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Diagnostic Methods to Assess the Nutritional Status of the Carrot Crop

Luciel Rauni Dezordi[1], Leonardo Angelo de Aquino[2]*, Rosiane Filomena Batista de Almeida Aquino[3], Junia Maria Clemente[1] and Natália Silva Assunção[1]

[1] Universidade Federal de Viçosa, Instituto de Ciências Agrárias, *Campus* de Rio Paranaíba, Programa de Pós-graduação em Agronomia - Produção Vegetal, Rio Paranaíba, Minas Gerais, Brasil.
[2] Universidade Federal de Viçosa, Instituto de Ciências Agrárias, *Campus* de Rio Paranaíba, Rio Paranaíba, Minas Gerais, Brasil.
[3] Instituto de Pesquisa Agrícola do Cerrado, Rio Paranaíba, Minas Gerais, Brasil.

ABSTRACT: Leaf analysis may identify nutrient deficiency or excess in plant tissue. The aim of this study was to determine reference values and generate nutritional diagnosis from the results of leaf analysis of the carrot crop (*Daucus carota* L.) by the methods of Critical Level, Sufficiency Range, Diagnosis and Recommendation Integrated System, and Compositional Nutrient Diagnosis. Contents of N, P, K, Ca, Mg, S, B, Cu, Fe, Mn, and Zn were determined in 210 carrot fields in the 2012 and 2013 crop seasons in the Alto Paranaíba region, MG, Brazil. The whole plant shoot was sampled at harvest time to generate reference values for diagnosis. The high yielding subpopulation showed yield higher than 87.8 Mg ha^{-1}. The four diagnostic methods generated similar reference values of nutrients, but different from those found in the literature. Leaf diagnosis through nutrient content in the shoot at harvest time indicated Mn as the most limiting nutrient for growing carrot, followed by Mg, K, and Ca.

Keywords: *Daucus carota* L., leaf critical level, DRIS, CND.

*** Corresponding author:**
E-mail: leonardo.aquino@ufv.br

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INTRODUCTION

Interpretation of the results of leaf analysis may identify nutrient deficiency or excess in plant tissue, thus allowing adjustments in the crop fertilization program (Malavolta et al., 1997). Despite the importance and application of leaf analysis, adequate contents for carrot (*Daucus carota* L.) growing found in the literature are out of date as they do not consider high yielding crops (Malavolta et al. 1997; Hanlon and Hochmuth, 2009). In the Alto Paranaíba region, yields are high over 80 Mg ha^{-1} (Aquino et al., 2015), twice that suggested in official recommendations for the state of Minas Gerais, Brazil (CFSEMG, 1999).

The carrot crop has a short cycle (90-130 days) and continually puts forth leaves during the growing season. There are no phenotypic changes over the market cycle (it does not include the reproductive phase). Thus, establishing a phenological stage to collect leaf samples is complex. The youngest fully expanded or physiologically mature leaf, around 40 days after sowing, as the diagnostic leaf (Malavolta et al., 1997). However, climatic conditions and the cultivar may lead to distinct growth and nutrient uptake characteristics after this period of cultivation. Nevertheless, definition of leaf physiologically maturity (recommended for sampling) is difficult because the period among leaf emergence is short. In order to overcome these problems, sample collection time can be established at harvest time (Hanlon and Hochmuth, 2009), and instead of sampling specific leaves, the whole shoot of the plants can be collected. The standardization of leaf collection time minimizes factors that may interfere in interpretation of analyses, such as nutrient dilution and concentration (Malavolta et al., 1997).

The methods most used for interpretation of leaf analysis have been Critical Level (CL), Sufficiency Range (SR), Diagnosis and Recommendation Integrated System (DRIS), and Compositional Nutrient Diagnosis (CND) (Coelho et al., 2013; Gott et al., 2014). The CL method consists of mathematical models to establish the leaf nutrient contents that allow production of a certain fraction (90, 95, or 99 %) of maximum yield. The SR establishes an optimal range of leaf content in accordance with the average content of nutrients in the leaves of the most productive plants (or fields). Although these are classical methods and represent the main information contained in the literature (Camacho et al., 2012; Santos et al., 2013), they are univariate and analyze each nutrient separately.

The DRIS and CND methods stand out, in comparison to the CL and SR methods, through being bivariate and multivariate, respectively. Diagnosis and Recommendation Integrated System was developed by Beaufils (1973), and it is based on comparison of indices calculated in accordance with the mutual relationship between two nutrients. An advantage of this method is minimizing dilution or concentration effects because it is based on a relationship of equilibrium between nutrients (Beaufils, 1973; Jones, 1981). Furthermore, this method enables identification of nutritional imbalances even when all nutrients are above the critical level (Baldock and Schulte, 1996).

The Compositional Nutrient Diagnosis (CND) method, developed by Parent and Dafir (1992), differs from DRIS because the content of each nutrient in the sample is corrected in accordance with the geometric mean of nutritional composition. Therefore, CND is based on multiple interactions among all the nutrients under diagnosis.

The CND method initially presents advantages compared to DRIS since it considers the interaction of all nutrients simultaneously, and not only dual interactions (Parent and Dafir, 1992). The DRIS and CND methods have been proposed for nutritional diagnosis in crops such as soybean (Urano et al., 2007), cotton (Serra et al., 2010), sugarcane (Santos et al., 2013), and common bean plants (Partelli et al., 2014). However, diagnostic indices for leaf diagnosis in growing high yielding carrots have not yet been established.

Method bivariate (DRIS) or multivariate (CND) may be more effective in the univariate nutritional diagnosis (CL and SR). The objective of this study was to establish reference values through the methods of Critical Level (CL), Sufficiency Range (SR), Diagnosis and Recommendation Integrated System (DRIS), and Compositional Nutrient Diagnosis (CND) for interpretation of results of leaf analysis in the carrot crop, as well as to generate diagnosis of nutritional limitations.

MATERIALS AND METHODS

To determine reference values for diagnosis of nutritional status based on leaf nutrient content, a database with information from commercial carrot fields in the Alto Paranaíba region, MG, was created. Two hundred and ten commercial planting fields were sampled (155 in the winter and 55 in the summer) in the 2012 and 2013 crop seasons in areas ranging from 2 to 15 ha. The average duration of the crop growth cycle for summer and winter cultivars was 105 and 125 days, respectively.

The average application rates of N, P_2O_5, and K_2O were as follows: 118, 650, and 398 kg ha^{-1}, respectively. The main summer cultivars were Juliana and Poliana, and the winter cultivars were Baltimore, Belgrado, Maestro, Músico, Nancy, Nandrim, and Soprano. Winter is the preferred carrot growing season since it provides better weather conditions, which explains the larger area and number of cultivars.

