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Article

New Nitrogen Use Efficiency Indices for Biomass Formation and Productivity in Beans Under Foliar Fertilization with Molybdenum Nanofertilizer

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Abstract: Most crops are fertilized with high amounts of nitrogen, and have an alarmingly low utilization efficiency. For this reason, the coordination between the fertilizer contribution and the nitrogen requirements of the crop is very important. Therefore, the objective of this study was to determine the nitrogen use efficiency, and create new NUE indices that can determine and calculate the final destination of the assimilated nitrogen, for the formation and development of green bean plant organs, fertilized with molybdenum nanofertilizer applied foliarly and combined with soil fertilization of ammonium nitrate. The plants were grown in a greenhouse covered with anti-aphid mesh and irrigated with nutrient solution. Three sources of foliar Molybdenum (Nano fertilizer, Molybdenum Chelate and Sodium Molybdate) were applied in four doses of 0, 5, 10 and 20 ppm Mo, complemented with edaphic fertilization of NH_4NO_3 (0, 3, 6 and 12 mM of N). As results, the NUE indices showed that with the application of the nanofertilizer, the total biomass production increased 41.65% more than with the application of the chelate, and 36.84% more than with the application of molybdate. Finally, it is concluded that the use of NUE indices is an important approach that evaluates the fate of nitrogen and accurately estimates plant yield.

Keywords: NUE indices; nanofertilizer; molybdenum; nitrogen; *Phaseolus vulgaris* L.

1. Introduction

Nowadays, the role of nitrogen (N) for the growth, development and productivity of crops is essential. This essential macroelement has a direct effect on the production of biomass and dry weight of plants, by influencing, in addition to many other physiological processes, the efficiency of the photosynthetic system of the leaves. In this way, a strong photosynthetic system allows optimal development; In contrast, a limited or stunted photosynthetic system results in low photosynthetic efficiency, causing rickets and plant death. [1,2].

For this reason, it is essential to improve nitrogen use efficiency (NUE) within crop agronomic management programs. The increase in NUE through agronomic management practices, and with the use of high-performance technologies, manages to reduce intensive applications of nitrogen, increase its use and reduce environmental pollution caused by nitrogen fertilizers. [3]. In parallel, it must be considered that N is a key component of living cells, and that nitrogen fertilizer is the second largest requirement after water in crop production [4]. For this reason, the relationship between the applied N and its absorption and fixation efficiency must be high, and not inefficient or on a scale higher than the plant's requirements [5].

It is then that the elements that are key for nitrogen fixation must also be taken into account. In this sense, molybdenum (Mo) stands out for being an essential micro-element that plays a fundamental role in N metabolism, and that regulates and optimizes the activities and expressions of the enzymes responsible for N assimilation. In addition, it participates in different biosyntheses responsible for the normal functioning of plant growth and development processes [6].

To enhance the effect of Mo, its application must be foliar and can be effectively combined with the use of nanotechnology, in this way a nanofertilizer is created capable of penetrating plant tissues and intelligently releasing the active ingredient. (Mo), so that it is fully available to the plant [7]. The harmony of these tools aligns agricultural systems with global needs to protect natural resources [3]. Therefore, the objective of this study was to determine the efficiency of nitrogen use, and to create new NUE indices that can determine and calculate the final destination of the assimilated nitrogen, for the formation and development of the organs of fertilized green bean plants. with molybdenum nanofertilizer applied foliarly and combined with soil fertilization of ammonium nitrate.

2. Materials and Methods

2.1. Location and Growth Conditions

The crop was growth in a greenhouse covered with anti-aphid mesh located in Lázaro Cárdenas, Meoqui, Chihuahua, Mexico (Latitude: N 28°23′ 9.80232′′, Longitude: W 105°36′ 58.09392′′), starting on September 2 of 2020 to harvest on November 3, 2020, with an average temperature of 28.6 °C. Bean seeds cv. Short cycle strike (60 days until physiological maturity) in polystyrene trays with 200 cavities; 12 days after germination, the plants were transplanted into polyethylene bags (two plants per bag) of 400 caliber and 10 kg capacity, which contained vermicu-lite and perlite as substrate in a 2:1 ratio. A complete nutrient solution pH 6.0 was applied for 20 days according to [8] as proposed by [9] from the germination of the plants, which had the following composition: NH_4NO_3 6,0 mM, K_2HPO_4 1,6 mM, K_2SO_4 0,3 mM, $CaCl_2{\cdot}2H_2O$ 4,0 mM, $MgSO_4$ 1,4 mM, Fe-EDDHA 5,0 µM, $MnSO_4{\cdot}H_2O$ 2,0 µM, 1,0 µM de $ZnSO_4{\bullet}7H_2O$, $CuSO_4{\bullet}5H_2O$ 0.25 µM, Na_2MoO_4 0.3 µM y H_3BO_3 0.5 µM (all reagents J.T. Baker, State of Mexico, Mexico); With the aim of ensuring that the plants were well nourished in their early stages of development, 500 mL of the nutrient solution per bag was applied every third day. After 20 days, differentiated nitrogen treatments were applied to the nutrient solution every three days and until the end of the crop. Molybdenum treatments were applied foliarly every seven days from the appearance of true leaves. The entire experiment was carried out in a single time (from September 2 to November 3, 2020), the application of all treatments (Sub splits, sub-sub splits and sub-sub-sub splits) was carried out in a simultaneous.

2.2. Experimental Design and Treatments

An experimental design was established with a divided plot arrangement in a completely randomized design with four repetitions. The organs of the plant were considered as Split: leaves, stems, fruits and roots, the sources of Molybdenum represented the Sub splits (BROADACRE® Zn Mo Nanofertilizer, Agrichem de México, Mazatlán, Sinaloa, México), GRO Bo Mo® Chelate (Fertilizados Tepeyac, Delicias, Chihuahua, Mexico) and Sodium Molybdate (J.T. Baker, State of Mexico, Mexico); Nitrogen doses as ammonium nitrate (NH_4NO_3): 0, 3, 6 y 12 mM accounted for the Sub- sub split, and molybdenum doses: 0, 5, 10 y 20 ppm represented the Sub- sub- sub split. With a total of 48 treatments, and 384 experimental units (plants) (two plants per bag represent one repetition, which gives a total of four repetitions) (Figure 1). Five foliar applications of the three different sources of molybdenum were made starting on day 21 after germination, and 16 applications of the nutrient solution differentiated in nitrogen starting on day 22 after germination. The additive linear model was the following:

$$\Upsilon ijklm = \mu + \theta i + \mathcal{E} im + \Omega j + (\theta\Omega)ij + \lambda ijm + \beta k + (\theta\beta)ik + (\Omega\beta)jk + (\theta\Omega\beta)ijk + Zijkm + \Sigma l + (\theta\Sigma)ij + (\Omega\Sigma)jl + (\beta\Sigma)kl + (\theta\Omega\beta\Sigma)ijkl + \Psi ijkl \quad (1)$$

where:

i= Bean plant organs (Split)
j= Molybdenum Source (Sub split)

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k= Nitrogen doses (Sub- sub split)
l= Molybdenum doses (Sub- sub- sub split)
m= Repetition

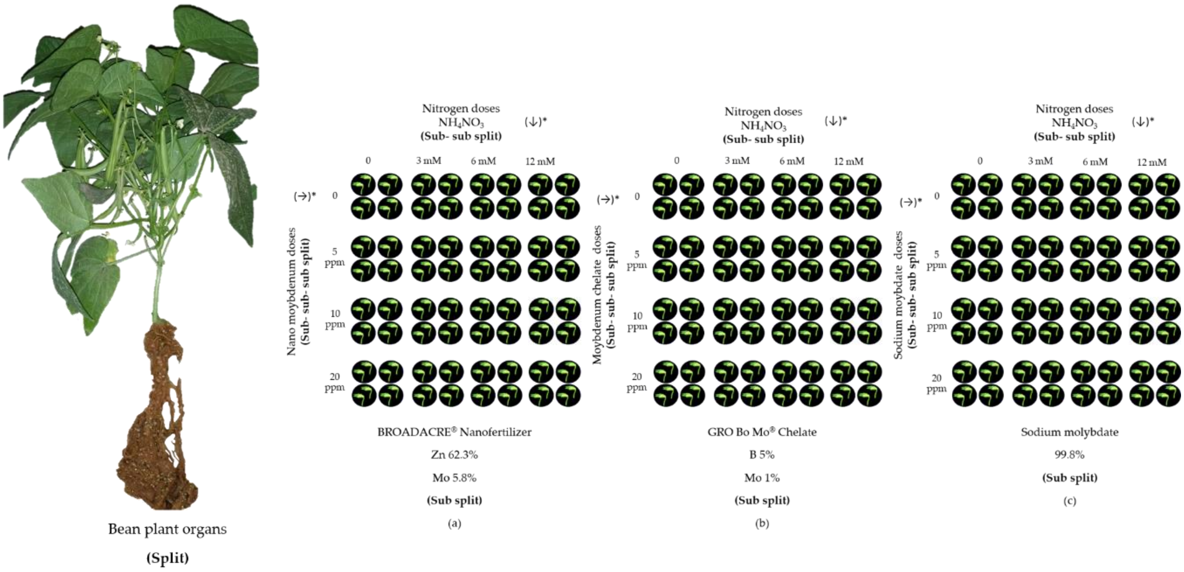

Figure 1. Experimental design. Soil nitrogen fertilization supplemented with foliar molybdenum fertilization in green beans cv. Strike. The figure shows how the experiment was laid out inside the greenhouse. The organs of the plant were considered as Spit: leaves, stems, fruits and roots, (a) Sub split where nanofertilizer was applied, (b) Sub split where chelate was applied, (c) Sub split where sodium molybdate was applied. (↓)* Direction of application in columns of nitrogen doses, (→)* Direction of application in rows of molybdenum doses.

2.3. Plant Analysis

2.3.1. Biomass

After environmental decontamination, the samples were placed in a drying oven at 70° C (Felisa® Oven St. Livonia, Michigan, USA) for 24 h until completely dry. Total biomass production was calculated based on the dry weight of the plant material expressed in grams (g plant^{-1}) [10].

2.3.2. Yield

The yield was obtained based on the fresh weight of the fruits per plant. Green beans were collected from each of the cultivated plants and weighed at the time of sampling (Analytical Balance, Precision Electronic Balance AND Comapany Limited, Milpitas, CA, USA). The total yield was expressed in grams per plant (g plant^{-1}) [10].

2.3.3. Total nitrogen content

Once dry, the samples were ground in a blender (Osterizer® Blender, Milwaukee, Wisconsin, USA) and placed in plastic bags (Nasco Whirl-Pak®, Cin-cinnati, Ohio, USA) for subsequent analysis. The total nitrogen concentration was determined using the Flash 2000 Organic Elemental Analyzer (Thermo Scientific® Corporation, Cambridge, UK), which bases its operation on the method initially described by Jean-Baptiste [11]. A tin capsule was placed on a microbalance (Mettler Toledo®, Columbus, Ohio, USA), 9 mg of vanadium pentoxide (JT Baker, State of Mexico, Mexico), and 3 mg of the finely ground sample were weighed, a Once the weight was taken, the capsule was closed. The samples were then placed in the Flash 2000 autosampler for analysis; Two certified standards of Me-thionine and Sulfanilamide (Thermo Scientific® Corporation, Cambridge, UK) were also analyzed in

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order to guarantee the accuracy of the results. The concentration of total organic N was expressed as a percentage (%).

2.4. Efficiency Indices of Absorption, Distribution and Utilization of Nitrogen for the Formation of Plant Biomass

The indices are the following:

- Total weight (*TW*)

Refers to the total weight of the plant biomass.

$$\boldsymbol{TW = Wl + Ws + Wf + Wr = g} \quad (1)$$

where *Wl* is the total weight of the leaves (g), *Ws* is the total weight of the stems (g), *Wf* is the total weight of the fruit (g), *Wr* is the total weight of the root (g). The result was expressed in grams (g).

- Percentage by weight (*PxW*)

It refers to what each organ represents in the total weight of the plant, in percentage (%).

$$\boldsymbol{PxW} = \left(\frac{\boldsymbol{Wh}}{\boldsymbol{Wl+Ws+Wf+Wr}}\right)\boldsymbol{100} = \% \quad (2)$$

where *Wh* is the total weight of the leaves (g), *Ws* is the total weight of the stems (g), *Wf* is the total weight of the fruit (g), *Wr* is the total weight of the root (g), between 100. The result was expressed as a percentage (%).

- Milligrams of nitrogen (*mg-N organs*)

It refers to the milligrams of nitrogen necessary to form each organ of the plant.

$$\boldsymbol{mg - N\ organs} = \left(\left(\frac{\boldsymbol{CON}}{\boldsymbol{100}}\right)\boldsymbol{PO}\right)\boldsymbol{1000} = \boldsymbol{mg} \quad (3)$$

where *CON* is the nitrogen concentration of the plant organ (leaf, stem, fruit or root), *PO* is the weight of the plant organ, all multiplied by 1000. The result was expressed in milligrams (mg).

- Quantity of nitrogen in percentage (%) (*CaN*)

It refers to the amount of nitrogen in percentage (%) to form each organ of the plant.

$$\boldsymbol{CaN} = \left(\frac{\boldsymbol{mgNO}}{\boldsymbol{mgNl+mgNs+mgNf+mgNr}}\right)\boldsymbol{1000} = \% \quad (4)$$

where *mgNO* are the milligrams of nitrogen necessary to form the plant organ, *mgNl* are the milligrams of nitrogen necessary to form the leaves, *mgNs* are the milligrams of nitrogen necessary to form the stems, *mgNf* are the milligrams of nitrogen necessary to form the fruits, *mgNr* are the milligrams of nitrogen necessary to form the root, all multiplied by 1000. The result was expressed as a percentage (%).

- Nitrogen fixation efficiency (*NFiE*).

It refers to efficiency as the maximum yield produced per unit of nitrogen used by the plant for the production of biomass.

$$\boldsymbol{NFiE} = \left(\frac{\boldsymbol{Pl}}{\frac{\boldsymbol{NTl}}{\boldsymbol{1000}}}\right) = \% \quad (5)$$

where *Pl* is the dry weight of the leaf, *TNl* is the total accumulation of nitrogen in the leaf, divided by 1000. The result was expressed as a percentage (%).

