Cassava Diets and Performance of Rats over Two Reproductive Cycles

by

O. O. TEWE, T. A. JOB, J. K. LOOSLI† AND V. A. OYENUGA

*Department of Animal Science
University of Ibadan
Ibadan*

SUMMARY

FRESH and dried bitter (high HCN) cassava, dried sweet (low HCN) cassava and a commercial pellet diet were fed to rats during two life-cycles.

First litter birth weights of rats on fresh cassava diets were significantly ($P < 0.05$) lower than on other diets. Performance of rats on sun dried cassava diets were probably more affected by the fungal growth on the sundried chips, rather than influence of its hydrocyanic acid content.

Histopathological examination of thyroid tissue revealed that thyroidal epithelial heights were significantly ($P < 0.05$) lower in the second litter offspring of the fresh cassava group than in others. Alopecia was also observed in these rats.

INTRODUCTION

The symptoms of chronic cassava toxicity which has hitherto been largely reported in humans, are not only manifested as pathological changes, but also bothers on the metabolic integrity of the organism under such repeated intakes of cassava. These biochemical changes, which may be due either to the direct effect of hydrocyanic acid or its biotransformation products, have been demonstrated to draw on various nutrients in the human or animal metabolic pool, for the accomplishment of the processes which they undergo in these subjects.

Cyanide detoxification causes an increased demand for sulphur containing amino acids (Maner and Gomez, 1973). Sizeable literature has been published on the goitrogenic role of thiocyanate which is the main detoxification product of cy-

anide in animals (Honour, Myant and Rowlands, 1952; Funderburk, 1966; Ekpechi, Dimitriadou and Fraser, 1966; Delange, 1974). This evidence therefore calls for a need to elucidate the role of the cyanogenetic glucoside fraction of cassava in the long term feeding of animals. This study was aimed at investigating the effect of cassava in its fresh and dried forms on the performance of rats over two reproductive cycles. Histological studies were also carried out on the thyroid tissue as it has been established that this organ affects the general metabolism and growth promoting developmental action in animals (Turner and Bagnara's 1971).

EXPERIMENTAL PROCEDURES

Twenty Sprague-Dawley strain female rats having an average body weight of 179g and age of about 100 days were randomly allotted on a weight basis to four experimental groups. The gross and proximate composition of the dietary variables are shown in Tables 1 and 2. The proximate composition of fresh and sun-dried cassava of the two varieties utilized are also shown in Table 3.

The fresh cassava diet was prepared by grating fresh tubers daily and mixing this thoroughly with a supplement containing fish meal, rice bran, groundnut oil, mineral and vitamin mix to provide 20% crude protein diet on a dry matter basis. The fresh cassava (Oyanrugbapupa variety) had an average HCN content of 402.3 mg/kg on a fresh basis while the dried meal of the Oyanrugbapupa and Isunikan-

†-Present Address: Department of Animal Science, University of Florida, Gainesville, Florida 32607, U.S.A.

kiyan varieties contained 57.6 and 12.6 mg/kg HCN on air dry basis respectively.

Management during gestation and lactation

The female rats in each treatment were group fed in breeding cages with a dimension of 80 cm by 30 cm. Two male rats were also run with each group of female rats using one of the two males every other day, thus allowing a day resting period on alternate days. The feed intake of the male and female rats were also recorded daily on the fresh diet and weekly on the others.

Feed and water were offered *ad libitum*. Pregnant rats were identified visually and individually kept in 40 x 20 cm. wire cages where they were offered their respective diets and water *ad libitum* throughout the remaining part of the gestation and lactation periods.

Management during the post-weaning growth phase

Six weaning rats, three of each sex, were selected from each treatment group and individually maintained in 25 x 20cm wire cages for a 28-day growth trial.

TABLE 1

Gross composition of cassava diets fed to rats in gestation, lactation and post-weaning growth

Ingredient	%
Cassava*	65.0
Fish meal	22.5
Rice bran	3.0
Groundnut oil	5.0
Mineral Mix**	4.0
Vitamin Mix**	0.5

* The percentage of cassava in the diet is expressed on 90% dry matter basis. Correction was therefore made for the moisture content of fresh cassava.

