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AGRO PRODUCTIVIDAD

Practices and perceptions of fall
ARMYWORM
(*Spodoptera frugiperda* Smith)
and
STRIPED GRASS LOOPER
(*Mocis latipes* Guenée)
management in maize

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Contacto principal
Jorge Cadena Iñiguez
Guerrero 9, esquina avenida Hidalgo, C.P. 56220, San Luis Huexotla, Texcoco, Estado de México.
agroproductividadeditor@gmail.com

Contacto de soporte
Soporte
5959284703
agroproductividadsoporte@gmail.com

Directrices para Autores/as

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Agronomic characterization of pigmented native corn populations (*Zea mays* L.)

Sánchez-Ramírez, Francisco J.[1*]; Rincón-Sánchez, Froylán[1]; Ruiz Torres Norma A.[1,2]; Sánchez-Sánchez Graciela[1]

[1] Universidad Autónoma Agraria Antonio Narro.Departamento de Fitomejoramiento. Buenavista, Saltillo, Coahuila, México. C. P. 25315.
[2] Universidad Autónoma Agraria. Centro de Capacitación y Desarrollo en Tecnología de Semillas. Buenavista, Saltillo, Coahuila, México. C. P. 25315.
[3] Universidad Autónoma Agraria Antonio Narro. Maestría en Ciencias en Fitomejoramiento. Buenavista, Saltillo, Coahuila, México. C. P. 25315.
* Correspondence: javier.sanchez@uaaan.edu.mx

ABSTRACT

Objective: To characterize agronomically 52 populations of pigmented native corn (*Zea mays* L.) from Coahuila, Mexico, in order to identify varieties with outstanding agronomic potential and to establish an improvement program with potential for grain yield, with the ability to adapt, and superior nutritional bioactive content.

Design/Methodology: Two experiments were evaluated through an incomplete block design in lattice alpha arrangement, in two localities that are representative of the agricultural area of southeastern Coahuila: a) Yellow populations, mostly of Tuxpeño, Ratón and Tuxpeño Norteño; and b) Anthocyanin populations (blue, red and purple), represented primarily by Cónico Norteño, Elotes Cónicos and Ratón.

Results: There is an environmental effect that contrasts between localities, with differences in grain yield of up to 78%; 25 days of difference in flowering, 53 cm in plant height, and up to seven plants without cobs. The response was product of the agricultural potential in each locality. The outstanding yellow populations due to their superior yield expression in both localities were the landraces Tuxpeño: COAH068, COAH089, COAH177 and COAH215, and Celaya: COAH075. The outstanding anthocyanin populations were Ratón: COAH23 and COAH203; Elotes Cónicos: COAH246 and COAH019; and Elotes Occidentales: COAH021; in addition, the study found populations adapted to a locality.

Conclusion: Among the diversity of the pigmented native corn in Coahuila, there are populations with superior agronomic expression that can be the basis for improving the production, the nutritional quality of the grain, and therefore, of its byproducts.

Keywords: *Zea mays* L., carotenoids, anthocyanins, nutritional quality.

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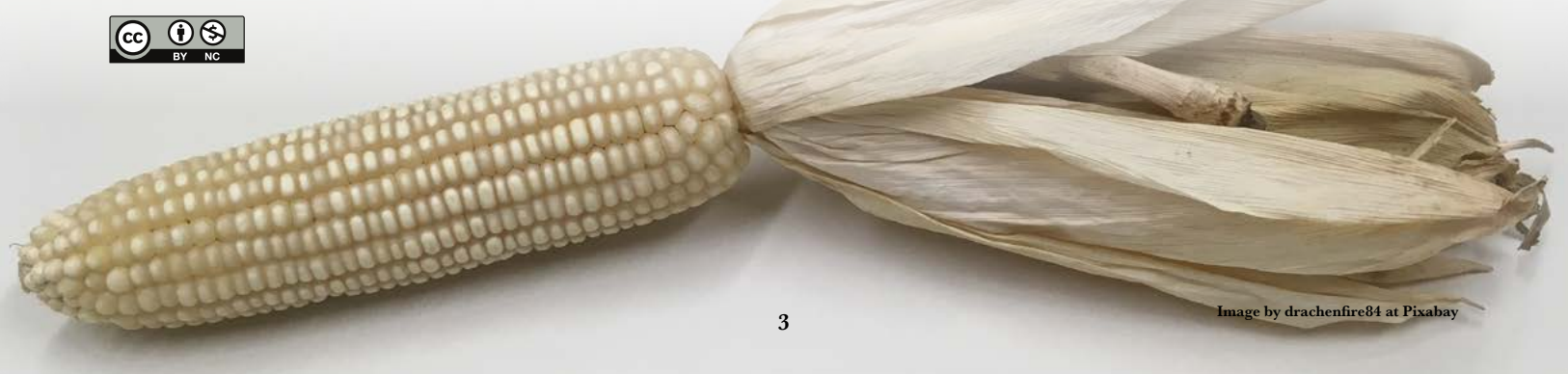
INTRODUCTION

Corn (*Zea mays* L.) is one of the most important species in Mexico and, due to the large number of races and its broad variation and distribution, it is considered center of origin and diversification of the species (Kato *et al.*, 2009). Among the broad diversity of native corns there is variation in the grain color, and this pigmentation gives them unique phytochemical properties (Serna-Saldivar *et al.*, 2013).



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The main pigments associated with corn are carotenoids and anthocyanins; yellow corn is rich in carotenoids and among them β-carotene is an important antioxidant considered of great relevance in human nutrition (Serna-Saldívar, 2010). Corns with blue, purple, red and black grains have antioxidant activity derived from anthocyanins (Mendoza-Díaz *et al.*, 2012). Corn pigments are innocuous colorings for human consumption and of wide interest for the food, pharmaceutical and cosmetic industry. Presently, given the need to improve the quality of human health, the nutritional bioactive properties of pigmented corns have attracted scientific interest, where a broad variation and wealth has been found.

The state of Coahuila, Mexico, is not an important agriculture and livestock production state, and it has the lowest annual precipitation (332 mm) under which corn can be cultivated. This makes it relevant since it values the adaptation of phytogenetic resources to these conditions. According to SIAP (2019), 30,400 ha of grain corn were planted in the spring-summer 2018 cycle, of which 85% were established under rainfed conditions (25,800 ha), mainly with native populations, concentrated (95%) in five municipalities in the southeastern region of the state: Saltillo, General Cepeda, Arteaga, Parras and Ramos Arizpe.

According to Rincón *et al.* (2015), the diversity of native populations in Coahuila were grouped into eight race groups: Celaya, Cónico Norteño, Elotes Cónicos, Olotillo, Ratón, Tuxpeño Norteño, Elotes Occidentales and Tuxpeño. The native populations were found at an altitude interval between 248 and 2557 masl. Because of the frequency of the populations, the corn races that predominate are Cónico Norteño, Ratón and Tuxpeño Norteño, and the highest yield potential, according to Nájera *et al.* (2010), has been observed in the groups Tuxpeño, Tuxpeño Norteño and Ratón.

According to the catalogue of native corns from Coahuila (Rincón *et al.*, 2010), there are accessions with variants of pigmented corns or with high frequency of pigmented grain. These populations, because of their adaptation to the conditions of regional production, can be the basis of the genetic improvement of varieties for grain production with nutritional bioactive contents appropriate for the elaboration of quality foods, and therefore, the study consisted in defining the accessions of pigmented native corn from Coahuila, and evaluating their agronomic behavior with the objective of selecting the best ones to establish production programs with higher grain quality due to their nutritional bioactive content.

MATERIALS AND METHODS

Biological material

Among the local diversity, 52 native populations of pigmented grain were identified with at least one population in each race group; they derived from four areas of adaptation, defined by the altitude of the collection sites. The populations belong to the collection of the state protected in the Conservation Center for Orthodox Seeds Northern Region. In the study, 27 native populations were used with high frequency of yellow pigmentation grains (carotenoids), 25 anthocyanins (blue and red), and eight control populations (Table 1).

Table 1. Frequency of the pigmented native corn populations from Coahuila, Mexico, in each racial group and their area of adaptation.

Corn races	Populations		Adaptation
	Yellow	Red&Blue	
Conico Norteño	3	12	Highland, Int-Highland, Intermediate
Raton	8	5	Lowland, Intermediate
Tuxpeño	10	0	Lowland, Intermediate
Elotes Conicos	0	5	Highland, Int-Highland, Intermediate
Tuxpeño Norteño	4	1	Intermediate, Lowland
Elotes Occidentales	0	2	Intermediate
Celaya	1	0	Intermediate
Olotillo	1	0	Intermediate
Control	3	5	Highland, Intermediate
Total	30	30	

† Highland (>2000 masl); Intermediate-Highland (1801-2000 masl); Intermediate (1001-1800 masl); Lowland (0-1000 masl).

The agronomic evaluation was carried out in the spring-summer (SS) 2019 cycle, under irrigation conditions, in two environmentally contrasting localities: El Mezquite, municipality of Galeana, Nuevo León, and General Cepeda, Coahuila. For the agronomic characterization, each set (yellow and anthocyanins) was established in an experiment with n=30 populations (Table 1), through an experimental design of incomplete blocks with alpha lattice arrangement (0,1) with two repetitions by locality with fertilization doses of 120-60-60 units of N, P and K, respectively. The agronomic characterization was conducted with mean values by experimental unit of plant height (PTH) (cm), days to male flowering (DMF), number of plants without cobs (PWC), and grain yield (GY) expressed in t ha^{-1} and at 14% moisture.

Information analysis

Given the diversity of the populations studied, associated with the area of adaptation, groups were defined within which the corresponding populations by ecological area were nested (Table 1). The analysis of variance (ANOVA) was conducted according to the experimental design with the GLM procedure of SAS (SAS Institute, 2004), where the effects of localities, adaptation groups, and corresponding interaction were tested, with their respective means comparison through the Tukey test. With ANOVA for populations as fixed effects, the superior ones were identified with a decision value determined by the mean plus once the standard error (μ+SE).

RESULTS AND DISCUSSION

Analysis of variance

The contrast of environmental conditions of production was made evident through the significance between localities ($P \leq 0.01$ and $P \leq 0.05$), which represented the ecological scenario of the southeastern region of the state of Coahuila, under which corn production

with native populations takes place. The variation between adaptation groups was also significant and it was associated with the racial and ecological origin of the populations; this condition defined the significance ($P \leq 0.05$) of the interaction Localities × Group, where a differentiated response of the groups was observed through the localities of evaluation (Table 2).

Effect of the localities of evaluation

Tables 3 and 4 show the effect between localities; for the case in study, by altitude and climate characteristics, General Cepeda was considered a locality of the intermediate adaptation area and El Mezquite as transition. In the populations of yellow corn, the environmental effect of the site General Cepeda compared to the site El Mezquite produced a decrease of 71.3% in grain yield (GY), 25 d of difference in masculine flowering (DMF), 53.4 cm in plant height (PTH), and a significant increase of non-productive plants (from 0.35 to 5.4 PWC). Correspondingly, in the anthocyanin populations between localities there was an affectation in GY of 78%, 19 d of difference in DMF, 53.8 cm in PTH and presence of the trait of PWC per experimental unit of up to 38%.

The results obtained between localities were contrasting and corresponded to the climate characteristics and the production potential in each locality. Authors such as Espinosa *et al.* (2019) reported a difference in yield of 29.7% in native populations of Coahuila produced in the same localities under evaluation in this study; that is, a detrimental effect 50% lower than the one found in this study; these contrasts were associated to a higher hydric stress and overpopulation of the fall armyworm (*Spodoptera frigiperda*), affecting the cob weight and increasing the number of plants without cob.

Table 2. Mean squares of the analysis of variance for the agronomic characteristics of the native populations of pigmented corn in the state of Coahuila. El Mezquite, Nuevo León and General Cepeda, Coahulia, Mexico (2019).

Source of variation	DF	Yellow accessions				DF	Blue&Red accessions			
		GrYd t ha^{-1}	DtoA d	PHt cm	NPP		GrYd t ha^{-1}	DtoA d	PHt cm	NPP
Environments	1	282.1 **	7183 *	37675 *	357.5 **	1	712.6 **	9652 **	69362 **	1033 **
Repetition	2	4.29 *	75.75 **	1541 *	19.6 **	2	8.81 *	16.14	6204 **	0.9
Block/Repetition × Environments	8	1.05	2.86 *	424.5	5.28	8	1.53	7.38	264.8	5.03
Adaptation groups	3	13.86 **	148 *	2165 **	7.48	2	14.23 **	65.04 *	237.8	153 **
Populations/Groups	26	2.66 *	48.76 **	789.3 **	5.35	27	6.84 **	82.45 **	1372 **	8.55
Environments × Groups	3	0.21 *	69.76 **	1226 **	3.17	2	7.77 *	32.45 **	464	45.2 *
Populations/Group × Environment	26	1.47 *	5.67 *	165.9	5.35	27	5.59 **	13.89 *	370.5	6.53
Error	50	1.1	2.79	199.4	3.54	50	1.13	6.54 *	264.6	5.98
Coefficient of variation (%)		24.11	2.2	7.55	65.07		27.45	3.41	8.05	55.9

**, *=significance at 0.01 and 0.05 levels of probability, respectively; DF=Degrees of freedom; GY=Grain yield; DMF=Days to masculine flowering; PTH=Plant height; PWC=nonproductive plants.

Among experiments, it was observed that the anthocyanin populations were more affected agronomically in the site General Cepeda; the differences were related to the racial origin and altitudinal level of the populations (Table 1), where the populations of yellow grain belonged primarily to the racial groups Ratón and Tuxpeño, with adaptation to Low and Intermediate altitudes, and highlighted by Nájera *et al.* (2010) as those with highest yield potential among the diversity found in Coahuila, while the anthocyanin populations were primarily of the racial group Cónico Norteño, constituted with populations of Elevation and Transition.

Agronomic characteristics of the pigmented populations and interaction with the environment

Table 3 shows the mean values for GY, DMF and PTH of the outstanding yellow populations in at least one locality of evaluation. The values, according to the conditions of each locality, showed the genetic diversity of the native populations evaluated and the

Table 3. Mean values of agronomic characteristics of the outstanding yellow populations in each locality of evaluation.

Corn races	Accession	General Cepeda, Coah.			El Mezquite, N.L.		
		GrYd t ha^{-1}	DtoA d	PHt cm	GrYd t ha^{-1}	DtoA d	PHt cm
Intermediate†							
OL	COAH 040	2.4 *	62.5	155.5	4.1	87	191
TX	COAH 044	2.3 *	63.5	170.5 *	6.4	89	216.5
CE	COAH 075	3 *	70 *	190.5 *	8.8 *	98 *	241 *
CM	POOL33	0.5	69 *	135.5	8.3 *	94 *	193.5
CM	POOL34	2.6 *	65 *	171 *	7.8 *	90	192.5
Lowland†							
RA	COAH 070	2.8 *	62	159.5	6.7	88	227 *
RA	COAH 077	2.4 *	64	145.5	5.6	88	210
RA	COAH 223	1.3	62.5	150	7.4 *	89.5	219.5 *
TX	COAH 068	2.8 *	62.5	161.5	7.6 *	87.5	224.5 *
TX	COAH 078	2	63.5	147	8.2 *	88.5	220.5
TX	COAH 083	2.2	65.5 *	156.5	7.8 *	94.5 *	214
TX	COAH 089	3.3 *	64.5 *	186 *	9.4 *	93 *	246 *
TX	COAH 177	3.8 *	62.5	177 *	7.5 *	89	222 *
TX	COAH 182	3.2 *	64	157.5	6.9	92.5 *	209
TX	COAH 215	2.8 *	64	196 *	7.7 *	92.5 *	225.5 *
TN	COAH 069	1.7	61.5	151.5	8.3 *	88.5	194.5
TN	COAH 178	2.3 *	62	162.5	6.6	90.5 *	226.5 *
Mean (μ)		1.9	63.1	160.2	6.8	88.8	213.7
Standar error (SE)		0.2	0.5	3.1	0.2	1.0	3.6

†=Adaptation group: Intermediate (1001-1800 m); Lowland (0-1000 m); OL=Olotillo; CE=Celaya; CM=CIMMYT; RA=Raton; TX=Tuxpeño; TN=Tuxpeño Norteño; GrYd=Grain yield; DtoA=Days to anthesis; PHt=Plant height. *=μ+EE; μ=mean; SE=Standar error.

possibility of selecting outstanding germplasm for grain production in each locality. Among the populations, two desired expressions were identified: populations with superior grain yield in both localities, according to the conditions of each one, which were considered stable, and populations with specific adaptation that presented superior yield in at least one locality. Both expressions stood out and these were considered as the outstanding populations to continue in the genetic improvement of the production and the improvement of grain quality.

From the genetic diversity of the populations of yellow grain associated with the racial group and of adaptation, the outstanding populations were from Low to Intermediate, similar to what most of the populations of this coloring represented; the populations of Elevation and Transition, in lower frequency, showed limited expressions.

The populations of the yellow grain considered stable were from the racial group Tuxpeño: COAH068, COAH089, COAH177 and COAH215, with mean yield of 3.1 and 8.0 t ha^{-1} precocious cycle (<65 DMF), and intermediate-late (87.5-93.0 DMF), PTH lower than 2.0 m and higher than 2.2 m, in the sites General Cepeda and El Mezquite, respectively. From the racial group Celaya, the genotype COAH075 stood out with a mean yield between localities of 5.9 t ha^{-1} and the control POOL34 with 5.2 t ha^{-1}.

In addition, six populations showed adaptation to General Cepeda, and they were considered relevant given the climate conditions of the locality and that they showed more than 2.0 t ha^{-1} in it: COAH070, COAH077, of racial origin Ratón; COAH044, COAH182, Tuxpeño; COAH178, Tuxpeño Norteño; and COAH040, Olotillo.

In the site of El Mezquite, with more than 7.0 t ha^{-1}, the ones that stood out were: COAH223, Ratón; COAH078 and COAH083, Tuxpeño; COAH69, Tuxpeño Norteño and the control POOL33. The agronomic expression of the outstanding anthocyanin populations is presented in Table 4, which shows that the variation in yield between these and the yellow populations was similar in the site General Cepeda, but in El Mezquite the variation in the first was higher, although there was a superior expression of yield.

Among the anthocyanin populations, there were also stable populations and with specific adaptation, with the group of Intermediate adaptation standing out: COAH23 and COAH203, Ratón; COAH246, Elotes Cónicos; and COAH019 and COAH021, Elotes Occidentales; these populations showed an interval of variation in yield of 1.4 to 3.0 t ha^{-1} and 7.6 to 11.5 t ha^{-1}, DMF of 61.5 to 71 d and 70 to 90 d; the variation in PTH was from 161 cm to 213 cm and from 212 cm to 269 cm, in General Cepeda and El Mezquite, respectively.

In the specific adaptation a response was observed according to the ecological origin. In the site General Cepeda populations stood out primarily from the Intermediate [COAH025, Ratón; COAH002 and COAH048 (Transition), Elotes Cónicos; COAH031, Tuxpeño Norteño, with mean yield of 2.1 t ha^{-1}], while in El Mezquite for altitude it was COAH011, COAH188, COAH191, COAH311, all of them from the racial group Cónico Norteño, with mean yield of 8.3 t ha^{-1}.

Among the pigmented populations studied, the presence of the racial groups Ratón, Tuxpeño and Tuxpeño Norteño was frequent in the yellow ones, and of Cónico Norteño and Elotes Cónicos in the anthocyanins; this frequency agreed partially with the diversity

Table 4. Mean values of agronomic characteristics of the outstanding anthocyanin populations in each locality of evaluation.

Corn races	Accession	General Cepeda, Coah.			El Mezquite, N. L.		
		GrYd t ha^{-1}	DtoA d	PHt cm	GrYd t ha^{-1}	DtoA d	PHt cm
Higland†							
CN	COAH188	0.6	63	151	9.2 *	80.5	243 *
CN	COAH191	0.3	68 *	156	9 *	80.5	220
CN	COAH198	0.3	67 *	157.5	6	79.5	216.5
CN	COAH311	1.2	70.5 *	183 *	7.8 *	91.5 *	290 *
PE	PERU2	2.4 *	71 *	203.5 *	7.6 *	90 *	230.5
Intermediate-Highland †							
CN	COAH017	0.7	60	150	8.1 *	74	210
Intermediate†							
EC	COAH048	3.2 *	66.5	207.5 *	5.4	87 *	212
RA	COAH023	2.1 *	68 *	189 *	10.5 *	86	243.5 *
RA	COAH025	1.8 *	70 *	163	3.2	97 *	215
RA	COAH203	1.7 *	64	176.5	9.2 *	80.5	219.5
RA	COAH227	1.3	69.5 *	191 *	8 *	95 *	243 *
EC	COAH002	1.5 *	60.5	177	6.9	82.5	218
EC	COAH246	1.4 *	61.5	161	8.5 *	78	212
EC	COAH1019	0.9	60	172.5	8 *	78	229.5
EO	COAH019	3 *	64.5	202.5 *	7.7 *	90 *	247 *
EO	COAH021	1.8 *	67.5 *	213.5 *	11.5 *	87.5 *	269.5 *
TN	COAH031	2.2 *	63.5	187 *	2.7	92.5 *	206.5
Mean (μ)		1.1	65.4	175	6.6	84.4	228.8
Standar Error (SE)		0.15	0.75	3.66	0.45	1.15	4.01

†=Groups of adaptation: Elevation (>2000 m); Transition (1801-200); Intermediate (1001-1800 m); POP=Population; CN=Cónico Norteño; OL=Olotillo; TX=Tuxpeño; EC=Elotes Cónicos; BO=Bolita; CM=; RA=Ratón; TN=Tuxpeño Norteño; EO: Elotes occidentales; GY=Grain yield; DMF=Days to male flowering; PTH=Plant height; *: μ+SE; SE=Standard Error.

of the predominant racial groups reported by Nájera *et al.* (2010), since the Tuxpeño race stood out in the yellow and Cónico Norteño in the anthocyanins, according to the component of pigmentation.

