Compositional Nutrient Diagnosis (CND) Standards for *Opuntia ficus-indica* (L.) Miller Variety 'Rojo Pelón' Fruiting

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ABSTRACT

In this study, the aim was to develop the compositional nutrient diagnosis (CND) norms for *Opuntia ficus indica* variety 'Rojo Pelón' fruiting. Statistical analyses involved a dataset of fruit yield and macro-nutrient concentrations of 228 1-year old cladodes from healthy plants. A cutoff yield (1,166.67 g cladode^{-1}) between the low- and high-yield subpopulations was determined after examining six cumulative variance ratio functions related to yield per 1-year old cladode. Means and standard deviations of row-centered log-ratios V_X^* of five nutrients (N, P, K, Ca, and Mg) and a filling value R_d, which includes all nutrients not chemically analyzed. Estimated preliminary CND norms (mean ± standard deviation of the row-centered log-ratios) are: $V_N^* = -1.114 \pm 0.219$, $V_P^* = -2.194 \pm 0.076$, $V_K^* = 0.163 \pm 0.259$, $V_{Mg}^* = -0.708 \pm 0.157$, $V_{Ca}^* = 0.401 \pm 0.100$, and $V_{Rd}^* = 3.452 \pm 0.095$. These CND standards are associated with the following 1-year old fruiting cladode mean concentrations: N=0.958%, P=0.318%, K=3.507%, Ca=4.228%, and Mg=1.448%. Then, the order of macro-nutrient requirements is as follows: Ca>K>Mg>N>P.

Keywords: Macro-nutrients; Fruit yield per cladode; Nitrogen; Phosphorus; Potassium; Calcium; Magnesium.

INTRODUCTION

Nutrient status in plants may be identified through tissue analysis. Chemical analysis of tissue of reference allows identifying optimum nutrient concentrations or nutrient norms (or standards) linked to maximize crop yields. So, nutrient diagnosis taking into account

identified nutrient optimum concentrations as a reference may allow estimating nutrient imbalances or disorders. Commonly, nutrient disorders are not observable, e.g. hidden hunger or toxicity (Barker and Pilbeam, 2015). The observable symptoms of nutrient imbalance often appear when significant and irreversible damage has been generated; these damages may cause scarce growth and low crop yield. Therefore, the opportune determination of nutrient status through correct diagnosis under the basis of nutrient norms could result in practical recommendations that increase yields and improve fruit quality (Habib, 2000; Mostashari et al., 2018).

There are widely known nutrient norms developed locally that may allow for correct field diagnoses useful to improve plant nutrient status through agricultural practices such a foliar or soil fertilization (Blanco-Macías et al., 2010). Some researchers have shown the precision in diagnosing imbalances when using locally developed norms (Bendaly Labaied et al., 2018). Nutrient norms can be developed according to various techniques. The most known techniques are the Critical Value Approach (CVA) (Bates, 1971), the Diagnosis and Recommendation Integrated System (DRIS) (Walworth and Sumner, 1987), and the Compositional Nutrient Diagnosis (CND) (Parent and Dafir 1992; Blanco-Macías et al., 2006; García-Hernández et al., 2006).

The CVA method does not consider interactions between or among mineral nutrients; these interactions may be important issues for plant nutrient balance. On the other hand, the DRIS and CND approaches involve nutrient interactions (Bhaduri and Pal, 2013). Various authors indicate that there are little differences between DRIS and CND for establishing nutrient norms (Khiari et al., 2001b; Barłóg, 2016; Mostashari et al., 2018; Barros de Morais et al., 2019; Bendaly Labaied et al., 2020). Nonetheless, other researchers endorse the CND arguing that it is more efficient to determine the nutritional status of crops because of its sound mathematical and statistical bases (Kumar et al., 2003; Blanco-Macías, 2009; René et al., 2013; Valdez-Cepeda et al., 2013; Barros de Morais et al., 2019).

Up to now, CND norms for nutrient status diagnosis have been developed for various crops such as *Carya illinoinensis* (Wangenh.) K. Koch (García-Hernández et al., 2009), *Aloe vera* L. (García-Hernández et al., 2006), *Coffea arabica* L. and *Coffea canephora* Pierre ex A. Froehner (Wairegi and Van Asten, 2012), *Phoenix dactylifera* L. (Bendaly Labaied et al., 2020), *Vitis vinifera* L. (Mostashari et al., 2018), *Eucalyptus grandis* W. Hill ex Maid. (Costa da Silva et al., 2004; Barros de Morais et al., 2019), *Zea mays* L. (Khiari et al., 2001a; Magallanes-Quintanar et al., 2006), *Musa* spp. AAA (Wairegi and Van Asten, 2011), and *Opuntia ficus-indica* L. Miller (Magallanes-Quintanar et al., 2004; Blanco-Macías et al. 2006; Valdez-Cepeda et al., 2013), among others. Recently, the Boundary-Line Approach (B-LA) was used by Hernández-Vidal et al. (2021) to estimate macro-nutrient standards for *O. ficus-indica* (L.) Miller variety 'Rojo Pelón' fruiting; the estimated optimum concentrations were N=1.02%, P=0.304%, K=3.518%, Ca=3.665%, and Mg=1.383% as linked to maximum fruit yield that varies between 1901.13 and 1984.41 g cladode^{-1}. However, the B-LA does not consider interactions between mineral nutrients like the CNA does.