- Nitrogen conduction (translocation) efficiency (*NCoE*).

Se refiere a la eficiencia de traslocación de nitrógeno para la producción de biomasa.

$$\boldsymbol{NCoE} = \left(\frac{\boldsymbol{Ps}}{\frac{\boldsymbol{NTs}}{\boldsymbol{1000}}}\right) = \% \quad (6)$$

where *Ps* is the dry weight of the stem, *NTs* is the total accumulation of nitrogen in the stems, divided by 1000. The result was expressed as a percentage (%).

- Nitrogen absorption efficiency (*NAbE*)

Refers to the efficiency of nitrogen absorption for biomass production.

$$\boldsymbol{NAbE} = \left(\frac{\boldsymbol{Pr}}{\frac{\boldsymbol{NTr}}{\boldsymbol{1000}}}\right) = \% \quad (7)$$

where *Pr* is the dry weight of the root, *NTr* is the total accumulation of nitrogen in the root, divided by 1000. The result was expressed as a percentage (%).

- Productivity.

It refers to efficiency as the maximum yield produced per unit of nitrogen used by the plant for fruit production.

$$\boldsymbol{Productivity} = \left(\frac{\boldsymbol{Number\ of\ fruits}}{\boldsymbol{TNA}}\right) = \boldsymbol{Fruits} * \boldsymbol{mg} - \boldsymbol{N}\ (8)$$

Where Number of fruits is the number of fruits produced per plant, and *TNA* is the total nitrogen accumulation. The result was expressed in number of fruits per milligrams of nitrogen. (Fruits · mg-N).

2.6. Statistical Analysis

The data obtained were subjected to an analysis of variance based on the proposed additive linear model, the probabilities of impact were Pr > 0.05 not significant, 0.05 ≤ Pr ≤ 0.01 significant, Pr < 0.01 highly significant, the rank test was obtained. multiple. Tukey test (α 0.05) to separate the treatment means within each factor (division, subdivision and sub-subdivision); Subsequently, a response surface analysis of the split x sub split interaction was carried out for the split cell factor with greater statistical relevance [12].

The response surface analysis included the following steps: 1) model adjustment and analysis of variance to estimate the parameters. The estimated surface will typically be curved, a hill whose peak occurs at the single estimated point of maximum response, a valley, or a saddle-shaped surface with no maximum or minimum; It is determined 1) if the types of effects are linear, quadratic or cross products, what part of the residual error is due to the lack of fit and what is the contribution of each factor in the statistical fit; 2) canonical correlation to investigate the shape of the predicted response surface, calculating whether the fixed point is a maximum, a minimum or a saddle point and which factor or factors are the most sensitive predicted responses and 3) ridge analysis for the search for the optimal response. The eigenvalues and eigenvectors of the canonical analysis characterize the shape of the response surface; The eigenvalues indicate the direction of the primary orientation of the surface, and the signs and magnitudes of the associated eigenvectors give the shape of the surface in those directions. Positive eigenvalues indicate upward curvature directions and negative eigenvalues indicate downward curvature directions. The eigenvector for the largest eigenvalue gives the direction of the steep rise from the fixed point, if positive, or the steep fall, if negative. Eigenvectors corresponding to small or zero eigenvalues indicate directions of relative flattening. To determine whether the solution is a maximum or a minimum, we observe the sign of the eigenvalues: if the eigenvalues are all negative, the solution is a maximum; If they are all positive the solution is a minimum, if they have mixed signs the solution is a saddle point and if they contain zeros the solution is a flattened area [13].

Once the statistical analysis was carried out, the SigmaPlot 14.0 program was used to obtain the graphs with the results predicted by the SAS program. The graphs are for those variables that were significant, whether in linear, quadratic regression and interaction of factors.

3. Results

3.1. Effect of Nitrogen Fertilization Supplemented with Molybdenum Foliar Nanofertilizer on NUE Indices for Biomass Formation

3.1.1. NUE Indices in Leaves

In the present study, the results showed that with the application of the molybdenum nanofertilizer (NanoMo), it was possible to increase the efficiency of nitrogen use for the efficient formation of the total biomass of the plant (Table 1). The nitrogen use efficiency indices showed that, with the application of molybdenum nanofertilizer, total biomass production increased 41.65% more than with the application of molybdenum chelate, and 36.84% more than with the application of

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sodium molybdate. This Biomass index allowed us to observe how the nanofertilizer achieved greater nitrogen assimilation, which translated into an increase in the accumulation of dry matter, in this case, greater development and number of leaves. The clear advantage of nanofertilizer over the most commonly used conventional fertilizers is presented as a reliable alternative that increases the efficient use of nitrogen for the benefit of the crop (Figure 2).

Figure 2. Effect of edaphic nitrogen fertilization, complemented with foliar fertilization with molybdenum nanofertilizer on nitrogen use efficiency (NUE).

Table 1. Effect of edaphic nitrogen fertilization supplemented with foliar fertilization of molybdenum nanofertilizer on leaf NUE indices.

	Indices NUE (Leaf)				
	PTl	**PxPl**	**mg-Nl**	**CaNl**	**NFiE**
Mo Source	<0.0001[U]	<0.0001	0.0008	0.0719	0.6624
Nano Mo	**8.93 a**[V]	52.50 a	**103.92 a**	42.58 a	21.54 a
Mo Chelate	5.21 b	**54.05 a**	77.46 b	**48.06 a**	**23.49 a**
Na Molybdate	5.64 b	43.52 b	51.81 c	40.44 a	22.24 a
MSD	1.35[W]	3.91	24.51	8.19	5.95
Nitrogen[X]	<0.0001	0.0649	<0.0001	0.0064	<0.0001
0	4.74 c	46.25 b	51.76 b	40.36 b	**30.90 a**
3	7.22 ab	51.17 ab	53.65 b	39.71 b	27.11 a
6	**8.20 a**	49.32 ab	101.23 a	44.14 ab	17.27 b
12	6.19 bc	**53.35 a**	**104.26 a**	**50.57 a**	14.42 b
MSD	1.62	7.07	15.64	8.54	7.49
Molybdenum[Y]	<0.0001	0.1327	0.9602	0.1924	0.0009

0	5.34 c	50.34 a	75.89 a	44.68 a	17.86 b
5	6.07 bc	**52.35 a**	**79.30 a**	**46.80 a**	22.43 ab
10	**7.76 a**	47.88 a	77.49 a	40.99 a	**26.77 a**
20	7.20 ab	49.56 a	78.30 a	43.30 a	22.64 ab
MSD	1.16	4.94	16.90	7.48	5.51
SoMo*N	0.0817	0.5016	0.0001	0.0926	<0.0001
SoMo*Mo	<0.0001	0.9408	0.0025	0.3620	0.0378
N*Mo	0.0675	0.2929	0.0256	0.0415	0.0327
SoMo*N*Mo	0.2956	0.0519	0.0085	0.0022	<0.0001
μ	6.59	50.02	77.73	43.69	22.42
C.V.	33.08	18.57	40.83	32.17	46.18
R^2	0.7872	0.6217	0.7704	0.5990	0.7705

[U]Probability not significant Pr > 0.05, significant 0.05 ≤Pr≤0.01, highly significant Pr <0.0001; [V]Means with the same letter are statistically equal; [W]Least significant difference; [X]Mm edaphic nitrogen concentration; [Y]Leaf ppm concentration of molybdenum, overall average μ, CV coefficient of variation, R^2 coefficient of determination; Regression Analysis: Linear L, Quadratic C, P N*Mo interaction.

It is important to highlight the benefit of the interaction of edaphic nitrogen with foliar NanoMo (Figure 3a). Although the joint action of nitrogen and molybdenum naturally is essential for the plant, the strategy of applying NanoMo enhanced the assimilation and fixation of nitrogen (doses of 6 mM-N and 10 ppm-Mo), and could be used more efficiently for the formation of a greater number of and larger leaves. With a greater amount of foliage, the leaf area of the plant was considerably increased, this allowed for greater light capture and an efficient photosynthetic system.

In the NUE index that estimates the amount of milligrams of nitrogen necessary for the formation of leaves (mg-N-leaf), the difference in the use of nitrogen for the formation of dry biomass of the NanoMo compared to the chelate and molybdate, ranges from 25.46 % and 50.14% respectively (Table 1). With the use of this index, it could be clearly seen how nitrogen, under the effect of NanoMo, was easily metabolized and transformed to form part of the amino acids and proteins necessary for the optimal development of the plant. In Figure 3b, you can see the positive effect of NanoMo at the dose of 10 ppm, and how it has a quite favorable response when interacting with a high dose of nitrogen (12 mM). This may mean that, as there is a greater amount of molybdenum available in the plant’s metabolism, there is a greater probability of metabolizing a high amount of nitrogen for the formation of foliage, which helps to mitigate to a certain extent the toxic effects of a plant. nitrogen supersaturation.

In relation to the NUE index that indicates the percentage of nitrogen used to form leaves (CaN), the plants treated with the chelate source were those that required the highest percentage of nitrogen for foliage formation (Table 1). Likewise, in Figure 3c, it can be seen how the doses of 3 mM and 6 mM of nitrogen supplemented with NanoMo foliar fertilization were the most efficient in leaf production per unit of nitrogen applied. Under the principle of efficiency, with these low doses of nitrogen and NanoMo, the plants developed and managed to produce a greater number of leaves; unlike the plants where the double dose of nitrogen (12 mM) was applied (Figure 4).

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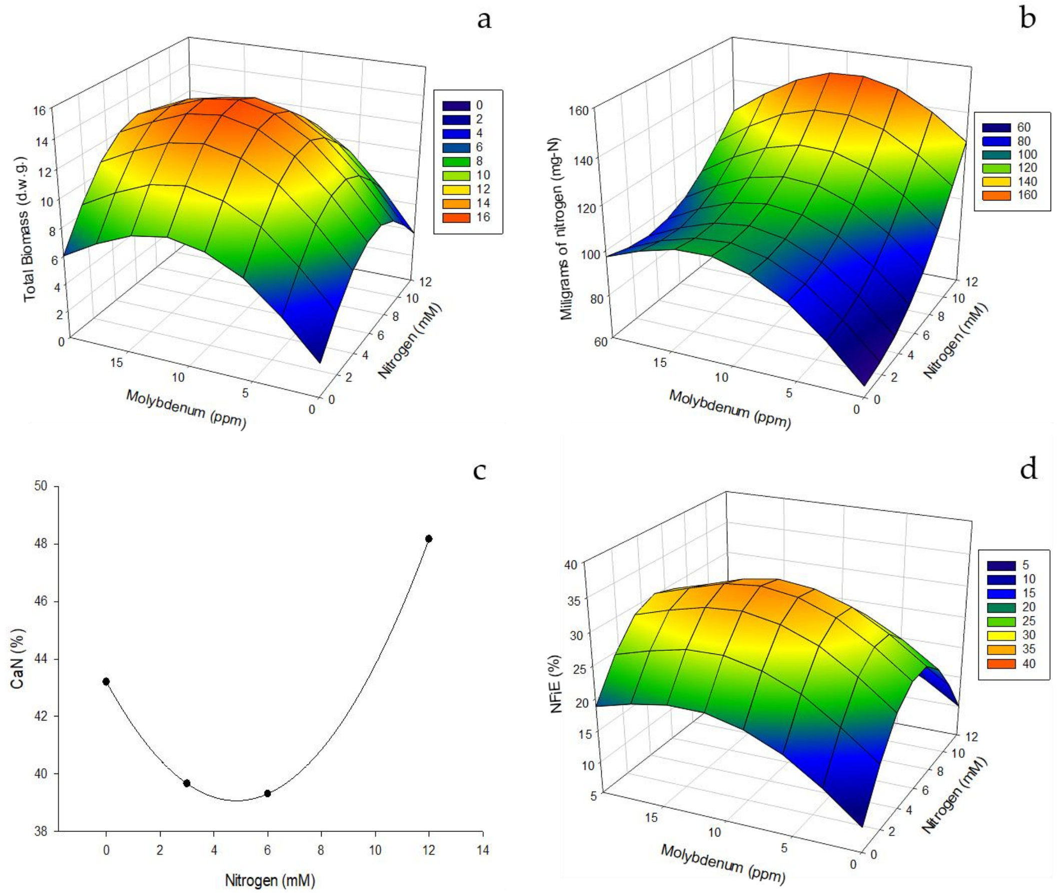

Figure 3. Effect of edaphic nitrogen fertilization, complemented with foliar fertilization with molybdenum nanofertilizer on NUE indices for biomass formation. (a) Total foliage biomass (grams dry weight). (b) Milligrams of nitrogen needed to form the leaves of the plant (mg-N). (c) Quantity of nitrogen to form the leaves (CaNl). (d)Nitrogen fixation efficiency (NFiE).