Among the populations, a general agronomic behavior was found according to the racial origin and their adaptation conditions; that is, Ratón, Tuxpeño and Tuxpeño Norteño with adaptation to Low and Intermediate altitudes, associated with similar conditions to General Cepeda and Cónico Norteño to areas of Transition and Elevation similar to El Mezquite; this behavior was also reported by Nájera *et al.* (2010).

Among the yellow populations, the racial groups Tuxpeño and Celaya stood out for their stability, while with specific adaptation the races that stood out were Ratón, Tuxpeño, Tuxpeño Norteño and Olotillo; in the anthocyanins, the stable ones were Ratón, Elotes Cónicos and Elotes Occidentales, while for adaptation the ones that stood out were Ratón,

Elotes Cónicos, Tuxpeño Norteño and Cónico Norteño. Based on this, it was defined that although there was a predominance of racial groups among the diversity of the state, the smaller groups are also important in the component of diversity and particularly in the production of pigmented grain.

CONCLUSIONS

Among the diversity of native populations characterized for having a high frequency of pigmented grain, there are populations with outstanding agronomic expression that can be the basis for the improvement in production and the increase in the nutritional-functional bioactive content in the grain destined to the elaboration of foods of better quality.

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Economic evaluation of a business model of a *Vanilla planifolia* biofactory using BIT® bioreactors

Rodríguez-Deméneghi, Marco Vinicio[1]; Gheno-Heredia, Yaqueline A.[1]; Aguilar-Rivera, Noé[1]; Ramírez-Mosqueda, Marco Antonio[2*]

[1] Universidad Veracruzana Facultad de Ciencias Biológicas y Agropecuarias Región Orizaba-Córdoba de la, C.P. 94945, Amatlán de los Reyes, Veracruz. México.
[2] Centro de Investigaciones Biológicas del Noroeste S.C., Unidad Guerrero Negro. Independencia y Paseo Eucalipto s/n, 23940, Guerrero Negro, Baja California Sur, México.
* Correspondence: marcomosqueda02@hotmail.com

ABSTRACT

Objective: To evaluate the economic and financial viability for the implementation of a biofactory responsible for the *in vitro* propagation of *Vanilla planifolia* with TIB® bioreactors.

Design/methodology/approach: A completely randomized design was used. The data were processed with the IBM SPSS Statistics software (version 21). A Mann-Whitney U test was performed ($p \leq 0.05$). An Economic-Financial Evaluation was carried out, determining the main economic indicators: Profitability, Minimum Acceptable Rate of Return (MARR), Internal Rate of Return (IRR), Net Present Value (NPV) and Benefit-Cost Ratio (B/C R).

Results: TIB® temporary immersion bioreactors were used in this study. An average multiplication rate of 18.37 shoots per explant was obtained. When performing the economic-financial analysis of this agribusiness model over a five-year horizon, it yielded an Internal Rate of Return of 12.36%, a Benefit-Cost Ratio of 1.81, and a Net Present Value of MX$286,506.73 pesos (US$14,474.93), with a payback period of two years and seven months.

Limitations on study/implications: Using semi-solid culture media in the multiplication stage in vanilla decreases the production capacity and significantly lowers the profitability of the biofactory.

Findings/conclusions: The profitability of a biofactory for the production of vanilla depends on the multiplication rate, achieved in this case satisfactorily through the use of TIB®. This vanilla biofactory agribusiness model using bioreactors can be adopted by investors as an economic development strategy.

Keywords: biofactory, bioreactors, economic evaluation.

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INTRODUCTION

Vanilla (*Vanilla planifolia*) has economic importance due to the compound extracted from its processed pods, which is called "vanillin", used mainly as flavoring with sweet and soft aroma (Banerjee and Chattopadhyay, 2019). It has even been used for its medicinal qualities as antimicrobial and antioxidant (Andrade-Andrade *et al.*, 2018). It is a very prized compound among the cosmetic, pharmaceutic, gastronomic industries, and it is currently one of the three most demanded and costly species in the world (Cardone *et al.*, 2020;

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Khoyratty et al., 2018); as a producing country, Mexico occupies the fourth place globally (FAOTAST, 2020). However, this species presents serious reproduction problems, because of the low germination percentage of its seeds (≤1 %) and the limited number of cuttings that can be obtained from an adult plant (Ramírez-Mosqueda and Iglesias-Andreu, 2016; Ramírez-Mosqueda and Bello-Bello, 2021).

Three methods are used in Mexico to propagate vanilla. 1) Cuttings or stem segments of 70-80 cm, 2) *In vitro* culture, and 3) Seeds. The first is the one used commercially for its propagation. In the vanilla-producing regions of Mexico, the cuttings are obtained from commercial plantations, since there are no nurseries devoted exclusively to their production (Santillán-Fernández, 2019). Convential propagation is slow, it requires much work and consumes a lot of time for the growth and development of the mother plant (Geetha and Shetty, 2000; Giridhar and Ravishankar, 2004). The second case refers to *in vitro* culture, which has been developed recently in greater proportion; these micropropagation techniques can attain a large amount of propagules in small spaces and in less time (Loyola-Vargas and Ochoa-Alejo, 2018). The third case, seeds, is limited by the low viability and germination rate of the seeds (Sasikumar, 2010).

In this sense, the micropropagation obtained from plant tissue culture (PTC) techniques has become the basis of a large industry of commercial plant propagation that involves hundreds of laboratories around the world. It is crucial to exploit the *in vitro* culture potential to multiply and supply the necessary amount of planting materials at a large scale (Abebe *et al.*, 2009). This technology can also be used to satisfy the market's demand for vanilla and to surpass the difficulties in alternative propagation methods.

Achieving the micropropagation of *V. planifolia* in a most efficient way has been attained through biotechnology, with the use of Temporary Immersion Bioreactors (TIB) (Ramírez-Mosqueda and Iglesias-Andreu, 2016). However, these scientific advances have not been used in the implementation of a biofactory, to supply producers from the Totonacapan region with commercial propagules. Biofactories can be defined as centers for the large-scale production of improved plants and seeds to obtain specimens with novel scientific techniques that guarantee their quality (CONACYT, 2016). Biofactories operate as productive businesses, which is why their development is based on the economic results from their sales (Suarez-Castellá *et al.*, 2009). In this sense, the implementation of a biofactory to cultivate vanilla using the TIB bioreactor would be an extremely efficient alternative in the commercial propagation of this valuable crop.

MATERIALS AND METHODS

Plant material

To conduct this research study, the methodology reported by Ramírez-Mosqueda and Iglesias-Andreu (2016), on the evaluation of different temporary immersion systems in the shoot proliferation phase, was replicated. *In vitro* plants of *V. planifolia* were used, obtained in the Plant Micropropagation Laboratory (Laboratorio de Micropropagación Vegetal, LAMICROVE) of the Facultad de Ciencias Biológicas y Agropecuarias Región Orizaba-Córdoba at the Universidad Veracruzana (UV) in Amatlán de los Reyes, Veracruz, Mexico.

Evaluation of two temporary immersion systems in the shoot proliferation phase

Nodal segments (1-2 cm of length) were cultivated in two temporary immersion systems: RITA® 100 ml (150×130 mm) and TIB® 940 ml (180×100 mm). In all the experiments, the MS (Murashige and Skoog, 1962) culture medium was used, added with 50 mg L^{-1} of cysteine hydrochloride, 100 mg L^{-1} of ascorbic acid, 30 g L^{-1} of sucrose. The pH of the medium was adjusted to 5.8±0.2 and the culture jars were sterilized in an autoclave at 1.5 kg cm^{-2} of pressure and 121 °C for 15 min. Then, 2.1 mg L^{-1} of benzyladenine (BA) was added and an immersion frequency of 2 min every 8 hours was used, with a volume of medium of 25 ml per explant. The cultures were incubated at a temperature of 25±2 °C, under radiation of 50 μmol m^{-2} s^{-1}, provided by LED lamps. For each immersion system, 50 explants were used (10 explants per jar with five repetitions). After six weeks of culture, the following variables were evaluated: number and length of shoots formed and number of leaves.

Rooting and acclimation

Individual shoots with 2 cm length were rooted in TIB bioreactors, using MS medium at 50% of its concentration, without plant growth regulators. The pH of the medium was adjusted to 5.8±0.2 and the culture jars were sterilized in an autoclave at 1.5 kg cm^{-2} of pressure and 121 °C for 15 min. The time, frequency of immersion, and incubation conditions were the same as those described before. For the acclimation, shoots with a height of 8-10 cm previously rooted *in vitro* were rinsed with running water and sown in (1:1 v/v) 1:1 peat moss mixture (Premier, Rivière-du-Loup, Canada) and agrolite® (Agrolita, Tlalnepantla de Baz, Mexico) as substrate, using trays of 50×30×5 cm. The seedlings were kept under greenhouse conditions (shade at 50%, relative moisture between 80-95%, temperature 28-32 °C), Nitrofoska® (N: 25 P: 10 K: 17) (PS, COMPO, Zapopan, Mexico) was applied as leaf fertilizer once per week, as well as irrigation with running water three times per week. When the plants reached 30 cm of height, they were transferred to individual containers with the same substrate.

Statistical analysis

A completely randomized design was used in these experiments. The data that were obtained from this experiment were processed with the IBM SPSS Statistics software (version 21). A Mann-Whitney U test was conducted ($p \leq 0.05$).

Economic-Financial Evaluation

An Economic-Financial Evaluation was carried out based on an agribusiness model described by Pavón (2012), where the implementation of a biofactory model is considered, establishing the main economic indicators: profitability, minimum acceptable rate of return (MARR), internal rate of return (IRR), net present value (NPV), and benefit-cost rate (B/C R) to understand the viability of this project.

RESULTS AND DISCUSSION

Evaluation of two different temporary immersion systems in the shoot proliferation phase

At six weeks of culture, significant differences were observed between the two temporary immersion systems evaluated (Table 1). The largest number of shoots/explants (18.37) was obtained in the BIT® bioreactor, followed by RITA® (12.62). However, longer shoots were obtained (1.67 cm) in the RITA® bioreactor compared to BIT® with 1.43 cm. No significant differences were observed for the number of leaves in BIT®.

The values represent the mean ±SE (standard error). Means with different letter are significantly different (U de Mann-Whitney ($p \leq 0.05$). Figure 1 shows the development of shoots/explants of *V. planifolia* in the temporary immersion bioreactor TIB®.

Based on the results obtained in our study, it was corroborated that the methodology proposed by Ramírez-Mosqueda and Iglesias-Andreu (2016) is efficient for the micropropagation of *V. planifolia*, using TIB® bioreactors. The TIB® bioreactor has been efficient for micropropagation of sugarcane (*Saccharum* sp.) (Lorenzo *et al.*, 1998), production of potato microtubers (*Solanum tuberosum* L) (Tapia *et al.*, 2020), and papaya (*Carica papaya* L.) (Gómez-Carrera, 2018). Ramos-Castellá *et al.*, determined that the RITA® bioreactor was efficient for the commercial micropropagation of *V. planifolia*. However, our study shows how TIB® generated higher biological yields, expressed in number of shoots. Ramírez-Mosqueda and Bello-Bello (2021) determined that shoots of

Table 1. Effect of two types of temporary immersion systems in the proliferation phase of *Vanilla planifolia* shoots at six weeks of culture, using the methodology proposed by Ramírez-Mosqueda and Iglesias-Andreu (2016).

Bioreactor Type	Number of shoots/explant	Shoot length (cm)	Number of leaves/ shoot
RITA®	12.62 ± 0.26^{b}	1.67 ± 0.09^{a}	1.85 ± 0.19^{a}
BIT®	18.37 ± 0.19^{a}	1.43 ± 0.10^{b}	1.77 ± 0.20^{a}

Values represent mean ± SE (standard error). Means with different letters are significantly different (Mann-Whitney U test ($p \leq 0.05$).

Figure 1. Result from the temporary immersion system in the shoot proliferation phase of *Vanilla planifolia* in TIB®: A) development of *V. planifolia* shoots, B) and C) specimens of *V. planifolia* established in bioreactor TIB® in acclimation stage.

V. planifolia generated in the SETIS™ bioreactor are taller in comparison with TIB®. However, in the multiplication phase, what is sought is to obtain the greatest number of commercial shoots. On the other hand, the TIB® bioreactor has a lower cost per unit than all bioreactors offered commercially. In this sense, Alamilla-Magaña *et al.* (2019) mention that the decrease in the initial cost of a biofactory is essential to ensure its success. Therefore, the TIB® bioreactors fulfill these qualities of being economic and functional.

Rooting and acclimation

On hundred percent rooting was achieved from the *V. planifolia* shoots using TIB® bioreactors. In addition, 100% of survival was attained during the acclimation process (Figure 1).

In vitro rooting using reduced culture means, without the addition of RCV, is an option that reduces the production costs (Alamilla-Magaña *et al.*, 2019). In this sense, our study was successful in this phase of micropropagation, which could be implemented in a biofactory. The acclimation phase is one of the most difficult in PTC, and therefore, its success reflects the efficiency of micropropagation protocols. The plants generated in TIS also present physiological advantages to those propagated in conventional systems (semi-solid), among them the stomatic functionality due to gas exchange (Georgiev *et al.*, 2014). In the present study, the efficiency of the protocol described is corroborated, with the attainment of 100% survival of the plants generated.

Economic-Financial Evaluation

To determine whether this agribusiness model of vanilla biofactory using TIB® bioreactors is feasible, an economic evaluation was carried out where the financial indicators of a private investment project were determined and where a source of financing from an external institution was contemplated. To start the operations of the biofactory, an initial investment of MX$ 605,380.07 is required, equivalent to US$ 30,585.10 (at an exchange rate of $19.7933 MX pesos per dollar, 26/05/22, BANXICO). In this investment, the construction of facilities was not considered, since a segment is contemplated for their rental within the operation expenses, although this investment includes equipment, materials, reagants, and assemblage of a greenhouse area for acclimation, required for the establishment of the biofactory. The fixed investment estimated is MX$ 440,380.07 (US$ 22,248.94), a deferred investment of MX$ 15,000.00 MX pesos (US$ 757.83), which is attributed to the legal construction of the company, as well as the specialized training on the operation of the TIS and working capital of MX$ 150,000.00 (US$ 7,578.32). It is expected that, for every productive year (in a horizon projected of 5 years), there will be an annual production of 20,000 vanilla seedlings and sales of MX$ 500,000.00 (US$ 25,261.07). The recovery period was achieved at two years and seven months after the project began. Table 2 shows the calculation memory used to define the financial indicators presented in this same study. A MARR of 10% was obtained, which is a minimal measure of profitability that was demanded from the

Table 2. Calculation memory to obtain financial indicators in a vanilla biofactory business model.

Concept	Unit of measurement	Calculation memory		Total (MX$)
		Quantity	Amount ($)	
Equipment	Batch	1	363,671.60	363,671.60
Reagents for propagation	Batch	1	47,590.16	47,590.16
Consumables	Batch	1	8,314.24	8,314.24
Area equipment	Batch	1	20,804.07	20,804.07
			TOTAL	$ 440,380.07
Other expenses				**Cost (MX$)**
Production and labor				209,531.56
Administration and sale				59,600.00
Financial expenses				18,000.00
Performance and sale price				**Total**
Multiplication rate		(units)		20,000
Unit price				$25.00

project and was obtained from the inflation rate for the year 2021, issued by Banco de México plus a risk premium (7.36+2.64=10). An IRR of 12.36% was obtained and this is equivalent to the interest rate provided to investors, this benefit percentage should be higher than the MARR and it reflects that the initial investment has been recovered. Regarding the NPV obtained of MX$286,506.73 (US$ 14,474.93), this criterion suggests that the project should be accepted since its net present value (NPV) is equal or higher than zero, where the NPV is the difference between all its revenues and expenditures expressed in present currency. After having conducted the corresponding operation for the calculation of the Benefit/Cost Rate (B/C R), the value obtained is positive and reaches $1.81, which indicates that for every peso invested, $0.81 cents of benefit will be obtained, where the percentage represents a profitability of 81%. The main indicators which express the economic yield of an investment were determined, and decisions can be made based on these to determine whether a project is viable or, if aplicable, to make a profitability assessment of such an investment. Chino (2011) points out that the most frequently used economic indicators are those that consider the value of money over time, among which there is: Minimum Acceptable Rate of Return (MARR); Internal Rate of Return (IRR); Net Present Value (NPV); and Benefit/Cost Rate (B/C R). In this sense, Baca (2001) mentions that the MARR is the real growth rate of the company above inflation, which is why in this project it is attained with the calculation. Meanwhile, Pavón (2012) indicates that the project gives a higher profitability than the minimum required profitability. The NPV considers the value of money over time; it is the numerical difference between the updated value of the benefits (utility) and the updated value of the costs at a specific updated rate, and when the numerical difference is positive, the investment is recommendable (Chino, 2011). Chino (2011) indicates that the B/C R value should be higher than zero, which indicates that the activity is generating utilities, so in this project it is fulfilled with this parameter.

CONCLUSIONS

Based on the results obtained in our study, the efficiency of the TIB® temporary immersion systems in the micropropagation of *V. planifolia* using the methodology by Ramírez-Mosqueda and Iglesias-Andreu (2016) was demonstrated. The use of TIB® bioreactors, which because of their acquisition cost that is rather lower than other types of temporal immersion systems, sets the path to be used in the assemblage of a commercial vanilla biofactory, which was the objective of this study. The results from the economic-financial evaluation indicators of a biofactory business model for *Vanilla planifolia* using TIB® bioreactors are positive, indicating the acceptability of the project based on an agribusiness model. The economic projections reflected the viability of this project, since it assures to be completely profitable based on the economic indicators. There are many research studies about the cultivation of vanilla, however, no studies have been conducted about the implementation of a vanilla biofactory company and the value of its costs in an investment project in this agricultural sector. This agribusiness model of a vanilla biofactory using TIB® bioreactors can be adopted by investors as an economic development strategy, considering the potential for growth in producing zones and in face of the need that producers have for quality plant material that is resistant to pests and diseases.

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Colegio de Postgraduados

Optimization of a production unit of the dairy agroindustry: Chapingo dairy technology unit case

Cortéz-Reyes, Carlos M.[1]; Arana-Coronado, Oscar A.[1*]; Garza-Bueno, Laura E.[1]; Martínez-Damián, Miguel A.[1]; Rodríguez-Yam, Gabriel A.[2]

[1] Colegio de Postgraduados Campus Montecillo, Carretera México-Texcoco km 36.5, Montecillo, Texcoco, Estado de México, México, C.P. 56264.
[2] Universidad Autónoma Chapingo, División de Ciencias Forestales, Chapingo, Texcoco, México, México, C.P. 56230.
* Correspondence: aranaosc@colpos.mx

ABSTRACT

Objective: To evaluate the optimization level of a dairy production unit (DPU) through a mathematical programming model (MP). It is expected that by maximizing net income by at least 10%, the DPU will be more profitable than without optimized management.

Design/Methodology/Approach: The analysis was carried out under the economic approach of agricultural production, taking into consideration 11 decision variables in the objective function (OF), which was subject to 20 constraints. The variables were based on the requirements of the demand for dairy by-products, using technical coefficients (input-output coefficient). Excel® Solver® was used to develop the sensitivity report and to analyze the shadow prices and reduced costs.

Results: Three scenarios were modeled. Between the first and third scenario, the income increased to $58,000.00 (41.02%). Between the second and third scenario, the income increased to $63,840.00 (46.16%).

Study Limitations/Implications: Dairy food processing is an important industry in the economies of the world.

Findings/Conclusions: Panela cheese recorded the highest shadow price ($72.85), which indicates that the DPU should concentrate on this type of dairy product. In conclusion, the optimization of the DPU guarantees the efficient use of scarce resources and therefore generates a higher profit.

Keywords: Mathematical programming, optimization, constraints, reduced cost, shadow prices.

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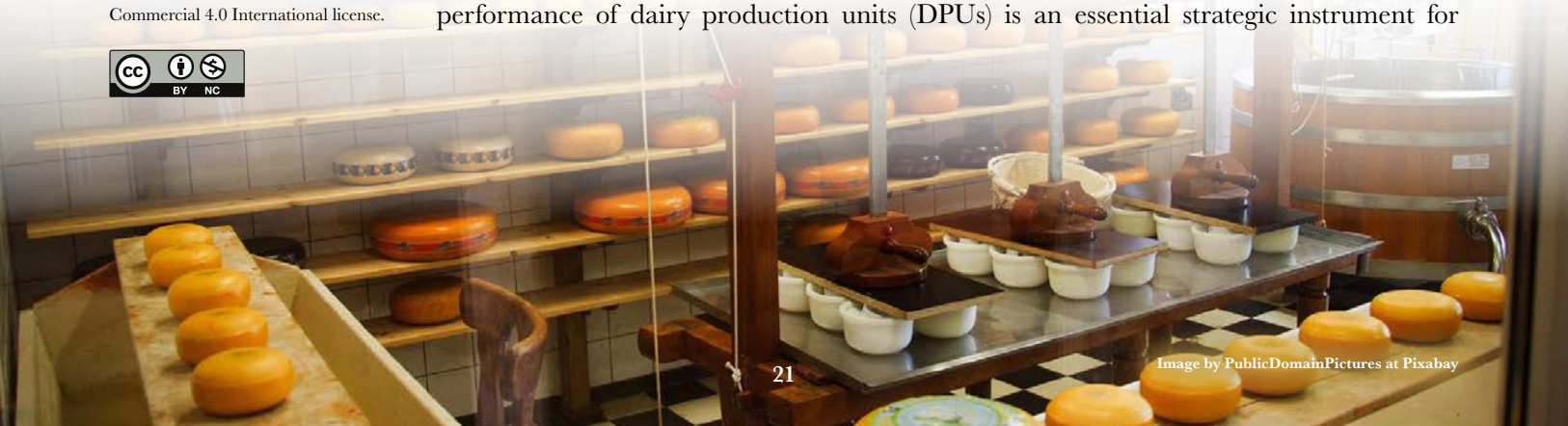
INTRODUCTION

Dairy food processing is an important industry in the developed and developing economies of the world. A wide range of dairy products meets the diverse consumer tastes and trends. Production planning in the dairy industry is usually a challenging task (Bilgen and Çelebi, 2013).

