Korean J. Ginseng Sci.
Vol. 14, No. 2, 317–331 (1990)

Nitrogen Compounds of Korea Ginseng and Their Physiological Significance

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Abstract□ Nitrogen compounds of *Panax ginseng* and their biological activities in plant and animal were reviewed. Major nitrogen compounds found in *P. ginseng* are free amino acids, water soluble proteins, insoluble proteins and peptides. Minor nitrogen compounds are dencichine, glycoproteins, amines, alkaloides, methoxy or alkyl pyrazine derivatives, free nucleosides and nucleic acid bases, 4-methyl-5-thiazoleethanol and pyroglutamic acid. The contents of total nitrogen and protein in root increased until 13 years old which was the highest age under investigation. Soluble protein content increased with the root weight and was higher in xylem pith than cortex-epidermis indicating the close relation with root growth. Arginine which covered 58% of total free amino acids may serve as a storage nitrogen. Arginine seems to be changed into proline in rhizome, threonine in stem and again threonine and arginine in leaf. The greater the root weight the higher the polyamine content. Polyamine stimulated the growth of root callus. Physiological roles of other minor nitrogen compounds are unknown although dencichine content is relatively high (0.5% d.w). Biochemical and pharmacological activities of some nitrogen compounds for animal were more investigated than physiological role for plant itself. Radiation and U.V. protective function (heat stable protein), insulin-like activity in lipogenesis and lipolysis (adenosine and pyroglutamic acid), depression of blood sugar content (glycopeptide), hemostatic and neurotoxic activity (dencichine) and, sedative and hypnotic activity (4-methyl-5-thiazoleethanol) are reported. Heat stable protein increased with root age. The traditional quality criteria appear to be well in accordance with biological activities of nitrogen compounds. Chemical studies of nitrogen compounds seem relatively rare, probably due to difficulty of isolation, subsequently the investigations of biological activities are little.

Key words□ *Panax ginseng* C.A. Meyer, nitrogen compounds, review.

Introduction

Since biologically active compounds are very important criteria for medicinal plants, chemical analysis has been concentrated to find indigenous compounds in medicinal herbs. Some indigenous compounds used to be considered as biologically active principles without full test of biological activities. The ideosyncracy of chemical composition can not be fully proved because number of unknown compounds in plant kingdom are much greater than that of known compounds. The test of biological activities of isolated compounds are so limited that the collation of activity with the traditional use is difficult in the most cases. Such difficulty pronouncedly arises in the herbs with high frequency in recipe, especally such as Korea ginseng.

Since nitrogen compounds are much abundant, mostly primary metabolites and difficult to handle than saponins pharmaceutical analysts seemed to choose saponin as unique compounds in Korea ginseng. Through the series on traditional quality control in production and processing of Korea ginseng in relation to chemical constituents[1-5] nitrogen compounds appeared to be very important not only for the growth of ginseng but also for medicinal efficacy. Although nitrogen compounds have been reviewed[6-8] it has been done only in various sections with all other compounds. This fact indicates that no special attention was given on nitrogen compounds.

We believe that nitrogen compounds deserve a single review than any other compounds. Biological activities of nitrogen compounds are considered in two ways, for ginseng plants and human.

1. Nitrogen compounds found in Korea ginseng

Analytical approach to biologically active compounds of *P. ginseng* through more than one century was reviewed to investigate the concept of biologically ative compounds[4]. In that review amino acids and vitamins were not included since those are commonly found in other plants. When such common compounds and other rare compounds that found recently are included all compounds reported except common organic acids, fatty acids, and some lipids, are grouped as Table 1 in relation to nitrogen compounds. Number of nitrogen compounds are smaller than that of non-nitrogen compounds. Non-nitrogen compounds are mostly secondary metabolites while most nitrogen compounds are primary ones.

Table 1. Major biological active compounds in *Panax*

Group of compound	Number of compound	Efficacy
Nitrogen compounds		
Alkaloids	8	Antitumor
Choline	1	Hypotonic action on blood pressure, Lipotropic agent
Amino acids	24	Insulin-like Hemostatic effect
Amines	3	Growth regulator in plants
Nucleosides and bases	8	Antilipolytic
Peptides and proteins	9	Radiation protective, Decrease blood sugar
Vitamins	6	
Pyrazines	14	
Thiazols	1	Sedative, hypnotic
Non nitrogen compounds		
Ginsenosides	26	Various efficacies
Phenolics	8	Antioxidant, Hematopoietic
Phytosterols	4	Cholesterol inhibitor
Polyacetylenes	10	Antitumor, Antiplantelet
Polysaccharides	5	Antitumor, Hypoglycemic
Sesquiterpene	2	
Volatile oils	2	

It may be the first reason for pharmaceutical chemist to neglect nitrogen compounds. Number of nitrogen compounds known are slightly less than that of other compounds, even under such neglected situation. This strongly suggests that there are a large number of unknown nitrogen compounds.