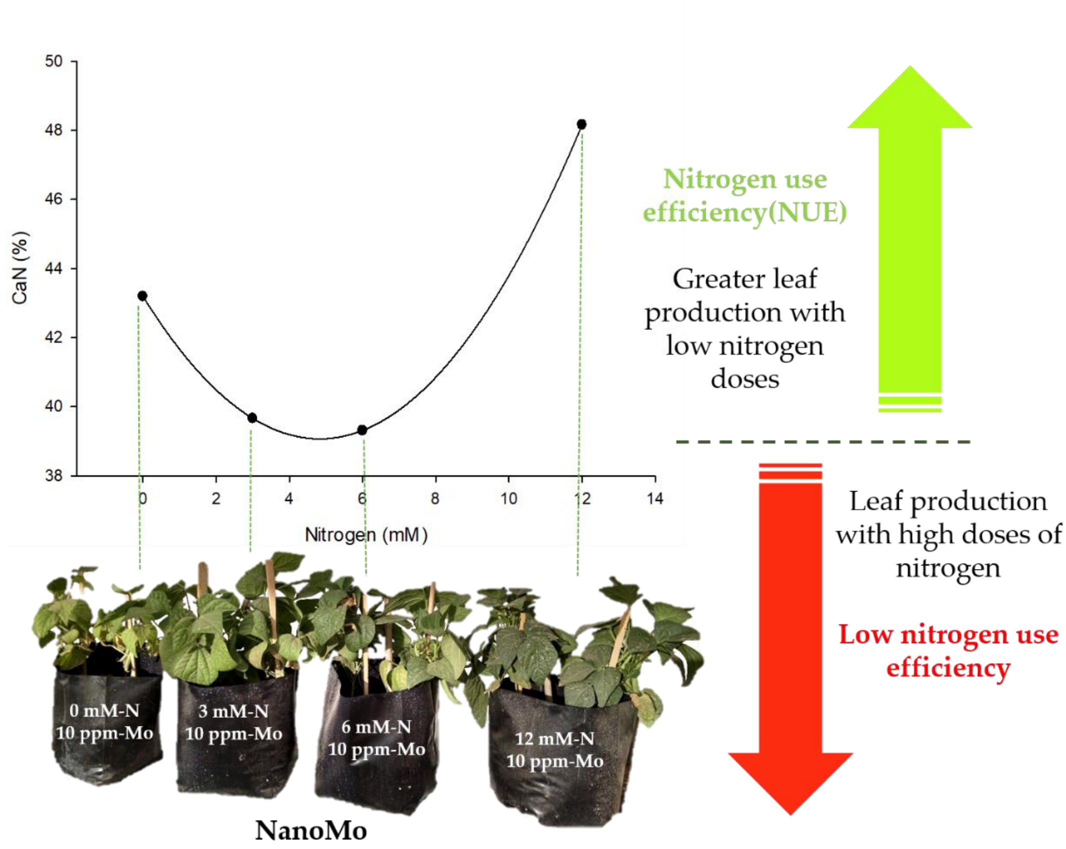

Figure 4. Nitrogen use efficiency in biomass production as an effect of foliar application of NanoMo. The image shows how the combination of low doses of nitrogen (3mM) and low doses of NanoMo (10 ppm) favors biomass production, increasing productivity efficiency per unit of fertilizer applied.

Regarding the nitrogen fixation efficiency (NFiE) (Figure 3c), the favorable effect of NanoMo and nitrogen is clearly seen at the doses of 6mM-N and 10 ppm-Mo. This interaction potentiates the transformation of absorbed nitrogen into abundant leaf tissue (greater number of leaves and larger size) which will be essential for the survival and optimal development of the plant. The efficient use of nitrogen transformed into dry matter is an unequivocal indicator of adequate use of nitrogen fertilizers.

3.1.1. NUE Indices for Stems

NanoMo had a quite favorable response in stem production, unlike the other two sources of molybdenum (Table 2). This difference was 43.52% more for the chelate and 23.84% more for the molybdate. Likewise, in Figure 4a we can observe the effect of the NanoMo doses directly on this index. The graph shows how the greatest development of stems was obtained with the dose of 10 ppm, and that a higher dose can cause a significant reduction in the formation of stems, in thickness, height and number.

In the indices that determine the percentage by weight of the plant organ (WxWs) and the amount of nitrogen to form each organ (CaNs), no significant statistical difference was found between the sources of molybdenum (Table 2); For this reason, the use of nanofertilizer continues to be prioritized due to its high efficiency in nitrogen assimilation and fixation. In the response surface analysis (Figure 4c) the importance of efficient nitrogen fixation can be observed, since this nitrogen is responsible for the formation and proper development of the stems.

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Table 2. Effect of edaphic nitrogen fertilization supplemented with foliar fertilization of molybdenum nanofertilizer on NUE indices in stems.

	Indices NUE (Stems)				
	PTs	**PxPs**	**mg-Ns**	**CaNs**	**NCoE**
Mo Source	0.0004[U]	0.2506	0.0003	0.4423	0.0074
Nano Mo	**4.32 a**[V]	23.29 a	**58.91 a**	24.82 a	9.52 b
Mo Chelate	2.44 c	23.88 a	45.49 b	**27.26 a**	**9.89 b**
Na Molybdate	3.29 b	**25.53 a**	34.25 c	26.24 a	12.90 a
MSD	0.80[W]	3.59	10.31	5.10	2.45
Nitrogen[X]	0.0149	0.0104	<0.0001	<0.0001	<0.0001
0	2.68 b	22.52 b	24.23 b	17.65 b	**14.47 a**
3	3.53 ab	25.41 ab	33.98 b	26.79 a	13.31 a
6	**4.10 a**	**25.86 a**	62.06 a	29.45 a	9.07 b
12	3.09 ab	23.14 ab	**64.59 a**	**30.55 a**	6.24 c
MSD	1.15	2.98	14.04	5.53	1.73
Molybdenum[Y]	<0.0001	0.4451	0.1723	0.0065	0.0078
0	2.77 b	**25.39 a**	**50.77 a**	**29.29 a**	9.14 b
5	2.85 b	24.00 a	47.76 a	27.59 ab	10.40 ab
10	**4.08 a**	23.57 a	43.34 a	22.77 b	**12.29 a**
20	3.70 a	23.97 a	42.99 a	24.78 ab	11.26 ab
MSD	0.74	3.10	10.59	5.15	2.41
SoMo*N	0.6515	0.8421	0.0004	<0.0001	<0.0001
SoMo*Mo	<0.0001	0.4362	0.2316	0.0287	0.9227
N*Mo	0.0857	0.3859	0.0111	0.1252	0.4571
SoMo*N*Mo	0.2070	0.8777	0.0022	0.0001	0.1112
μ	3.35	24.23	46.21	26.11	10.77
C.V.	41.40	24.03	43.01	37.06	42.09
R^2	0.7113	0.4407	0.7834	0.6900	0.7300

[U]Probability not significant Pr > 0.05, significant 0.05 ≤Pr≤0.01, highly significant Pr <0.0001; [V]Means with the same letter are statistically equal; [W]Least significant difference; [X]Mm edaphic nitrogen concentration; [Y]Leaf ppm concentration of molybdenum, overall average μ, CV coefficient of variation, R^2 coefficient of determination; Regression Analysis: Linear L, Quadratic C, P N*Mo interaction.

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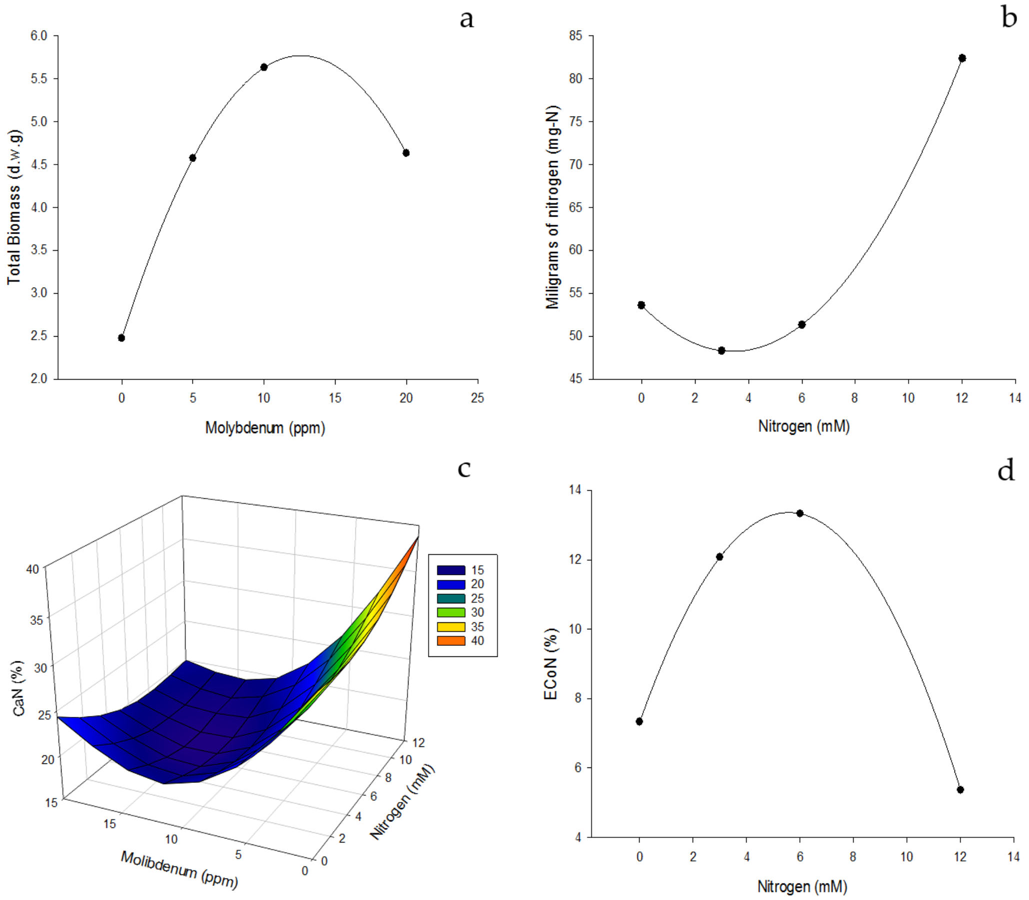

Figure 4. Effect of edaphic nitrogen fertilization, complemented with foliar fertilization with molybdenum nanofertilizer on NUE indices for stem formation. (a) Total stem biomass (grams dry weight). (b) Milligrams of nitrogen needed to form the plant stems (mg-Ns). (c) Amount of nitrogen to form the stems of the plant (CaNs). (d) Nitrogen conduction efficiency (NCoE).

On the other hand, in Figure 4b referring to the index that determines the milligrams of nitrogen necessary to form the stems, a behavior similar to that recorded for the leaves can be seen. Where, the low doses of nitrogen (3 and 6 mM) were the most efficient in the use of nitrogen, since, with a smaller amount and efficiently used in the plant metabolism, the development of a system was promoted. Stems large enough to hold the size of the entire foliage of the plant. Similarly, the graph shows how plants that are subjected to excessive nitrogen fertilization (12 mM) use a greater amount of nitrogen to produce dry matter, breaking with the principle of nitrogen use efficiency.

Finally, the index that measures the efficiency of nitrogen conduction through the stems (NCoEt) shows how plants fertilized with NanoMo and with low doses of nitrogen (3 and 6 mM) had the highest conduction efficiency (Figure 4d). And, on the contrary, high doses of nitrogen (12 mM) led to a dramatic drop in translocation efficiency by approximately 61% compared to the 6 mM dose.

3.1.2. NUE Indices for Root

In the present investigation, root development was favored by the application of NanoMo. The difference in total biomass varied from 2.29% with respect to sodium molybdate and 25.96% with respect to molybdenum chelate. These differences in root volume allowed the plants fertilized with the nanofertilizer to develop a larger and more efficient root system. This efficiency of nitrogen use in the root can also be verified with the index that determined the milligrams of nitrogen used to form the dry matter of the root (mg-Nr), where the difference in this index in the NanoMo with respect to the other sources of molybdenum was around 50% (Table 3).

On the other hand, it is important to highlight how the source of sodium molybdate registers a higher percentage of the root weight (WxWr) compared to the chelate and NanoMo; Similarly, the

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nitrogen absorption efficiency index (NAbE) shows how molybdate led to the highest absorption efficiency, where the difference is 36.56% more than the chelate and 56.50% more than the nanofertilizer. To explain this behavior, it is necessary to analyze the data from all the plant organs together. This apparent superiority of the effect of molybdate in these two parameters did not translate into a positive effect on the total development of the plant, since the greatest production of foliage (leaves) and productivity (fruit production) was obtained with the NanoMo application. This means that the amount of nitrogen absorbed by the plants under molybdenum fertilization did not necessarily translate into development of aerial biomass and fruit formation, and only had an effect of nitrogen overaccumulation that could affect optimal plant development.

Table 3. Effect of edaphic nitrogen fertilization supplemented with foliar fertilization of molybdenum nanofertilizer on NUE indices in roots.

Indices NUE (Roots)					
	PTr	**WxWr**	**mg-Nr**	**CaNr**	**NAbE**
Mo Source	0.0141[U]	<0.0001	<0.0001	0.3945	<0.0001
Nano Mo	**1.31 a**[V]	15.22 c	**47.30 a**	**21.84 a**	5.75 c
Mo Chelate	0.97 b	18.74 b	21.91 b	19.42 a	8.40 b
Na Molybdate	1.28 a	**24.22 a**	21.83 b	21.66 a	**13.24 a**
MSD	0.27[W]	3.00	7.52	5.24	2.39
Nitrogen[X]	0.0006	<0.0001	0.0002	<0.0001	<0.0001
0	1.31 a	**25.80 a**	31.75 a	**35.50 a**	**16.70 a**
3	**1.34 a**	18.39 b	32.14 a	23.40 b	9.30 b
6	**1.34 a**	18.26 b	**36.09 a**	16.02 c	6.36 c
12	0.85 b	15.13 b	21.40 b	8.98 d	4.15 c
MSD	0.31	3.39	7.95	5.87	2.29
Molybdenum[Y]	0.0995	0.8598	0.1130	0.2469	0.0318
0	1.12 a	20.11 a	28.25 a	19.98 a	7.57 b
5	1.05 a	19.34 a	27.97 a	19.19 a	8.54 ab
10	1.31 a	19.40 a	33.34 a	23.04 a	10.54 a
20	1.26 a	18.73 a	31.83 a	21.70 a	9.88 ab
MSD	0.30	4.14	6.86	5.39	2.81
SoMo*N	0.1897	0.0274	0.5581	<0.0001	<0.0001
SoMo*Mo	0.0719	0.1580	0.0004	0.3757	0.4295
N*Mo	0.0264	0.6066	0.0079	0.0027	0.3090
SoMo*N*Mo	0.0006	0.0922	0.0001	0.0403	0.2646
μ	1.19	19.39	30.35	20.98	9.13
C.V.	48.65	40.10	42.49	48.24	57.94
R^2	0.6052	0.6114	0.7714	0.7642	0.7653

[U]Probability not significant Pr > 0.05, significant 0.05 ≤Pr≤0.01, highly significant Pr <0.0001; [V]Means with the same letter are statistically equal; [W]Least significant difference; [X]Mm edaphic nitrogen concentration; [Y]Leaf ppm concentration of molybdenum, overall average μ, CV coefficient of variation, R^2 coefficient of determination; Regression Analysis: Linear L, Quadratic C, P N*Mo interaction.

