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γ-Aminobutyric Acid (GABA) Content of Selected Uncooked Foods

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Abstract

We analyzed the γ-aminobutyric acid (GABA) content of a selection of uncooked foods. Foods with GABA concentrations in excess of 100 nmole per g dry weight included: brown rice germ, brown rice sprouts, barley sprouts, bean sprouts, beans, corn, barley, brown rice, spinach, potatoes, sweet potatoes, yams, kale and chestnuts. Cereals included: brown rice germ, brown rice sprouts, barley sprouts, bean sprouts, beans, corn, barley, and brown rice and had GABA concentrations of 718, 389, 326, 302, 250, 199, 190, and 123 nmole per g dry weight (DW), respectively. The vegetables: spinach, potatoes, sweet potatoes, yams and kale contained 414, 166, 137, 129, 122 nmole GABA per g DW, respectively. The GABA concentration of chestnut was 188 nmole per g DW. However, oatmeal, adlay, broccoli, squash, carrots, onion, apples, lentinus edodes, green laver, and lactobacillus had GABA concentrations of less than 100 nmole per g DW. These results show that brown rice germ, sprouted cereals and spinach are good sources of plant-derived GABA. These data will be useful in selecting foods for the manufacturing of uncooked foods containing a relatively high concentrations of GABA.

Key words: GABA, uncooked foods, cereals, vegetables

INTRODUCTION

γ-Aminobutyric acid (GABA) is a ubiquitous non-protein amino acid (1,2). The presence of GABA in plants has been known for at least half a century (2). It has been reported that the levels of GABA in plants are low, but enhanced during high stress conditions such as: mechanical stimulation, hypoxia, cytosolic acidification, water, darkness, and drought (3-5). The role of GABA in plants is still obscure, whereas its role as an inhibitory neurotransmitter in animals is well understood (1,2).

GABA is involved in the regulation of cardiovascular functions such as blood pressure and heart rate, and plays a role in the sensations of pain and anxiety (6). Many neurological disorders, such as seizures, Parkinson's disease, stiff-man syndrome, and schizophrenia have been shown to be related to alterations of the GABA and glutamate decarboxylase (GAD) levels in the brain (6,7). Alcoholics have remarkably low plasma GABA concentrations and reduced expression of GABA receptor proteins (6).

Recently, there has been increased interest in the utilization of GABA as a bioactive plant component. Several lines of evidence suggest that plant extracts containing high levels of GABA are effective for blood pressure regulation (8,9) and in the recovery from alcohol-related symptoms (9,10).

Saengsik is a simple type Korean food product made by mixing several uncooked plant foods such as: cereals, legumes, fruits, vegetables, mushrooms, seaweeds, etc. Total cholesterol and triglyceride are significantly lower in the serum of vegetarians who eat uncooked foods than in non-vegetarians (11). It has been known that the uncooked foods contain bioactive polysaccharides such as: arabinoxylan in cereals, arabinogalactan in legumes, pectin in fruits and vegetables, γ-glucan in mushrooms, and fucoidan in seaweeds (12). However, few attempts have been made to investigate bioactive non-protein amino acids such as GABA in uncooked foods (13,14).

We investigated the GABA content of commonly used ingredients in uncooked food products. This study enhances our knowledge about the contents of GABA in uncooked foods and also seeks to provide a database for selection of ingredients in the manufacture of uncooked foods containing a high concentration of GABA.

MATERIALS AND METHODS

Materials

γ-Aminobutyric acid (GABA) standard was obtained from Sigma Chemical Co. (St. Louis, USA). Uncooked foods were supplied by El-Chitosan Korea Co., Ltd. in Jeonju, Korea. The selected foods included: cereals, vegetables, mushroom, fruits, seaweed, and functional foods. All other reagents were purchased from commercial sources

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and were of the highest grade available.

Extraction and analysis of GABA

GABA was extracted using the procedures of Baum et al. (15) with modifications as previously described (16). The sample powders (~200 mg) were resuspended in 800 μL of a mixture of methanol : chloroform : water 12 : 5 : 3 (v/v/v). The mixture was vortexed and then centrifuged at 13,000×g at 4°C for 15 min. The supernatant was collected and then 200 μL chloroform and 400 μL water were added to the pellet. The resulting mixture was vortexed and centrifuged for 15 min at 13,000×g. The supernatant was collected and combined with the first supernatant and recentrifuged upper phase collected. The collected samples were dryed in a freeze dryer and redissolved in water. The resulting samples contained the GABA and other amino acids. Each sample was passed through a 0.45 μm filter and analyzed by HPLC (Waters, USA) after 6-amino-quioly-N-hydroxysuccinimidyl carbonate (AQC) derivatization.

Data analysis

Each sample was determined in duplicate and GABA content was calculated by using the program of Young-Lin Autochro WIN and was expressed as an average value.

