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Yield response of a polycropping system with maize to fermented foliar fertilizers

Efecto de biofertilizantes foliares sobre el rendimiento de un policultivo con maíz

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ABSTRACT:

Anaerobically fermented foliar fertilizers (FFF) are made of local plant or animal inputs. The impact of four different FFF formulations on the output of an intercropping system with fava bean, maize, and squash was assessed in the Toluca Valley, Central Mexico. A fertilizer made of cow manure and fermented agave cactus juice had an elevated N and P content and resulted in the highest maize output. A fertilizer made of cow manure and maize stalks contained most K and delivered the highest squash yield. A FFF made of cow manure and stinging nettle delivered the highest fava bean output. There are no ideal formulations of FFF, but their formulation depends on available resources and treated crops.

KEYWORDS: Organic plant nutrition, biofertilizer, anaerobic digestion, sustainable horticulture, Mexico.

RESUMEN:

plantas o animales. En el valle de Toluca se evaluó el impacto de cuatro diferentes BFF sobre un policultivo con maíz, haba y calabaza. De todos los BFF comparados, el elaborado de estiércol de ganado y pulque mostró el mayor contenido de N y P y resultó en la mayor cosecha de maíz. Un fertilizante hecho de estiércol de vaca y mazorcas de maíz mostró más K y estimuló el rendimiento de la calabaza. Otro BFF, elaborado de estiércol de vaca y ortiga picada, generó la mayor cosecha de haba. No existen recetas estándar para los BFF, sino su formulación apropiada varía según los recursos disponibles y los cultivos tratados.

PALABRAS CLAVE: nutrición vegetal orgánica, biofertilizante, horticultura sustentable, México.

1. Introduction

1. 1. Definition

Fermented foliar fertilizers, FFF (also known as fermented leaf fertilizers, FLF), describe anaerobically digested animal and plant residues used as organic foliar fertilizers. Essentially, FFF nourish crops directly (Ebel & Kissmann, 2019). In Latin America, FFF are widely called *biol.*, although this term is also used for the (partially overlapping) concept of bio-fertilizers, which do not directly provide nutrients to the plants but increase the soil fertility through an enhancement of the soil microorganism activity (Chojnacka, 2015).

The use of FFF is viable for all kinds of annual and perennial crops (Galindo *et al.*, 2007). Hence, their use in horticultural production is most common. Increased yields because of the use of FFF are proved for bean (Bejarano & Méndez, 2004), maize (Vázquez *et al.*, 2014), and spinach (Siura *et al.*, 2009). If FFF are used, compared to conventional solid organic fertilizers, there is a remarkable immediate nutrient supply, high enough to meet the micronutrient demand of numerous horticultural and ornamental plants (Burnett *et al.*, 2016). These fertilizers are normally produced on-farm (Ito, 2006). Therefore, their composition is not specified but depends on the resources available on a farm and the demands of the therewith treated crops.



1.2. Formulation

Despite a diverse potential resources for generating FFF, there are four principal components every FFF consists of: Organic nutrient sources, energy sources, microorganism sources (inoculants), and water. Concerning nutrient sources, cattle manure is a common source. Potential alternatives are manures of other animals as well as vegetative nutrient sources such as stinging nettle, fruit mash, pods of legumes, oat and wheat groats, arnica, or comfrey. Pulverized charcoal can be added to nourish microorganisms. For energy sources, cane molasses or unrefined sugar are the standard ingredients. Alcoholic beverages may be used as well. Fresh cow milk is the most common microorganism source. Further options include forest humus, compost, fermented food, rice, and corncobs. Vegetative ash or rock flour may be added to raise the nutrient content. The addition of lime ensures a neutral pH (Ebel, 2017).

The most basic FFF formulation, called from here on the standard formulation, consists of 50 kg fresh cow manure, 4 L fresh cow milk, 4 L cane molasses, and 150 L rainwater, which are anaerobically digested to produce approximately 200 L FFF (Table 1).

TABLE 1 Nutrient content of standard FFF formulations after a minimum of 30 days of digestion

Formulation	N (a)	P	K	Ca	Mg	Fe	Mn	Cu	Zn	Reference
	Ppm									
S (b)	2 000	733	4 531	2 042	486					(Ito, 2006)
SSY (c)	818	93	2 037	191	483		4	1	1	(Galindo et al., 2007)
P (d)	1 200	50	340				<10		<10	(Lee et al., 2012)
SA(e)	480	28	1 651	978	348	8				(Zagoya et al., 2015)

Source: own elaboration.

Note: (a) Total N; (b) Standard formulation; (c) Standard formulation, salts and yeast added; (d) FFF made of pig manure; (e) Standard formulation, ash added.