In times of food crisis, global economic crisis, and increased poverty, improving the performance of dairy production units (DPUs) is an essential strategic instrument for



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development (Alvarado *et al.*, 2020). Demand for dairy products is expected to double by 2050, as a result of increasing population, rising incomes, and nutritional concerns. Therefore, DPUs will have to increase their production, in order to meet the growing demand, without sacrificing their profitability versus the competition (Salinas *et al.*, 2020). Guevara and Guevara (2015) affirm that the dimensionality of milk production systems makes milk production one of the major contributors to food security. SADER (2020) defines production units as the set of land, infrastructure, machinery, equipment, animals, and other goods used in agricultural activities. SADER (2020) also recognizes the contribution of production units to the reversion of productivity and competitiveness problems in the agricultural sector, given Mexico's high dependency on food imports. OECD/FAO (2019) points out that the increase in productivity within the DPUs will accelerate agricultural growth, since it is an effective factor for both economic development and poverty reduction. Rueda and Rueda (2017) and Alvarado *et al.* (2020) agree on the advantages of the DPUs for the welfare in rural areas through the development of the economic-productive factors to generate jobs and income, which directly contribute to the increase in GDP. In Mexico, there are more than 300,000 DPUs that represent more than 78% of dairy farms (SAGARPA, 2018).

With regard to the Mexican economy, DPUs play an important role in food security: since they contribute approximately 35% of the national milk production and generate permanent paid employment opportunities, they have been promoted to alleviate poverty (Salinas *et al.*, 2020). The DPUs usually include teaching, service, and research activities; dairy production; milk pasteurization; and Oaxaca, panela and Chapingo cheeses production (Hernández, 2018). The objective of this research was to optimize the profitability of the DPU, through a mathematical programming model (MP). It is expected that by maximizing its net income, the DPU will be higher than the current situation without optimized management.

MATERIALS AND METHODS

The research was carried out in the dairy production unit of the Universidad Autónoma Chapingo (UACh), known as the Chapingo dairy technology unit (DTU), in Texcoco, State of Mexico (19° N, 98° W). The experimental fields were located at 2,252 m.a.s.l. The objective of the DTU is to contribute to the practical training of the students of the Departamento de Ingeniería Agroindustrial (DIA).

According to Terrazas (2012), in order to carry out an analysis under the economic approach of agricultural production through the construction of a MP model, the decision variables to be optimized in the objective function (OF), subject to production constraints, must be taken into consideration. First, the said variables were defined. The OF will determine the values of the technical coefficient of the decision variables (Jebelli *et al.*, 2016). The general form of OF (Z) is expressed mathematically as follows:

$$Maximize\ Z = \sum_{j=1}^{n} a_j x_j \qquad \text{where: } j = 1 \text{ until } n \tag{1}$$

Subject to 20 constraints and 11 decision variables, as follows:

$$\begin{aligned} a_{11}X_1 + a_{12}X_2 + a_{13}X_3 + \ldots + a_{1\,11}X_{11} \leq = \geq b_1 \\ a_{21}X_1 + a_{22}X_2 + a_{23}X_3 + \ldots + a_{2\,11}X_{11} \leq = \geq b_2 \\ a_{31}X_1 + a_{32}X_2 + a_{33}X_3 + \ldots + a_{3\,11}X_{11} \leq = \geq b_3 \\ \vdots \quad \vdots \quad \vdots \quad \vdots \quad \vdots \\ a_{20\,1}X_1 + a_{20\,2}X_2 + a_{20\,3}X_3 + \ldots + a_{20\,11}X_{11} \leq = \geq b_{20} \end{aligned} \tag{2}$$

And non-negativity constraint: $X_j \geq 0$ (3)

The constraints for the DPU are based on the requirements of the demand for dairy by-products and production restrictions (Anderson *et al.*, 2011). Subsequently, a sensitivity report was developed directly from Excel® Solver® for the analysis of the shadow prices and reduced costs. The dairy processes of the DPU that are considered are the decision variables whose level of production must be optimized and sold (Table 1).

The limits of the constraints were established based on field research at the DPU. These limitations included the technical coefficients, the minimum demand for each by-product, the amount of raw milk received weekly in the DPU, and the intensity of the labor requirements for scenario 1 (Table 2).

Three scenarios were proposed: 1) the model was calibrated to obtain results similar to the current production situation; 2) the optimal combination of production activities was determined; and 3) slack variables were introduced.

The objective function of the three scenarios was:

$$\begin{aligned} Maximize\, Z = &-7LB - 0.6LP + 16VLPC - 17.35PY + 32VY \\ &-47.15PQP + 120VQP - 84.73PQO + 140VQO \\ &-112.16PQCH + 160VQH \end{aligned} \tag{4}$$

Table 1. Decision variables for the objective function.

Variable	Abbreviation	Notation	Coefficient	Element in the OF
Raw milk	LB	X_1	c_1	$c_1 * X_1$
Pasteurized milk	LP	X_2	c_2	$c_2 * X_2$
Pasteurized milk for sale	VLPC	X_3	c_3	$c_3 * X_3$
Yogurt production	PY	X_4	c_4	$c_4 * X_4$
Yogurt sale	VY	X_5	c_5	$c_5 * X_5$
Panela cheese production	PQP	X_6	c_6	$c_6 * X_6$
Panela cheese sale	VQP	X_7	c_7	$c_7 * X_7$
Oaxaca cheese production	PQO	X_8	c_8	$c_8 * X_8$
Oaxaca cheese sale	VQO	X_9	c_9	$c_9 * X_9$
Chapingo cheese production	PQCH	X_{10}	c_{10}	$c_{10} * X_{10}$
Chapingo cheese sale	VQCH	X_{11}	c_{11}	$c_{11} * X_{11}$

Source: Dairy production unit (DPU) data for the year 2020. c_i = net price = product price$_j$ × quantity produced$_i$ − variable cost$_i$

Table 2. Technical coefficients for the restriction limitations, scenario 1.

Product	Sign	Quantity required	Unit	Yield	Price
LB	=	22,400	L	1	7
LP	≤	22,400	L	1	0.60
VLPC	≤	14,000	L	1	16
PY	≤	3,000	L	0.9	32
PQP	=	300	kg	6.67	120
PQO	=	200	kg	10	140
PQCH	=	160	kg	12.5	160
Mano de obra	≤	42	h		
Agua	≤	15	m^3		
Cultivo láctico	≤	60	g		
Cloruro de calcio	≤	160	ml		
Sal	≤	350	kg		
Cuajo	≤	100	ml		

Data directly provided by the DPU, 2020.

For the three scenarios, the signs were changed; as shown below, the first sign belonged to the first scenario and so on:

Subject to:

Raw milk received: $1LB = = \leq 22{,}400$

LP Production: $1LP \leq = \leq 22{,}400$

Sale of LP for consumption: $1LP \leq = \leq 22{,}400$

Yogurt production: $1PY \leq = \leq 3{,}000$

Panela cheese production: $1PQP = = \leq 3{,}000$

Oaxaca cheese production: $1PQO = = \leq 200$

Chapingo cheese production: $1PQCH = = \leq 160$

Transfer from LB to LP: $-1LB + 1LP \leq \leq \leq 0$

Transfer from LP to by-products: $-1LP + 0.9PY + 6.67PQP + 10PQO + 12.5PQCH \leq \leq \leq 0$

Restaurant markets: $-1LP + 1VLP \leq \leq \leq 0$

Yogurt market: $-1LP + 1VLP \leq \leq \leq 0$

Panela cheese market: $-1PQP + 1VQP \leq \leq \leq 0$

Oaxaca cheese market: $-1PQO + 1VQO \leq \leq \leq 0$

Chapingo cheese market: $-1PQCH + 1VQCH \leq \leq \leq 0$

Labor: $0.000022VQCH + 0.00018LP + 0.00255PY + 0.01861PQP + 0.03708PQO + 0.054687PQCH \leq \leq \leq 42$

Water: $0.00022VQCH + 0.00022LP + 0.00033PY + 0.00333PQP + 0.005PQO + 0.00625PQCH \leq \leq \leq 15$

Lactic culture: $0.01PY + 0.18PQCH \leq \leq \leq 60$

Calcium chloride: $0.2PQP + 0.3PQO + 0.25PQCH \leq \leq \leq 160$

Salt: $0.5PQP + 0.5PQO + 0.6PQCH \leq \leq \leq 350$
Rennet: $0.5PQP + 0.5PQO + 0.6PQCH \leq \leq \leq 350$

RESULTS AND DISCUSSION

When comparing the levels of production and sales in scenario three, a reduction of 8,400 L of the raw milk received weekly was observed; therefore, the pasteurization diminished to 37.5%. Other by-products remained at the same levels in the three scenarios (Table 3).

Between the first and third scenario, the net income had a difference of $58,800.00 and Z increased by 41.02%. Between the second and third scenario, a difference of $63,840.00 meant that the income increased by 46.16%.

The net income obtained in the scenarios was similar to the percentage used by Salinas *et al.* (2020) to optimize their economic benefit in stratum I (46%) and stratum III (73%). The results of Arauco and Arauco (2016) match those obtained in the model; they optimized the production of the Mantaro S. A. dairy plant, increasing its income from $3,030.00 to $93,000.00 biweekly soles, through the application of optimization techniques and the efficient use of available resources.

The third scenario obtained $202,113.00 per week, which means that the DPU must sent its entire production to the market, as a consequence of the reduction of 8,400 L of LB received per week. Likewise, the variable costs (with savings of $58,800.00) were optimized by 22.08% between the first and third scenario.

When contrasting the second and third scenario, the optimization was 23.52% and diminished weekly by $63,840.00. When comparing the second and third scenario, the pasteurization process decreased 23.52% and $5,040.00 were saved per week. Alvarado (2011) maximized his benefit by 5.40%, with an increase from $109,704.00 USD to

Table 3. Results of the optimized OF in the tree scenarios of the DPU (2020).

Scenarios	Optimized quantity			$ MX		
	1	2	3	1	2	3
LB	22,400	22,400	14,000	−156,800	−156,800	−98,000
LP	14,000	22,400	14,000	−8,400	−13,440	−8,400
PY	14,000	14,000	14,000	−52,050	−52,050	−52,050
PQP	3,000	3,000	3,000	−14,145	−14,145	−14,145
PQO	3,000	3,000	3,000	−16,946	−16,946	−16,946
PQCH	300	300	300	−17,946	−17,946	−17,946
VLP	300	300	300	224,000	224,000	224,000
VY	200	200	200	96,000	96,000	96,000
VQP	200	200	200	36,000	36,000	36,000
VQO	160	160	160	28,000	28,000	28,000
VQCH	160	160	160	25,600	25,600	25,600
Max Z				143,313	138,273	202,113

Data obtained directly from the MP model. The amounts with sign (−) correspond to variable cost.

$115,634.00 USD, obtaining similar results to those found in this research. Terrazas (2012) determined that the application of mathematical models to industrial and business problems makes the application of mathematical programming an imperative.

The optimal production of the three scenarios of the DPU is similar to that found by Arauco and Arauco (2016), who established that the optimal quantities for a biweekly production are the following: 202 kg of semi-integral fresh cheese, 506 kg of fresh cheese, 1,668 L of yogurt, and 498 kg of manjar blanco.

Processing 14,000 L of raw (unpasteurized) milk weekly, in the first and third scenario, provided similar results to those of Jablonsky and Skocdopolova (2017), when 56,810 L of raw milk were collected or bought each month to be processed.

The variable costs of by-products remain at the same levels in all three scenarios. The second part of the sensitivity report showed changes in the resources, which are the values to the right of the constraints (Right Hand Side-RHS-). These results represent the availability of resources of the DPU (LB, LP, Y, QP, QO, QCH, labor, water, inputs, raw materials, etc.). Changes in these values also affect the results of the feasible region and consequently the value of the optimal solution.

Among the results found, the final value obtained stands out, as well as the shadow prices generated (Table 4).

The shadow price of the LB in the first and second scenario is −$7.00, which is equal to the market price of the product. In the first scenario, 12,880 L more can be received of LB, and in the second 4,480 L more; however, the first can receive 8,400 L less, but the second scenario cannot receive any less L. The shadow price of the VLPC was $15.40, $16.00 and $8.40, in the first, second, and third scenarios, respectively. The VLPC in the first scenario is 54.54% more profitable than the third scenario; the VLPC is 47.5% more efficient in the second scenario than in the third; and the VLPC can increase up to 8,400 L per week in the three scenarios. In the first and third scenario, the VLPC can diminish by 5,299 L, and in the second by 14,000 L, without affecting the optimal solution found by the model.

Producing an additional liter of fruit yogurt implies an increase in profit of $14.65 in the three scenarios and the weekly production can be increased by 120 L. According to the shadow prices, PQP is 50.5% more profitable in first and second scenario —since the optimal value increases $72.85 for each additional kilo of PQP (only $36.00 in the third scenario). Therefore, production in third scenario can increase up to 24 kg more without affecting the optimal value.

The PQO in scenarios 1 and 2 had a shadow price of $52.27.

Increasing an additional kilo of the PQCH increases income in first and second scenario by $47.84 and in third scenario by $1.74.

In the third scenario, transferring an additional liter from LB to LP has a shadow price of $7.00; therefore, for each liter transferred from LB to LP, the optimal value will increase by $7.00. The same phenomenon takes place when a liter of LP is transferred to VLPC, but in this case the income increases by $7.60.

According to the shadow prices calculated in the scenarios, selling an additional liter of yogurt or a kilo of panela, Oaxaca or Chapingo has a positive impact on the profit of $32.00, $120.00, $140.00, and $160.00, respectively (because these are the market prices

Table 4. Contrast between the final value and the shadow-price of the scenarios.

Scenarios	Final Value			Slack Variable	Shadow Price		
	1	2	3	S1−S20	1	2	3
LB	22,400	22,400	14,000	8,400	−7	−7	0
LP	14,000	22,400	14,000	8,400	0	−0.60	0
VLPC	14,000	14,000	14,000	0	15.40	16.00	8.40
PY	3,000	3,000	3,000	0	14.65	14.65	14.65
PQP	300	300	300	0	72.85	72.85	36
PQO	200	200	200	0	55.27	55.27	0
PQCH	160	160	160	0	47.84	47.84	1.78
Transfer LB to LP	−8,400	0	0	0	0	0	7
Transfer LP to By products	−5,299	−13,699	−5,299	5,299	0	0	0
Transfer LP to VLPC	0	−8,400	0	0	0.60	0	7.60
VY	0	0	0	0	32	32	32
VQP	0	0	0	0	120	120	120
VQP	0	0	0	0	140	140	140
VQCH	0	0	0	0	160	160	160
Labor	32.42	33.93	32.23	9.76	0	0	0
Water	12.13	14	10.25	4.75	0	0	0
Lactic culture	58.80	58.80	58.80	1.2	0	0	0
Calcium chloride	160	160	160	0	0	0	184.23
Salt	346	346	346	4	0	0	0
Rennet	92	92	92	8	0	0	0

Data obtained from the sensitivity analysis of the MP model.

of products). Despite the unlimited labor in the DPU, operating in the red is not convenient for the university (UACh); therefore, this resource should be constrained. Salinas *et al.* (2020) determined that, according to the producers (who agree with this argument), labor was the second most important element. However, when the DPUs are seen as companies, they generate work and self-employment positions, often with an above minimum payment (Posadas *et al.*, 2014).

Water does not have a shadow price, because it is used without restrictions. However, if it were constrained, it would effectively represent a problem for the DPU, since the dairy industry is one of the biggest spenders or consumers of this resource. The lactic culture, calcium chloride, salt, and rennet resources did not have a shadow price, because the DPU is subsidized by the University.

CONCLUSIONS

The optimization of the DPU with mathematical programming is a tool that improved the efficiency in its production. The following strategy was proposed: not receiving LB allowed an increase in the profit up to $58,800.00 per week. In its turn, this also increased

the optimal value, since if no more liters of LB were received, they would not be pasteurized, which reduces variable costs.

By optimizing the income of the DPU, a benefit of $143,313.00 was obtained between the first and third scenario; the aim was to obtain the maximum value of Z per week (an increase of 41.02%).

On the production side, the cheese that should be produced more is panela (PS=72.85/kg) and the cheese that should be produced less is Chapingo. When deciding whether to use VLPC or PY, VLPC turned out to be a better choice, since it increases the optimum by $1.35; additionally, the variable costs of VLPC are 96.54% lower than those of PY. Another significant aspect is that selling fruit LPC or PY is not as profitable as the production of certain types of cheese.

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Use of firewood in an indigenous community from the Sierra Norte de Puebla: a gender perspective on the access to natural resources

Rosas-Mancilla, María E.[1]; Pérez-Ramírez, R.[2]

[1] Universidad Autónoma Chapingo, Carretera Federal México - Texcoco Km 38.5, Texcoco, Estado de México, C.P. 56235, Mexico.
[2] Colegio de Postgraduados Campus Montecillo, Carretera México -Texcoco km 36, Montecillo, Texcoco, Estado de México, C. P. 56264, Mexico.
* Correspondence: rodrigo_perez_r@yahoo.com

ABSTRACT

Objective: To analyze several key elements in order to understand the rights of use, access, and ownership of natural resources, based on the social roles assigned to each gender in the community of Reyeshogpan, Puebla, Mexico.

Design/Methodology/Approach: A participant observation methodology was used, complemented by the application of 24 semi-structured interviews in local households.

Results: The surface ranges of the plots are 0.25±3.0 ha. Out of all the interviewees, 40.79% own less than 0.5 ha, which is not enough to cover their annual energy needs (10,752 m^3 per household). Therefore, they rent and loan their land, as well as purchase of firewood. Regarding the access to land category, 82.14% is owned by men and 17.86% by women. However, this percentage of female holders does not imply that they can exercise their rights as owners, because the land is generally administered by men. This situation confirms that the access to land influences the availability of firewood and its energy use.

Study Limitations/Implications: The information provided by the interviewees was the basis for the analysis of the use and amount of firewood used by households; however, the interviewees may have underestimated or overestimated the amounts.

Findings/Conclusions: In the Reyeshogpan community, the gathering of firewood is carried out according to gender, based on the physical differences between men and women. Tasks specifically assigned to men are related to their physical strength, while the tasks performed by women are considered complementary and are not given any economic value.

Keywords: Gender, dendroenergy, household energy use.

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INTRODUCTION

Firewood is one of the main energy sources for the rural population in Mexico. According to data from the Instituto Nacional de Estadística y Geografía (INEGI, 2017), 20% of the population uses firewood to cook their food and heat their homes. Over time, the

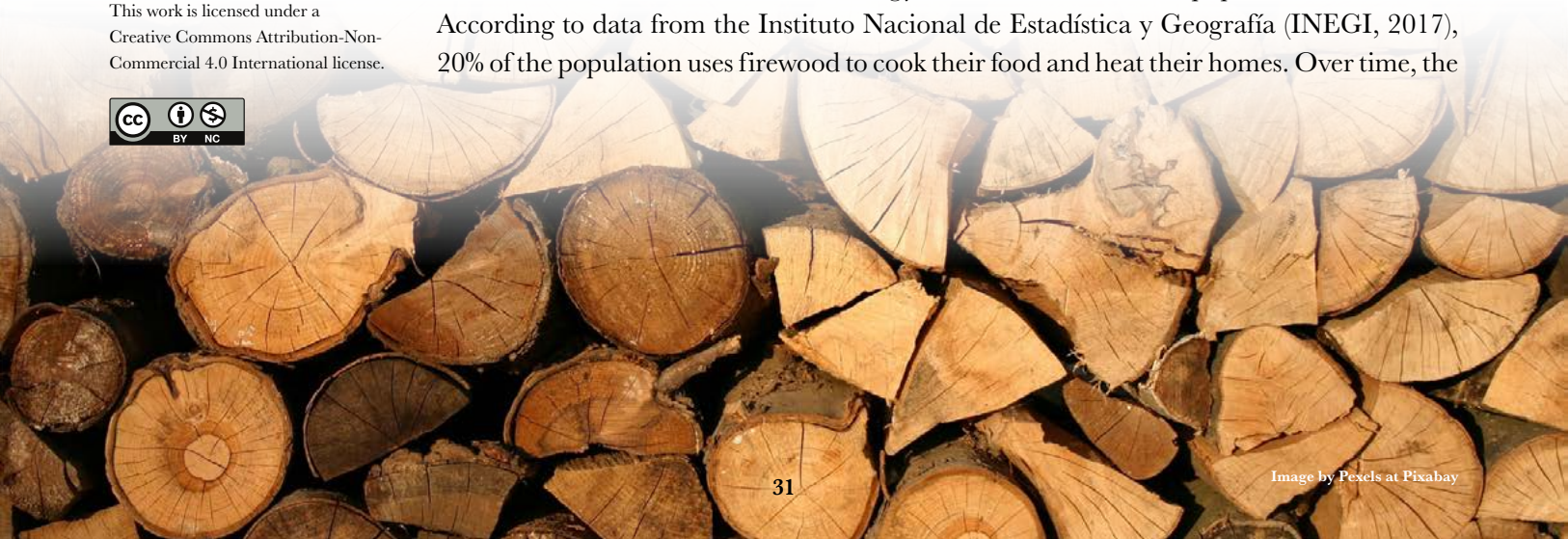

Image by Pexels at Pixabay

use of firewood has caused pressure on forest resources, generating availability issues and increasing the time that people spend gathering it. Studies carried out in two communities in Chiapas by Santos *et al.* (2012) conclude that the search and gathering of firewood falls mainly on women, since men only participate in this activity when they are not otherwise engaged. However, Escobar *et al.* (2009) indicate that the task of collecting firewood falls mainly on adult men; therefore, they conclude that this activity does not seem to be linked to gender.

