



Biomass and Harvest Index of Two Quality Protein Corn Varieties with Bio-Fertilization in Two Luvisols of Yucatan, Mexico

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Abstract

This work aimed to evaluate the above-ground Total Dry Biomass (TDB) and the Harvest Index (HI) of two quality protein maize varieties. Biofertilizers (*Bio*) in combination with chemical fertilizers (*Chem*) were applied in two Luvisols with low (Lot 1) and high (Lot 2) intensive agricultural use. Eight treatments resulted from combining the two varieties: *Chichen Itza* (*Chich*) and *Sac Beh* (*Sac*) with 1) a chemical fertilizers dose (60-80-00): (N-P₂O₅-K₂O) alone; 2) supplemented with biofertilizers (60-80-00 + Mycorrhizae + *Azospirillum*); 3) a half nitrogen dose plus biofertilizers (30-80-00 + Mycorrhizae + *Azospirillum*) and 4) the control (00-00-00). At physiological maturity, the TDB (grain, leaves, stalks and husk) in t·ha⁻¹ was used to calculate the Harvest Index (HI). The relationship between partial biomass (PB) on GY was assessed. No statistical differences were found. Regardless of treatments, the general average of TDB, PB and GY, of both varieties was higher in Lot 1. *Sac* produces more PB than *Chich* in all treatments including the *Control*. The maximum GY's, in Lot 1 for both *Chich* (5.88 t·ha⁻¹) and *Sac* (5.83 t·ha⁻¹) were practically the same. T5 (*Chem 1-Bio-Chich*) and T6 (*Chem 1-Bio-Sac*) were the best treatments. However, *Sac* obtained the maximum HI (0.49) whilst *Chich* had 0.43. No effect on HI was found when applying *Bio* to *Chich* as *Sac* showed. The lowest HI of both varieties was found in the *Control* and those treatments with half nitrogen. In Lot 2, *Sac* continues with the highest PB (6.6 t·ha⁻¹) with practically the same GY (4.63 t·ha⁻¹) as *Chich* (4.84 t·ha⁻¹). However, *Chich*, and no *Sac*, showed the best HI (0.46) performance with T5 (*Chem 1-Bio-Chich*). It seems that the GY of *Sac* can be more predictable if PB is used as an indicator according to high Determination Coefficients (R²) in both lots.

Subject Areas

Agricultural Engineering

Keywords

Intensive Use, *Sac Beh*, *Chichen Itza*, Mycorrhizae, *Azospirillum*

1. Introduction

In America, the United States produces an estimated (2016/2017) 382.47 million tons of maize followed by Brazil with 83.5, Argentina with 36.5 and Mexico with 24.5. Mexico is a country with the lowest yields of 3.2 t·ha⁻¹ in contrast to the yields of the USA and Argentina with 9.3, 8.0 t·ha⁻¹, respectively, but similar to Brazil with 3.5 t·ha⁻¹.

Even though the Mexican production is low, there is a tendency to increase since in 1993: it was 1.8 t·ha⁻¹ and currently it is 3.2 t·ha⁻¹; cultivating nowadays is approximately 7 million 157 thousand 586 hectares [1]; it is mainly in sub-humid tropical, temperate humid and sub-humid zones [2].

Most of the corn is produced under rainfed conditions [1] for self-consumption and it is grown by 2 million small producers [2] contributing to more than half of the national food security of the poorest rural strata [3]. These producers are still using native varieties with large genetic diversity [4], but with very low yield potential and poor protein quality.

INIFAP has released improved Creole varieties converted to protein quality adapted to the stony soils of Yucatan, Mexico such as *Sac Beh* and *Chichen Itza*, which have more than 50% Lysine and Tryptophan than the common Creole maize. Average yields of 2.23 to 3.33 t·ha⁻¹ have been reported on rocky soils and can reach more than 5.0 t·ha⁻¹ on better deep soils such as the *Luvisols* (LV) [5].

Using those improved varieties can be more profitable and environmentally friendly if cheaper in-puts such as biofertilizers (*Mycorrhizae* and *Azospirillum*) are to be incorporated into the traditional production systems with the idea of partially replacing the chemical fertilizers.

