

GOITROGENIC PRINCIPLE FROM CASTOR SEEDS

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Abstract—Ricinine, a principal alkaloid isolated from the castor-cake, has been studied for its antithyroid activity. Its effect on goiter production and thyroid hormonogenesis has been compared with the known goitrogen, propylthiouracil. The data presented herein reveal that ricinine is a potent goitrogen.

Presence of goitrogens in plants was first reported by Chesney *et al.* [1], who observed enlarged thyroids in rabbits fed cabbage (genus: *Brassica*), as a major part of their diet. Related *Brassica* seeds were also found to induce goiters in rats [2]. It was further found that most plants of *Cruciferae* family belonging to *Brassica* genus contain thio-glucosides which are responsible for the observed antithyroid activity [3].

Ricinus communis L, commonly known as castor plant, belongs to *Euphorbiaceae* family. The various parts of the plant comprising root and shoot are used for Ayurvedic and other medicinal preparations as has been detailed elsewhere [4, 5]. Castor oil is commonly used as a purgative in medicine. Besides, the castor-cake is commonly employed in fields as manure and cattle-feed after little detoxification.

One of the principal alkaloids isolated from the castor-cake is ricinine (RCN) which is a cyano compound. 20 mg% RCN is found to be present as dry weight basis of the castor-cake.

RCN is reported to have a number of toxic effects such as vomiting, nausea, hemorrhagic gastro-enteritis etc. [6]. Presence of cyanide group in the structure of RCN (3-cyano,4-methoxy-N-methyl-2-pyridone), prompted us to study its goitrogenic action. The observations made in the present communication do suggest that RCN is a potent goitrogen in rats, and its goitrogenic activity is comparable to other known goitrogens like propylthiouracil (PTU).

MATERIALS AND METHODS

PTU was obtained from Koch-Light Laboratories, England. Iodo amino acids and methimazole (MMI) were purchased from Sigma Chemical Co., U.S.A. Pronase (70,000 PUK/g) was obtained from E. Merck, Germany. Whatman No. 3MM paper was obtained

from Whatman Co. U.S.A. Na¹³¹I was supplied by Isotope Division of Bhabha Atomic Research Centre, Bombay. All the other reagents used were of analytical grade.

Crystalline RCN was isolated from castor-cake by solvent extraction method. Its purity was checked by (i) Melting point, 201.5° [6] and (ii) paper chromatography in two different solvent systems as reported by Bregoff [7]. The structural identity of RCN was further confirmed by u.v., i.r. and n.m.r. spectra.

Young male rats of Wistar strain weighing about 140–170 g were taken and were divided into the following three groups (4 rats/group).

- Control group—received 2 ml saline by stomach tube.
- PTU group—received 2 ml suspension of PTU in saline (10 mg/day) by stomach tube.
- RCN group—received 2 ml suspension of RCN in saline (10 mg/day) by stomach tube.

Oral feeding of the drugs and saline was continued for 3 weeks. During this period all the animals received normal colony diet. The rats were then injected (i.p.) with Na¹³¹I (5 µCi per 100 g body weight). 24 hr after the administration of radio-iodine, the rats were sacrificed under ether anaesthesia. The blood was collected by heart puncture, while thyroid lobes and liver were collected in chilled saline. Blood and liver were processed for protein bound radio-iodine as described by Studer *et al.* [8]. The thyroid lobes were homogenised in Tris-Cl buffer, pH 8.6 (0.05M), containing MMI (0.03M), and the thyroid homogenate was hydrolysed by pronase [9]. The thyroid hydrolysate was further analysed by paper chromatography as reported by Inoue *et al.* [10].

RESULTS AND DISCUSSION

Four-fold enlargement of the thyroid gland was induced by RCN when fed for 3 weeks (10 mg/day); which is quite comparable to the results obtained with the treatment of a known goitrogen, PTU (control thyroid—6 mg; RCN—24.2 mg and PTU—24.8 mg; per 100 g body weight of the rat). The difference in the

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Table 1. Effect of RCN and PTU on thyroid-weight and $PB^{131}I$ in serum and liver

	Thyroid-wt (mg/100 g body wt)	Serum $PB^{131}I$ (cpm/ml)	Liver $PB^{131}I$ (cpm/g)
Control	6.07 ± 0.28	830 ± 34	490 ± 167
PTU	24.80 ± 0.96*	92 ± 50*	79 ± 13†
RCN	24.17 ± 1.91*	97 ± 7.0*	86 ± 14†

Results are mean ± S.D.