Average altitude of the growing areas was around 1,100 m. The regional climate is classified as Cwa according to the Köppen-Geiger system, which is characterized by a dry season and a well-defined rainy season between October and March. The temperature and rainfall data during the sampling times for data collection are shown in the figure 1.

The soils were classifed as *Latossolos Amarelos*, *Vermelhos* and *Vermelho-Amarelo* (Oxisol), with clayey or very clayey texture. The vast majority of the fields under study have high fertility soils according to CFSEMG (1999). The physical and chemical properties of the soils are shown in table 1.

The database was composed of root yield and leaf nutrient content. Root yield in each field was obtained by harvesting four samples from 4 m of double rows, and their average weight was converted into tons per hectare. The time of sampling was that stipulated by farmers of the region based on growth, size standardization and firmness of the roots (an average of 105 days in the summer and 125 days in the winter).

Leaf collection consisted of the whole shoot at the time of harvest. Collection of the whole shoot was adopted to minimize sampling errors because the juxtaposed emergence of

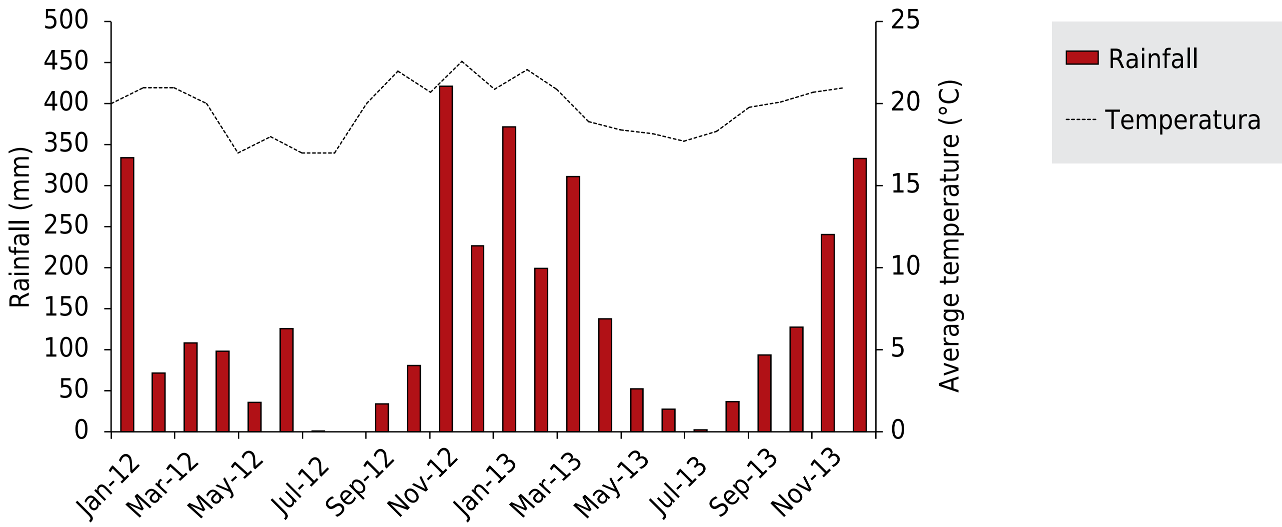

Figure 1. Average temperature and monthly rainfall in the Alto Paranaíba region during collection for database formation.

Table 1. Average and standard deviation of the main physical and chemical properties of the soil at the 0.00-0.20 m depth in carrot fields sampled in the 2012 and 2013 crop seasons

Property[(1)]	Average	Standard deviation
pH(H_2O)	6.3	0.3
Organic carbon (g kg^{-1})	20.0	0.3
P_{rem} (mg L^{-1})	10.6	3.2
P (mg dm^{-3})	28.0	15.1
K^+ ($mmol_c$ dm^{-3})	3.1	0.8
Ca^{2+} ($mmol_c$ dm^{-3})	33.9	5.8
Mg^{2+} ($mmol_c$ dm^{-3})	10.7	3.0
SO_4^{2-} (mg dm^{-3})	7.5	4.5
T ($mmol_c$ dm^{-3})	82.3	8.2
V (%)	58.0	7.0
B (mg dm^{-3})	0.52	0.21
Cu (mg dm^{-3})	2.5	1.4
Fe (mg dm^{-3})	38.0	12.2
Mn (mg dm^{-3})	3.2	2.3
Zn (mg dm^{-3})	6.8	3.0
Total sand (g kg^{-1})	25.7	5.7
Silt (g kg^{-1})	12.1	4.3
Clay (g kg^{-1})	62.2	12.1

[(1)] pH in water (1:2.5); Organic carbon: Walkley-Black method; P, K, Cu, Fe, Mn, and Zn: extractor Mehlich-1; Ca and Mg: KCl 1 mol L^{-1}; S: $Ca(H_2PO_4)_2.H_2O$ in AcOH; B: hot water; total sand, silt, and clay: pipette method.

leaves hampers the individualization of each leaf position on the plant. Futhermore, the effects of nutrient dilution and concentration are minimized by collection at harvest time due to greater leaf dry matter (Hanlon and Hochmuth, 2009).

The leaves were washed and rinsed in deionized water, followed by drying in an air circulation laboratory oven at 70 °C for 72 h. They were then ground in a Willey mill equipped with a 1.27 mm sieve. Nutrient content was determined according to methods described by Malavolta et al. (1997). Contents of N, P, K, Ca, Mg, S, B, Cu, Fe, Mn, and Zn and root yields were critically analyzed (outlier analysis) and processed to generate reference values for leaf diagnoses by the CL, SR, DRIS, and CND methods.

To determine the CL, the yield data were organized in ascending order and grouped into 14 classes (number and classes = $\sqrt{n}$; in which "n" is the number of samples).

The maximum root yield obtained among the 14 classes was 111.7 Mg ha^{-1} and this was used to fit regressions of CL_s. Thus, the CL was established for the yield of 100.5 Mg ha^{-1}, which is 90 % of the maximum yield. The interval composing the classes was obtained by dividing the total amplitude of the nutrient by the number of classes. Subsequently, linear mathematical models were fitted to study the relationship between the leaf content of each nutrient and the yield of the 14 classes, as performed by Gott et al. (2014). Each nutrient was evaluated separately from others.

The significance of the parameters of the mathematical models was evaluated by the *t* test at 5 % probability. When the slope of the linear regression was significant by the *t* test and positive, the CL was estimated as the content corresponding to 90 % of the maximum yield (Gott et al., 2014). When the slope of the linear regression was negative and/or not significant, the CL was considered as the average content of the nutrient in the database.