1. 3. Preparation and application

Usually, self-made biodigestors (made of adapted barrels) are used to mineralize the nutrients in the raw material, but large-scale industrial production is conceivable. Before digestion, all vegetative material must be shredded to a length of a maximum of 2 cm. The biodigestor is then filled with the shredded material, the fresh animal excrements, and 100 L water. Ash may be added. Simultaneously, the energy sources and the milk are mixed with 20 L water in a separate container and subsequently added to the digestor. Afterward, the barrel is filled with water (leaving enough space for the digestion gases) and locked (Ebel & Kissmann, 2019). The digestion process lasts two to eight weeks (Ito, 2006). During digestion, the bio-digestor must be protected from sunlight as it stimulates the oxidation of the organic compounds (Restrepo & Hensel, 2009). After locking the digestor, aerobic microorganisms cause a quick consumption of the remaining oxygen, and anaerobic digestion starts immediately (Siura *et al.*, 2009). Thereby, organic substances are converted into humic and non-humic biomass, minerals, and volatile inorganic compounds (Gerardi, 2003). Before applying them, FFF must be diluted 1:20 with water. During a crop cycle of rainfed bean and maize production, six to eight applications of FFF are recommended (Restrepo & Hensel, 2009).



1. 4. Nutrient content and absorption

There is evidence that leaves can absorb inorganic nutrients supplied in aqueous forms (Kannan, 2010). Foliar-applied nutrients are lipid-insoluble ions and, therefore, enter the plant metabolism following an aqueous pathway through a leaf's cuticular wax or the stomata. The ultimate parameter controlling the penetration of foliar-applied substances is the intrinsic permeability of the leaf surface. This is a passive process driven by concentration gradients (Fernández & Eichert, 2009) and stimulated by light and soil moisture (Fageria *et al.*, 2009).

A standard FFF formulation (Table 1) contains 2 000 ppm N, 733 ppm P, and 4 531 ppm K. Most FFF show a slightly acidic pH (Zagoya *et al.*, 2015). The electric conductivity of FFF is around 5 dS m⁻¹ (Orellana *et al.*, 2013).

2. Justification

Although conventional monocropping is economically predominant, traditional polycropping still characterizes small-scaled farms in central Mexico. Apart from the traditional Mesoamerican *milpa* polycropping system (maize, bean, and squash), the polycropping of maize, fava bean, and squash is common (Ebel *et al.*, 2017). Increased agrobiodiversity of a production system has multiple benefits: increased yield; a longer harvest period; a diversified production; time-efficient crop management; stimulation of the soil microflora (Gliessman, 1985); the maximization of the exploitation of available nutrients and water; sequestration of atmospheric carbon; as well as improved resilience to pests and meteorological perturbations (Altieri, 1994). In short, polycropping is an excellent approach to increase the sustainability of farming systems.

The use of FFF is an equally sustainable plant nutrition strategy. Although FFF have been used in crops such as maize and legumes (Zagoya *et al.*, 2015), which are frequently intercropped, evidence of their use in polycropping systems is scarce. Thus, FFF are prevailingly used in monocropped vegetables (Ito, 2006). Furthermore, the sustainability of FFF is based on the use of locally available resources. As FFF are most common in the Latin American tropics, common formulations are based on resources (such as molasses), which are not available in the template Central Mexican highlands or similar climates.

The present trial deals with FFF made of resources available in the Toluca Valley, located in the Mexican Highlands. It assesses the impact of their application on the yield and growth of a polycropping system consisting of maize, fava beans, and squash. Fava beans were chosen due to their economic significance in the region as well as their adaptation to the prevailing template climate.

3. Materials and methods

3. 1. Location

The trial was implemented in 2016 at the Campus El Cerrillo Piedras Blancas of the Autonomous University of the State of Mexico (92° 42' E and 19° 24' N) in the Valley of Toluca, central Mexico, 18 km north of the city of Toluca, at an altitude of 2 611 m above sea level. There is a dry-winter highland climate (Instituto Nacional de Estadística y Geografía, 2018). In the city of Almoloya de Juárez, located 9.5 km from the place of the experiment, the average temperature is 13.3° C and the annual precipitation 744 mm (Comisión Nacional del Agua, 2018). Annual frost is an environmental limitation for agriculture in this area, where rainfed maize is the most common cropping system. The area lies on extrusive igneous rocks. The prevailing soil is vertisol.



3. 2. Experimental set-up

There were six treatments with eight replications each; hence, the field was divided into 48 plots of 100 m², arranged in a randomized block system. The treatments differed in terms of fertilization: After initial incorporation of cow manure to the entire field four weeks before seeding, different FFF were applied in four treatments; a further treatment was fertilized with bokashi, a solid organic fertilizer; the control treatment did not receive additional fertilization management.

3. 3. Formulation and preparation of examined fertilizers

The preparation of all four experimented FFF was based on the standard formulation as defined by Restrepo & Hensel (2009), which was modified based on farmer recommendations regarding the availability of the required resources in the region (Table 2).

TABLE 2
Raw material used for the preparation of FFF compared in the trial

	Nutrient and micro- organism sources (a)	Energy sources	Inoculants	Sources of organic matter	Additional ingredients
B1	Cow manure, ash	Unrefined brown sugar	Milk	Maize stalks	Rainwater
B2	Cow manure, ash	Pulque (b)	Oat bran, yeast	Forestal soil, oat straw, charcoal	Rainwater
В3	Cow manure, stinging nettle	Refined sugar	Milk, beer	Forestal soil, maize stubbles	Lime, rainwater
B4	Chicken dung	Unrefined brown sugar	Milk, yeast	Fruit waste (c)	Rainwater

Source: own elaboration.

