

# COMPARISON BETWEEN CND NORMS AND BOUNDARY-LINE APPROACH NUTRIENT STANDARDS: *Opuntia ficus-indica* L. CASE

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## ABSTRACT

It is a need to know optimum concentrations and/or optimum ranges of nutrients useful for correct diagnosis and improvements of nutrient status of cultivated plants. Recognizing the need to develop correct nutritional standards, it is convenient to improve or to ensure the reliability of those already determined. Having in mind that aim, boundary-line approach standards were developed taking into account data acquired from a field experiment to study the effects of three fertilization treatments on yield of *Opuntia ficus-indica* L. The developed boundary-line approach standards were compared with already developed compositional nutrient diagnosis norms for N, P, K, Ca, and Mg concentrations in one-year old cladodes, and cladodes fresh matter as yield in order to define correct nutrient standards. The developed boundary-line approach optimum nutrient concentrations (associated to estimated maximum yield) for *Opuntia ficus-indica* L. are: N = 1.42 %, P = 0.38 %, K = 4.55 %, Ca = 2.83 % and Mg = 1.41 %. Results suggest that boundary-line approach estimated standards could be trustful and that lower and upper concentrations of the boundary-line approach sufficiency nutrient ranges could be also reliable as well as those for compositional nutrient diagnosis norms.

**ADDITIONAL KEY WORDS:** plant nutrition, CND norms, BLA standards.

## COMPARACIÓN ENTRE NORMAS DNC Y ESTÁNDARES NUTRIMENTALES DE LA TÉCNICA DE CURVA LÍMITE: CASO *Opuntia ficus-indica* L.

## RESUMEN

Es necesario conocer las concentraciones óptimas e intervalos óptimos de nutrientes útiles para el diagnóstico correcto y el mejoramiento de la condición nutrimental de las plantas cultivadas. Reconocer la necesidad de identificar normas nutrimentales correctas, es conveniente para mejorar o garantizar la confiabilidad de las ya definidas. Al tomar en cuenta ese objetivo, la técnica de línea límite se usó para identificar concentraciones óptimas y rangos de suficiencia al 95 % del rendimiento máximo al tomar en cuenta los datos obtenidos de un experimento de campo desarrollado con el fin de estudiar los efectos de tres tratamientos de fertilización sobre el rendimiento de *Opuntia ficus-indica*. Las normas de la técnica de línea límite, producto del presente trabajo, se

compararon con las normas de diagnóstico de nutrimento compuesto ya desarrolladas para los casos de concentraciones de N, P, K, Ca y Mg en cladodios de un año de edad, en su relación con materia fresca de cladodios como rendimiento con el fin de identificar normas nutrimentales correctas. Las concentraciones óptimas identificadas con la técnica de línea límite para *Opuntia ficus-indica* L. son las siguientes: N = 1.42 %, P = 0.38 %, K = 4.55 %, Ca = 2.83 % y Mg = 1.41 %. Los resultados sugieren que las normas estimadas mediante la técnica de línea límite son confiables; además, las concentraciones inferior y superior de los intervalos de suficiencia de los cinco nutrientes son tan confiables como las asociadas a las normas de diagnóstico de nutrimento compuesto.

**PALABRAS CLAVE ADICIONALES:** nutrición vegetal, normas de nutrimento compuesto, normas de la técnica de línea límite.

## INTRODUCTION

In many respects, improvements of soil fertility and efficient use of mineral nutrients is of high ecological and economic importance. Use of basic and practical research information provided through plant nutrition studies will play a decisive role in establishing efficient and ecologically based nutrient management systems. Special attention should be paid to plant nutrients to ensure further increases in yield and sustained soil fertility. Development and application of precise cultural practices that ensure an adequate and balanced supply of nutrients to crop plants is a high priority research area of plant nutrition in order to increase productivity and maintain the fertility of cultivated soils.

Among these precise practices are the correct diagnosis and improvements of the nutrient status of plants. They provide further valuable impacts on the health and quality of plants, animals and humans. For instance, there are several excellent examples showing that plants with high-balanced levels of nutrients are better adapted to biotic and abiotic stress factors, such as fungal diseases, metal toxicities, salinity and photo-oxidative stress caused by low temperature, drought stress and high irradiation (Loneragan, 1997; Cakmak, 2000). In addition, it is expected a good relationship between the plant nutrient status and crop performance (Dow and Roberts, 1982) when involved nutrient is a limiting factor. By these reasons, it is a need to know optimum concentrations and/or optimum ranges of nutrients useful for correct diagnosis and improvements of nutrient status of cultivated plants.