Rodríguez-Eugenio *et al.* (2019) [6] comment that soil contamination due to excessive applications of chemical fertilizers reduces food security; and nutrients such as nitrogen and phosphorus are transported to the surface and groundwater, contaminating the water.

Carcaño-Montiel *et al.* (2006) [7] argue that with biofertilizers the native phosphorus and potassium of the soil are exported to the plant and the acidifying effect of ammoniacal nitrogenous fertilizers is reduced.

On the other hand, as has been mentioned by Aguilar Carpio *et al.* (2015) [8], crop growth is influenced mainly by climate and nutrients; so the influence of biofertilizers on productivity can be studied from the analysis of the dry matter accumulation and its relationship with other factors such as nitrogen, soil chem-

ical characteristics and environmental conditions. One way to do it is by studying the Harvest Index (HI) defined as the ratio of grain to total shoot dry matter as a valuable parameter to measure reproductive efficiency. It is determined by interactions between genotypes (G), environment (E) and crop management (M) and can measure the physiological efficiency and ability of a crop for converting the total dry matter into economic yield [9].

Even though general studies indicate that the inoculation with *Azospirillum*, *Glomus* and the use of nitrogen increases dry matter production and grain (as a result of higher growth indexes) in both native and hybrid maize, there is a lack of information on this subject in the tropical regions of Mexico.

Thus, the purpose of this work was to evaluate the total biomass production and the Harvest Index of two improved quality protein native corn varieties when biofertilizers in combination with chemical fertilizers were applied in two different *Luvisols* with low and high intensive agricultural use.

2. Materials and Methods

The work was carried out in the state of Yucatan, Mexico in the spring-summer 2017/2017 season under favorable rainfed conditions at the INIFAP-UXMAL Experimental Station located at 20°29'08.1" North Latitude and 89°24'39" West Longitude, in an altitude of 50 meters above sea level [10].

The yellow grain *Chichen Itza* (**Chich**) and the white grain *Sac Beh* (**Zac**) were the corn varieties classified as Quality Protein ones, and used as phytometers in two different soils classified as *Luvisols*.

2.1. Selection of Experimental Plots

The first Lot 1 had a low intensive agricultural use and maize has been grown every 4 to 5 years with long fallow periods. In the second Lot 2 corn has been grown every year with intensive use of chemical fertilizers. Both Lots have contrasting chemical characteristics such as salinity, electrical conductivity and phosphorus contents.

The soil attributes, analyzed by Phytomonitor (2018) [11], were compared with reference data from Nom-021-Semarnat-2000 [12]. Even when the pH's are neutral, the electrical conductivity of Lot 1 is lower (EC = 0.66 mS/cm) than Lot 2 (1.53 mS/cm). Sodium (Na) is higher in Lot 2 (330 vs. 165 ppm). The organic matter (OM) is satisfactory in both lots, but it is higher in Lot 1 (2.78% vs. 2.11%).

According to the official Mexican standards [12] Phosphorus (P) in Lot 1 is in the optimal range (17 ppm) however, in Lot 2, with more intensive use, P is in excess with 80 ppm. Potassium (K) is in excess in both Lots with more than 1000 parts per million (ppm).

2.2. Treatments, Variables and Statistical Analysis

Eight treatments were studied in experimental units of 5 m × 4 m (20 m²) with

four rows of 1 m wide and 5 m long. The corn population density was estimated in 50,000 plants ha⁻¹ (**Figure 1** and **Figure 2**). The treatments resulted when the two varieties (**Figure 3** and **Figure 4**) named: *Chichen Itza* (**Chich**) and *Sac Beh* (**Sac**) were combined with the following four levels of fertilization: 1) chemical (**Chem 1**) fertilization (N-P₂O₅-K₂O): (60-80-00), 2) chemical fertilization (**Chem 1**) with biofertilizers (**Bio**): (60-80-00 + Mycorrhizae + *Azospirillum*), 3) second dose of chemical fertilization (**Chem 2**) with biofertilizers (**Bio**): (30-80-00 + Mycorrhizas + *Azospirillum*) and 4) The control (00-00-00). The treatments were distributed in a completely randomized block design with three repetitions in each Lot and were identified according to **Table 1**.