** The mineral and vitamin mix meet the requirements for rats as indicated in N.R.C. (1972).

TABLE 2

Proximate composition of fresh and sun-dried Cassava and pellet diets fed to rats on gestation, lactation and post-weaning growth

Dietary Variables	Dry Matter (as fed) %	Dry matter basis				
		Crude Protein %	Ether extract %	Crude Fibre %	Nitrogen free extract %	Ash %
Commercial pellet						
-control diet	88.1	26.5	4.1	4.5	55.8	9.1
Fresh Cassava						
-Oyanrugbapupa variety	44.5	20.7	6.8	3.8	61.7	7.0
Sundried Cassava						
-Oyanrugbapupa variety	87.6	20.6	6.2	3.8	62.4	7.0
Sun-dried Cassava						
-Isunikankiyan variety	87.5	20.2	6.5	3.0	62.5	7.8

TABLE 3

Proximate composition of fresh and sun-dried Cassava of two varieties

Form of Cassava	Dry Matter %	Dry matter basis				
		Crude Protein %	Ether extract %	Crude Fibre %	Nitrogen free extract %	Ash %
Fresh Cassava -Oyanrugbapupa variety	35.0	2.3	0.5	3.1	93.8	2.4
Sun-dried cassava Oyanrugbapupa variety	88.5	4.4	0.9	5.1	86.6	3.0
Sun-dried Cassava -Isunikankiyan variety	88.5	3.7	0.8	5.0	87.6	3.0

They were offered the diets consumed by their Does in 100 ml beakers attached to the inner side of their cages. Feed and water were given *ad libitum* throughout the study. Feed intake was recorded daily on the fresh cassava diets and weekly on the other treatments. Body weights were recorded weekly.

Second litter studies

After weaning, the Does were rested for a two week period and carried through the same procedure as described above for another gestation and lactation period. As equal number of sexes could not be obtained for the post-weaning growth phase, female rats alone were used in the second litter study.

Histological Studies

At the end of the second litter studies, all surviving Does, first and second litter offspring were individually anesthetized and their thyroid glands removed.

These were immediately fixed, dehydrated and stained using the Masson's trichrome stain. The epithelial heights of the tissue were then measured. They were also examined for thyroid lesions at the Animal health laboratory, Centro Internacional de Agricultural Tropical (CIAT) Cali, Colombia.

Statistical analysis

Variance and covariance analysis were carried out using the method described by Snedecor and Cochran (1967).

RESULTS AND DISCUSSION

First litter studies

Performance of rats on the four experimental diets during gestation, lactation and post-weaning growth phases are presented in Table 4. Litter size at birth was least on the fresh cassava diet, although this was not significant. It is however interesting that the lowest birth weight was recorded in this group. Covariance analysis in which correction was made for litter size also confirmed the same trend. This suggests that the fresh cassava diet contains a factor or factors which makes it inferior to the dried meals. This suggestion is further buttressed by the fact that a slightly higher average feed consumption of rats on the fresh cassava diet as compared to those on the dried flour was recorded during the gestation period.

Variance analysis showed that there was no significant difference in the weaning weight of rats on the commercial pellet and fresh cassava diets. Covariance analysis correcting for litter size however

TABLE 4
Performance of Rats Fed Fresh and Dried Cassava Diets During Gestation*, Lactation and Post-Weaning Growth

1st Litter Studies

Parameters	Control	Cassava		
		Fresh	Dried	
			High HCN	Low HCN
<i>Does</i>				
Number of female mated	5	5	5	5
Length of breeding period, days	27	31	35	31
Daily feed intake during gestation, g	17.9	11.4	10.4	10.1
Daily feed intake during lactation, g**	23.7 ^a	13.7 ^b	14.9 ^{ab}	15.1 ^{ab}
Weight change of Does during lactation, g	-11.9	-8.6	+16.0	-13.0
<i>Suckling rats</i>				
Litter size at birth	6	5	7	6
% Mortality at birth	0	16.7	5.7	6.7
Litter size at weaning	5 ^a	3 ^b	5 ^a	5
% Mortality at weaning	16.7	37.5	28.7	21.9
Birth Weight, g	4.8 ^a	3.8 ^c	4.2 ^b	4.6 ^a
Weaning weight, g	26.3 ^a	25.9 ^a	20.5 ^b	19.6 ^b
<i>Post-weaning growth***</i>				
Daily feed intake, g**	12.9 ^a	8.6 ^b	7.2 ^c	7.0 ^c
Daily weight gain, g	2.7 ^a	2.6 ^a	1.9 ^c	2.1 ^{bc}
Feed efficiency (Feed/gain)	4.8 ^a	3.3 ^c	3.8 ^b	3.3 ^c
Total protein efficiency (TPE)	0.8 ^b	1.4 ^a	1.3 ^a	1.5 ^a

* Since female rats in each treatment were group fed during gestation, data collected at this phase could not be statistically analyzed.

