Effects of Combined Administration of Quercetin, Rutin, and Extract of White Radish Sprout Rich in Kaempferol Glycosides on the Metabolism in Rats

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Quercetin, rutin, the extract of white radish sprout rich in kaempferol glycosides, and their combination were intragastrically administered to Wistar rats to investigate the interactive metabolism of these flavonoids. The combined administration of these flavonoids changed the concentrations of the metabolites in plasma as compared with the concentrations after the administration of a single compound.

Key words: rutin; quercetin; kaempferol; white radish sprout; isorhamnetin

The metabolic analyses of flavonoids have been significantly studied around the world, because of their preventive effects on degenerative diseases. Regular diets contain a variety of compounds including flavonoids such as quercetin- and kaempferol-glycosides, and some constituents may interfere or promote the metabolism of the others. Study on the metabolic analysis of a single compound is unavailable that reflect the metabolism of the dietary constituents in our regular diets. In the present study, the interactive effect of dietary constituents on the metabolism was investigated using quercetin, rutin, and the extract of white radish sprout (Raphanus sativus L.) rich in kaempferol glycosides.

Quercetin and rutin (quercetin-3-0-rutinoside) were purchased from Nacalai Tesque (Kyoto, Japan) and Wako Pure Chemical Industries (Osaka, Japan), respectively. The extract of the white radish sprout was prepared using 90% methanol, and the amount of kaempferol glycosides was determined by HPLC as kaempferol aglycone following acidic hydrolysis as previously described.1) Quercetin, rutin, and the extract of white radish sprout were dissolved in 0.1% carboxymethylcellulose for intragastric administration.

All animal treatments in this study conformed to the “Guidelines for the care and use of experimental animals, in Rokkodai Campus, Kobe University”. Male Wistar rats (7 weeks old, purchased from Japan SLC, Shizuoka, Japan, n = 3) were intragastrically administered quercetin (50 mg/kg body weight (B.W.) equal to 148 μmol/kg B.W.), rutin (100 mg/kg B.W. equivalent to 50 mg quercetin aglycone/kg B.W.), a combined mixture of quercetin and rutin (25 and 50 mg/kg B.W., respectively, equal to 74 μmol kaempferol glycosides/kg B.W.), or a mixture of rutin and the extract of white radish sprout (100 mg/kg B.W. of each). The venous blood was collected from the tail. The flavonoid metabolites in the plasma were deconjugated to the aglycone form with /C12-glucuronidase/sulfatase according to the method of Azuma et al.2) After extraction with acetonitrile, the concentrations of aglycones were determined by HPLC equipped with an amperometric electrochemical detector (Nanospace SI-2, Shiseido, Tokyo, Japan) at +800 mV as previously described3) with a slight modification. An experiment using control plasma spiked with a quercetin standard showed the 95% recovery for quercetin.

After the administration of quercetin (50 mg/kg B.W.), the plasma concentrations of the quercetin- and isorhamnetin (a methylated form of quercetin)-conjugates rapidly increased, and reached peaks of 4.1 ± 0.2 and 0.39 ± 0.11 μM, respectively, at 2 h (Fig. 1A). Both concentrations then decreased and diminished at 12 h. On the other hand, 2 h after the administration of rutin (100 mg/kg B.W.), the plasma concentration of the quercetin- and isorhamnetin-conjugates slightly increased (Fig. 1B). These conjugates then increased to 20.4 ± 4.1 and 13.5 ± 2.9 μM, respectively, at 12 h, the end time

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Abbreviation: HPLC, high performance liquid chromatography
shown in Fig. 1A. These conjugates further increased (Fig. 1C) as well as the metabolism of the singly administered rutin as shown in Fig. 1B. The profiles of the conjugates detected early might belong to the quercetin in the mixture, and that of the conjugates detected late might belong to the rutin in the mixture. The variations in the concentrations of quercetin- and isorhamnetin-conjugates in the plasma within 6 h were almost the same as the sum of the variations in the singly administered quercetin and rutin. The quercetin- and isorhamnetin-conjugates further increased to 20.4 ± 4.1 and 13.5 ± 2.9 μM, respectively, 12 h after the administration of rutin alone (Fig. 1B). Thus, quercetin and rutin were constantly absorbed until 6 h. On the other hand, 12 h after the administration of the mixture, the quercetin conjugates in the plasma decreased to 5.6 ± 2.3 μM (Fig. 1C). This decrease might have been due to the decrease in the amount of rutin-derived quercetin absorbed from the intestine.

After the administration of the white radish sprout extract rich in kaempferol glycosides (100 mg/kg B.W.), the kaempferol metabolites in the plasma slightly increased to 0.14 ± 0.03 μM at 2 h, then increased to 3.8 ± 0.6 μM at 6 h, and decreased to 1.7 ± 0.3 μM at 12 h (Fig. 2A). On the other hand, when the extract was intragastrically administered with rutin (100 mg/kg B.W.), as shown in Fig. 2B, the kaempferol metabolites in the plasma increased up to 6 h (3.3 ± 0.3 μM) as well as that in the singly administered extract (Fig. 2A). However, 12 h after the administration of the mixture, the kaempferol metabolites in the plasma increased to 8.2 ± 4.1 μM. On the other hand, the quercetin conjugates in the plasma were lower than that after the administration of the single rutin, i.e., 4.1 ± 0.3 μM at 6 h and 10.7 ± 3.5 μM at 12 h after the administration of the mixture (Fig. 2B), and 8.7 ± 1.8 μM at 6 h and 20.4 ± 4.1 μM at 12 h after the administration of rutin alone (Fig. 1B). These results indicate that the metabolism of rutin and the kaempferol glycosides interact with each other. de Vries et al.7 demonstrated that the absorption of quercetin from tea containing quercetin- and kaempferol-glycosides was half that from onion containing only quercetin glycosides. This supports our data that the metabolism of rutin was suppressed by the white radish sprout extract rich in the kaempferol glycosides. It is unknown whether the antagonistic compound in the extract against the metabolism of rutin is the kaempferol glycoside, because the extract includes not only the kaempferol glycosides (89 μmol/100 g fresh edible part) but also other compounds such as cinnamic acids (197 μmol/100 g). On the other hand, our data indicated that the metabolism of the kaempferol glycosides was promoted by the co-administration with rutin. There is also the possibility of additive, synergistic, and antagonistic interactions among the different constituents in the white radish sprout extract.

In a human study, kaempferol derived from endive was reported to be absorbed from the distal section of

![Fig. 1. Quercetin Metabolites in Plasma of Rats after Intragastric Administration of Quercetin and Rutin.](image-url)
the small intestine and the colon. This indicates the possibility that kaempferol glycosides interact with rutin, which is also absorbed from these sections, but their interactive absorption mechanism was unclear. A recent study demonstrated that the simultaneous administration of quercetin and catechin aglycones altered their absorption and decreased the concentration in the plasma. To elucidate the interactive metabolism of rutin and kaempferol glycosides, the sugar and the deglycosidases are also required to be identified.

The present study indicates that the administration of the same kinds of flavonoids shows their reasonable absorption. However, the administration of flavonoids with vegetable extracts altered their absorption, because of the interactive metabolism of the flavonoids and the other constituents in vegetables. Not only the metabolism of a singly administered flavonoid, but also the metabolism of flavonoids in complexes should be discussed in order to understand the metabolism of dietary constituents in our regular diets and the preventive effects of the diets on degenerative diseases.

References