* $P < 0.001$ † $P < 0.005$

weight of thyroids from normals and RCN-fed rats is statistically significant ($P < 0.001$).

Table 1 further shows that RCN had also lowered $PB^{131}I$ in serum and liver to the same extent as in case of PTU treatment. A five-fold decrease in $PB^{131}I$ in liver which is known to be a store house for thyroid hormone, further indicated that overall thyroid function was lowered by RCN treatment.

The percent radio-iodine incorporation into iodothyrosines and iodothyronines within the thyroid gland of control and experimental animals is shown in Fig. 1. It is evident that radio-iodine incorporation in iodothyronines is considerably lowered in both PTU and RCN treated thyroids (3 and 4 per cent respectively of the total) compared to that in control (13 per cent of the total). However RCN does permit more organification than PTU, as is reflected by more radio-iodine incorporation in iodotyrosines (RCN—31 per cent and 17 per cent vs PTU—18 per cent and 5.3 per cent; in MIT and DIT respectively). The free radio-iodine in the RCN treated thyroids is also found to be significantly lower (40%) than that in PTU (65%); again suggesting that RCN is not as potent as PTU in blocking organification.

Goiter production in PTU-treated rats appears to be growth-related as it is accompanied by the lesser gain in their body weight, as has been already reported [11]. However the goitrogenic effect of RCN, though comparable to PTU, as far as goitre size is concerned, does not seem to be growth-related as there is no significant change in the body weight of rats compared to that of control. It remains further to be studied whether effects observed by RCN treatment, can be compared with thiocyanate (SCN) which is also known to induce goiters [12].

It is interesting to note that no toxic effects of RCN-ingestion were observed when 10 mg/day of this alkaloid was fed to the rats for 3 weeks. It would be worthwhile to study the cattle, fed on castor-cake, for its goitrogenic action, and how much of ingested RCN is excreted through milk.

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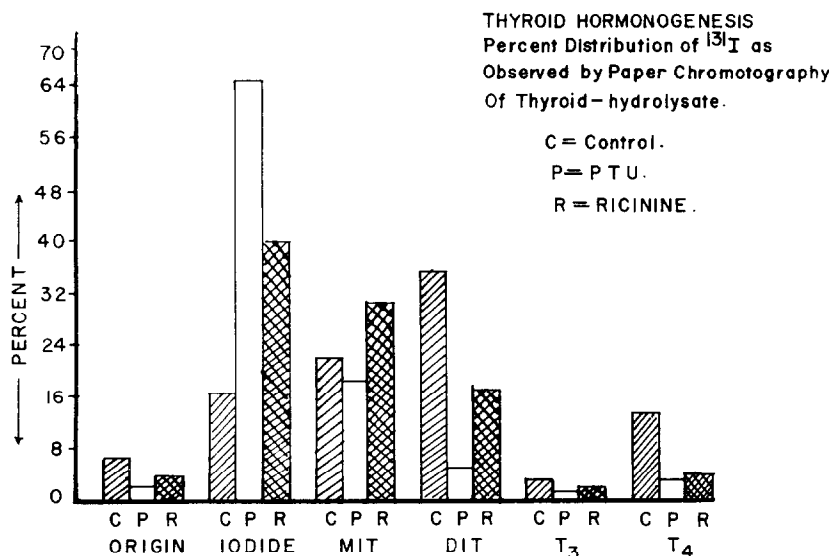


Fig. 1. The per cent ^{131}I in iodo amino acids in the thyroid gland. MIT = Mono iodo tyrosine; DIT = Di iodo tyrosine; T₃ = Tri iodo thyronine; T₄ = Thyroxine.

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