Note: (a) The categorization is approximate: cow manure, for example, is primarily a source of nutrients but also provides microorganisms and organic matter; (b) Fermented agave cactus juice; (c) Diverse fruits (except citrus).

For all experimented FFF (Table 3), fresh animal excrements and fresh milk were used. Maize stalks and stubbles, as well as oat bran and straw, were ground before being added to the fermenter. Fresh stinging nettle (only leaves) and the fruit waste were squashed manually to a size of a maximum of 2 cm. The charcoal was smashed, and the soil was sieved before adding. Yeast and sugar were each dissolved in 2 L warm water before adding them to the respective FFF. Ash and lime were both dissolved in 4 L rainwater. Immediately after fermentation, they were added to the FFF.

TABLE 3
Amounts of resources used for the preparation of 1 L of the experimented FFF

					1 1		1	
	Manure	Ash (a)	Sugar	Cow milk	Other liquids	Herbal inputs	Other inputs	Rain-water
	g L ⁻¹	g L ⁻¹	g L-1	ml L−1	ml L−1	g L ⁻¹	g L ⁻¹	ml L−1
В1	250 (b)	20	20 (c)	20		25 ^(d)		900
В2	250 (b)	20			10 (e)	25 (f), 15 (g)	5 (h), 5 (i), 5 (j)	900
В3	150 (b)		20 (k)	10	10 (1)	100 ^(m) , 25 ⁽ⁿ⁾	10 (j),10 (o)	900
B4	75 (p)		10 (c)	20		225 (q)	5 (h)	900

Source: own elaboration.

Note: (a) Added after fermentation; (b) Fresh cow manure; (c) Unrefined brown sugar; (d) Maize stalks; (e) Pulque; (f) Oat bran; (g) Oat straw; (h) Yeast; (i) Charcoal; (j) Soil; (k) Refined sugar; (l) Beer; (m) Stinging nettle; (n) Maize stubbles; (o) Lime; (p) Chicken dung; (q) Fruit waste.



All four FFF were digested in airtight barrel-like containers of 200 L with an airlock. After adding all ingredients, the barrels were filled with rainwater and locked. Twenty-one days afterward, the barrels were opened for a few minutes to check the consistency, color, smell as well as the temperature. Subsequently, they were stirred. This procedure was repeated weekly for five to nine weeks (depending on the formulation): B1 and B2 were ready after 39 days, B3 after 46 days, and B4 after 60 days of fermentation. The following criteria were applied to identify a completely fertilized product: Vastly liquid (and not viscous) consistency, the absence of visible solid elements, widely translucent coloring, and smell like silage.

Simultaneously, a solid fermented organic fertilizer (bokashi) was elaborated. 500 kg bokashi were produced of 200 kg fresh sheep manure collected from local stables; 5 kg unrefined brown sugar; 150 kg maize stubbles; 50 kg maize stalks; 200 kg approved forestal soil; 50 kg charcoal; and 1 kg yeast. Water was added as needed to moisturize the bokashi entirely.

3. 4. Agrobiodiversity and crop management

As common in traditional polycropping arrangements in central Mexico, maize (landrace 'Criollo Blanco'), fava bean ('San Pedro Tlaltizapan'), and a squash landrace ('Criollo de guia') were seeded simultaneously. Maize and fava bean were each seeded at a density of 20 000 plants ha⁻¹, squash at a density of 5 000 plants ha⁻¹.

All plots were exclusively rainfed. There were no explicit disease control measures. Weeds were removed manually, and pests were monitored with a yellow sticky trap (Table 4).

TABLE 4
Timetable of crop preparation, management, and harvest activities

Date	Activity
20.02.2016	Elaboration of FFF and bokashi
22.02.2016	Incorporation of well-rotted cow manure
03.03.2016	Application of bokashi
17.03.2016	Seeding of maize, fava bean, and squash
01.04.2016 - 01.08.2016	Two-weekly application of FFF (total of 9 applications)
06.06.2016 + 25.06.2016	Application of garlic tea (organic pesticide)
27.06.2016 - 04.09.2016	Harvesting of squash (9 harvestings)
10.08.2016 - 15.09.2016	Harvesting of fava beans (4 harvestings)
31.08.2016 - 25.11.2016	Harvesting of maize (8 harvestings)

Source: own elaboration.

The following pests were observed: *Agriotes* sp., *Aphis fabae* Scopoli, *Apion godmani* Wagner, *Chaetocnema pulicaria* F. E. Melsheimer, *Dalbulus maidis* De Long, *Diabrotica longimaizeis* Say, *Helicoverpa zea* Boddie, *Helix aspersa* Müller, *Macrodactylus* spp., *Phyllophaga* spp., *Rhopalosiphum maidis* Fitch, *Sphenophorus* spp., and *Spodoptera frugiperda* Walker (all at minor incidence). Only *Diabrotica undecimpunctata* Mannerheim showed a precariously growing population on fava beans, which is why it had to be controlled with organic pest management consisting of a tea of garlic and wood ash, which was applied twice (dose: 2 L ha⁻¹ per treatment at a dilution of 1:50 with rainwater) in June. Thanks to the use of this organic pesticide and due to increasing precipitations in summer, no additional pest control measures were necessary.

