Research paper

Tartary, but not common, buckwheat inhibits α -glucosidase activity: its nutritional implications.

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ABSTRACT

This study shows the presence of α -glucosidase inhibitor in Tartary buckwheat. The present study shows that standard Tartary buckwheat flour exhibited a high level of α -glucosidase inhibitory activity, whereas newly-developed Manten-kirari var., which exhibited a very low level of rutin-degrading enzyme, exhibited no α -glucosidase inhibitory activity. On the other, no α -glucosidase inhibitory activity was found in common buckwheat flour. Nutritional implications of these findings in view of diabetes mellitus prevention were discussed.

INTRODUCTION

Life-style diseases such as diabetes mellitus are a current, major nutritional problem globally. Prevention from these diseases is a subject of much interest in nutrition science. Much attention in food components, which, if any, might be effective for preventing from such life-style diseases, will be paid. **Buckwheat** (Fagopyrum spp.) is an important crop in many countries of the world (Ikeda, 2002; Kreft et al., 2003). There is a large variety of buckwheat products globally. There are two species of cultivated buckwheat, i.e., buckwheat common and Tartary buckwheat. R. Lin and his group (1992) showed that Tartary buckwheat, but not common buckwheat, lowers blood sugars in patients suffering from diabetes mellitus and that Tartary buckwheat flour also lowers serum lipid in patients suffering from hyperlipidemia (Lin et al., 1992). However, the exact mechanisms responsible for the observed beneficial effects for prevention of diabetes mellitus and hyperlipidemia remains uncertain. α-Glucosidase is a major enzyme responsible for the

MATERIAL AND METHODS

Material

Two different types of Tartary buckwheat (*Fagopyrum tataricum* (L.) Gaertn.) were used in this study. One type of Tartary buckwheat, harvested in China, was used

gastrointestinal digestion of saccharides into glucose. α-Glucosidase inhibitor is used as drug curing diabetes mellitus. Although there are many factors involved in prevention from diabetes mellitus, aglucosidase inhibitors in foods, if any, might inhibit the intestinal absorption of glucose, so maybe leading to the prevention of diabetes mellitus. In this connection, increasing attention in polyphenols present in red wine and various plant foods for beneficial effects in human health is currently paid (Renaud and De Lorgeril, 1992). It is well known that buckwheat, especially Tartary buckwheat, contains a high level of polyphenols such as rutin and quercetin. Polyphenols in common and Tartary buckwheat may have profoundlybeneficial effects on human health.

This study was undertaken to identify α glucosidase inhibitor in Tartary buckwheat in view of clarifying a factor responsible for the report by Lin's group showing that the intake of Tartary buckwheat lowered blood sugar in patients suffering diabetes mellitus.

and was abbreviated as the standard Tartary buckwheat. Another type of Tartary buckwheat (var. Manten-Hikari) (HARC, 2017), which was bred by the group of Hokkaido Agriculture Research Center, was obtained. Since this Tartary buckwheat seed (var. Manten-kirari Tartary buckwheat) has less or substantially no rutin-degrading enzyme (HARC, 2017). Therefore, this Tartary buckwheat contains a high level of rutin but lower level of quercetin. Common buckwheat (Fagopyrum esculentum Moench, var. Hashikami-wase), was used in this study. These buckwheat seeds were separately milled with a Brabender Quadromat milling machine with 231 µm sieving. Each flour obtained was subjected for analysis. Rice flour and wheat flour used were commercial products. Catechin and tannic acid, which were obtained from Nakalai Tesque Co., Kyoto, Japan, were of analytical grade.

Extraction

Buckwheat flour samples were extracted with 80% methanol for 1 h at room temperature with rotating. After extraction, the suspensions were centrifuged at 10,000 rpm for 10 min. After centrifugation, the supernatants obtained were subjected for analysis.