Figure 1. Topological arrangement of corn plants.



Figure 2. Corn materials at flowering.



Figure 3. *Chichen Itza* yellow grain variety.



Figure 4. *Sac Beh* white grain variety.

Table 1. Treatments studied in Lot 1 and Lot 2 with *Chichen Itza* and *Sac Beh*.

(N°)	Treatment	Fertilization (N-P ₂ O ₅ -K ₂ O)
1	<i>Control-Chich</i>	(00-00-0) No Bio
2	<i>Control-Sac</i>	(00-00-0) No Bio
3	<i>Chem 1-Chich</i>	(60-80-00) No Bio
4	<i>Chem 1-Sac</i>	(60-80-00) No Bio
5	<i>Chem 1-Bio-Chich</i>	(60-80-00) with Bio
6	<i>Chem 1-Bio-Sac</i>	(60-80-00) with Bio
7	<i>Chem 2-Bio-Chich</i>	(30-80-00) with Bio
8	<i>Chem 2-Bio-Sac</i>	(30-80-00) with Bio

Six plants with complete competence were selected, and at the end of the physiological cycle the Total Dry Biomass (**TDB**) production (stems, leaves, husk and grain) were measured and converted into t·ha⁻¹. The stems, leaves, and husk were considered as the Partial Biomass (**PB**) weighed under field conditions (**Figure 5**) and considered as the main variable influencing Grain Yield (**GY**) production. With the **TDB** and **GY**, all in dry base, the HARVEST INDEX (**HI**) was calculated with the formula: **GY/TDB**. An Analysis of Variance (**ANOVA**) with their Coefficients of Variation (**CV**) were performed to all variables.

2.3. Inoculation of Biofertilizers and Chemical Fertilization

The seeds were inoculated with a mixture (1:1 ratio) of both: 1) INIFAP™ brand biofertilizer with *Rhizophagus intraradices* (*Mycorrhizae* fungus) at a concentration of ≥60 spores and 2) *Azospirillum brasilense* (Bacterium) at a concentration of 1 × 10⁻⁶ Colony Forming Units (CFU) mL⁻¹. After inoculation the seeds were dried at room temperature for 8 hours to be planted in the experimental plots. 15 days after sowing, the chemical fertilizer was applied to the corresponding treatments. The fertilizer was buried 10 cm from the corn stem in the form of Urea (N) and Triple Calcium Superphosphate (P₂O₅) in a single application.



Figure 5. Weighing dry partial biomass under field conditions.

3. Results and Discussion

The variables to be discussed below are highly implicated in the efforts or capacity of plants, as biological machines, to convert most of their biomass into grain. The Harvest Index (HI) is a very important trait for plant breeding. The higher the capacity of corn plants to produce economic yields the higher the probability to be selected for breeding programs. In that way, the food self-sufficiency of a country can be ensured.

3.1. Statistic Analysis

Table 2 shows the Mean Squares and the statistical significance when submitting the grain yield and the other biomass components ($t\cdot ha^{-1}$) to the corresponding Analysis of Variance (ANOVA). No statistical differences were found ($p = 95\%$); therefore, applying chemical fertilizers (*Chem*), alone or combined with biofertilizers (*Chem-Bio*) and even not applying any treatment (*Control*) is statistically the same. However, the information will be discussed later on when considering the arithmetic data of the investigation. However, it seems that further research is needed since the variables studied, under the specific conditions of this work, did not implicate any substantial change.

The CV's (%) ranged from 0.014 in Partial Biomass Lot 2 to 18.23 in Partial Biomass Lot 1. Authors such as Pimentel (1985) [13] comment that the CV's can be different depending on the type of experiments. Other authors such as: [14] [15] [16] Gómez and Gómez (1984) [14]; Martínez (1988) [15]; Patel *et al.* (2001) [16] suggest that when the CVs are greater than 30%, the experiments have low precision.