To establish standard scores or norms by the RS, DRIS, and CND methods, the population was divided into two subpopulations based on root yield. The threshold of these classes was defined as the average root yield plus $^2/_3$ of the standard deviation of this variable, which corresponded to 87.8 Mg ha^{-1}. After the data normality test, the subpopulation with the greatest yield was considered nutritionally balanced or a reference for establishment of norms/standard scores for interpretation of the methods. The subpopulation with high yield was represented by 64 fields (30.5 % of the fields) and low yield by 146 fields (69.5 % of the fields).

The average ($\bar{x}$) and standard deviation (s_x) of the leaf nutrient contents were estimated in the high yielding population. The threshold levels of the SR were defined by the average content plus or minus the standard deviation of this variable (FS = $\bar{x} \pm S_{\bar{x}}$), as performed by Gott et al. (2014). As for the CL, each nutrient was evaluated separately from the others.

The mean (x) and variance (s^2) of the dual relationships of the leaf nutrient contents were determined by the DRIS method, which composed the DRIS norms. The relationships were obtained from the ratio of macro- and micronutrient contents in g kg^{-1} and mg kg^{-1}, respectively. The functions of the dual relationships were obtained as described by Jones (1981) and chosen through the F method (Letzch, 1985):

$$f(X/Y) = \frac{(X/Y - x/y)}{s}$$

$$f(Z/X) = \frac{(Z/X - z/x)}{s}$$

in which X = nutrient for which the index is to be calculated; Y_1, Y_2, ..., Y_n = nutrients that are in the denominator of relationships with nutrient X; Z_1, Z_2, ..., Z_m = nutrients that are in the numerator of relationships with nutrient X; Z/X = relationship between the contents of the nutrients Z and X of the sample to be subjected to DRIS; X/Y = relationship between the contents of the nutrients X and Y of the sample to be subjected to DRIS; z/x = average relationship between the contents of the nutrients Z and X provided by DRIS norms; x/y = average relationship between the contents of the nutrients X and Y provided by DRIS norms; and s = standard deviation of the dual relation in the reference population.

The DRIS index for each nutrient was calculated in accordance with the equation proposed by Beaufils (1973):

$$Index_{DRIS}X = \frac{[f(X/Y_1) + f(X/Y_2) + ... + f(X/Y_n)] - [f(Z_1/X) + f(Z_2/X) ... + f(Z_m\ X)]}{n + m}$$

in which X/Y and Y/X = functions of the dual relationships chosen for calculation of the DRIS index; m = number of functions in which the nutrient X is in the denominator; and n = number of functions in which the nutrient X is in the numerator.

For establishment of the Compositional Nutrient Diagnosis (CND), the proposal of Khiari et al. (2001) was adopted, which includes adaptations of the original model as described by Parent and Dafir (1992):

$$V_{xi} = \ln \frac{X_i}{G}$$

in which V_X = multinutrient variable for the nutrient X; X_i = leaf concentration of the nutrient to be analyzed (mg kg^{-1}); and G = geometric mean of dry matter constituents.

To calculate the geometric mean (G), the following equation was used:

$$G = (X_1.X_2. ... .X_n.R)\ \frac{1}{n+1}$$

in which G = geometric mean of dry matter constituents; X_1, X_2, ..., Xn = concentration of the nutrients in the dry matter (%); R = residue or concentration of the unmeasurable nutrients in the dry matter (%); and n = number of nutrients evaluated.

The CND index for each nutrient was calculated by the following equation:

$$CND_{xi} = \frac{VX_i + VX'}{S_{X'}}$$

in which CND_{Xi} = CND index for the nutrient; VX_i = multinutrient variable of the field evaluated; VX' = average of the multinutrient variable of the reference population; and $S_{X'}$ = standard deviation of the multinutrient variable of the reference population.

For fields that composed the reference population, optimum leaf content was obtained by the DRIS and CND methods through fitting a regression equation between the nutrient index (independent variable) and its respective leaf content (dependent variable). Subsequently, nutrient content was obtained in the leaf that provides a null or balanced index.

The optimum range for each nutrient was established by leaf contents corresponding to ± $^2/_3$ indexes, the normal ranges of Beaufils (Urano et al., 2007; Santos et al., 2013). The order of nutritional limitation was generated in two forms: in accordance with the optimum range of leaf content or in accordance with fertilization response potential. From the optimum range, the order of nutritional limitation was estimated for the SR, DRIS, and CND methods, as well as from existing literature for purposes of comparison. From the optimum content ranges of each method and from leaf nutrient concentration in the samples, the percentage of fields in imbalance (deficient or excessive) for each nutrient and diagnostic method was established.

Nutritional restriction was considered as deficiency or excess when the leaf nutrient content was below or above the optimum range established by diagnostic methods, respectively. The order of nutritional limitation by deficiency was generated in descending order of nutrients in regard to the frequency of fields under deficiency (DRIS and CND). The order of nutritional limitation was also generated as a function of potential response to fertilization for DRIS and CND. In this case, the fields were first placed in five groups in accordance with fertilization response potential (FRP), namely: positive, positive or zero, zero, negative or zero, and negative (Wadt et al., 1998).

Positive FRP occurs when the DRIS or CND index of a nutrient is negative and greater in magnitude than the medium DRIS or CND. Positive or zero FRP occurs when the DRIS or CND index is negative and its module is larger than the mean DRIS or CND; however, it is not the nutrient with the lowest index. Zero FRP occurs when the DRIS or CND index is negative, but the module is lower than the mean DRIS or CND. Negative or zero FRP occurs when the nutrient index is positive and lower than the mean DRIS or CND. Negative FRP occurs when the nutrient index is positive and simultaneously the greatest of the indexes and greater than the mean DRIS or CND. Subsequently, the frequency of fields grouped into classes with positive and positive or zero response for each nutrient was calculated. In these classes of fertilization response, the nutrient can be considered as limiting for cultivation.

Field rankings in regard to nutritional status were performed by grouping the fields in three classes for each nutrient, namely: limiting by deficiency (LD), limiting by excess (LE) and not limiting (NL). The ranking was also generated by two different methods: as a function of the optimum content range and of the fertilization response potential.

In regard to the first method, those fields that presented leaf contents below or above the range limits were classified as LD or LE, respectively. When the nutrient content was in the range of the interval, it was considered NL for the field. The ranking of fertilization response potential was conducted for DRIS and CND. The fields with positive and positive or zero response were classified as LD for the respective nutrient. The fields with zero response were classified as NL, and the other classes of fertilization response (negative and negative or zero) were grouped in the LE class (Urano et al., 2006).