Enzyme assay

α-Glucosidase assay was performed with maltose as the substrate according to the method described (Bergmeyer, 1974). Glucose released from the enzyme assay was analyzed with a commercial enzyme assay kit with glucose oxidase plus peroxidase. Inhibition assay for aglucosidase was determined as follows: sample solutions tested were preincubated with enzyme solutions for 10 min at 37°C After pre-incubation, remaining enzyme assay was determined with the substrate at 37°C for 30 min. Inhibitor constants (Ki) were estimated by the Lineweaver-Burk plot.

Statistical analysis

Statistical analysis was conducted using a personal computer with the program Excel (Microsoft Co., USA).

RESULTS AND DISCUSSION

Inhibitory activity against α -Glucosidase by methanol extract of Tartary buckwheat flour



Fig. 1 Effect of Tartary and common buckwheat extracts on the activity of α-glucosidase. (A), common buckwheat extract; and (B), Tartary buckwheat extract.

Figure 1 shows the effects of methanol of common extracts and Tartary buckwheat flours. As shown in Fig. 1, the methanol extract of Tartary buckwheat exhibited inhibitory activity against a-Glucosidase, whereas the methanol extract of common buckwheat exhibited less inhibitory activity against this enzyme. The effect of heating for inhibitory activity against α-glucosidase by the methanol extract of Tartary buckwheat flour was examined. Heating did not influence the inhibitory activity by the methanol extract of Tartary buckwheat flour (data not shown). This finding shows that the observed inhibitory activity against α -glucosidase by the methanol extract of Tartary buckwheat flour may be due to heat-stable components such as polyphenols present in buckwheat, but not to heat-unstable proteinaceous components.

Inhibitory activity of various polyphenols

It is well known that common and Tartary buckwheats contain various kinds of polyphenols such as rutin and quercetin. We tried to assay some polyphenols against α -glucosidase. Table 1 shows the inhibition constant, as the Lineweaver-Burk plot method, of some polyphenolics against α-glucosidase activity. As shown in Table 1, quercetin exhibited extremly high inhibitory activity with very low inhibition constant, so indicating that quercetin may be a strong inhibitor against α -glucosidase. Tannic acid also exhibited inhibitory activity against α -glucosidase. On the other hand, no inhibitory activity against this enzyme was found with rutin and catechin. The observed inhibitory behavior (Table 1) with quercetin and rutin was very important as discussed below.

Tahle	1	Inhibition	constant	٥f	various	nolynhend	hle	anainst	n-ul	ucosidase
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Components	Inhibition constant ¹⁾	
Quercetin	1.68 X 10 ⁻⁶ M	
Tannic acid	4.34 X 10 ⁻⁷ M	
Rutin	No inhibition ²⁾	
Catechin	No inhibition ²⁾	

¹⁾ Inhibition constant was estimated by the Lineweaver-Burk plot method.

²⁾ The word "no inhibition" means that no inhibition against α-glucosidase was found under the assay conditions used.

No inhibitory activity against $\alpha\mbox{-glucosidase}$ by the Manten-kirari Tartary buckwheat



Fig. 2 Effect of Tartary buckwheat var. Manten-kirari extracts on the activity of α-glucosidase.

Figure 2 shows the effect of the Mantenkirari Tartary buckwheat extract against α glucosidase. No inhibitory activity against α -glucosidase was found with the Mantenkirari Tartary buckwheat. The Manten-kirari Tartary buckwheat exhibits no rutindegrading enzyme. Therefore this Tartary buckwheat contains a high level of rutin, which exhibited inhibitory activity α -glucosidase (Table 1), and contains less or no quercetin which have inhibitory activity (Table I).

α-Glucosidase is a major enzyme responsible for the gastrointestinal digestion of saccharides into glucose. α-Glucosidase inhibitor is used as a medicine curing diabetes mellitus through inhibition of intestinal absorption of ingested glucose. Although there are many factors involved in the prevention from diabetes mellitus, α glucosidase inhibitors in foods, if any, might inhibit the intestinal absorption of glucose, so maybe leading to the prevention of diabetes mellitus.

This study concludes that quercetin may be a factor responsible for the report by Lin's group showing that the intake of Tartary buckwheat lowered blood sugar of patients suffering diabetes mellitus.

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