Similar statistical results were reported by Uribe Valle and Dzib Echeverria (2006) [10] and Uribe-Valle *et al.* (2007) [17]. They did not find any statistical differences between yields when corn was treated with Mycorrhizae + *Azospirillum*, a chemical treatment (N-P₂O₅-K₂O) (40-100-00) or a control (00-00-00) in a *Luvisol* of Yucatan, Mexico.

However, other authors in north Mexico like Díaz Franco *et al.* (2012) [18] have found important statistical differences suggesting that mycorrhizal inoculation alone was very competitive in relation to chemical fertilization. The

Table 2. Mean squares and statistical significance between treatments evaluated for grain yield and partial biomass (t·ha⁻¹) through the analysis of variance.

Source of Variation	Df	Yield Lot 1	Yield Lot 2	Partial Biomass Lot 1	Partial Biomass Lot 2
Treatments	7	1.946ns	4.199ns	1.592 ns	1.025ns
Repetitions	2	0.067ns	2.918ns	0.471ns	0.177ns
Error (EE)	14	10.883	3.441	2.114	0.786
CV (%)		16.07	10.88	18.23	0.014

ns =Statistically no significant at $p = 95\%$; EE = Experimental Error, CV = Coefficient of Variation.

combined inoculation of *G. intraradices* and *A. brasilense*, did not present any additive effect on corn growth. In addition, of the ecological advantage, it is more profitable by reducing the cost production of corn as compared to the use of chemical fertilizers.

3.2. Biomass Production and Harvest Index (HI)

3.2.1. Lot 1 vs. Lot 2

The production of biomass (t·ha⁻¹) and the HI are observed in **Table 3** and **Table 4**. Regardless of the treatments, the average partial biomass production in Lot 1 was 2.36 t·ha⁻¹ higher than that of Lot 2 and so was the grain yield with more than 0.76 t·ha⁻¹. This difference may be due to the high sodium (Na) content and higher Electrical Conductivity (EC) of Lot 2. The sensitivity of corn to salinity has been argued by Ayala-Contreras (2015) [19]. Despite the contrasting results with biomass, the **HI** was similar in each Lot, ranging from 0.41 and 0.42; indicating that of the total biomass, a little more than 40% refers to the grain in both experimental Lots. It seems that the extraction process of photo-assimilates, to form grain, is equally efficient in both Lots regardless of their contrasting chemical soil conditions. Studies on this subject have been reported by López-Castañeda (2011) [20] in barley crop growing in soils with different moisture conditions.

3.2.2. Chichen Itza and Sac Beh in Lot 1

The contrasting agronomic behavior between *Chich* and *Sac*, in Lot 1, are shown in **Table 3**. *Sac* produces more **PB** than *Chich* in all treatments including the *Control*. The maximum **GY**, in Lot 1 for both *Chich* and *Sac* varieties was found with the same formula *Chem1-Bio* where 60 kilos N ha⁻¹, 80 kilos of phosphorus as P₂O₅ plus biofertilizers were applied as it is reflected in T5 (*Chem 1-Bio-Chich*) and T6 (*Chem 1-Bio-Sac*) with 5.88 and 5.83 t·ha⁻¹ respectively. Even with practically the same **GY**, it was *Sac* which obtained the maximum **HI** (0.49) whilst *Chich* had 0.43 with the same abovementioned treatments. It was no found any effect on **HI** when applying *Bio* to *Chich* as it is observed when comparing T3 (0.44) vs. T5 (0.43). However, *Sac* showed better response to *Bio* as it is seen in T4 (0.41) vs. T6 (0.49).

Table 3. Biomass, yield production and Harvest Index (HI) with biofertilizers in a low intensive agricultural use *Luvisol* (Lot 1) in the Experimental Field Station at Uxmal Yucatan, Mexico.