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It is important to highlight the direct effect that the NanoMo doses had on the total biomass (Figure 5a). The graph shows how with the dose of 10 ppm of molybdenum the maximum biomass production was achieved. Likewise, we can see how a double dose (20 ppm) causes a drastic drop in leaf production, most likely due to overfertilization with molybdenum. In the percentage index by weight of the root (WxWr) (Figure 5b), it can be seen that with the increase in nitrogen doses, the percentage by weight of the root decreases. The direction of the graph expresses how the volume of the root decreases as there is a greater contribution and consequently an overaccumulation of nitrogen, which results in a decrease in the production of root biomass. Figure 5c (mg-N indice) shows how the 10 ppm dose of molybdenum had greater efficiency in the use of nitrogen to form the root. On the other hand, in Figures 5d (CaNr indice) and 5e (EAbN indice) a similar behavior can be seen, where the dose of 3 mM of nitrogen had the highest absorption efficiency (5e), and greater formation root with the lowest dose of nitrogen (5d).

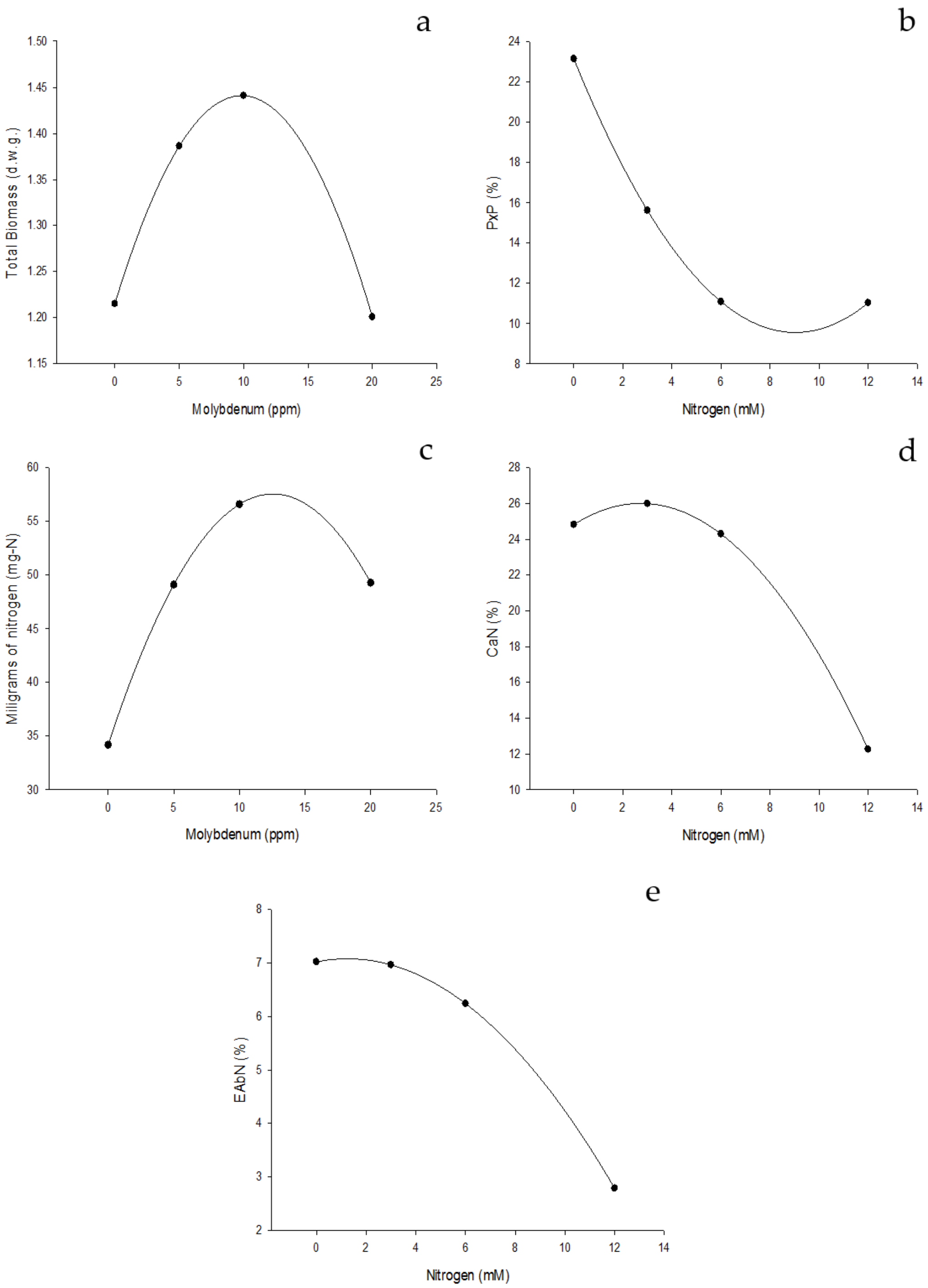

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Figure 5. Effect of edaphic nitrogen fertilization, complemented with foliar fertilization with molybdenum nanofertilizer on NUE indices for root formation. (a) Total biomass (grams of dry weight). (b) Percentage by weight, which represents the root of the total weight of the plant (%). (c) Milligrams of nitrogen needed to form the plant root (mg-N). (d) Amount of nitrogen to form the root of the plant (CaNr). (e) Nitrogen absorption efficiency (NAbE).

3.1.3. Indices NUE Para Fruit.

The highest efficiency of nitrogen use for fruit production was achieved with the application of NanoMo (Table 4). The difference in production with respect to the chelate source was 45.78% more, and 17.77% more than with the molybdate source. The ease of absorption and translocation of NanoMo allowed the plant to have the molybdenum necessary to assimilate the absorbed nitrogen through the enzymatic metabolism responsible for its transformation; and in this way it can be used by the plant for its growth and fruit production.

Another determining index to measure the efficiency of nitrogen use for fruit formation was the mg-N index, which shows that with the application of NanoMo, the plants achieved greater productivity and use of nitrogen by 60.22% more milligrams used. for fruit formation than the chelate source, and 11.43% more than plants treated with molybdate. Similarly, the productivity efficiency of the nanofertilizer exceeded the chelate and molybdate sources by 45.10% and 20.19% respectively. These results show the capacity of the nanofertilizer to increase the efficiency of nitrogen use in favor of the crop, to produce more fruits per unit of applied nitrogen.

Table 4. Effect of edaphic nitrogen fertilization supplemented with foliar fertilization of molybdenum nanofertilizer on NUE indices in fruit.

Indices NUE (Fruit)					
	PTf	**WxWf**	**mg-Nf**	**CaNf**	**Productivity**
Mo Source	0.0020^{U}	0.0145	0.0923	0.0334	0.0059
Nano Mo	**2.25 a**^{V}	**8.97 a**	**28.78 a**	10.73 ab	**0.0510 a**
Mo Chelate	1.22 b	3.31 b	11.45 a	5.25 b	0.0280 b
Na Molybdate	1.85 a	6.72 ab	25.49 a	**12.00 a**	0.0407 ab
MSD	0.55^{W}	4.24	20.5	6.28	0.0147
Nitrogen^{X}	0.5207	0.3797	0.0476	0.5260	0.0005
0	1.62 a	5.41 a	7.91 a	6.47 a	**0.0543 a**
3	1.72 a	5.01 a	15.76 a	10.09 a	0.0490 a
6	**2.05 a**	6.54 a	29.40 a	**10.38 a**	0.0356 ab
12	1.70 a	**8.36 a**	**34.56 a**	10.37 a	0.0209 b
MSD	0.84	5.62	27.33	8.47	0.0203
Molybdenum^{Y}	0.0001	0.0030	0.0095	0.0030	0.0008
0	1.43 bc	1.18 b	13.73 b	6.04 b	0.0229 b
5	1.39 c	4.30 b	12.80 b	6.40 b	0.0357 ab
10	**2.28 a**	**9.13 a**	**33.16 a**	**13.19 a**	**0.0524 a**
20	1.99 ab	7.72 ab	27.93 ab	11.68 ab	0.0488 a
MSD	0.58	1.13	18.82	6.05	0.0201
SoMo*N	0.7188	0.5879	0.6899	0.8514	0.0011
SoMo*Mo	0.0097	0.0011	0.0224	0.0048	0.0094
N*Mo	0.3347	0.7546	0.5063	0.9849	0.2249
SoMo*N*Mo	0.1443	0.9891	0.9834	0.9164	0.0687
μ	1.77	6.33	21.91	9.33	0.03

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C.V.	61.29	122.40	161.29	121.71	94.54
R^2	0.62	0.56	0.56	0.54	0.61

[U]Probability not significant Pr > 0.05, significant 0.05 ≤Pr≤0.01, highly significant Pr <0.0001; [V]Means with the same letter are statistically equal; [W]Least significant difference; [X]Mm edaphic nitrogen concentration; [Y]Leaf ppm concentration of molybdenum, overall average μ, CV coefficient of variation, R^2 coefficient of determination; Regression Analysis: Linear L, Quadratic C, P N*Mo interaction.

On the other hand, the direct effect of NanoMo on nitrogen indices should be highlighted. The graphs in Figure 6 show a similar trend, where the dose of 10 ppm was the most efficient for fruit production. With the application of 10 ppm, the plants achieved greater efficiency in the use of fertilizers to produce a greater amount of fruit (parameter PTf, Figure 6a). Similarly, the highest nitrogen use efficiency was achieved, where a greater number of fruits were produced per milligrams of nitrogen used (mg-N indice, Figure 6c). The greater efficiency of utilization of absorbed nitrogen for fruit production (Productivity indice) per unit of nitrogen and molybdenum applied was also favored by the application of 10 ppm of NanoMo. It is important to highlight that applying a higher dose of NanoMo (20 ppm or more) causes a drop in yield, this may be caused by the overaccumulation of molybdenum in the plant, derived from the high absorption efficiency that the plant has nanofertilizer. This particular characteristic should be considered essential, and special care should be taken in fertilization programs to avoid toxicity in plants.

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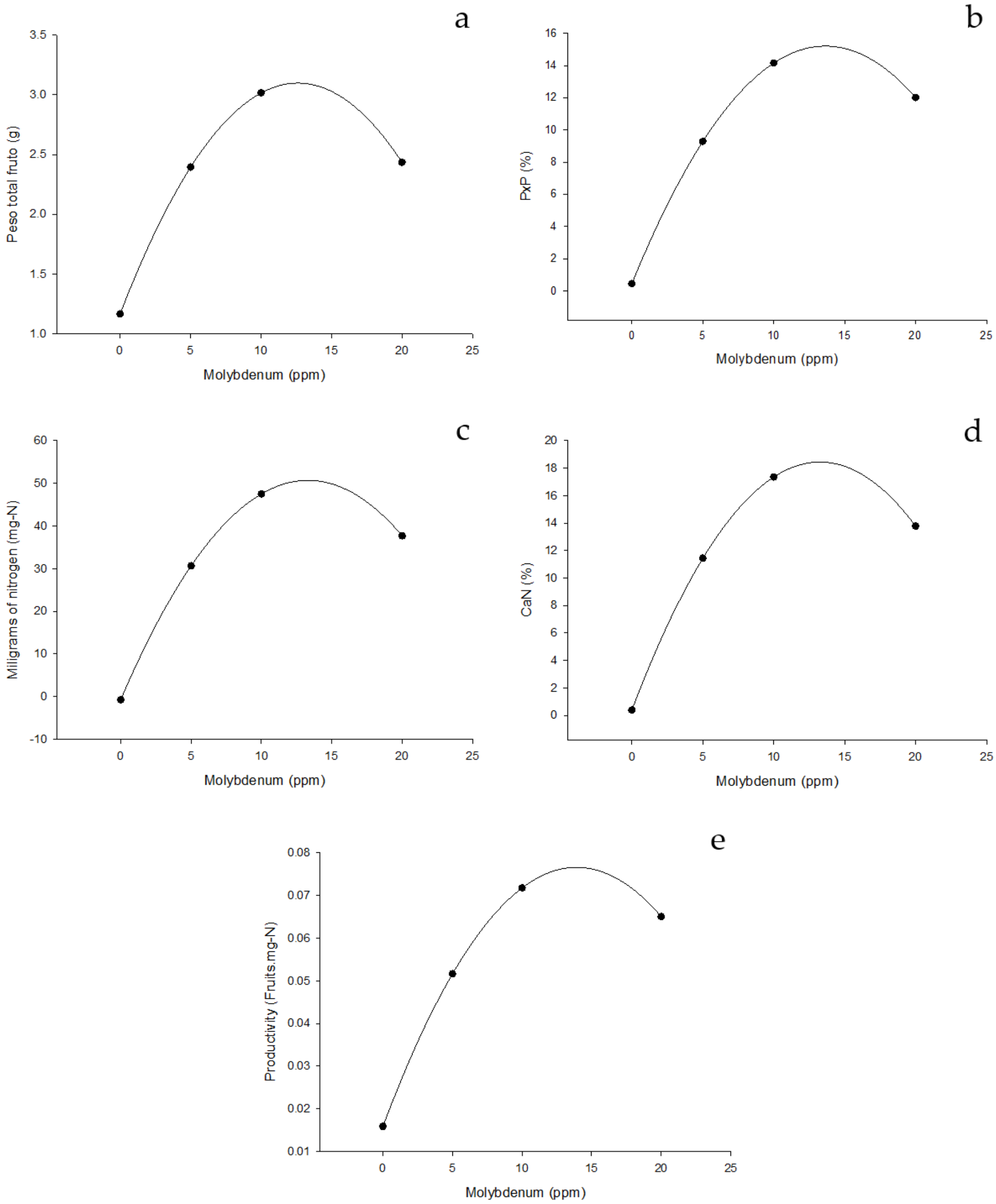

Figure 6. Effect of edaphic nitrogen fertilization, complemented with foliar fertilization with molybdenum nanofertilizer on NUE indices for fruit formation. (a) Total biomass (grams of dry weight). (b) Percentage by weight, which represents the fruit of the total weight of the plant (%) (c) Milligrams of nitrogen required for fruit formation (mg-N). (d) Amount of nitrogen to form the fruits of the plant (CaNf). (e) Productivity (Fruits.mg-N).