With the onset of the raining season in June, the following diseases were observed: Common rust (*Puccinia sorghi* Schweinitz) with maize; chocolate spot disease (*Botritis fabae* Sardiña), fusarium wilt (*Fusarium* spp.), and *Peronospora viciae* de Bary with fava bean; as well as *Sphaerotheca* sp. with squash (all at minor incidence). At the end of the harvesting period, faba bean rust, *Uromyces fabae* (Pers.) Schröt, seriously affected fava beans but did not harm the output.



As for weeds, the following species were identified: *Medicago lupulina* L. *Medicago polymorpha* L., *Melilotus indica* (L.) All., *Trifolium repens* L., Trifolium pratense L. (legume species, which were partially tolerated), *Asclepias linaria* Cav., *Brassica nigra* (L.) W. D. J. Koch, Brassica rapa L., *Commelina erecta* L., *Lopezia racemosa* Cav., and *Rumex crispus* L. (observed at high density); as well as *Avena fatua* L., *Bidens odorata* Cav., *Calandrinia micrantha* Schltdl., *Chenopodium album* L., *Hilaria cenchroides* Kunth, *Phalaris minor* Retz., *Sinapis alba* L., and *Taraxacum* spp. (at minor incidence).

To provide basic fertilization for germination and initial vegetative growth, 23 days before seeding, a basic fertilization of 10 t ha⁻¹ rotted cow manure was incorporated at a depth of 15 cm. In the treatment with bokashi, additional 10 t ha⁻¹ of bokashi were incorporated 14 days before seeding.

Before their application, all FFF were diluted 1:20 with rainwater. Fourteen days after seeding (DAS), the respective treatments received the first application of the liquid fertilizers. This was repeated eight times in a two-week interval. Through these applications, each plant received approximately 100 ml of the correspondent FFF.

3. 5. Response variables and statistical analysis

The following variables were evaluated in the analysis of the FFF and the bokashi: pH; electric conductivity; organic matter; and content of C, N, P, K, Ca, Mg, Na, and Fe. The carbon content was determined using dry combustion. Total Kjeldahl N was determined spectrophotometrically using chromotropic acid for extraction. P was also analyzed with a spectrophotometer. K (wavelength: 766 nm) and Na (589 nm) were determined using a flame photometer. Standard solutions were prepared by the dilution of stock solutions containing K⁺ and Na⁺ respectively. For the analysis of Ca (423 nm), Mg (285 nm), and Fe (510 nm), an atomic absorption photometer with direct aspiration was used. For the analysis of Ca and Mg, standard solutions with Ca²⁺, Mg²⁺, and lanthanum chloride were used. Regarding Fe, diluted nitric acid was added instead of lanthanum chloride (Secretaría de Medio Ambiente y Recursos Naturales, 2002).

In the case of maize, the weight of fresh cobs was measured 135 and 200 DAS. 200 DAS, the number of cobs and leaves per maize plant as well as the plant height were also determined. Fresh pods of the fava bean were weighed at 70, 90, and 110 DAS. The plant height and the number of leaves per fava bean plant were determined at 154 DAS. For squash, the number of flowers per plant was measured ten times between 30 and 178 DAS and then summarized. For all crops, the total yield per area was determined. Maize was harvested nine times between 135 DAS and 220 DAS. Fava bean was harvested four times in a biweekly interval, starting at 160 DAS and ending at 202 DAS. Squash was harvested eight times, 45 to 178 DAS (Table 4).

Additionally, the Land Equivalent Ratio (LER) was calculated. It refers to the monoculture area required to generate the same yield as in 1 ha of the analyzed polyculture system and provides a standardized basis to compare monocropping and polycropping systems:

$$LER = \sum_{i=1}^{n} \frac{YP_i}{YM_i},$$

where n corresponds to the number of associated crops, YP to the yield per hectare of a crop in polyculture and YM to the yield of the same crop in monoculture (grown under similar conditions). An LER > 1.0 expresses a productive advantage of a polyculture. Maize, fava bean, and squash yields in nearby low-input monocropping systems were used as reference values.

The normality of distribution of means was tested using the Shapiro-Wilk test. The homogeneity of variances was assessed with the Levene test. Significant differences were determined using a one-way ANOVA and post-hoc analysis with a Tukey test ($p \le 0.05$) for the following parameters: total yield per



area of each crop, fruit weight of each crop at different stages, cobs per plant (maize), flowers and harvestable fruits per plant (squash), plant height (maize and fava bean), and the number of leaves (maize and fava bean).

4. Results

4. 1. Analysis of FFF

As Table 5 shows, B1, B3, and B4 were slightly alkaline, and B2 was strongly alkaline. The four applied FFF showed a salinity between 0.13 dS cm⁻¹ (B3) and 1.02 dS cm⁻¹ (B2). With 2306 ppm, B2 showed the highest mineral N content. Regarding P, Ca, and Mg content, B2 was also among the most nutritive treatments, while B1 showed the highest K content.

