

Studies on a Virus Disease of Tapioca (*Manihot esculenta* Crantz)

I. Water Relations and Mineral Metabolism*

by

M. N. ALAGIANAGALINGAM¹ and K. RAMAKRISHNAN²

A virus disease of tapioca (cassava, *Manihot esculenta* Crantz) found to be prevalent in a severe form through out Kanyakumari District in Tamil Nadu was reported by Alagianagalingam and Ramakrishnan (1967). The virus was identified as cassava mosaic virus from the symptoms, host range and transmission of the disease. A detailed study of the physiology of diseased cassava plants has been made and the results are presented below.

Materials and Methods: Physiological experiments were carried out with the cassava variety T. 108. The plants were maintained in an insect proof glass house. The moisture content of the healthy and cassava mosaic virus (CsMV) infected cassava was estimated at four times a day viz., 6 a. m., 10 a. m., 2 p. m. and 6 p. m. The leaf samples from both healthy and diseased plants of the same age, growing under identical conditions were collected and their fresh weight recorded immediately. The leaves were then dried in a hot air oven at 105°C till a constant weight was recorded.

The moisture loss of the diseased and healthy leaves was determined by the quick weighing method of Stocker (1956). The loss in weight of individual leaf was recorded in a sensitive balance for 5 minutes at intervals of one minute.

The transpiration rate of the cassava plant was assessed by the gravimetric method of Miller (1953).

For mineral analysis the healthy and diseased leaves were dried at 100°C and then powdered. The powdered leaf was incinerated in a muffle furnace at 525°C for 45 min. The pH of the ash eluate was determined by the method described by Ulrychova and Limberk (1964). Analysis of total phosphorus, calcium, magnesium, potassium, sodium and manganese were made as per the methods described by Jackson (1962). The presence of inorganic P, and total iron were estimated as per the methods of Lowry and Lopez (1946) and Humpries (1956) respectively. The activity of phosphatase was estimated by the method of Promekanon *et al* (1963).

Results: The CsMV infected plants showed at all times a significant decrease of moisture content when compared to healthy cassava. The fresh weight/dry weight ratio also was found to be less. The moisture content

1. Assistant Lecturer in Mycology, 2. Associate Dean, Agricultural College & Research Institute, Coimbatore-3.

* Part of Thesis for ph. D of Madras University.

of both healthy and diseased cassava leaves were found to be maximum at 6 a. m. and thereafter steadily declined till it reached a minimum at 6 p. m. (Table 1). The CsMV infected cassava leaves lost moisture more rapidly and

TABLE 1. *Moisture content of healthy and infected cassava leaves*

Time of sampling	Moisture content in %			Fresh Wt. / Dry Wt. ratio		
	Healthy	Diseased	% decrease over healthy	Healthy	Diseased	% decrease over healthy
6 a.m.	73.47	72.26	-1.65	3.76	3.62	-3.91
10 a.m.	71.36	70.38	-1.38	3.49	3.37	-3.69
2 p.m.	71.08	69.83	-1.78	3.41	3.29	-3.44
6 p.m.	71.32	67.71	-5.07	3.35	3.26	-2.72

was higher by 25.76% by weight and 36.97% by area when compared to healthy leaves (Table 2).

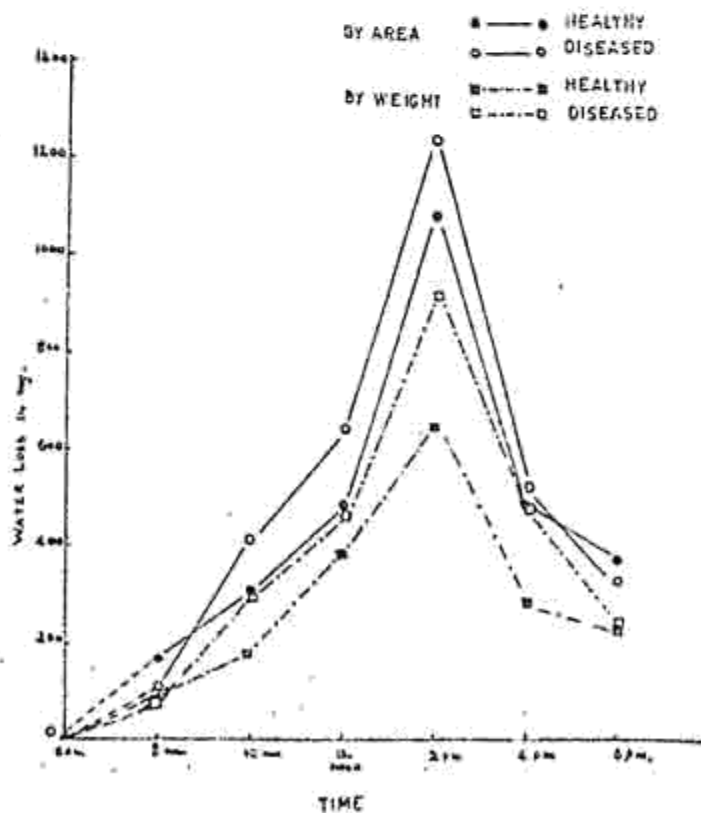
TABLE 2. *Water loss in detached cassava leaves (mg)*

