Original article

Effects of cassava on thyroid gland in rats

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Abstract:

The root of cassava has been consumed as a food in many tropical countries. In pharmaceutical industry, cassava starch can be used as a binder in tablet formulation. Since cassava contains high concentration of linamarin, a cyanogenic glucoside which can be metabolized to thiocyanate, it has been suspected as a cause of endemic goiter. However, controversial results from various study indicated the unclear effect of cassava on goiter. The main objective of this study is to determine the effect of cassava ingestion on the thyroid gland in Wistar rats. The rats were fed with different diets which were fresh cassava root, steamed cassava flour, and standard (control) diet for the duration ranging from 1 day to 3 weeks. The results showed that the serum thiocyanate concentration was significantly elevated in the rats fed with fresh cassava root whereas the rats fed with steamed cassava flour showed the similar level of serum thiocyanate as compared to the control group. Both fresh cassava root and steamed cassava flour did not cause significant thyroid gland enlargement as compared to the standard diet. These findings suggest that the elevation of serum thiocyanate from fresh cassava root ingestion may be insufficient to cause goiter in Wistar rats. The results also showed that steamed cassava flour did not increase serum thiocyanate level compared to the control group, indicating that most of the cyanogenic glucosides were eliminated during the preparation process.

Key words: Cassava; Goiter; Thiocyanate; Thyroid gland

Introduction

Cassava (*Manihot esculenta* Crantz), a shrubby tree of the Euphorbiaceous family, is an extensively cultivated plant in many tropical countries. Cassava root is a major source of dietary carbohydrate for human and livestock. Several plants, used as food, contain cyanogenic glucosides; these glucosides when ingested release hydrocyanic acid (HCN) which is one of the most potent cytotoxins. Linamarin is a cyanogenic glucoside that found in high concentration in cassava. There is also a small amount of another glucoside, lotaustralin [1-2]. In cassava, HCN is most easily liberated by the linamarase enzyme, which also contained in the plant. The contact between linamarin and the enzyme is sufficient to initiate the hydrolysis of the glucoside and liberation of HCN [3]. Although the relation of iodine deficiency to goiter is well established, other factors may involve. Various natural agents such as cyanogenic glucoside and thiocyanate have been identified that they might be goitrogenic [4-5]. The goitrogenic action of cassava and the effect of thiocyanate on thyroid function were reported [6]. Although cassava does not contain thiocyanate or its precursor; the degradation of the linamarin results in HCN which is internally metabolized by the body to less toxic substances, principally thiocyanate [7-8]. It is widely accepted that plants grown in various location produced different quality and quantity of substances. Thus, the cyanogenic glucoside content in cassava may be affected by the plant location. Until now, the antithyroid effect of cassava is still unclear, since several controversial effects of cassava on thyroid gland have been reported [1, 9].

Known cases of acute HCN poisoning from the consumption of cassava are rare, probably because preparation process of cassava for consumption can destroy the linamarase and remove much of the free HCN. Nevertheless, the possibility of chronic toxicity has not been eliminated [10-12]. Due to the toxic cyanogenic glucoside, several processing methods have been used to process fresh cassava root in order to reduce the cyanide content. The methods include grating, sun drying, ensiling, fermentation, oven drying, and boiling [13-18]. Previous studies reported that ensiling and fermentation are the most effective ways to eliminate cyanogenic substances whereas oven drying method is the least effective way [13, 15, 16]. The residual level of cyanogenic glucoside in processed cassava would therefore depend upon the processing method used. Determination of cassava toxicity is of interest, since cassava cultivated in Thailand is considerably achieved in agricultural importance as one of the major exports. Cassava is not only considered to be a good source of carbohydrate as a diet, but also has been used in pharmaceutical industry as a binder for tablet formulation [19, 20]. Besides the unclear goitrogenic effect of cassava, there is still endemic goiter in the North-East rural area of Thailand. In the present study, two experiments were conducted to determine the effects of fresh cassava and steamed cassava on the anatomy of thyroid gland and serum thiocyanate concentration. In an attempt to facilitate the use of this economic plant, this study may lead to an increase of cassava usage with safety in food and pharmaceutical industries.

Materials and Methods

Animals

The nine-week-old albino Wistar rats, weighed between 190 and 250 g, were obtained from the National Laboratory Animal Center, Mahidol University, Nakornpatom province, Thailand. The rats were housed in a temperature and light control room.

Diets

Ten groups of eight rats were fed for 1-21 days on different diets, which were (1) fresh cassava root; unpeeled fresh cassava root (from Rayong province, Thailand) cut into small pieces, weighed 2 g per piece approximately, (2) steamed cassava; processed cassava flour (pellet pearl tapioca), steamed for about 20 minutes, and (3) standard diet for rat purchased from F.E. Zuellig (Bangkok) Ltd.