TABLE 5 Chemical properties and composition of the examined fermented liquid fertilizers (B1-B4) and the bokashi (BO)

	рН	C org	OM (a)	EC (p)	И (c)	P
		%	%	dS cm ⁻¹	ppm	
BO (d)	8.27 ± 0.21	33.54 ± 2.1	29.13 ± 3.5	9.05 ± 0.07	$19\ 100 \pm 674$	$4\ 404 \pm 111$
B1	$7.68 \pm 0.14 b^{(e)}$	$0.49 \pm 0.03 \text{ a}$	$32.1 \pm 1.1 \text{ a}$	$0.82 \pm 0.01 \text{ b}$	$2\ 101 \pm 103\ a$	$2709 \pm 806 a$
B2	$9.21 \pm 0.09 \text{ a}$	$0.17 \pm 0.02 \text{ b}$	$24.2 \pm 5.6 \text{ b}$	1.03 ± 0.04 a	$2\ 306 \pm 87\ a$	$2\ 306 \pm 717\ a$
В3	$7.33 \pm 0.02 \text{ b}$	$0.46 + \pm 0.09$ a	$12.4 \pm 7.0 \text{ b}$	0.31 ± 0.06 c	$1\ 288 \pm 205\ b$	$949 \pm 52 \text{ b}$
B4	7.52 ± 0.12 b	$0.33 \pm 0.1 a$	$2.5 \pm 4.3 \text{ c}$	$0.13 \pm 0.1 d$	$132 \pm 87 c$	$113 \pm 29 c$
	K	Ca	Mg	Na	Fe	
	K 	Ca 				
BO (d)			Mg	Na	Fe	
BO (d) B1			Mg 	Na 	Fe	
	 30 963 ± 1 385	 22 014 ± 1 400	Mg 2 097 ± 88	Na 1 292 ± 44	Fe 1 769 ± 31	
В1	30 963 ± 1 385 4 493 ± 103 a	22 014 ± 1 400 3 716 ± 122 b	Mg 2 097 ± 88 1 499 ± 96 a	Na 1 292 ± 44 12 ± 2 a	Fe 1 769 ± 31 0.49 ± 0.02 a	

Source: own elaboration.

Note: (a) Organic matter; (b) Electric conductivity; (c) Total Kjeldahl N; (d) Since (compared to FFF) bokashi is characterized by a different consistency and preparation process, it was not considered in the statistical analysis; (e) Different letters in the same column indicate significant differences based on a one-way ANOVA and a post-hoc analysis using Tukey-Test ($p \le 0.05$).

4. 2. Growth and yield performance

The highest maize yield was obtained in treatment B2 (Table 6). Since there were minor differences regarding fava bean and squash yield among all treatments, maize output was decisive for the total yield performance of the polycropping system. B2 consequently provided the highest *LER* of 1.26, while the other treatments showed *LER* below 1.0 (Table 7).



TABLE 6
Total maize yield per area, weight of single corncobs, number of cobs per plant, plant height, and number of leaves per maize plant in control treatment (c), and treatments with four different fermented liquid fertilizers (B1-B4) and with bokashi (BO)

	Total yield	Fruit weight 135 DAS ^(a)	Fruit weight 200 DAS	Cobs per plant	Plant height 200 DAS	Number of leaves 200 DAS
	kg ha ^{–1}	g	g	g	cm	
С	$1766 \pm 44 \ c^{(b)}$	$66 \pm 31 \text{ c}$	$189 \pm 9 \text{ a}$	$1.13 \pm 0.11 \text{ b}$	$130 \pm 3 \text{ b}$	6 ± 0 a
ВО	$1636.4 \pm 31 \text{ c}$	$53 \pm 2 c$	$182 \pm 31 \text{ a}$	$1.09 \pm 0 \text{ b}$	$125 \pm 4 b$	7 ± 0 a
B1	$2261.4 \pm 56 b$	104 ± 13 bc	$229 \pm 18 \text{ a}$	$0.79 \pm 0.08 \text{ b}$	$131 \pm 2 b$	$7 \pm 1 a$
B2	$4788.9 \pm 167 \text{ a}$	$180 \pm 4 \text{ ab}$	$171 \pm 9 \text{ a}$	$1.02 \pm 0.01 \text{ b}$	$154 \pm 4 a$	$6 \pm 1 a$
B3	$2217.5 \pm 102 \text{ b}$	$232 \pm 14 a$	$195 \pm 15 \text{ a}$	$2.42 \pm 0.03 \text{ a}$	$150 \pm 5 a$	6 ± 0 a
B4	$1535.4 \pm 90 \text{ c}$	$122 \pm 8 b$	$126 \pm 14 b$	$0.77 \pm 0.02 \text{ b}$	$156 \pm 3 \text{ a}$	6 ± 0 a
p	0.027	0.043	0.039	0.012	0.037	0.758

Source: Own elaboration.

Note: (a) Days after seeding; (b) Different letters in the same column indicate significant differences based on a one-way ANOVA and a post-hoc analysis using Tukey-Test ($p \le 0.05$).