In the case of *O. ficus-indica*, the CND norms have been determined for dry and fresh matter production (Magallanes-Quintanar et al., 2004; Blanco-Macías et al., 2006; Valdez-Cepeda et al., 2013). In fact, the species *O. ficus-indica* is an important crop around the world (López-García et al., 2016). It is widely used for vegetable production in Mexico and fruit production in at least 25 countries around the world (Valdez-Cepeda et al., 2013). Hitherto, *Opuntia ficus-indica* L.variety 'Rojo Pelón' is being used for fruit production in Mexico's Northern-Central Region. Despite its social and economic importance, there is little information about the nutrient requirements of this variety; this means that its CND norms remain unknown yet. Therefore, this work aimed to develop CND norms for *O. ficus-indica* variety 'Rojo Pelón' for fruit production.

MATERIALS AND METHODS

Study site
This research was carried out in the experimental field of the 'Centro Regional Universitario Centro Norte' of the 'Universidad Autónoma Chapingo'. It is located at 22° 44' 49.6'' North latitude, 102° 46' 28.2'' West longitude, and 2 296 masl, near the Zacatecas City, Mexico. The regional climate is classified as BS1kw (w), with an average annual temperature that varies between 12°C and 18°C, and an average annual rainfall of 472 mm. Most of the precipitation (65%) occurs from June to August.

An orchard was established in 2006, using 20 mother cladodes. The plant density was 625 plants ha^{-1}. Then, 20 trees with a natural vessel-shaped structure were growing. The management of the orchard consisted of removing weeds each year at the end of spring and summer through minimum tillage. Fertilization, irrigation, and other agronomic practices were not performed.

Experimental data
To develop CND norms, a data set of nutrient concentrations and fruit yield per 1-year old cladodes was used. A sample of two hundred twenty-eight fruiting cladodes of *O. ficus-indica* variety 'Rojo Pelón' and a total of 1744 fruits were considered in this research. All cladodes were selected from the uppermost part of the trees to ensure they were 1-year old. Fruiting cladodes and their fruits were taken for four consecutive years (2012-2015). Sampling involved the selection of cladodes having from 1 to 15 fruits, and four cladodes having each of these numbers of fruits were selected when possible. Sampled cladodes with each number of fruits were taken from sections of the plant oriented to North, South, East, and West when most of the fruits showed peel color breakage.

Weights of each cladode and their fruits were registered. Besides, all 228 detached fruiting cladodes were cleaned with distilled water and immediately weighted. Afterward, the detached cladodes were cut into slices and dehydrated to constant weight in an oven at 75°C for 36 hours, and then their dry weights were registered.

Dry tissue of cladode samples was milled and then digested with a mix of acids. Afterward, these samples were used to determination of nutrient concentrations. The N concentration was determined by the Kjeldahl method, whereas that the P content was estimated by reduction with the molybdo-vanadate technique using an optical photo spectrometer (Thermo Spectronic, Helios Epsilon model, USA®). The K, Ca, and Mg concentrations were determined with an atomic absorption spectrophotometer (UNICAM Solar model 9626).

Theory of the CND approach
The following description of the theory of the CND approach was modified from Khiari et al. (2001). Plant tissue composition forms a *d*-dimensional nutrient arrangement, *i.e.*, simplex (S^d) made of *d*+1 nutrient proportions including *d* nutrients and a filling value defined as (Parent and Dafir, 1992):

$$S^d = \left[(N, P, K,..., R_d) : N > 0, P > 0, K > 0,..., R_d > 0, N + P + K + ... + R_d = 100 \right], \quad (1)$$

where 100 is the dry matter concentration (%); *N*, *P*, *K*, … are nutrient proportions computed as:

$$R_d = 100 - (N + P + K + ...). \quad (2)$$

The nutrient proportions become scale invariant after they are divided by the geometric mean (*G*) of the $d + 1$ components, including R_d (Aitchinson, 1986), as follows:

$$G = \left[N \cdot P \cdot K \cdot ... \cdot R_d \right]^{\frac{1}{d+1}}. \quad (3)$$

Row-centered log ratios are computed as:

$$V_N = ln\left(\frac{N}{G}\right), V_P = ln\left(\frac{P}{G}\right), V_K = ln\left(\frac{K}{G}\right), ..., V_{R_d} = ln\left(\frac{R_d}{G}\right), \text{ and} \quad (4)$$

$$V_N + V_P + V_K + ... + V_{R_d} = 0, \quad (5)$$

where V_X is the CND row-centered log-ratio expression for nutrient *X*. The sum of tissue components is 100%, as in Equation 1, and the sum of their row-centered log ratios, including the filling value must be zero, as in Equation 5.

Thereafter, the database is partitioned between two subpopulations using the Cate-Nelson procedure, once the observations have been ranked in a decreasing yield order (Khiari et al., 2001ab). In the first partition, the two highest yield values form one group (group A) and the remainder of yield values forms another group (group B); thereafter, the three highest yield values form the group A. This process is repeated until the two lowest yield values form group B, and the remainder of yield values forms the group A. At each iteration, group A comprises n_1 observations, and group B comprises n_2 observations for a total of *n* observations ($n = n_1 + n_2$) in the whole database. For the two subpopulations obtained during

each iteration, one must compute the variance of the CND V_X values. Then the variance ratio for component X can be estimated as:

$$f_i(V_X) = \frac{Variance\ of\ V_X\ n_1\ observations}{Variance\ of\ V_X\ n_2\ observations},$$
(6)

where $f_i(V_X)$ is the variance ratio function between two subpopulations for nutrient X at the ith iteration $(i = n_i - 1)$ and the V_X is the CND row-centered log-ratio expression for nutrient X. The first variance ratio function computed for the two highest yields is put on the same line as the highest yield, and so on, thus leaving three empty bottom lines.

