischemic heart was mediated by oxygen radical.

Maltol significantly inhibited the MDA production and this effect was compared with of BHT and tocopherol.

LDH leakage also protected by maltol.

The protective effect of maltol against oxygen radial *in vivo* was tested. The ethane from expired air of animal was concentrated with trapping device. Ethan was determined with gas chromatography.

We already confirmed that maltol did not react with superoxide anion and H_2O_2. Then we tested on singlet oxygen. When Rose bengal reacted with NADPH under UV irradiation, NADPH was oxidized by singlet oxygen. This was confirmed by inhibitory effect of DABCO, singlet oxygen scavenger. In contrast maltol did not show any effect, meaning that maltol did not react with singlet oxygen.

Fe and H_2O_2 produces hydroxyl radical which produces ethylene in the presence of methional. The maltol inhibited the ethylene production dose dependently suggesting maltol may interact with hydroxyl radical. H_2O_2 degraded in the presence of Fe and this H_2O_2 degradation was inhibited by EDTA or DETAPAC but not inhibited by maltol.

And other hydroxyl radical scavengers also showed similar effect to maltol in this reaction system. This reslt suggest that maltol, although it has metal chelating action, its antioxidant action was not due to its chelating action but due to hydroxyl radical scavenging action. This was further supported by absorption spectrum of maltol, its peak at 270 nm and this peak completly disappeared by treatment of hydroxyl radical. This result suggested that maltol seems to interact hydroxyl radical and changed into a different compound.

Conclusion

1. Although its potency was low compared to that of other known antioxidant, maltol has an antioxidant effect strong enough to suppress oxidation-damaging processes in biological system.

2. The removal of hydroxyl radical may be a possible mechanism of its antioxidant action.