The weight of dehydrated cassava root without fiber was approximately the same as processed cassava flour [21].

Experimental design

Two experiments were performed.

Experiment I: Sixty four rats were divided into two groups. One group was fed the fresh cassava root (15 g/day) and the control group was fed the standard diet (15 g/day). Each group were divided into 4 subgroups (n=8/each) which were fed for 1, 4, 7 and 21 days, respectively.

Experiment II: Two groups of eight rats were fed the following diets for 7 days. (a) fresh cassava root 15 g/day plus standard diet 10 g/day, (b) steamed cassava 10 g/day plus standard diet 10 g/day. For the control group, the serum thiocyanate and thyroid gland data were obtained from the experiment I at the seventh day.

At the end of the experimental period, rats were bled by laparotomy under diethyl ether anesthesia and venepuncture of the inferior vena cava. Measurement of serum thiocyanate was determined by the method originally described by Schreiber [22] and modified by Chesley [23]. Optical absorption was determined at 470 nm by spectrophotometer. The thyroid glands were examined for general appearance, then weighed and collected for histological examination.

Statistical analysis

The data were descriptively presented using mean ± S.E.M. The concentration of serum thiocyanate and the weight of thyroid glands were compared across the different feeding groups by independent t-test or one-way ANOVA. Differences of the means were considered to be significant when p-value < 0.05.

Results

Effects of fresh cassava root on serum thiocyanate concentration and thyroid gland

Eight subgroups of rats were fed with fresh cassava root or standard diet. The results showed that the serum thiocyanate concentration in rats from the groups which were fed with fresh cassava root for 4, 7, and 21 days was significantly higher than that in the control groups ($p < 0.05$) as shown in Figure 1.

The ingestion of fresh cassava roots did not significantly modify the thyroid gland weight of the rats as compared to the controls ($p > 0.05$) as shown in Table 1. The general appearance of the skin, eyes, tails and thyroid glands of all groups were normal. Histological examination of the thyroid glands was normal; there was no evidence of hypotrophy or hyperplasia of the thyroid glands.

Table 1 Effects of fresh cassava and standard diets on weight of the thyroid gland in rats. Data are presented as mean ± S.E.M. (n = 8).

Duration of Feeding	Weight of thyroid gland (mg/kg body weight)		
(Day)	Standard diet	Fresh cassava	*P*-value
1	89.99 ± 2.13	89.56 ± 1.73	0.877
4	90.05 ± 2.48	86.54 ± 1.63	0.257
7	90.41 ± 2.80	91.04 ± 6.31	0.929
21	91.99 ± 2.53	89.22 ± 0.45	0.298

Effects of fresh and steamed cassava on serum thiocyanate concentration and thyroid gland

Two groups of eight rats were fed with standard diet supplemented with fresh cassava or steamed cassava for 7 days. The physical examination of both groups showed normal appearance of the eyes, skin, tails and thyroid glands. Histological examination of the thyroid glands was normal and there was no evidence of hypotrophy or hyperplasia of the thyroid glands. Serum analysis showed significant increase of thiocyanate concentration in rats fed with standard diet plus fresh cassava (fresh cassava group) ($p < 0.05$), whereas in rats fed with standard diet plus steamed cassava (steamed cassava group) had no difference ($p > 0.05$) comparing to control group (Figure 2). As expected the data of thyroid gland weight showed no significant difference in either fresh cassava group or steamed cassava group as compared to the control group (Table 2).

Discussion and Conclusion

Several evidences on the pathogenesis of goiter indicated the close association between low iodine content in food and incidence of the disease in the population [4, 24]. Also, other factors have been implicated to be a possible cause of goiter. Due to the association between goiter and the large amount of cassava consumption, then linamarin, a cyanogenic glucoside presented in cassava, was identified and tested for the antithyroid activity [24].

Until now, the goitrogenic effect of cassava is still in controversial. The cyanogenic glucosides, linamarin and small amount of lotaustralin, are absorbed by the body and converted to thiocyanate, which is excreted in the urine [1, 4, 24]. Although thiocyanate is widely accepted to cause reduction in iodine uptake by thyroid gland; other factors affecting serum thiocyanate level including ingestion, detoxification, and excretion are also needed to be concerned. The data reported by Hershman *et al.*[6] indicated that even intake of cassava root could increase urine thiocyanate excretion and serum thiocyanate levels. Surprisingly, the thyroid glands of mice fed with cassava were not enlarged. Moreover, the thyroid iodide trap was similar in mice fed on cassava and low iodine diets [9]. The present study indicated the significant elevation of serum thiocyanate in Wistar rats fed with fresh cassava root without any change of thyroid size. The possible explanation of these cases may be that the thiocyanate produced from ingestion of cassava is insufficient to inhibit thyroid iodide transport or organification of iodide.

