

Yield of corn hybrids in response to foliar fertilization with biostimulants

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Abstract

The use of biostimulants has been an agricultural strategy for increasing crop yields and quality. The objective of this work was to evaluate the effect of biostimulants on the yield and components of corn hybrids (*Zea mays* L.) in the High Valleys of the State of Mexico. The sowing was carried out during the spring summer 2017 cycle in three environments (Jocotitlán, Temascalcingo, Jilotepec). Eleven corn hybrids were evaluated (TSIRI PUMA; ATZIRI PUMA; TLAOLI PUMA; IXIM PUMA, H-50, #46#48; H-66; H-76; H-77; H-47AE and H-49AE). The foliar treatments with biostimulants were the following: B1= control; B2=Eurobor; B3= Eurologo; B4= Eurodual; B5= Euroalg. Evaluations included: grain yield, straw production, rows per cob, grains per row, grains per cob, volumetric weight, weight of 200 grains, and cob diameter. For all variables, a combined analysis of variance, a mean comparison test (Tukey) and a correlation analysis (Pearson) were performed. Between environments, hybrids and biostimulants there were significant differences. In Temascalcingo, higher grain yield was observed with 13.5 t ha⁻¹. The hybrids had yields higher than those of studies reported in the literature, and the genotypes H-66, H-50 and H-76 stand out in the present study. Biostimulants increased grain yield from 7.9 to 11.4%, with respect to the control, and positively affected the agronomic components of the hybrids evaluated. Biostimulants are an alternative in complementary fertilization to increase production in the cultivation of corn.

Keywords: *Zea Mays* L, high productivity, mineral nutrition, sustainable agriculture.

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Introduction

Corn (*Zea mays* L.) in the High Valleys of Mexico (2 200 to 2 600 masl) is produced by small and medium producers who do not have economic resources and adequate technologies in the conduct of their cultivation, this limit reaching maximum potentials of grain yields. Although there are already improved materials with yield stability (Vázquez *et al.*, 2020), the lack of agricultural innovations in the agronomic management of corn contributes to its low productivity, in addition, adverse edaphoclimatic conditions in contrasting environments significantly reduce production (Turrent *et al.*, 2016).

The above, it will be necessary to consider actions of adjustments in the technological packages in the agronomic management and plant breeding programs that allow the producer to select their seed and adopt the best agricultural practices such as the efficient management of chemical and organic fertilization, finer practices of foliar nutrition to the foliage (Fernández *et al.*, 2015; Zamudio *et al.*, 2018) and other technological innovations that provide better answers in the agronomic components of the crop.

In recent years, the use of biostimulants has been a strategy within the fertilization program as a complement to fertilization applied to the soil (Zamudio *et al.*, 2018). Studies on the effects of biostimulants have been intensified in different crops (Du Jardin, 2015), although most of the research is reported in horticultural crops and under greenhouse conditions (Grabowska *et al.*, 2012; Mattner *et al.*, 2013; Petrozza *et al.*, 2013), with few studies associated with corn (Quezada *et al.*, 2015; Tejada *et al.*, 2018; Zamudio *et al.*, 2018).

According to Calvo *et al.* (2014), biostimulants can be synthetic or natural and are composed of substances such as plant hormones, macro and micronutrients, amino acids, proteins and microorganisms, so they can be defined by their composition and mode of action, by their impact on the plant (Yakhin *et al.*, 2017), by the functions they exert on plants, or by the final response on crop yield (Du Jardin, 2015). These compounds affect the physiology of the plant when applied in small amounts. Du Jardin (2015) classified biostimulants into three main groups: algae extracts, hydrolyzed proteins (peptides and free amino acids (FAs)) and humic substances (humic and fulvic acids).

According to Battacharyya *et al.* (2015), algae extracts are a mixture of organic and inorganic compounds from seaweed biomass that contain, in the source material, carbohydrates (mannitol), minerals, osmolytes (betaines), secondary metabolites (eg. phenols), amino acids, vitamins and plant growth hormones. Algae extracts have high resistance to osmotic stress, reduction in protein degradation, preventing oxidation of chloroplasts and a delay in foliar senescence that prolongs the photosynthetic activity of the plant (Jannin *et al.*, 2013). In addition, by functioning as metal ion chelates, they improve the absorption of nutrients when applied under suboptimal growing conditions or under environmental stress (Crouch and Van Staden, 1994).

The industrial synthesis of amino acids includes chemical or enzymatic hydrolysis, or both, of agro-industrial by-products of animal or vegetable origin and biomass of specific crops (Cavani *et al.*, 2006). According to Rai (2002), physiological processes in plants are regulated by short-chain free

amino acids. Amino acids can act as anti-stress agents (Mladenova *et al.*, 1998), source of nitrogen and hormone precursors (Rai, 2002; Zhao, 2010; Maeda and Dudareva, 2012), as additives with insecticides and fungicides (Cavani *et al.*, 2006) and as chelating agents (Ashmead, 1986). The application of any nutrient together with amino acids is most effective during the process of its assimilation and incorporation into the vegetable tissues of the plant. Nutrients chelated with amino acids form very small, electrically neutral molecules that accelerate their absorption and transport within the plant (Ashmead, 1986).