2. Proteins

Water soluble nitrogen ranges 60 to 90% of total nitrogen depending on the parts, and about 20% of soluble N is soluble protein[3]. The content of soluble protein increased with the weight of root[3] indicating that soluble protein has certain role for root growth. Furthermore, protein content in *P. ginseng* root gradually increased with the age until 13 years old that was the oldest age under investigation[9] as shown in Table 2.

Soluble protein showed radiation protective effect on X-ray irradiated animals but saponin fraction had no such effect[10]. Similar result was obtained in other experiments with γ-ray irradiation[11]. The water soluble extract of whole root gave the best protection and the isolated protein and carbohydrate fractions gave less protective while the saponin fraction did not protect. This fact indicates that saponin may serve only sinergistically to protect radiation with protein and carbohydrate if

Table 2. Protein content of *P. ginseng* root (%)

Age	7	8	9	11	12	13
Crude	13.96	14.09	14.83	14.83	14.98	15.72
Precipitated*	9.14	9.16	9.64	9.64	9.73	10.22

*1.25% NaOH, 6% H_2SO_4 Hong and Kim (1971).

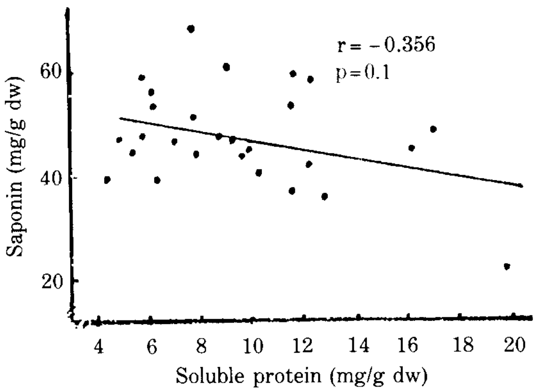


Fig. 1. Relationship between soluble protein and saponin pith-xylem and Heaven grade percentage in *Panax ginseng* root.

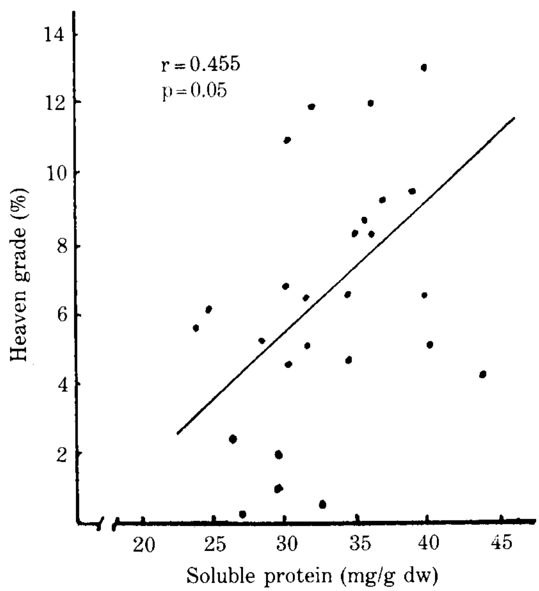


Fig. 2. Relationship between soluble protein content in pith-xylem and Heaven grade percentage in *Panax ginseng* root.

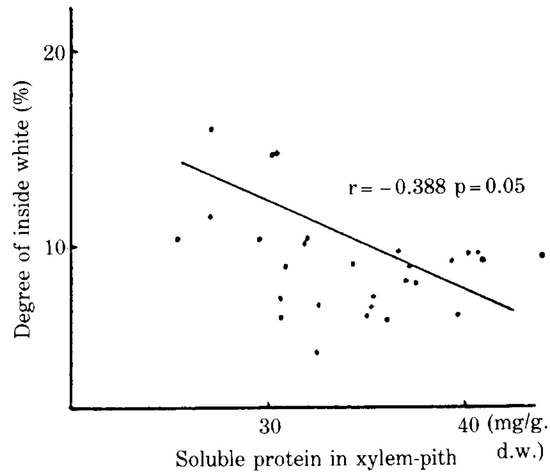


Fig. 3. Relationship between soluble protein in xylem-pith and degree of inside white.

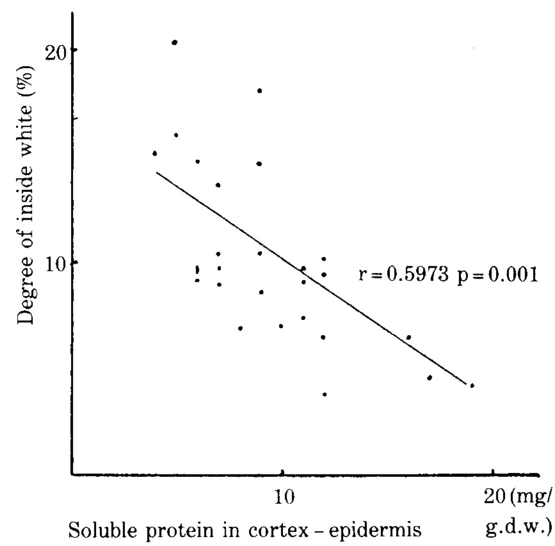


Fig. 4. Relationship between soluble protein in cortex-epidermis and degree of inside white.

saponin is concerned. The same protein fraction increased survival rates and reduced chromosome aberrations of UV irradiated cells[12]. One of radiation protective protein did not contain saponin[10] but it is not clear yet that all radioprotective protein fraction did not contain saponin.