In diagnosis of nutrient status, the analysis of plant tissues has been used because plant composition is an indicator of plant response more sensitive than yield, but at the same time is much more difficult to interpret (Melsted *et al.*, 1969). The diagnosis of plant nutritional status was first based on the interpretation of single-nutrient concentrations (Bates, 1971) and later on bi-variate ratios. Used approaches are critical values for the single-nutrient for the first case; and simple ratios, and diagnosis and recommendation integrated system (DRIS), among others, for the second case. However, recently Parent and Dafir (1992) introduced compositional nutrient diagnosis (CND) approach to consider the effects of the multivariate nature of nutrient interactions. Such an approach, CND ensures that the variation in one element in plant tissue inevitably changes the proportion of the other elements. This method allows the computation of multivariate nutrient ratios that

are more representative of the compositional nature of plant tissue (Aitchison, 1986). CND has been proven with yearly crops (Parent y Dafir, 1992; Parent *et al.*, 1994; Khiari *et al.*, 2001ab; Magallanes *et al.*, 2006), and perennial species (Magallanes *et al.*, 2004; Vizcaíno-Soto y Côté, 2004; Blanco-Macías *et al.*, 2006; Quesnel *et al.*, 2006).

Recognizing the need to develop correct nutrient standards, it is convenient to improve or to ensure the reliability of those already determined. There are known several ways to do it. One of them is to modify the standards by increasing the data base. Also, new optimum values can be determined through other techniques such as regression analysis of relationships between site index and foliar-nutrient concentrations from several stands at local or regional level (Quesnel *et al.*, 2006). Another way is to contrast nutritional standards derived from different techniques. By using this strategy, Vizcayno-Soto and Côté (2004) found that boundary-line approach (BLA) yielded nutritional standards for sugar maple (*Acer saccharum* Marsh.) comparable to those already computed by the CND technique.

Blanco-Macías *et al.* (2006) developed CND nutrient standards for *Opuntia ficus-indica* L. (Table 1), an important crop in Mexico due to its tender pads are widely used for human consumption as vegetables, whereas mature cladodes are used for animal feed; in Mexico, 11,344 ha are used for tender pads production and 4,672 ha for forage production (SAGARPA-SIAP, 2006). However, there remains the idea of whether these standards are correct. Then, to contrast such nutrient norms with others estimated through at least a different approach deserve be performed. Therefore, the main aims of this research were i) to determine nutrient standards using BLA, and ii) to contrast both CND and BLA nutrient norms.

## MATERIALS AND METHODS

### Data

This study is based on data acquired from a field experiment to test three fertilization treatments and three varieties of *Opuntia ficus-indica* ('Jalpa', 'Villanueva' and 'Copena V1'). Within the experimental plot, a plant density of 10,000 plants per hectare was used. We are considering data from 36 plants (12 of each variety). Data corresponds to a database (n = 252) of the concentration of N, P, K, Ca and Mg in one-year old cladodes of *Opuntia ficus-indica*,

and cladodes fresh matter as yield. All data are associated to one-year cladodes harvested from plants having the same structure through formation-pruning practice. The cut cladodes were growing on nine cladodes at the second level from the mother cladode, from February to March of each year: 2001, 2002, 2003, and 2004. Nutrient concentrations were estimated through conventional approaches after acid digestion of the dry tissue samples: N by vapor efflux, P by reduction with molybdo-vanadate, and K, Ca and Mg by spectrophotometric techniques. Interested readers are encouraged to review Magallanes Quintanar *et al.* (2004) and Blanco-Macías *et al.* (2006) papers.

### Compositional nutrient diagnosis norms

Compositional nutrient diagnosis norms, developed by Blanco-Macías *et al.* (2006), were compared with BLA standards developed in this research work. These CND norms were calculated using CND approach, as described in Parent and Dafir (1992), Khiari *et al.* (2001ab), García-Hernández *et al.* (2004), Magallanes-Quintanar *et al.* (2004), García-Hernández *et al.* (2005), Magallanes-Quintanar *et al.* (2006) and Blanco-Macías *et al.* (2006), and using the database ( $n = 252$ ) described *ut supra*. The consigned CND norms as means and standard deviations of the row-centered log ratios  $V_N$ ,  $V_P$ ,  $V_K$ ,  $V_{Ca}$ ,  $V_{Mg}$ , and  $V_{R_d}^*$ , and the corresponding nutrient optimum concentrations and their standard deviations for cladodes fresh matter production with a yield cutoff value of reference of  $46.7 \text{ kg} \cdot \text{plant}^{-1}$  are presented in Table 1.