TABLE 7
Total maize, fava bean, and squash yield and consequent land equivalent ratio (based on referential monocropping yields) in control treatment (c) and treatments with four different fermented liquid fertilizers (B1-B4) as well as bokashi (BO)

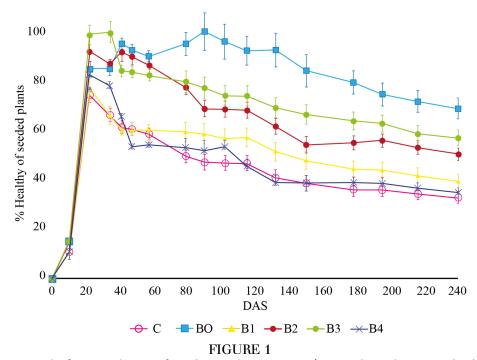
Treatment	Maize total yield	Fava bean total yield	Squash total yield	Land Equivalent Ratio
	kg ha ⁻¹	kg ha⁻¹	kg ha ^{–1}	
С	1766	1330.4	3670.2	0.90
ВО	1636.4	740.3	2129	0.58
B1	2261.4	633.35	5677	0.98
B2	4788.9	1397.4	3809	1.26
B3	2217.5	1741.97	2085	0.89
B4	1535.4	1194.75	1486	0.62
Reference Yield (a)	9000	4000	10000	

Source: Own elaboration.

Note: (a) The reference yield considers the monocropping output of the respective crops (same varieties, similar crop management) in central Mexico.

Maize germinated earliest in B3 (Figure 1). 135 DAS, B3 also showed the highest cob number and weight. Gradually, more and more plants died in this treatment. 240 DAS, 43% of the maize plants in B3 had died. In contrast, bokashi resulted in most vital maize plants 240 DAS. Yet, its yield performance was poor (Table 7). From 30 DAS, B1 and B4 showed a continuously declining maize population. B1 and bokashi resulted in shorter maize plants compared to treatments B2, B3, and B4 (Table 6).

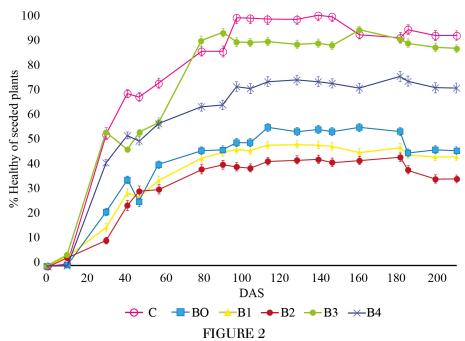




Survival of maize plants referred to each treatment (scatter bars show standard deviation): B3 caused the fastest germination, bokashi the highest survival rate Source: own elaboration.

Regarding fava bean, the control treatment was the fastest germinating and resulted in most living plants (Figure 2) as well as in the highest number of leaves per plant (Table 8). B3 showed a similar survival rate but generated a higher yield than B2 due to the production of the heaviest pods of all treatments. The other treatments underperformed B2 and B3 in all parameters.





Survival rate of fava bean plants referred to each treatment (scatter bars show standard deviation): The control treatment caused the fastest germination; together with treatment B3, it also showed the highest survival rate 240 DA Source: own elaboration.

TABLE 8
Total yield per area, weight of single pods, plant height, and number of leaves of fava bean in control treatment (c) and treatments with four different fermented liquid fertilizers (B1-B4) as well as bokashi (BO)

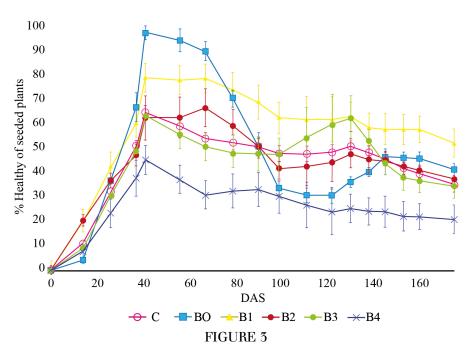
Treatmen	t Total yield (fresh pod)	Fresh pod weight 70 DAS ^z	Fresh pod weight 90 DAS	Fresh pod weight 110 DAS	Plant height 154 DAS	Number of leaves 154 DAS
	kg ha ⁻¹	g	g	g	cm	
С	$1\ 330.4 \pm 117\ ab^{(b)}$	16.45 ± 0.43 a	$15.28 \pm 0.94 \text{ ab}$	17.02 ± 0.06 ab	94 ± 2 ab	20.43 ± 1.03 b
ВО	$740.3 \pm 39 \text{ b}$	17.96 ± 0.95 a	$18.02 \pm 0.29 \text{ a}$	$15.96 \pm 0.41 \text{ ab}$	$81 \pm 6 b$	$15.75 \pm 1.11 \text{ c}$
B1	$633.35 \pm 44 \text{ b}$	$12.53 \pm 0.8 \text{ a}$	$13.84 \pm 0.11 \text{ b}$	$15 \pm 0.11 \text{ ab}$	$83 \pm 1 b$	$17.04 \pm 0.99 \mathrm{b}$
B2	$1397.4 \pm 86ab$	13.58 ± 0.61 a	15.47 ± 0.66 ab	$15.31 \pm 0.45 \text{ ab}$	111 ± 5 a	30.42 ± 1.56 a
B3	$1.741.97 \pm 75 a$	19.31 ± 0.31 a	$17.31 \pm 0.51 \text{ ab}$	$19.32 \pm 1.18 a$	$104 \pm 2 a$	$21.4 \pm 0.86 b$
B4	$1\ 194.75 \pm 224\ ab$	18.23 ± 0.16 a	$13.39 \pm 0.21b$	$14.08 \pm 0.7 \ b$	$95 \pm 7 \text{ ab}$	$17.25 \pm 0.29 \text{ b}$
p	0.054	0.027	0.038	0.26	0.013	0.044

Source: own elaboration.