Treatments	Partial Biomass (t·ha ⁻¹)	Corn Yield (t·ha ⁻¹)	Total Biomass (t·ha ⁻¹)	HI
<i>T1 (Control-Chich)</i>	8.00	5.66	13.66	0.41
<i>T2 (Control-Sac)</i>	8.75	5.05	13.80	0.36
<i>T3 (Chem 1-Chich)</i>	6.80	5.48	12.28	0.44
<i>T4 (Chem 1-Sac)</i>	8.25	5.77	14.02	0.41
<i>T5 (Chem1-Bio-Chich)</i>	7.50	5.88	13.38	0.43
<i>T6 (Chem 1-Bio-Sac)</i>	8.42	5.83	14.25	0.49
<i>T7 (Chem 2-Bio-Chich)</i>	7.20	5.00	12.20	0.40
<i>T8 (Chem 2-Bio-Sac)</i>	8.78	5.36	14.14	0.37
<i>Average</i>	7.96	5.50	13.49	0.41

Table 4. Biomass, grain yield and Harvest Index (HI) with biofertilizers in a low intensive agricultural use *Luvisol* (Lot 2) in the experimental field station at Uxmal Yucatan, Mexico.

Treatments	Partial Biomass (t·ha ⁻¹)	Corn Yield (t·ha ⁻¹)	Total Biomass (t·ha ⁻¹)	HI
<i>T1 (Control-Chich)</i>	5.60	4.58	10.18	0.44
<i>T2 (Control-Sac)</i>	6.40	4.13	10.53	0.39
<i>T3 (Chem 1-Chich)</i>	6.70	5.27	11.97	0.44
<i>T4 (Chem 1-Sac)</i>	6.70	4.94	11.64	0.42
<i>T5 (Chem1-Bio-Chich)</i>	5.60	4.92	10.52	0.46
<i>T6 (Chem 1-Bio-Sac)</i>	6.00	4.17	10.17	0.41
<i>T7 (Chem 2-Bio-Chich)</i>	6.40	4.62	11.02	0.41
<i>T8 (Chem 2-Bio-Sac)</i>	7.30	5.30	12.60	0.42
<i>Average</i>	5.60	4.74	11.08	0.42

The lowest HI's of both varieties *Chich* and *Sac* were found in the *Control* T1 and T2 with 0.41 and 0.36 respectively, as well as those treatments (T7) and (T8) where N was reduced by half. The difference between treatments can be related to the root growth and the high exploring volume when biofertilizers are applied. By instance, works in the north of Mexico [18] have indicated that the root volume of corn can be 75% higher when *Mycorrhizae* fungus was applied as compared to the *Control*. The higher root volume of 155 cm³ was obtained with *Mycorrhizae* alone while the *Control* just had 40 cm³; and the total fresh fodder was exceeded by more than 60%.

3.2.3. Chichen Itza and Sac Beh in Lot 2

As it is observed in **Table 4** of Lot 2, *Sac* continues with a general trend of having the highest **PB** with practically the same **GY** ($4.63 \text{ t}\cdot\text{ha}^{-1}$) as *Chich* ($4.84 \text{ t}\cdot\text{ha}^{-1}$). However, in this Lot 2, *Chich*, and no *Sac*, showed the best **HI** (0.46) performance. In these soils, of highly intensive agricultural use, the **HI** of *Sac* is practically the same (0.41 to 0.42) in all treatments referred to *Chem* alone or combined with *Bio*. However, the Control (T2) showed the lowest **HI** with 0.39. On the other hand, *Chich* obtained the highest **HI** (0.46) with T5 (*Chem 1-Bio-Chi*) and the lowest one (0.41) with T7 (*Chem 2-Bio-Chi*).

Similar works, but with a corn hybrid (H-526), have indicated higher **HI**'s, ranging from 0.49 to 0.63 [21] compared with the varieties *Chich* and *Sac*. The lower **HI** of 0.49 was related to the *Control* whilst the higher one was for a chemical fertilizer of (120 N-160 P_2O_5 -000 K_2O). However, when chemical fertilizer (120 N-80 P_2O_5 -000 K_2O) was complemented with *chicken manure*, the **HI** of the hybrid was similar (0.49) as the *Control* [21]. It seems that applying more fertilizers does not necessarily increase the **GY**, but it does increase the production of **PB** [21]. Studies related to the agronomic behavior of tropical corn hybrids, in the state of Veracruz, Mexico [22] have indicated that the **HI**'s can range from 0.2 to 0.5 depending on the corn material.