The absorption of amino acids by plants has an energy advantage by avoiding the processes of chemical transformation of nutrients in their different forms of assimilation. Amino acids are absorbed and assimilated by the plant, quickly incorporated into plant metabolism, as if they were synthesized by them without energy expenditure (Jones and Kielland, 2002). In this sense, it improves crop nutrition, especially in the critical stages of plant development, exhibits higher yield and improves the quality of the grain or fruit harvested (Parrado *et al.*, 2008; Tejada *et al.*, 2016). In the application of biostimulants, it is expected that the plants show optimal development and present better characteristics and grain yield at the end of the production cycle.

Although in other countries research work is carried out in corn to observe the yield response as a function of the application of biostimulants (Quezada *et al.*, 2005; Gazola *et al.*, 2014; Galindo *et al.*, 2015), in Mexico there is no information regarding the foliar fertilization of biostimulants (amino acids and seaweed extracts) for the cultivation of corn that verifies its efficiency in the agronomic characteristics of the crop. In this context, the hypothesis was that the foliar application of biostimulants will differentially impact the genetic response of the corn seed in a higher grain yield and better responses in agronomic characteristics by favoring the synthesis and assimilation of photosynthates. The objective of this research was to determine the effect of foliar fertilization of biostimulants based on amino acids and seaweed extracts on the yield and agronomic characteristics of corn hybrids in the High Valleys of the State of Mexico.

Materials and methods

The experiments were established in three environments of the High Valleys of the State of Mexico in the spring-summer (SS) 2017 agricultural cycle. The first experiment was carried out in Jocotitlán (JOC) (19° 43' north latitude, 99° 51' west longitude and 2 700 masl), with average annual rainfall of 669.0 mm and the temperature ranges between 3 and 24 °C and the sowing was carried out on April 10, the second experiment was located in Temascalcingo (TEM) (19° 55' north latitude, 100° 00' west longitude, 2350 masl), with average annual rainfall of 874.6 mm and 10.6 and 23 °C. The sowing was done on April 22 and the third experiment corresponded to Jilotepec (JIL) (19° 59' north latitude, 99° 30' west longitude and 2 500 masl) with average annual rainfall of 850 mm and the temperature varies from 8 °C to 24 °C, the sowing was carried out on May 20, 2017. In the three experimental areas, the soils are classified as Phaeozem dark color phase (haplic Phaeozem (Jocotitlán), luvisc Phaeozem (Temascalcingo) and gleyic Phaeozem (Jilotepec)), of loam-clay texture (30, 38, 32% of clay, silt and sand respectively) (Sotelo *et al.*, 2011). A soil analysis was performed in all three environments to determine its chemical characteristics (Table 1).

Table 1. Chemical characteristics of the soil in three production environments for the cultivation of corn, at depths from 0 to 20 cm. Agricultural cycle, spring-summer, 2017.

Environment	pH	MO	N-NO ₃	P ¹	K	S	Ca	Mg	Al ³⁺	H+Al	CEC	V ²	m ³
	Agua	(%)	(mg dm ⁻³)				(cmol _c dm ⁻³)				(%)		
Jocotitlán	5.6	2.1	11.9	46.8	205	5.6	5.25	2.32	0.2	0.8	10.1	80.9	1.8
Temascalcingo	7	1.9	15.9	72.2	350	2.8	6.64	3.24	0	0	11	95	0
Jilotepec	6.8	2.8	28	137	420	7	14.8	6.52	0	0	22.7	98	0

¹= method of Bray 1 (Bray and Kurtz, 1945). ²= base saturation. ³= aluminum saturation. CEC= cation exchange capacity; micronutrients (mg dm⁻³), Jocotitlán: B= 0.18; Cu= 1.01; Fe= 96.1; Mn= 35.8; Zn= 0.57; Temascalcingo: B= 0.65; Cu= 1.01; Fe= 46.4; Mn= 39.7; Zn= 2.57; Jilotepec: B= 0.28; Cu= 2.74; Fe= 45.6; Mn= 35.3; Zn= 2.28.

Eleven white grain hybrids were evaluated, four released by UNAM (TSIRI PUMA; ATZIRI PUMA; TLAOLI PUMA; IXIM PUMA) and seven released by the National Institute for Forestry, Agricultural and Livestock Research (INIFAP, for its acronym in Spanish) (H-50, #46#48; H-66; H-76; H-77; H-47AE; H-49AE), the latter two and the four of UNAM are trilinear hybrids generated with 'androsterile' progenitors. The sowing density was 85 000 plants ha⁻¹, each experimental plot had four furrows of 0.8 m by 8 m in length and the two central lines (4.8 m²) were considered as useful plot.

Prior to sowing, the seeds were treated with Crusier[®] (thiamethoxam 50 ml ha⁻¹) and Force[®] insecticide (10 tefluthrin 15 kg ha⁻¹) for soil pest control. The fertilization dose was 250-60-60 kg ha⁻¹ of N-P-K, and the application was carried out in three moments. At the sowing, 60 kg ha⁻¹ of N was applied, 100% of phosphorus (P₂O₅) and potassium (K₂O) at a rate of 60 kg ha⁻¹ for both cases, based on urea, diammonium phosphate and potassium chloride, respectively. The second and third applications used urea with 120 kg ha⁻¹ of N between V₄-V₆ and 70 kg ha⁻¹ of N between V₁₀-V₁₂, respectively.