Likewise, Soussan (1991) mentions that the use of this resource depends on the amount of firewood to which the population has access and the level of control they have over its sources. For rural households, land constitutes the main asset and is the means for food production, housing, and income generation. Access to land encompasses access to other natural resources; therefore, having rights to the former implies having rights to the latter. In this context, this research focused on the gender dynamics of the firewood use in the community of Reyeshogpan, Puebla, Mexico, in order to determine to which degree do the rights of use, access, and ownership of natural resources influence the consumption of firewood.

MATERIALS AND METHODS

During the field work of this qualitative research, a descriptive and interpretative analysis was carried out from the gender perspective, using the participant observation methodology, complemented with semi-structured interviews. This tool collects social, economic, access, resource use, and sexual division of labor data in households. During this stage of the research, four temporary stays were also carried out in the community (1st quarter of 2020). In the first visit, participant observation was carried out as a qualitative method and, in the remaining stays, n=24 semi-structured interviews were applied (12 women and 12 men) from a total of n=91 households in the community. A purposive sampling was used, based on the willingness of the inhabitants to participate in the research. Two complementary semi-structured interviews were applied to key informants, to determine the forest species used as fuel in the community and to stablish its organizational structure.

Description of the study site

Reyeshogpan de Hidalgo is a Nahua community, located in the municipality of Cuetzalan, Puebla (20° 00' N and 97° 29' W). It is part of the Sierra Norte de Puebla region and the northeastern socioeconomic region of the state. According to the INEGI (2020), its population is made up of 487 inhabitants, out of which 224 and 263 are men and women, respectively. In average, every household has five members.

The predominant economic activity of the community is agriculture in small agricultural units. There are no common areas for the use of natural resources or for land endowment. To obtain additional income, the inhabitants engage in several activities: trade, embroidery, firewood gathering, food production, and raising backyard animals for self-consumption. In addition, wage labor, subsidies, and remittances have become important income sources that satisfy household needs.

RESULTS AND DISCUSSION

The community uses firewood to carry out most of the domestic tasks (*e.g.*, cooking and making tortillas and drinks. It is used mainly for its local availability, apparent free access, and the difficulty in acquiring other energy sources. The average household owns 0.25-3.0 hectares, although most of the population (40.74%) owns less than 0.5 hectares, 37% has 0.5 to one hectare, 7.4% has one to two hectares, and only 14.8% has more than two hectares.

Regarding property ownership, 82.14% of the land belongs to men and only 17.86% is owned by women, who mostly acquire property as an inheritance.

The interviews revealed that 83.3% of the total households use firewood as the main and only source of fuel, while 16.7% combine it with liquefied petroleum (LP) gas. Regarding the technology used, 75% use a traditional stove as the only device or combine it others, 58.3% have an efficient stove, and 12.5% have a gas stove (Table 1).

The households that still use a traditional stove as the only device mention that the main reason why they have not changed this technology is that they do not have the financial resources to build an efficient stove, while others consider that cooking takes longer in other stoves and that food tastes different. Finally, households that have both traditional and efficient stoves combine their use to save time. Based on the sample, the average monthly consumption per household is 0.5 *tareas* (local measure consisting of thin firewood or firewood splits) equivalent to 10.752 m^3. The maximum monthly consumption per household is 1.5 *tareas*, which represents a volume of 32.25 m^3.

Identification of combustible species

To determine which species are the best for generating energy, the quality criteria used by the residents is "firewood that makes embers", which means that the best quality firewood is the one that burns quickly and completely, leaving no unburned residues. Based on this characteristic, Table 2 shows a list of 26 local species used as fuel.

Six out of the 16 most used species are in greater demand as quality fuel: *chalahuite* (24%), allspice (*pimienta*) and coffee (*café*) (19%), orange (*naranja*) (13%), and *cedro bambú* (6%). Since there are no common use areas in Reyeshogpan, firewood can only be obtained from their own plots, from rented plots or bought. Inhabitants who do not own land or whose plots do not cover their needs choose to make informal arrangements —such as *trato a medias*, a lease-like agreement on the use of natural resources. In this research, 57.14%

Table 1. Wood burning technologies used in the study community.

Type of technology	Use (%)
Traditional stove	44
Efficient stove	16
Traditional stove and efficient stove	28
Efficient stove and gas stove	12
Gas stove	12.5

Source: table developed by the authors.

Table 2. List of species used as firewood with good ember quality.

Spanish name	Scientific name	Nahuatl name	Quality of the ember
Pimienta	*Pimienta dioca* (L) Merril	Unidentified	Good
Caoba	*Swietenia macropylla* King	Ayacach	Good
Huaje	*Leucaena leucocephala* (Lam)	Huaxi	Good
Café	*Coffea arabica* L.	Unidentified	Good
Encino	*Quercus* spp.	Ahuatl	Good
Guayaba	*Psidium guajava* L.	Xalxokolt	Good
Chalahuite	*Inga edulis* Mart.	Chalahui	Good
Garrochilla	*Cuponia dentata* DC	Koetzalouit	Good
Sangregrado	*Crotón draco* Schl tdl	Eskouitl	Good
Palo mulato	*Bursera simaruba* (L.) Sarg	Chaca	Good
Cedro	*Cedrela odorata* (L.)	Tiokouitl	Good
Cocuite	*Glyricidia sepium* (Jacq.) Konth ex warp.	Couite	Good
Mango	*Manguifera indica* L.	Unidentified	Good
Xiloxochitl	*Pseudobombax ellipticum* Dugand	Unidentified	Regular
Jonote blanco	*Heliocarpus appendiculatus* Turez	Xonot	Regular
Maicillo	*Pleuranthodendron Lindenii* (Turez) Sleumer	Taolkoutl	Regular
Mamey	*Poutena sapota* (Jacq) H.B. Moore et Stearn	Koutzapot	Regular
Zapote negro	*Diospyrus diyna* Jacq	Tiltzapot	Regular
Carboncillo	*Persea* sp.	Alauak	Regular
Capulín	*Conostegia xalapensis* (Bonpl)	Capoli	Regular
Hormiguillo	*Cecropia ostusifolia* Bretol	Chikikis	Regular
Tarro	*Bambusoideae*	Unidentified	Regular
Xicalcuahuitl	*Alchornea latifolia* swartz	Xicalcuahuitl	Low
Matacaballo	*Trema micrantha* (L.) Blume	Totocouitl	Low
Mala mujer	*Cridoscolus multilobus* (Pax) I.M. Jonhston	Tetzonkilitl	Low
Anayo	*Beilsch miedia anay* (S.F. Blake) Kostermans	Anay	Low
Atzizikas	Unidentified	Atzizikas	Low
Zapote corona	Unidentified	Unidentified	Low

Source: table developed by the authors based on the interview with Mr. José Antonio Matamoros Hernández, from the community of Reyeshogpan, on March 21, 2020.

of the interviewees said that they obtain firewood from their own plots, 35.71% combine it with gathering, and 7.14% buy it.

Likewise, households that do not own plots or whose surfaces are insufficient mostly purchase firewood to meet their requirements. They either buy it on retail (bundles of firewood of 30-40 thin logs) or per volume (1,792 m^3 as trace or split). Households that buy firewood get it within the community (from their acquaintances and relatives) or from sellers in neighboring communities, including Tecoltepec, Xalcuahuta, Tepetitán, Santa Rosa, Huitziltepec, Amatlán, and Rancho Morelos. The firewood is directly commercialized between the producer and the consumer, who set its price according to supply and demand, the species in question, and the negotiation.

The average price of a *tarea* is MX$700.00 pesos, with minimum and maximum prices of $600.00 pesos and $800.00 pesos, respectively. The maximum price has been reported in periods of scarcity. The firewood that is sold includes a mixture of species; however, the price changes, if a specific type of firewood is required. The most valued species —based on their high potential heat and their quality— are allspice and orange, which report the highest market prices. The price of a *tarea* of either species ranges between MX$800.00 or MX$1,000.00 pesos.

The firewood gathering tasks are assigned according to physical strength. In this case, men are responsible for the so-called "heavy work" (*e.g.*, cutting, splitting, and transportation). Therefore, the greatest amount of firewood is gathered by men.

The women take part as "helpers." They carry out activities that require less physical effort, such as transporting and drying firewood. Occasionally women also gather firewood to complement the household's firewood demand.

The data found show that, in 86% of households, men responsible for the gathering, in 54% is a shared task, 39% report that other family members also participate, and 25% declare that they hire *jornales* (workers on a daily-wages basis).

Although, based on these percentages, men are directly responsible for the supply of firewood in most households, the participation of other members of the family is not totally passive and they get involved in the supply in some way or another, especially during the gathering season.

However, the increase in the hiring of *jornales* points out the changes that households have undergone as a consequence of the seasonal migration arrangements they make to obtain a higher income. Forty-six-point-six percent of the interviewees reported that they hire *jornales* to carry out activities related to the supply of firewood. The activities they pay for are cutting, splitting, and to a lesser extent transportation. All these paid activities are carried out by men.

The households that hire *jornales* for gathering activities indicate that the lack of a male labor force in the household forces them to take this measure. However, if they do not own the resource or there is a shortage, they must purchase firewood.

Therefore, the availability of resources constitutes a factor that directly interferes with the assignment of tasks within the household. Its composition determines the intensity of the tasks that each of its members must carry out.

The changes that take place when the sources of household income are modified do not only have an impact on the sexual division of labor, but also on the income. García and Dolors (1990) point out that the flexibility in the assignment of tasks proves that the division of labor does not occur naturally or as a consequence of the role that women have in the reproductive sphere. Instead, the forms in which a given social group organizes work are determined by the material and social conditions of the households, as can clearly be seen in the case of Reyeshogpan.

CONCLUSIONS

In the context of a community whose members continually migrate to neighboring towns in search of employment, whose plots are increasingly pulverized and measure less

than 0.5 ha, and where overexploitation of the resources has led to scarcity, the sustained demand for firewood has caused the community's main energy resource to enter a market that assigns a value to it and therefore it no longer is a free resource. The results of this research indicate that the availability of the resource directly depends on the rights of use, access, and ownership of natural resources. Households with less than 0.5 hectares do not meet their energy demand and therefore must buy firewood, which limits the amount the can spend on other basic needs. Regarding the sexual division of labor, "physical strength" is the factor that determines which sex is responsible for gathering firewood. Under this premise, men are responsible for this process and the rest of the household members complement the gathering sporadically. Households that do not have a male presence at the time of gathering must resort to hiring *jornales*. In conclusion, the gathering of firewood in the Reyeshogpan community is carried out according to gender, based on the physical differences between men and women: specific tasks are assigned to men based on their physical strength, while the tasks performed by women are considered complementary and do not have any economic value.

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Mexico's agricultural policy in the American context (1995-2020)

Olvera-Avendaño, Aminta[1]; Salas-González, José M.[1*]; Zavala-Pineda, María J.[2]; Martínez-Bautista, Humberto[3]; Abelino-Torres, Gonzalo[4]; Sagarnaga-Villegas, Leticia M.[1]; Leos-Rodríguez, Juan A.[1]

[1] Universidad Autónoma Chapingo, Km 38.5 Carretera México-Texcoco, Chapingo, Estado de México, México, C.P. 56230.
[2] Prepa en Línea - SEP Servicio Nacional De Bachillerato En Línea, Av. Revolución 1425, Tlacopac, Álvaro Obregón, Ciudad de México, CDMX, C.P. 01040.
[3] Centro de Investigación en Matemáticas CIMAT-AGS, Fray Bartolomé de las Casas 314, Barrio de la Estación, Aguascalientes, Ags., C.P. 20259.
[4] Universidad Autónoma Chapingo, Unidad Regional Universitaria de Zonas Áridas, Carretera Gómez Palacio-Chihuahua km 40, Bermejillo, Durango, México, C.P. 35230.
* Correspondence: jmsalasgonzalez@gmail.com.

ABSTRACT

Objective: To analyze the long-term relationship of two groups of agricultural policy instruments classified by the OECD-Producer Support Estimator (PSE) and General Services Support Estimator (GSSE)-OECD classification on Agricultural Gross Domestic Product (AGDP) in Mexico, USA, Canada, Chile and Brazil during the period 1995-2020, to generate information that contributes to the design of agricultural policies.

Design/Methodology/Approach: The information used in this work was developed by the OECD and was integrated into a time series for the 1995-2020 period. A quantitative analysis was carried out based on the econometric method, applying the cointegration test.

Results: The Canadian, Brazilian, and Mexican series are cointegrated, because the error of the model has a unit root (*i.e.*, individual variables are not of order I(0)); however, the combination of their variables show that the error is a process I(0), with a zero mean. However, the Chilean and USA variables were not cointegrated.

Study Limitations/Implications: An open market environment requires the development and implementation of policies that include the use of diverse and relevant instrument groups, guaranteeing that the resources transferred to the sector generate the expected results.

Findings/Conclusions: In comparison with the PSE, the GSSE has a closer long-term relation with the growth of the agricultural GPB in most countries; therefore, using this group of instruments to transfer resources to the sector is assumed to improve its performance to a greater degree.

Keywords: Cointegration, Producer Support Estimate, General Services Support.

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INTRODUCTION

The agricultural sector contributes to national income, employment, foreign trade, and its dependent and related industries (Escobar, 2016; OECD and FAO, 2019). This situation has laid the foundations for governmental intervention in agriculture, through national policies and support mechanisms (Schaelicke, 2019). Governments can adopt different measures to improve the productivity, sustainability, and resilience of the agricultural sector (OECD, 2020).

The OECD (2016) points out that the Producer Support Estimate (PSE), the General Services Support Estimate (GSSE), and the Consumer Support Estimate (CSE) quantify the subsidy transferred to the producers, general services, and consumers in the agricultural sector. This categorization system considers the following criteria for the implementation

and allocation of the transfers: producers, consumers, or the overall agricultural sector (Effland, 2011). The OECD does not only follow up the policies applied in each country, it makes recommendations that seek to provide policy makers with inputs for their decision-making (OECD, 2010). Among its policy prescriptions, the OECD (2001, 2013, 2022) has pointed out the importance of diminishing the application or even progressively eliminating PSE policies —because they include support mechanisms that directly distort production and trade— and increasing the use of GSSE (Morris *et al.*, 2020; OECD, 2018).

In recent years, the support for the agricultural sector (as a GDP percentage) has shown a downwards trend worldwide (Pawlak, 2018). From 2019 to 2021, the total support for agriculture in the 54 countries analyzed by the OECD amounted to an average of over $817 billion dollars per year. Seventy-five percent of this total was transferred to the PSE, while the remaining 25% was transferred in almost equal measures between the GSSE and the CSE (OECD, 2022). The Unites States and Canada are two of the most important markets for the exportation of Latin American food products (Gurria *et al.*, 2016). These two countries are also the main corn and wheat exporters worldwide (Espinosa-Cortés, 2022). The support percentage they provide to their agricultural sectors regarding their production value (28.9% for the USA and 13% for Canada) is higher than other countries in the Americas (OECD, 2022). Morris *et al.* (2020) point out that almost 25% of the world's agricultural and fishing exports come from Latin America and the Caribbean (LAC). Brazil has consolidated as the biggest exporter of agricultural and food products in the region, followed by Argentina, Mexico, Chile, Ecuador, and Peru (Escobar, 2016). Additionally, Mexico is one of the world's major importers of corn, soy, dairy products, pork, and poultry meat, while Brazil is one of the major wheat importers of the world (OECD and FAO, 2019). Currently, Chile is the sixth most important fruit exporter of the world and is the South American leader in this sector (Boza *et al.*, 2020). For its part, Mexico has historically reported the highest support levels in LAC (Gurria *et al.*, 2016). The OECD (2022) reported that, regarding the value of its production, Mexico provided a 10.8% support to its agricultural sector, while Chile and Brazil provided a 9.7% and 3.8% support, respectively.

The relevance of each of the abovementioned countries for the global agricultural trade justifies the comparative study of their agricultural policies. The behaviour of the agricultural GDP has been found to have a long-term relationship with the general services support policy, unlike the producer support. Therefore, the objective of this work was to analyze the long-term relationship (1995-2020) between two sets of agricultural policy instruments (PSE and GSSE) and the agricultural GDP in Mexico, USA, Canada, Chile, and Brazil, in order to generate information that contributes to the design of agricultural policies. The CSE policy instrument was not taken into consideration in this analysis, since this group generally has a negative impact as an implicit tax on market prices, although it compensates the food subsidies for the consumer (OECD, 2004).

MATERIALS AND METHODS

The analysis was performed with information from the OECD, which was integrated in an Excel® database, creating a 26-year time series (1995-2020) based on data from Mexico,

USA, Canada, Brazil, and Chile. The database includes information about the agricultural GDP and the amount transferred to the PSE and GSSE of the analyzed countries.

The amounts were determined at nominal prices and in national currency; therefore, the exchange rate and USA inflation rate available in the database of the World Bank (2021b, 2021a) were used to deflect the values to the 1995 Monetary Base. The Stata statistical software (Release 17) was used to carry out the statistical estimates (StataCorp, 2017).

Definition of variables

The support instruments of the agricultural sector are classified in three groups, based on the implementation methods and its type of users: producers, consumers, and services provided to the agricultural sector (Effland, 2011). This research only included the following variables: agricultural GDP, PSE, and GSSE (OECD, 2016). The GSSE is considered to be the agricultural support mechanism that less distorts the market, unlike the PSE (Table 1).

Times series – cointegration test model

One of the major problems with time series regressions is senseless or spurious results (Gujarati and Porter, 2010). A way to avoid this problem is to determine if the times series are cointegrated (Jordan and Philips, 2018). Additionally, cointegration can be applied with two series of order I(1), although their lineal combination is I(0) (Wooldridge, 2009). Consequently, the result of the regression is not spurious and the series has a long-term relationship (Gujarati and Porter, 2010). The GSSE is expected to be cointegrated, since the OECD (2018, 2022a) points out that supports for this kind of instruments do not have a direct impact on agricultural income or consumer expenses; however, they do affect long-term agricultural production or consumption. The following model was used to meet the objective of the analysis:

$$PIBAg_t = \beta_0 + \beta_1 EAP_t + \beta_2 LEASG_t + t + \mu_t$$

Where $PIBAg_t$ is the time series for the agricultural GDP; EAP_t and $EASG_t$ are the series of producer support and general services support policies, t refers to the lineal trend; and μ_t, is the term of the error. Nevertheless, even if individual variables are assumed to be

Table 1. Description of variables. Source: (OECD, 2016).

Variable	Concept	Description
GDPAg	Agricultural GDP	Market value of all final goods and services produced by the agricultural sector.
PSE	Producer Support Estimate	The annual monetary value of gross transfers from consumers and taxpayers to agricultural producers.
GSSE	General Services Support Estimate	The annual monetary value of gross transfers arising from policy measures that create enabling conditions for the primary agricultural sector through development of private or public services, and through institutions and infrastructures.

Source: OECD (2016).

non-stationary [I (0)], a lineal combination of two or more time series can still be stationary (Wooldridge, 2009). Therefore:

$$\mu_t = PIBAg_t - \beta_0 - \beta_1 EAP_t - \beta_2 LEASG_t - t$$

Consequently, the error of the model can be cointegrated, representing a long-term balance (Gujarati and Porter, 2010). The expression of the long-term balance relationship between the variables can prove that the clash in one of them alters the behaviour of the other to a similar degree (Rios, 2014). A simple test to analyze the cointegration is the application of the Augmented Dickey Fuller test to the residues estimated from the cointegrating regression (Gujarati and Porter, 2010; Jordan and Philips, 2018; Montero-Granados, 2013). Logarithms (both in the dependent and the independent variables) were calculated to limit the range of the variables to a smaller amount and to reduce the sensibility of extreme or atypical observations (Schuschny and Soto, 2009).

RESULTS AND DISCUSSION

Brazil has the most dynamic agricultural sector among the countries analyzed. Its initial annual growth was 5% (1995), but by 2020 it had reached 5.09%, which indicates an average annual growth of 0.63%. Meanwhile, the annual growth of other countries diminished throughout the same period. Overall, the support for the agricultural sector has diminished (Figure 1). However, Mexico and the United States recorded higher support levels (0.78% and 0.54%, respectively) than the other countries regarding their GDPs. Producers support is the instrument group with the highest amount (>70%) of the total support for the agricultural sector. Canada and Mexico allocated a little more than 13% of their gross agricultural receipts to this group of instruments (% PSE) from 1995 to 2020; for their part, Brazil and Chile provide the lowest support (2.9% and 4.5% of their gross agricultural receipts, respectively).

Gurria *et al.* (2016), the OECD (2022a), and other sources report that support for market price prevail in the PSE, influencing the said prices. This kind of instruments are considered the least efficient, because they influence the price that the producers obtain (Nguyen and Grote, 2018; Orden *et al.*, 2007; Pawlak, 2018). Additionally, they generate a reduction of importation products and increase subsidized exportations, diminishing world market prices (Dewbre *et al.*, 2001).

Subsidies for variable inputs (*e.g.*, energy and fertilizers) stand out among the PSE supports provided by Chile, Brazil, and Mexico. An important element of Brazil's producers support policy has been the Programa Nacional de Fortalecimiento da Agricultura Familiar (PRONAF), which has contributed to the strong expansion of the agricultural sector, through the strengthening of credits that stimulate agricultural investments (Guanziroli, 2014; Gurria *et al.*, 2016; Trentin and Quaresma, 2022). Overall, producers support is still relevant, while general services support still has minimal importance in the budget transfers of the countries analyzed.