The cumulative variance ratio function is the sum of variance ratios at the ith iteration from the top. The cumulated variance ratios for a given iteration are computed as a proportion of the total sum of variance ratios across all iterations to compare the discrimination power of the V_X between low- and high-yield subpopulations on a common scale. The cumulative variance ratio function $F_i^C(V_X)$ can then be computed as:

$$F_i^C(V_X) = \left[\frac{\sum_{i=1}^{n-1} f_i(V_X)}{\sum_{i=1}^{n-3} f_i(V_X)}\right] \cdot [100],$$
(7)

where $n-1$ is the partition number and n is the total number of observations $(n_1 + n_2)$. The denominator is the sum of variance ratios across all iterations and is a constant for nutrient X. The cumulative function $F_i^C(V_X)$ related to yield (Y) shows a cubic pattern:

$$F_i^C(V_X) = aY^3 + bY^2 + cY + d.$$
(8)

The inflection point is the point where the model shows a change in concavity. It is obtained by the second derivation of Equation 8:

$$\frac{\partial F_i^C(V_X)}{\partial Y} = 3aY^2 - 2bY + C$$
(9)

$$\frac{\partial^2 F_i^C(V_X)}{\partial Y^2} = 6aY - 2b.$$
(10)

The inflection point is then obtained by equating the second derivative Equation 10 to zero. Therefore, the solution for the yield cutoff value is $-b/3a$. The highest yield cutoff value across nutrient expressions can be selected to determine what minimum yield target for a high-yield subpopulation will be classified as high yield, whatever the nutrition expression. CND norms are computed using means and standard deviations corresponding to the row-

centered log-ratios V_X of d nutrients for high-yield specimens, that is, V_N^*, V_P^*, V_K^*, …, V_R^*, and SD_N^*, SD_P^*, SD_K^*, …, SD_R^*, respectively.

Once CND norms have been developed, an independent database can validate them. Validation of CND norms has been reported by Parent and Dafir (1992), Parent et al. (1994), and Khiari et al. (2001ab). CND norms can also be used for diagnostic purposes:

$$I_N = \frac{\left(V_N - V_N^*\right)}{SD_N^*}, I_P = \frac{\left(V_P - V_P^*\right)}{SD_P^*}, I_K = \frac{\left(V_K - V_K^*\right)}{SD_K^*}, ..., I_{R_d} = \frac{\left(V_{R_d} - V_{R_d}^*\right)}{SD_{R_d}^*}, \tag{11}$$

where I_N, …, I_{R_d} are the CND indices.

Independence among compositional data is ascertained by row-centered log-ratio transformation (Aitchison, 1986). CND indices, as defined by Equation 11, are standardized and linearized variables as dimensions of a circle ($d + 1 = 2$), a sphere ($d + 1 = 3$), or a hypersphere ($d + 1 > 3$) in a $d + 1$ dimensional space. The CND nutrient imbalance index of a diagnosed specimen is its CND r^2 and is computed by:

$$r^2 = I_N^2 + I_P^2 + I_K^2 + ... + I_{R_d}^2. \tag{12}$$

Its radius, r, computed from the CND nutrient indices, thus characterizes each specimen. The sum of $d + 1$ squared independent, unit-normal variables produces a new variable having a chi-square distribution with $d + 1$ degrees of freedom (Ross, 1987). Because CND indices are independent, unit-normal variables, the CND r^2 values must have a chi-square distribution function. This is why it is recommended that the highest yield cutoff value (highest discrimination power) among $d + 1$ nutrient computations be retained to calculate the proportion of the low-yield subpopulation below yield cutoff used as a critical value for the chi-square cumulative distribution function. As defined by Eqs. [11] and [12], the closer to zero that CND indices and thus the CND r^2 or chi-square values are, the higher the probability to obtain a high yield. Theoretically, at the critical chi-square value of zero where the ideal nutrient balance is reached, 100% of the population would be expected to produce low target yields when a high critical chi-square value is set.

RESULTS AND DISCUSSION

Descriptive statistics
In this research work, fruit yield per cladode and macro-nutrient concentrations in 1-year old cladodes of *O. ficus-indica* variety 'Rojo Pelón' was studied to define its CND standards.

Basic statistic estimators of fruit yield per cladode and macro-nutrient concentrations in 1-year old fruiting cladodes can be appreciated in Table 1.

Table 1. Basic statistics of *Opuntia ficus-indica* L. Miller variety 'Rojo Pelón' fruit yield per cladode and N, P, K, Ca, and Mg concentrations in 1-year old fruiting cladodes (*n* = 228).

