# ROLE OF COPPER AND MANGANESE APPLICATION ON VARIOUS PIGMENT LEVELS OF *ZINGIBER OFFICINALE* ROSC. VAR-1

## A. KSHERODA DEVI and P.K. SINGH

Department of Life Sciences, Manipur University, Canchipur, Imphal-795003, Manipur, India.

Field experiments were carried out during the period of April to December 2002 and 2003 to improve the qualitative and quantitative characters of ginger (*Zingiber officinale* Rosc.)VAR-1 with soil and foliar application of copper (CuSO<sub>4</sub>) manganese (MnSO<sub>4</sub>) and combination of the two as total micronutrients (TM) at different concentrations, in a randomized block design. Estimation of chlorophyll from the fresh leaf samples of various treatments was done at four intervals of 90, 120, 150 and 180 days after plantation (DAP). The concentration of total chlorophyll (T-chl) and chlorophyll b (chl-b) were found to be highest at the early growth stage but the concentrations of chlorophyll a (chl-a) and carotenoid (ca) were found to be highest at the latter growth stages. Among the treatments, minimum level was recorded in control stages (T<sub>1</sub>). In order to enhance the yield, micronutrients play an important role in carotenoid and chlorophyll levels, both in soil and foliar sprayed plants.

Keywords : Ginger; Micronutrients; Soil and foliar application.

# Introduction

. 1.

Micronutrients play specific roles that enhance the production of vegetables. Wallace and Romney' reported that due to synergistic trace metal effects in plants, any individual trace elements, when supplied at low or high quantity, affects the concentration of other elements in plants. The most well known example of the involvement of manganese is in an enzyme protein of the water splitting system associated with PSII and in its deficiency, the functioning of the protein is impaired, thus affecting photosynthesis. In severe manganese deficiency, a decrease in chlorophyll content along with disturbances in ultra structure of thylakoids have been reported as a consequence of inhibition of carotenoid biosynthesis in such condition. Copper acts as a prosthetic group of the enzymes. It is also a component of plastocyanin, which takes important role in electron transport system in photosynthesis. Copper deficiency causes curling of young leaves. The present study is to investigate the effect of various concentrations of Mn, Cu and TM in ginger (Zingiber officinale Rosc. L.)VAR-1, cultivation.

# **Materials and Methods**

The experiment was conducted at the Life Sciences Experimental field, Manipur University Campus, during the period of April to December 2002 and 2003. The plot was laid out in a randomized block design (RBD) with four replications each having a plastic bag (size 15 cm diameter, 8cm height and 2 kg soil capacity) and spacing of 15cm. The size of the experimental area is 6.3m length and 1.75m breadth. Number of plots were 24 with 6 treatments and a population of 240 plants. Application of micronutrients were performed on soil and foliar (40 and 70 DAP). The treatments are  $T_1$  (control without micronutrients),  $T_2$  (with copper-0.012 g/ bag),  $T_3$  (manganese - 0.12g/bag). Foliar application was done with 0.05% of spraying reagents Cu, Mn and TM (0.05% : 0.05%); they are  $T_4$ ,  $T_5$  and  $T_6$  respectively. Estimation of chl / and ca / were done by the methods of Arnon<sup>3</sup> and Jayaraman<sup>4</sup>. Representation and statistical calculations were made by following the techniques of Cochran and Cox<sup>5</sup> and Gomez and Gomez<sup>6</sup>.

1. 1. 2.

#### **Results and Discussion**

The levels of T- chl in the fresh leaves of test plants were variable. In 90 DAP and 150 DAP, the highest T-chl concentrations were 0.673 mg/g and 0.346 mg/g respectively and occurred when the plant received the treatment  $T_6$  (Total micronutrients in foliar sprayed plants). At 120 and 180 DAP, the highest T-chl value was observed in  $T_4$  (plants treated with copper as foliar spray) i.e. 0.311 mg/g and 0.470 mg/g. The lowest T-chl concentration was recorded by plants in  $T_1$  (control without micronutrients) in all age groups (Table 1).

The concentration of chl - a was highest in  $T_4$  (Cu in foliar sprayed plants) at 90, 120 and 180 DAP. At 150 DAP, the highest concentration of chl - a was observed in  $T_6$ . The lowest concentration of chl - a was observed in  $T_1$ (control without micronutrients) in all stages (Table 2). The levels of chl- b decreased from 90 to 180 DAP. The highest chl- b was observed in  $T_6$  i.e. 0.526 mg/g at 90 DAP. At 120 DAP and 180 DAP, the concentrations of chl- b were observed highest in  $T_2$  (Cu in soil treated plants) i.e. 0.170 mg/g and 0.143 mg/g. At 150 DAP, the highest concentration of chl- b was observed in  $T_4$  (Cu foliar sprayed plants) i.e.