**TABLE 1.** *Opuntia ficus-indica* L. compositional nutrient diagnosis (CND) norms (row-centered log ratios, RCLR, and standard deviations, SD) for  $d = 5$  nutrients in a high-yield subpopulation producing more than  $46.7 \text{ kg} \cdot \text{plant}^{-1}$  of cladodes fresh matter, and their associated nutrient mean concentrations and standard deviations (SD) (Blanco-Macías *et al.*, 2006).

| CND nutrient norms |       |      | Associated mean nutrient concentrations and standard deviations |      |      |
|--------------------|-------|------|---|------|------|
| RCLR               | Mean  | SD   | Nutrient  | Mean | SD   |
| $V_N^*$            | -0.98 | 0.35 | N (%)   | 1.29 | 0.47 |
| $V_P^*$            | -2.23 | 0.21 | P (%)   | 0.36 | 0.08 |
| $V_K^*$            | 0.25  | 0.19 | K (%)   | 4.24 | 0.88 |
| $V_{Ca}^*$         | 0.37  | 0.35 | Ca (%)  | 4.96 | 1.73 |
| $V_{Mg}^*$         | -0.71 | 0.17 | Mg (%)  | 1.61 | 0.27 |
| $V_{R_5}^*$        | 3.29  | 0.09 |   |      |      |
| $\Sigma V_X$       | 0     |      |   |      |      |

### The boundary-line approach standards

The boundary-line approach standards were developed taking into account data ( $n = 252$ ) acquired from the field experiment described above. The boundary-line is formed when all values for two variables are plotted and a line enclosing these points is established (Michael *et al.*, 1985). The line represents the limiting effect of the independent variable on the dependent variable (Webb, 1972; Lark, 1997), and thus is assumed that all values below it result from the influence of another independent variable or a combination of variables that are limiting the dependent variable (Webb, 1972; Hinkley *et al.*, 1978).

There are several approaches to select the points to define the boundary-line. Involving the Blanco-Macías *et al.* (2006) database ( $n = 252$ ) for *Opuntia ficus-indica*, the boundary-line for each nutrient (N, P, K, Ca and Mg) was estimated when used as independent variable vs. yield as response, according to the following procedure:

The first step consisted in plotting data of cladodes nutrient concentration (%) vs. cladodes fresh matter ( $\text{kg} \cdot \text{plant}^{-1}$ ) as yield. Then, each bivariate relation was used in order to analyze the distribution pattern, to determine its suitability and potential use, and to remove obvious outliers. For instance, Figure 1a shows this step results for nitrogen (N) concentration.

The second step was the selection of the points located on the upper limit of the scatter diagram. It was performed by dividing the nutrient concentration range (independent variable) in classes by using the OpenStat software (2008) and selecting only the highest point for each interval. The rationale for taking into account representative points of classes was based on two criteria: (1) to use at least 10 representative observations to develop the model in order to limit the selection of points to the superior boundary of the scatter of points, and (2) to maximize the likelihood of developing statistically significant models by increasing the number of observations ( $>10$ ). The choice of a number of boundary points which are used to estimate a boundary-line in one scatter diagram represents a compromise between the two aims of big group sizes and a high number of boundary points (Schmidt *et al.*, 2000). These conditions are prerequisites for a reliable definition of boundary points and estimation of boundary lines, respectively. As a result we considered at least 10 points enough by taking into account the distribution of points in each scatter diagram and sample size ( $n = 252$ ). For instance, Figure 1b shows the 18 representative observations relating N concentration and *Opuntia ficus-indica* yield.