The weed was controlled in the initial vegetative stage (V₂-V₄) mechanically with hoeing and subsequently Lumax[®] herbicide (S-metolachlor, atrazine and mesotrione 4 L ha⁻¹) was applied. For the control of foliage insects, Karate Zeon[®] (Lambda cyhalothrin 250 ml ha⁻¹) and Denim[®] (emamectin benzoate 100 ml ha⁻¹) were applied. For the prevention of diseases, Priori[®] Xtra (azoxystrobin, cyproconazole 350 ml ha⁻¹) was applied. The biostimulants were applied foliarly with the use of a backpack sprayer in two phenological stages of the crop, between V₅-V₆ and V₈-V₁₀, respectively. Foliar applications were made in the early hours of the morning (07:00 to 09:00 am). The chemical composition of the biostimulants (amino acids and seaweed extracts) of each foliar treatment are shown in Table 2.

Once physiological maturity was reached (R₆), the harvest was carried out to quantify the yield (YIE) (t ha⁻¹) with the dry weight of the grain, adjusted to 14% moisture and extrapolated per hectare (Tadeo *et al.*, 2015). In addition, the following agronomic components were recorded: straw production (SP) (t ha⁻¹), weight of 200 g (W200) (g), rows per cob (RC), grains per row (GR), number grains per cob (NGC), cob diameter (mm) (CD) and volumetric grain weight (VW) (kg hl⁻¹).

Table 2. Chemical composition of biostimulants and doses applied in corn plants in three environments of the State of Mexico.

Bioestimulants	Chemical composition	Dose (kg ha ⁻¹)
Control (B1)	Only water (test)	0
Eurobor (B2)	5% organic N (ON), 25% free amino acids (FAs) and 3% boron;	2.5
Eurologo (B3)	5% ON, 25% FAs and M (Zn 1%, Mn 0.5%, Fe 0.5%, B 0.1%, Mo 0.01%);	2.5
Eurodual (B4)	3% ON, 10% organic carbon (OC), 15% FAs, 8% CaO and 2% MgO;	2.5
Euroalg (B5)	10% OC, 1% ON, pH 8.9, organic substance with molecular weight <50 kDa (KiloDaltons) 30%, concentrated extract of alga <i>Ascophillum nodosum</i> and yeast extracts (dry substance: 37%), macronutrients and micronutrients.	3

The experimental design was completely randomized blocks with four repetitions, in a 3×5×11 factorial arrangement and environments (E), biostimulants (B), hybrids (H) and their interactions, as sources of variation. To determine the simple effects and interactions of the treatment design in relation to the response variables, a combined analysis of variance was performed with the SAS statistical program version 9.4 (SAS, 2002). The comparison of means was with the Tukey test ($p \leq 0.05$) and a correlation analysis (Pearson).

Results and discussion

Differences in grain yield and yield components due to the environment (E) were significant ($p \leq 0.01$), except for RC and CD. Among the hybrids (H), there was a highly significant difference ($p \leq 0.01$) in all variables. In the biostimulant (B) factor, two groups of significance were identified in YIE, VW, NGC and W200 ($p \leq 0.01$) and in RC and GR ($p \leq 0.05$). In the interaction E*H, it had a significant effect on all variables ($p \leq 0.01$); in the interaction E*B, in YIE, SP, RC, GR, NGC and W200 there was significance ($p \leq 0.05$); in the interaction H*B in YIE, VW, SP, NGC and W200, and in the interaction E*H*B in YIE, SP, W200 and RC there was high significance ($p \leq 0.01$) (Table 3).

In the interactions (E*H, E*B, H*B and E*H*B), there were significant differences for grain yield, which means that the hybrids evaluated present differential behavior in yield for both the environment factor and the biostimulants, and at least three biostimulants (B2, B3 and B4) had positive response in the three environments (Table 3). As for the coefficient of variation, it was 6.4% for grain yield, which guarantees the reliability of the statistical results.

The final average yield was 12.3 t ha⁻¹, higher than the region's mean of 6 t ha⁻¹ (Tadeo *et al.*, 2015). The yields on average by environments, the highest production with 13.5 t ha⁻¹ corresponded to Temascalcingo, in contrast, in Jilotepec, it presented lower yield with 10.2 t ha⁻¹. The grain yield in Jilotepec decreased 24.5% (3.3 t ha⁻¹) with respect to Temascalcingo, due to the delay in the sowing, which led to fewer days to complete the cycle and probably affected the grain yield due to the unfavorable weather conditions, such as poor distribution of rains, low temperatures and frosts.

Table 3. Combined analysis of variance for grain yield and yield components of eleven white-grain corn hybrids as a function of foliar fertilization with biostimulants in three environments of the High Valleys of the State of Mexico. Agricultural cycle, spring-summer, 2017.

SV	DF	YIE (t ha ⁻¹)	VW (kg hl ⁻¹)	SP (t ha ⁻¹)	RC (num.)	GR (num.)	NGC (num.)	W200 (g)	CD (mm)
E	2	686.1**	505**	995.4**	0.47 ns	554.1**	134278.2**	1117.3**	14.9 ns
Bloc (E)	9	0.56 ns	1 ns	8.6 ns	0.61 ns	13.9 ns	5934.4 ns	2**	5.9 ns
H	10	11.6**	46.84**	19.8**	23.3**	49.6**	23521.4**	351.2**	52.7**
B	4	34.2**	10.6**	12.6 ns	7*	43.5*	33532.9**	77.5**	16.9**
E×H	20	19.9**	34.1**	45.9**	10**	48.5**	21652.9**	198.6**	47.3**
E×B	8	9.1*	0.33 ns	20.9*	4.65*	18.9*	9854.5*	53*	4.2 ns
H×B	40	1.9**	1.84**	16.7**	2.2 ns	12.9 ns	6475.5*	18.6**	7.2 ns
E×H×B	80	2**	1.12 ns	10.9*	3.05**	12.2 ns	5568.1 ns	18.1**	6.4 ns
Error	486	0.6	0.92	7.74	1.8	10.3	4550.5	7.3	6.4
Total	659	-	-	-	-	-	-	-	-
CV (%)	-	6.4	1.2	18.5	8.4	10.5	13.5	4.1	5.2
Mean	-	12.3	80.2	15.08	16.4	30.5	499.5	65.8	48.3

