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Nutrient imbalance indices are closely related with susceptibility of pomegranate to bacterial blight disease

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ABSTRACT

Bacterial blight disease caused by *Xanthomonas axonopodis* pv. *punicae* is a major threat to pomegranate cultivation causing heavy losses. An attempt was made to establish nutrient sufficiency range for imparting moderate disease resistance and diagnosing nutrient imbalance causing disease development. Two approaches i.e. Diagnosis and Recommendation Integrated System (DRIS) and Compositional Nutrient Diagnosis (CND) were considered to evaluate the nutrient balance status in pomegranate. Nutritional survey revealed that concentration of Ca, Mg, Mn and Cu in leaves were significantly high in moderately resistant germplasms, while N, K and S concentration were observed to be high in susceptible ones. Deficiency of Ca was observed as the cause of disease in 80% of the susceptible germplasms studied. The next important nutrient deficiencies in the susceptible germplasm were Cu (77.14%), Fe (77.14%), Mn (68.57%) and Mg (65.71%). Nutrient imbalance indices of susceptible germplasm diagnosed through DRIS and CND had linear relationship ($R^2 = 0.93$ and $R^2 = 0.82$) with bacterial blight disease severity however, DRIS approach is superior for diagnosis of nutrient imbalances. As per DRIS analysis, leaf nutrient status of N 1.56–2.05%, P 0.11–0.28%, K 0.83–1.20%, Ca 1.60–2.16%, Mg 0.38–0.82%, S 0.09–0.16% and micronutrients viz. Fe 132.50–187.00 mg kg^{-1}, Mn 31.60–58.40 mg kg^{-1}, Zn 13.20–27.40 mg kg^{-1} and Cu 26.00–47.80 mg kg^{-1} could result in imparting moderate resistance to bacterial blight disease. The nutrient norms were validated and found to have linear relation between nutrient imbalance indices and bacterial blight disease severity.

1. Introduction

Pomegranate (*Punica granatum* L.), being an ancient medicinal fruit of tropical and subtropical regions of the world has emerged as an important commercial fruit in many Indian states including Maharashtra, Karnataka, Andhra Pradesh, Tamil Nadu, Gujarat, Rajasthan and Uttar Pradesh (Mondal and Mani, 2009). In the past, variety 'Ganesh' was predominantly cultivated in the state of Maharashtra, Karnataka and Andhra Pradesh. Recently improved variety such as 'Bhagwa' bred out of 'Ganesh' and 'Gulesha Red' was introduced in Maharashtra, Karnataka and Andhra Pradesh where it cover more than 90 per cent of cultivated area (Mondal and Singh, 2009).

Intensive cultivation practices, lack of judicious nutrient management led to severe incidence of bacterial blight disease caused by *Xanthomonas axonopodis* pv. *punicae* (Xap), causing substantial damage to the pomegranate production and economic losses to the pomegranate growers (Mondal and Sharma, 2009). Various management options involving the application antibiotics, chemicals and cultural practices have been investigated. Treatment by only chemicals had limited success against the disease (Kumar et al., 2009). Presently, orchard health management (OHM) strategies were found to be effective to some extent. Improving plant resistance hold promise in OHM strategies for effective management of bacterial blight disease. Although plant disease resistance is genetically controlled (Agrios, 2005), it is affected by the environment and especially by nutrient deficiencies and toxicities (Marschner, 1995 Krauss, 1999). Information on nutrient status of diverse germplasm showing different degree of resistance to the disease could be utilized for developing nutrient norms for improving plant resistance against bacterial blight disease.

Foliar analysis has frequently been used to be an important tool to monitor nutrient status of plant. Of several approaches, two

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approaches, the Diagnosis and Recommendation Integrated System (DRIS) (Walworth and Sumner, 1987) and Compositional Nutrient Diagnosis (CND) (Parent and Dafir, 1992) were established according to the principle of nutrient balance in plant and both of them could provide an approach to reflect the balance, sufficiency or deficiency of plant nutrients. DRIS and CND are based on the bivariate and multivariate diagnosis respectively (da Silva et al., 2004). These two approaches are more precise than sufficiency range approach (SRA). However, no attempt has been reported so far to diagnose the deficiency or toxicities of nutrient responsible for making plant susceptible to bacterial blight disease in pomegranate.

The objectives of this research were: (i) to compare nutritional difference, if any, between moderately resistant germplasm and susceptible germplasm; (ii) to compare two different approaches (DRIS and CND) in diagnosis and evaluation of the nutrient status of pomegranate germplasm in relation to bacterial blight disease severity and in identifying the suitable form of these two approaches; (iii) to validate suitable approach in diagnosing nutrient status in relation to bacterial blight disease severity.

2. Materials and methods

2.1. Location and description

The study consisted of two parts: (1) establishment of nutrient norms for improving resistance in plant against bacterial blight disease, (2) validation of nutrient norms in the farmers' field. For the first part studied area was at pomegranate field gene bank, ICAR-National Research Centre on Pomegranate research farm, Solapur, Maharashtra State, India located at 17°48′N latitude and 75°91′E longitude, at an altitude of 457 m above mean sea level. And the second part was conducted at pomegranate orchards of the most susceptible cv. Bhagwa located in three talukas i.e. Mohol, Sangola and Pandharpur of Solapur district, Maharashtra, India. The area under the study was semi-arid, showing hot summer and moderate winter with a mean annual maximum and minimum temperature of 40.4 °C and 14.9 °C, respectively and the average annual rainfall of 694 mm (approximately), occurring mostly during the months of July–September with 35–60 numbers of rainy days. The soil of the field gene bank at ICAR-NRCP, Solapur is loamy-skeletal Typic Ustorthents and properties are shown in Table 1.