The highest GSSE percentage regarding the total support for the agricultural sector was recorded in Chile (average: 30%) in the period under study. This group of instruments

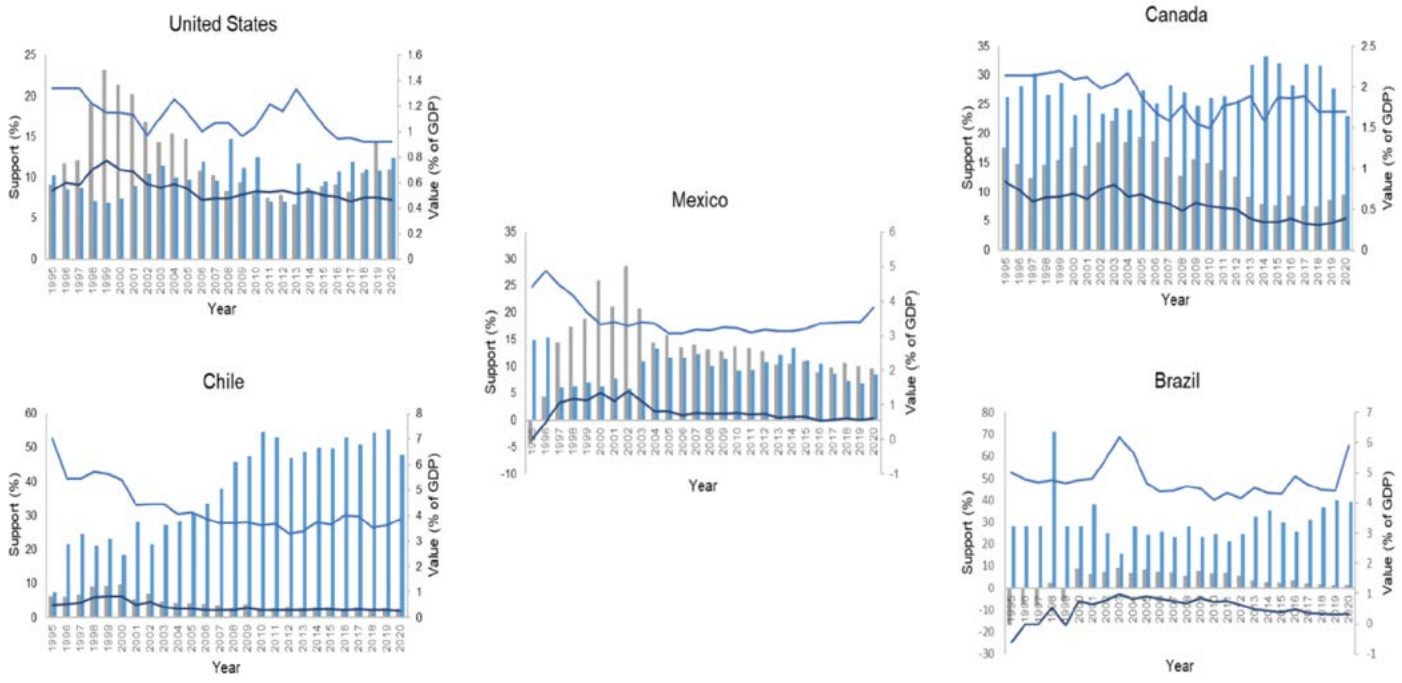

Figure 1. Agricultural GDP Growth, Total Agricultural Support (TSE), PSE (% of Gross Farm Receipts), and GSSE (% of TSE). Source: Figure developed by the authors based on data from the OECD (2022b).

diminished by 2% in Mexico, while in Chile the budgeted amount increased by 7% (annual average for 1995-2020). The OECD (2022a) points out that, with regard to the size of the sector, the general services expenses diminished from 5.4% to 3.5% of the agricultural production value; these results suggest that these expenses did not follow the growth pace of the sector. Morris *et al.* (2020) mention that the public expenditure in support of agriculture in Brazil and Chile has been focused on agricultural research, outreach services, and plant and animal inspection services.

Cointegration test

The results show that the Canadian, Brazilian, and Mexican series are cointegrated, since the error of the model has an unit root (*i.e.*, the individual variables are not of order I(0)). However, according to Gujarati and Porter (2010), Rios (2014), and Wooldridge (2009), the combination of variables shows that the error is a process I(0) with a zero mean.

This phenomenon expressed the long-term balance relationship between the variables and indicates that a clash in one of the variables alters the behavior of the agricultural GDP to a similar degree. For their part, the variables in Chile and the USA were not cointegrated. The results show that the growth of the agricultural GDP of Chile and the USA has not been influenced by the PSE and the GSSE (Table 2); therefore, it could be subject to other elements that benefit the dynamics of the sector in those countries. In Mexico and Canada, the GSSE policies were closer to $p<0.05$. In Brazil, the PSE and GSSE variables were significant ($p<0.05$).

Table 1. Parameters obtained from the cointegrating regression.

Country	Variable	Coefficient	t	P>t	R^2	DW	DF	Portmanteau (Q)
Mexico	LnPSE	0.019	0.520	0.607				
	LnGSSE	0.049	1.160	0.260				
	Time	0.011	7.150	0.000				
	Constant	9.428	33.400	0.000				
	Error L.1	−0.756	−3.640	0.001	0.347	1.805	0.002**	0.062
	Constant	−0.001	−0.090	0.927				
Brazil	LnGSSE	0.113	2.890	0.010**				
	Time	0.496	8.900	0.000***				
	Constant	0.040	12.820	0.000				
	Error L.1	5.518	14.270	0.000				
	Constant	−0.815	−3.540	0.002	0.378	1.667	0.001**	0.167
	LnGSSE	−0.006	−0.300	0.767				
Canada	LnGSSE	−0.124	−0.680	0.503				
	Time	0.424	1.900	0.070				
	Constant	0.015	4.230	0.000				
	Error L.1	7.528	7.840	0.000				
	Constant	−0.723	−3.830	0.001	0.363	1.993	0.000***	0.309
	LnGSSE	−0.008	−0.350	0.732				
Chile	LnGSSE	0.391	2.090	0.048				
	Time	0.077	0.620	0.539				
	Constant	0.028	2.560	0.018				
	Error L.1	5.511	5.730	0.000				
	Constant	−0.399	−3.350	0.003	0.298	1.662	0.225	0.000
	LnGSSE	−0.020	−0.990	0.333				
USA	LnGSSE	0.016	0.060	0.956				
	Time	−1.555	−3.370	0.003**				
	Constant	0.048	3.430	0.002				
	Error L.1	23.781	5.070	0.000				
	Constant	−0.570	−2.590	0.016	0.192	1.748	0.091	0.574
	LnGSSE	−0.005	−0.100	0.920				

*p<0.05 **p<0.01 ***p<0.001. Source: Own elaboration with outputs from STATA.

Therefore, the design and application of policies focused on general services support (GSSE) are important, because the results indicate that they have the greatest influence in the behaviour of the sector —*i.e.*, a clash in this group of policy instruments alters the behaviour of the agricultural GDP to a similar degree. Bielik *et al.* (2007) and the OECD (2018) stress the importance of guaranteeing the availability of public services, because they benefit society as a whole (*i.e.*, producers and consumers). The services provided can include efficient systems that improve the sector, through agricultural innovation systems,

research, education, appropriate infrastructure, health, and quality control of agricultural products (Arisoy, 2020; Morris *et al.*, 2020; Sánchez *et al.*, 2022).

CONCLUSION

In most of the analyzed countries, the group of policy instruments for the general services support have a long-term relationship with the agricultural GDP. A clash in this group of policy instruments can alter the agricultural GDP to a similar degree. According to the OECD, this group of policy instruments has a long-term relationship with the behaviour of the agricultural GDP. Therefore, the use of this group of instruments should be emphasized within agricultural policy. The results can be taken into consideration for the redesign of national agricultural policies and for the compliance with the international commitment to deregulate the world's agricultural market. Further studies aimed at the identification of the dynamism factor of the Chilean agriculture should be carried out, since the variables employed did not show cointegration.

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Horses' welfare during transport

Vázquez-Díaz, Rigoberto[1]; Ortega-Cerrilla, María Esther[1*]; Bárcena-Gama J. Ricardo[1]; Ramírez-Bribiesca, J. Efrén[1]; Pérez-Rodríguez, Paulino[2]; Lombardero-Goldaracena, Germán[3]; Ayala-Rodríguez, Julio M.[1]

[1] Colegio de Postgraduados, Posgrado Recursos Genéticos y Productividad-Ganadería, Carretera México-Texcoco km 36.5, Montecillo, Estado de México, CP 56264.
[2] Colegio de Postgraduados, Posgrado de Socioeconomía, Estadística e Informática, Colegio de Postgraduados Carretera México-Texcoco km 36.5, Montecillo, Estado de México, CP 56264.
[3] Universidad Autónoma Metropolitana, Xochimilco. Calzada del Hueso 1100. Colonia Villa Quietud, Alcaldía Coyoacán, C.P. 04960.
* Correspondence: meoc@colpos.mx

ABSTRACT
Objective: To review the literature related to horses' welfare during transport.
Approach: Transporting horses is a common practice in the equine industry and obeys to different reasons and activities. Therefore, understanding the factors that intervene during transport and management of the horses is critical.
Limitations on study/implications: Horses are the most transported animals globally, this being a very stressful event. Different factors affect their welfare during this process, such as the type of vehicle, the driver's expertise, the length of the trip, the orientation of the animal in the vehicle, lesions and diseases.
Conclusions: The factors involved in horses' transport should be known and taken into account, to ensure the horses' welfare.

Keywords: horses' transport, horses' welfare, stress.

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INTRODUCTION

Horses are transported more than any other type of livestock, for different reasons: races, exhibitions, breeding, sale, or sacrifice (Riely *et al.*, 2022). It is a stressful event that implies the separation from their familiar environment and mixing with unknown animals. Mixing can cause aggressions between the animals (Hartmann *et al.*, 2009). The groups ought to be limited and from the same place, mares with offspring should be kept together with added space, and the studs ought to be separated from other males (Roy, 2014). During transport, horses are exposed to confinement, vibrations, inadequate ventilation, changes in temperature and moisture, and they usually do not receive food

Image by Thomas G. at Pixabay

and water (Wara, 1993; Dai *et al.*, 2021). Transport is among the main causes of lesions during loading, transport and disembarking; the physical stressors include movement of the vehicle, characteristics of the floor, environmental moisture, temperature, and restricted space (Giovagnoli *et al.*, 2002). The animals can be transported by ship, train, truck or plane and this depends on the destination and objective, for example, for competition, reproduction or sacrifice.

Horses began to be transported 3500 years ago. Xerxes moved his cavalries in Ancient Greece. Hannibal transported his horses through the Rhone River in 218 BC. During the 14th century, the horse trade between England and the rest of Europe increased, and at that time the horses were transported primarily by ship. In the 18th century transport was primarily by land and in the 19th century the horses were transported by train (Friend, 2001; Warran *et al.*, 2007). Currently the most common means of transportation is by land and air for long distances.

Many countries regulate horses' transport. Europe has the EC Regulation No 1/2005 for animal protection during transport (EPRS, 2018). In the United Kingdom, there is regulation for the welfare of animals during transport, especially of horses, ponies and other domestic equids (Defra, 2006). In USA, eleven states have laws that regulate horses' transport prohibiting the use of two-story trailers (Wish, 2014). There is also a practice code for their transport in Australia (Department of Agriculture, 2003).

Factors that influence the welfare of horses during transport

Different factors affect horses' welfare. The preparation of the horse and the vehicle can generate stress, affecting the horse's health and causing economic losses (Oikawa *et al.*, 2004; Padalino *et al.*, 2017b). Social isolation, age, sex, familiarity with the other animals that travel, the driver's expertise, duration of the trip, state of hydration, ventilation, air quality, temperature and moisture, prior experiences with transport, medications, the animal's temperament, restraint methods, as well as the place where the horse travels, in the front or back of the vehicle, and the road conditions, can affect horses' welfare (McGreevy, 2012; Nivelle *et al.*, 2020). Stress can favor lesions and fractures (Ferguson and Rosale-Ruiz, 2001; Mansmann and Woodie, 1995), as well as respiratory (Austin *et al.*, 1995) or gastrointestinal infections (McClintock and Begg, 1990).

Loading tends to be one of the most stressful events during transport, since by nature horses are neophobic (Dai *et al.*, 2021) and they present fear in face of new experiences. Most of the lesions in extremities at the time of loading are associated to the ramp (Dai *et al.*, 2021), so it is important accustom the horses to loading when they are foals to prevent behavior problems. During transport, horses tend to be restrained. In this regard, it is important for the distance of the cord between the tying point and the halter to be enough for the horses to lower their head freely. Restraining the horse without allowing it to keep its head low is associated with the increase in respiratory infections (Stull and Rodiek, 2002). During transport, the horses try to gain balance in the presence of changes in speed and direction, moving their head and legs quickly, which can cause leg wounds as they lose their balance when the vehicle stops, turns or accelerates abruptly; the noise and other factors also stress them, so it is very important to have an adequate

design of the vehicles, since it is one of the highest risks (Colborne *et al.*, 2021; Riley *et al.*, 2018; Giovagnoli *et al.*, 2002).

The dynamics of the vehicle will depend on the driver, road conditions, climate and other factors during the route (Riley *et al.*, 2016), and in the effort to adjust their posture horses can record muscular and emotional stress (Warran, 1993; Padalino, 2015b). The correction in the horse's posture can be insufficient to avoid lesions or distress, which will depend on the physical and emotional ability of each animal to respond to the stressful factors (Colborne *et al.*, 2021). The physical and behavioral problems are also affected by the driver's behavior, the inability of the horse to face unexpected movements of the vehicle, and the lack of outfitting of the vehicles taking into account the horses' welfare (Padalino *et al.*, 2018; Riley *et al.*, 2018).

Type of vehicle

Horses destined to sports and to reproduction are frequently transported by air, although transport by road is often used. In a study carried out with six horses (five castrated males of Durch Warmblood race and a Belgian Warmblood female, of approximately 9.2±2.1 years) that were transported by land and air, they showed variations in heart rate. The trip consisted of 24 h of rest in the stable, 4.5 h of travel by road, then 5.5 h of wait in a stable and, then, 11 h of travel by plane. The heart rate was significantly higher during air transport than during rest in the stable (Ohmura *et al.*, 2012), reaching the conclusion is that air transport caused a slight increase in the heart rate than the transport by road (Munsters *et al.*, 2015).

Duration of the trip

Sometimes horses are transported for long distances, exposing them to health and welfare problems (Marlin *et al.*, 2011). In a study with 180 horses transported between Sydney and Perth (4000 km), 97.2% of the horses arrived without signs of disease or lesions. The animals that presented health problems were diagnosed with respiratory problems (27%), gastrointestinal problems (27%), pyrexia (19%), traumatic lesions (15%) and death (12%). The duration of the trip and the season have a significant effect. During the spring and in trips of more than 20 h, respiratory problems and deaths increased ($p<0.05$). Long trips increase the health problems and compromise the horse's welfare (Paladino *et al.*, 2015a).

Some physiological parameters were studied in horses between 4 and 10 years old transported in Poland. Blood samples were taken 8 h before transport and 24 h after having disembarked. The values of urea, cholesterol, creatinine, magnesium and alkaline phosphatase were similar. The albumin, total protein, aminotransferase alanine, aminotransferase aspartate, total bilirubin, kinase creatinine, and triglycerides were different as a result of transport. The metabolic response was lower in horses that exercised moderately or were transported for 24 h (Niedzwiedz *et al.*, 2012).

In two different trips with distances of 50 and 200 km with duration of 1 and 3 h, respectively, the blood of 12 Standarbred horses was analyzed, at rest, during the loading and disembarking, and 2 and 4 h after returning from the trips. The following were analyzed:

mean cell volume, cortisol, transaminase aspartate, transaminase alanine, creatinine, glucose, triglycerides, cholesterol, ureic nitrogen in blood, calcium, phosphorus, chlorine, total proteins, albumin, and alkaline phosphatase. The heart rate, respiratory frequency and body temperature were also measured. The loading always increased the respiratory frequency and the mean cell volume. In both cases, the distance of the trip increased the glucose, cortisol, mean cell volume and heart rate, while the kinase creatinine increased two hours after the return from the trip of 200 km. The water intake was higher in horses that travelled 200 km compared to those that travelled 50 km.

The adaptation to a new stable was easier when food and water were offered after arriving, and one of the conclusions of this study was that horses should arrive at least four hours at their destination before they start physical activity (Tateo *et al.*, 2012).

Stress is associated with long trips and the increase in the time of transport; however, it can also increase in short trips. Fazio *et al.* (2008) measured the ACTH hormone, β endorphin and cortisol in studs travelling different distances (100, 200 and 300 km). The β endorphin increased ($P<0.01$) after 100 km, the ACTH increased ($P<0.001$) after transport of 100 and 200 km, and the levels of cortisol were higher ($P<0.001$) after 100, 200 and 300 km of transport by road. Wessely-Szponder *et al.* (2015) observed the physiological, immunological and metabolic changes that are produced in young and older horses during short and long distance trips. The study was carried out in 24 mares divided into four groups. Six female colts of 12-18 months and six mares of 6-12 years were transported 500 km. Blood samples were taken before, immediately after transport and during sacrifice.

The samples were analyzed to determine fibrinogen, malondialdehyde (MDA), aminotransferase aspartate, and kinase creatinine. The fibrinogen increased especially in the female colts after the long-distance transport, reaching the maximum level during sacrifice. Changes in the muscular enzymes were produced and the level of MDA also increased; the parameters of oxidative stress and the muscular lesions were related to the age and the distances in transport.

Seven castrated males and seven females, 5 to 15 years old, with the same body condition were studied in a trip of 4000 km. The heart and respiratory rate, rectal temperature, number of neutrophils, serum albumin and antioxidant state of the plasma increased. The results showed that long distances favor the induced response mediated by deteriorated cells (Padalino *et al.*, 2017a).

Horses have been given vitamin C to reduce the stress. Although horses can synthesize it from glucose or galactose, it cannot be stored in large quantities and it is used in prolonged stressful situations such as transport, which can favor the presence of diseases such as shipping fever. Ralston and Stives (2012) supplemented weaned colts with vitamin C during 5 and 10 days after 50 h of transport, and they found that supplementation with this vitamin for five days was adequate yet prolonged supplementation is not advisable.

Orientation of the horse in the vehicle

Gibbs and Friend (1999) researched the preferred orientation of the horses transported by road. The horses were tied to the left side of the trailer, to the right, or unleashed. The

horses tied to the right spent 59% of the time looking forward, the horses tied to the left 52% of the time looking backward, and unleashed horses 57% of the time looking forward and 35% looking backward. However, there was great variation between the horses.

Kusunose and Turikai (1996) studied the behavior and orientation of the horses that travelled unleashed. They observed that while the vehicle was in movement, the horses reduced food consumption and spent more time standing than when the vehicle was parked, in addition to increasing the frequency of animals looking backward. Smith *et al.* (1994) studied four horses that were left unleashed in the vehicle during transport, allowing them to choose the orientation; the results showed that they spent significantly more time looking to the back during a trip of 32 km by road. Horses can prefer three positions: looking forward, looking backward or at an angle (Colborne *et al.*, 2021). Different studies have attempted to determine the effect of the orientation on the ability of the horses to maintain their balance during transport and the results are contradictory due to the differences in the vehicle's design and performing simultaneous comparisons (Toscano and Friend, 2001). The orientation in which the horses travel does not significantly affect the ability of the horses to keep their balance (Fried, 2001), although it seems that travelling with their back to the direction of the trip offers allows them to maintain their balance better (Clark *et al.*, 1993).

Lesions and diseases

Some diseases, such as equine herpes virus, salmonellosis and pneumonia can increase during transport (Mc Greevy, 2012); in addition, lesions such as fractures, swelling, abrasions and hematomas. Roy *et al.* (2019) evaluated the hematomas ante-mortem after transport to a slaughterhouse using digital infrared thermography and they observed a significant percentage of horses with lesions on the skin, although this methodology had a modest sensitivity for the detection of hematomas.

Dehydration is also frequent, due to the lower intake of water and because it is rarely offered during transport and has been related to acute laminitis and colon impacting. Friend (2000) evaluated the effects of transport on the hydration status of the horses and found that the horses transported for 30 h showed a dehydration status higher than 10%; the study indicated that transport longer than 24 h causes a marked state of dehydration and extreme dehydration after 28 h of travelling with temperatures of 24 to 37 °C and relative humidity conditions of 32 to 94.

The horses subjected to trips for more than 24 h face higher risk of developing serious diseases or even dying (Padalino *et al.*, 2017). Sometimes they supply electrolyte solutions to avoid gastrointestinal impact, yet it is important to supply electrolytes directly into the animal's mouth and not in the water. Two or three percent dehydration can affect the yield during the competitions. The water supply during transport, especially when it is warm, prevents dehydration and stress (Mc Greevy, 2012).

It is important to keep the horses adequately hydrated so it is recommended to offer water at least every two hours of travel, and preferably from the place of origin, so there should always be water deposits. The restriction both of food and water during transport can trigger negative emotional responses (Roy, 2014).

Respiratory diseases are related to transport (Oikawa *et al*., 2004); prior sub-clinical respiratory diseases and restraining with the position of the head upwards, which prevents the mechanisms of pulmonary cleaning, can favor these diseases. The stress that affects the immune system, the presence of toxic gases, the high concentrations of dust and bacteria in the air, the orientation of the animal during transport, the duration of the trip, and the body status can also cause the disease.

Oikawa *et al*. (2005) studied the orientation of the head (to the front or to the back), the periods of rest (30 min or 2 h per 4 h of travel, or 1 h every 5 h of travel), and the state of cleanliness of the truck used for horses' transport along 1500 km; in two seasons of the year, April and August. The results showed that the increase in rest time and cleanliness inside the truck were the main factors to prevent respiratory diseases.