Ns= no significant; * = $p \leq 0.05$; ** = $p \leq 0.01$. Bloc= block; E= environment; H= hybrid; B= biostimulant; SV=source of variation; CV= coefficient of variation; DF= degrees of freedom; YIE= grain yield; VW= volumetric weight; SP= straw production; RC= rows per cob; GR= grains per row; NGC= number of grains per cob; W200= weight of 200 grains; CD= cob diameter.

In Jocotitlán and Temascalcingo, a difference of 0.5 t ha⁻¹ was observed, which may be influenced by the date of sowing close to each other. Tadeo *et al.* (2015) evaluated four corn genotypes on different sowing dates (May 17; June 1) in Cuautitlán Izcalli (2 240 masl) and verified that the sowing carried out on May 17 had a higher grain yield in relation to the second sowing. Therefore, as the sowing of corn in the High Valleys is delayed, it is affected by the irregular distribution of rainfall, reduction of solar radiation and low temperatures (frosts) during the cycle, thus reducing the productive potential of the crop.

In relation to hybrids, they presented a highly significant difference in all the agronomic variables evaluated (Table 4), which indicates that the genotypes used present a great phenotypic diversity. The yield of the hybrids ranged from 11.5 (Tsiri Puma) to 13 (H-66) t ha⁻¹, with an average yield of 12.3 t ha⁻¹ of grain. The hybrids H-66, H-50 and H-76 stand out with average yield of 13, 12.6 and 12.5 t ha⁻¹, respectively. The hybrids Tsiri Puma (11.5 t ha⁻¹) and Atziri Puma (11.7 t ha⁻¹) had relatively low yields in this study (Table 4).

Table 4. Comparison of means of grain yield (t ha⁻¹) of eleven corn hybrids as a function of foliar treatments with biostimulants. Average of three environments of the High Valleys of Mexico. Agricultural cycle, spring-summer, 2017.

Hybrid	Foliar biostimulants				
	B1	B2	B3	B4	B5
Tsiri Puma	10.9 bcdB	12 bA	11.9 cA	11.7 cAB	10.9 dB
Atziri Puma	10.9 bcdB	11.9 abA	12.2 bcA	12 bcA	11.7 cdAB
Tlaoli Puma	11.5 abcC	12.8 abA	11.9 cBC	12.6 abcAB	11.9 bcdABC
Ixim Puma	11.6 abcC	11.9 bBC	13.1 abA	12.6 abcAB	12.7 bcAB
H-47 AE	11.2 abcdB	13 aA	13.2 abA	12.9 abA	11.9 bcdB
H-49 AE	10.7 dB	12.4 abA	12.7 abcA	11.8 cA	12.3 bcA
H-50	11.2 abcdC	12.5 abB	13 abAB	13.4 aA	12.9 abAB
H-66	12 aC	12.9 aB	13.2 abAB	13 abB	13.9 aA
H-76	11.9 abC	12.4 abABC	13.3 aA	12.2 bcBC	12.9 abAB
H-77	11.5 abcB	12 abAB	12.6 abcA	12.2 bcAB	12 bcAB
#46#48	11.9 abB	13 aA	12.9 abcA	13 abA	12 bcB
M ^f B	11.4 C	12.4 B	12.7 A	12.5 B	12.3 B
CV (%)	6.4				

Means with the same lowercase letter in the column (hybrids) and uppercase letter in the row (biostimulants) are statistically equal (Tukey; $p \leq 0.05$). CV= coefficient of variation; M^f= means; B= biostimulants; B1= control; B2= Eurobor; B3= Eurologo; B4= Eurodual; B5= Euroalg.

The results of this study exceed the average yield of the region and those reported in other studies (Martínez *et al.*, 2018; Tadeo *et al.*, 2020; Vázquez *et al.*, 2020), in which yields were relatively low for these same hybrids. This was due to foliar fertilization with biostimulants that directly causes a significant increase in grain yield (Quezada *et al.*, 2015; Tejeda *et al.*, 2018; Zamudio *et al.*, 2018). However, the continuous study of the foliar fertilization of biostimulants in the cultivation of corn is still pertinent in order to know their mechanisms of action and the morphological and physiological response of the plants in the different hybrids and depending on the contrasting environments.

Regarding biostimulants, in the localities of Temascalcingo and Jilotepec, the best responses were observed with the foliar application of B2 and B3. In both localities, they present slightly alkaline soils with $\text{pH} \geq 6.8$ (Table 1). In contrast, in Jocotitlán, it is characterized by soils of pH of 5.6, (Table 1), it allowed foliar fertilization with biostimulant (B4) to exceed the other foliar treatments, which contains in its composition 8 and 2% of % of CaO and MgO, respectively (Table 2). In these soils that have characteristics such as low or high pH, presence of scarcely available minerals, plants lack optimal levels of essential nutrients, particularly micronutrients, which could be provided by foliar fertilization.