4. Discussion

Nitrogen shortage or overfertilization severely affects the physiological, molecular and biochemical responses of the plan. In addition, the general metabolism and the distribution of metabolites and general resources necessary for their survival are affected [14]. For this reason, nitrogen is considered among all essential nutrients, the most limiting nutrient for agricultural production [5]. However, its intensive and unbalanced use is strongly related to the losses that cause low efficiency in the use of nitrogen fertilizers (NUE) [15]. Given the urgency of solving the problem of low nitrogen use efficiency, techniques were used that allowed raising the level of use of this nutrient to the maximum. The use of microelements as important as molybdenum, essential in the

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nitrogen assimilation process; and the use of cutting-edge tools such as nanotechnology, have proven to be nitrogen use efficiency.

Nitrogen use efficiency represents the ability of plants to use the mineral nitrogen that is available. For this reason, its definition implies the increase in yield per unit of nitrogen applied, absorbed and used by the plant for the production of economically viable fruit [16]. However, it is also used to determine the production efficiency of the entire plant biomass. For this reason, the different parameters calculated in the present research were able to demonstrate the positive effect of NanoMo on the efficiency of nitrogen use in the production of leaves, stems, roots and fruit in bean plants. The particular characteristics of NanoMo facilitated its penetration through the surface of the leaves, and its nanometric size allowed it to be distributed throughout all tissues, which guaranteed an effective concentration required by the plant for its growth (Figure 7) [17].

Figure 7. Effect of NanoMo on Nitrogen Use Efficiency in green beans cv. Strike on the different NUE indices.

The importance of Mo being available in the cells lies in the fact that it is a metallic component that plays a fundamental role in the biosynthesis of the cofactor Moco, which binds to the molybdoenzymes (enzymes that require molybdenum) responsible for the reduction, assimilation and nitrate fixation (Nitrate reductase (NR) and Nitrite reductase (NiR)), in addition to the regulation of the enzymes Glutamine synthetase (GS) and glutamate synthase (GOGAT) responsible for the assimilation of ammonium [6]. In this way, a sufficient amount of Mo allowed the plant to use it in the metabolic process of nitrogen assimilation, and that the activity of the enzymes responsible for its assimilation and fixation was not stopped or decreased. Likewise, nitrogen used efficiently allowed the development of meristems, the formation and growth of leaf tissue in the increase in the growth rate and elongation of the leaves [18] (Figure 8).

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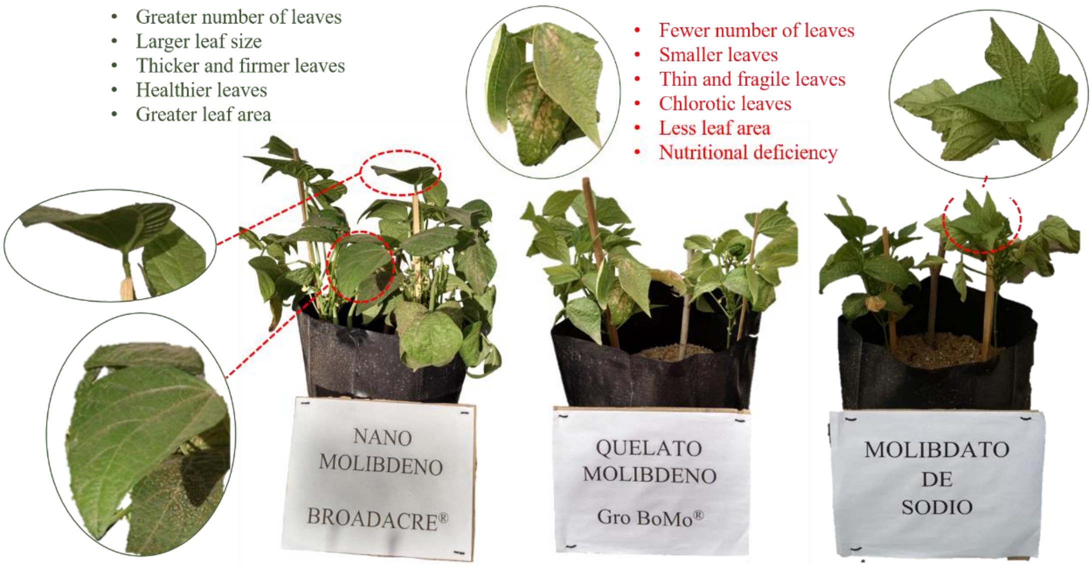

Figure 8. Effect of NanoMo on NUE in green beans cv. Strike on the growth and development of leaves.

The adequate supply of nutrients and especially nitrogen, allows the plant to develop a support system strong enough so that it can remain upright, and prevent the stems from breaking despite the overturning forces that the plant can generate. wind and the weight of the plant itself; The stems must have the ability to redirect and transmit those forces to the anchoring system in the ground [19].

Likewise, the effect of NUE was reflected in the formation of strong and resistant stems in the plants treated with NanoMo, which were not prevented from developing properly. On the other hand, the deficiency in the growth of plants treated with Chelate and Molybdate, supposes a low assimilation of nitrogen, which consequently affects the production of proteins and various nitrogen products essential for the development of the plant. The low assimilation of nitrogen triggered various metabolic effects, which strongly impact metabolic pathways, and specific actions that involve some macronutrients that act on the specific development of the stems. In this case, it can be assumed that there was an affectation in the phosphorus (P) cycle, which results in a reduction in the synthesis of cellulose, starch and sucrose, which affects the formation of stems, in such a way that the plants may present dwarfism. Likewise, the activity of potassium (K) could be affected; the inaction or deficiency of this element caused by the N-K relationship produces a stagnation in the development of the plant, especially by shortening the internodes of the stems, making them weaker [20] (Figure 8).

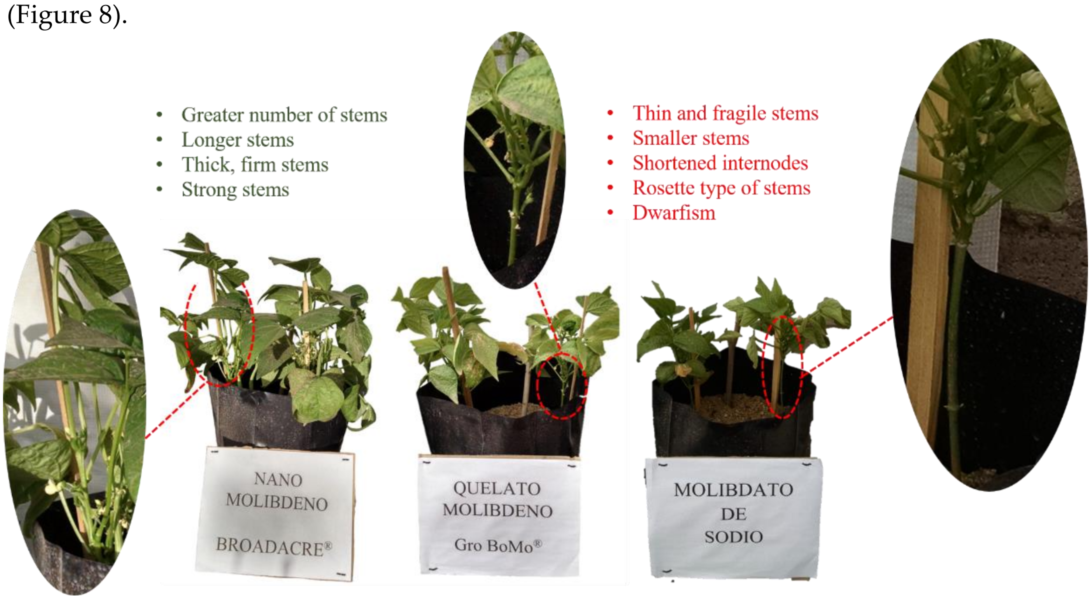

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Figure 9. Effect of NanoMo on Nitrogen Use Efficiency in green beans cv. Strike on the growth and development of stems.

The root is the fundamental organ of the plant that anchors it to the soil. Furthermore, it has the indispensable function of capturing water and nutrients from the soil, and is a site of great interaction with biotic and abiotic factors that are frequently determinants for crop productivity [21]. For good root development to occur, there must be an adequate supply of nutrients, especially nitrogen. The absence or shortage of this element drastically reduces root growth, which directly impacts the development and quality of the plant [22].

The development of a strong and abundant root system facilitates the absorption and translocation of nitrogen for the development of the entire vegetative system, and especially in fruit formation [23]. The adequate nutritional status of the plants fertilized with NanoMo, allowed, among many other plant mechanisms, the timely activation of nutrient transporters, which are distributed in all their organs. In the particular case of this research, nitrate transporters, responsible for the absorption and transport of nitrate to assimilation sites within cells [24]. In such a way, plants with a high nitrogen use efficiency develop a strong and extensive root system during their growth and vegetative development; which is an essential basis for the continuous absorption and translocation of nitrogen, until culminating in the stage of fruit formation and filling [25,26] (Figure 10).

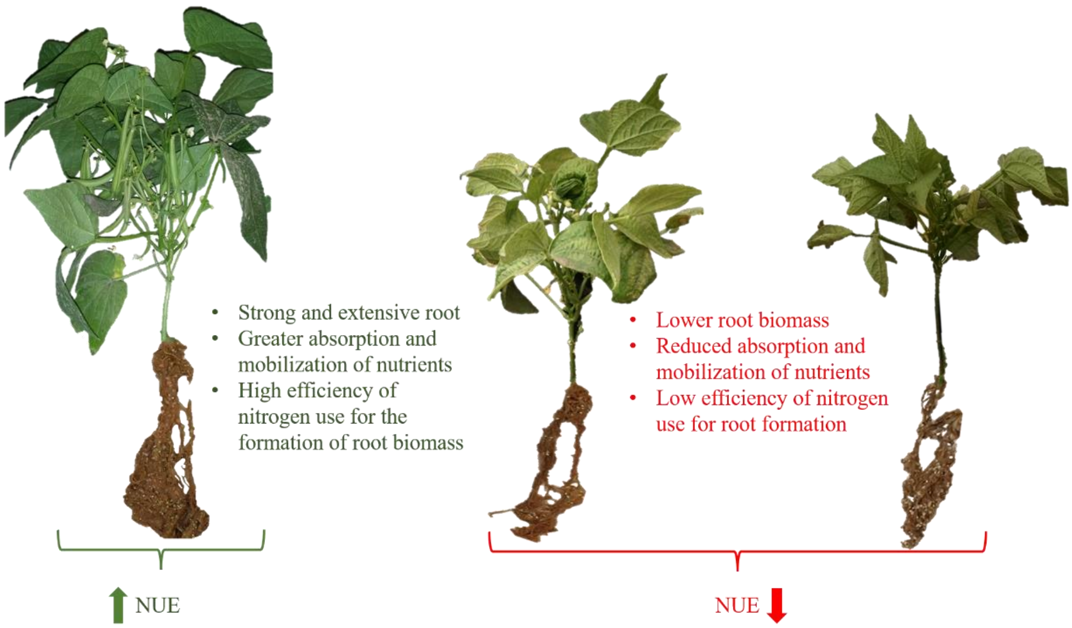

Figure 10. Effect of NanoMo on NUE in green beans cv. Strike on root growth and development.

Nitrogen is vital in increasing crop yields. Adequate supply of this nutrient can increase performance by up to 40% [27]. Furthermore, the quality of the fruits depends on the quantity and proportion of amino acids, proteins, vitamins, sugars and other metabolites that depend on the supply of nitrogen by the plant [28]. In addition, it is necessary to increase the efficient use of nitrogen to improve the productivity and quality of the fruits.

It is very important to highlight that the high quality of the fruits is vital for consumers and food processors. In addition, high-quality fruits are mainly desired in the world market, where they are expected to have good flavor, excellent appearance, firmness, size and, above all, be rich in nutrients essential for health. All this can be achieved with proper crop management, and especially fertilization management [29]. For this reason, the efficient use of nitrogen and nanomolybdenum allowed the bean plants to produce a greater quantity of high-quality fruit; since nitrogen directly impacts fruit growth and regulates its quality [27]. Furthermore, the high assimilation efficiency of nanomolybdenum allowed optimizing the metabolic functions of the enzymes and cofactors responsible for the reduction, assimilation and fixation of nitrogen, which are intended for plant

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growth and fruit development [6]. . Efficiently assimilated nitrogen is an important component of the chemical structure of proteins, chlorophylls, some phytohormones, nucleic acids and secondary metabolites responsible for the quality of the fruits [30]. In this way, a higher content of sugars, antioxidant compounds and firmness are guaranteed [31,32].

On the other hand, [33] reported that high nitrogen applications combined with low efficiency caused a decrease in phenolic content, yield and nutritional content. In addition, the size, shape, color and general appearance of the olive fruit was affected. Likewise, most crops are undoubtedly affected by nitrogen, and it is for this reason that the calculation of nitrogen parameters is a reliable tool to effectively determine the use of nitrogen and other essential nutrients for plant development and performance.