Statistic	Yield (g cladode^{-1})	N (%)	P (%)	K (%)	Ca (%)	Mg (%)
Mean	795.16	1.167	0.286	3.206	3.845	1.365
Standard Deviation	469.79	0.358	0.055	1.169	0.947	0.285
Coefficient of Variation (%)	59.08	30.690	19.380	36.480	24.620	20.880
Minimum	60.00	0.600	0.156	1.312	1.300	0.594
Maximum	2186.00	2.170	0.420	6.536	6.350	2.150

The estimated fruiting cladode mean nutrient concentrations are N = 1.167%, P = 0.286%, K = 3.206%, Ca = 3.845%, and Mg = 1.365%, whereas the mean fruit yield per cladode is 795.16 g. Besides, results suggest that fruit yield per cladode show high variability (CV=59.08%), P, Mg, and Ca concentrations have moderate variability (CV=19.38%, CV=20.88%, and CV=24.62%, respectively), and K and N show high variability (CV=36.48% and CV=30.69%, respectively). Variability is an important issue to get the planned objective, that is, our database (*n*=228) can be used to identify the CND standards for *O. ficus-indica* variety 'Rojo Pelón' fruiting.

The compositional nutrient diagnosis norms for simplex S^5

The S^5, i.e., six-dimensional (d + 1) *O. ficus-indica* variety 'Rojo Pelón) simplex comprised the five nutrients N, P, K, Ca, and Mg and the filling value R. The R values were estimated using Eq. [1]. Nutrient concentrations were transformed into CND row-centered log ratios V_N, V_P, V_K, V_{Ca}, and V_{R_d} through Eqs. [2 to 4]. Eq. [6] was used to estimate the $F_i^C(V_X)$ values.

The descriptive statistics of the yield were as follows: mean = 795.16 g cladode^{-1}, minimum = 60 6 cladode^{-1}, maximum = 2186 g cladode^{-1}, and a standard deviation = 469.79 g cladode^{-1} (Table 1). The cutoff yield between the low- and high-yield subpopulations was determined after examining the six cumulative variance ratio functions [$F_i^C(V_N)$, $F_i^C(V_P)$, $F_i^C(V_K)$, $F_i^C(V_{Ca})$, $F_i^C(V_{Mg})$ and $F_i^C(V_R)$] related to yield per 1-year old cladode (Table 2). All six relationships showed a cubic pattern with inflection points at –*b/3a*. Yield cutoff values were 200 g cladode^{-1} for $F_i^C(V_N)$, 1,166.67 g cladode^{-1} for $F_i^C(V_P)$, 333.33 g cladode^{-1} for $F_i^C(V_K)$, 1,666.67 g cladode^{-1} for $F_i^C(V_{Ca})$, 23,333.33 g cladode^{-1} for $F_i^C(V_{Mg})$, and 33.33 g cladode^{-1} for $F_i^C(V_R)$. The theory of the CND approach recommends that the highest yield cutoff value (highest discrimination power) among *d* + 1 nutrient computations be retained to calculate

the proportion of the low-yield subpopulation below yield cutoff used as the critical value for the chi-square cumulative distribution function. Notably, the highest value (23,333.33 g cladode[-1]) and the lowest value (33.33 g cladode[-1]) are out of the explored yield range, so each was not considered a target yield. This flaw has been found previously by several researchers (e.g. Khiari et al., 2001ab; Magallanes-Quintanar et al., 2004; García-Hernández et al., 2005, 2007; Magallanes Quintanar et al. 2006ab; Hernández-Caraballo et al., 2008). To avoid such a flaw, Valdez-Cepeda et al. (2013) recommended using the unrestricted Boltzmann equation instead of the traditional cubic function.

Table 2. Fruit yields of *Opuntia ficus indica* variety 'Rojo Pelón' at inflection points (–b/3a) of cumulative variance functions [$F_iC(V_X)$] for row-centered log-ratios (*n*=225) in the survey population (*n*=228).

Nutrient	$F_i{}^c(V_x)= aY^3+bY^2+cY+d$	R^2	Yield at –b/3a (g cladode[-1])
N	$F_i{}^c(V_N) = -5E{-}09x^3 + 3E{-}06x^2 - 0.0236x + 100$	0.99	200.00
P	$F_i{}^c(V_P) = 2E{-}08x^3 - 7E{-}05x^2 - 0.0019x + 100$	0.99	1,166.67
K	$F_i{}^c(V_K)= -3E{-}09x^3 + 3E{-}06x^2 - 0.0078x + 100$	0.98	333.33
Ca	$F_i{}^c(V_{Ca}) = 6E{-}09x^3 - 3E{-}05x^2 - 0.0026x + 100$	0.99	1,666.67
Mg	$F_i{}^c(V_{Mg})= 1E{-}10x^3 - 7E{-}06x^2 - 0.0118x + 100$	0.98	23,333.33
R	$F_i{}^c(V_R)= -6E{-}09x^3 + 6E{-}07x^2 - 0.0206x + 100$	0.99	33.33

As a consequence, 1,166.67 g cladode[-1] was used to define the high-yield subpopulation. This implies that 22.37% of the population (51 observations) is considered as the high-yielding subpopulation, while the low-yield sub-population includes 77.63% of the population (177 observations). This result agrees with those from other researchers, who found that a high-yielding subpopulation represents a lower percentage of the whole population (e.g. Parent et al., 1994; Magallanes-Quintanar et al., 2004; Blanco-Macías et al., 2006; García-Hernández et al., 2004, 2006; Blanco-Macías et al., 2009).