Absorption by aerial parts is the only practical means of supplying specific nutrients. The availability of micronutrients in the soil is closely related to their solubility, since the leaf and root tissue have the same morphological structure (they originate in the meristem tissue), plants

can quickly absorb dissolved minerals (Martinka *et al.*, 2014). Therefore, micronutrient deficiency in soils can be successfully managed by foliar fertilization (Barbosa *et al.*, 2013; Tejada *et al.*, 2018). The effects of foliar biostimulants were favored during application in all three environments by low temperatures, higher relative humidity, the combination of both factors implies a lower vapor pressure deficit (Fernández *et al.*, 2015) and at the same time is associated with stomatal opening, which allowed greater efficiency in the absorption of foliar biostimulants.

In corn components, biostimulants produced statistically different effects, except for straw production (Table 4). Regarding the control (B1), for each biostimulant, an increase in corn grain yield of 11.4, 9.6, 8.8 and 7.9% was detected for B3, B4, B2, B5, respectively (Table 4). In a study of foliar fertilization with biostimulant based on free amino acids from chicken epithelial tissue, on average of two agricultural cycles, Tejada *et al.* (2018) observed a 14% increase in corn grain yield. Ahmad *et al.* (2007), in a field study with compost enriched with N and tryptophan to the soil, observed a 21% increase in corn grain yield.

In contrast, Quezada *et al.* (2015) did not observe an increased in grain yield in soil fertilization of tryptophan and lysine biosynthesis by-product, but they did detect a significant difference for harvest index (HI) in relation to the control, so they suggest that there is potential in amino acids as sources of N in complement to nitrogen fertilization (nitrate and ammonium sulfate) to increase grain yields.

On average of the eleven hybrids, the best response was observed in B3 with an 11.4% increase in yield, which represents 1.3 t ha^{-1} of corn grain (Table 4) compared to B1, followed by B4, B2 and B5. B3, in addition to free amino acids, contains micronutrients (in percentage Zn 1, Mn 0.5, Fe 0.5, B 0.1, Mo 0.01) of easy assimilation for plants due to the chelating effect of amino acids on micronutrients (Cu, Mn, Zn and B) within the plant, which facilitates their absorption and transport when applied together, also positively affect the permeability of the cell membrane (Ibrahim *et al.*, 2010), they are also particularly involved in the reproductive phase of the plant and therefore, in the determination of the yield and quality of the harvested crop (Parrado *et al.*, 2008; Tejada *et al.*, 2018).

Amino acid-based biostimulants combined with essential micronutrients such as Zn and B represent an alternative for the soils of the High Valleys due to the low concentrations of these micronutrients for these agroecosystems (Table 1). It is observed that, when applying B2, which contains 3% boron, it exceeds the control (B1), with 8.8% grain yield that represents 1.1 t ha^{-1} of corn grain production. The presence of these nutrients in biostimulants allows correcting their deficiency in soils, optimizing vegetative and reproductive activities to plants. Micronutrients once assimilated by plants and incorporated into plant tissues are effectively transported by amino acid complexes, and affect grain yield (Ashmead, 1986).

B5, which has in its composition seaweed extracts, presented an increase of 7.9% in grain yield with respect to the control and presented a similar response with B2 and B4 of this study. Although the mechanism of action of seaweed extract to improve agricultural crop productivity is unknown (Mohanty *et al.*, 2013), positive responses in grain yield have been observed with foliar fertilization

of biostimulant based on seaweed extracts as a complement to soil fertilization. Galindo *et al.* (2015) observed an increase in the yield of corn grain with *Egeria densa* extract, compared to the control, of 8 and 6.2%, in a single application (VT) and in two applications (VT and R2), respectively. Kumar and Sahoo (2011) reported higher yield in *Triticum aestivum* in the foliar application of 20% *Sargassum wightii* extract, with a 13.69% increase in seed number and a 22.86% increase in the dry weight of the seeds compared to the control.

In this study, B5, in addition to its composition (10% organic carbon and 1% organic nitrogen), contains in the source materials a source rich in minerals, vitamins, carbohydrates and growth promoters, which would be an alternative for use as an organic foliar fertilizer since it impacts on grain yield (Table 4). The agronomic variables evaluated in this study presented significant differences for the biostimulant factor, except for straw production, which did not vary between treatments (Table 5). This is attributed to the high percentage of CV measured for straw production (18.5%), compared to the other variables that ranged from 1.2 to 13.5% (Table 3), acceptable values of experimental variability.

Table 5. Comparison of means of eight variables as a function of five foliar treatments with biostimulants. Average of environments and hybrids in the High Valleys of Mexico. Agricultural cycle, spring-summer, 2017.

Foliar bioestimulants	Agronomic variables							
	YIE (t ha ⁻¹)	VW (kg hl ⁻¹)	SP (t ha ⁻¹)	RC (num.)	GR (num.)	NGC (num.)	W200 (g)	CD (mm)
B1	11.4 c	79.9 b	14.6 a	16 b	29.7 b	473 c	64.8 c	47.7 c
B2	12.4 bc	80.6 a	15.5 a	16.5 ab	30.5 ab	502 bc	66.9 a	48.9 a
B3	12.7 a	80.3 ab	14.9 a	16.4 ab	31 a	507 ab	65.5 ab	48.2 ab
B4	12.5 ab	80 b	15.3 a	16.5 ab	30.4 ab	499 bc	66 ab	48.4 ab
B5	12.3 bc	80 b	14.7 a	16.6 a	31.1 a	516 a	65.7 ab	48.1 ab

Means with different letters in a column are statistically different (Tukey; $p \leq 0.05$). YIE= grain yield; VW= volumetric weight; SP= straw production; RC= rows per cob; GR= grains per row; NGC= number of grains per cob; W200= weight of 200 grains; CD= cob diameter; B1= control; B2= Eurobor; B3= Euroligo; B4= Eurodual; B5= Euroalg.