For both ranking methods (optimum range or fertilization response potential), the frequency of fields and average yield of each class (LD, LE, and NL) were presented for all nutrients, and leaf diagnosis methods (SR, DRIS, and CND) and data presented in the literature (Hanlon and Hochmuth, 2009) were evaluated. The *t* test was carried out for comparing the mean yields of the LD and LE classes with the yield of the NL class.

The calculations for determining reference values (content and optimum ranges) by the CL, SR, DRIS, and CND methods were carried out using Microsoft Excel® software. The significance of the parameters of the fitted equations was analyzed by the *t* test at 5 % probability by SAS software 8.2.

RESULTS AND DISCUSSION

Analysis of chemical properties of the soil (Table 1) showed that acidity was corrected (pH in water 6.3) in addition to adequate P contents, available S, and exchangeable cations (average concentration of P and K in the very high fertility class, and medium contents of Ca, Mg, and S in the high fertility class), according to CFSEMG (1999).

In contrast, the soils showed imbalances in micronutrients since the average Mn content was classified as low, B as medium, Fe as adequate, and Cu and Zn as high, in accordance with the classification proposed by CFSEMG (1999). Moreover, the soils had medium contents of organic C (2 dag kg^{-1}) and low remaining P – P-rem (10.6 mg L^{-1}). The low P-rem value shows that the soils of the region are highly buffered for this nutrient, i.e., it is necessary to add large amounts to the soil to increase the P fraction available to the plants (Bedin et al., 2003; Broggi et al., 2011).

Regression equations with positive and significant slope for K, B, and Mn were fitted; thus, CL was established for these nutrients in the crop shoots. The CL for K was 49.7 g kg^{-1}, 56.2 mg kg^{-1} for B, and 79.9 mg kg^{-1} for Mn (Table 2). It was not possible to fit a regression equation with significant parameters for the CL of Ca, Mg, and Cu. Thus, the CLs of these nutrients were established as their average contents in the database. The higher proportion of roots/leaves in high yielding cultivars along with higher Ca partition to leaves may explain the lack of fit of a positive regression for Ca content.

High yielding plants may have lower Ca content in the leaves due to lower transpiration (smaller number of leaves) and higher allocation of Ca in the roots. Magnesium is allocated primarily in the roots and its content in leaves may decline with increasing yield (Cecílio Filho and Peixoto, 2013).The equations fitted to describe the leaf content of N, P, S, Fe, and Zn as a function of root yield were decreasing, i.e. with a slope lower than zero. Thus, the CLs of these nutrients were also defined as the average of their contents in the database. The fitting of decreasing functions means that there were dilution effects of the contents of these nutrients in the leaves as yield increased.

The root yield of the reference class for the SR, DRIS, and CND methods ranged from 87.8 to 120.5 Mg ha^{-1}, with an average of 98.2 Mg ha^{-1}. Averages and variances of the dual relationships (DRIS norms) of the high yielding population (>87.8 Mg ha^{-1}) are shown in table 3 and the CND norms in table 4.

Among the mathematical models to describe the relationship between indices (DRIS and CND) and nutrient contents in leaf (Tables 4 and 5), quadratic models obtained the best fit for cationic micronutrients (except for Zn in the CND index), whereas for the other nutrients, linear models were fitted. As for S, it was not possible to fit a significant mathematical model by the CND method (Table 4).

Table 2. Leaf content corresponding to 90% of maximum yield (CL), mathematical model fitted to describe leaf content as a function of yield, and coefficient of determination of the models

Nutrient	CL	Mathematical model	R^2
	g kg^{-1}		
N	21.5	N = 24.187 - 0.0351* yield	0.393
P	1.8	P = 2.2978 - 0.0065** yield	0.529
K	49.7	K = 33.881 + 0.1573** yield	0.484
Ca	27.1	Ca = 27.1	-
Mg	2.9	Mg = 2.9	-
S	2.3	S = 2.8311 - 0.0072* yield	0.442
	mg kg^{-1}		
B	56.2	B = 45.16 + 0.1101* yield	0.309
Cu	89.0	Cu = 89.0	-
Fe	1,019.7	Fe = 2,387.6 - 18.147** yield	0.797
Mn	79.9	Mn = 8.5441 + 0.7098** yield	0.543
Zn	47.1	Zn = 67.621 - 0.2726** yield	0.659

* and **: significant by the *t* test at 5 and 1 %, respectively.

The contents and optimum ranges of the nutrients in the carrot shoots at harvest (Table 6) had little influence on the diagnostic method. In regard to optimum content, small variations were observed between adequate levels obtained by CL compared to DRIS and CND for cationic micronutrients. In this case, the CL established optimum contents with concentrations greater than the other methods. This is due to CL being more sensitive than CND to uncontrolled effects of environment, such as nutrient concentration or dilution in plant tissue (Wadt, 2008). Included in the calculation method of CND is the effect of variations in dry matter on interpretation of nutrient contents, correcting them and thus reducing the effects of dilution or concentration (Parent and Dafir, 1992).

An additional explanation for the difference in contents of cationic micronutrients between the methods is the high variability (coefficient of variation >46 % - data not shown) obtained for the content of these micronutrients in the plant tissue. Since the DRIS and CND methods are not univariate, i.e., they do not analyze each nutrient individually, they are not highly influenced by the variability of leaf nutrient concentrations. For the other nutrients (macronutrients and B), there were practically no differences between the optimum contents established by the CL, DRIS, and CND methods.

The sufficiency range of leaf nutrients, intervals and amplitudes were similar for the SR and DRIS methods, except for Fe (Table 6). The CND method showed narrower ranges that were included more in the optimum range established by SR and DRIS. For Fe, the SR established optimum range with an amplitude greater than DRIS, and these methods, for their part, established wider ranges than the CND. The smaller amplitudes of optimum ranges generated by CND favored more accurate diagnosis of the nutritional status of the carrot plants (Gott et al., 2014). However, small ranges generated for some nutrients may lead to wrong diagnosis due to error that arises in the analysis of determination of leaf content (laboratory errors).

Optimum ranges of leaf contents established by Hanlon and Hochmuth (2009) for carrot growing in the United States have higher upper limits for N and P compared to the limits of the present study. However, for K, Ca, and B, the optimum range stated in this study have higher limits (upper and lower) than those indicated by Hanlon and Hochmuth (2009). In regard to Mg, the optimum range proposed by these authors is wider and encompasses the range generated by SR, DRIS, and CND in this study.