As a remarkable result, the preliminary CND norms as means and standard deviations (V_X^* and SD_X^*) of the CND row-centered log-ratios for the high-yield (>1,166.67 g cladode[-1]) subpopulation, as well as their corresponding nutrient optimum ranges (means and standard deviations) in 1-year old fruiting cladodes, are shown in Table 3. The estimated mean concentrations are N=0.958%, P=0.318%, K=3.507%, Mg=1.448%, and Ca=4.228%. As a remarkable result, the order of nutrient requirements is as follows Ca> K> Mg> N> P. This means that *O. ficus-indica* variety 'Rojo Pelón' plants tend to concentrate more Ca and K than N in their 1-year old fruiting cladodes, confirming they are calcitrophic organisms (Lüttge, 2004). This result also indicates that they may have a high nitrogen use efficiency (Raven and Spicer, 1996).

Table 3. The preliminary compositional nutrient diagnosis (CND) norms (V_X^* means and their standard deviations) for $d = 5$ nutrients in a high-yield (>1,166.67 g cladode⁻¹) subpopulation of *Opuntia ficus-indica* (L.) Miller variety 'Rojo Pelón', and their corresponding nutrient optimum ranges (mean nutrient concentrations and their standard deviations) in 1-year old fruiting cladodes.

Row-Centered Log-Ratio	Mean	Standard Deviation	Nutrient	Mean (%)	Standard Deviation (%)
V_N^*	-1.11463	0.21916	N	0.95823529	0.20446717
V_P^*	-2.19444	0.07689	P	0.31807843	0.03920808
V_K^*	0.16356	0.25908	K	3.50713725	0.93183052
V_{Mg}^*	-0.70845	0.15711	Mg	1.44847059	0.28891351
V_{Ca}^*	0.40159	0.10077	Ca	4.2284902	0.62320226
V_{R5}^*	3.45238	0.09502			
$\sum^* V_X$	0.00000				

The estimated mean nutrient concentrations (Table 3) differ slightly from those proposed by Hernández-Vidal et al. (2021) as optimum concentrations for *O. ficus-indica* (L.) Miller variety 'Rojo Pelón' fruiting through the B-LA. There can be noted that estimated N and K mean concentrations linked to the CND standards are slightly lower than the optimum concentrations estimated by Hernández-Vidal et al. (2021) (0.958% versus 1.02%, and 3.507% versus 3.518%, respectively). On the other hand, the calculated P, Mg, and Ca mean concentrations associated with the CND standards are higher than the optimum concentrations proposed by Hernández-Vidal et al. (2021), that is, 0.318% versus 0.304%, 1.448% versus 1.383, and 4.228% versus 3.665%, for P, Mg, and Ca, respectively. Those differences may be due to we used a target fruit yield =1,166.67 g cladode⁻¹ whereas that Hernández-Vidal et al. (2021) linked their estimations to maximum fruit yield that varies between1,901.13 and 1,984.41 g cladode⁻¹. In other words, we used a target fruit yield lower than those considered by Hernández-Vidal et al. (2021). Nonetheless, our results have the advantage of involving multivariate nutrient ratios through the CND approach whereas those from Hernández-Vidal et al. (2021) do not.

The preliminary CND norms were used to estimate nutrient indices I_N, I_P, I_K, I_{Ca}, I_{Mg}, and I_{Rd} through Eq. [11]. Also, CND r² values were computed using Eq. [12]. The CND r² values were distributed like chi-square values ($R^2 > 0.988$; $p < 0.001$; Figure 2). Eighty-five percent of the observations were below the yield cutoff of 1,166.67 g cladode⁻¹, and the corresponding chi-square value was 2.5. Then, this value must be considered when validating the preliminary CND norms because the independent dataset ought to be characterized by a similar value. By taking into account that more observations from high-yielding cladodes or specimens must be added to the database, the chi-square value of 5.1 could change due to the high-yielding subpopulation may provide more weight for defining

yield cutoff than the low-yielding subpopulation according to the theory of the CND approach.
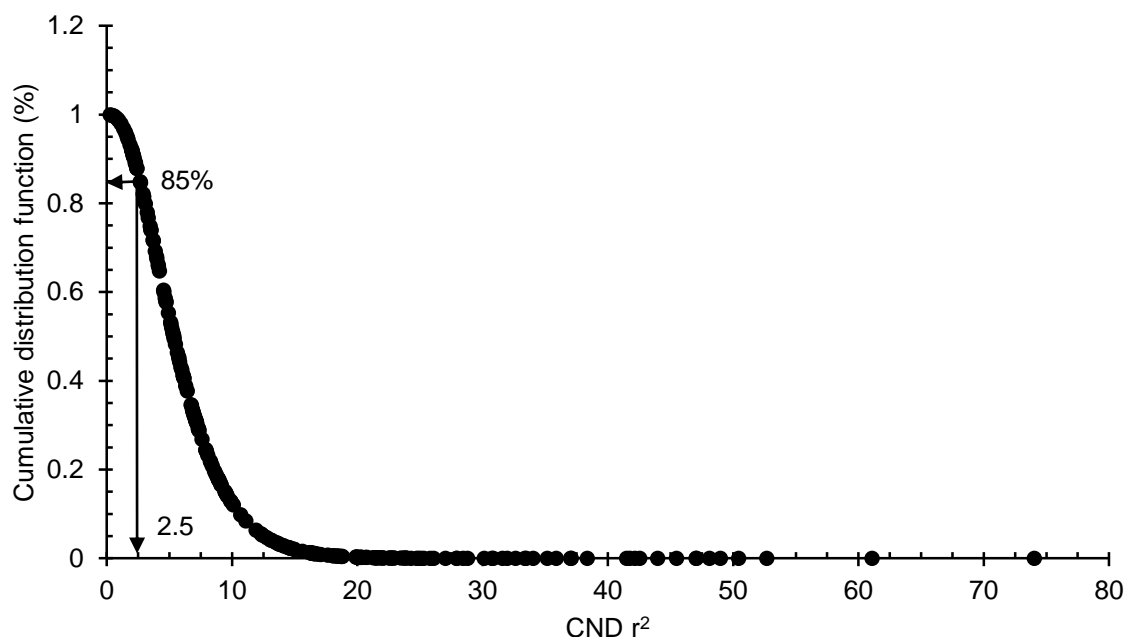
Figure 1. The chi-square cumulative distribution function with 6 degrees of freedom for obtaining theoretical threshold compositional nutrient diagnosis (CND) r² value (2.5) in S^5 for a yield cutoff at 85% of the low-yield subpopulation.