## Devi & Singh

0.182 mg/g. The lowest concentration of chl-b was observed in control in all treatments (Table 3). The levels of ca increased gradually from 90 to 180 DAP. The concentration of ca was observed highest in  $T_4$  (Cu in foliar sprayed plants) at 90 and 180 DAP. At 120 and 150 DAP, the highest concentrations were observed in  $T_6$  (Total micronutrients in foliar sprayed plants). The lowest concentration of ca was observed in control in all treatments (Table 4).

Chlorophylls are the plant pigments present in the chloroplast of the cells. They are responsible for the process of photosynthesis of plants which is one of the most important vital metabolic processes of the plants. The rate of photosynthesis is governed by the chlorophyll level as well as the presence of appropriate amount of precursor molecules which are associated with number of elements such as Mg <sup>2+</sup>, Fe <sup>2+</sup> etc.

In the present investigation, the highest accumulation of T-chl and chl- b are found in  $T_6$  (TM in foliar sprayed plants). Then the accumulation of chl- a was found in  $T_4$  (Mn in foliar sprayed plants). Mn<sup>+2</sup> has been reported to increase the enzyme activities<sup>7</sup>. Cu plays an important role in electron transport system of photosynthesis.

In all cases there is a gradual decrease in chlorophyll content as the plant get ageing 120 to 180 DAP. At 90 DAP, the highest accumulation of T-chl and chl- b were found in treatment  $T_6$  (TM in foliar sprayed plants) with 0.673 mg/g and 0.526 mg/g respectively. In chl- a, highest accumulation was found in  $T_4$  (Cu in foliar sprayed plants) with 0.166 mg/g.

Manganese plays an important role in splitting of water associated with PSII and in its deficiency the functioning of protein is impaired thus affecting photosynthesis. Increased chlorophyll content and the presence of efficient photosynthetic electron transport system could be an indication of increased photosynthesis. At 120 DAP, highest accumulation of T- chl, chl- a and chl-b were found in T<sub>4</sub> and T, with 0.311 mg/g 0.183 mg/g and 0.170 mg/g respectively. At 150 DAP highest is found in T<sub>c</sub> for chlorophyll a and T-chl with 0.206 mg/g and 0.346. But for chl- b, highest accumulation is found in T, with 0.180 mg/g. At 180 DAP, highest accumulation for T-chl and chl-a are found in T, with 0.470mg/g and 0.284 mg/g respectively. For chl- b, highest is found in T, with 0.143 mg/g. In the present finding, total micronutrients (TM) plays an important role in the synthesis of T-chl, chl- a and chl- b at the early stages. But at the latter stage,  $T_4$  (Cu in foliar sprayed plants) are more distinctive then the combined one. This may be due to functional importance of individual micronutrients in various enzymatic reactions.

These findings are in agreement with the finding

of Miziorko and Lorimer<sup>8</sup> and Sorte *et al.*<sup>9</sup>. The effect of plant growth regulators and micronutrients on chlorophyll was studied by Meyyappan *et al.*<sup>10</sup>. Miller *et al.*<sup>11</sup> reported that in Fe deficient plants, ferredoxin would be limiting and could directly affect chlorophyll synthesis.

Increased chlorophyll content due to NAA application is recorded by Mohamed Yasin<sup>12</sup> in cotton, and by Karthikeyan<sup>13</sup> in groundnut. Plant growth regulators had increased the photosynthetic rate<sup>1415</sup>.

Carotenoids are the pigments involved in the initial absorption of water, which ruled as an environmental factor of growth and development. The result of brown colouration or less greenery on the surface of leaves may be a consequence of the formation of carotenoid pigments. The finding was in agreement with the depicted results of earliar workers<sup>16</sup>.

Regarding the carotenoid content, it is found to be highest in  $T_4$  at 90 and 180 DAP with 2.036 mg/g and 2.886 mg/g. At 120 and 180 DAP, highest accumulation of carotenoid are found in T6 with 1.964 and 1.493 mg/g respectively.

The present finding indicates the role played by Mn and TM in the different stages of growth phase of the test crop. Carotenoid signify the enzymes viz. catalase, peroxydase and the cytochrome, which have increased electron transport in proteins in photosynthesis<sup>17</sup>. Mishra and Shrivastava<sup>18</sup> reported that the supply of inorganic nitrogen prevents degradation of chlorophyll and carotenoid in maize leaves. Nieman and Paulson<sup>19</sup> studied the alleviation of carotenoid level by either form of nitrogen indicated structural requirement of chloroplast which might be deformed under salinity.

