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Changes in Sugar, Amino Acid and Mineral Contents of Leaves of Two Mango Varieties Affected by Quick Decline Disease

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Abstract: Quick decline disease causes collapse of healthy mango plants within a few days. The menace has been observed in the entire mango growing areas of Pakistan. The leaf samples of healthy and diseased mango plants of two major varieties viz. chaunsa and langra were analyzed to asses the variations in the contents of sugars, free amino acids and metals in relation to quick decline. In general, the contents of total sugars, reducing sugars, non-reducing sugars, free amino acids and minerals were found to be decreased in the diseased leaves as compared to that of healthy leaves of both the mango varieties. Quantitative analysis of macro and micro-nutrients was carried out by atomic absorption and flame emission spectrometry to find the status of these elements in healthy and diseased leaves. Mg contents were found to be decreased in the diseased leaves of both the mango varieties whereas Cu and Zn contents were found to be increased. No consistent trend was observed in case of other metals.

Key words: Variations, biological constituents, leaves, mango, quick decline

Introduction

Mango (*Mangifera indica* L.) is one of the popular seasonal fruits found mainly in the tropical and subtropical countries. It is widely grown in different countries of the world such as India, Pakistan, China, Nigeria, Egypt, Sri Lanka, Brazil, Mexico, Florida (USA) Malaysia, Philippines, Indonesia, Thailand, Vietnam, Australia, Java and Queensland (Srivastava, 1998). Mango grows well in the tropics and subtropics in well drained soils. The crops can be grown on various types of soil with pH 5.5–6.0. It can tolerate temperatures as low as 4°C and as high as 46°C. Ideal location for mango growing is the area where there is a consistent 1-2 month drought to trigger flowering and fruiting.

Pakistan is a second largest producer of mango in the Asia with annual production of 917,000 tons of mangoes (Anonymous, 1999). At present, area under mango cultivation in Pakistan is more than 82000 hectares. Mango varieties have been known for attractive colors, savouring smell, delightful taste and high nutritive value. Pakistani mangoes include: anwar ratole, bengen phali, chaunsa, collector, dasehri, fajri, langra, maldah, neelam, gulab khas, rumani, vanraj, fazli, safeda, zardalu, mulgoa, samar bahist, saroli, sindhri, tuta and, pari, etc. sindhri and chaunsa are leading export varieties mangoes from Pakistan reach Europe, the Gulf States, Central Asia and the United States.

A mango plant suffers from several diseases at all stages of its life. All parts of the plant, namely, trunk, branch, twig, leaf, petiole, flower and fruit are attacked by a number of pathogens including fungi, bacteria and algae. They cause several kinds of rot, die back, anthracnose, scab, necrosis, blotch, spots, mildew, etc. Major diseases of mango plant are powdery mildew, anthracnose, die back, phoma blight, bacterial canker, red rust and sooty mould etc., (Persley, 1993). More than 492 species of insects, 17 species of mites and 26 species of nematodes have been reported to be infesting mango trees, about 45% of which have been reported from India. Almost a dozen of them have been found damaging the crop to a considerable extent causing severe losses and, therefore, may be termed as major pests of mango. These are hopper, mealy bug, inflorescence midge, fruitfly, scale insect, shoot borer, leaf webber and stone weevil. Of these, insects infesting the crop during flowering and fruiting periods cause more severe damage (Khan *et al.*, 1996).

Now-a-days a new disease called quick decline (actually known as collar rot, stem rot or crown rot) has been observed (Ellis, 1997; Gove, 1997; Tevioltdafe and Gubler, 1995). This is a serious disease and its intensity is increasing day by day. The healthy plants under attack collapse within days. It damages the mango orchards which is a discouraging aspect for mango growers as well as economic loss to the national economy. Pest control work has been done by different research institutions in Pakistan (Khan *et al.*, 1996) but little attention has been paid towards chemical analysis of the mango plants in relation to quick decline.

This study was aimed to look at variations in some biologically important constituents such as the contents of total sugars, reducing sugars, non-reducing sugars, total free amino acids and metals in healthy and diseased (quick decline) leaves of two mango varieties i.e. chaunsa and langra. Such an information was thought to be helpful in order to develop strategies to rehabilitate the disease affected mango plants.