CONCLUSIONS

This is the first study to our knowledge carried out to estimate Compositional Nutrient Diagnosis (CND) standards for *Opuntia ficus-indica* (L.) Miller variety 'Rojo Pelón' fruiting taking into account a target yield of 1,166.67 g cladode^{-1} or higher. The proposed preliminary CND norms (mean ± standard deviation of the row-centered log-ratios) are: $V_N^* = -1.114 \pm 0.219$, $V_P^* = -2.194 \pm 0.076$, $V_K^* = 0.163 \pm 0.259$, $V_{Mg}^* = -0.708 \pm 0.157$, $V_{Ca}^* = 0.401 \pm 0.100$, and $V_{Rd}^* = 3.452 \pm 0.095$. These CND standards are associated with the following 1-year old fruiting cladode mean concentrations: N=0.958%, P=0.318%, K=3.507%, Ca=4.228%, and Mg=1.448%. Then, the order of macro-nutrient requirements is as follows: Ca>K>Mg>N>P. Future works should be focused on validation of these macro-nutrient norms taking into account a database involving more high-yielding cladodes or specimens to change the estimated chi-square value of 5.1 and to provide more weight for defining a yield cutoff to divide the population into high- and low- yield subpopulations. Also, this process could be improved by increasing the population and estimating the target yield through the unrestricted Boltzmann equation to describe the relationship between each cumulative variance ratio function and the yield per fructification cladode.

ETHICS STATEMENT

Not apply.

CONSENT FOR PUBLICATION

Not apply.

AVAILABILITY ON SUPPORTING DATA

Data might be available upon request addressed to RD V-C.

COMPETING INTERESTS

The research has no financial or commercial purpose that must be interpreted as a potential conflict of interest in the future.

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AUTHOR CONTRIBUTIONS

Conceptualization, EH-V, RDV-C, and FB-M; Project administration, fieldwork and data registration, RDV-C, and FBM; data organization and statistical analyses, EH-V, RDV-C, FB-M, and AG-T; writing and reviewing of the original draft, EH-V, RDV-C, and AG-T; review, editing of the last manuscript, EH-V, FB-M, FGV-D, LG-A, AG-T, and RDV-C. All authors have read and agree to approve the final version of the manuscript.

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REFERENCES

Aitchison, J. 1986. The Statistical Analysis of Compositional Data. Chapman and Hall, New York.
Barker, A.V.; Pilbeam, D. J. (Eds.). 2015. Handbook of Plant Nutrition. Second Edition. CRC Press. Boca Raton, FL, USA.

Bates, T.E. 1971. Factors affecting critical nutrient concentrations in plants and their evaluation: A review. Soil Science, 112, 116-130.

Barros de Morais, T.C.; de Melo Prado, R.; Franklin Traspadini, E. I.; Salvador Wadt, P.G. S.; de Paula, R.C.; Rocha, Souza Rocha, M. 2019. Efficiency of the CL, DRIS and CND methods in assessing the nutritional status of *Eucalyptus* spp. rooted cuttings. Forests, 10(9), 786. https://doi.org/10.3390/f10090786

Barłóg, P. 2016. Diagnosis of sugar beet (*Beta vulgaris* L.) nutrient imbalance by DRIS and CND-clr methods at two stages during early growth. Journal of Plant Nutrition, 39(1), 1-16. https://doi.org/10.1080/01904167.2014.964366

Bendaly Labaied, M.; Serra, A. P.; Ben Mimoun, M. 2018. Establishment of nutrients optimal range for nutritional diagnosis of mandarins based on DRIS and CND methods. Communications in Soil Science and Plant Analysis, 49(20), 2557-2570. https://doi.org/10.1080/00103624.2018.1526944

Bendaly Labaied, M.; Khiari, L.; Gallichand, J.; Kebede, F.; Kadri, N.; Ben Ammar, N., Ben Hmida, F.; Ben Mimoun, M. (2020). Nutrient diagnosis norms for date palm (*Phoenix dactylifera* L.) in Tunisian Oases. Agronomy, 10(6), 886. https://doi.org/10.3390/agronomy10060886

Bhaduri, D.; Pal, S. 2013. Diagnosis and Recommendation Integrated System (DRIS): Concepts and applications on nutritional. Journal of Soil and Water Conservation, 12(1), 70-79.