Kind of tissue	Minutes after removal from plant		Water loss per 5 minutes	
	0	5	Per g of leaf	Per 100 sq. cm of leaf area
Healthy	533.1	522.7	21.85	13.57
Diseased	394.6	3284.4	27.48	18.59
% increase over healthy			+25.76	+36.99

The pattern of transpiration in both healthy and diseased cassava were found to be similar (Fig. 1). Both on leaf area basis and leaf weight basis the transpiration rate was found to be enhanced in diseased plants when compared to healthy plant. The transpiration rate increased steadily from 6 a. m. to 2 p. m. when it reached a peak and thereafter declined upto 6 p. m.

The ash content of healthy and CsMV infected cassava was analysed and the results indicated an increase of 33.75% in the diseased cassava,

TEXT FIG. 1. *TRANSPIRATION RATE OF CASSAVA.*



The pH of the ash eluate of healthy leaves was 10.8 and in diseased 11.1.

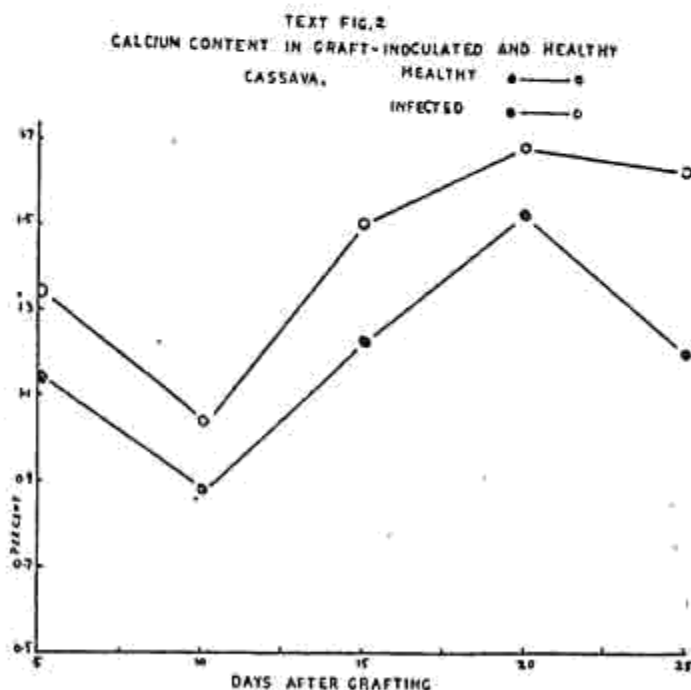
Phosphorus: All the three forms of P were found to be more in diseased leaves. Inorganic P showed an increase of 105.70% over control while total P and organic P showed an increase of 8.04% and 2.10% respectively over healthy leaves (Table 3). The activity of phosphatases also was found to be higher in the case of diseased leaves. Among the phosphatases B-glycero-phosphatase showed an increase of 281.86% while the acid and alkaline pyrophosphatases showed an increase of 61.57 and 69.07% respectively over healthy leaves (Table 3).

TABLE 3. *Phosphorus status and Phosphatase activity in healthy and diseased cassava*

	Healthy	Diseased	% increase (+) or decrease (-) over healthy
Total Phosphorus	0.2151	0.2324	+ 8.04
Inorganic P	0.0123	0.0253	+105.70
Organic P	0.2028	0.2071	+ 2.10
B-Glycero Phosphatase	47.49	181.35	+281.86
Acid Pyrophosphatase	64.99	105.01	+ 61.57
Alkaline Pyrophosphatase	95.09	160.77	+ 69.07

Phosphorus expressed as per cent on dry weight basis.