In the present study chlorophyll and carotenoid accumulation in various treatments particularly in  $T_6$  and  $T_4$  indicates the suitable treatment in ginger crop. The general objectives of this experiment is to forcast the yield of the crop. So in  $T_6$  treatment maximum photosynthate will be available because of the maximum level of chlorophyll. In the two treatments  $T_6$  and  $T_4$  highest concentration and accumulation of chlorophyll was observed. From the above findings it can be concluded that the maximum crop yield will be in the two treatments ( $T_6$  and  $T_4$ ).

The application of micronutrients on ginger become important for the efficient utilization and better performance of the crop<sup>20</sup>. It is thus suggested that Mn application (MnSO<sub>4</sub> as a source) such as combination with Cu (CuSO<sub>4</sub> as a source) may counteract the toxic effect of Copper sulphate in the leaves of *Zingiber officinale*, has been in aggrement with the observation of Day and Shrivastava<sup>21</sup>.

#### Acknowledgements

The authors are grateful to the Head, Department of Life

#### 216

Treatments	Levels of total chlorophyll (mg/g)				
	90 DAP	120 DAP	150 DAP	180 DAP	
Ti	0.357±0.048	$0.203 \pm 0.005$	0.249±0.003	0.358±0.002	
T <sub>2</sub>	0.499±0.036	0.232±0.025	0.312±0.008	0.428±0.004	
T <sub>3</sub>	0.454±0.028	$0.259 \pm 0.002$	0.273±0.093	$0.371 \pm 0.006$	
T <sub>4</sub>	0.418±0.095	0.311±0.008	0.254±0.006	0.470±0.006	
T <sub>s</sub>	0.519±0.023	0.245±0.013	0.316±0.002	0.403±0.026	
T <sub>6 &lt;</sub>	0.673±0.000	0.263±0.018	0.346±0.000	0.309±0.031	
CD at 5% CD at 1%	0.042 0.076	0.022 0.037	0.020 0.033	0.016	

Table 1. Effect of Cu, Mn and TM application (Treatments) on T-chl content of Zingiber officinale Rosc.L. VAR-1 at different stages of days after plantation (DAP).

Table 2. Effect of Cu, Mn and TM application on Chl - a content of Zingiber officinale Rosc. L. VAR-1 at different stages of days after plantation (DAP).

Treatments	Levels of chlorophyll a (ma/a)			-)
	90 DAP	120 DAP	150 DAP	180 DAP
T <sub>1</sub>	$0.120 \pm 0.007$	0.117±0.003	0.143±0.003	0.178±0.015
T	0.142±0.016	$0.133 \pm 0.001$	0.169±0.006	0.265±0.012
T <sub>3</sub>	0.149±0.005	0.139±0.008	0.144±0.008	0.209±0.018
T <sub>4</sub>	0.166±0.022	0.183±0.006	0.159±0.008	0.284±0.005
T,	0.137±0.011	$0.131 \pm 0.013$	0.179±0.001	0.225±0.024
T <sub>6</sub>	0.154±0.002	$0.151 \pm 0.005$	0.206±0.004	0.188±0.005
CD at 5 % CD at 1 %	0.010 0.016	0.006 0.010	0.004 0.006	0.014 0.023

Table 3. Effect of Cu, Mn and TM application on Chl - b content of Zingiber officinale Rosc. L. VAR-1 at different stages of days after plantation (DAP).

Treatments		Levels of chlorophyll $h(mg/g)$			
	a., ,	90 DAP	120 DAP	150 DAP	180 DAP
T <sub>1</sub>	⊂ a 3 <sup>8</sup>	$0.192 \pm 0.021$	0.085±0.002	0.132±0.017	0.095±0.011
T_2		$0.358 \pm 0.020$	0.170±0.001	0.165±0.017	0.142±0.003
T <sub>3</sub>		0.311±0.002	0.117±0.004	0.162±0.007	$0.086 \pm 0.003$
T <sub>4</sub>		0.369±0.000	$0.128 \pm 0.002$	0.182±0.005	0.107±0.008
T,		$0.369 \pm 0.028$	$0.105 \pm 0.009$	0.179±0.002	0.137±0.002
T <sub>6</sub>		$0.526 \pm 0.003$	0.112±0.013	0.164±0.011	$0.139 \pm 0.003$
CD at 5 % CD at 1 %		0.012 0.020	0.004	0.010 0.016	0.008

Devi & Singh

COME.

Table 4. Effect of Cu, Mn and TM application on Carotenoid (Ca) content of Zingiber officinale Rosc. L.VAR-1 at different stages of days after plantation(DAP).