There was negative correlation between soluble protein content and saponin content as shown in Fig. 1[13] suggesting that saponin may not be much concerned in radiation protection. Soluble protein has close relation to red ginseng quality. The higher the soluble protein content in xylem-pith of raw ginseng, the greater the percentage of Heaven grade (the best grade) of red ginseng[13] as shown in Fig. 2. The roots bearing the inside white after processing were eliminated from red ginseng. The higher the soluble protein content in xylem-pith (Fig. 3) or cortex-epidermis (Fig. 4) the lesser the degree of inside white occurrence. Thus the traditional red ginseng quality measure appears to be close relation to soluble protein fraction, probably radiation protec-

Table 3. Various protein fraction

(mg/g.d.w)

		Water soluble	Heat stable	TCA stable	Heat ppt.	TCA ppt.	Insoluble N
P. ginseng	Fresh	43.4	24.4	12.7	12.7	28.6	4.8
	Red	19.7	18.1	19.5	0.1	0.01	9.1
P. quin	Fresh	27.4	19.1	7.6	3.0	15.3	3.9
	Wild	23.7	18.1	8.7	5.1	6.5	4.3

Table 4. Thermostable protein in *P. ginseng*

Content or M.W.	Biological activity	Reference
G1 (1.7% to extract)	Haematological recovery of	Yonezawa 1980
G3 (3.1% to extract)	erythrocyte and thrombocyte,	
G2 (1.6% to extract)	not effective	
G1 200K, 100K, 52K, 51K G2 41K G3 28K, 19K, 14K	Radioprotective protein	Kim 1986

tive protein. The radiation protective protein was thermostable[10,12]. Soluble protein in red ginseng was almost heatstable as shown in Table 3 (unpublished) since red ginseng was heat processed. Furthermore most heatstable protein in red ginseng was not precipitated with trichloroacetic acid (Table 3), indicating relatively low molecular peptides. The contents[14] and molecular weight[15] of thermostable radiation protective protein were shown in Table 4. There was no information about TCA solubility of each fraction. The relationship between thermostability and TCA solubility of ginseng protein must be further studied.

The radiation protective activity per unit weight of extract was much greater in the central part than in the cortex part[16], because the soluble protein content was higher in the central part[3] and probably due to higher content of effective protein fraction. Since the ratio of central portion to the outside pro-

Table 5. Relationship between root weight and soluble or thermostable protein content in *P. ginseng* (1988)

	Xylem – pith	Cortex – epidermis	Main body
Soluble (S)	0.446	0.212	0.453
Thermostable (T)	0.613	0.786*	0.749*
T/S	0.398	0.887**	0.772*

**,*: significant at p=0.05 and 0.1 (n=6).

tein increases with the increase of root weight it is well expectable that the greater the root weight the more effective in radiation protection. Thus it is rational that the larger the root the higher the price per unit weight as it has long been such in the tradition[4]. It is well understood from the fact that the larger the root the higher the content of soluble or thermostable protein[17] as shown in Table 5. The radiation protective activity may exert in two ways, direct[18] and indirect[19]. In the latter case active protrigger to produce second principle.

Probable repair effect of damaged DNA by mutagen or increased stability of DNA tested on cell level seemed to indicated a direct effect. The increase of Th/Ts (helper/supressor) in immune system by red ginseng powder[19] seemed to suggest a triggering effect. The cancer patients received radiation therapy decreased Th/Ts.

3. Peptides

Gstirner and Vogt[20] first reported Korea ginseng peptides in 1966. They investigated only amino acid compostition of two peptides. Biological activity of ginseng peptide fraction was first reported[21]. That fraction, however, was known as pyroglutamate[22]. Kim *et al.*[23] reported antilipolytic fraction that contained oligopeptides.

A tetradeca peptide was isolated and its amino acid sequence and secondary conformation were investigated[24]. This peptide decreased levels of blood sugar and hepatic glycogen. Studies on peptides are very rare (Table 6). It may be due to difficulties for isolation and elucidation of molecular structure. The fact that a predicted peptide was known as pyroglutamate after 10 years studies[22] indicates the

Table 6. Peptides in *P. ginseng*

Name	M.W. or content	Biological activity	Reference	
Five peptides			Gstirner	1966
Oligopeptides	P-F-2 fraction	Antilipolytic	Kim	1987
Tetradeca peptide	1620	Decrease blood sugar and hepatic glycogen	Zhang	1988
Panaxan A,B,C,D,E	A:14 K (1.7% protein in glycan)	Hypoglycemic in mice	Tomoda	1984
P.G. 5-1	105K (11.5% protein in polysaccharide)	Carbon clearence of	Ohtani	1988
P.G. 5-2	29K (17.9% protein in polysaccharide)	Reticuloendotherlial system	Ohtani	1988