3.3. Correlations Coefficients (R^2) between Grain Yield and Partial Biomass ($\text{t}\cdot\text{ha}^{-1}$)

Table 5 shows the Determination Coefficients (R^2) when the **GY**'s were compared with the **PB**. The highest R^2 was obtained with *Sac* in both experimental lots with 0.75 and 0.84 for Lot 1 and Lot 2 respectively; while *Chich* showed very low Determination Coefficients of 0.19 and 0.27 in each lot. This indicates that the **GY** of *Sac* can be associated more intensely with the above ground **PB** than that of *Chich*.

The above analysis indicates that the **GY** may not always be highly associated with the production of **PB** but also depends on the genetic material and other factors that need further study. It has been found [23] that the dry matter and

Table 5. Determination Coefficients (R^2) for grain yield as dependent variable (Y) vs. partial biomass as independent variable (X) ($\text{t}\cdot\text{ha}^{-1}$) for both *Chichen Itza* and *Sac Beh* varieties in lots with low (Lot 1) and high intensive agriculture use (Lot 2).

Variety/Lot	Corn Yield ($\text{t}\cdot\text{ha}^{-1}$) (Y)	Partial Biomass ($\text{t}\cdot\text{ha}^{-1}$) (X)	Determination coefficient (R^2)
<i>Chich/Lot 1</i>	5.51	7.38	0.19
<i>Chich/Lot 2</i>	4.84	6.08	0.27
<i>Average</i>	5.17	6.73	0.23
<i>Sac/Lot 1</i>	5.50	8.55	0.75
<i>Sac/Lot 2</i>	4.64	6.60	0.84
<i>Average</i>	5.07	7.57	0.80

grain yield in the corn hybrid H-562 (applying 160 kg N ha⁻¹) was mainly related to the size and duration of the photosynthetic biomolecular apparatus (Leaf Area and Duration of the Total Leaf Area) which induced a highest Growth and Assimilation Rate.

On the other hand, with the Vandeño corn variety, better response was obtained with biofertilizer and the application of nitrogen; but even when the dry matter increased this was not substantially reflected in the grain yield. This behavior is due to a higher expansion, duration and speed growth of the plant canopy [23].

Referring to previous works of Ramírez *et al.* (2020) [24] related to the contents of amino acids *Lysine* and *Tryptophan* in both *Chich* and *Sac*, it would be very important to quantify the relationship between those amino acids and the variables studied in this work. But in a first glance, it seems that the HI's of *Sac*, in both lots, are better associated to the amino acids than that of *Chich*. However, there is a trend for the amino acids to decrease while the HI's increase. This contrasting agronomic and biochemical behavior needs a further and deep understanding to improve the corn breeding programs.

4. Conclusions

No statistical differences were found between treatments in any *Luvisol*. However, regardless of the treatments, the general average of TDB, PB and GY of both varieties were higher in Lot 1 with the lower intensive agriculture use. *Sac* produces more PB than *Chich* in all treatments including the *Control*.

The maximum GY, in Lot 1 for both *Chich* and *Sac* varieties, was found with the same formula *Chem 1-Bio* as reflected by T5 (*Chem 1-Bio-Chich*) and T6 (*Chem 1-Bio-Sac*) with 5.88 and 5.83 t·ha⁻¹ respectively.

Even with practically the same GY, in Lot 1, it was *Sac* that obtained the maximum HI (0.49) whilst *Chich* had 0.43. No effect on HI was found when applying *Bio* to *Chich* as *Sac* showed. The lowest HI of both varieties was found in the *Control* and those treatments with half N.

In Lot 2, *Sac* continues with the general trend of having the highest PB (6.6 t·ha⁻¹ vs. 6.0 t·ha⁻¹) with practically the same GY (4.63 t·ha⁻¹) as *Chich* (4.84 t·ha⁻¹). However, *Chich*, and no *Sac*, showed the best HI (0.46) performance with T5 (*Chem 1-Bio-Chich*).

It seems that the GY of *Sac* can be more predictable if associated with the PB production due to the high Coefficient of Determination (R²) in both lots.

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Conflicts of Interest

The authors declare no conflicts of interest.

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