** Feed intake is recorded on a 100% dry matter basis.

*** 28-day study period.

a,b,c Values without common superscripts in horizontal rows are significantly different ($P < 0.05$).

revealed that the weaning weight of rats on the control diet was significantly higher than in the other three groups. The apparently high weaning weight of rats on the fresh cassava diet is explained by the higher mortality in this group. This will ensure less competition for maternal milk among the suckling rats thus enhancing their growth rate.

Post-weaning growth studies show that daily weight gain was significantly higher on the fresh cassava than on dried cassava diets. Feed intake was also significantly higher in this group. Studies by Mancr and Gomez (1973) indicate that when fresh cassava, sun-dried and sucrose diets, were fed to rats, growth rate of the fresh cassava fed rats were significantly higher than those on sun-dried cassava and sucrose diets. It should be remarked

that sun-dried cassava is highly susceptible to fungal attack (Clerk and Cauric, 1968), as well as appreciable alterations in meal quality. This may cause remarkable growth depressions in rats when offered this diet. It is however possible that the initial higher weaning weight of rats on the fresh cassava diet as compared to those on cassava meal diets might have conferred some advantage on the former group.

The feed efficiency and total protein efficiency revealed that the poorest feed utilization was observed on the commercial pellet diet, obviously due to the high dry matter intake in this group. The palatability of the feed might have caused an over consumption of the pelleted diet.

Second litter studies

Performance of rats during the second gestation, lactation and post weaning growth phases are presented in Table 5.

The average breeding period, that is the period between mating with males and parturition, was longest on the dried cassava diets especially on the low HCN variety. During this breeding period which lasted 132 days, all the five rats (100%) on the control diet kindled while only three (60%) on the fresh cassava diet casted their young. On the dried high HCN variety cassava diet, only 1 (20%) kindled, while only two of the five rats (40%) on the dried low HCN cassava produced litters. Since the dried cassava diets have considerably lower HCN concentration than the fresh form, other dietary factors may be responsible for the poorer performance of rats on the dried cassava diets during breeding.

Proximate analysis of fresh and dried cassava samples as presented in Table 3 show that the dried samples contain higher crude protein than the fresh form when values are expressed on dry matter basis. This increase may be due to fungal growth, which may by themselves constitute toxic factors in the cassava diets. This might largely explain the poorer performance observed in the female rats fed the dried cassava diets.

Litter size at birth and weaning were not significantly different in all treatment groups. Covariance analysis revealed that birth and weaning weights were significantly higher on the control diet. This is obviously due to the larger feed intake during gestation and lactation in this group. Other factors like HCN of cassava and probably fungal contamination might have lowered performance of rats on the cassava diets.

TABLE 5

Performance of Rats Fed Fresh and Dried Cassava Diets During Gestation, Lactation and Post-Weaning Growth
2nd Litter Studies

Parameters	Control	Cassava		
		Fresh	High HCN	Low HCN
<i>Does</i>				
Number of females mated	5	5	5	5
Number of Does that kindled	5	3	1	2
Length of breeding period, days	27	37	41	91
Daily feed intake during gestation*, g	16.6	11.7	11.4	11.7
Daily feed intake during lactation*, g	21.5a	15.0b	14.5b	20.1a
Weight change of Does during lactation, g	+27.4	-0.8	-0.8	-12.0
<i>Suckling rats</i>				
Average Litter size at birth	7	5	7	6
% Mortality at birth	0	0	0	0
Average Litter size at weaning	5	4.5	5	4.5
% Mortality at weaning	29.7	12.5	28.5	25.0
Birth weight, g	5.1a	4.1b	4.3b	5.1a
Weaning weight, g	28.6a	17.3b	19.0b	18.7b
<i>Post-weaning growth**</i>				
Daily feed intake*, g	10.9a	7.1b	6.9b	7.2b
Daily weight gain, g	1.5a	1.2b	1.0c	1.0c
Feed efficiency (Feed/gain)	7.4bc	6.2d	7.1c	7.9a
Total protein efficiency	0.5b	0.8a	0.7ab	0.7ab

* Feed intakes are recorded on 100% dry matter basis.