Table 7. Some examples of biologically active peptides

Name	Number of amino acid residue	Function
Methionine-enkephalin	5	Inhibits pain sense (brain)
Angiotensin II (horse)	8	Pressor or hypertensive also stimulates release of aldosterone from adrenal gland
Plasma bradykin (bovine)	9	Vasodilator peptide
Vasopressin (antidiuretic hormone)	9	Causes kidney to retain water from urine (pituitary gland)
Substance P	10	Neurotransmitter
Glucagon (bovine)	29	Pancreatic hormone regulating glucose metabolism
Calcitonin	32	Lowering of blood Ca^{2+} (parathyroid and thyroid gland)
ACTH (Adrenocorticotropic hormone)	39	Stimulates secretion of steroid hormone (adrenal cortex)
CRF (Corticotropin releasing factor)	41	Stimulates ACTH secretion (anterior pituitary)
Insulin	50	Sugar metabolism
PTH (Parathyroid hormone)	84	Causes kidney excrete phosphate

difficulty of peptide study. As seen in Table 3 TCA insoluble fraction may be peptides.

There are biologically active peptides working as hormones (Table 7)[25]. Some peptides in ginseng may work directly or indirectly as hormones. Pyroglutamic acid, once suspected as a peptide, was reported as a selective modulator in lipid metabolism[22].

There are two hormone peptides that contain pyroglutamic acid, thyrotropin releasing factor (TRF), and little gastrin[25]. Their primary structures are shown in Table 8. Generally accepted ginseng effect for stomach may be related with similar peptides to little gastrin. The existence of pyroglutamic acid contained peptide seems to be highly probable. TCA soluble fraction of ginseng protein (Table 3) may be such peptides. Some peptides may be more related to root growth.

Konno *et al.* isolated glycans (Panaxan A,B,C,D,E) having hypoglycemic activity[27]. Panaxan A consisted of mainly D-glucose (92.1%) and a peptide moiety (1.7%)[28] indicating that active prin-

Table 8. Biologically active pyro-Glutamate peptides

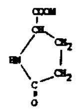
Amino acid sequence	Name	Function	Reference	
	Pyro-glutamate acid	Inhibition of lipolysis	Okuda	1988
pyroGlu-His-Pro (NH_2)	Thyrotropin releasing facter	Secreted by hypothalamus and causes pituitary gland to release thyrotropic hormone	Devlin	1986
pyroGlu-Gly-Pro-Trp-Leu-Glu-Glu-Glu-Glu-Glu-Ala-Try-Gly-Trp-Met-Asp-Phe (NH_2)	Little gastrin (human)	Hormone secreted by mucosal cells in stomach and causes parietal cells of stomach to secrete acid	Devlin	1986

Table 9. Amino acid content of *Panax* root

(mg/g dw)

	P. ginseng		*P. quinquefolius*		*P. notoginseng*	
	Xylem & pith	Cortex & epidermis	Xylem & pith	Cortex & epidermis	Xylem & pith	Cortex & epidermis
Free. a.a	39.9	21.4	18.6	14.9	10.8	11.8
Total a.a	88.9	101	120	82.2	84.2	58.6
F/T (%)	44.9	21.2	15.5	18.1	12.8	20.1

ciples are glycopeptides (peptidoglycans).

Polysaccharides (PG5-1 and PG5-2) were isolated from *P. ginseng* and found to potentiate reticuloendotherial system[29]. Both compounds contain 11.5 and 17.9% proteins suggesting that they are a kind of glycopeptides. Thus they are included into peptides as shown in Table 6. Studies on active site of these glycoproteins will elucidate the significance of peptide moiety. In searching biologically active peptides a question about biological activities arises on lipoproteins.

4. Amino acids

Amino acids of *P. ginseng* exist as free and in peptide and protein. The contents of total and free amino acids (FAA) in Panax species were shown in Table 9[30]. FAA are about 45% of total amino acids (TAA) in xylem-pith of *P. ginseng*. *P. ginseng* showed highest free amino acids content. In some cases FAA to TAA was higher than 45%[3], according to season[31]. FAA increased for wintering and shooting and decreased root growing season[31]. The increase of FAA during winter seems to be protective mechanism. Low temperature storage increased FAA but decreased protein AA in *P. ginseng* root[32]. Main free FAA of Panax species were shown in Table 10[30]. Arginine content was highest and it was more pronounced in *P. ginseng* (59-62% of total FAA). Second abundant FAA was different only in the central part of *P. ginseng*. FAA patterns are not much different between species as shown in Table 11. It is interesting that similarity is very high between cortex-epidermis of *P. ginseng* and *P. notoginseng*, while the lowest between xylem-pith of *P. ginseng* and contex-epidermis of *P. notoginseng*.

