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"Kuro-mame," which belongs to Glycine Soya Benth, is given the special name because of its delicacy and of the specially deep colour of the seed coat. As it is favoured as food and medicine, the chemical study of its colouring matter is of great importance as well as of much interest. However no literature has been found upon the subject.

As the result of qualitative tests made by the authors it was found that this colouring matter belongs to the anthocyan group and therefore the investigation was undertaken accordingly, first starting from the water soluble portion which is the main constituent of the colouring matter. From the reddish-purple aqueous solution obtained by leaving the beans immersed in cold water, bluish-green precipitate was formed on addition of aqueous lead acetate solution, which was then filtered, dried at room temperature, and powdered (yield: 0.5% of beans).

When the above lead salt was treated by cold methyl alcoholic hydrochloride, a red solution was obtained, and the chloride of the colouring substance was precipitated by addition of ether (yield: 9% of lead salt).

The precipitate was purified into a crystalline-like state by adding ether into its absolute methyl alcoholic solution. When separated and dried, it yielded reddish-purple powder which was further purified either by the repetition of this process or by the addition of hydrochloric acid into its aqueous solution. As the compound thus obtained is a glucoside, it was decomposed by the following method: the substance (1 g) was dissolved in 0.7% warm dilute hydrochloric acid (30 c.c) and into the solution conc. hydrochloric acid (28 c.c) was added; the mixture was then boiled for 3 minutes. After cooled, a reddish brown product crystallized out (yield: 0.6 g). When purified by ethyl or methyl alcoholic hydrochloride, crystals were obtained in plates.

1) Kuro means black and mame, bean in Japan.
2) The sample used was from Shiriuchi Hokkaido.
Analytical results agreed with cyanidin chloride \( \text{C}_{15}\text{H}_{11}\text{O}_6 \text{Cl H}_2\text{O} \) and the qualitative reactions were identical.

The above consideration was also confirmed by decomposition products, namely phloroglucin and protocatechuic acid. After removing the aglucon, the aqueous hydrochloric acid solution (the ethereal extract of the solution showed only a trace of a green colouration with ferric chloride) was neutralized and the formation of osazone was carried out, the product melted at 203° corresponding with glucosazone, the mixed melting point showing no depression; and the analytical result was also in agreement. After alkali fusion of the glucoside (1.5 g) the reaction product was dissolved in water, acidified with hydrochloric acid, neutralized with sodium bicarbonate, and extracted with ether. On evaporation the residue was recrystallized from water with the aid of animal charcoal. When recrystallized from dilute methyl alcohol and dried in the air, the product was identical with phloroglucin (M. P. 205°), giving no depression in admixture with the authentic sample. Analysis and qualitative tests were also in agreement with the result. Upon removing the phenol, the bicarbonate solution was acidified, extracted with ether, and the brown residue obtained was purified by water. The compound melted at 193° agreeing with protocatechuic acid, giving a green colouration with ferric chloride. To glucoside (1 g) dissolved in aqueous solution of sodium carbonate (70 c.c), 3% hydrogen peroxide (70 c.c) was added. After left for a few hours, it was acidified and extracted with ether. The residue of evaporation was recrystallised from water and quickly washed with ether and dried in the air for analysis. The analytical result, melting point (193° C alone or with an authentic specimen), and colour reaction with ferric chloride agreed with protocatechuic acid.

From these studies it is certain that the water-soluble colouring matter in the seed coat of “Kuro-mame” contains Cyanin. Further studies are in progress regarding the sugar and other colouring matter of the bean etc.

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