** 28-day growth study.

a,b,c,d Values without common superscripts in horizontal rows are significantly different ($P < 0.05$).

Post weaning growth studies showed that growth rates of rats were significantly lower on dried cassava than on the fresh cassava diet. This is in spite of the similar feed intake on these three groups. This therefore suggests that the growth depression caused by fungal proliferation in dried cassava may be more marked than that caused by high HCN concentration in the fresh cassava diet. Higher feed intake perhaps explains the better live weight performance of rats on the commercial pellet diet.

Alopecia in rats

During the post-weaning growth phase of the second litter studies, poor hair growth was observed around the thoracic and abdominal areas in rats fed the fresh cassava diets. This condition was most noticeable by the second and third week of study. Most of these rats started recovering by the fourth week of study, but this condition was still noticeable, though to a lesser extent by the end of the study.

Histological Studies

Table 6 shows the epithelial heights of thyroid follicles in the experimental animals. While no significant differences were obtained in the Does (mother rats) and first litter offspring. Statistically significant differences were obtained on the second litter offspring. In this group, cell epithelial heights were highest on the control diet, while those on the fresh cassava diet had significantly lower values than on the dried cassava and commercial pellet diets. While absolute

conclusions cannot be made on the effects observed, considering the absolute and relative differences between groups, this may indicate a reduction in thyroid activity in the second litter offspring of the fresh cassava group. The alopecia observed in these rats also lend support to this view.

It should be remarked that malfunctioning thyroid glands do not always exhibit striking differences in their epithelial heights. This histological examination may therefore not suffice to establish the functional state of the gland. A detailed histopathological examination of the prepared thyroid tissue cells was subsequently carried out at the animal health section of the Centro Internacional de Agricultura Tropical (CIAT) Cali, Colombia. The results obtained showed that apart from the low epithelial heights observed in some groups as shown in Table 6, the thyroid follicular cells showed apparently normal activity. While this may suggest that feeding fresh or dried cassava diets over two reproductive cycles does not cause major pathological changes in the thyroid gland of adult female rats or their offspring, the lower epithelial heights and alopecia in the second litter offspring of the fresh cassava group indicate that there is an initiation of thyroid hypofunction at this stage. The major pathological changes confirming lowered activity may therefore take a longer period of study to become manifested. Indeed, epidemiological studies by Osuntokun, (1968) and Delange, (1974); reveal that the effects of cassava toxicity are long term, involving decades in a number of instances. These workers were however

TABLE 6

Epithelial Heights of Thyroid Follicle Cells Obtained from Rats on Different cassava Diets During 2 Reproductive cycles

Test Samples	Commercial Pellets Control	Fresh (High HCN) Cassava	Dried (High HCN) Cassava	Dried (Low HCN) Cassava
Does	6.2	6.4	5.8	6.0
1st litter offspring	6.0	5.6	5.8	6.2
2nd litter offspring	6.4a	6.0c	6.3a	6.2b

a,b,c. Values without common superscripts in horizontal rows are significantly different ($P < 0.05$).

able to show biochemical changes in subjects fed cassava diets over a short period of study.

This study therefore indicates that two factors of paramount importance in cassava diets for the rats were:

- (1) The hydrocyanic acid content, and
- (2) Fungal proliferation in dried cassava diets.

While a careful microbial seeding of ensiled fresh cassava roots has revealed that tremendous amount of nutritionally valuable protein can be produced (Gray and Abou-El-Seoud, 1966; and McPherson, 1966), sundrying of cassava roots especially in rainy seasons can promote the growth of undesirable and toxic micro organisms in the dried chips (Clerk and Caurie, 1968). Indeed sundried cassava chips serves as the major form in which this root crop is exported into world markets for livestock feeding (Phillips, 1974).

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