Table 3. Average (AVER) and variance (VAR) of the dual relationships of leaf content in carrot shoots (DRIS) for the high yield (<87.8 Mg ha^{-1}) population

Relationship	AVER	VAR	Relationship	AVER	VAR	Relationship	AVER	VAR	Relationship	AVER	VAR
N/P	13.814	14.846	K/Mn	0.729	0.305	S/Cu	0.023	0.004	Zn/Ca	1.564	0.770
N/K	0.458	0.034	K/Fe	0.102	0.005	S/Zn	0.034	0.002	Zn/Mg	11.805	43.555
N/Ca	0.788	0.029	Ca/N	1.329	0.084	S/Mn	0.018	0.001	Zn/S	12.742	293.992
N/Mg	5.963	3.165	Ca/P	18.193	35.774	S/Fe	0.002	<0.001	Zn/B	0.705	0.219
N/S	5.896	42.026	Ca/K	0.601	0.071	B/N	2.562	0.908	Zn/Cu	0.571	0.525
N/B	0.366	0.019	Ca/Mg	7.84	8.100	B/P	35.758	278.659	Zn/Mn	0.519	0.327
N/Cu	0.36	0.211	Ca/S	8.082	94.257	B/K	1.146	0.299	Zn/Fe	0.074	0.003
N/Zn	0.697	0.180	Ca/B	0.471	0.029	B/Ca	1.974	0.514	Mn/N	3.372	5.917
N/Mn	0.316	0.062	Ca/Cu	0.487	0.392	B/Mg	15.059	42.525	Mn/P	45.209	1130.76
N/Fe	0.041	0.001	Ca/Zn	0.934	0.452	B/S	14.819	358.403	Mn/K	1.396	0.833
P/N	0.078	0.001	Ca/Mn	0.415	0.115	B/Cu	0.827	1.112	Mn/Ca	2.581	3.087
P/K	0.035	<0.001	Ca/Fe	0.052	0.001	B/Zn	1.804	1.801	Mn/Mg	20.107	233.941
P/Ca	0.061	<0.001	Mg/N	0.165	0.002	B/Mn	0.793	0.450	Mn/S	19.035	1109.632
P/Mg	0.462	0.033	Mg/P	2.281	0.869	B/Fe	0.104	0.005	Mn/B	1.171	0.855
P/S	0.522	0.361	Mg/K	0.075	0.001	Cu/N	4.363	10.973	Mn/Cu	1.018	1.701
P/B	0.028	<0.001	Mg/Ca	0.128	0.001	Cu/P	57.251	1834.378	Mn/Zn	2.17	2.027
P/Cu	0.029	0.002	Mg/S	1.017	1.476	Cu/K	1.924	2.999	Mn/Fe	0.13	0.012
P/Zn	0.052	0.001	Mg/B	0.059	0.001	Cu/Ca	3.329	6.908	Fe/N	29.302	471.231
P/Mn	0.023	<0.001	Mg/Cu	0.056	0.005	Cu/Mg	25.418	446.499	Fe/P	371.250	47914.983
P/Fe	0.003	<0.001	Mg/Zn	0.11	0.005	Cu/S	29.117	1700.404	Fe/K	12.606	88.537
K/N	2.492	0.706	Mg/Mn	0.051	0.002	Cu/B	1.538	1.823	Fe/Ca	22.252	289.453
K/P	34.052	208.852	Mg/Fe	0.007	<0.001	Cu/Zn	2.541	4.589	Fe/Mg	172.272	18040.712
K/Ca	1.933	0.472	S/N	0.054	0.003	Cu/Mn	1.203	2.252	Fe/S	224.475	119443.43
K/Mg	14.544	36.832	S/P	0.652	0.514	Cu/Fe	0.175	0.030	Fe/B	9.417	44.776
K/S	12.708	226.583	S/K	0.028	0.001	Zn/N	2.014	1.286	Fe/Cu	11.634	486.387
K/B	0.865	0.139	S/Ca	0.043	0.002	Zn/P	27.243	307.305	Fe/Zn	19.179	363.234
K/Cu	0.932	1.725	S/Mg	0.344	0.151	Zn/K	0.893	0.390	Fe/Mn	8.013	62.902
K/Zn	1.653	1.043	S/B	0.018	<0.001	-	-	-	-	-	-

For Mn and Zn, the ranges obtained by SR, DRIS, and CND in this study were wider and encompassed the optimum ranges obtained in the United States. However, there was high discrepancy between the optimum ranges obtained by Hanlon and Hochmuth (2009) and those generated by the methods of SR, DRIS, and CND for Cu and Fe. For these two nutrients, the contents indicated by the authors are lower than those obtained in this study. This disagreement demonstrates the importance of having regionalized standard scores for leaf diagnosis, or generated for each growing condition (Serra et al., 2010; Gott et al., 2014). Similar conclusion regarding the importance of regionalization of optimum leaf nutrient levels for the Pera orange and sugarcane crops, respectively, was came by Camacho et al. (2012) and Santos et al. (2013).

The content and the optimum ranges generated by different methods (CL, SR, DRIS, and CND) were consistent for nearly all the nutrients in this study. Similar results were obtained by René et al. (2013), who proposed leaf diagnostic rates by the CL, DRIS, and CND

Table 4. Compositional Nutrient Diagnosis (CND) norms (average - AVE, and standard deviation - SD) of the multinutrient variables (Vi) and geometric mean of nutritional composition (G), mathematical model, amplitude of the indices, and coefficient of determination (R^2) of the regressions fitted to describe the leaf content of carrot shoot as a function of the CND index for the population of reference (<87.8 Mg ha^{-1})

Variable	AVE	SD	Mathematical Model	Amplitude			R^2
V_N	1.373	0.22	$N = 20.792 + 0.7918^{*}I_N$	-2.05	$< I_N <$	1.79	0.920
V_P	-1.215	0.30	$P = 1.6181 + 0.3441^{**}I_P$	-2.65	$< I_P <$	2.32	0.491
V_K	2.224	0.33	$K = 50.64 + 11.208^{**} I_K$	-2.67	$< I_K <$	2.19	0.608
V_{Ca}	1.634	0.26	$Ca = 27.158 + 2.5796^{**}I_{Ca}$	-2.05	$< I_{Ca} <$	2.70	0.335
V_{Mg}	-0.424	0.25	$Mg = 3.5117 + 0.4139^{**}I_{Mg}$	-2.31	$< I_{Mg} <$	2.59	0.349
V_S	-0.966	0.40	$S = 2.1456 - 0.0214^{ns}I_S$	-2.31	$< I_S <$	2.51	0.010
V_B	-2.242	0.26	$B = 55.448 + 5.5405^{**}I_B$	-2.05	$< I_B <$	2.15	0.363
V_{Cu}	-1.914	0.75	$Cu = 82.383 + 72.541^{**}I_{Cu} + 17.554^{**} I_{Cu}^2$	-2.83	$< I_{Cu} <$	1.63	0.896
V_{Fe}	-2.697	0.45	$Fe = 570.77 + 303.3^{**}I_{Fe} + 61.453^{**} I_{Fe}^2$	-1.92	$< I_{Zn} <$	3.00	0.840
V_{Mn}	-2.007	0.46	$Mn = 69.337 + 41.098^{**}I_{Mn} + 9.2941^{**} I_{Mn}^2$	-2.46	$< I_{Mn} <$	1.79	0.879
V_{Zn}	0.036	0.48	$Zn = 41.796 + 21.811^{**}I_{Zn}$	-1.82	$< I_{Fe} <$	2.14	0.855
G	0.530	0.11	-	0.30	$< G <$	0.85	-

ns: not significant by the *t* test; * and **: significant by the *t* test at 5 and 1 %, respectively