Orchard soils of Mohol, Sangola and Pandhar were loamy iso-hyperthermic Lithic Ustorthent. Soil characteristics of Mohol were: pH 8.0–8.4, free $CaCO_3$ 4.48–10.84%, organic carbon 0.83–1.59% and cation exchange capacity 14.6–17.8 cmol(p^+) kg^{-1} soil and those of Sangola were: pH 7.7–8.1, free $CaCO_3$ 7.47–22.72%, organic carbon 0.57–1.29%, and cation exchange capacity 12.8–15.6 cmol(p^+)kg^{-1} soil while those of Pandharpur were: pH 7.7–8.2, free $CaCO_3$ 5.56–16.84%, organic carbon 1.08–1.82% and cation exchange capacity 18.8–20.5 cmol(p^+)kg^{-1} soil.

2.2. Experimental layout

Sixty germplasm population comprising of IC318753, IC310790, IC318702, IC318712, IC318749, IC318740, IC1201, IC1203, IC318707, IC310728, IC318779, Accession no. 1, Accession no. 2, Accession no. 8, Accession no. 9, Accession no. 10, Accession no. 12, Accession no. 13, EC24686, EC104349, 1255, 1195, 1262, 1180, G526, P13, P16, P23, 17/2, Jodhpur collection, Double flower, Almoda, Bedanasuri, Kerala collection, Amlidana, Kalpitya, Yearcud HRS, Bedana thinsl, Bedanasri, Co-white, Gulesha red, Kabuli yellow, KRS, Pune collection, Sirin anar, Nimali, Dholka, Kasuri, Muskat, Spendanader, Bassain seedless, GR pink, Surat anar, Tabesta, Bas kalinsi, Dorsta, Spin sakaharir, Kabuli canoor, Damini, Bedanasedana was randomly planted in three blocks following randomized complete block design (RCBD) in the Pomegranate field gene bank at ICAR-National Research Centre on Pomegranate showing varying degree of response to bacterial blight disease (differing degree of severity) were studied during July to November 2014 in the *Mrigh bahar* (rainy season flowering) which coincided the peak period of bacterial blight disease incidence. Four years old trees were selected for the investigation. Bacterial blight disease severity was assessed and recorded four times during the period. The plants were spaced at 4.5 m and 3.0 m in between and within the rows, respectively. The plants were provided with routine cultural practices suitable for commercial pomegranate production including pruning, defoliation and irrigation uniformly for *Mrigh bahar* crop (i.e. June-July flowering). One-third of the recommended dose of N-P_2O_5-K_2O (625-250-250 g per plant) along with 40 kg farmyard manure were applied immediately after the harvest of the previous crop and the remaining two-thirds of the recommended dose of N-P_2O_5-K_2O were applied in two equal splits, one at 45 days after flowering and other at 120 days after flowering. The source of N, P_2O_5 and K_2O were urea, single super phosphate/di-ammonium phosphate, and sulphate of potash respectively. For validation of nutrient norms, seven pomegranate orchards cv. Bhagwa from each of the three talukas viz. Mohol, Sangola and Pandharpur were surveyed and bacterial blight disease severity were recorded with the collection of leaf samples.

2.3. Sampling and analysis

In the nutritional status survey, the 8th leaf pair from the non-fruiting new flush was collected in field gene bank as well as in farmers' orchards in Solapur district during August 2014 at flowering stage. The leaf samples were washed with tap water, deionized-water, 0.1 mol L^{-1} HCl and deionized-water (Alvarez-Fernandez et al., 2001). Then they were oven-dried at 65 °C for 48 h to constant weight and grounded with agate mortar. The samples were digested with H_2SO_4 to determine N and HNO_3/$HClO_4$ (v/v, 4:1) to determine P, K, Ca, Mg, S, Fe, Mn, Zn and Cu. The leaf N, P, K, Ca, Mg, S, Fe, Mn, Zn, Cu were measured with the methods of AOAC (2005).

2.4. Establishment of DRIS parameters

The calculation of DRIS is derived from Walworth and Sumner (1987). Field observation indicated that severity of disease on fruits is positively correlated with that on the foliage ($r = 0.76^*$). It was also observed that bacterial blight disease severity above 20% causes considerable yield loss ranging from 30–80% depending on the severity of disease and prevailing weather condition and hence, 20% disease severity was considered as cut off value for separating moderately resistant and susceptible subpopulation (Singh et al., 2015; Anonymous, 2009). Bacterial blight disease severity on foliage and leaf nutrient concentration built a databank, which was divided to moderately resistant (disease severity $\leq$20%) and susceptible (disease severity >20%) sub-populations based on their impact on yield loss. Mean, standard deviation and variance were calculated for each subpopulation. The mean values (in moderately resistant subpopulation) of 10 nutrients expression were selected as the diagnostic norms for imparting moderate resistance to the pomegranate plant and their respective variance ratios is listed in Table 2.

The DRIS norms established from the moderately resistant population of pomegranate germplasm were further employed to compute DRIS indices for the foliar mineral nutrient composition of susceptible germplasm and surveyed pomegranate orchards. DRIS