Treatments	Levels of carotenoid (mg/g)			
1. 1	90 DAP	120 DAP	150 DAP	180 DAP
T .	0.884±0.012	1.333±0.026	0.971±0.023	$2.088 \pm 0.302$
T, David 28	0.999±0.018	$1.517 \pm 0.051$	$1.256 \pm 0.062$	$2.543 \pm 0.092$
T <sub>i</sub> er t	1.074 ±0.015	1.582±0.062	0.912±0.023	2.376±0.128
T Carto	2.036±0.214	1.990±0.104	$1.131 \pm 0.010$	2.886±0.015
T, EXTLA	$1.061 \pm 0.068$	1.575±0.147	1.032 ±0.019	2.684±0.143
T	$1.858 \pm 0.025$	1.964±0.116	1.493±0.025	1.956±0.174
CD at 5% CD at 1%	0.066 0.111	0.096 0.161	0.030 0.050	0.163 0.272

Sciences, Manipur University, Canchipur, Imphal, for laboratory facilities.

## References

- 1. Wallace A and Romney E M 1977, Synergistic trace metal effects in plants. *Soil Science*.
- Nable R O, Bar Akiva A and Loneragan J F 1984, Functional nutrient requirement and its use as a critical value for diagnosis of Mn deficiency in Subterranean cloves. *Ann. Bot.* 54 39-49.
- Arnon DI 1949, Copper enzymes in isolated chloroplast polyphenoloxidase in *Betavulgaris*. *Plant Physiol*. 249 1-15.
- Jayaraman J 1992, Laboratory Manual in Biochemistry. H.S. Proplairfor Wiley Eastern Limited, New Delhi, pp:176.
- Cochran W G and Cox G M 1965, Experimental Design. John Wiley, New York.
- Gomez K A and Gomez A A 1976, Statistical procedure for Agricultural Research with emphasis on rice. IARI. LOS-Banos. Philippines.
- 7. Marschner H 1995, Mineral Nutrition of Higher Plants. Academic Press, New York.
- Miziorko HM and Lorimer GH. 1983, Ribulose-1,5biphosphate carboxylase- oxygenase. Ann. Rev. Biochem. 52 507-535.
- 9. Sorte N V, Ratnaparkhi V P and Shastri N R 1989, Effect of foliar application of cycocel on Peanut(*Arachis hypogea* L.). *Ann. Plant Physiol.* **3** 203-211.
- Meyyappan M, Vaiyapuri V and Alagappan R M 1991, Proc. of Intt. Conf. Pl. Physiol. 7-11.
- Miller G W, Pushnik J C and Welkie G W 1984, Iron chlorosis a world wide problem, the relation of chlorophyll biosynthesis to iron. J. Plant Nutr. 7 1-22.
- 12. Mohamed Y 1987, Effect of tricontanol and other

growth regulators on Bud and Boll shedding, yield and quality of MCU of cotton. M. Sc. (Agri)Thesis, TNAU, Coimbator.

- Karthikeyan M 1988, Effect of tricontanol and other growth regulators on growth yield and quality in groundnut(*Arachis hypogea* L.)JL-24.M. Sc.(Agri)Thesis, TNAU, Coimbatore.
- Erickson A B, Selton G, Shogen D and Nelson S 1981, Comperative analysis of the effects of triacetanol on photosynthesis. *Planta* 152 44-49.
- Rao B C 1985, Physiological effects of Tricontanol on first crop rice at its late growth stages under high temp conditions. *Plant Physiol. Commun.* 1 28-29.
- Spurr A R and Harris W M 1968, Ultrastructure of chloroplast and chromoplast in *Capsicum annuum*. *Ann. J. Bot.* 55 1210-1224.
- 17. Ting I P 1982, Plant Physiology, Addition Wesley Publishing Company.
- Mishra S N and Srisvastava H S 1983, The role of inorganic nitrogen in the synthesis and degradation of chlorophyll and carotenoid in maize leaves. *Biol. Plant.* 25 21-27.
- Nieman R H and Paulson L L 1971, Plant growth suppression in saline medium : Interaction with light. *Bot. Gaz.* 132 14-19:
- Devi A K and Singh P K 2005, Role of copper and manganese application on nitrate reductase and protease activities of *Zingiber officinale*. J. Curr. Sci. 7(1)67-70.
- 21. Dey B and Srivastava R C 2005, Ameliorative role of certain naturally occurring protective substances in antagonizing the phytotoxic effects of copper sulphate on nitrate reductase activity in *Pisum sativum L. Indian J. Plant Physiol.* **10**(1) 70-72.