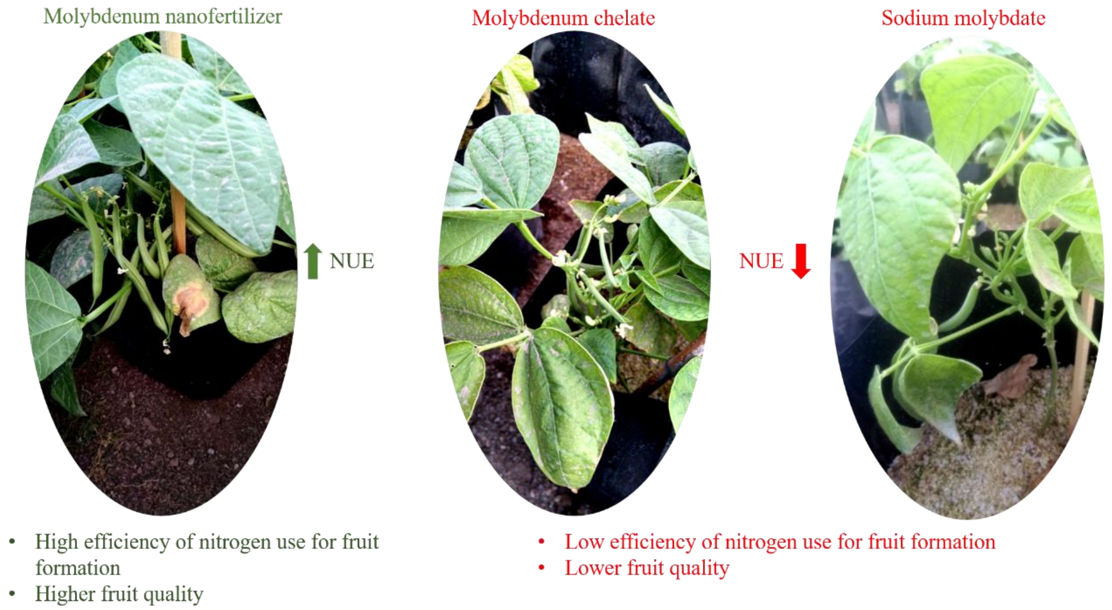

Figure 11. Effect of NanoMo on NUE in green beans cv. Strike on the growth and development of the fruit.

5. Conclusions

Foliar applications of Nanomolybdenum considerably increased the efficiency of nitrogen use, which increased the productivity of bean plants per unit of applied nitrogen by 42% and 37% more in the leaf, 44% and 24% more in the stem, 26% and 2% more in roots, and 46% and 18% in fruit production than chelate and molybdate respectively.

Furthermore, the determination of the NUE through the use of the different efficiency indices allowed us to specify the final destination of the assimilated nitrogen and its use in the production of leaves, stems, fruit and roots of the green bean plants.

Finally, the results prove that the use of NUE indices is an important approach to evaluate the efficiency of nitrogen applied to crops; and that can be used to estimate the growth, development and yield of cultivated plants.

Author Contributions: E.S, J.M.S.P. and E.M.M. designed the study. E.S., R.P.L. and J.M.S.P. analyzed the data, E.S and E.M.M. prepared the manuscript, while ǀwed E.M.M., R.P.L. forks. conducted the experiments. E.S., J.M.S.P. and E.M.M. organized the data and performed the statistical analysis. All authors read and approved the final manuscript.

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Conflicts of Interest: The authors declare no conflict of interest.

Appendix A

The data from the response surface analysis are shown below to give greater clarity and statistical support to the graphs.

1. Indices NUE leaves

Table 5. Response surface analysis for molybdenum sources in leaves total biomass (TWl).

TWl											
Nano Mo				Mo Chelate				Na Molybdate			
CV	23.26	**R^2**	0.7288	**CV**	44.96	**R^2**	0.1651	**CV**	51.16	**R^2**	0.1684
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	<0.0001	N	Mo	L	0.9319	N	Mo	L	0.0529	N	Mo
C	<0.0001	<0.0001	<0.0001	C	0.0075	0.0239	0.5817	C	0.0748	0.0582	0.2753
P	0.1850	L, C	L, C	P	0.4143	L, C		P	0.7155	L	
Model	<0.0001	µ 8.93		Model	0.0571	µ 5.21		Model	0.0521	µ 5.64	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	2.4162	0.7485	0.0021	**Int**	4.4112	0.8439	<0.0001	**Int**	3.4782	1.0398	0.0014
N	1.5667	0.2217	<0.0001	**N**	0.7443	0.2499	0.0042	**N**	0.7424	0.3080	0.0191
Mo	1.0716	0.1330	<0.0001	**Mo**	- 0.1231	0.1499	0.4148	**Mo**	0.0376	0.1848	0.8392
N*N	-0.1212	0.0162	<0.0001	**N*N**	-- 0.0562	0.0183	0.0033	**N*N**	- 0.0524	0.0226	0.0239
Mo*N	0.0106	0.0079	0.1850	**Mo*N**	- 0.0073	0.0089	0.4143	**Mo*N**	0.0040	0.0109	0.7155
Mo*Mo	-0.0445	0.0058	<0.0001	**Mo*Mo**	0.0073	0.0066	0.2697	**Mo*Mo**	0.0017	0.0081	0.8308
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	-4.0867	0.7547	0.6559	**Eigenva**	0.7537	- 0.0789	0.9968	**Eigenva**	0.1818	0.0582	0.9983
	-4.7302	-0.6559	0.7547		- 2.0440	0.9968	0.0789		- 1.8950	0.9983	- 0.0582

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 6. Response surface analysis for molybdenum sources in leaves percentage by weight (*PxWl*).

PxWl											
Nano Mo				Mo Chelate				Na Molybdate			
CV	16.37	**R^2**	0.2795	**CV**	15.69	**R^2**	0.0758	**CV**	2907	**R^2**	0.0744
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	

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L	0.0162	N	Mo	L	0.2688	N	Mo	L	0.4616	N	Mo
C	0.0979	0.0004	0.0235	C	0.6304	0.2331	0.6860	C	0.3894	0.2179	0.7378
P	0.0044	L, C		P	0.2910			P	0.2825		
Model	0.0016	μ 52.50		Model	0.4554	μ 54.05		Model	0.4669	μ 43.52	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	44.0200	3.0966	<0.0001	**Int**	53.0349	3.0567	<0.0001	**Int**	46.8362	4.5592	<0.0001
N	3.2661	0.9172	0.0007	**N**	0.8457	0.9054	0.3542	**N**	-1.7058	1.3504	0.2116
Mo	-0.0010	0.5503	0.9985	**Mo**	-0.1596	0.5432	0.7699	**Mo**	-0.4726	0.8102	0.5619
N*N	-0.1392	0.0673	0.0431	**N*N**	-0.0626	0.0664	0.3499	**N*N**	0.1357	0.0991	0.1761
Mo*N	-0.0971	0.0327	0.0044	**Mo*N**	0.0344	0.0323	0.2910	**Mo*N**	0.0523	0.0482	0.2825
Mo*Mo	0.0181	0.0242	0.4564	**Mo*Mo**	-0.0048	0.0239	0.8387	**Mo*Mo**	0.0073	0.0356	0.8385
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	2.8915	-0.3457	0.9383	**Eigenva**	-0.0131	0.4185	0.9081	**Eigenva**	5.4142	0.9482	0.3176
	-6.0879	0.9383	0.3457		-2.7318	0.9081	-0.4185		0.2050	-0.3176	0.9482

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 7. Response surface analysis for molybdenum sources in milligrams of nitrogen (*mg-N leaves*).

mg-N leaves											
Nano Mo				**Mo Chelate**				**Na Molybdate**			
CV	44.23	**R^2**	0.2173	**CV**	48.86	**R^2**	0.4384	**CV**	51.14	**R^2**	0.5062
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0073	N	Mo	L	<0.0001	N	Mo	L	<0.0001	N	Mo
C	0.1025	0.0127	0.1879	C	0.0023	<0.0001	0.0346	C	0.0122	<0.0001	0.1929
P	0.4235	L	L, C	P	0.1731	L, C	L	P	0.0581	L, C	
Model	0.0124	μ 103.92		Model	<0.0001	μ 77.46		Model	<0.0001	μ 51.83	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	65.0628	16.5603	0.0002	**Int**	61.936	13.6358	<0.0001	**Int**	17.3333	9.5470	0.0746
N	0.3056	4.9052	0.9505	**N**	14.3558	4.0389	0.0008	**N**	11.1359	2.8252	0.0002
Mo	6.4568	2.9431	0.0323	**Mo**	-7.0825	2.4233	0.0049	**Mo**	0.3015	1.6997	0.8595
N*N	0.4027	0.3601	0.2681	**N*N**	-0.8409	0.2965	0.0063	**N*N**	-0.6285	0.2076	0.0037
Mo*N	-0.1411	0.1751	0.4235	**Mo*N**	0.1988	0.1441	0.1731	**Mo*N**	0.1951	0.1009	0.0581
Mo*Mo	-0.2421	0.1296	0.0668	**Mo*Mo**	0.2493	0.1067	0.0230	**Mo*Mo**	0.0448	0.0747	0.5508
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	14.9555	0.9944	-0.1074	**Eigenva**	25.5730	0.1062	0.9943	**Eigenva**	-2.7597	0.2826	0.9592
	-24.6749	0.1074	0.9942		-30.9122	0.9943	-0.1062		-24.3541	0.9592	-0.2826

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 8. Response surface analysis for molybdenum sources in quantity of nitrogen in percentage (%) (*CaNl*).

CaNl											
Nano Mo				Mo Chelate				Na Molybdate			
CV	31.21	**R^2**	0.1391	**CV**	30.90	**R^2**	0.1904	**CV**	45.17	**R^2**	0.1788
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.4004	N	Mo	L	0.0753	N	Mo	L	0.0173	N	Mo
C	0.1628	0.0487	0.2009	C	0.3370	0.0249	0.0299	C	0.3900	0.0181	0.2998
P	0.0572		L	P	0.0172		L	P	0.1618	L, C	
Model	0.1129	μ 42.58		Model	0.0278	μ 48.05		Model	0.0390	μ 40.44	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	41.1870	4.7883	<0.0001	**Int**	57.4500	5.3505	<0.0001	**Int**	33.3585	6.5817	<0.0001
N	-0.8474	1.4183	0.5525	**N**	-1.4097	1.5849	0.3774	**N**	1.6225	1.9495	0.4087
Mo	-0.2934	0.8509	0.7315	**Mo**	-2.2164	0.9509	0.0233	**Mo**	0.7249	1.1697	0.5379
N*N	0.1767	0.1041	0.0950	**N*N**	0.0794	0.1163	0.4977	**N*N**	-0.0787	0.1431	0.5843
Mo*N	-0.0982	0.0506	0.0572	**Mo*N**	0.1388	0.0565	0.0172	**Mo*N**	0.0986	0.0696	0.1618
Mo*Mo	0.0348	0.0374	0.3562	**Mo*Mo**	0.0554	0.0418	0.1909	**Mo*Mo**	-0.0654	0.0515	0.2094
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	8.2041	0.8480	-0.5298	**Eigenva**	8.5766	0.5887	0.8083	**Eigenva**	-1.1965	0.8748	0.4843
	1.6456	0.5298	0.8480		-0.1741	0.8083	-0.5887		-8.1796	-0.4843	0.8748

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 9. Response surface analysis for molybdenum sources in Nitrogen fixation efficiency (*NFiE*).

NFiE											
Nano Mo				Mo Chelate				Na Molybdate			
CV	50.26	**R^2**	0.3463	**CV**	78.08	**R^2**	0.3554	**CV**	38.33	**R^2**	0.5644
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.1641	N	Mo	L	0.0001	N	Mo	L	<0.0001	N	Mo
C	<0.0001	0.0002	0.0559	C	0.0106	<0.0001	0.1565	C	<0.0001	<0.0001	0.7685
P	0.4688	L, C	L	P	0.2757	L, C	L	P	0.6558	L, C	
Model	0.0001	μ 21.54		Model	<0.0001	μ 2348		Model	<0.0001	μ 22.24	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	8.8866	3.9011	0.0264	**Int**	31.6067	6.6101	<0.0001	**Int**	34.8012	3.0713	<0.0001
N	4.8201	1.1555	0.0001	**N**	-6.1431	1.9579	0.0027	**N**	-5.8532	0.9097	<0.0001
Mo	1.8566	0.6933	0.0096	**Mo**	2.6727	1.1747	0.0266	**Mo**	0.4776	0.5458	0.3852
N*N	-0.3935	0.0848	<0.0001	**N*N**	0.3612	0.1437	0.0148	**N*N**	0.3491	0.0667	<0.0001

Mo*N	-0.0300	0.0412	0.4688	**Mo*N**	-0.0769	0.0699	0.2757	**Mo*N**	-0.0145	0.0324	0.6558
Mo*Mo	-0.0679	0.0305	0.0300	**Mo*Mo**	-0.0973	0.0517	0.0650	**Mo*Mo**	-0.0139	0.0240	0.5629
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	-6.6862	-0.1197	0.9928	**Eigenva**	13.2354	0.9949	-0.0999	**Eigenva**	12.5825	0.9995	-0.0312
	-14.2760	0.9928	0.1197		-9.9666	0.0999	0.9949		-1.4127	0.0312	0.9995

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

2. Indices NUE stems

Table 10. Response surface analysis for molybdenum sources in total biomass stems (TWs).

TWs											
Nano Mo				**Mo Chelate**				**Na Molybdate**			
CV	32.89	**R^2**	0.5219	**CV**	54.33	**R^2**	0.1124	**CV**	58.34	**R^2**	0.1545
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0010	N	Mo	L	0.7350	N	Mo	L	0.0558	N	Mo
C	<0.0001	0.0003	<0.0001	C	0.0673	0.0912	0.6508	C	0.1132	0.1104	0.2415
P	0.0072	L, C	L, C	P	0.3044			P	0.9512		
Model	<0.0001	µ 4.32		Model	0.2137	µ 2.44		Model	0.0759	µ 3.29	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	2.2763	0.5128	<0.0001	**Int**	2.0692	0.4791	<0.0001	**Int**	1.8780	0.6915	0.0106
N	0.4043	0.1519	0.0100	**N**	0.3085	0.1419	0.0338	**N**	0.4645	0.2048	0.0271
Mo	0.4436	0.0911	<0.0001	**Mo**	-0.0299	0.0851	0.7266	**Mo**	0.0414	0.1229	0.7371
N*N	-0.0406	0.0111	0.0006	**N*N**	-0.0234	0.0104	0.0279	**N*N**	-0.0318	0.0150	0.0385
Mo*N	0.0151	0.0054	0.0072	**Mo*N**	-0.0052	0.0050	0.3044	**Mo*N**	0.0004	0.0073	0.9512
Mo*Mo	-0.0207	0.0040	<0.0001	**Mo*Mo**	0.0028	0.0037	0.4532	**Mo*Mo**	0.0011	0.0054	0.8370
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	-1.2235	0.8828	0.4659	**Eigenva**	0.3048	-0.1356	0.9907	**Eigenva**	0.1120	0.0107	0.9999
	-2.3173	-0.4659	0.8828		-0.8675	0.9907	0.1356		-1.1465	0.9999	-0.0107

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 11. Response surface analysis for molybdenum sources in percentage by weight (*PxWs*).