The controversial reports may be that the goiter caused by cassava ingestion in some areas may cause by the coincidence with the iodine deficiency. Moreover, the widely accepted concept that the plants cultured in different locations produce different chemical components, suggests that cassava in some areas may contain high level of cyanogenic glucosides which are sufficient to cause goiter.

In order to reduce toxicity of cassava, several processing methods have been proposed. It seem clear according to Tewe *et al.* [8] and Obioha and Anikwe [13] that ensiling and fermentation are the most efficient methods to reduce cyanogenic glucoside content, while oven-drying is the least efficient method. Since widely used cooking method for cassava in Thailand are steaming and boiling, thus the effect of steaming on serum thiocyanate content was investigated. The results clearly indicated that steaming is a good method for eliminating cyanogenic glucoside in cassava. Also, the serum of rats fed with steamed cassava showed similar level of thiocyanate as compared to the rats fed with standard diet. As expected the rats fed with steamed cassava did not show any pathological signs including thyroid enlargement.

Since cassava is a good source of carbohydrate, the present study shows that well processed cassava is safe for consumption. Moreover, many studies reported that cassava starch has a good property for using as a binder in pharmaceutical industry [25]. Hopefully, this study may facilitate the use of this economic plant by providing the safety information.

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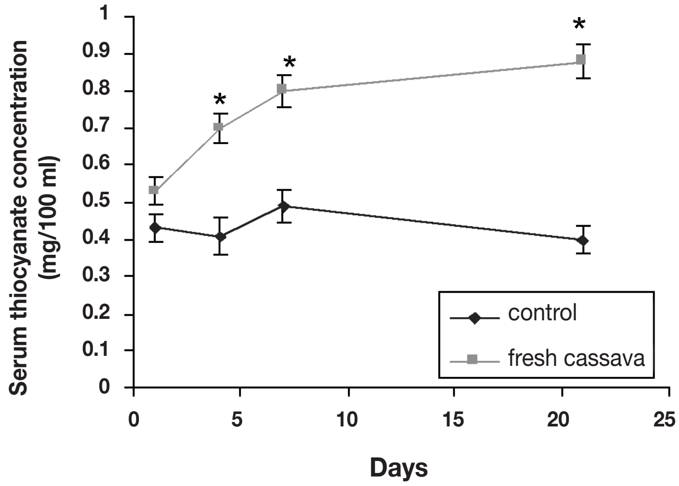

Figure 1 Effects of fresh cassava and standard diets (control) on serum thiocyanate concentration in rats. Data are presented as mean ± S.E.M. (n = 8), * indicates significant at *P* value < 0.05.

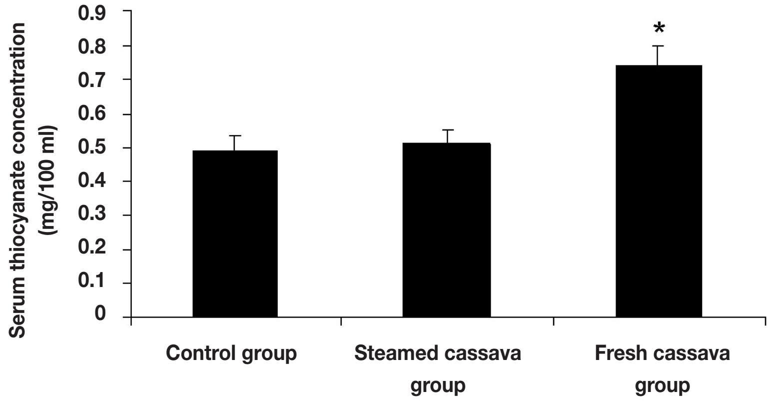

Figure 2 Effects of fresh cassava, steamed cassava, and standard diets (control) on serum thiocyanate concentration in rats, all groups were fed with indicated diets for 7 days. Data are presented as mean ±S.E.M. (n=8), * indicates significant at *P* value < 0.05.

Table 2 Effects of fresh cassava, steamed cassava, and standard diets on weight of the thyroid gland in rats after feeding for 7 days. Data are presented as mean ± S.E.M. (n = 8).

Type of diet	Weight of thyroid gland (mg/kg body weight)	*P*-value
Standard diet	90.41 ± 2.80	
Standard diet supplemented with steamed cassava	90.08 ± 3.58	0.743
Standard diet supplemented with fresh cassava	87.52 ± 2.09	

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