B2 stands out, which resulted in higher density of the corn grain (80.6 kg hl⁻¹), rows per cob (16.5), weight of 200 grains (66.9 g) and cob diameter (48.9 mm) followed by B3, B4, B5, respectively, compared to the control (B1). This B2 response is probably due to the presence of 3% B, which, among other functions, participates in the development of fruits and seeds, responsible for sugar transport, hormone development and cell division (Goldbach, 2001). In pioneering studies on the effect of boron on cob formation and corn grain yield components, Mozafar (1987); Vaughan (1977) conclude that, in the deficiency of B, the stigmas become little receptive to pollen, this is because the B is immobile in the phloem of the plant, which leads to a lower number of rows and grains per row on the cob, so the application of B is recommended for better pollination and grain formation, which is evident in the present study (Table 5).

The significant differences for rows per cob, grains per row and number of grains per cob in the biostimulant factor show consistency of the three variables, which influenced the grain yield both in the treatment with the highest yield (B3) and the control (B1) that presented lower yield. A positive correlation was detected between RC vs NGC ($r= 61$) and GR vs NGC ($r= 77$). Rows per cob, grains per row, and number of grains per cob provided values of 16-16.6; 29.7-31.1; 473.3-515.6, respectively (Table 5). This last variable, on average of the four treatments with biostimulants (B2, B3, B4 and B5), represents an increase of 6.9%, compared to the control (B1). Tejada *et al.* (2018) verified an increase of 8.9 to 15.8% in the number of grains per cob when they applied increasing doses of amino acid-based biostimulants, in two agricultural cycles (Table 5).

The above data also exceed those reported by Martínez *et al.* (2018), Zamudio *et al.* (2018); Tadeo *et al.* (2020); Vázquez *et al.* (2020), with hybrids adapted to environmental conditions similar to this study. The positive responses observed in RC, GR and NGC are associated with the genetic quality of the seed of the hybrids, complementary foliar fertilization (Zamudio *et al.*, 2018) and the timely application of nutrients in times of increased demand. The foliar biostimulants affected the volumetric weight, presenting very dense grains that range from 79.9 (B1) to 80.6 (B2) kg hl⁻¹ (Vázquez *et al.*, 2020), these values exceed what was reported by Zamudio *et al.* (2018) in a work with foliar fertilization with different products enriched with nitrogenous organic material.

Thus, what is required by the dough and tortilla industry (DTI) and the nixtamalized flour industry (NFI), which demand grains with a VW greater than 74 kg hl⁻¹ (SE, 2002), is met. In addition, a close positive correlation between VW and YIE ($r= 60$) was observed through Pearson's correlation analysis, which allows deducing that the hybrids evaluated with efficient agronomic management such as foliar fertilization, specifically with the use of biostimulants, may be suitable or favorable for the industry.

There was a significant difference in the weight of 200 grains between the biostimulants and it ranged from 64.8 in B1 to 66.9 g in B2. The increase in W200 was probably due to the application of nitrogen products that maintained the photosynthetic activity of the plant for a longer period, allowing greater accumulation of reserves in the grains and finally greater weight of the harvested grains. The result of this study corroborates what was reported by Gazola *et al.* (2014), on the positive correlation between grain mass and corn yield ($r= 60$) (Table 5).

The cob diameter showed significant differences for the biostimulant factor (Table 5) and ranged from 47.7 (B1) to 48.9 (B2) mm, respectively. Like the previous variables, all four treatments exceed the control (B1). The observed values of CD are similar to those reported by Zamudio *et al.* (2018). The cob diameter is associated with the genetic factor of the seeds, agronomic management and the environmental conditions that prevail during the cycle. The results of this study coincide with those of Quezada *et al.* (2015); Galindo *et al.* (2015); Zamudio *et al.* (2018); Tejada *et al.* (2018) on the effect of biostimulants on corn plants, which significantly increased grain yield and agronomic characteristics of the crop.

Conclusions

The application of biostimulants in the critical moments of growth and development of the plant favors the productive potential of the hybrids evaluated for the environments of the High Valleys of the State of Mexico, especially in conditions of osmotic stress. The environment with the best response corresponds to Temascalcingo, followed by Jocotitlán and Jilotepec. On average, the hybrids H-66, H-50 and H-76 stand out, however, all genotypes exceeded the regional yield average. In the application of biostimulants, grain yield increased from 0.9 to 1.3 t ha⁻¹. Technically, the best responses were observed in the biostimulants, B2, B3 and B4. Within B2, the hybrids with the best responses were Tlaoli Puma, H-47AE, H-66 and #46#48, for B3 (Ixim Puma, H-47AE, H-66, #46#48), in B4 (Tlaoli Puma, Ixim Puma, H-47AE, H-50, H-66 and #46#48), and for B5 in H-66. The easy assimilation due to amino acid complexes is responsible for the increase in grain yield. In this sense, foliar biostimulants are an alternative in complementary fertilization to increase production in the cultivation of corn.