The third step consisted of fitting a second degree polynomial function. It can be appreciated in Figure 1c taking into account N concentration vs. yield. When the second-degree function is not appreciated by eye and instead appears a triangular shape, a set of two linear functions

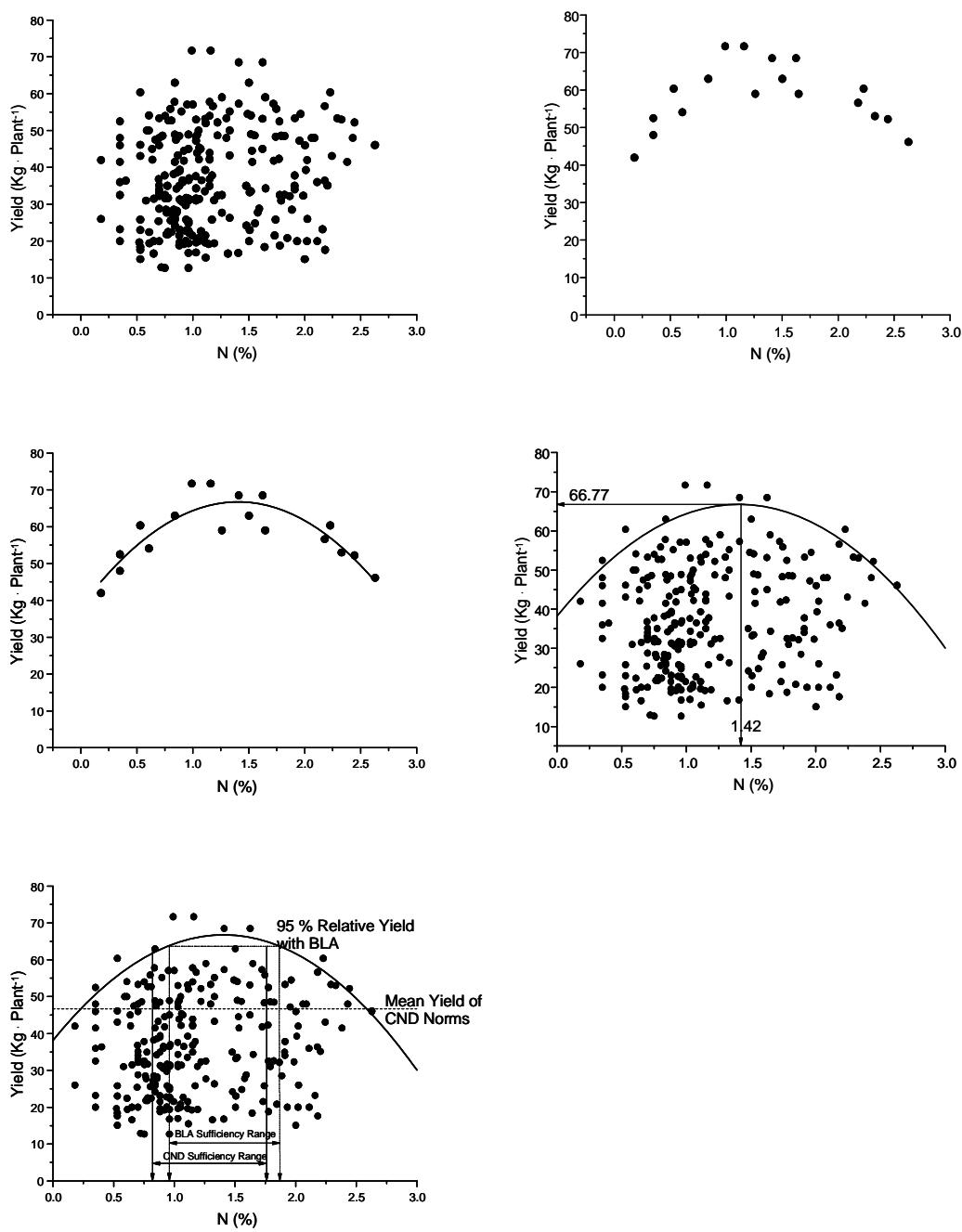


FIGURE 1. The different steps of the boundary-line approach as applied to nitrogen (N) concentration and yield ( $\text{kg} \cdot \text{plant}^{-1}$ ) of *Opuntia ficus-indica* L.

could be used. It is logical to search survey data for the characteristic triangular pattern among maximum values of level of nutrient and yield (Walworth *et al.*, 1986; Evanyo and Sumner, 1987; Righetti *et al.*, 1990). It is possible to estimate the optimum nutrient concentration by calculating the value corresponding to a slope of zero for the second-degree polynomial regression as is clearly appreciated in Figures 1c, d, e for N concentration. In the case that triangular shape appears, the optimum concentration could be estimated by choosing the crossing point between the two straight lines. The optimum range was assessed by solving the second-degree polynomial regression for the

nutrient concentrations corresponding to 95 % of relative yield, as shown for N case in Figure 1e. When a couple of straight lines is used, this nutrient range could be elucidated under the basis of 95 % of relative yield value for both linear functions.