Relative abundance of main amino acids of root protein was shown in Table 12. Glutamic acid was most abundant in *P. ginseng* but arginine was in *P. ginseng* or *P. notoginseng*. Arginine was second aboundant in *P. ginseng* and especially high in cor-

Table 10. Relative abundance of main free amino acids in *Panax* roots

(%)

P.ginseng Xylem & pith		*P.ginseng* Cortex & epidermis		*P.quinquefolius* Xylem & pith		*P.quinquefolius* Cortex & epidermis		*P.notoginseng* Xylem & pith		*P.notoginseng* Cortex & epidermis	
Arg.	59.4	Arg.	61.7	Arg.	51.6	Arg.	51.6	Arg.	47.3	Arg.	43.9
Thr. (+ser)	11.9	His.	8.8	His.	12.9	His	11.6	His.	24.3	His.	24.2
His.	7.0	Asp.	8.0	Ala.	8.0	Pro.	7.0	Asp.	10.5	Asp.	11.6
Asp.	6.4	Thr.	6.1	Thr.	5.4	Ala.	6.6	Tyr.	3.5	Ala.	3.2
Ala.	5.1	Ala.	4.3	Asp.	5.4	Glu.	5.4	Ala.	3.3	Phe.	3.0

Table 11. Similarity of amino acid composition of root among *Panax* species

Free a.a. (above diagonal); Protein a.a (below diagonal)

		P.ginseng Xylem & pith	*P.ginseng* Cortex & epidermis	*P.quinquefolius* Xylem & pith	*P.quinquefolius* Cortex & epidermis	*P.notoginseng* Xylem & pith	*P.notoginseng* Cortex & epidermis
P.ginseng	Xylem & pith		0.9846****	0.9609****	0.9479****	0.9586****	0.8520****
	Cortex & epidermis	0.8192****		0.9755****	0.9698****	0.9117****	0.8974****
P.quinquefolius	Xylem & pith	0.2917	0.6341**		0.9919****	0.9224****	0.9060****
	Cortex & epidermis	0.1043	0.4968*	0.8309****		0.9072****	0.8894****
P.notoginseng	Xylem & pith	0.5039*	0.8833****	0.7675***	0.6172**		0.9948****
	Cortex & epidermis	0.3137	0.6266**	0.5923**	0.3625	0.6738***	

****,***,**,*; p=0.001, 0.01, 0.05 and 0.1

Table 12. Relative abundance of main amino acids of protein in *Panax* roots

(%)

P.ginseng Xylem & pith		*P.ginseng* Cortex & epidermis		*P.quinquefolius* Xylem & pith		*P.quinquefolius* Cortex & epidermis		*P.notoginseng* Xylem & pith		*P.notoginseng* Cortex & epidermis	
Glu.	29.3	Glu.	17.0	Arg.	14.0	Try.	10.3	Arg.	13.3	Arg.	32.3
Arg.	12.9	Arg.	14.3	Phe.	13.5	Arg.	10.1	Glu.	11.3	Glu.	10.3
Asp.	9.7	Asp.	10.0	Glu.	11.1	Phe.	10.0	Asp.	11.0	Leu.	9.4
Pro.	8.1	Leu.	8.8	Leu.	9.7	Glu.	9.5	Leu.	9.7	Asp.	7.2
Lys.	7.4	Lys.	6.8	Try.	8.5	Leu.	8.7	Phe.	8.1	Try.	5.9
Leu.	7.3	Ala.	5.5	Asp.	7.3	Ile.	8.1	Try.	7.1	Phe.	5.6

tex-epidermis of *P. notoginseng.* The composition of protein amino acids was quite different between Panax species, especially between *P. ginseng* and *P. quinquefolius* as seen in Table 11. Although amino acid compoisitions of some peptides[20,24] showed low arginine content most proteins of Panax species contain large amount of arginine. Physiological role of high content of arginine for ginseng plant may be the nitrogen storage as it is in other plant seeds[33].

Physiological role[34,35] of arginine for human was known as Table 13, various efficacies of ginseng are

Table 13. Biological activity of arginine for animal

Increases growth hormone	
	Burns fat and builds muscle
	Speeds wound heeling
Stimulates thymus gland (Immune system)	
	Activation of helper lymphocytes, provably helpful for AIDS: (supressor lym./helper lym.)
Inhibits cancer, (antitumor)	
Increases sexual fertility in males	
	Increases sperm count & motility
Generates endotherial-derived relaxing factor (NO)	

similar to the arginine effect. Since ginseng proteins contain large amount of arginine, it may be released after protein hydrolysis. Difference in efficacy of free arginine from protein arginine is unknown. The efficacy of ginseng must be reevaluated by the efficacy of arginine. It can not be ruled out that various activities of ginseng reported may be attributed to arginine and other compounds. It is also very interesting that *Codonopsis lanceolata* that has long been called as vine seng, with high price in the case of wild, contains large amount of free arginine (37% of total free amino acids) about 6 times than other wild vegetable plants.[36)]

Feee amino acid composition of other part of ginseng is different from root. In rhizome proline was 77% of total free amino acids about one month before emergence and after emergence threonine + serine and ammonium increased.[30)] Arginine in root seems to change into proline in rhizome and then into threonine + serine and ammonium especially in stem. After emergence argiline increased in leaf. It is not clear how reserved arginine in root are used in rhizome, stem and leaf. The basic free amino acids were not found in ginseng leaf[37)] but it was not true[30)].