Cited literature

- Ahmad, R.; Khalid, A. H.; Arshad, M.; Zahir, A. and Mahmood, T. 2007. Effect of compost enriched with N and L-tryptophan on soil and maize. *Agron. Sustain. Dev.* 2(28):299-305.
- Ashmead, H. D. 1986. The absorption mechanism of amino acid chelates by plant cells. *In*: Ashmead, H. D. Foliar feeding of plants with amino acid chelates. Noyes publications, park ridge, NY. 219-235. pp
- Battacharyya, D.; Babgohari, M. Z.; Rathor, P. and Prithiviraj, B. 2015. Seaweed extracts as biostimulants in horticulture. *Sci. Hortic.* 1(196):39-48. Doi: 10.1016/j.scienta.2015.09.012.
- Barbosa, R. H.; Tabaldi, L. A.; Miyazaki, F. R.; Pilecco, M.; Kassab, S. O. and Bigaton, D. 2013. Foliar copper uptake by maize plants: effects on growth and yield. *Ciência Rural.* 9(43):1561-1568.
- Bray, R. H. and Kurtz, L. T. 1945. Determination of total, organic and available forms of phosphorus in soil. *Soil Sci.* 59(1):39-45.
- Calvo, P.; Nelson, L. and Kloepper, J. W. 2014. Agricultural uses of plant biostimulants. *Plant Soil.* 1-2(383):3-41.
- Cavani, L.; Ter-Halle, A.; Richard, C. and Ciavatta, C. 2006. Photosensitizing properties of protein hydrolysates-based fertilizers. *J. Agr. Food Chem.* 24(54):9160-9167.
- Crouch, I. J. and Van-Staden, J. 1994. Evidence for rooting factors in a seaweed concentrate prepared from *Ecklonia maxima*. *J. Plant Physiol.* 3(137):319-322.
- Du Jardin, P. 2015. Plant biostimulants: definition, concept, main categories and regulation. *Sci. Hortic.* 1(196):3-14. Doi: 10.1016/j.scienta.2015.09.021.
- Fernández, V; Sotiropoulos, T. y Brown, P. 2015. Fertilización foliar: principios científicos y práctica de campo. Paris, Francia, Asociación Internacional de la Industria de Fertilizantes (IFA). 49-82 p
- Galindo, S. F.; Nogueira, M. L.; Bellote, J. L. M.; Gazola, R. N.; Alves, C. J. and Teixeira, F. M. C. M. 2015. Desempenho agrônômico de milho em função da aplicação de bioestimulantes à base de extrato de algas. *Tecnol. Ciên. Agropec.* 1(9):13-19.

- Gazola, D.; Zucareli, C.; Silva, S. R. and Fonseca, C. B. I. 2014. Aplicação foliar de aminoácidos e adubação nitrogenada de cobertura na cultura do milho safrinha. *Rev. Bras. Eng. Agríc. Ambient.* 1(18):700-707.
- Goldbach, H. E.; YU, Q.; Wingender, R.; Schulz, M.; Wimmer, M.; Findeklee, P. and Baluska, R. Rapid response reactions of roots to boron deprivation. 2001. *J. Plant Nutr. Soil Sci.* 2(164):173-181.
- Grabowska, A.; Kunicki, E.; Sekara, A.; Kalisz, A. and Wojciechowska, R. 2012. The effect of cultivar and biostimulant treatment on the carrot yield and its quality. *Veg. Crops Res. Bull.* 1(77):37-48.
- Ibrahim, S. M. M.; Taha, L. S. and Farahat, M. M. 2010. Influence of foliar application of pepton on growth, flowering and chemical composition of *Helichrysum bracteatum* plants under different irrigation intervals. *Ozean J. Appl. Sci.* 3(1):143-155.
- Jannin, L.; Arkoun, M.; Etienne, P.; Laíné, P.; Goux, D.; Garnica, M.; Fuentes, M.; Francisco, S. S.; Baigorri, R. and Cruz, F. 2013. Brassica napus growth is promoted by *Ascophyllum nodosum* (L.) Le Jol. Seaweed extract: microarray analysis and physiological characterization of N, C, and S metabolisms *J. Plant Growth Regul.* 1(32):31-52.
- Jones, D. L. and Kielland, K. 2002. Soil amino acid turnover dominates the nitrogen flux in permafrost-dominated taiga forest soils. *Soil Biol. Biochem.* 2(34):209-219.
- Kumar, P. and Sahoo, D. 2011. Effect of seaweed liquid extract on the growth and yield of *Triticum aestivum* var. Pusa gold *J. Appl. Phycol.* 2(23):251-255.
- Maeda, H. and Dudareva, N. 2012. The shikimate pathway and aromatic amino acids biosynthesis in plants. *Annu. Rev. Plant Biol.* 1(63):73-105. Doi: 10.1146/annurev-arplant-042811-105439.
- Martinka, M.; Vaculík, M. and Lux, A. 2014. Plant cell responses to cadmium and zinc. In *Applied plant cell biology: cellular tools and approaches for plant biotechnology, plant cell monographs*; Nick, P. and Opatrny, Z. (Ed.) Springer: Berlin/Heidelberg, Germany. 209-246 pp.
- Martínez, G. A.; Zamudio, G. B.; Tadeo, R. M.; Espinosa, C. A.; Cardoso, G. J.; Vázquez, C. G. y Turrent, F. A. 2018a. Rendimiento de híbridos de maíz grano blanco en cinco localidades de valles Altos de México. *Rev. Mex. de Cienc. Agríc.* 7(9):1447-1458.
- Mattner, S. W.; Wite, D.; Riches, D. A.; Porter, I. J. and Arioli, T. 2013. The effect of kelp extract on seedling establishment of broccoli on contrasting soil types in southern Victoria, Australia. *Biol. Agric. Hortic.* 4(29):258-270.
- Mladenova, Y. I.; Maini, P.; Mallegni, C.; Goltsev, V.; Vladova, R. and Vinarova, K. 1998. Siapton-anamino-acid-based biostimulant reducing osmotic stress metabolic changes in maize. *Agro Food Ind. Hi-Tech.* 6(9):18-22.
- Mohanty, D.; Adhikary, S. P. and Chattopadhyay, G. N. 2013. Seaweed liquid fertilizer (slf) and its role in agriculture productivity. *Ecoscan. Special Issue.* (3):147-155.
- Mozafar, A. A. 1987. Effect of boron on ear formation and yield components of two maize (*Zea mays* L.) hybrids. A. Institute of plant sciences, swiss federal institute of technology (ETHZ), 8092, Zurich. 319-332 pp.
- Parrado, J.; Bautista, J.; Romero, E. J.; García-Martínez, A. M.; Friaza, V. and Tejada, M. 2008. Production of a carob enzymatic extract: potential use as a biofertilizer. *Biores. Technol.* 7(99):2312-2318.
- Petrozza, A.; Summerer, S.; Di-Tommaso, G.; Di-Tommaso, D. and Piaggese, A. 2013. Evaluation of the effect of Radifarm® treatment on the morpho-physiological characteristics of root systems via image analysis. *Acta Hortic.* 1009(18):149-153.