Note: (a) Days after seeding; (b) Different symbols in the same column indicate significant differences based on a one-way ANOVA and a post-hoc analysis using Tukey-Test ($p \le 0.05$).

The survival rate of squash was generally low: 180 DAS, B1 provided 51% vital squash plants, while the other treatments showed values below 50% (Figure 3). As shown in Table 9, B1 produced the highest squash yield. B1 and the control treatment resulted in most flowers per plant.





Survival rate of squash plants referred to each treatment (scatter bars show standard deviation): The use of bokashi resulted in the fastest germination. Treatment B1 showed the highest survival rate 240 DAS Source: own elaboration.

TABLE 9

Total yield per area, weight of single fruits, number of flowers, and number of harvestable fruits per plant of squash in a control treatment (c), and treatments with four different fermented liquid fertilizers (B1-B4) as well as bokashi (BO)

	Total yield	Fruit weight	Flowers per plant	Harvestable fruits per plant
	kg ha ⁻¹	g		
C	$3670.2 \pm 87 \ b^{(a)}$	$244 \pm 11b$	36 ± 1^a	$21 \pm 3 a$
ВО	$2129.9 \pm 144 \text{ b}$	$195 \pm 10 \text{ b}$	17 ± 0.2 c	$13 \pm 2 b$
B1	$5677.6 \pm 309 a$	$262 \pm 53 \text{ ab}$	31 ± 3 ab	$23 \pm 1 a$
B2	$3800.9 \pm 284 \text{ b}$	$357 \pm 40 \text{ a}$	$24 \pm 0.2 \ b$	$15 \pm 0.1 \text{ b}$
В3	$2085.3 \pm 140 \text{ b}$	$231 \pm 21 \text{ b}$	$25 \pm 2 b$	$15 \pm 2 b$
B4	1486.2 ± 365 c	$223 \pm 8 b$	$21 \pm 0.1 b$	$11 \pm 2 b$
p	0.038	0.072	0.009	0.045

Source: own elaboration

Note (a) Different symbols in the same column indicate significant differences based on a one-way ANOVA and a post-hoc analysis using Tukey-Test ($p \le 0.05$).

5. Discussion

5. 1. Analysis of FFF

Although FFF usually have a pH of around 6.0 (Zagoya *et al.*, 2015), all analyzed FFF were alkaline (Table 5). The elevated pH in B1 and B2 is related to the used wood ash originating from the domestic fuel of pine wood. Such ash is commonly alkaline, especially if the burning temperatures are not extremely high



(Etiégni & Campbell, 2005). Charcoal made of pine wood (as used in B2) is a further trigger of alkalinity (Briggs, 2005). In the case of B3, high pH is related to the use of lime (Arman & Munfakh, 1970). In B4, the used chicken manure increased the pH (Wang *et al.*, 2012). The pH of 9.2 measured in B2 requires special attention because such elevated alkalinity decreases the microbial activity. Likely, anaerobic digestion resulted in accumulation of alkaline biogenic amines (Chen *et al.*, 2008). Unique ingredients of B2 were *pulque*, oat bran, oat straw, and charcoal.

B1, B2, and B3 showed a higher EC than common in FFF (Orellana *et al.*, 2013), which is prevailingly related to the high nutrient content of these treatments (Tables 1, 5).

B2 showed the highest, B1 the second-highest N content. The same amount of cow manure was used in both treatments. The main difference between B1 and B2 consisted in the used energy sources and inoculants: unrefined brown sugar was used as the energy source in B1, *pulque* in B2; respectively, milk was the inoculant in B1, while oat bran and yeast were used in B2. The total raw protein content of these ingredients is 131.8 g L⁻¹ unfermented raw material in B2, but only 12.56 g L⁻¹ in B1 (De Leon *et al.*, 2005), which explains the higher N content of B2. Denitrification is a potential trigger of N losses but was not assessed.

Concerning P content, B1 and B2 also outperformed the other treatments. The high content of mineralized P is an indicator of a complete fermentation process (Galindo *et al.*, 2007). Cow manure was the most significant P source in both treatments. Additionally, the ash added to B1 and B2 supplied 8.4 g kg⁻¹ P but also 0.6 g kg⁻¹ Na (Misra *et al.*, 1993), which explains why these treatments contained most Na.

Apart from cattle manure and wood ash, fermented maize stalks are an important K source (Darwish *et al.*, 2012). Consequently, B1 showed the highest K content. Finally, B2 contained most Ca and Mg, which is related to the use of ash and charcoal (Briggs, 2005).

5. 2. Growth and yield performance of maize

Since young leaves are more penetrable for nutrients than fully-expanded ones (Fernández & Eichert, 2009), the application of FFF affected the vegetative development of all crops positively at an early stage. A quick vegetative growth (enabled through foliar N-input) is the base for a satisfactory maize output (Aguilar *et al.*, 2015). The foliar supply of P at the final stage of vegetative growth is a further stimulus of a maize output (Fageria *et al.*, 2009). Consequently, B2, the treatment that contained most N and P, provided the highest maize yield (Table 6).