Materials and Methods

Sample collection and storage: Leaf samples were collected from the experimental orchard of Mango Research Station Shujabad, Punjab, Pakistan. The leaves were picked from the healthy and diseased (affected by quick decline) plants of two varieties viz. chaunsa and langra. Each variety had three single tree replications. The leaves were dried under shade and packed in airtight plastic bags. Before the analysis, the samples were powdered in an agate pestle and mortar. The powdered samples were stored in pre-cleaned polyethylene containers.

Extraction and quantitative analysis of sugars and free amino acids: Sugars and free amino acids were extracted and quantified as follows: 2g of each powdered sample was taken and socked separately in 75% ethanol (100 ml). After 24 h, the samples were ground and filtered. The residue was washed with a few ml of 75% ethanol and the volume was made upto 100 ml. The filtrate obtained in each case was tested for presence of sugars and free amino acids using molish and ninhydrin reagents respectively. These showed positive tests in each case. The filtrates were then preserved in refrigerator for further analysis. Total sugars were determined spectrophometrically using anthrone reagent (Travelyan and Harrison, 1952). Reducing sugars were determined by ferricyanide method (Hulme and Narian, 1931). Free amino acids were quantified using ninhydrin method (Pandey, 1984).

Sample preparation and metal analysis: For sample preparation,

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0.5 g of dried powdered samples was ashed in a muffle furnace (Gallenkamp, England), at 500^{0}C for 5 h. After heating the samples were cooled down to room temperature in a desicator and ash contents were weighed. The ash contents were dissolved in aqua regia (5ml). The sample was heated to near dryness and 5 ml of HNO_3 was added to it. Solution was filtered using Whatman filter paper-42 and filtrate was transferred to 50 ml measuring flask and volume was made upto the mark with deionized water. The solution was stored in clean polyethylene bottle for metal analysis by flame spectrometric technique. Metals such as Ca, Mg, Fe, Cu, Mn and Zn in sample solutions were estimated by atomic absorption spectrometry (A-1800 AAS, Hitachi, Japan) following wavelengths (nm) Ca, 422.7; Mg, 285.2; Fe, 248.3; Cu, 324.8; Ma, 279.6 and Zn, 213.8. Na and K were estimated by flame emission at wavelengths 589.0 and 766.5 nm respectively under conditions specified by the manufacturer.

Results and Discussion

The contents of total sugars were found to be decreased in case of diseased leaves of both the mango varieties. A significant decrease in total sugar contents i.e. 27.61 and 37.84% was observed in diseased leaves of both varieties viz. chaunsa and langra respectively (Table 1). Contents of reducing sugars were also found to be decreased i.e. 33.33 and 35.43% in case of diseased leaves of chaunsa and langra respectively. However decrease in non-reducing sugars contents was found to be 10.34 and 42.10% in diseased leaves of chaunsa and langra respectively compared to the healthy leaves of the mango plants. In general, more profound decrease was observed in the contents of total sugars, reducing sugars and non-reducing sugars in case of diseased leaves of langra plant. A uniform trend was observed in the decrease of sugars in diseased leaves of the each mango variety. Decrease in the sugar contents may be attributed to the decrease in the rate of anabolism (photosynthesis etc.) or higher rate of degradation of sugars in the diseased leaves caused by the pathogens. Free amino acids were also found to be decreased 33.61 and 24.14% in diseased leaves of chaunsa and langra varieties respectively. Decrease in the contents of free amino acids was more profound in case of diseased leaves of chaunsa compared to the diseased leaves of langra. The decrease in free amino acid contents may be due to more utilization of these by the disease causing pests. Ash contents were lower in the diseased leaves of langra compared to that of chaunsa.

Macro and micronutrient levels found in the healthy leaves of both varieties, i.e. chaunsa and langra were lower than the levels critical for growth of a multicellular plant which may be due to intensive cultivation and imbalanced fertilizer use causing depletion of a number of nutrients from the soil (Table 2). Percent decrease in ash contents in diseased leaves of chaunsa and langra were found to be as 0.47 and 2.71% respectively compared to healthy leaves of these varieties.