#### Comparison between CND norms and BLA nutrient standards

Comparison of results from both approaches was performed taking into account optimum concentrations and ranges of sufficiency.

## RESULTS AND DISCUSSION

### The boundary-line approach standards

In each of the scatter diagrams most of the data points grouped at the bottom, i.e. at a low yield level (Figures 1, 2, 3, 4, 5). High-yields were only rarely measured across each interval considered, thus selection of representative points was easy. So, we were able to choose  $>10$  points at the upper edge of each data body. However, drawing boundary-lines by hand is an uncertain and not reproducible method. Thus, procedure proposed in this study was developed to overcome this uncertainty.

There, each of all five nutrients vs. yield scatter diagrams showed to have a boundary-line model developed without correction for outliers. The boundary-lines were estimated using representative observations for all defined classes. The boundary-line fitted by a second-degree function was observed for N, P, and K, with 18, 17, and 11 intervals, as appreciated in Figures 1, 2, and 3, respectively. The Boundary-lines for Ca with 14 observations, and Mg with 11 points were described through two straight lines, because they had triangular-shaped scatter distributions (Figures 4 and 5).

Blanco-Macías *et al.* (2006) data base ( $n = 252$ ) allow for the successful application of a boundary-line approach to assess nutrient standards through fitting significant ( $P < 0.01$ ) second-degree polynomial or straight functions for N, P, K, and Mg (Table 2); exception corresponds to Ca ( $P < 0.09$ ). Models for N, K, and Mg had high  $R^2$  values ( $R^2 > 0.6$ ), whereas those for P and Ca were low ( $R^2 < 0.6$ ) as appreciated in Table 2. These results may be due to the wide range of Ca concentration in cladodes of *Opuntia ficus-indica* L. and to the fewer data in the upper region of the scatter diagram. This late aspect also applies for P case. In this context, Vizcayno-Soto and Côte (2004) pointed out improvements in  $R^2$  by increasing intervals, but lower levels of probability. Thus, BLA would appear to be particularly appropriate for N, P, K, and Mg. In addition, it appears scatter point distributions with large spread of nutrient concentrations (for instance Ca in this study) require a large number of points to optimize the yield of the BLA. It deserves be mentioned *Opuntia ficus-indica* is a rare calcitrophic species (Lüttege, 2004) and that experimental soil is rich in available Ca (Blanco-Macías *et al.*, 2006) which explain the large spread of Ca concentrations.

It is interesting to point out all five optimum nutrient concentrations related to maximum yield scores estimated by the BLA (Figures 1-5) are  $>38\%$  higher than target yield ( $46.7 \text{ kg} \cdot \text{plant}^{-1}$ ) used for dividing the whole database ( $n = 252$ ) and to develop the CND norms by Blanco-Macías *et al.* (2006). This result suggests that BLA estimated optimum nutrient concentrations could be trustful. Moreover, yields associated to sufficiency nutrient ranges estimated through

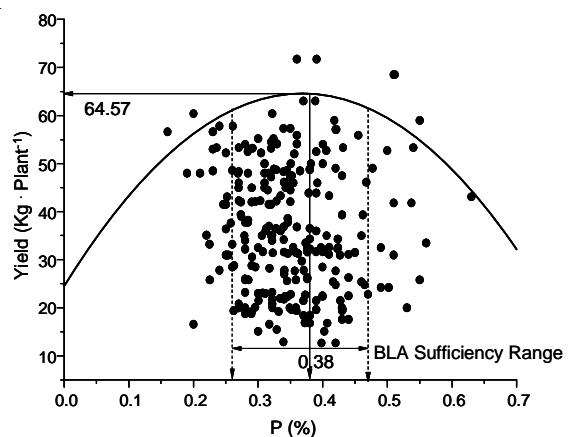


FIGURE 2. The relationship between phosphorus (P) nutrient concentration and *Opuntia ficus-indica* L. yield ( $\text{kg} \cdot \text{plant}^{-1}$ ) showing boundary line described by a second degree function.