Except 20 common amino acids hydroxy-proline, γ-amino butyric acid asparagine and glutamine were found in flower[38)] and seed[39)] and methionine sulfoxide, pipecolic acid in the dehisced seeds.[39)] Norleucine was found in fresh ginseng[40)]. The unusual amino acids are pyroglutamic acid[22)] as mentioned above and dencichine[41,42)]. Dencichine is 3-N-oxalo-L-α,β-diaminopropionic acid and has hemostaitic and neurotoxic activity such as antihemorrhagic[41)].

Table 14. Special amino acids in *P.ginseng*

Name	Content	Biological activity	Reference
Dencichine	0.31% in P.q		Zheng 1990
	0.5% in P.q	Hemostatic	Personal
	0.9% in *P.n*	Neurotoxic	Communication
Pyroglutamic acid		Inhibition of lipolysis	Okuda 1988

Table 15. Content of polyamine in root and their relation to root weight in *P.ginseng* (nmol/g.d.w.)

	Putrescine (P)	Spermidine (S)	P+S
Mean	11.6±3.9	13.6±2.7	23.9±6.1
Max	20.0	17.5	33.7
Min	7.5	8.1	15.7
R Tap root	0.750**	0.677*	0.688*
R Whole root	0.819***	0.622*	0.761**

R: simple correlation
***,**,*: significant at p=0.01, 0.05 and 0.1 (1989)

It was found in *P. notoginseng* and other six medicinal plants. Its content in root was 0.50%, 0.31% and 0.90% for *P. ginseng. P. quinquefolius* and *P. notoginseng*, respectively (Table 14). Physiological role and metabolic pathway are not investigated eventhough its content is relatively high. Dencichine may serve as nitrogen reserve as arginine or protective agent.

5. Amines

Polyamines are found in *P. ginseng*[43)]. Spermidine content was about 14 nmol/g.d.w. and putrescine content was about 12 nmol/g.d.w. as shown in Table 15. Spermine existed in trace and was not quantitated. Spermidine content was highest in xylem-pith of tap root and least in cortex-epidermis of lateral root (Table 16). The content of spermidine and putrescine showed significant positive correlation with root weight as shown in Table 15 and Fig. 5. The close positive relationship bet-

Table 16. Content of spermidine in various parts of *P.ginseng*

(nmol/g.f.wt)

	Tap root		Lateral root		Fine root
	Xylem & pith	Cortex & epidermis	Xylem & pith	Cortex & epidermis	
Mean	16.6	14.7	12.6	12.1	14.1
S.D.	2.05	1.91	1.29	1.47	2.25

n=3

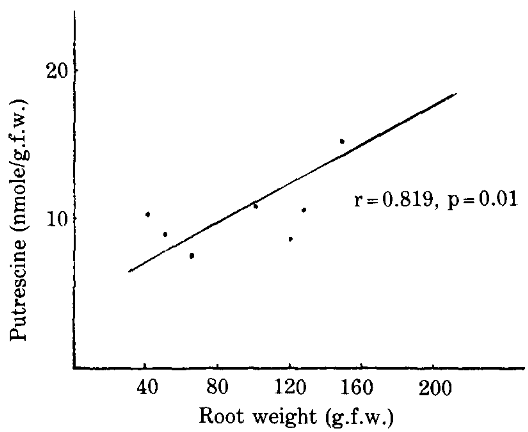


Fig. 5. Relationship between root weight and content of putrescine in root of *P. ginseng.*

Table 17. Effect of polyamines on growth of ginseng callus

(g fw/flask)

PAS conc.	Putrescine	Spermidine	Spermine
(ng/ml)			
10	7.81±1.7	7.81±1.5	8.29±1.3*
50	7.97±1.1	7.70±1.2	7.52±1.8
100	7.56±1.8	7.48±1.5	7.17±1.2
500	8.07±1.5	8.31±1.4	7.07±1.9

f.w. in control: 6.91±1.3

*F test: significant at 0.05 to control

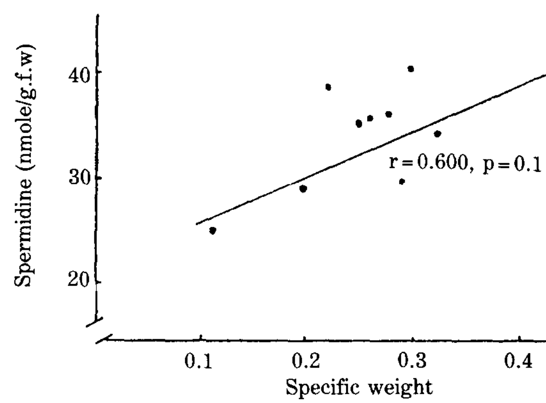


Fig. 6. Relationship between specific weight (g.d.w/cm³ F.V.) and spermidine content in root of *P.ginseng.*

ween root weight and putrescine content suggests growth stimulation by polyamines.

The growth of ginseng callus affected by putrescine concentration as shown in Table 17. Spermine that exists in trace showed significant growth stimulation at the lowest concentration. Main polyamines showed highest effect on the callus growth at highest concentration but there was no consistency among concentrations. Further experiments are needed for more precise information, however the role of polyamine as a growth hormone is highly probable in ginseng. Spermidine content in tap root was significantly and positively correlated with specific weight of tap root (Fig. 6) indicating that spermidine keeps the tissues of especially xylem and pith young. Specific weight indicates filling degree of tissue and low specific weight causes inside white of red ginseng[13].