Table 5. Mathematical model, range of indices and coefficient of determination of regressions (R^2) fitted to describe the leaf contents in carrot shoots as a function of the Diagnosis and Recommendation Integrated System (DRIS) index for the high yield (<87.8 Mg ha^{-1}) population

Mathematical model	Amplitude			R^2
$N = 20.805 + 3.5396^{**}I_N$	-0.91	$< I_N <$	0.79	0.333
$P = 1.6212 + 0.7795^{**}I_P$	-1.19	$< I_P <$	1.58	0.705
$K = 50.694 + 21.439^{**} I_K$	-1.39	$< I_K <$	1.49	0.798
$Ca = 27.193 + 6.7308^{**}I_{Ca}$	-1.08	$< I_{Ca} <$	1.54	0.557
$Mg = 3.5164 + 1.17^{**}I_{Mg}$	-1.17	$< I_{Mg} <$	1.27	0.635
$S = 2.1225 + 1.0281^{**}I_S$	-1.69	$< I_S <$	1.01	0.778
$B = 55.504 + 17.349^{**}I_B$	-1.07	$< I_B <$	0.96	0.408
$Cu = 81.539 + 91.848^{**}I_{Cu} + 26.236^{**}I_{Cu}^2$	-2.32	$< I_{Cu} <$	1.47	0.911
$Fe = 592.04 + 468.41^{**}I_{Fe} + 98.705^{**}I_{Fe}^2$	-1.5	$< I_{Fe} <$	2.34	0.887
$Mn = 67.348 + 66.716^{**}I_{Mn} + 24.112^{**}I_{Mn}^2$	-1.76	$< I_{Mn} <$	1.2	0.914
$Zn = 37.646 + 33.067^{**}I_{Zn} + 9.3153^{**}I_{Zn}^2$	-1.41	$< I_{Zn} <$	1.53	0.931

**: significant by the *t* test at 1 %.

methods for poplar hybrid (*Populus maximowiczii*) and concluded that the methods generate consistent indices. Among the reasons that led to consistency in the ranges proposed by different diagnostic methods, the high yields of carrot obtained in the Alto Paranaíba region, MG, and the large number of fields sampled stand out.

The yield potential of the carrot crop is from 100 to 120 Mg ha^{-1} (Cecílio Filho and Peixoto, 2013). These high yields may be related to high soil fertility and the nutrient balance conditions of the crop. It is possible that most of these fields were nutritional balanced because to establish standard scores for leaf analysis by SR, DRIS, and CND, the reference population covered areas with yields higher than 87.8 Mg ha^{-1} (and reached 120.5 Mg ha^{-1}).

Possibly the use of bi- or multivariate methods compared to univariate methods in generating reference values in fields with high yield is not justified. High yields require such nutrient balance where effects of concentration, dilution, and relationship among nutrients are less expressive. Therefore, simpler methods, such as the CL and SR, can be used for accurate diagnosis. Moreover, the number of fields sampled

Table 6. Content and optimum range for the concentration of nutrients in the shoots of the carrot crop grown in the Alto Paranaíba region at the time of harvest, generated by the Critical Level (CL), Sufficiency Range (SR), Diagnosis and Recommendation Integrated System (DRIS), and Compositional Nutrient Diagnosis (CND) methods, in comparison to values in the literature

Nutrient	Optimum content			Optimum range			
	CL	DRIS	CND	SR	DRIS	CND	Literature[1]
	g kg^{-1}						
N	21.5	20.8	20.8	18.2 - 23.4	18.5 - 23.1	20.3 - 21.3	22.0 - 40.0
P	1.8	1.6	1.6	1.1 - 2.1	1.1 - 2.1	1.4 - 1.8	3.0 - 7.0
K	49.7	50.7	50.6	36.3 - 65.0	36.5 - 64.8	43.2 - 58.0	15.0 - 30.0
Ca	27.1	27.2	27.2	22.7 - 31.6	22.8 - 31.6	25.5 - 28.9	10.0 - 20.0
Mg	2.9	3.5	3.5	2.8 - 4.2	2.7 - 4.3	3.2 - 3.8	2.5 - 6.0
S	2.3	2.1	2.1	1.4 - 2.8	1.4 - 2.8	1.7 - 2.6	-
	mg kg^{-1}						
B	56.2	55.5	55.5	46.3 - 64.6	44.1 - 67.0	51.8 - 59.1	30.0 - 50.0
Cu	89.0	81.5	82.4	35.9 - 164.0	32.3 - 153.6	42.2 - 137.9	3.0 - 5.0
Fe	1,019.7	592.0	570.8	248.0 – 1,016.5	325.9 - 944.2	397.4 - 797.7	30.0 - 60.0
Mn	79.9	67.3	69.3	36.5 - 120.7	33.8 - 121.9	46.3 - 100.5	50.0 - 80.0
Zn	47.1	37.6	41.8	18.2 - 65.4	19.9 - 63.5	37.4 - 66.2	30.0 - 50.0

[1] Hanlon and Hochmuth (2009).

in this study (210 plots) may have contributed to obtaining similar results among the diagnostic methods evaluated. More representative sampling of carrot growing conditions (management practices and climate) may have reduced the influence of various effects that justify the use of bi- or multivariate methods.

For common bean (*Phaseolus vulgaris* L.), Partelli et al. (2014) found that methods based on nutrient balance (DRIS and CND) showed high agreement in leaf diagnoses, though different from those obtained by SR. These authors evaluated 55 commercial fields and used those with yields higher than 2.7 Mg ha^{-1} as a reference population. The potential yield of the bean crop in Brazil estimated by the Blackman method is 6 Mg ha^{-1} (Oliveira et al., 2011). The low number of fields evaluated (compared to this study) and the limit yield of the reference class far below the maximum potential estimated for the crop, may have influenced the results obtained by Partelli et al. (2014).