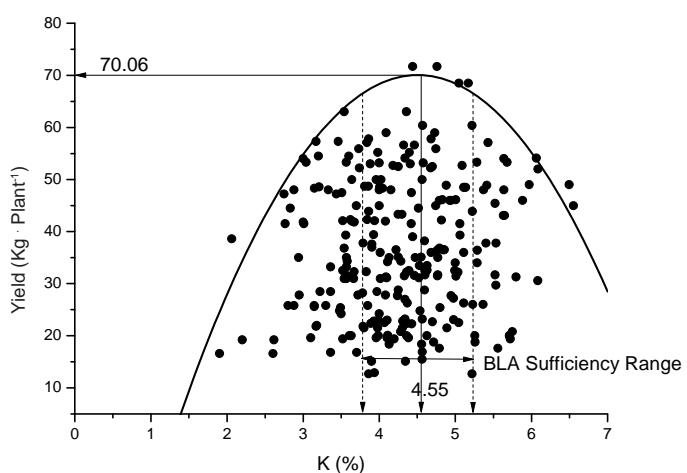


FIGURE 3. The relationship between potassium (K) nutrient concentration and *Opuntia ficus-indica* L. yield ( $\text{kg} \cdot \text{plant}^{-1}$ ) showing boundary line described by a second degree function.

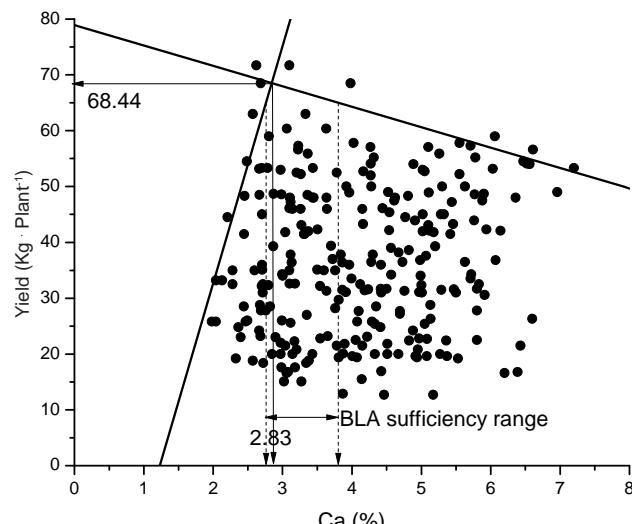


FIGURE 4. The relationship between calcium (Ca) nutrient concentration and *Opuntia ficus-indica* L. yield ( $\text{kg} \cdot \text{plant}^{-1}$ ) showing boundary lines (straight lines).

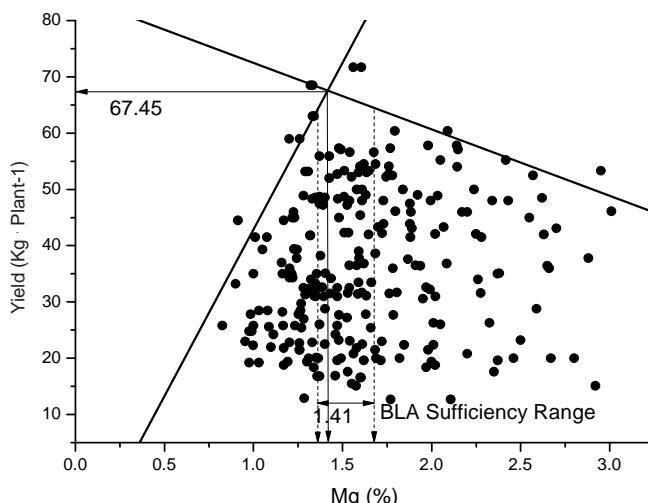


FIGURE 5. The relationship between magnesium (Mg) nutrient concentration and *Opuntia ficus-indica* L. yield ( $\text{kg} \cdot \text{plant}^{-1}$ ) showing boundary lines (straight lines).

