RECENT ADVANCES OF KOREAN RED GINSENG STUDIES IN JAPAN

Akira Kumagai

Toyama Medical and Pharmaceuticl University

Nearly 2000 years have passed since Korean Red Ginseng (KRG) was described as medical plant in the first time in China.

The evidence that KRG has been used as medicinal purposes for such a surprisingly long period, is most likely ascribable to its efficacy and harmlessness.

By the modern scientific approaches which in recent years rapidly developed, biochemical and pharmacological analysis of KRG has been made possible. The structures of ginseng saponins, major components of KRG, have been intensively investigated and determined by Shibata and his co - workers. Then pharmacological efficacy of each ginseng saponins has now been clarified. This means that this ancient Chinese medical plant shrouded in mystery has been now unveiled.

Eight years ago, "The Medical Society for Red Ginseng Research" was founded and since then every years, congress for the medical and pharmacological study related to KRG is regularly held in Kobe, Japan and also annual reports about KRG(Ginseng Review) are published.

[I]

1. Anti - atherogenic and anti - thrombotic action of Korean Red Ginseng

KRG has been said to have anti - atherogenic and anti - thrombotic action and has been intensively investigated until now.

The mechanism of anti - thrombotic action of KRG has been investigated in detail by Tamura and his group (Chiba University Medical School).

Increased TXA_2 formation and hyper - aggregability of platelet is said to be closely related to thrombogenesis. TXA_2 is well known to have a potent platelet aggregative action and play a very important role in collagen or arachidonic acid(AA) - stimulated platelet aggregation. Actually platelet aggregation induced by these aggregants is reported to be abolished by inhibition of TXA_2 biosynthesis.

4 weeks administrations of KRG significantly decreased collagen - induced platelet aggregation and TXB_2 formation in patients with various cardiovascular thrombotic disorders. Interesting finding is that when U - 46619 (a stable analogue of TXA_2) stimulated platelets obtained from these patients, both platelet aggregation and TXB_2 formation were suppressed. In *in vitro* study, both Ginsenoside - Rg_1(Rg_1) or Ginsenoside - Rg_3(Rg_3) were found to have a potent anti - aggregative action when platelets were stimulated by collagen, AA or U - 46619. On the other hand, Rg_1 has no antagonistic action for TXA_2 - receptor. Interesting finding is that an increase of cytosalic Ca^{++} in platelets stimulated by U - 46619 was dose - dependently decreased by in vitro addition of Rg_1 with concomitant decrease of platelet aggregation. This result indicate that ginsenosides apparently may impaire Ca^{++} influx and thereby suppress platelet aggregation.

It can be concluded that Rg_1 may decrease TXA_2 production stimulated by collagen or AA or U - 46619, most likely by inhibiting Ca influx.

It was reported that Ginsonoside - Rb_1(Rb_1), - Rb_2(Rb_2), - Rc(Rc) or - Rd(Rd) has no anti - platelet aggregative action in *in vitro*, but these saponins can be converted to Rg_3 in acid fluid(such as gastric juice), and exert anti - aggregative action. Therefore these saponins may act as pro - drug.

As for the effect of ginseng on biosynthesis of PGI_2, a potent vasodilatory and anti - platelet aggregative eicosanoid, Rc enhanced the production of PGI_2 by cultured rat vascular smooth muscle cells. Urinary excretion of 11 - dehydro - TXB_2, (TXA_2 - M) one of the major metabolites of TXA_2 and 2, 3 - dinor - 6 - keto - $PGF_{1\alpha}$, (PGI_2 - M) one of the major metabolites of PGI_2 in trial were measured. When KRG was orally given for 4 weeks to the patients with cardiovascular thrombotic disorders, urinary 2, 3 - dinor - 6 - keto - $PGF_{1\alpha}$ was significantly increased, while that of 11 - dehydro - TXB_2 was unchanged. Therefore the ratio of PGI_2 - M/TXA_2 - M was significantly increased after 4 weeks administration of KRG. This would certainly contribute to the improvement of peripheal blood circulation by ingestion of KRG and thereby lead to anti - thrombogenesis.

Endotoxin - induced thrombosis has been widely used as experimental model of thrombosis. Death due to thrombosis is considerably lessened by prior administration of KRG extracts. This could be explained by anti - thrombin and fibrinolytic action of KRG besides its anti - platelet aggregative action.

In ancient china, a large amounts of KRG was administered to critically ill patients. This may implicate the prevention of DIC from the viewpoint of modern medicine.