Levels of macronutrients (Ca, K, Mg) found in healthy leaves of both the mango varieties differ with the earlier reported data (Khan *et al.*, 1996). Calcium and magnesium levels were found to be decreased 47.59 and 12.26% respectively in diseased leaves compared to the healthy leaves of chaunsa whereas sodium and potassium levels were found to be increased 90.36 and 26.23% respectively in diseased leaves of chaunsa compared to the healthy leaves. Calcium and potassium levels were found to be increased 27.22 and 6.99% respectively in diseased leaves of langra. The contents of Mg and Na were decreased 53.40 and 25.64% respectively in case of diseased leaves as compared to healthy leaves of langra. Magnesium levels found to be decreased in the diseased leaves of both the mango varieties. No consistent pattern was found in case of other metals.

Concentrations of micronutrients (Zn, Fe, Mn) found in the healthy leaves of chaunsa and langra varieties are similar to the earlier reported work (Khan *et al.*, 1996). Copper and zinc levels were found to be increased in the diseased leaves of both the mango varieties. On the other hand, iron levels were found to be decreased (41.41%) and increased (39.89%) in diseased leaves of chaunsa and langra respectively. Manganese levels were found to be decreased (41.03%) and increased (32.36%) in the diseased leaves of chaunsa and langra respectively.

Keeping in view the results of this study, it is suggested that in addition to the pest control strategies, soil of the disease affected

Table 1: Concentration of, free amino acids, sugars and ash contents in healthy and diseased leaves of two mango varieties in relation to quick decline

	Chaunsa			Langra		
Constituents	Healthy leaves	Diseased leaves	% Decrease	Healthy leaves	Diseased leaves	% Decrease
Free amino acids (g/100g)	2.38±0.39	1.58±0.39	33.61	2.61±1.38	1.98±0.70	24.13
Total sugars (g/100g)	1.34±0.23	0.97±0.09	27.61	1.85±0.15	1.15±0.15	37.83
Reducing sugars (g/100g)	1.05±0.16	0.70±0.08	33.33	1.27±0.34	0.82±0.28	35.43
Non reducing sugars (g/100g)	0.29±0.10	0.26±0.04	10.34	0.57±0.19	0.33±0.23	42.10
Ash contents (%)	86.27±0.52	85.86±1.72	0.47	90.30±4.30	87.85±0.83	2.71

Table 2: Levels of essential nutrients critical for growth of multicellular plant (Bonner and Varner, 1976) and metals estimated in healthy and disease (quick decline) affected leaves of two mango varieties

		Metal concentration					
		Chaunsa			Langra		
Metals	Levels of essential nutrients critical for growth of multicellular plant	Healthy leaves	Diseased leaves	% Decrease/ increase	Healthy leaves	Diseased leaves	% Decrease/ increase
Ca (mg g^{-1})	5.0	9.49±2.79	5.00±3.58	47.31	3.16±0.94	4.02±0.52	21.39
K (mg g^{-1})	10.0	2.49±0.88	4.74±2.17	47.46	5.86±5.35	6.27±0.84	6.53
Mg (mg g^{-1})	2.0	7.75±1.06	6.80±4.17	12.25	5.88±2.72	2.74±1.58	53.40
Na (mg g^{-1})	-	1.22±5.20	1.54±0.32	20.77	1.56±0.56	1.16±0.13	25.64
Cu (μg g^{-1})	6.0	9.20±1.6	11.20±2.8	17.85	8.50±1.4	12.30±2.5	30.89
Fe (μg g^{-1})	100.0	496.00±70.0	291.00±107.0	41.33	367.00±117.0	516.00±19.0	28.87
Mn (μg g^{-1})	50.0	28.80±3.1	17.00±8.2	40.97	16.80±3.2	22.20±1.0	24.32
Zn (μg g^{-1})	20.0	10.00±3.3	23.00±11.2	56.52	11.80±9.5	21.00±1.3	43.81

mango orchards should be enriched by organic as well as inorganic matter in order to make up the deficiency of the biologically important components and to rehabilitate the diseased plants. Application of animal manure and synthetic fertilizers (after plant and soil analysis because these are accepted methods for the determination of fertilizer requirement of citrus and other fruits) which may prove to be beneficial in this regard. It is also suggested that intensive cultivation of other crops in the mango orchards should be avoided.

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