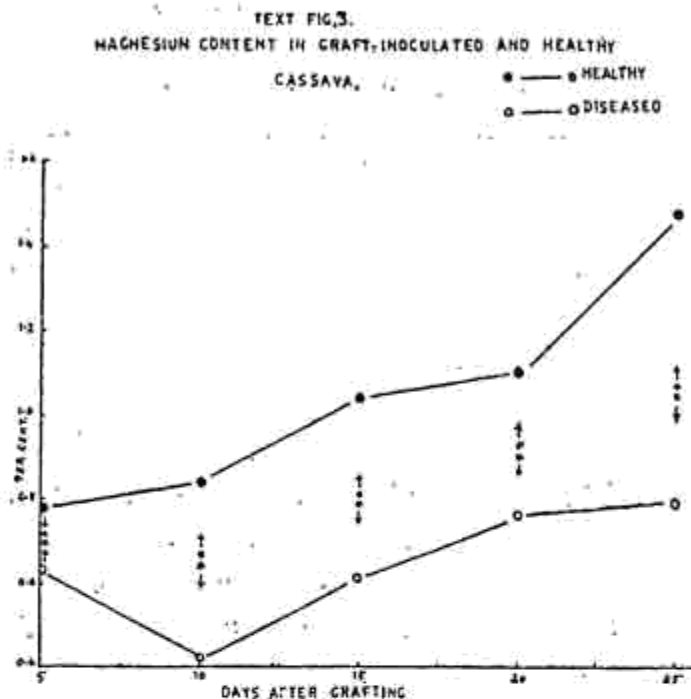
Phosphatases expressed as mg of inorganic P per g of dry weight of leaves.



Calcium: Calcium content was found to be 6.48% higher in the diseased plant than in healthy ones (Table 4). Progressive changes in calcium content in the graft-inoculated cassava was estimated at 5, 10, 15, 20 and 25 days after grafting. The results indicated that the graft-inoculated plants always showed higher calcium content. Both healthy and diseased leaves showed a similar pattern of calcium content. (Fig. 2). The cal-

cium content was found to increase steadily from 10th day onwards and reached a maximum on 20th day and then it declined. A significant maximum calcium content irrespective of disease was found in 20th day sample.

Magnesium: Diseased leaves had significantly lesser magnesium content than healthy leaves, the difference being of the order of 32.98% (Table 4). In the graft-inoculated plants the magnesium content was observed to be less at all stages when compared to healthy plants. In the graft-inoculated cassava the magnesium content was found to drop suddenly on the 10th day and thereafter gradually increased up to 25th day. A maximum difference was noted in diseased leaves on 25th day when compared to healthy leaves (Fig. 3).



Manganese: No significant difference in manganese content was observed between healthy and diseased leaves (Table 4).

Potassium: There was a significant increase of 33% potassium in the diseased cassava over healthy ones (Table 4).

Sodium: The diseased leaves contained significantly higher quantity of sodium. An increase of 6.79% was recorded by diseased plants (Table 4).

Iron: There was a decrease of 15.38% of total iron noted in mosaic infected plants when compared to healthy ones (Table 4).

TABLE 4. Mineral constituents of healthy and diseased cassava

Constituents	Expressed as per cent on dry wt. basis		% increase (+) or decrease (-) over healthy
	Healthy	Diseased	
Ash	5.57	7.45	+ 33.75
Calcium	1.10	1.17	+ 6.48
Magnesium	0.97	0.65	- 32.91
Pottassium	1.10	1.47	+ 33.00
Sodium	0.59	0.63	+ 6.79
Iron	0.26	0.21	- 15.38
Manganese	0.13	0.19	+ 56.00

The phosphorus/iron ratio in healthy and diseased cassava leaves was worked out and the results are presented in Table 5. The results indicated an increased P/Fe ratio in the diseased cassava than that of healthy cassava.