RESULTS AND DISCUSSION

GABA content of cereals

GABA concentrations of cereals are shown in Table 1. Brown rice germ, brown rice sprouts, and brown rice contained 718, 389, 123 nmole GABA per g dry weight (DW), respectively. Barley sprouts and barley contained 326 and 190 nmole GABA per g DW, respectively. The concentrations of GABA in bean sprouts, beans, and corn were 302, 250, 199 nmole per g DW, respectively. As shown in Table 1, brown rice germ contains relatively high concentrations of GABA compared to other samples, and sprouted cereals contain more GABA than normal cereal samples. Previously, we demonstrated that the germination of brown rice in chitosan or glutamic acid solution increases GABA about 8-fold above non-germinated brown rice (14,17). By germinating the brown rice in a chitosan/glutamic acid solution, 2,011 nmole GABA/g fresh weight was obtained, which is 13-fold increase above non-germinated brown rice (18). Yun et al. (13) demonstrated that the GABA content of germinated barley can be increased by an anaerobic treatment.

Table 1. GABA content of cereals

Foods	GABA (nmole/g DW[1))
Brown rice germ	718
Brown rice sprout	389
Brown rice	123
Barley sprout	326
Barley	190
Bean sprout	302
Bean	250
Corn	199
Oatmeal	98
Adlay	55

[1)]DW means dry weight.

GABA is produced by the enzymatic α-decarboxylation of glutamate; the activity of glutamate deccarboxylase (GAD) is increased during germination (14,18). For example, germination of brown rice in a glutamic acid solution, especially when combined with chitosan, increases GAD activity and GABA accumulation more than the other organic acids, even with chitosan (18). These results suggest that glutamic acid, the precursor of GABA, and chitosan, an elicitor of plant cells, synergistically increase GAD activity and GABA synthesis (18). Overall, these results suggest that brown rice germ and sprouted cereals are good sources of plant GABA, and that germination of uncooked cereals under specific conditions can further enhance the concentrations of GABA in foods.

GABA content of vegetables

GABA concentrations in vegetables are shown in Table 2. Spinach, potatoes, sweet potatoes, yams, and kale contained 414, 166, 137, 129, 122 nmole GABA per g dry weight (DW), respectively. Other vegetables: broccoli, squash, carrots, onions contained less than 100 nmole per g DW of GABA. Previously, it was reported that radish leaves contain about 1,000 nmole GABA per g fresh weight (19). They also demonstrated that the GABA content of the radish leaves can be increased to 4,000~5,000 nmole per g fresh weight by an anaerobic treatment for 6 hr (19). Recently, we demonstrated that freeze-dried Chinese cabbage leaf and root contain 4,690 and 7,020 nmole of GABA per g DW, respectively (20). Further, the Chinese cabbage diet decreased liver triglycerides and total lipids, and blood LDL-cholesterol and γ-GTP levels that were increased due to the chronic ethanol administration (20). There is a substantial body of literature demonstrating that large amounts of GABA accumulates rapidly in many plants under several environmental stress conditions, such as mechanical stimulation, damage, cold shock, heat shock, hypoxia, cytosolic acidification, darkness, water stress, phytohormones, and drought stress (3, 4,21). It has been suggested that GABA accumulation in plant tissues may have a role in pH regulation (21), N storage (22), plant development (15), and in the plant's defense against phytophagous insects (23). Therefore, the levels of GABA in uncooked vegetables can be drastically increased depending upon treatment conditions. Although, GABA is stable at high temperatures, anaerobically treated

Table 2. GABA content of vegetables

Foods	GABA (nmole/g DW)
Spinach	414
Potato	166
Sweet potato (tuberous root)	137
Yam	129
Kale	122
Broccoli	77
Squash (ripen)	43
Carrot	28
Onion	12

Table 3. GABA content of fruits, mushroom, seaweed and lactobacillus

Foods	GABA (nmole/g DW)
Chestnut	188
Apple	2
Lentinus edodes	44
Green laver	37
Lactobacillus	12

samples at low temperature, for example, freeze-dried samples will have more GABA than samples treated at room temperature (20).

GABA content of fruits, mushroom, seaweed and lactobacillus

GABA concentrations in fruits, mushroom, seaweed and lactobacillus are shown in Table 3. Chestnuts, lentinus edodes, green laver, and apples contained 188, 44, 37, 2 nmole GABA per g dry weight (DW), respectively. GABA concentrations in fruits and vegetables are typically low (24). As shown in Tables 2 and 3, the levels of GABA in apples, squash, carrots, broccoli, and onion are very low (ranging from 2 to 77 nmole per g DW), but chestnut contained relatively high amounts of GABA. The GABA concentration of lactobacillus was 12 nmole per g DW. Attempts to increase GABA concentrations by culturing lactic acid bacteria with sodium glutamate have been conducted (24,25). Sodium glutamate, the precursor of GABA, is a substrate for glutamate decarboxylase (GAD). Kim et al. (24) used *Lactobacillus hilgardii* K-3 isolated from kimchi and sodium glutamate enriched media to produce a lactobacillus-fermented extract containing a high amount of GABA. Strains of *Lactobacillus sakei* and *Lactobacillus brevis* producing high amounts of GABA were also isolated from *kimchi* (25). These lactobacillus strains produced over an 80% GABA yield from added sodium glutamate (3.0%) in the growth media. These results suggest that lactobacillus-fermented extracts are potentially rich sources of GABA in uncooked foods.

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