2. Arterial cells and red ginseng

Intimal thickening is one of the characteristic features commonly observed in atheromatous lesions. This thickened lesion consists of various components, especially smooth muscle cells and macrophages. Increase in smooth muscle cells in the intimal layer is caused by, at first, migration of smooth muscle cells from the media to the intima. The migration is initiated by migration factors secreted after injuries of endothelium. As migration factors, well known are platelet - derived growth factor(PDGF) or 12 - hydroxyeicosatetraenoic acid(12 - HETE) derived from platelets. Furthermore, smooth muscle cells themselves secret

a migration factor called smooth muscle cell - derived migration factor(SDGF) (Saito and Co - workers). Ginsenosides inhibited migration of smooth muscle cells induced by PDGF (3.2 ng/ml), Rc and Rb_2 being inhibitory by 20% at 0.1 μg/ml and 60% at 10 μg/ml.

Proliferation of smooth muscle cells also contributes to intimal thickening. Proliferation of smooth muscle cells require a growth factor. As growth factors, well known are PDGF, fibroblast growth factor(FGF) and epidermal growth factor (EGF) and so on secreted from various types of cells. Recently smooth muscle cell - derived growth factor(SDGF) secreted by smooth muscle cells has also been reported. The proliferation of smooth muscle cells induced by PDGF was inhibited by 50% by a ginsenoside, Ro(5 μg/ml), but not by other ginsenosides.

Next, we examined the *in vivo* effect of ginseng on the proliferation of smooth muscle cells. Blood was collected from subjects to whom 2.7g/day of red ginseng was administered for 4 weeks. This serum (up to 2.5%) inhibited proliferation of smooth muscle cells by 30% compared to the control. Thinking of these results, red ginseng may be beneficial in terms of prevention or treatment of the intimal thickening.

3. From cell formation and red ginseng

Observed in the atheromatous lesions are foam cells which accumulate lipids, mainly cholesterol ester. Foam cells are originated from macrophages and smooth muscle cells. Lipid accumulation is caused by uptake of denatured LDL in macrophages and LDL in smooth muscle cells. Disturbances of LDL metabolism in smooth muscle cells cause lipid accumulation, i.e., inhibition of cholesterol ester hydrolysis, increase in esterification of free cholesterol, and inhibition of cholesterol removal from cells to extracellular spaces. Ginsenosides increased cholesterol ester hydrolysis, Ginsenoside - Re(Re) being the strongest (30% increase over the control). The order of inhibitory potency was Re>Ginsenoside - Rg_2>Ginsenoside - Ro>Rb_2. These data suggest that red ginseng improves intracellular lipid metabolism and prevents lipid accumulation.

On the other hand, denatured LDL is specifically incorporated into macrophages through a specific receptor. This incorporation was inhibited by ginsenosides such as Rb_2 and Rc.

From these results, it was suggested that ginsenosides act as inhibitory to formation of atheromatous lesions.

[II] PSYCHOTOROPIC ACTION OF RED GINSENG IN ANIMALS

The therapeutic efficacy of KRG in a wide variety of mental illnesses ranging from a lack of appetite to psychosomatic diseases such as anxiety neurosis, depressive state and insomnia has been demonstrated through its long history of use in oriental medicine. However, experimental proof in support of these clinical applications, especially as a psychotropic drug, is limited.

1. Effects of intermale aggression

Ever since the classic finding that benzodiazepine anxiolytics have a taming effect on savage wild animals without producing excessive sedation, various experimental models of aggression have been used for evaluating the psychotropic action of a drug. To generate intermale aggression, a resident - intruder paradigm has recently been introduced into the behavioral analysis of drug action. Using this ethological paradigm, several series of experiment were conducted to investigate whether or not ginseng saponins alter social interaction between the attacking animal and the attacked animal. This kind of approach is considered to be useful for differentiating the pharmacological profile of psychotropic drugs. When the attacking resident mouse was treated with crude ginseng saponins (25, 50 100 mg/kg, i.p.), aggressive episodes such as offensive sideways posture, attack bite and tail rattle were significantly suppressed in a dose - dependent manner. However, the aggressive behavior was not altered when the intruder was treated with crude ginseng saponins. Rb_1 (2.5, 5 and 10 mg/kg, i.p.) also significantly suppressed aggressive episodes when given to the resident, whereas Rg_1 (2.5, 5 and 10 mg/kg, i.p.) was ineffective. Neither Rb_1 nor Rg_1 given to the intruder caused any significant changes in the behavior of the resident. Both benzodiazepine anxiolytics and tricyclic antidepressants suppressed the resident aggressive behavior without causing motor dysfunction when the resident was drugged. When the attacked intruder was drugged, however, benzodiazepine anxiolytics increased the resident's attack bite, while antidepressants had no such effect. Therefore, it appears that the effects of ginseng saponins on intermale aggression are comparable to those of the antidepressants.

