

## IDENTIFICATION OF HEAVY METAL DEPOSIT IN THE CABBAGE TISSUES OF DISEASED COCONUT PALMS (*COCOS NUCIFERA* L.) BY USING ELECTRON MICROPROBE X-RAY MICRO-ANALYSER

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### ABSTRACT

The deposition of Cr, Ti, Cu, Pb, Bi and Ga in the cabbage tissue of root (wilt) diseased coconut palms were investigated by employing scanning electron x-ray microprobe analyser. The x-ray oscillogram and back scattered electron image of specimen indicated the high deposition of Cr, Ti, Cu, Pb, Bi and Ga in the cabbage of diseased palms which were absent in the healthy ones. This was also confirmed in the chemical analysis of a large number of tissue and soil samples under identical conditions.

### INTRODUCTION

VARIOUS biotic factors such as fungi<sup>1</sup>, bacteria<sup>2,3</sup>, nematodes<sup>4</sup>, etc are implicated in the incidence of root (wilt) disease of coconut, which are still to be confirmed. Recently, metal deposit in the roots of the diseased coconut palms has also been studied by using X-MA technique<sup>5</sup>. This technique has been successfully employed in studying the mode of entry and localisation of different heavy metals in the rice crops<sup>6-10</sup>. Rasmussen<sup>11</sup> and Libanati and Tandler<sup>12</sup> observed the deposition of phosphate by aluminium and lead respectively.

The present investigation was undertaken to elucidate the deposition of heavy metals in the cabbage tissues of root (wilt) diseased coconut palms by using X-MA technique.

### MATERIALS AND METHODS

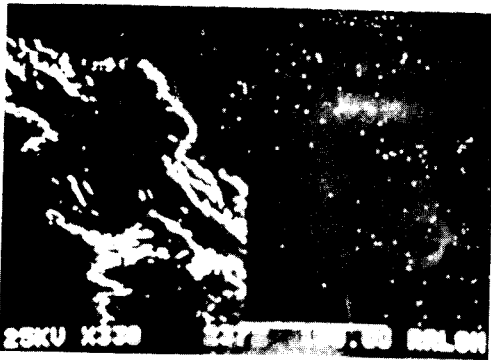
Cabbage tissues (young meristematic) have been collected from healthy and root (wilt) diseased coconut seedlings for this study. The tissue samples were chilled and thin sections were prepared. The tissue sections were mounted on pre-cooled electron microscope grids and further chilled immediately in liquid nitrogen at  $-170^{\circ}\text{C}$  before X-MA analysis. The electron microprobe analysis was carried out in SEM model JEOL 35. The topography of specimen was observed through secondary electron image with accelerating voltage of 25 KV attached with energy dispersive x-ray detector. The selected area was irradiated with an electron beam and the intensity of the characteristic x-ray generated from each metal was integrated and detected. The soil was extracted with DTPA, and 0.1N  $\text{HNO}_3$ , and the cabbage tissue samples

were digested in a di-acid mixture ( $\text{HNO}_3 + \text{HClO}_4$ ). The heavy metals in the extract and digest were estimated using atomic absorption spectrophotometer.

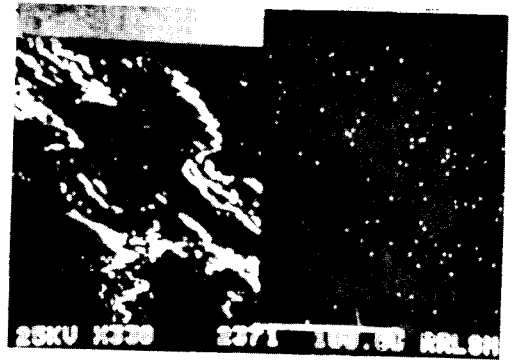
### RESULTS AND DISCUSSION

Results pertaining to the back scattered electron image and x-ray oscillogram for Cr, Ti, Cu, Pb, Mn, Bi and Ga in the cabbage tissue of diseased palms have been illustrated in figures 1 and 2. The X-MA photograph presented in figure 1 reveals the dense distribution of x-ray in the diseased cabbage tissue compared to the healthy suggesting a higher concentration of Cr in the diseased tissue. The intensity of x-ray oscillogram for Ti in the diseased (figure 1) cabbage tissue is far greater than the healthy revealing the accumulation of Ti in the diseased palms. Similar results were also recorded for Pb, Bi, Ga, Mn and Cu where the contents in the diseased tissues were high while the same could not be detected in the healthy cabbage (plants). Though Cu is one of the essential micronutrients for coconut, its concentration in the cabbage tissue of healthy palm was in the range of non detectable range whereas the diseased tissue showed high x-ray intensity for Cu suggesting that Cu is also being accumulated in large amounts under diseased conditions.