Horses' transport destined to sacrifice

The commercial transport conditions of horses for meat jeopardize their welfare and lesions are frequently generated, causing both physiological and behavioral disorders (Marlin *et al*., 2011; Roy *et al*., 2015a). Globally, 50% of the horses transported for sacrifice suffer lesions (Roy, 2014). The conditions of management and transport cannot be compared with the horses that are transported with the purpose of sports and leisure; however, in these animals the transport also generates negative conditions in their health and yield (Leadon, 1994). In many countries, horse meat is a common food. In France, Italy, Germany, Austria, Belgium, Netherlands, Iceland, Norway, Sweden, Poland, Ukraine, Russia, Hungary, Bulgaria and Slovenia its consumption is common. In the United Kingdom, the sacrifice and consumption are not prohibited, although it is infrequent since the 1930s. Nowadays, horses are protected from their export for sacrifice (Warran, 1993). Horse meat is also consumed in many countries of Asia (China, Japan, Indonesia, South Korea, Kazakhstan, Kyrgyzstan and Mongolia). Although in most countries horse meat is not consumed, in Canada horses are sacrificed to obtain meat. In the United States, this meat is not used for human consumption, and plants to sacrifice horses are prohibited since 2007 (Roy and Cockram, 2015). However, the horses are sent to Canada and Mexico to be sacrificed (Stull, 2012). Mexico is one of the main producers of horse meat; it is exported but not consumed. Since 1960, Mexico is characterized for remaining in the first places as producer of this meat (Jastrzębska *et al*., 2019; FAOSTAT, 2020). In South America, it is the main source of meat for indigenous peoples of Chile; and Argentina is producer and exporter, although the meat is not consumed.

The annual consumption of horse meat is 500,000 t on average (Belaunzaran, 2015). However, it has increased to 700,000 t annually due to the increase in production in Asia and Europe. Horse meat represents 0.25% of the total meat consumed in the world. Asia is the main producer of horse meat (46%), followed by America 30%, Europe 18%, Oceania 4% and Africa 2% (Belaunzaran *et al*., 2015). It is estimated that 800,000 horses are sacrificed globally each year (Padalino and Riley, 2020).

The largest transporter and producer of horse meat in the world is China; others are Kazakhstan, Mexico, Russia and Argentina, which represent 58% of the world production of horse meat. The main consumers are Mongolia, Kazakhstan, Switzerland, Italy and

Russia. The composition of horse meat is similar to that of beef; it is composed of 68-70% water, 22% protein, 0.5-6% fat, and around 1.5% minerals (Venegas and Gutiérrez, 2016). Some factors that affect its quality are race, age, sex, live weight, months to weaning, diet and months of fattening (Belaunzaran *et al.*, 2015).

Sometimes, horses that will be sacrificed travel unleashed in the vehicles towards the slaughterhouse; in addition, the number of animals transported is variable. In this case, horses can suffer falls during the trip and end up hurt or dead, or there can be aggressive interactions between the horses. It is frequent for them to have to travel long distances without food and water, which causes adverse results in relation to the welfare, such as excessive dehydration, thirst, hunger and fatigue (Roy and Cockram, 2015).

In a study carried out by Stull (1999) in horses transported to slaughterhouses for long distances (96 to 2496 km), an average weight loss of 4% was observed in two types of commercial trailers (trailers with straight cover or livestock trailer). The type of trailer used affected the percentage of horses hurt. The percentage was higher ($P<0.05$) in the case of the livestock trailers (29.2%) than in those with straight cover (8%).

The stress, indicated by the level of cortisol and the *neutrophil:lymphocyte* rate, as well as the rectal temperature were higher ($P<0.05$) in a surface of 1.14 to 1.31 m^2 per animal than in those of 1.40 to 1.54 m^2. The physiological responses, such as the recount of white blood cells, the total concentration of proteins, and the *neutrophil:lymphocyte* rate were lower in horses with a greater surface per animal.

Nivelle *et al.* (2020) observed the conditions of the horses transported for sacrifice in Argentina and Uruguay. The horses were transported during 294 ± 153 min at a distance of 295 ± 250 Km, with a surface of 1.40 ± 0.33 m^2 per horse.

The study included 23 horse transports from loading to the slaughterhouse with 596 mestizo horses during one year. The results showed that loading and disembarking were carried out without problems. However, they found that it is necessary to improve the training of drivers. The soil surface should also be levelled with the loading ramp, to avoid confusion in the horses. They found that often animals were transported during the warmest hours of the day and that animals of different ages were not separated. The conclusion was reached that the degree of aggressiveness was not associated to the characteristics of the environment or transport, but rather to the animals loaded in the back or front spaces. It is very important to also consider that the behavior of the drivers towards the animals during loading, travelling and disembarking is affected by the payment they receive for the transport (Broom, 2007).

Transportation affects the horses' welfare, as seen in a study conducted in Iceland. The horses travelled between 0.33 and 3.10 h; the animals were healthy and no wounds were seen before transport. However, after transport, 1.6% of the horses presented superficial bleeding wounds, they were stressed and the respiratory frequency and blood lactate were high, which decreased the blood glucose. Of the adult horses, 45% and 17% of the colts presented contusions. The horses also showed a slight dehydration. These observations suggest that the manipulation, transport and stabling were inadequate and should be improved (Roy *et al.*, 2015b). There is evidence of the scarce welfare of horses transported for sacrifice, which show dehydration, severe lameness, lesions and even death, in addition

to the grave lack of compliance of the EC no. 1/2005 about the protection of animals during transport (Marlin *et al.*, 2011).

CONCLUSIONS

Transport is a stressful activity for horses where different factors intervene. Therefore, there should be suitable management of the animals during the different stages of transport, ideal vehicles in excellent conditions, and training and adequate salary for the drivers.

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Colegio de Postgraduados

Effect of silicon and humic substances on the productivity and absorption of minerals in cucumber

Osuna-Zárate, Alberto Paul[1]; Robledo-Torres, Valentín[1*]; Mendoza-Villarreal, Rosalinda[1]; Sandoval-Rangel, Alberto[1]

[1] Universidad Autónoma Agraria Antonio Narro, Calzada Antonio Narro1923, C.P. 25315, Saltillo, Coahuila, México.
* Correspondence: robledo3031@gmail.com

ABSTRACT

Objective: To evaluate the effect of potassium silicate and humic substances in the mitigation of salt stress in cucumber grown in a greenhouse.

Design/Methodology/Approach: A completely randomized design with factorial arrangement was used. The first factor was the cucumber varieties (Var): Induran (I) and SV2516 (SV). The second factor was potassium silicate (Si), with doses of 0, 10, and 20 $mL \cdot L^{-1}$. The third factor was humic substances (SH), with doses of 0, 10, and 20 kg/ha.

Results: The best interaction for the NFP and Y variables was SV*SH20. The N and Ca content in the fruit was I*SH10, while the K and P content was SV*Si20. Meanwhile, Mg, Fe, and Cu interactions stood out with SV*Si10, SV*Si20, and I*Si10, respectively. The best interaction in leaf mineral content was I*SH20 (N), SV*SH10 (P and K), and SV*Si20 (Cu).

Study Limitations/Implications: There was no significant difference in Ca, Mg, and Fe in leaf.

Findings/Conclusions: At least one of the interactions between cultivars and bio stimulant doses favored agronomic traits, quality, and mineral absorption in fruit.

Keywords: salt stress, humic substances, potassium silicate.

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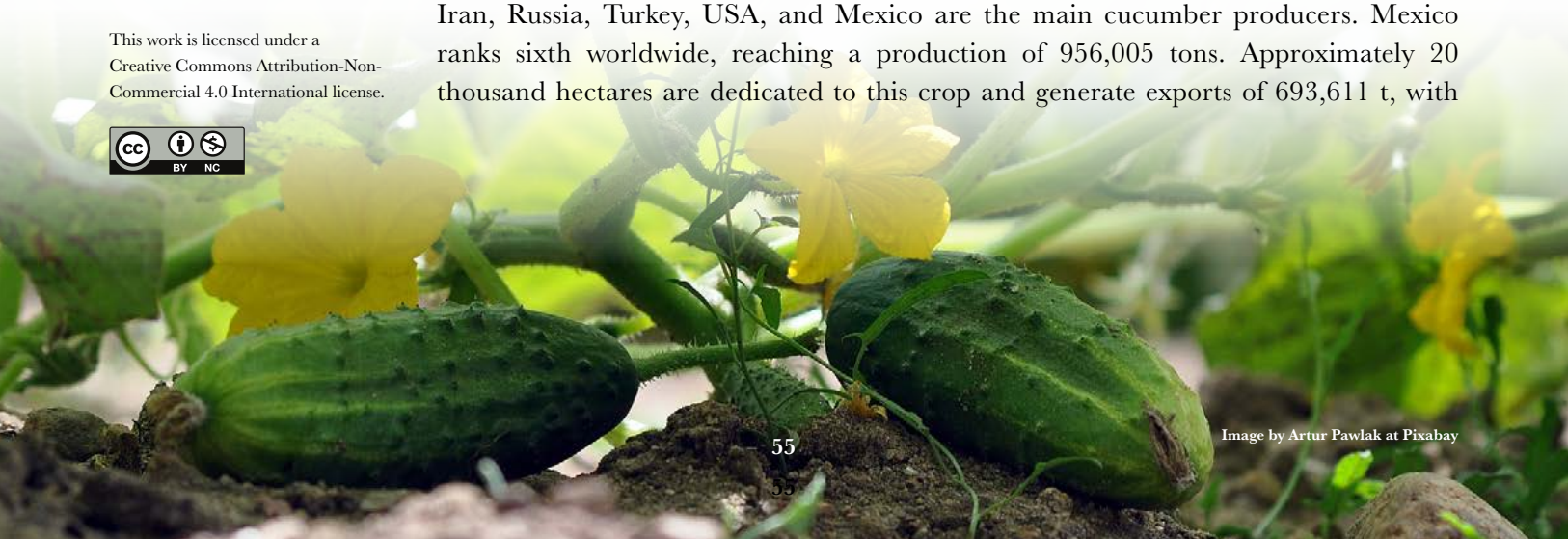
INTRODUCTION

Cucumber (*Cucumis sativus* L.) is one of the most popular vegetables cultivated all over the world (Singh *et al.*, 2017). There are several cultivars on the market that are mainly characterized by the different sizes, shapes, colors, flavors, and vegetative characteristics of their fruits. The most common types of cucumber are classified as: American, European, Middle Eastern, Dutch, and Oriental (López *et al.*, 2015). China, Iran, Russia, Turkey, USA, and Mexico are the main cucumber producers. Mexico ranks sixth worldwide, reaching a production of 956,005 tons. Approximately 20 thousand hectares are dedicated to this crop and generate exports of 693,611 t, with



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a value of around 483 million dollars (FAOSTAT, 2017). Kumar *et al.* (2017) mention that high levels of soil salinity can inhibit seedling germination and growth, as a result of the combined effect of high osmotic potential and specific ion toxicity. They also affect short- and long-term photosynthesis. In the short term, stomatal limitations can affect photosynthesis, leading to a decrease in carbon assimilation (Acosta *et al.*, 2017). Salt stress also causes excessive generation of reactive oxygen species (ROS), such as superoxide anions, hydrogen peroxide, and hydroxyl radicals (Zhang *et al.*, 2014). Biostimulants are increasingly integrated into production systems with the aim of modifying physiological processes in plants. This modification optimizes their productivity, offering a potentially novel approach to the regulation and modification of physiological processes in plants, in order to stimulate growth, mitigate stress-induced limitations, and increase yield (Yakhin *et al.*, 2017). The role and importance of humic substances (SH) in soils have been proven for a long time. Their multiple properties especially their capacity to "sequester" (adsorbent and chelate) organic and mineral compounds (pesticides and metals) allows them to play an essential role in the solubilization, bioavailability, degradability, transport, and exchange of these compounds in water and soil (Ouni *et al.*, 2014). This role results from the greater cation exchange capacity of the soil that contains SH and the increased availability of phosphorus that interferes with the precipitation of calcium phosphate (Du Jardin, 2015). Likewise, the beneficial properties of silicon are better documented in terms of its positive effects on tolerance to abiotic stress and pathogen resistance. It is mainly deposited at flow endpoints in cell walls and intercellular spaces. These silicon or phytolith depositions increase the mechanical resistance and erection of the leaves, which increases the interception of light and photosynthesis (Albrecht, 2019).

MATERIALS AND METHODS

This research project was carried out during the 2020 spring-summer in the Departamento de Horticultura at the Universidad Autónoma Agraria Antonio Narro, in the city of Saltillo, Coahuila, Mexico (1790 m.a.s.l., 25° 21" N 101° 01" W).

Crop management

Cucumber cultivars from two commercial American-type cucumber hybrids (Induran and SV2516) were planted in polystyrene trays with 72 square cavities. The cultivars were transplanted 20 days after planting, when the seedlings had two true leaves. The planting density was 2.7 plants/m^2. The two cucumber cultivars were staked to a single stem. Initial formative pruning and pruning were performed throughout the cycle, as axillary buds and tendrils grew.

The solution proposed by Steiner (1969) was chosen for the fertilization stage. The nutrient solution was supplied by fertigation and set at 1.8 dS m^{-1} and 6 pH at the beginning of the transplant. Two weeks after transplant, 25 mM sodium chloride was added to cause salt stress. No more sodium chloride was added after the E.C. reached 6 dS m^{-1} in the nutrient solution in the soil.

A foliar application of potassium silicate (Genhydro® Armor Si™) was applied every 7 days as a biostimulant throughout the cycle. Regarding humic substances, Cosmocel® H-85™ was drenched thrice (in the vegetative, flowering, and development stages).

This project used a completely randomized block design with factorial arrangement. Three factors were included in this arrangement: 1) the Induran (I) and SV2516 (SV) cucumber varieties (Var); 2) potassium silicate (Si) with 0, 10, and 20 $mL \cdot L^{-1}$ doses; and 3) humic substances (SH) with 0, 10, and 20 kg/ha doses (Table 2). There were three replicates and a total of 54 experimental units.

Evaluation of yield variables

The following variables were evaluated: number of fruits per plant (NFP), fruit weight (FW/PF), fruit diameter (FD/DF), fruit length (FL/LF) and yield (Y/Rend). NFP was evaluated once the crop entered the fruit set and the development stages. In the case of FW, the fruits of a sample —taken by experimental unit and which had the characteristics required by the market— were weighed after they had been harvested. This process was carried out with a Steren® MED-080 scale, with a maximum weight of 5 kg. Regarding the FD, the maximum circumference of the fruit was evaluated with a Steren® digital vernier. The LF was measured from one end to the other using a Truper® FH-5M flexometer. Finally, regarding Y, the weight of all the fruits harvested from each treatment was measured per square meter.

Mineral evaluation

The macroelements and microelements evaluated in fruit and leaf were N, P, K, Ca, Mg, and Fe. Three repetitions were taken per experimental unit. A digestion, distillation, and titration process was carried out to analyze N, following the procedure described by Mckean (1993). First, a 0.050 g dry and ground vegetable sample was weighed; then it was placed in Kjeldahl micro digestors and 5 mL of digester solution were added; subsequently, the mix was taken to 400 °C to disintegrate the organic nitrogen. The sample was considered ready when it showed a transparent hue. The samples were subjected to 50% NaOH and the resulting distillate was poured into 30 mL of 2% H_3BO_3 until it increased to 60 mL. The result was titrated with 0.025 N H_2SO_4 until it turned pink. N was calculated using the following formula.

$$\%N = \frac{\left(ml\ acid\ titrant - ml\ blank\right) \times N\ of\ the\ acid \times 1.4007}{Sample\ weight\ in\ grams} \times 100$$

P, K, Ca, Mg, and Fe were subjected to acid or wet digestions. At this stage, a 0.5 g sample of dry and ground matter was weighed. The previously weighed sample was placed in a 100-mL beaker and 30 mL of HNO_3 with a purity of 96% was added. The results were then placed on a watch glass rack for digestion. Once the sample was transparent, it was measured in a 50-mL Erlenmeyer flask with deionized water. P was analyzed by

colorimetry with the amino-naphthol-sulfonic acid (ANSA) method in a Thermo® Bio Matte 5 UV-VIS spectrophotometer (Terán, 2016).

$$mg \mid gr = \frac{Curve\ reading \times 10^{-3}}{Grams\ of\ the\ sample \times the\ dilution}$$

Meanwhile, K, Ca, Mg, and Fe were analyzed by atomic absorption spectrometry with a GBC Scientific Equipment® Xplorra. Ca and Mg digestions were diluted to of the initial concentration (Williams, 1972). To determine the mineral concentration, the absorbance of each mineral was taken and later supported with the following formula.

$$Element\ (\mu \mid g) = \frac{(c)(V)(d.f.)}{(W)}$$

Where: c=Concentration of the element that contains the sample used; V=Volume of the sample used; $d.f. = \frac{Volume\ of\ the\ diluted\ sample\ in\ mL}{Volume\ of\ the\ aliquot\ taken\ from\ the\ dilution\ in\ mL}$; and W=Weight of the sample in grams.

Statistical data analysis

The results obtained from the determination of each variable were subjected to an ANOVA and the means were compared using Duncan's test ($p \leq 0.05$), using the 2018 version of the Infostat statistical software.

RESULTS AND DISCUSSION

Agronomics

The Var*Si interaction had a statistically significant difference ($p \leq 0.05$) in FW, FD, FL, and Y. In the Var*SH interaction, a statistically significant difference ($p \leq 0.05$) was observed in the evaluated variables of NFP, FW, FD, FL, and Y. In relation to NFP, the SV*Si10 interaction recorded the best average: 30.9% more than I*Si10 (Figure 1A). However, SV*SH20 registered the maximum mean: 33% more than the SV*SH10 interaction (Figure 1B). Al-madhagi (2019) reported that 100-mg L^{-1} doses of humic acids increase the number and the average weight gain of cucumber fruits. Previous experiments indicated that SH treatments improved some fruit characteristics —for example, fruit number, fruit weight, and dry weight per fruit— of several crops, including cucumbers, tomatoes, onions, eggplants, and peppers (Shehata *et al.*, 2016). The SV*Si10 interaction was the maximum mean for the FW analysis, with a value of 388.3 g. This results was 16% higher than I*Si0, which had a minimum value of 326.1 g (Figure 1C). SH did not behave in the same way as Si. The maximum value (403.2 g) was obtained by the SV*SH0 interaction, while the minimum value was recorded with the I*SH0 interaction (Figure 1D). Therefore, the SH

did not infer on the FW variable. Regarding FD, the interaction with the highest mean was SV*Si0. This result was 5.5% higher than the I*Si20 interaction, which recorded the lowest value (Figure 1E). Meanwhile, in SH, the interaction with the highest value was SV*SH10. This result was 7.7% higher than the I*SH0 interaction that obtained the lowest value (Figure 1F). Therefore, it can be inferred that SH promoted FD in cucumber. Ekinci *et al*., (2015) analyzed several types of biostimulants and determined that the application of different SH doses to cucumber plants obtained similar results regarding the diameter and length of the fruit. Regarding FL, the SV*Si0 interaction obtained the highest value, which was 7.5% higher than the result of the I*Si10 interaction with the lowest value (Figure 1G). Consequently, it can be inferred that the Si doses do not directly favor the increase of FL. Meanwhile, the SV*SH10 interaction showed the highest SH value: 6.1% higher than the I*SH10 interaction that obtained the lowest value (Figure 1H). These data are consistent with those previously published by Ameta *et al.* (2017), who applied 10kg/ha of humic acids to the soil + 0.1% humic acids and micronutrients to the leaves, obtaining cucumber crop with very similar fruit lengths. Regarding Y, the SV*Si10 interaction had the highest value, obtaining 33.1% more than the I*Si10 interaction (Figure 1I); therefore, the assumption is that the Si supply is not related to the increase in Y —which is attributed to the genetic aspects of the varieties used. The SV*SH20 interaction had the highest SH value: 32% higher than I*SH10, which was the interaction with the lowest value (Figure 1J). For their part, Abd Elkareem *et al.* (2017) obtained higher yields (15-20%) with the direct application of HS into the soil and Benyamin Esho and Saeed (2017) reported that the application of different HS doses to several pumpkin (*Cucurbita pepo* L.) varieties resulted in a higher yield.

Minerals

The Var*Si interaction had a highly statistically significant difference ($p \leq 0.01$) in the P and Mg variables and a statistically significant difference ($p \leq 0.05$) for N, K, Ca, Fe, and Cu. Regarding Var*SH, a statistically significant difference ($p \leq 0.05$) was observed in N, P, Ca, Mg, and Fe. Regarding the N variable in fruit, the SV*Si10 interaction was the maximum mean with a value of 20.2 $g \cdot kg^{-1}$. It was 34.1% higher than SV*Si10, which had a value of 13.33 $g \cdot kg^{-1}$ (Figure 2A). Meanwhile, I*SH10 obtained the maximum N mean, with a value of 22.56 $g \cdot kg^{-1}$. This result was 34.4 % higher than SV*SH0, which had a value of 14.78 $g \cdot kg^{-1}$ (Figure 2B). Leaves had the best interaction with I*Si20 and I*SH20 (Figure 3A and 3B). Yasir *et al.* (2016) have suggested that using organic fertilizers based on humic substances in the soil plus the application of amino acids into the leaves favor the absorption of N. Humic acids make nutrients available for absorption; therefore, they facilitate growth, the accumulation of carbohydrates, and the increase in the photosynthetic rate and chlorophyll production.