2. Effects on female aggression

Ginseng - containing prescriptions such as Unkei - tou and Nyoshin - san have been thought to be effective in the management of indefinite complaints, postpartum depressive states and menopausal disorders in women. In this context, it is interesting whether or not ginseng saponins alter social behavior in female animals. Although in preclinical psychopharmacology little attention has been paid to female behavior in rodents, increasing evidence indicate that parturient female mice and rats which have been housed singly after a mating period display intense aggressive behavior towards on male intruder during early lactation. Using the maternal aggression in female mice, Yoshimura et al. found that acute administration of crude ginseng saponins (50 and 100 mg/kg, i.p.) and Rb_1 (2.5 and 5 mg/kg, i.p.) significantly suppressed maternal aggression in a dose - dependent manner, whereas Rg_1 was ineffective. As psychological factors during pregnancy seem to play an essential role in the manifestation of maternal aggression, the effects of chronic administration of drugs were also studied. As compared with the vehicle - treated group, chronic treatment with crude ginseng saponins (50 mg /kg/day, i.p.) and Rb_1 (2.5 mg/kg/day, i.p.) significantly suppressed postpartum maternal aggression, whereas Rg_1(2.5 mg/kg/day,

i.p.) showed a tendency to facilitate maternal aggression. It has been reported that acute administration of benzodiazepine anxiolytics increased maternal aggression, whereas serotonin - related anxiolytics and antidepressants suppressed it in a dose - dependent manner. Moreover, chronic administration of antidepressants and serotonin - related anxiolytics caused a significant decrease in the frequency of attack bite. These pieces of evidence suggest that the suppressive effect of Rb_1 on maternal aggression is similar in nature to the effect of antidepressants or serotonin - related anxiolytics.

3. Effects on copulatory disorder

The root of ginseng is well known to replenish a vital energy leading to the strengthening of sexual motivation in males. There is, however, little scientific approach on this field because an animal model for copulatory disorder has not been available. Recently, it was found that prolonged individual housing altered copulatory behavior of male mouse to a receptive female : the incidence and the frequency of copulatory behavior(mounting, intromission and penis licking) was remarkably low in individual housing group compared to aggregate housing group. The copulatory disorder in male mice following prolonged individual housing was recovered by acute administration of crude ginseng saponins(50 and 100 mg/kg, i.p.) and Rg_1 (5 and 10 mg/kg, i.p.). Chronic treatments with crude ginseng saponins (50 and 100 mg/kg/day, i.p.) during individual housing period also prevented the development of copulatory disorder in male mice. Interestingly, Rb_1 which was one of the psychoactive ingredients of ginseng root failed to show the alleviation of copulatory disorder. More recently, Yoshimura and Kimura (1991) established a new model which was caused by a repeated experience of defeat : repeatedly attacked male mice rarely showed copulatory behavior, whereas their attacking counterparts display more copulatory behavior. And they found that acute administration of crude ginseng saponins (25 and 50 mg/kg, i.p.) and Rg_1 (5 and 10 mg/kg, i.p.) significantly recovered copulatory behavior in defeated male mice. In this model, neither Rb_1, nor Ro altered copulatory disorder.

4. Effects on experimental dementia

Besides the therapeutic effects of KRG on psychosomatic, Sakanaka and co - workers recently concentrate on if and how crude ginseng saponins amend the impairment of memory acquisition and/or retention in gerbils with transient forebrain ischemia. Gerbils subjected to 5 min occlusion of the bilateral common carotid arteries exhibit a significant loss of pyramidal neurons within the hippocampal CA1 region, and concomitant disorder of memory retention when examined with a step - down passive avoidance apparatus. Preliminary experiments showed that treatment of the animals with crude ginseng saponins(100 mg/kg) before and after 5 min ischemia not only rescues the degeneration of CA1 pyramidal neurons but also prevents significantly the impairment of memory retention (Sakanaka et al., Unpublished observation). These findings are in line with previous in vitro studies suggestive of neurotrophic functions of KRG. It is of value to determine the chemical component of ginseng saponins, which exerts a prophylactic or therapeutic effect on cerebrovascular dementia.