Blanco-Macías, F.; Lara-Herrera, A.; Valdez-Cepeda, R.D.; Cortés-Bañuelos, J.O.; Luna-Flores, M.; Salas-Luévano, M.A. 2006. Interacciones nutrimentales y normas de la técnica de nutrimento compuesto en nopal (*Opuntia ficus-indica* L. Miller). Revista Chapingo Serie Horticultura, 12(2), 165-175.

Blanco-Macías, F.; Magallanes-Quintanar, R.; Valdez-Cepeda, R.D.; Vázquez-Alvarado, R.; Olivares-Sáenz, E.; Gutiérrez-Ornelas, E.; Vidales-Contreras, J.A. 2009. Comparison between CND norms and Boundary-Line Approach nutrient standards: *Opuntia ficus-indica* L. case. Revista Chapingo Serie Horticultura, 15(2), 217-223.

Blanco-Macías, F.; Magallanes-Quintanar, R.; Valdez-Cepeda, R.D.; Vázquez-Alvarado, E.; Olivares-Sáenz, E.; Gutiérrez-Ornelas, E.; Vidales-Contreras, J.A.; Murillo-Amador, B. 2010. Nutritional reference values for *Opuntia ficus-indica* determined by means of the Boundary-Line Approach. Journal of Plant Nutrition and Soil Science, 173(6), 927-934. https://doi.org/10.1002/jpln.200900147

Costa da Silva, G.G.; Lima Neves, J.C.; Alvarez V, V.H.; Palha Leite, F. 2004. Nutritional diagnosis for eucalypt by DRIS, M-DRIS, and CND. Scientia Agricola, 61(5), 507-515. https://doi.org/10.1590/S0103-90162004000500008

García-Hernández, J.L., Valdez-Cepeda, R.D., Murillo-Amador, B., Nieto-Garibay, A., Beltrán-Morales, L.F., Magallanes-Quintanar, R., Troyo-Diéguez, E. 2004. Compositional Nutrient Diagnosis and main nutrient interactions in yellow pepper grown on desert calcareous soils. J. Plant Nutr. Soil Sci. 167, 509-515. https://doi.org/10.1002/jpln.200320370

García-Hernández, J.L.; Valdez-Cepeda, R. D.; Ávila-Serrano, N. Y. Murillo-Amador, B.; Nieto-Garibay, A.; Magallanes-Quintanar, R.; Larrinaga-Mayoral, J.; Troyo-

Diéguez, E. 2005. Preliminary Compositional Nutrient Diagnosis norms for cowpea (*Vigna unguiculata* (L.) Walp.) grown on desert calcareous soil. Plant and Soil, 271, 297-307.

García-Hernández, J.L.; Valdez-Cepeda, R.D.; Murillo-Amador, B.; Beltrán-Morales, F.A.; Ruiz-Espinoza, F.H.; Orona-Castillo, I.; Flores-Hernández, A.; Troyo-Diéguez, E. 2006. Preliminary Compositional Nutrient Diagnosis norms in *Aloe vera* L. grown on calcareous soil in an arid environment. Environmental and Experimental Botany, 58(1-3), 244-252. https://doi.org/10.1016/j.envexpbot.2005.09.001

García-Hernández, J.L.; Valdez-Cepeda, R.D.; Servín-Villegas, R.; Troyo-Diéguez, E.; Murillo-Amador, B.; Rueda-Puente, E. O.; Rodríguez-Ortiz, J. C.; Magallanes-Quintanar, R. 2007. Nutrient interactions and Compositional Nutrient Diagnosis in a semi-domesticated variety of *Capsicum frutescens*. Revista Chapingo Serie Horticultura, 13(2), 133-140.

García-Hernández, J.L.; Orona-Castillo, I.; González-Cervantes, G.; Valdez-Cepeda, R. D.; Murillo-Amador, B.; Troyo-Diéguez, E.; Fortis-Hernández, M.; Segura-Castruita, M.A. 2009. Interacciones nutrimentales y normas de Diagnóstico de Nutrimento Compuesto en nogal pecanero (*Carya illinoensis*). Revista Chapingo Serie Horticultura, 15(2), 141-147.

Habib, R. 2000. Modeling fruit acidity in peach trees effects of nitrogen and potassium nutrition. Acta Horticulturae, 512(2), 141-48.

Hernández-Vidal, E.; Blanco-Macías, F.; A. González-Torres; Véliz-Deras, F.G.; Gaytán-Alemán, L.; Valdez-Cepeda, R.D. 2021. Boundary-Line Approach nutrient standards for *Opuntia ficus-indica* (L.) Miller variety 'Rojo Pelón'. Journal of Soil Science and Plant Nutrition, 21, 467-475. https://doi.org/10.1007/s42729-020-00374-z

Kumar, P.S.S.; Geetha, S.A.; Savithri, P.; Mahendran, P.P.; Ragunath, K.P. 2003. Evaluation of DRIS and CND indexes for effective nutrient management in Muscat grapevines (*Vitis vinifera*). Journal of Applied Horticulture, 5(2), 76-80. http://www.horticultureresearch.net/jah/2003_5_2_76_80.PDF