PxW stems											
Nano Mo				**Mo Chelate**				**Na Molybdate**			
CV	18.70	**R²**	0.2977	**CV**	23.37	**R²**	0.0397	**CV**	26.32	**R²**	0.1064
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.1913	N	Mo	L	0.6679	N	Mo	L	0.5903	N	Mo
C	0.0003	0.0049	0.0088	C	0.4896	0.6688	0.8190	C	0.1591	0.1343	0.3760
P	0.1230			P	0.7106			P	0.1576		
Model	0.0008	μ 23.29		Model	0.7900	μ 23.88		Model	0.2445	μ 25.53	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	23.0404	1.5695	<0.0001	**Int**	22.7406	2.0108	<0.0001	**Int**	22.9347	2.4215	<0.0001
N	1.6305	0.4649	0.0009	**N**	0.6078	0.5956	0.3117	**N**	1.5549	0.7172	0.0343
Mo	-0.6720	0.2789	0.0192	**Mo**	-0.1346	0.3573	0.7076	**Mo**	0.1805	0.4303	0.6763
N*N	-0.1166	0.0341	0.0012	**N*N**	-0.0478	0.0437	0.2782	**N*N**	-0.1016	0.0526	0.0585
Mo*N	-0.0259	0.0165	0.1230	**Mo*N**	0.0079	0.0212	0.7106	**Mo*N**	-0.0366	0.0256	0.1576
Mo*Mo	0.0326	0.0122	0.0103	**Mo*Mo**	0.0078	0.0157	0.6205	**Mo*Mo**	-0.0050	0.0189	0.7921
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	3.3416	-0.1027	0.9947	**Eigenva**	0.8061	0.0936	0.9956	**Eigenva**	-0.1565	-0.2995	0.9540
	-4.2813	0.9947	0.1027		-1.7458	0.9956	-0.0936		-4.0045	0.9540	0.2995

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 12. Response surface analysis for molybdenum sources in milligrams of nitrogen (*mg-Ns*).

Mg-Ns											
Nano Mo				**Mo Chelate**				**Na Molybdate**			
CV	49.72	**R²**	0.2290	**CV**	49.13	**R²**	0.6245	**CV**	50.98	**R²**	0.5077
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0031	N	Mo	L	<0.0001	N	Mo	L	<0.0001	N	Mo
C	0.1207	0.0054	0.3616	C	<0.0001	<0.0001	0.4950	C	<0.0001	<0.0001	0.2253
P	0.7796			P	0.5885	L, C		P	0.0912	L, C	
Model	0.0086	μ 58.90		Model	<0.0001	μ 45.49		Model	<0.0001	μ 34.25	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	57.1635	10.5530	<0.0001	**Int**	6.9024	8.0528	0.3949	**Int**	5.6542	6.2913	0.3725
N	-2.8816	3.1258	0.3604	**N**	16.1892	2.3852	<0.0001	**N**	12.1614	1.8635	<0.0001
Mo	0.2958	1.8755	0.8752	**Mo**	-1.6246	1.4311	0.2610	**Mo**	-0.6784	1.1181	0.5464
N*N	0.4628	0.2295	0.0484	**N*N**	-0.8435	0.1751	<0.0001	**N*N**	-0.6786	0.1368	<0.0001
Mo*N	-0.0313	0.1115	0.7796	**Mo*N**	-0.0463	0.0851	0.5885	**Mo*N**	-0.1142	0.0665	0.0912
Mo*Mo	-0.0467	0.0826	0.5734	**Mo*Mo**	0.0746	0.0630	0.2412	**Mo*Mo**	0.0609	0.0492	0.2207
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	16.7033	0.9990	-0.0439	**Eigenva**	7.5172	-0.0366	0.9993	**Eigenva**	6.4784	-0.1102	0.9939

	-4.7194	0.0439	0.9903		-30.4184	0.9993	0.0.66		-24.8101	0.9939	0.1102

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 13. Response surface analysis for molybdenum sources in quantity of nitrogen in percentage (%) (*CaNt*).

CaNt											
Nano Mo				**Mo Chelate**				**Na Molybdate**			
CV	38.4410	**R^2**	0.3074	**CV**	39.71	**R^2**	0.5089	**CV**	47.28	**R^2**	0.1865
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0025	N	Mo	L	<0.0001	N	Mo	L	0.1112	N	Mo
C	0.0067	0.4381	0.0001	C	0.0003	<0.0001	0.8399	C	0.0657	0.0105	0.2686
P	0.2221		L, C	P	0.9584	L, C		P	0.0873	L	
Model	0.0006	μ 24.82		Model	<0.0001	μ 27.26		Model	0.0312	μ 26.24	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	32.4641	3.4383	<0.0001	**Int**	10.3098	3.9002	0.0105	**Int**	16.7517	4.4699	0.0004
N	-0.2903	1.0184	0.7766	**N**	6.4605	1.1552	<0.0001	**N**	4.0890	1.3240	0.0031
Mo	-2.1151	0.6110	0.0010	**Mo**	0.3357	0.6931	0.6299	**Mo**	-0.3192	0.7944	0.6892
N*N	0.0684	0.0747	0.3636	**N*N**	-0.3608	0.0848	<0.0001	**N*N**	-0.2111	0.0972	0.0339
Mo*N	-0.0448	0.0363	0.2221	**Mo*N**	-0.0021	0.0412	0.9584	**Mo*N**	-0.0822	0.0472	0.0873
Mo*Mo	0.0555	0.0269	0.0024	**Mo*Mo**	-0.0209	0.0305	0.4946	**Mo*Mo**	0.0348	0.0349	0.3237
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	8.8353	-0.2067	0.9783	**Eigenva**	-2.0983	-0.0059	0.9999	**Eigenva**	4.0072	-0.2078	0.9781
	2.1808	0.9783	0.2067		-12.9902	0.9999	0.0059		-8.1260	0.9781	0.2078

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 14. Response surface analysis for molybdenum sources in Nitrogen conduction (translocation) efficiency (NCoE).

NCoE											
Nano Mo				**Mo Chelate**				**Na Molybdate**			
CV	48.73	**R^2**	0.3913	**CV**	53.65	**R^2**	0.4790	**CV**	32.58	**R^2**	0.6053
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0763	N	Mo	L	<0.0001	N	Mo	L	<0.0001	N	Mo
C	<0.0001	<0.0001	0.1681	C	0.0114	<0.0001	0.0774	C	0.0011	<0.0001	0.4110
P	0.6493	L, C		P	0.0807	L, C		P	0.3397	L, C	
Model	<0.0001	μ 9.52		Model	<0.0001	μ 9.89		Model	<0.0001	μ 12.90	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**

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Int	4.8628	1.6716	0.0051	**Int**	13.0843	1.9130	<0.0001	**Int**	18.5041	1.5153	<0.0001
N	2.2341	0.4951	<0.0001	**N**	-2.0986	0.5666	0.0005	**N**	-2.4152	0.4488	<0.0001
Mo	0.6342	0.2970	0.0370	**Mo**	0.6932	0.3399	0.0460	**Mo**	0.4130	0.2693	0.1305
N*N	-0.1939	0.0363	<0.0001	**N*N**	0.1201	0.0416	0.0054	**N*N**	0.1244	0.0329	0.0004
Mo*N	-0.0080	0.0176	0.6493	**Mo*N**	-0.0359	0.0202	0.0807	**Mo*N**	-0.0154	0.0160	0.3397
Mo*Mo	-0.0234	0.0130	0.0779	**Mo*Mo**	-0.0172	0.0149	0.2548	**Mo*Mo**	-0.0129	0.0118	0.2794
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	-2.3366	-0.0521	0.9986	**Eigenva**	4.5129	0.9853	-0.1704	**Eigenva**	4.5162	0.9968	-0.0793
	-6.9958	0.9986	0.0521		-1.9091	0.1704	0.9853		-1.3321	0.0793	0.9968

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

3. Indices NUE in roots

Table 15. Response surface analysis for molybdenum sources in roots total biomass (TWr).

					TWr						
	Nano Mo				**Mo Chelate**				**Na Molybdate**		
CV	53.05	**R^2**	0.2497	**CV**	52.13	**R^2**	0.1875	**CV**	50.83	**R^2**	0.2145
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0213	N	Mo	L	0.1647	N	Mo	L	0.0149	N	Mo
C	0.4849	0.0013	0.0170	C	0.1229	0.0286	0.0379	C	0.0415	0.0525	0.0626
P	0.0030		C	P	0.0247			P	0.8199		
Model	0.0044	µ 1.31		Model	0.0303	µ 0.97		Model	0.0135	µ 1.28	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	1.8635	0.2505	<0.0001	**Int**	0.9673	0.1833	<0.0001	**Int**	0.8429	0.2351	0.0007
N	-0.1119	0.0742	0.1367	**N**	0.0546	0.0542	0.3185	**N**	0.1309	0.0696	0.0651
Mo	0.0027	0.0445	0.9503	**Mo**	-0.0305	0.0325	0.3527	**Mo**	0.0675	0.0417	0.1117
N*N	-0.0012	0.0054	0.8151	**N*N**	-0.0034	0.0039	0.3904	**N*N**	-0.0123	0.0051	0.0188
Mo*N	0.0082	0.0026	0.0030	**Mo*N**	-0.0044	0.0019	0.0247	**Mo*N**	-0.0005	0.0024	0.8199
Mo*Mo	-0.0023	0.0019	0.2398	**Mo*Mo**	0.0027	0.0014	0.0628	**Mo*Mo**	-0.0017	0.0018	0.3522
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	0.1238	0.8230	0.5679	**Eigenva**	0.3133	-0.2930	0.9561	**Eigenva**	-0.1716	-0.0622	0.9980
	-0.4028	-0.5679	0.8230		-0.1563	0.9561	0.2930		-0.4462	0.9980	0.0622

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Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 16. Response surface analysis for molybdenum sources in roots percentage by weight (*PxWr*).

PxWr											
Nano Mo				Mo Chelate				Na Molybdate			
CV	34.66	R^2	0.5800	CV	33.85	R^2	0.2623	CV	41.14	R^2	0.1983
Regression		Factors		Regression		Factors		Regression		Factors	
L	<0.0001	N	Mo	L	0.0765	N	Mo	L	0.0040	N	Mo
C	0.0002	<0.0001	0.0002	C	0.0156	0.0007	0.0745	C	0.3434	0.0105	0.5598
P	0.0009	L, C	L	P	0.0148	C		P	0.8618		
Model	<0.0001	μ 15.22		Model	0.0028	μ 18.74		Model	0.0221	μ 24.22	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	30.0313	1.9017	<0.0001	**Int**	21.1344	2.2864	<0.0001	**Int**	26.2301	3.5898	<0.0001
N	-3.6255	0.5632	<0.0001	**N**	-0.6371	0.6772	0.0188	**N**	-0.5323	1.0633	0.6685
Mo	-1.2118	0.3379	0.0007	**Mo**	0.0172	0.4063	0.9663	**Mo**	0.8915	0.6379	0.1676
N*N	0.1666	0.0413	0.0002	**N*N**	0.1415	0.0497	0.0061	**N*N**	-0.0310	0.0780	0.6920
Mo*N	0.0702	0.0201	0.0009	**Mo*N**	-0.0607	0.0241	0.0148	**Mo*N**	-0.0066	0.0379	0.8610
Mo*Mo	0.0284	0.0148	0.0611	**Mo*Mo**	0.0164	0.0179	0.3619	**Mo*Mo**	-0.0399	0.0281	0.1607
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	7.0550	0.8942	0.4474	**Eigenva**	5.8788	0.9185	-0.3952	**Eigenva**	-1.1053	0.9976	-0.0687
	1.7893	-0.4474	0.8942		0.8615	0.3952	0.9185		-4.0073	0.0687	0.9976

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 17. Response surface analysis for molybdenum sources milligrams of nitrogen (*mg-Nr*).

Mg-Nr											
Nano Mo				Mo Chelate				Na Molybdate			
CV	34.20	R^2	0.3670	CV	57.73	R^2	0.2665	CV	67.25	R^2	0.1294
Regression		Factors		Regression		Factors		Regression		Factors	
L	0.249	N	Mo	L	0.0048	N	Mo	L	0.2772	N	Mo
C	0.0010	0.0016	<0.0001	C	0.0770	0.0018	0.0552	C	0.0629	0.0475	0.9400
P	0.0022			P	0.0504			P	0.6595		
Model	<0.0001	μ 47.30		Model	0.0025	μ 21.91		Model	0.1433	μ 21.83	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	41.7864	5.8287	<0.0001	**Int**	25.6685	4.5584	<0.0001	**Int**	22.1820	5.2901	<0.0001
N	1.0738	1.7264	0.5364	**N**	1.0441	1.3502	0.4425	**N**	2.5045	1.5669	0.1154
Mo	2.6922	1.0358	0.0118	**Mo**	-1.0066	0.8101	0.2190	**Mo**	-0.4682	0.9401	0.6204
N*N	-0.2801	0.1267	0.0310	**N*N**	-0.1143	0.0991	0.2535	**N*N**	-0.2726	0.1150	0.0211
Mo*N	0.1973	0.0616	0.0022	**Mo*N**	-0.0963	0.0482	0.0504	**Mo*N**	0.0247	0.0559	0.6595
Mo*Mo	-0.1486	0.0456	0.0019	**Mo*Mo**	0.0716	0.0356	0.0494	**Mo*Mo**	0.0178	0.0414	0.6676

		Eigenvectors				Eigenvectors				Eigenvectors	
Eigenva	-6.0970	0.8288	0.5594	Eigenva	7.8617	-0.2345	0.9721	Eigenva	1.8350	0.0636	0.9979
	-18.8587	-0.5594	0.8288		-4.8130	0.9721	0.2345		-9.8644	0.9979	-0.0636

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 18. Response surface analysis for molybdenum sources in quantity of nitrogen in percentage (%) (*CaN*r).