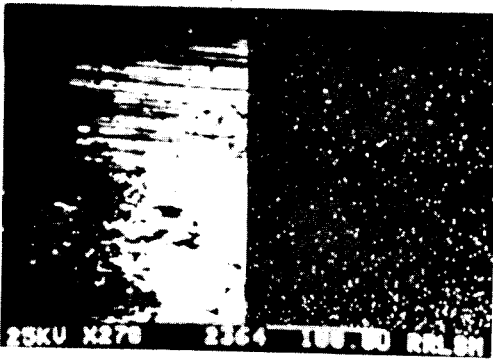
Further, the chemical analysis of soil and tissue samples from diseased and healthy palms also revealed a similar phenomenon. The results furnished in table 1 show the contents of heavy metals at two depths of soils collected from healthy and diseased palms. The  $\text{HNO}_3$  soluble fraction of Bi, Cr, Pb and Ga was high in the diseased soils while that of Cu and Ti showed a



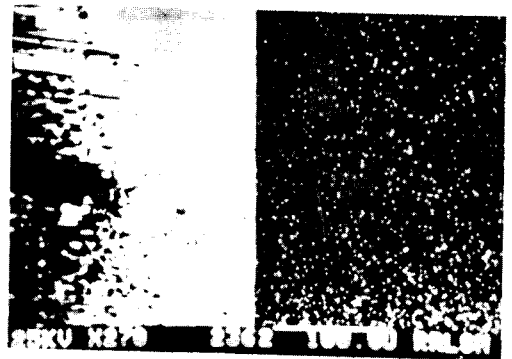
TITANIUM — HEALTHY



CHROMIUM — HEALTHY



TITANIUM — DISEASED



CHROMIUM — DISEASED

Figure 1. Deposition of titanium and chromium.

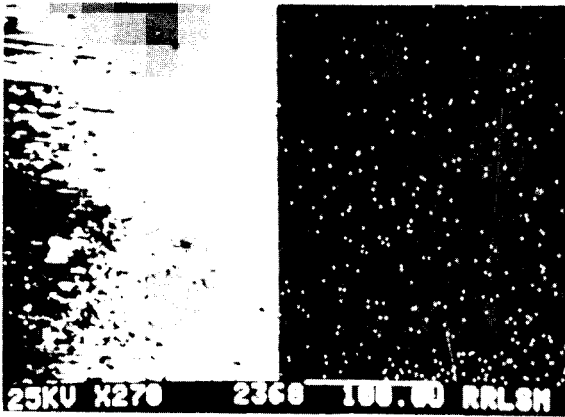
Table 1 Concentration of heavy metals in soils ( $\mu\text{g/g}$ )

Extractant	Depths (cm)		Elements					
			Cu	Bi	Cr	Pb	Ti	Ga
0.1N HNO <sub>3</sub>	0-50	H	2.80	0.17	0.16	0.51	0.21	tr
		D	1.14	0.19	0.19	1.30	0.15	tr
	50-100	H	2.90	0.15	0.15	0.65	0.20	tr
		D	1.28	0.82	0.15	0.03	1.00	0.12
0.005M DTPA	0-50	H	0.66	tr	0.13	0.14	0.33	0.22
		D	0.91	tr	0.05	1.52	tr	0.45
	50-100	H	0.63	tr	0.10	0.40	0.22	0.51
		D	1.28	tr	0.05	1.01	tr	0.35

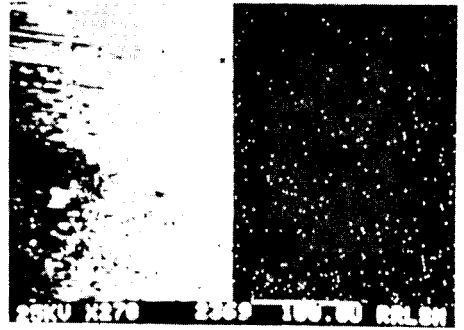
(H = Healthy; D = Diseased)

reverse trend. On the other hand, the DTPA extractable Cu, Pb and Ga were high for the diseased soils. However, Bi was not detected in the DTPA extract of the soils at both the depths. The results of heavy metal

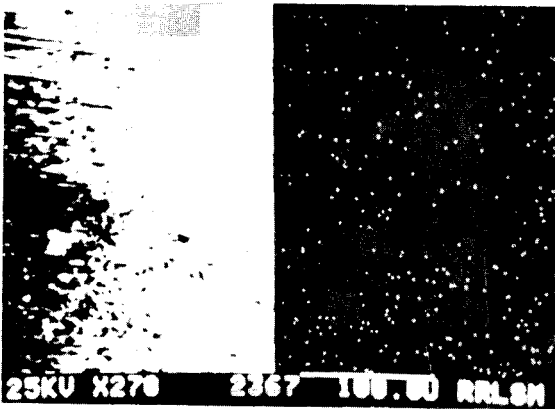
concentration in the leaves (table 2) showed a significant difference between the diseased and healthy palms. The content of all the heavy metals is higher in the diseased tissue compared to the healthy palms. The



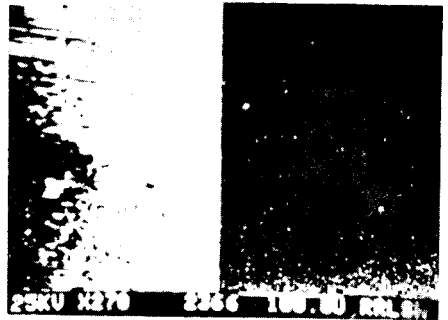
LEAD



BISMUTH



GALLIUM



COPPER

Figure 2. Deposition of lead, bismuth, gallium and copper in diseased tissues only.

Table 2 Heavy metals concentration in cabbage tissue ( $\mu\text{g/g}$ )

	Cu	Bi	Cr	Pb	Ti	Ga
H	21	7	6	tr	12	10
D	20	19	34	2.8	17	29

(H = Healthy; D = Diseased)

differences are more pronounced with respect to Bi, Cr, Pb and Ga. This strongly supports the findings of X-MA results where high concentration of localisation of Cu, Bi, Cr, Pb, Ti and Ga in the cabbage tissues of the diseased palms have been detected.

#### ACKNOWLEDGEMENTS

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