Khiari, L.; Parent, L.-E.; Tremblay, N. 2001a. Critical compositional nutrient indexes for sweet corn at early growth stage. Agronomy Journal, 93(4), 809-814. https://doi.org/10.2134/agronj2001.934809x

Khiari, L.; Parent, L.-E.; Tremblay, N. 2001b. Selecting the high-yielding subpopulation for diagnosing nutrient imbalance in crops. Agronomy Journal, 9(4), 802-808. https://doi.org/10.2134/agronj2001.934802x

López-García, R.; Mata-González, R.; Blanco-Macías, F.; Méndez-Gallegos, S. de J.; Valdez-Cepeda, R.D. 2016. Fruit attributes dependence on fruiting cladode dry or fresh matter in *Opuntia ficus-indica* (L.) Miller variety 'Rojo Pelón'. Scientia Horticulturae, 202, 57-62. https://doi.org/10.1016/j.scienta.2016.02.028

Lüttge, U. 2004. Ecophysiology of crassulacean acid metabolism (CAM). Annals of Botany, 93(6), 629-652. https://doi.org/10.1093/aob/mch087

Magallanes-Quintanar, R.; Valdez-Cepeda, R.D.; Blanco-Macías, F.; Márquez-Madrid, M.; Ruiz-Garduño, R.R.; Pérez-Veyna, O.; García-Hernández, J.L.; Murillo-Amador,

B.; López-Martínez, J.D.; Martínez-Rubín de Celis, E. 2004. Compositional Nutrient Diagnosis in nopal (*Opuntia ficus-indica*). Journal of The Profesional Association for Cactus Development, 6, 78-89. http://jpacd.org/jpacd/article/view/298/257

Magallanes-Quintanar, R.; Valdez-Cepeda, R.D.; Olivares-Sáenz, E.; Pérez-Veyna, O.; García-Hernández, J.L.; López-Martínez, J.D. 2006. Compositional Nutrient Diagnosis in maize grown in a calcareous soil. Journal of Plant Nutrition, 29(11), 2019-2033. https://doi.org/10.1080/01904160600928235

Mostashari, M.; Khosravinejad, A.; Golmohammadi, M. 2018. Comparative Study of DOP and CND methods for leaf nutritional diagnosis of *Vitis vinifera* in Iran. Communications in Soil Science and Plant Analysis, 49(5), 576-584. https://doi.org/10.1080/00103624.2018.1432633

Parent, L.-E.; Dafir, M. 1992. A theoretical concept of Compositional Nutrient Diagnosis. Journal of The American Society for Horticultural Science, 117(2), 239-242. https://doi.org/10.21273/JASHS.117.2.239

Parent, L.-E.; Cambouris, A.N.; Muhawenimana, A. 1994. Multivariate diagnosis of nutrient imbalance in potato crops. Soil Science Society of American Journal, 58(5),1432-1438. https://doi.org/10.2136/sssaj1994.03615995005800050022x

Raven, J.A.; Spicer, R.A. 1996. The evolution of crassulacean acid metabolism. pp. 360-385. In: Winter, K.; Smith, J.A.C. (Eds.), Crassulacean Acid Metabolism. Biochemistry, Ecophysiology and Evolution. Ecological Studies (Analysis and Synthesis), vol 114. Springer, Berlin, Heidelberg. https://doi.org/10.1007/978-3-642-79060-7_25

René, W.; Côté, B.; Camiré, C.; Burgess, M.; Fyles, J.W. 2013. Development and application of CVA, DRIS, and CND norms for three hybrids of *Populus maximowiczii* planted in Southern Quebec. Journal of Plant Nutrition, 36(1), 118-142. https://doi.org/10.1080/01904167.2012.736578

Ross, S.M. 1987. Introduction to Probability and Statistics for Engineers and Scientists. John Wiley & Sons. New York, USA.

Valdez-Cepeda, R. D.; Magallanes-Quintanar, R.; Blanco-Macías, F.; Hernández-Caraballo, E.A.; García-Hernández, J.L. (2013): Comparison among Boltzmann and cubic polynomial models for estimation of Compositional Nutrient Diagnosis standards: *Opuntia ficus-indica* L. case. Journal of Plant Nutrition 36(6), 895-910. https://doi.org/10.1080/01904167.2013.770020

Walworth, J.L.; Sumner, M.E. 1987. The Diagnosis and Recommendation Integrated System (DRIS). pp. 149-188. In: Stewart B.A. (eds) Advances in Soil Science. Advances in Soil Science, vol 6. Springer, New York, NY. https://doi.org/10.1007/978-1-4612-4682-4_4

Wairegi, L.; van Asten, P. 2011. Norms for multivariate diagnosis of nutrient imbalance in the East African highland bananas (*Musa* spp. AAA). Journal of Plant Nutrition, 34(10), 1453-1472. https://doi.org/10.1080/01904167.2011.585203

Wairegi, L.W.I.; van Asten, P.J. 2012. Norms for multivariate diagnosis of nutrient imbalance in arabica and robusta coffee in the East African Highlands. Experimental Agriculture, 48(3), 448-460. http://africasoilhealth.cabi.org/wpcms/wp-

content/uploads/2014/06/Norms_for_multivariete_diagnosis_of_nutrient_imbalance_in_Arabica_and_Robusta_coffee.pdf