Fertilization with bokashi resulted in more surviving maize plants than when FFF were used, which indicates a tendentially limiting effect of FFF on the maize survival rate. However, bokashi did not result in a satisfactory yield. This circumstance is mainly attributed to a low early maize yield (Table 6), a consequence of a low N and P supply during the initial growth stages.

The remarkable low survival of maize plants treated with B1 is attributed to the elevated K content of this FFF, which decreased the availability of other nutrients (Restrepo & Hensel, 2009). The equally low maize survival rate of B4 is related to generally low nutrient supply. Accordingly, plants treated with B4 showed the lowest cob weight.

5. 3. Growth and yield performance of fava bean

The yield response of the fava bean was relatively constant among all treatments, indicating that this legume (compared to maize and squash) demands less additional fertilization measures in a polycropping arrangement (Table 8).

Although B2 and B3 showed a similar fava bean survival rate, B3 produced heavier pods and a consequently higher yield. There is evidence that rhizospheric salinity affects the association between a fava bean and N-



fixing bacteria and that this condition favors the accumulation of macronutrients in the vegetative organs instead of the fruits (Cordovilla *et al.*, 2008). Simultaneously, salinity decreases the overall K, Mg, Ca, and S content of fava beans (Abdelhamid *et al.*, 2010). There is no evidence whereby the salinity of organic foliar applications has similar consequences. However, salinity may explain why B3 (with a lower EC and significantly less Na) yielded more fava bean than B2 (which was richer in N and P).

Additionally, B2 stimulated a quick maize growth, which resulted in stronger competition for sunlight between maize and the (shorter) fava beans. Since sunlight stimulates the stomatal opening (Fernández & Eichert, 2009), the higher the maize plants grew, the lower was the efficiency of the FFF applied to the fava beans.

5. 4. Growth and yield performance of squash

Competition for light affected the squash yield, causing a generally low survival rate of this crop. Only B1 provided more than 50% vital squash plants and was consequently the highest yielding treatment. Of all experimented FFF, B1 contained most K; and for cucurbits, there is evidence of a satisfactory yield response to foliar K supply (Bejarano & Méndez 2004).

5. 5. Land Equivalent Ratio

While all other treatments resulted in less yield than comparable monocropping arrangements with each, maize, squash, or fava bean (*LER* below 1.0), B2 provided 26% more output than comparable monocropping systems (Table 9). This was caused by high maize yield of B2 of 4788.9 kg ha⁻¹. Regarding fava bean and squash, additional FFF applications may have increased their competitiveness against maize and consequently provided higher yields (Bejarano & Méndez, 2004).

PROSPECTIVE ANALYSIS

Fermented foliar fertilizers, together with biofertilizers, are already standard practice in small-scale farms in tropical regions of South American countries such as Ecuador. The results of this trial can contribute significantly to the adoption of these techniques in other parts of the world, for example in Mexico. The significance of this trial lies in the exclusive use of resources available in a temperate climate, which amplifies the geographical applicability of FFF. The experiment proved the functionality of using resources such as fermented food (*pulque*) instead of the so far indispensable tropical molasses. Additionally, the trial centered on the intercropping of maize, a staple crop. Using FFF in both polycropping systems and staple crops is a new approach for this plant nutrition strategy, which is more common in horticultural monocropping. Due to the significance of polycropping maize in traditional Mesoamerican farming, FFF are a viable option for thousands of peasants in the Mexican highlands and similar climates.

Conclusions

The present trial assessed the effect of the use of four FFF made of resources available in template central Mexico on the yield and the vegetative development of a polycropping system consisting of maize, fava bean, and squash. Three out of four experimented FFF formulations (B1, B2, B3) showed satisfactory nutrient content. Especially B2, made of cow manure, ash, *pulque*, oat bran, and yeast, was outstandingly nutritive and consequently delivered the highest maize yield in the observed polycropping system. Remarkably, B2



did not include fresh cow milk, which is commonly used as an inoculant of FFF, but *pulque*. Accordingly, pulque is a promising and regionally available substitute for milk. Nevertheless, the considerable alkalinity of B2 requires further microbiological analysis. It was also shown that FFF rich in N and P had a positive impact on maize yield, which is expected to be related to the immediate supply of these nutrients during the vegetative growth stage of the crop. his opens the possibility of using FFF as a macronutrient source. Despite the sustainability of polycropping systems and their significance for Mexico, a lesson learned from this trial is that the assessment of new FFF formulations would be easier in monocropping arrangements due to crop interactions in polycropping. For example, B2 strongly stimulated maize growth and yield; yet, the sound maize growth affected the development of the associated crops (fava bean and squash), mainly due to competition for sunlight. Furthermore, it diverse crops responded differently to FFF application: Fava bean yielded most with B3 (made of stinging nettle and cow manure); squash delivered most yield with B1 (based on a standard formulation). This indicates the need for developing specific FFF formulations for each crop. Thanks to an uncomplicated preparation process and the use of locally available resources, FFF are a sustainable option for sustainable farmers. Due to their "regionality", there won't be a standard manual for the preparation of FFF. The successful use of FFF requires innovative researchers and farmers all over the world.

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