TABLE 5. *Phosphorus/iron ratio of healthy and diseased cassava*

Kind of material	Total phosphorus	Total iron	Phosphorus/iron ratio
Healthy	0.2151	0.2556	0.81
Diseased	0.2324	0.2163	1.07
% Increase (+) or decrease (-) over healthy			+31.48

Discussion: Enhanced transpiration, low moisture content, and a low fresh weight/dry weight ratio as observed in the cassava mosaic disease appear to be common among virus diseased plants (Huberger and Norton, 1933; Ainsworth and Selman, 1936; Owen, 1958; John, 1963; Narayanasamy, 1963). Enhanced transpiration appears to lead to a higher ash content also as observed by Freeland (1936, 1937). It would be interesting to investigate the reasons for such accelerated water loss. It may also be observed that increased transpiration is not peculiar to virus diseases alone but is observed in such diseases as powdery mildews and rusts.

Increases in phosphorus content have been observed in many virus diseased plants (Narayanasamy, 1963; Susuki and Hirai, 1963). Increased phosphatase activity has also been observed in virus diseased plants. The significance of such increase is still not clearly understood. Whether increases of the magnitude reported here could be accounted for by the phosphorus content of the virus particles or the increased energy requirements for virus protein synthesis are problems to be studied.

Considering the pronounced chlorosis of the leaves, one would expect changes in the magnesium and iron contents in the diseased leaves. This is in consonance with the results of Bergman and Boyl (1962) and Narayanasamy (1963) in the case of TMV infected tomato and PsMV infected pigeon pea respectively. The high P/Fe ratio in chlorotic cassava leaves is in corroboration with the results of Dekock, 1956.

Summary: Cassava mosaic infected cassava leaves had lower moisture content and fresh weight/dry weight ratio. The leaves also transpired more rapidly. The diseased leaves contained higher quantities of phosphorus, calcium, potassium, and sodium while magnesium and iron were found to be less when compared to healthy leaves.

REFERENCES

- Ainsworth, C. C., and I. W. Selman. 1936. Some effects of tobacco mosaic virus on the growth of seedling tomato plants. *Ann. appl. Biol.*, **23** : 89-98.
- Alagianagalingam, M. N. and K. Ramakrishnan. 1967. Cassava mosaic in India. *South Indian Hort.*, **14** : 71-2.
- Bergman, E. L. and J. S. Boyle. 1962. Effect of tobacco mosaic virus on the mineral content of tomato leaves. *Phytopathology.*, **52** : 956-7.
- Dekock, P. C. 1956. Heavy metal toxicity and iron chlorosis. *Ann. Bot. Lond.*, **20** : 133-41.
- Freeland, R. O. 1936. Effects of transpiration upon the absorption and distribution of mineral salts in plants. *Am. J. Bot.*, **23** : 355-62.
- . 1937. Effects of transpiration upon the absorption of mineral salts. *Ibid* , **24** : 373-4.
- Huberger, J. W. and J. B. S. Norton. 1933. Water loss in tomato mosaic. (Abstr.) *Phytopathology.*, **23** : 15.
- Humphries, E. C. 1956. Mineral components and ash analysis. *Modern Methods of Plant Analysis* . **1** : 468-502. Springer-Verlag, Berlin.
- Jackson, M. L. 1962. *Soil Chemical Analysis*. Constable & Co. Ltd., London.
- John, V. T. 1963. Physiology of virus infected plants. *Bull. nat. Inst. Sci. India*, **24** : 103-14.
- Lowry, O. H. and J. A. Lopez. 1946. Determination of inorganic phosphates in the presence of labile phosphate esters. *J. biol. Chem.*, **162** : 421-8.
- Miller, E. V. 1953. *Within the living plants*. The Blakiston Company, Inc., New York.
- Narayanasamy, P. 1963. Studies on redgram sterility mosaic disease. *Doctorial Thesis*, Univ. Madras. (Unpubl.)
- Owen, P. C. 1958. Some effects of virus infection on leaf water contents of *Nicotiana* species. *Ann. appl. Biol.*, **46** : 205-9.
- Promeکانon, Y., C. K. Barat and N. B. Das. 1963. Distribution of phosphatase in leaf, stem and root of wheat plant during reproductive period. *Indian J. exp. Biol.*, **1** : 95-7.
- Sasaki, K. and T. Hirai. 1963. Changes in the amounts of phosphorus compounds of tobacco leaves infected with TMV with special reference to the effect of Dinitrophenol. *Phytopath. Z.*, **46** : 343-50.
- Stocker, O. 1956. Messmethoden der Transpiration. *Handb. Pflphysiol.*, **3** : 697-741.
- Ubrychova, M. and J. Limberk. 1964. Some metabolic disturbances in tomato plants infected with potato witch's broom. *Biol. Plant.*, Praha **6** : 291-8.
-