There is consensus among the methods (SR, DRIS, and CND) that Mn and Mg are the most limiting nutrients for carrot growing in the general and low yield populations (Table 7). After these nutrients, K and Ca are considered limiting for production. However, these elements were not highlighted by all the diagnostic methods in sequence to Mn and Mg. Therefore, one can induce the need to increase K, Ca, and Mg supply for carrot cultivation. However, because of the limitation caused by Mn, the supply of K, Ca, and Mg should be made from sources that do not alter the pH of the soil. Increasing pH in this case would worsen the deficiency limitation posed by Mn.

The Soil Fertility Commission of Minas Gerais suggests at least 8 mmol dm^3 of Mg^{2+} in the soil for carrot cultivation (CFSEMG, 1999). In the soils sampled, the Mg^{2+} content was above this critical level (Table 1), and it was still the macronutrient most limiting to cultivation. In addition to content in the soil, another factor which must be taken into account is cation ratios. The average ratio of Ca:Mg and Mg:K was 3.2:1 and 3.5:1, respectively (Table 1). The limitation from Mg deficiency was greater than that of Ca and K for the carrot crop (Table 7). Future studies should investigate whether lower Ca:Mg and greater Mg:K ratios may provide nutritional balance conditions for Mg and increased yield. Among the factors that may have contributed to limitation of carrot growing by Mn deficiency, the low content of this nutrient in the soil may be highlighted (Table 1). In addition, lime application resulted in an increase in pH, which, combined with high

Table 7. Order of nutritional limitation by deficiency for the carrot crop in the Alto Paranaíba region, MG, generated by the Compositional Nutrient Diagnosis (CND), Diagnosis and Recommendation Integrated System (DRIS), and Sufficiency Range (SR) methods, compared to the literature

Method	Order of nutritional limitation
	General population
CND(1)	Mn > Mg > K > Ca > Zn > Cu > B > S > N > Fe > P
DRIS(1)	Mn > Mg > K > Ca > Fe > Cu > S > Zn > P > B > N
CND(2)	Mg > Mn > K > B > N > Zn > Ca > P > Cu > S > Fe
DRIS(2)	Mg > Mn > K > Ca > N > B > P > Cu > Fe > S > Zn
SR(2)	Mg > Mn > B > K > Ca > Cu > N > P > S > Zn > Fe
Literature(2)	P > N > Mn > Mg > Zn > Cu > B > K = Ca = Fe
	High yield population (>87.8 Mg ha^{-1})
CND(1)	Zn > B > Fe = Ca > S > N = P > Mg > Mn > K > Cu
DRIS(1)	Fe > Zn > K > P > Ca > Mg > S > B > N > Mn > Cu
CND(2)	Zn > N > Mg > K = Ca > B > P > Fe > S > Mn > Cu
DRIS(2)	K = Ca > Zn > N > Fe > S > Mn > Cu > B > Mg > P
SR(2)	B > K = Ca > Cu > Zn > N > S > Mn > Mg > P > Fe
Literature(2)	P > N > Zn > Mn > Mg > K = Ca = B = Cu = Fe
	Low yield population (<87.8 Mg ha^{-1})
CND(1)	Mn > Mg > K > Ca > Cu > S > B > N > Zn > P > Fe
DRIS(1)	Mn > Mg > K > Ca > Fe > Cu > S > P > B > N > Zn
CND(2)	Mg > Mn > B > K > Ca > N > Zn > P > Cu > S > Fe
DRIS(2)	Mg > Mn > K > Ca > B > N > Cu > P > S > Fe > Zn
SR(2)	Mg > Mn > K > B > Ca > Cu > N > P > S > Fe > Zn
Literature(2)	P > Mn > N > Mg > Zn > Cu > B > K = Ca = Fe

(1) Ranking generated as a function of fertilization response potential; Ranking generated as a function of optimum leaf content; (2) Hanlon and Hochmuth (2009).

rates of P at sowing (average of 640 kg ha^{-1} of P_2O_5), reduced Mn availability. High pH and high P application rates are factors that lead to reduced Mn availability to plants (Moreira et al., 2006; Gonçalves et al., 2011).

The importance of nutritional limitations caused by Fe and Zn was not significant, and the importance of B and Cu was moderate (Table 7). The lesser importance of these micronutrients compared to Mn can be explained by their levels in the soil (Table 1). In accordance with the classification proposed by CFSEMG (1999), Zn and Cu contents are considered high in the soil, while those of B and Fe are moderate and good, respectively.

Analysis of the order of nutritional limitation (deficiency or excess) in the high yield population (>87.8 Mg ha^{-1}) does not designate the nutrients most limiting for carrot cultivation. The order of limitation varied as a result of the method of diagnosis. There was no agreement among the optimum ranges for most of the nutrients established in this study and those proposed by Hanlon and Hochmuth (2009). Thus, there was also no agreement among the orders of limitation obtained from the diagnostic indices generated in this study compared to the contents considered adequate by Hanlon and Hochmuth (2009).

The SR and DRIS methods showed higher frequency of fields as NL (non-limiting) compared to the frequency shown in the CND (Table 8). In contrast to this study, Serra et al. (2010) found that CND classified a higher number of fields as NL compared to DRIS for cotton plants (*Gossypium hirsutum* var. *latifolium* Hutch). In regard to the manners of interpretation (fertilization response potential [FRP] and optimum range) for DRIS and CND, the FRP classified a small number of fields as limiting (LD, limiting by deficiency or LE, limiting by excess) compared to the classification given by the optimum range of leaf contents.

In relation to average yield of the classes of nutrient limitation for K, Mg, and Mn, a tendency for the fields classified as LE to exhibit the highest yields was observed. Therefore, it is possible that adequate concentrations and ranges of these elements in carrot leaves are truly higher than those obtained in the present study. The diagnostic methods used may have underestimated the optimum range because the fields exhibited a general limitation of these nutrients (Table 7). An opposite tendency was observed for Fe and Zn, in which fields grouped as LD exhibited the highest yields (Table 8). This result may indicate that these elements reached toxic levels for carrot and caused yield reduction. However, visual symptoms of this possible excess were not observed.