When analyzing the P variable in fruit, the SV*Si20 interaction recorded the maximum mean with a value of 6.7 $g \cdot kg^{-1}$. This result was 48.4% higher than SV*Si10, which had a value of 3.48 $g \cdot kg^{-1}$ (Figure 2C). For its part, SV*SH10 recorded the maximum mean with a value of 5.9 $g \cdot kg^{-1}$. This result was 27.9% higher than I*SH0, which registered a value of 4.25 $g \cdot kg^{-1}$ (Figure 2D). SV*Si20 and SV*SH10 recorded best absorption of P in the leaves (Figure 3C and 3D). El-Nemr *et al.* (2012) have reported that, compared with their

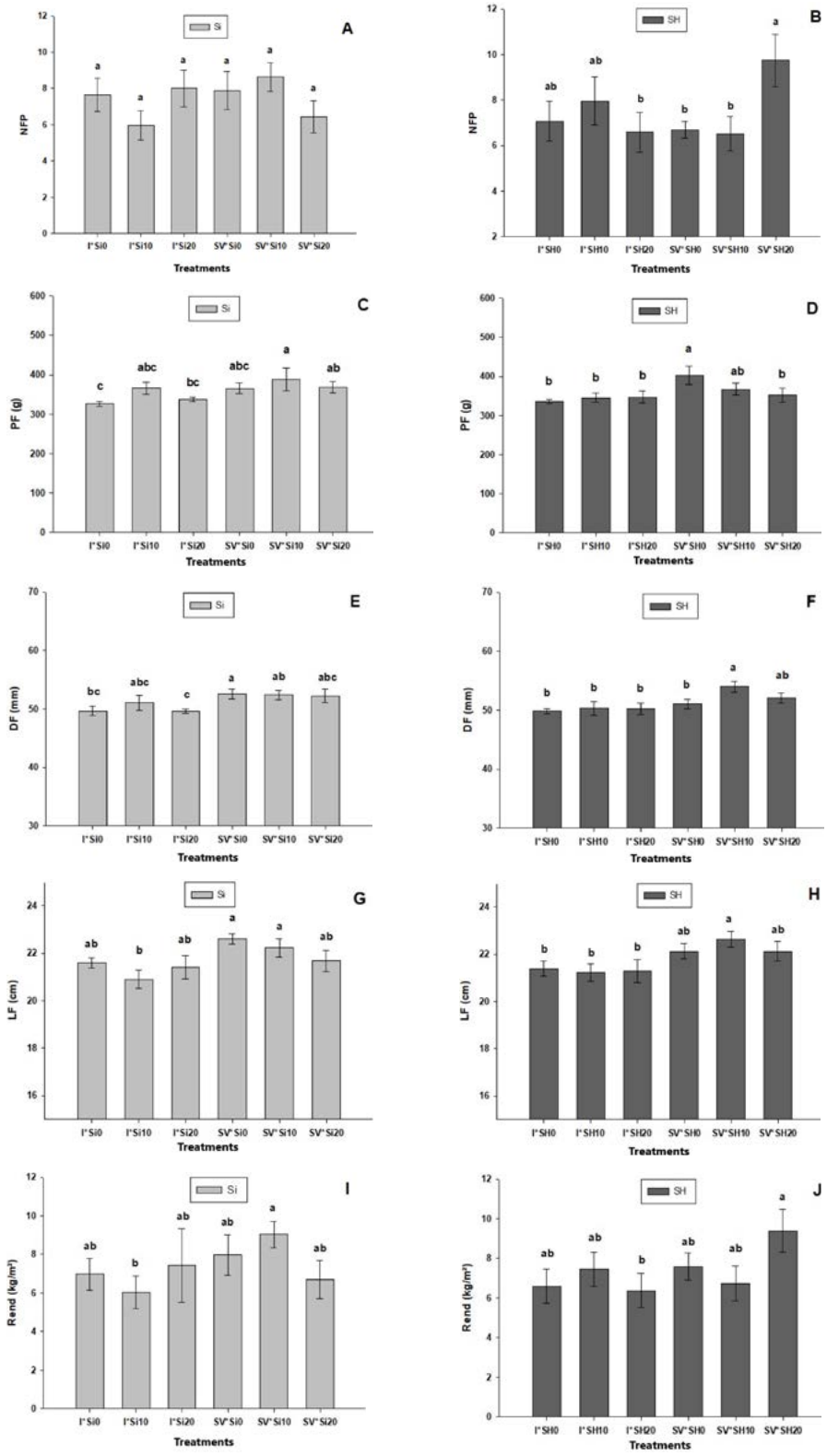

Figure 1. Tests of means. A and B (number of fruits); C and D (fruit weight); E and F (fruit diameter); G and H (fruit length); I and J (yield); Si=Silicon (0, 10, 20 mL L^{-1}); SH=humic substances (0, 10, 20 kg ha^{-1}); I=Induran; SV=SV2516. Different letters indicate significant differences between treatments, according to Duncan ($p \leq 0.05$), n=9 ± standard error.

controls, the application of humic acids increased the content of minerals such as N, P, and K in cucumber leaves, and found that the applications of these substances can stimulate the uptake of macro and microelements.

For the variable of K in fruit, the SV*Si20 interaction recorded the maximum mean, with a value of 28.3 $g \cdot kg^{-1}$. This result was 24.4% higher than SV*Si10, which registered a value of 21.39 $g \cdot kg^{-1}$ (Figure 2E). In contrast, the I*SH20 interaction obtained the maximum mean, with a value of 26.3 $g \cdot kg^{-1}$, which was 10.6% higher than the 23.5 $g \cdot kg^{-1}$ value of I*SH0 (Figure 2F). Regarding the K content in leaves, the SV*Si10 treatment had the highest value (Figure 3F). In many plant species, foliar application of silicic acid in the flag leaves of cereal crops increase N, P, and K absorption in grain (Soratto *et al.*, 2012). Another plausible explanation for SH activity is the hormone-like behaviour of their structures, which facilitates the nutrient translocation throughout the plant. Subsequently, the formation of complexes with metal ions increases their solubility and availability to plant roots (Khaled and Fawy, 2011). Regarding Ca in fruit, the I*Si20 interaction recorded the maximum mean with a value of 5.2 $g \cdot kg^{-1}$. This result was 53.8% higher than SV*Si20, which registered a value of 2.4 $g \cdot kg^{-1}$ (Figure 2G). The I*SH10 interaction obtained the maximum mean with a value of 5.7 g kg^{-1}. This result was 49.1% higher than I*SH0, which registered a value of 2.9 $g \cdot kg^{-1}$ (Figure 2H). In addition, Tripathi *et al.* (2014) recorded a significant decrease in the Ca an K concentrations in plants grown under a high NaCl treatment; however, their concentrations were raised to the required level by the addition of Si in both shoots and roots. In relation to Mg in fruit, the SV*Si10 interaction had the maximum mean with a value of 3.3 $g \cdot kg^{-1}$. This result was 36.3% higher than I*Si20, which registered a value of 2.1 $g \cdot kg^{-1}$ (Figure 2I). However, SV*SH0 had the maximum mean with a value of 3.5 $g \cdot kg^{-1}$. This result was 34.2% higher than I*SH20, which registered a value of 2.3 $g \cdot kg^{-1}$ (Figure 2J). Under salt stress (150 mM NaCl), Si (2 mM Na_2SiO_3) increased the K, Ca, and Mg content and decreased the Na and Cl content in tomato roots, stems, and leaves. This phenomenon was not mediated by a reduced translocation from root to stem or stem to leaf, but rather by a salt dilution effect triggered by improved growth —*i.e.*, higher shoot biomass accumulated under salt stress and Si application (Li *et al.*, 2015).

Regarding Fe in fruit, the SV*Si20 interaction had the maximum average with a value of 117.7 ppm. This result was 43.4% higher than SV*Si20, which registered a value of 66.6 ppm (Figure 4A). However, the I*SH0 interaction recorded the maximum average with a value of 116.2 ppm. This result was 56.1% higher than I*SH20, which registered a value of 65.3 ppm (Figure 4B). Studies prove the ability of Si to modulate the Fe absorption activity of cucumber at an early stage of stress, as result of the Fe deficiency, through the regulation of the gene expression levels of the proteins involved in this process (Pavlovic *et al.*, 2013). It has also been shown that Si application can facilitate Fe mobility and translocation from the xylem to the shoot, along with the accumulation of Fe-chelating compounds, such as citrate in xylem sap and leaf tissues (Bityutskii *et al.*, 2014). Regarding the means of Cu in fruit, the I*Si0 interaction had the maximum mean with a value of 11.9 ppm. This result was 42.8% higher than SV*Si20, which registered a value of 6.8 ppm (Figure 4C). However, the maximum mean value for I*SH10 was 11.3 ppm. This result was 30.9%

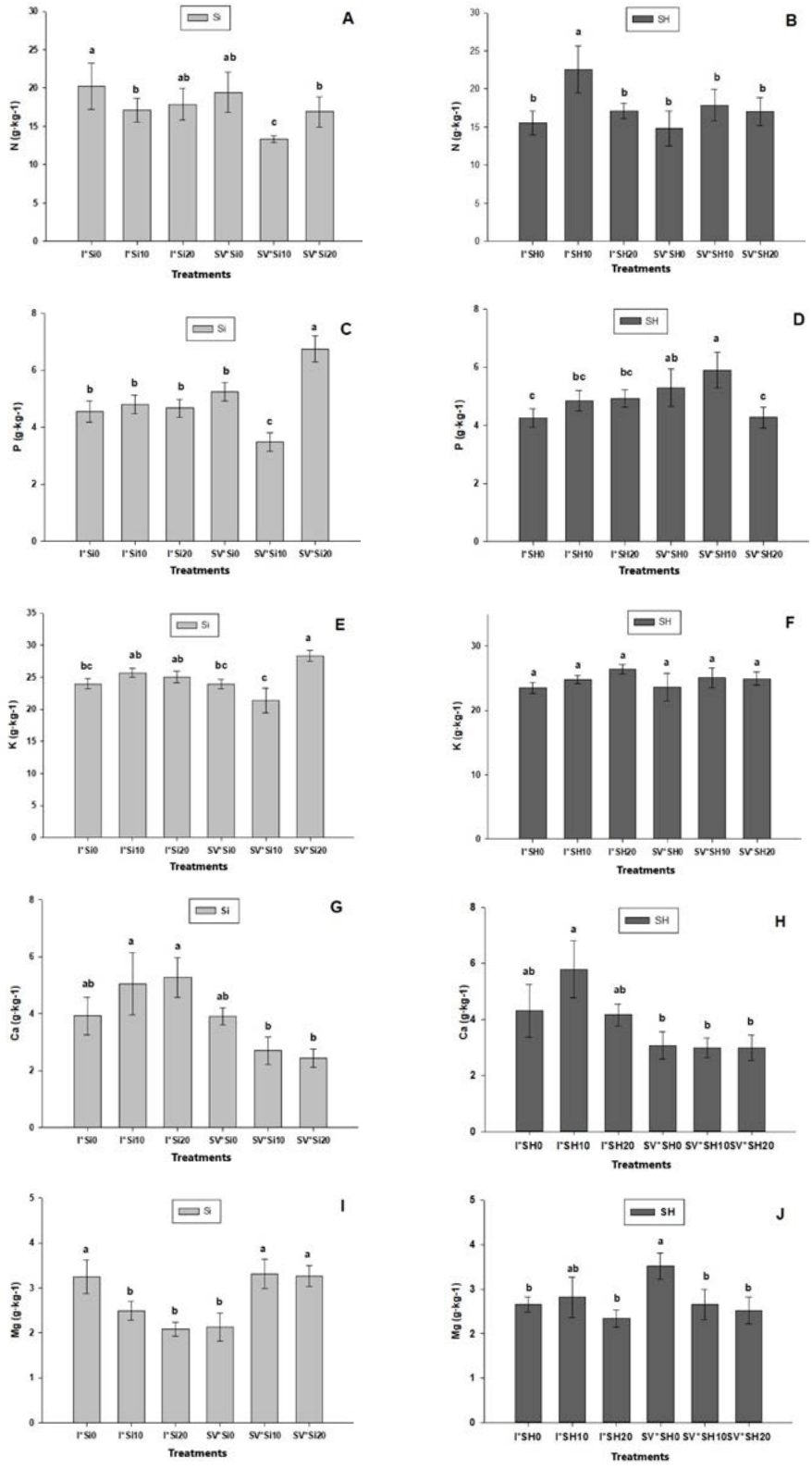

Figure 2. Tests of means on macrominerals in fruit. A and B (Nitrogen); C and D (Potassium); E and F (Phosphorus); G and H (Calcium); I and J (Magnesium); Si=Silicon (0, 10, 20 mL L^{-1}); SH=humic substances (0, 10, 20 kg ha^{-1}); I=Induran; SV=SV2516. Different letters indicate significant differences between treatments, according to Duncan ($p \leq 0.05$), n=9 ± standard error.

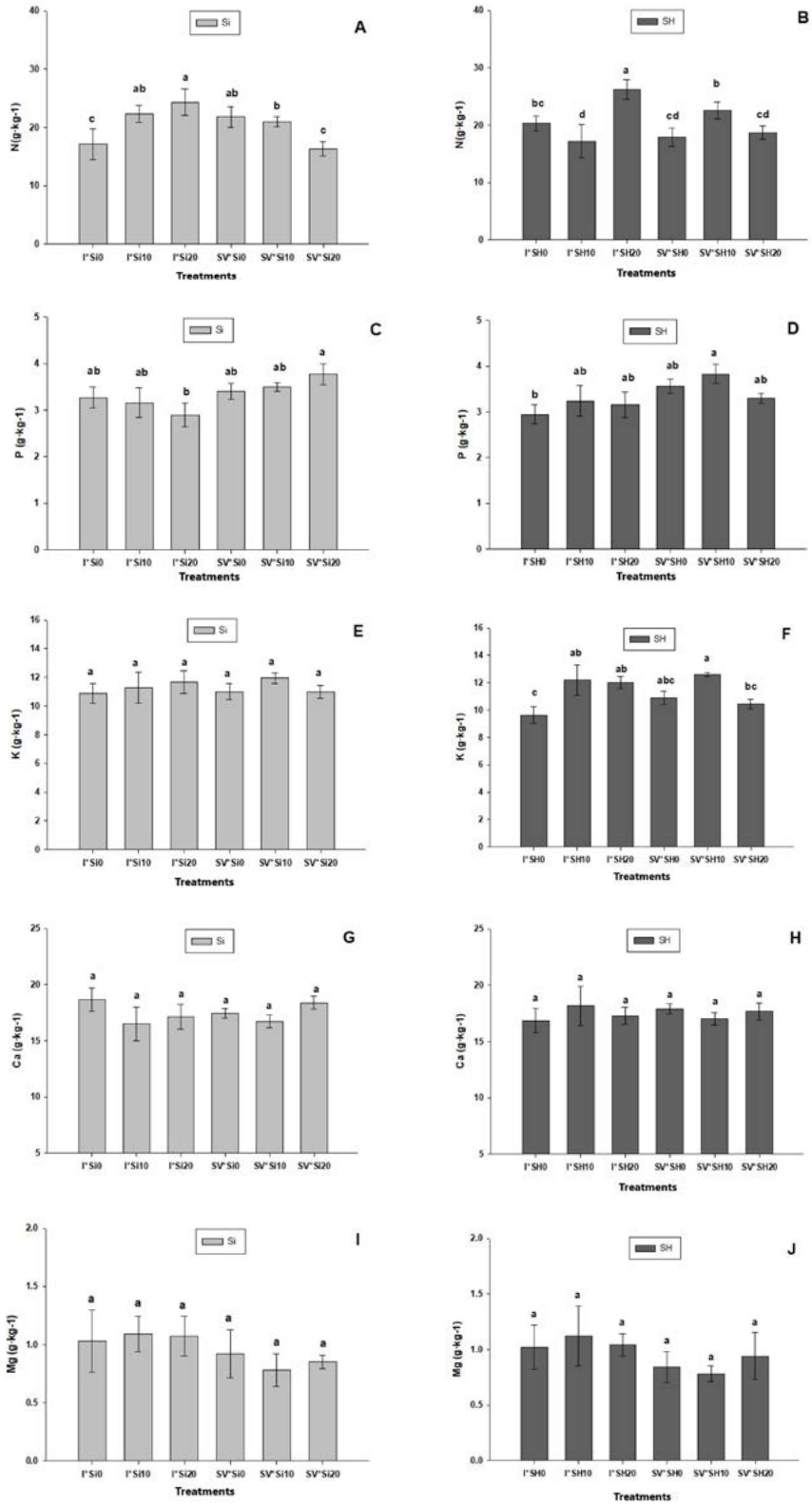

Figure 3. Tests of means on macrominerals in leaf. A and B (Nitrogen); C and D (Potassium); E and F (Phosphorus); G and H (Calcium); I and J (Magnesium); Si=Silicon (0, 10, 20 mL L^{-1}); SH=humic substances (0, 10, 20 kg ha^{-1}); I=Induran; SV=SV2516. Different letters indicate significant differences between treatments, according to Duncan ($p \leq 0.05$), n=9 ± standard error.

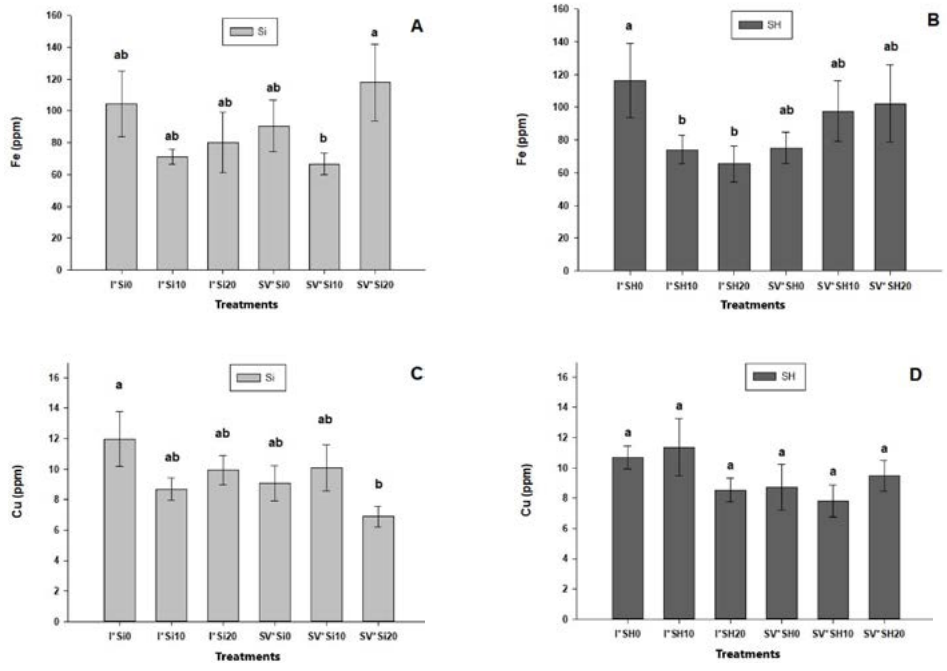

Figure 4. Tests of means on microminerals in fruit. A and B (Iron); C and D (Copper); Si=Silicon (0, 10, 20 mL L^{-1}); SH=humic substances (0, 10, 20 kg ha^{-1}); I=Induran; SV=SV2516. Different letters indicate significant differences between treatments, according to Duncan ($p \leq 0.05$), n=9 ± standard error.

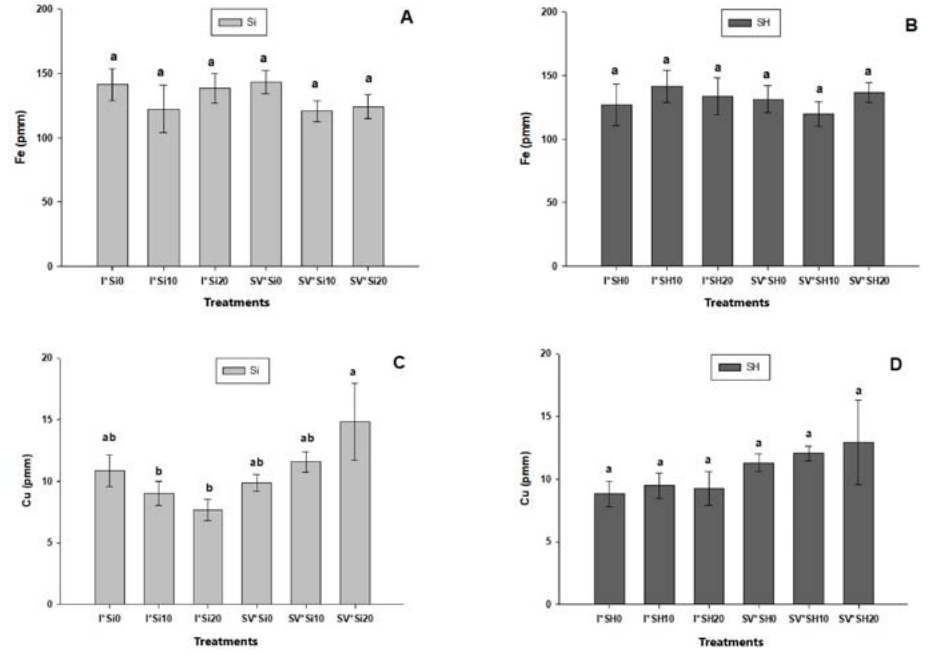

Figure 5. Tests of means on microminerals in leaf. A and B (Iron); C and D (Copper); Si=Silicon (0, 10, 20 mL L^{-1}); SH=humic substances (0, 10, 20 kg ha^{-1}); I=Induran; SV=SV2516. Different letters indicate significant differences between treatments, according to Duncan ($p \leq 0.05$), n=9 ± standard error.

higher than SV*SH10, which registered a value of 7.8 ppm (Figure D). The SV*Si20 interaction recorded the highest Cu content in leaves (Figure 5C). Cu absorption rates in alkaline soils are low. This is a consequence of the production of insoluble Cu complexes through the sequestration of oxides and hydroxides. An alternative to the low Cu activity in the soil is the use of SH to release carboxylic acid and favor its absorption (Turhan and Kuşçu, 2020). Protein hydrolysates (polypeptides, oligopeptides, and amino acids) and the components of SH have an impact on plant nutrition through the formation of complexes and chelates between peptides/amino acids and soil micronutrients (*i.e.*, Cu, Fe, Mn, and Zn). Consequently, they contribute to the availability of nutrients and its acquisition by the root system (De Pascale *et al.*, 2017).

CONCLUSIONS

The use of silicon and humic substances in different doses —along with the interaction of commercially available cucumber cultivars with improved resistance to different types of abiotic stress— can increase the productivity, quality, and absorption of nutrients and improve agronomic traits such as NFP, FW, FD, FL, and Y. Regarding mineral assimilation, it favors N, P, K, Ca, Fe, and Cu content and the N, P, K and, Cu content in the fruit and the leaves, respectively. Further research that involves various sources of silicon and humic substances, as well as their application techniques, their quantity, and their frequencies in cucumber cultivars is required to increase the mitigation of the stress caused by abiotic factors in regions of economic interest.