CaNr											
Nano Mo				Mo Chelate				Na Molybdate			
CV	42.55	**R²**	0.3226	**CV**	58.50	**R²**	0.6381	**CV**	55.19	**R²**	0.5541
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0003	N	Mo	L	<0.0001	N	Mo	L	<0.0001	N	Mo
C	0.0802	<0.0001	0.2097	C	0.0004	<0.0001	0.0028	C	0.0033	<0.0001	0.6620
P	0.0498			P	0.0018	L, C		P	0.4572	L, C	
Model	0.0003	μ 21.84		Model	<0.0001	μ 19.42		Model	<0.0001	μ 21.66	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	26.5884	3.3492	<0.0001	**Int**	30.2598	4.0934	<0.0001	**Int**	42.5829	4.3087	<0.0001
N	0.2507	0.9920	0.8014	**N**	-6.0937	1.2124	<0.0001	**N**	-6.9212	1.2762	<0.0001
Mo	0.0636	0.5952	9151	**Mo**	1.0038	0.7274	0.1729	**Mo**	0.2026	0.7657	0.7923
N*N	-0.1598	0.0728	0.0322	**N*N**	0.3770	0.0890	<0.0001	**N*N**	0.3556	0.0937	0.0010
Mo*N	0.7097	0.0354	0.0498	**Mo*N**	-0.1420	0.0432	0.0018	**Mo*N**	0.0341	0.0455	0.4572
Mo*Mo	-0.0176	0.0262	0.5029	**Mo*Mo**	0.0082	0.0320	0.7985	**Mo*Mo**	-0.0251	0.0337	0.4584
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	-0.8445	0.3977	0.9174	**Eigenva**	14.8666	0.9569	-0.2902	**Eigenva**	11.7976	0.9974	0.0712
	-6.6791	0.9174	-0.3977		-0.4701	0.2902	0.9569		-2.5913	-0.0712	0.9974

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 19. Response surface analysis for molybdenum sources in nitrogen absorption efficiency (NAbE).

NAbE											
Nano Mo				Mo Chelate				Na Molybdate			
CV	48.05	**R²**	0.3405	**CV**	64.13	**R²**	0.5907	**CV**	48.76	**R²**	0.61.32
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	<0.0001	N	Mo	L	<0.0001	N	Mo	L	<0.0001	N	Mo
C	0.1249	<0.0001	0.2208	C	0.0001	<0.0001	0.0015	C	<0.0001	<0.0001	0.1619
P	0.0803	L, C		P	0.0012	L, C		P	0.5020	L, C	
Model	0.0002	μ 5.75		Model	<0.0001	μ 8.40		Model	<0.0001	μ 13.24	

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Source	Es	SE	Pr> t	Source	Es	SE	Pr> t	Source	Es	SE	Pr> t
Int	7.4187	0.9957	<0.0001	**Int**	12.0365	1.9422	<0.0001	**Int**	21.4271	2.3263	<0.0001
N	-0.0712	0.2949	0.8100	**N**	-2.8226	0.5752	<0.0001	**N**	-4.0693	0.6890	<0.0001
Mo	0.0913	0.1769	0.6078	**Mo**	0.6268	0.3451	0.0745	**Mo**	0.9057	0.4134	0.0325
N*N	-0.0913	0.0216	0.0915	**N*N**	0.1944	0.0422	<0.0001	**N*N**	0.2184	0.0505	<0.0001
Mo*N	0.0187	0.0105	0.0803	**Mo*N**	-0.0702	0.0205	0.0012	**Mo*N**	-0.0166	0.0246	0.5020
Mo*Mo	-0.0091	0.0077	0.2469	**Mo*Mo**	-0.0019	0.0152	0.8976	**Mo*Mo**	-0.0327	0.0182	0.0771
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	-0.5235	0.5683	0.8227	**Eigenva**	7.5713	0.9651	-0.2617	**Eigenva**	7.8851	0.9990	-0.0446
	-1.7260	0.8227	-0.5683		-0.7676	0.2617	0.9651		-3.3005	0.0446	0.9990

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

4. Indices NUE in fruits

Table 20. Response surface analysis for molybdenum sources in fruit total biomass (TWf).

TWf											
Nano Mo				**Mo Chelate**				**Na Molybdate**			
CV	53.18	**R²**	0.3502	**CV**	77.85	**R²**	0.0626	**CV**	73.36	**R²**	0.1122
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0318	N	Mo	L	0.6210	N	Mo	L	0.0597	N	Mo
C	0.0027	0.0170	<0.0001	C	0.3970	0.2932	0.7787	C	0.5073	0.4413	0.2131
P	0.0017	C	L, C	P	0.3137			P	0.8545		
Model	0.0001	μ 2.25		Model	0.5723	μ 1.22		Model	0.2151	μ 1.85	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	1.7572	0.4316	0.0001	**Int**	1.0515	0.3443	0.0034	**Int**	1.0729	0.4896	0.0325
N	-0.0750	0.1278	0.5596	**N**	0.1314	0.1020	0.2026	**N**	0.1869	0.1450	0.2025
Mo	0.2275	0.0767	0.0044	**Mo**	0.0052	0.0612	0.9318	**Mo**	0.0192	0.0870	0.8260
N*N	-0.0041	0.0093	0.6593	**N*N**	-0.0100	0.0074	0.1830	**N*N**	-0.0120	0.0106	0.2629
Mo*N	0.0150	0.0045	0.0017	**Mo*N**	-0.0037	0.0036	0.3137	**Mo*N**	0.0009	0.0051	0.8545
Mo*Mo	-0.0121	0.0033	0.0007	**Mo*Mo**	0.0006	0.0026	0.8061	**Mo*Mo**	0.0011	0.0038	0.7586
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	0.0151	0.9390	0.3438	**Eigenva**	0.0934	-0.2361	0.9717	**Eigenva**	0.1198	0.0516	0.9986
	-1.3799	-0.3438	0.9390		-0.3903	0.9717	0.2361		-0.4348	0.9986	-0.0516

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Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 21. Response surface analysis for molybdenum sources in fruit percentage by weight (*PxWf*).

PxWf											
Nano Mo				**Mo Chelate**				**Na Molybdate**			
CV	83.20	**R^2**	0.4022	**CV**	173.38	**R^2**	0.0427	**CV**	146.44	**R^2**	0.1036
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0003	N	Mo	L	0.9317	N	Mo	L	0.0996	N	Mo
C	0.0007	0.0601	<0.0001	C	0.4145	0.7616	0.5593	C	0.4041	0.2497	0.4749
P	0.0685		L	P	0.4202			P	0.8116		
Model	<0.0001	μ 8.97		Model	0.7617	μ 3.30		Model	0.2604	μ 6.72	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	2.9054	2.6900	0.2846	**Int**	3.0906	2.1389	0.1538	**Int**	4.0041	3.5475	0.2637
N	-1.2707	0.7967	0.1162	**N**	0.1836	0.6335	0.7729	**N**	0.6826	1.0508	0.5185
Mo	1.8849	0.4780	0.0002	**Mo**	0.2771	0.3801	0.4688	**Mo**	-0.6000	0.6304	0.3452
N*N	0.0893	0.0585	0.1323	**N*N**	-0.0310	0.0465	0.5072	**N*N**	-0.0030	0.0771	0.9686
Mo*N	0.0528	0.0284	0.0685	**Mo*N**	0.0183	0.0226	0.4202	**Mo*N**	-0.0089	0.0375	0.8116
Mo*Mo	-0.0792	0.0210	0.0004	**Mo*Mo**	-0.0194	0.0167	0.2512	**Mo*Mo**	0.0376	0.0277	0.1803
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	3.4360	0.9904	0.1381	**Eigenva**	-0.8415	0.8940	0.4480	**Eigenva**	3.7852	-0.0690	0.9976
	-8.1434	-0.1381	0.9904		-2.2169	-0.4480	0.8940		-0.1283	0.9976	0.0690

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 22. Response surface analysis for molybdenum sources in milligrams of nitrogen (*mg-N fruits*).

mg-Nf											
Nano Mo				**Mo Chelate**				**Na Molybdate**			
CV	104.01	**R^2**	0.4010	**CV**	262.54	**R^2**	0.0858	**CV**	187.32	**R^2**	0.1506
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0001	N	Mo	L	0.7854	N	Mo	L	0.0203	N	Mo
C	0.0044	0.0031	<0.0001	C	0.0946	0.2620	0.7100	C	0.4633	0.0694	0.3698
P	0.0201		L, C	P	0.8356			P	0.5393		
Model	<0.0001	μ 28.78		Model	0.3765	μ 11.45		Model	0.0840	μ 25.49	

Source	Es	SE	Pr> t	Source	Es	SE	Pr> t	Source	Es	SE	Pr> t
Int	-3.3947	10.7834	0.7540	**Int**	-3.0095	10.8367	0.7822	**Int**	12.1972	17.2054	0.4812
N	1.1996	3.1940	0.7086	**N**	5.9183	3.2098	0.0703	**N**	2.6891	5.0962	0.5997
Mo	6.2976	1.9164	0.0017	**Mo**	1.8545	1.9259	0.3396	**Mo**	-3.1664	3.0577	0.3047
N*N	-0.0768	0.2345	0.7444	**N*N**	-0.4459	0.2356	0.0634	**N*N**	-0.0073	0.3741	0.9843
Mo*N	0.2725	0.1140	0.0201	**Mo*N**	0.0238	0.1145	0.8356	**Mo*N**	0.1123	0.1819	0.5393
Mo*Mo	-0.2904	0.0844	0.0011	**Mo*Mo**	-0.0979	0.0848	0.2530	**Mo*Mo**	0.1681	0.1347	0.2168
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	-0.4291	0.9615	0.2747	**Eigenva**	-9.7143	0.1123	0.9936	**Eigenva**	17.4602	0.1867	0.9823
	-31.3782	-0.2747	0.9615		-16.1360	0.9936	-0.1123		-0.9067	0.9823	-0.1867

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 23. Response surface analysis for molybdenum sources in quantity of nitrogen in percentage (%) (*CaNf*).

				CaNf							
	Nano Mo				**Mo Chelate**				**Na Molybdate**		
CV	87.20	**R²**	0.3944	**CV**	175.90	**R²**	0.0736	**CV**	128.72	**R²**	0.0816
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0005	N	Mo	L	0.9216	N	Mo	L	0.2420	N	Mo
C	0.0007	0.0469	<0.0001	C	0.1186	0.5937	0.4435	C	0.3627	0.7404	0.2640
P	0.0478		L	P	0.8801			P	0.6717		
Model	<0.0001	μ 10.73		Model	0.4733	μ 5.25		Model	0.4082	μ 12.00	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	-0.2437	3.3732	0.9426	**Int**	1.9793	3.3297	0.5545	**Int**	8.1903	5.5660	0.1466
N	0.8878	0.9991	0.3779	**N**	1.0432	0.9862	0.2946	**N**	0.8851	1.6486	0.5934
Mo	2.3455	0.5995	0.0002	**Mo**	0.8769	0.5917	0.1438	**Mo**	-0.8027	0.9892	0.4204
N*N	-0.0853	0.0733	0.2494	**N*N**	-0.0956	0.0724	0.1916	**N*N**	-0.0176	0.1210	0.8847
Mo*N	0.0721	0.0359	0.0478	**Mo*N**	0.0053	0.0352	0.8801	**Mo*N**	-0.0250	0.0588	0.6717
Mo*Mo	-0.1027	0.0264	0.0003	**Mo*Mo**	-0.0426	0.0260	0.1071	**Mo*Mo**	0.0622	0.0435	0.1583
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	-2.4728	0.9636	0.2672	**Eigenva**	-3.4141	0.9828	0.1844	**Eigenva**	6.3099	-0.1076	0.9941
	-10.8720	-0.2672	0.9636		-4.2965	-0.1844	0.9828		-0.7161	0.9941	0.1076

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

Table 24. Response surface analysis for molybdenum sources in fruit productivity (Productivity).

Productivity											
Nano Mo				Mo Chelate				Na Molybdate			
CV	65.34	**R^2**	0.3846	**CV**	185.92	**R^2**	0.2411	**CV**	74.33	**R^2**	0.2216
Regression		**Factors**		**Regression**		**Factors**		**Regression**		**Factors**	
L	0.0002	N	Mo	L	0.0.0066	N	Mo	L	0.0067	N	Mo
C	0.0006	0.0303	<0.0001	C	0.0340	0.0031	0.3667	C	0.2424	0.0333	0.0270
P	0.6964		L, C	P	0.6018	L		P	0.1075		
Model	<0.0001	μ 0.05		Model	0.0058	μ 0.02		Model	0.0108	μ 0.0407	
Source	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**	**Source**	**Es**	**SE**	**Pr> t**
Int	0.0162	0.0120	0.1827	**Int**	0.0481	0.0188	0.0131	**Int**	0.0354	0.0109	0.0019
N	0.0056	0.0035	0.1211	**N**	-0.0146	0.0055	0.0108	**N**	0.0021	0.0032	0.5079
Mo	0.0084	0.0021	0.0002	**Mo**	0.0060	0.0033	0.0778	**Mo**	0.0004	0.0019	0.8171
N*N	-0.0006	0.0002	0.0192	**N*N**	0.0008	0.0004	0.0385	**N*N**	-0.0002	0.0002	0.3865
Mo*N	0.0001	0.0001	0.6964	**Mo*N**	-0.0001	0.0002	0.6018	**Mo*N**	-0.0001	0.0001	0.1075
Mo*Mo	-0.0003	0.0001	0.0015	**Mo*Mo**	-0.0002	0.0001	0.1063	**Mo*Mo**	0.0001	0.0001	0.1486
		Eigenvectors				**Eigenvectors**				**Eigenvectors**	
Eigenva	-0.0224	0.9860	0.1662	**Eigenva**	0.0313	0.9984	-0.0563	**Eigenva**	0.0140	-0.2550	0.9669
	-0.0315	-0.1662	0.9860		-0.0243	0.0563	0.9984		-0.0089	0.9669	0.2550

Non-significant probability Pr > 0.05, significant 0.05 ≤Pr ≤0.01, highly significant Pr <0.0001; CV coefficient of variation, R^2 regression coefficient; Regression Analysis: Linear L, quadratic C, P N*Mo interaction; SE standard error; Int intercept; Es estimation; Eigenva: Eigenvalues; Eigenvectors.

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