Table 8. Frequency of fields and average yield of the classes limiting by deficiency (LD), non limiting (NL) and limiting by excess (LE) generated by the Compositional Nutrient Diagnosis (CND), Diagnosis and Recommendation Integrated System (DRIS), and Sufficiency Range (SR) methods, in comparison to the literature

Nutrient	Method	Nutritional Status (%)			Yield (Mg ha^{-1})		
		LD	NL	LE	LD	NL	LE
N	CND(1)	12.1	51.8	36.1	88.1*	74.1	73.6
	DRIS(1)	9.6	70.7	19.7	90.4*	74.5	72.2
	CND(2)	38.9	10.6	50.5	78.3	78.7	72.8*
	DRIS(2)	21.2	43.7	35.1	72.2	82.6	68.9*
	SR	20.2	49.0	30.8	71.1*	82.4	67.7*
	Literature(3)	55.8	44.2	0.0	79.6*	70.2	-
P	CND(1)	9.6	48.5	41.9	92.7*	76.7	70.0*
	DRIS(1)	11.9	51.0	37.1	85.5*	76.6	70.4*
	CND(2)	27.6	29.5	42.9	74.0*	83.3	70.9*
	DRIS(2)	15.7	52.9	31.4	65.2*	82.3	68.9*
	SR	19.1	48.4	32.5	65.2*	82.3	68.9*
	Literature(3)	92.4	7.6	0.0	76.4*	64.4	-
K	CND(1)	25.8	59.0	15.2	73.9	73.6	84.9*
	DRIS(1)	30.4	54.8	14.8	74.7	72.8	86.5*
	CND(2)	43.3	35.3	21.4	72.8	72.5	85.3*
	DRIS(2)	29.5	61.0	9.5	74.0	74.4	86.2*
	SR	29.5	61.0	9.5	74.0	74.4	86.2*
	Literature(3)	0.0	16.2	83.8	-	70.0	76.5
Ca	CND(1)	18.1	59.6	22.3	78.4	76.0	71.1*
	DRIS(1)	23.8	64.8	11.4	74.1	75.7	76.5
	CND(2)	28.1	35.2	36.7	74.4*	78.0	73.7*
	DRIS(2)	28.6	48.5	22.9	74.0*	77.9	71.7*
	SR	28.6	48.5	22.9	74.0*	77.9	71.7*
	Literature(3)	0.0	14.3	85.7	-	67.7	76.7*
Mg	CND(1)	32.2	58.6	9.2	71.7*	74.7	91.6*
	DRIS(1)	31.2	63.5	5.3	71.5*	75.4	96.3*
	CND(2)	58.0	29.3	12.7	70.4*	78.3	90.8*
	DRIS(2)	37.1	58.0	4.9	64.2*	80.5	97.5*
	SR	42.9	49.8	7.3	65.1*	80.6	97.7*
	Literature(3)	32.4	67.6	0.0	62.9*	80.1	-
S	CND(1)	12.8	61.1	26.1	80.0*	71.3	68.5*
	DRIS(1)	12.8	69.0	18.2	77.6*	70.3	72.8
	CND(2)	21.8	47.3	30.9	74.8	73.0	67.5*
	DRIS(2)	12.1	67.3	20.6	72.4	73.3	65.9*
	SR	12.8	64.0	23.2	72.4	73.3	65.9*
	Literature(3)	-	-	-	-	-	-

Continue

Table 8. Continuation

B	CND[1]	13.7	71.1	15.2	82.8*	72.0	81.4*
	DRIS[1]	10.3	81.9	7.8	82.8*	73.3	81.2*
	CND[2]	43.1	28.5	28.4	69.8*	80.6	76.9*
	DRIS[2]	20.1	67.2	12.7	66.2*	77.5	75.0
	SR	29.9	54.9	15.2	70.0*	77.1	76.6
	Literature[3]	0.5	38.2	61.3	59.3*	69.6	78.3*
Cu	CND[1]	13.9	71.7	14.4	71.0*	74.4	80.3*
	DRIS[1]	14.4	66.1	19.5	71.8	74.3	78.4*
	CND[2]	24.6	54.9	20.5	69.9*	76.0	77.3
	DRIS[2]	18.5	67.1	14.4	66.6*	76.6	76.5
	RS	22.1	68.2	9.7	70.0*	75.4	80.7*
	Literature[3]	3.6	1.5	94.9	56.2*	53.4	75.8*
Fe	CND[1]	10.1	51.8	38.1	94.5*	77.0	70.6*
	DRIS[1]	20.8	41.5	37.7	87.0*	77.0	69.8*
	CND[2]	21.3	37.2	41.5	86.0*	76.2	71.5*
	DRIS[2]	13.1	52.5	34.4	83.3*	79.1	69.5*
	RS	2.2	66.7	31.1	81.8	79.7	68.9*
	Literature[3]	0.0	0.0	100.0	-	-	76.4
Mn	CND[1]	44.7	36.5	18.8	67.0*	79.5	83.2*
	DRIS[1]	42.6	37.6	19.8	67.1*	78.5	83.5*
	CND[2]	50.3	29.4	20.3	67.8*	81.8	81.3
	DRIS[2]	36.5	50.3	13.2	66.0*	78.7	83.1*
	RS	39.6	46.7	13.7	66.5*	78.9	83.7*
	Literature[3]	52.3	20.3	27.4	68.5*	82.2	80.7
Zn	CND[1]	14.7	48.1	37.2	89.5*	77.4	69.8*
	DRIS[1]	12.5	57.1	30.4	93.3*	71.6	74.8*
	CND[2]	37.2	44.9	17.9	81.5*	72.2	71.5
	DRIS[2]	8.7	69.6	21.7	91.6*	75.2	70.2*
	RS	6.3	75.3	18.4	94.8*	74.8	72.1*
	Literature[3]	26.6	28.0	45.4	84.1*	74.4	71.3*

[1] Ranking generated as a function of fertilization response potential; [3] Ranking generated as a function of optimum leaf content; *: different from the yield of the NL class by the *t* test at 5 %; [3] Hanlon and Hochmuth (2009).

The frequency of fields of the LD, NL, and LE classes generated with data from the literature (Hanlon and Hochmuth, 2009) showed different values compared to the results of the SR, DRIS, and CND methods. As observed for the differences obtained in the optimum leaf ranges and order of nutritional limitation, these results support the idea that is necessary to regionalize reference values to carry out leaf diagnosis of the carrot crop.

It was not clear which method of leaf diagnosis (SR, DRIS, and CND) and, or, means of interpretation (optimum range or FRP) can rank the fields in LD, LE, and NL classes in such a way that significant differences can be detected in the yields of these groups (Table 8). However, all the methods tended to provide consistent results for interpreting nutritional status through optimum content or range of nutrients in the leaf tissue.

CONCLUSIONS

The univariate, bi and multivariate methods are equally effective in nutricional diagnosis of carrot crop.

The most limiting nutrients in carrot crop were Mn, Mg, K and Ca.

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