6. Alkaloids

Although alkaloid investigation started relatively early[44] most alkaloids except choline in ginseng were recently isolated[45-47] probably due to small quantity as shown in Table 18. An old report that *P. quinquefolius* did not contain an alkaloid[48] may partly attribute to the delaying of alkaloid investigation. From alkaloidal fraction pyrrolidone was isolated and identified but did not match to the activity of alkaloidal fraction[44]. Choline content of *P. quinquefolius* was quantitated and was found to have hypotonic action[49]. Choline content in *P. quinquefolius* was reported[48] but since it is unusually high it should be checked. Choline content in *P. ginseng* was not much different among years, parts and weights as shown in Table 19.

The role of choline in ginseng plants may be as

Table 18. Alkaloids of *P.ginseng*

Name	Content	Biological activity	Reference	
N_9-Formyl-1-methyl-β-carboline	0.13 μg/g.d.w.		Han	1986
1-Carbobuthoxy-β-carboline	0.11 μg/g.d.w.		Park	1987
1-Carboethoxy-β-carboline	0.14 μg/g.d.w.	c-AMP phosphodiesterase inhibition	Han	1986
1-Carbomethloxy-β-carboline	0.36 μg/g.d.w.		Park	1987
1-(5-Hydrooxymethyl-2-furyl)-β-carboline	0.64 μg/g.d.w.		Han	1986
Norharman	0.24 μg/g.d.w.		Park	1987
Norharman acetate Harman	0.15 μg/g.d.w.		Park	1987
Spinacine			Han	1987
Choline	6.2 (*P.q*)	lippotropic	Joseph	1977
	0.1-0.2%	hypotonoic action on B.P.	Takatori	1963
α-Pyrrolidone			Woo	1965
Alkaloidal fraction		activation or inhibition of dehydrogenase	Cho	1974

Table 19. Choline content in tap root of *P.ginseng* (mg/g dw)

Age	3	4	5	6		
root size				42	82	113
Xylem & pith	1.13	1.22	1.05	1.22	1.55	1.43
Cortex & epidermis	0.95	1.37	1.13	1.50	1.50	1.33

Table 20 Effect of choline for animal

Precursor: Lecithin-Cell membrane
Acetylcholine-neurotransmitter
(Alzheimer's disease)
memory inhancement.
Lipotropic agent
Hypotonic action on blood pressure.

precursor of lecithin for cell membrane as it is for animal. Choline activities[34,48,49] for animal were as Table 20. Lipotropic activity (preventing the desease of fat accumulation), memory inhancement and hypotonic action are all the effects with whole ginseng. Choline alone did not show such effect and some modification may be needed when using as nutritional administration[34]. This fact indicates choline in ginseng may exert such effect due to other compounds.

Minor alkaloids are 5 carbolines, 3 harmans and spinacine. Physiological role for ginseng may be as phytoalexins. Biological activity for animal is not investigated except some effect on enzymes as shown in Table 18. Spinacine in *P.g.* was the first case in plant kingdom[47].

7. Nucleosides and nucleic acid bases

Only Free nucleic acid bases and nucleosides were reported[48,50,52] as shown in Table 21. Hiyama *et al.* found three free nucleic acid bases (uracil, guanine, adenine) and two nucleosides (uridine, adenosine), from the butanol layer of water extracts of white ginseng and dried fine root[50]. There was no difference between tap root and fine root in qualitatively but they were quantitatively higher in fine root as shown in Table 22. Hong *et al.* found two more nucleic acid bases, nucleosides, and orotic acid, precursor of pyrimidine[51]. The content of bases or nucleosides were quite different according to the investigators. Most abundant compound was orotic acid in both white and red ginseng. The content of nucleic acid compounds were higher in red

Table 21. Free nucleosides and bases in *P.ginseng*

Name		Materials	Biological activity	Reference	
Adenosine	Structure	Red ginseng (R)	Antilipolytic	Okuda	1980
		Red & white ginseng (W)		Han	1985
		Fresh ginseng	growth regulator	Park	1989
Nucleosides	Adenosine	R & W	in plant	Hong	1980
	Cytidine	R & W		Hong	1980
	Uridine	R & W, W		Hong 1980,Hiyama 1978	
Base	Cytosine	R & W		Hong	1980
	Thymine	R & W		Hong	1980
	Guanine	R & W		Hong	1980
	Uracil	R & W, W		Hong 1980, Hiyama 1978	
	Adenine	R & W, W		Hong 1980, Hiyama 1978	
Precursor	Orotic acid	R & W		Hong	1980

Table 22. Free nucleic acid compounds in ginseng (mg/100g)

Cytidine	1.18	0.14	2.85	0.32	
Uridine	1.73 (13)	0.03	0.41	1.47	(47)
Adenosine	1.35	0.04	3.25	0.10	
Cytosine	1.13	0.08	0.62	0.24	
Uracil	7.52 (6)	0.65	20.20	0.18	(16)
Thymine	1.50	0.04	0.15	0.04	
Guanine	6.41	0.18	0.88	0.05	
Adenine	0.55 (1)	0.01	-	0.06	(13)
Orotic acid	20.94	1.57	38.73	10.53	

( ): The data from Hiyama (1978)

difference may be due to root age, since white gin-ginseng than white one as shown in Table 22. The seng is made of 4 years old root but red ginseng is made of 6 years old root. Nucleic acid-nitrogen may increases with age as protein nitrogen increases with age as mentioned above.