- Quezada, J. C.; Lenssen, A. W.; Moore, K. J.; Sawyer, J. E. and Summer, P. 2015. Amino acid biosynthesis byproducts are a suitable source of nitrogen for corn production. *Field Crop. Res.* 1(184):123-132.
- Rai, V. K. 2002. Role of amino acids in plant responses to stresses. *Biol. Plant.* 4(45):481-487.
- SAS Institute. 2002. The SAS system for Windows user's guide. Release 9.4. SAS Institute, Cary, NC.
- Sotelo, R. E. D.; González, H. A.; Cruz, B. G.; Moreno, S. F. y Cruz, C. G. 2011. Los suelos del estado de México y su actualización a la base referencial mundial del recurso suelo 2006. *Rev. Mex. Cienc. Forest.* 8(2):71-84.
- Tadeo, R. M.; Zamudio, G. B.; Espinosa, C. A.; Turrent, F. A.; Cárdenas, M. A. L.; López, L. C.; Arteaga, E. I. y Valdivia, B. R. 2015. Rendimiento de maíces nativos e híbridos en diferente fecha de siembra y sus unidades calor. *Rev. Mex. Cienc. Agríc.* 1(6):33-43.
- Tadeo, R. M.; Espinosa, C. A.; Canales, I. E.; López, L. C.; Zamudio, G. B.; Turrent, F. A.; Gómez, M. N.; Sierra, M. M.; Martínez, G. A.; Valdivia, B. R. and Andrés, M. P. 2020. Grain yield and population densities of new corn hybrids released by the INIFAP and UNAM for the high valleys of Mexico. *Terra Latinoam.* 3(38):507-515. Doi: <https://doi.org/10.28940/terra.v38i3.557>.
- Tejada, M.; Rodríguez, M. B.; Gómez, I.; Franco, A. L.; Benítez, C. and Parrado, J. 2016. Use of biofertilizers obtained from sewage sludges on maize yield. *Eur. J. Agron.* 1(78):13-19.
- Tejada, M.; Rodríguez, M. B. P. P. and Parrado, J. 2018. Effects of foliar fertilization of a biostimulant obtained from chicken feathers on maize yield. *Eur. J. Agron.* 1(96):54-59.
- Turrent, F. A.; Cortés, F.; Espinosa, C. A.; Turrent, T. C. y Mejía, A. H. 2016. Cambio climático y algunas estrategias agrícolas para fortalecer la seguridad alimentaria de México. *Rev. Mex. Cienc. Agríc.* 7(7):1727-39.
- Vaughan, A. K. F. 1977. The relation between the concentration of boron in the reproductive and vegetative organs of maize plants and their development. *Rhod. J. Agric. R.* 9(15):163-170.
- Vázquez, C. G.; Martínez G. A.; Zamudio, G. B.; Espinosa, C. A.; Tadeo, R. M.; y Turrent, F. A. 2020. Estabilidad de rendimiento y características fisicoquímicas de grano de híbridos de maíz en Valles Altos de México. *Rev. Mex. Cienc. Agríc.* 8(11):1803-1814.
- Yakhin, O. I.; Lubyantsev, A. A.; Yakhin, I. A. and Brown, P. H. 2017. Biostimulants in plant science: a global perspective. *Front. Plant Sci.* 1(7):1-32. Doi: 10.3389/fpls.2016.02049.
- Zamudio, G. B.; Félix, R. A.; Martínez, G. A.; Galvão, C. J. C.; Espinosa, C. A. y Tadeo, R. M. 2018. Producción de híbridos de maíz con urea estabilizada y nutrición foliar. *Rev. Mex. Cien Agríc.* 6(9):1231-1244.
- Zhao, Y. 2010. Auxin biosynthesis and its role in plant development. *Annu. Rev. Plant Biol.* 1(61):49-64. Doi: 10.1146/annurev-arplant-042809-112308.