TABLE 2. Statistics of boundary lines fitted by second-degree or linear functions used to estimate *Opuntia ficus-indica* L. nutrient standards.

| Nutrient, and function(s)              | $R^2$ | Probability |
|--|-------|-------------|
| N<br>$Y = 38.16 + 40.67x - 14.46x^2$   | 0.74  | 0.00001     |
| P<br>$Y = 24.46 + 217.62x - 295.19x^2$ | 0.45  | 0.00571     |
| K<br>$Y = -66.01 + 60.36x - 6.69x^2$   | 0.90  | 0.00004     |
| Ca<br>$Y = -52.27 + 42.49x$            | 0.73  | 0.0957      |
| <br>$Y = 78.93 - 3.66x$                | 0.58  | 0.00604     |
| Mg<br>$Y = -16.35 + 59.23x$            | 0.86  | 0.00516     |
| <br>$Y = 84.28 - 11.81x$               | 0.69  | 0.05164     |

TABLE 3. *Opuntia ficus-indica* L. compositional nutrient diagnosis (CND) and Boundary-line Approach (BLA) optimum nutrient concentrations and ranges of sufficiency.

| Nutrient | CND                   |             | BLA                   |                               |
|----------|-----------------------|-------------|-----------------------|-------------------------------|
|          | Optimum concentration | Range       | Optimum concentration | Range (95 % of maximum yield) |
| N (%)    | 1.29                  | 0.82 - 1.76 | 1.42                  | 0.92 - 1.89                   |
| P (%)    | 0.36                  | 0.28 - 0.44 | 0.38                  | 0.26 - 0.47                   |
| K (%)    | 4.24                  | 3.36 - 5.12 | 4.55                  | 3.78 - 5.23                   |
| Ca (%)   | 4.29                  | 3.23 - 6.69 | 2.83                  | 2.76 - 3.80                   |
| Mg (%)   | 1.61                  | 1.34 - 1.88 | 1.41                  | 1.36 - 1.71                   |

BLA (Table 3) are 31 % higher than target yield mentioned *ut supra*. Then, these results suggest lower and upper concentrations of the BLA sufficiency nutrient ranges could be also reliable.

### Comparison between CND norms and BLA nutrient standards

Contrasting the optimum CND scores with those estimated in this study using BLA allow to identify optimum CND scores for Ca (4.29 %) and Mg (1.81 %) are higher than optimum BLA concentrations (Ca = 2.83 %, Mg = 1.41 %) (Table 3). Curiously, both cases show to have triangular-shaped positively skewed distributions. Thus, such results could be attributed to skew, and suggest sample size must be increased to have a normal distribution of data or study them using a data transformation strategy. On the other hand, optimum CND concentrations for N (1.29 %), P (0.36 %), and K (4.24 %) were slightly lower than the optimum BLA scores (N = 1.42, P = 0.38, and K = 4.55 %). This latter difference indicates that *Opuntia ficus-indica* L. plants yield well at slightly higher concentrations than optimum CND scores reported by Blanco-Macías *et al.* (2006).

Concerning optimum ranges, there are several interesting differences. Lower and upper BLA scores are slightly higher than CND values. This implies CND under-estimated or BLA over-estimated N optimum range, but the real difference is almost nil. Both ranges for P are practically equal. The K case is similar to that of N. CND optimum range for Ca (3.23 - 6.69 %) is circumscribed to BLA optimum range (2.76 - 3.8 %), which indicates the former could be more sensitive when Ca diagnosis is performed. The lower BLA score for Mg is slightly higher than that associated to CND technique; however, the higher BLA Mg value is slightly lower than that for CND approach. There is also remarkable BLA ranges for N and P (0.9 and 0.21 %, respectively) are slightly larger than those estimated from CND method (0.94 and 0.16 % for N and P, respectively), and that BLA sufficiency ranges for K, Ca, and Mg (1.45, 1.04, and 0.35 %, respectively) are lower than optimum ranges (1.76, 3.46, and 0.54 % for K, Ca, and Mg, respectively) as defined by Blanco-Macías *et al.* (2006) using CND procedure.

### CONCLUSIONS

The boundary-line fitted by a second-degree function was observed for N, P and K. The Boundary-lines for Ca, and Mg were described through two straight lines, because they had skewed triangular-shaped scatter distributions.

Optimum boundary-line approach scores were as follows: N = 1.42 %, P = 0.38 %, K = 4.55 %, Ca = 2.83 % and Mg = 1.41 %. These for N, P, and K were slightly higher than those corresponding to CND norms. However, for Ca and Mg, BLA optimum concentrations were strongly lower than those associated to CND norms, especially for Ca.

When ranges of sufficiency for both approaches were compared, there appear no important differences although procedures for their estimation are really different.

Boundary-line approach estimated standards could be trustful, and lower and upper concentrations of the BLA sufficiency nutrient ranges could be also reliable as well as CND norms.

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