Adenosine was isolated from red ginseng and found to have insulin-like activity[52]. Adenosine was also isolated and molecular structure was determined in red and white ginseng[53]. In fresh ginseng adenosine was isolated (90 mg/100g d.w.) and identified for structure[43]. Since adenosine was reported as probable growth stimulator[54], further studies are needed in relation to root weight.

8. Pyrazines

Five methoxy pyrazine and eight alkylpyrazine derivatives were identified from the basic fraction of the ether extract or dried fine root of *P. ginseng*[55]. The characteristic earthy, green aroma with moldy undertone of ginseng root were assessed to be attributed to the methoxy pyrazine derivatives. Such compounds were not appeared in volatile flavor components of fresh ginseng[56]. From the either extract of freezing trap ice of fresh tap root two methyl-pyrazine derivatives were found by gas chromatography[57]. The nitrogen containing flavor compounds are shown in Table 23.

9. Other nitrogen containing compounds

Most nitrogen containing vitamins were quantitated in ginseng root by using microorganism in 1964[58-60] as shown in Table 24. Such amount of vitamins may not be negligible for biological effect. Vitamin B_{12} content increased up to 3 years old and folic acid up to 4 years old[60]. With more precise analytical method and sampling age-related investigation is worthwhile to assess age effect. The 4-methyl-5-thiazoleethanol was isolated from dried ginseng[61] and is an intermediate of thiamine synthesis and it is the first case of natural existence[62] suggesting that it is an intermedate in biosynthesis. This compound was used for sedative and hypnotic

Table 23. Nitrogen containing flavor compounds in *P. ginseng*

Group	Name	Reference	
Methoxy pyrazine	2-Isopropyl-3-methoxy pyrazine	Iwabuchi	1984
	3-Isopropyl-2-methoxy-5-methyl pyrazine		
	2-sec-Butyl-3-methoxy pyrazine		
	3-sec-Butyl-2-methoxy-5-methyl-pyrazine		
	2-Isobutyl-3-methoxy pyrazine		
Alkyl pyrazine	2-Ethyl-6-methyl pyrazine		
	2-Ethyl-5-methyl pyrazine		
	Trimethyl pyrazine		
	2,6-Diethyl pyrazine		
	2,3-Dimethyl-5-ethyl pyrazine		
	Tetramethyl pyrazine		
	2,6-Diethyl-3-methyl pyrazine		
	2-Ethyl-3,5,6-trimethyl pyrazine		
	2,6-Dimethyl pyrazine	Park	1990

Table 24. Other nitrogen compounds in *P.ginseng*

Name	Content	Biological activity	Reference	
4-methyl-5-thiazoleethanol	0.08 μg/g.d.w.	Sedative effect	Park	1987
Niacinamide	0.34 μg/g.d.w		Park	1987
Vitamin B_{12}			Kim	1964 b
Nicotinic acid			Kim	1964 b
Folic acid			Kim	1964 b
Pantothenic acid	0.66 mg/100g		Kim	1964 a
Biotin	0.92 μg/100g		Kim	1964 a

therapy.[63] Some experience falling long sleep after taking wild ginseng may be due to hypnotic action of this thiazole compounds.

10. Concluding remarks

Nitrogen containing compounds are recently attracted attention from researchers. Inspite of just begining in searching nitrogen compounds the prospect in both physiological role for ginseng growth, and biological activity for human being is very blight. Radiation protective activity seems to be profoundly concerned to antiaging mechanism, thus to antistress activity. This principles are thermostable proteins and not ginsenosides. Furthermore such fact is well in accordance with traditional quality control measures. Peptides appear to be more important than thermostable protein due to the hormone-like activity as seen in pyroglutamic acid since the existence of pyroglutamate-containing peptide can not be ruled out. High content of free arginine is very interesting point of nitrogen metabolism. Hemostatic and neurotoxic dencichine is another good example of biologically active amino acid. Immune enhancing polysaccharides may have small peptides as active sites. Polyamine could be used to break the slow growth rate of ginseng. Adenosine is the insulin-like compound but also another target compound in relation to plant growth rate. No one knows the biological activity of small amount of alkaloids. These compounds should not be neglected simply due to small content. Some biological activities of ginsenosides reported may be attributed to the adherent trace of nitrogen compounds. The compounding effect of all the biologically active nitrogen principles and, of those with other principles should be considered. The horison of nitrogen compound research is wide and just at dawn waiting for the rising sun. Nitrogen compound research will give answer to why Korea Ginseng grow so large and has long been in high reputation.

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