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Evaluation of fermentative activity of lactic cultures for dehydrated yogurt with the use of different additives

Colorado-Campos, Julio A.1; Bucio-Galindo, Adolfo1*; García-Alamilla, Pedro2*; Ramos-Juárez, Jesús A.1

[1] Colegio de Postgraduados Periférico Carlos A. Molina s/n, Km. 3.5 carretera Cárdenas-Huimanguillo. H. Cárdenas, Tabasco, México. CP 86500 Cárdenas, Tabasco. México
[2] División Académica de Ciencias Agropecuarias. Universidad Juárez Autónoma de Tabasco. Carretera Villahermosa- Teapa, kilómetro 25 Centro, Tabasco C. P. 86800.
* Correspondence: adbucio@colpos.mx; pedro.garciaa@ujat.mx

ABSTRACT

Objective: To evaluate the fermentative activity of dehydrated lactic cultures with the use of different additives and vacuum desiccation, using yogurt as model system.

Design/methodology/approach: The yogurt was elaborated with commercial lactic cultures (YF-L705 Yo-Flex CHR HANSEN) and whole milk incubated at 42 °C for 4 h. Yogurt was centrifuged at 6,000 rpm/15 min/4 °C. The supernatant was eliminated and with the precipitate, 6 treatments were established by addition of additives: T1, Without additive, T2, Glycerol, T3, Calcium carbonate, T4, Yeast extract, T5, Glycerol, and T6, Glycerol, Calcium carbonate and Yeast extract; non-dehydrated and freeze-dried yogurt was used as control: T7 and T8, respectively. The precipitate of the treatments with additives was dehydrated in a silica gel in a desiccator and under vacuum conditions. The weight loss was recorded at 0, 24, 48, 72 and 96 h. The precipitate with dehydrated additives was used as milk inoculates for yogurt elaboration. The change of pH was recorded at 0, 1, 2, 3, 4, 5, 6, 7, 8 and 24 h. With the pH and the fermentation time, a model was established to present the change curve in fermentation pH and the Fourier transform infrared spectroscopy (FTIR).

Results: The drying time to constant weight was 3 days. The fermentation pH change curve was a Boltzman sigmoidal function and analysis of variance was conducted with its parameters to assess the different fermentation speeds of the different treatments. The dehydrated cultures with Yeast Extract and Calcium Carbonate are associated with a higher fermentation activity of the milk ($p \leq 0.05$). The yogurts manufactured with fresh cultures take 4 to 5 h to ferment and the dehydrated ones take more than 10 h. The infrared spectra showed that the quality of the yogurts produced with fresh or dry cultures are similar, which agrees with other studies.

Limitations on study/implications: The dehydrated inoculated with the additives can be used to make yogurt with similar quality as to when inoculate with fresh culture is used, with the disadvantage of the fermentation time being longer. It is possible that this methodology can be used to dehydrate other inoculates based on lactic bacteria, but their effectiveness would have to be assessed experimentally.

Findings/conclusions: This study shows an alternative method to dehydrate lactic bacteria in the laboratory with equipment of relatively easy access for any laboratory.

Keywords: yogurt, drying, additives, lactic bacteria.

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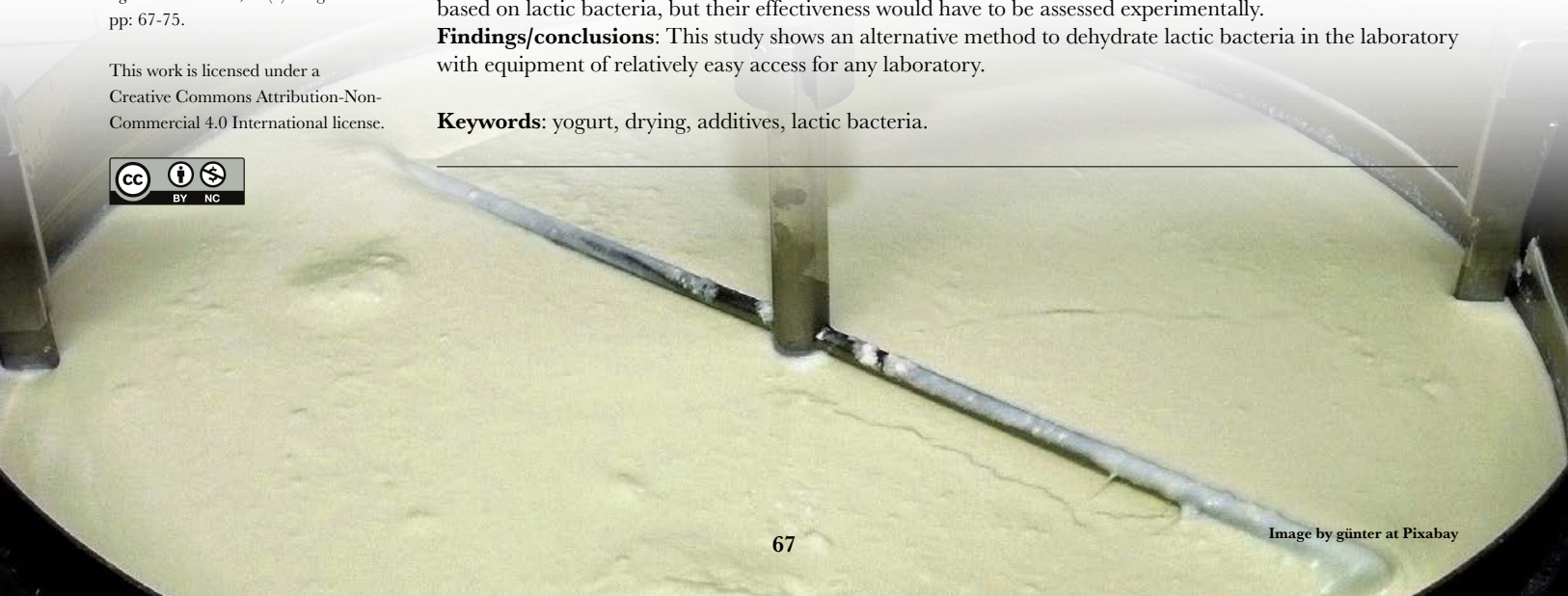

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INTRODUCTION

Acid-lactic bacteria are Gram-positive, non-sporulated, negative catalases, microaerophilic which produce lactic acid based on the fermentation of glucose or other carbohydrates (Mora-Villalobos *et al.*, 2020). They are broadly distributed in nature. One of the main uses is to elaborate fermented lactic products, such as cheese, jocoque, yogurt (Walstra *et al.*, 2005; Ramírez *et al.*, 2011), or products used in animal feeding such as vitafert (Lazo-Pérez *et al.*, 2017, Citalan Cifuentes *et al.*, 2016; Castillo Mercado *et al.*, 2020), which is an inoculate of lactobacillus from yogurt that is useful for the production of lactic acid and the reduction of pH in fermented foods.

Yogurt inhibits the pathogens transmitted by foods due to the action of organic acids (primarily lactic acid) and other compounds such as hydrogen peroxide, acetaldehyde, and bacteriocins that can act as bactericide/bacteriostatic agents (Papadopoulou *et al.*, 2021).

Historically, fermented lactic foods are prepared by inoculating fresh milk with a portion of a previously manufactured product that contains lactic bacteria and other fermented microorganisms. The inoculates used in the industrial production of many fermented products are dehydrated lactic cultures, which have functional advantages compared to fresh cultures, since in addition to being concentrated, they present low volume which makes their frozen storage and commercialization easy, and in addition, they can have several years of useful life.

To process 200 L of milk to yogurt, the inoculate must consist in 5 to 10 L of recently elaborated yogurt, but if a freeze-dried inoculate from some prestigious commercial brand is used, with 20 g of concentrated freeze-dried culture, it is enough to process the 200 L of milk. However, dehydrated inoculates are expensive for their application in animal production, particularly if they are freeze-dried; thus, for example, the cost of 20 g of the freeze-dried inoculate to process 200 L of milk for yogurt is approximately US$10.00.

To obtain inoculates, there are several drying processes (freeze drying and convection drying). Convection drying is a less costly method than freeze drying, and it is used to preserve lactic bacteria (Kets, 1997; Linders, 1996) with the advantage that it can be done at a small scale and with equipment that is relatively easy to get or fabricate. However, convection drying is a somewhat unknown method.

Water is the main constituent of bacteria, and their viability and fermentative capacity is decreased with dehydration; however, their viability can be improved with the addition of agents that protect from osmotic stress (Broeckx *et al.*, 2016). Those protective agents are compatible solutes such as betaine, carnitine, ectoines, sucrose and trehalose that make bacteria more resistant to dehydration (Kets, 1997; Prasad *et al.* 2003); these solutes can accumulate at high levels without interfering with cellular processes. Many compatible solutes have shown effectiveness in enzyme stabilization, under conditions of high concentrations of salt, high temperatures, freezing or thawing and with dehydration processes (Poolman *et al.*, 1998). The design of convection drying protocols can be evaluated using lactic yogurt bacteria as a model system. The fermentative activity of dehydrated inoculates in the substrate to ferment is an indirect, fast and practical measure to understand the functionality of the bacteria (Linders, 1996). FTIR spectroscopy can be used to evaluate the spectral signatures of many products, such as dairy products (Rodríguez-Saona and

Allendorf, 2011), and also to perform exploratory studies of changes in quality during the useful life of the products. In the case of yogurt, FTIR spectroscopy has been used to evaluate the changes during its shelf life; however, it has not been used in the assessment of the fermentative activity of dehydrated bacteria. Based on this, the use of different additives to preserve the fermentative activity of dehydrated lactic cultures was assessed in vacuum desiccation using yogurt as model system.

MATERIALS AND METHODS

The yogurt was formulated pasteurized cow milk of the brand Lala® (fat 3.3% g; protein 3.1% g; lactose 4.80%) inoculated with freeze-dried culture (0.1 g bacteria L milk) of the brand YF-L705 Yo-Flex® CHR HANSEN and incubated at a temperature of 44 to 45 °C in a FELISA FE-377 thermal bath for a period of 4 to 5 h. Then, it was stored in refrigeration for conservation before using it. The yogurt was centrifuged at 6000 rpm at 4 °C during 15 min; then the lactic sediment was collected, which contained lactic bacteria that were used for the experiments, and the supernatant was eliminated.

Treatments and experimental design

In a completely random design, six treatments (additives) with three repetitions were distributed. The treatments were: T1: without additive, T2: Glycerol, T3: Calcium carbonate, T4: Yeast extract, T5: Glycerol and Calcium carbonate, T6: Glycerol, Calcium carbonate and Yeast extract. The experimental unit consisted of aluminum trays of 5 cm diameter. Each tray was added with 2 g of the yogurt precipitate and 0.2 mL of the additive according to the treatment (Figure 1). The treatments were dried during 5 days

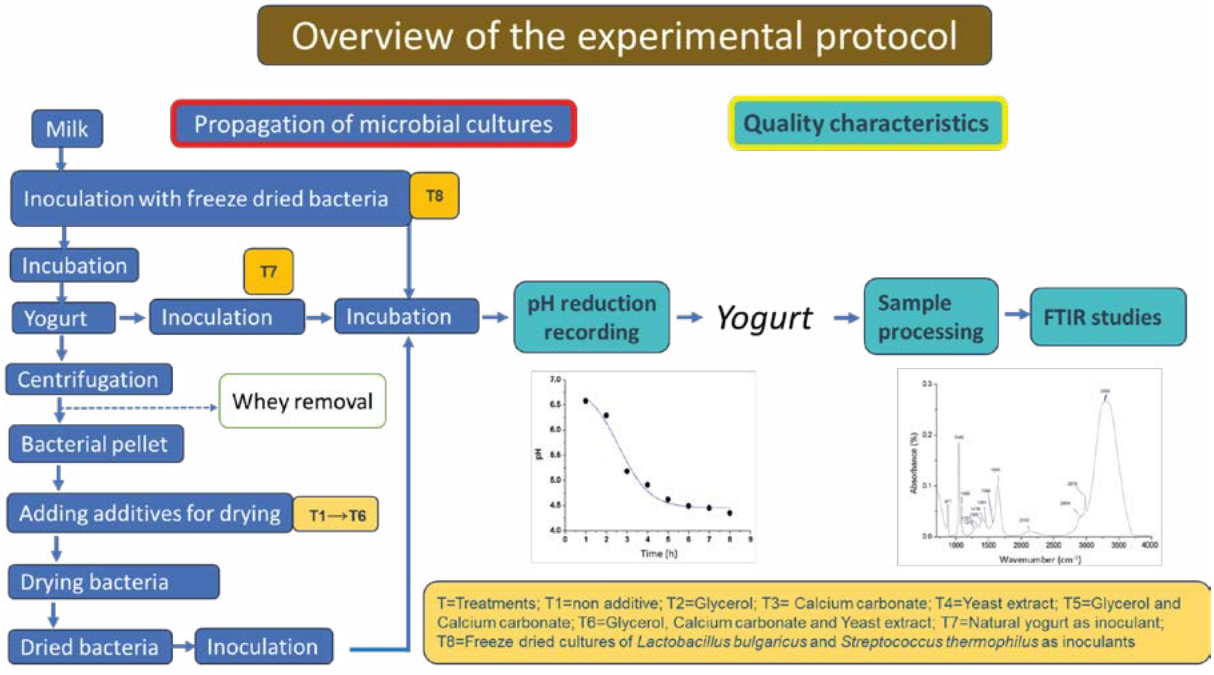

Figure 1. Flow diagram of the experiment and additives used in the experiment.

in a desiccator under vacuum conditions (Nalgene NALGE07022). The weight loss of the sample in the desiccator was measured on day 0, 1, 2, 3, 4 and 5. Fermentative activity tests of the treatments studied were conducted at 72 h of drying, since no changes were observed after that time in the weight of the samples. The dehydrated product (0.5 g) was used to inoculate 50 mL of whole milk (composition described earlier) and the pH was measured at 0, 1, 2, 3, 4, 5, 6, 7 and 8 h with a potentiometer (HANNA HI-2211, United States of America). In the fermentation tests, fresh yogurt inoculates (Treatment 7) and a freeze-dried culture (Treatment 8) were used as control groups. Treatment 7 was inoculated with 2 g of natural yogurt elaborated with the freeze-dried culture (without centrifuge and without dehydrating). Treatment 8 was inoculated with 0.005 g of a freeze-dried culture.

The pH values found at different times were graphed to model and monitor the fermentation in milk using the Boltzman equation with the Microcal Origin software version 6 (Navrátil *et al.*, 2004), adjusted in the following way:

$$pH = \frac{pH_{initial} - pH_{final}}{1 + e^{(x - x_0)/d_x}} + pH_{final}$$

The values of the constants ($pH_{initial}$, pH_{final}, x_0, d_x) were estimated through the non-linear adjustment curves of the Microcal Origin 6.0 software (Northampton, MA, USA). In the Boltzmann sigmoidal function, Y denotes the pH that changes in time t. The parameters $pH_{initial}$ and pH_{final} correspond to the positions of two asymptotes of the curve Y(t) (superior and inferior), x_0 is half the time that passes between $pH_{initial}$ and the pH_{final} (Figure 2). From this, it is inferred that $2x_0$ is the time at which yogurt reaches the finalization pH.

FTIR spectral signature

Following the procedures by Subramanian and Rodríguez-Saona (2009), 1 mL of yogurt was placed in a vial, centrifuged at 14,000 rpm for 3 min and the precipitate was discarded. Of the supernatant, 0.5 mL was taken and placed in a new vial, and after that

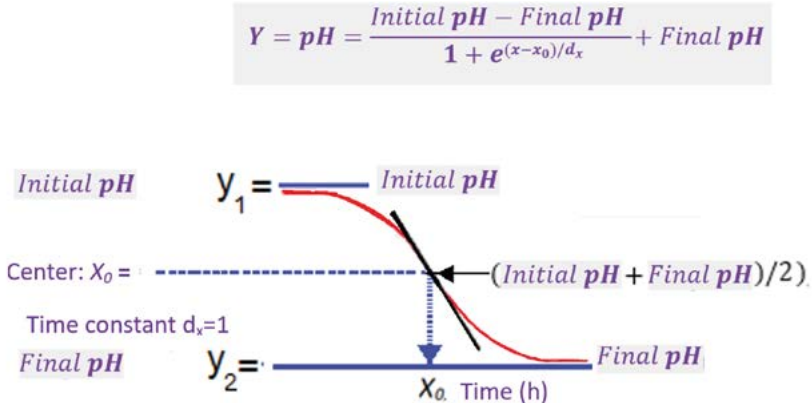

Figure 2. Boltzman function, modified from Microcal origin version 6.

0.5 mL of distilled water was added plus 0.5 mL of chloroform to separate the complex fat in the sample from the other portions of the sample; it was centrifuged again under the same conditions mentioned in previous steps, discarding the most viscous part that the chloroform contained with dissolved fats. Finally, 200 μL of the supernatant was collected and 200 μL of absolute ethanol from the last mixture was added to precipitate complex proteins of the supernatant; 100 μL was taken in a new vial to freeze it until its reading, using the trial sample in a FTIR analysis. The vibrational analysis was carried out by using Fourier transform infrared spectrophotometer (FTIR) (Perkin Elmer, Frontier, EUA), using diamond attenuated total reflectance (ATR) controlled with software for Windows©, in the wave number interval of 400 to 4000 cm^{-1} with a resolution of 1 cm^{-1} and 32 scan., A pre-treatment baseline correction and softening in the Spectrum software of the FTIR Perkin-Elmer spectrophotometer (Perkin-Elmer) was conducted with the spectra obtained. The data of the spectra were exported in ASCII format and analyzed using MagicPlot.

RESULTS AND DISCUSSION

Yogurt was produced with the typical fermentation kinetics with duration of 4 or 5 h incubating at 45 °C (Soukoulis *et al.*, 2007). With the yogurt centrifuge and the removal of the aqueous part, the precipitate was left with approximately 25% of the original weight of the yogurt. Precipitate drying on the trays was done for three days, and after the third day, the samples did not lose weight anymore (Figure 3).

The portions of milk inoculated with the dehydrated yogurt inoculates presented pH reduction profiles where the three phases present in the entire fermentation process are distinguished: 1) latency phase, lag or start of pH decrease, 2) logarithmic phase (quick decrease of the pH), and 3) final decrease of pH until reaching stable values. Two examples of fermentation kinetics are presented in Figure 4. The finalization time of the fermentation differed significantly between freeze-dried and dehydrated inoculates. These data could be modelled in a predictive way with the Boltzman function (Figure 4). The coefficients of determination reached are very high, indicating a good adjustment of the data to the model selected within the whole interval of values of the variables. This model has already been

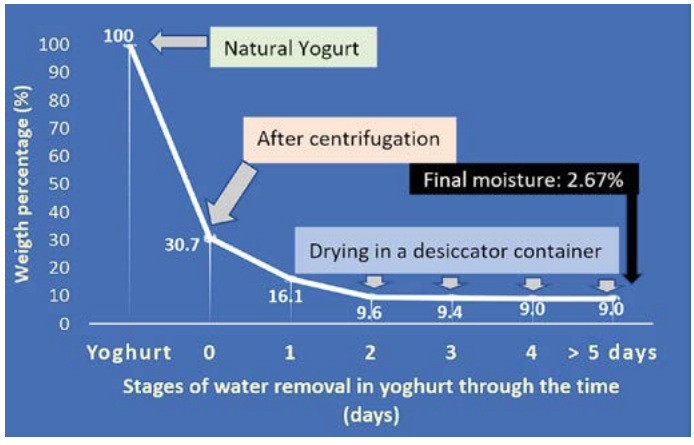

Figure 3. Stages of water removal from the yogurt.

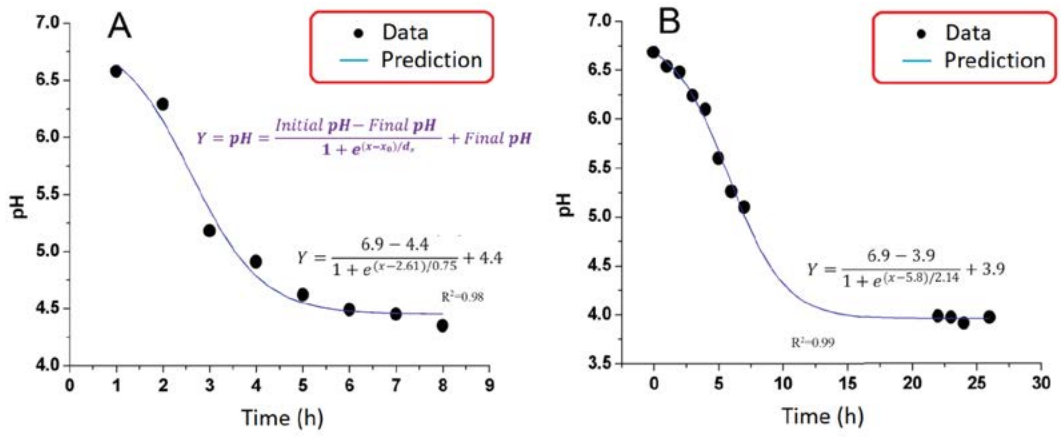

Figure 4. pH reduction curve of the yogurt inoculated with a) freeze-dried yogurt and b) dehydrated yogurt using the yeast extract as additive.

used previously when assessing the kinetics of lactose disappearance after fermentation of the Kombucha dairy product (Kanurić *et al.*, 2018), although they have not been used to describe the pH decrease. The yogurts manufactured with fresh cultures take 4 h to ferment, and the dehydrated exceed 10 h (Figure 4).

The parameters of pH reduction kinetics of the milk, fermented with different treatments of dehydrated inoculates and fresh yogurt, based on modelling with the Boltzman function equation, are presented in Table 1. The milks inoculated with fresh and freeze-dried yogurt reduced the pH significantly in less time (X_0) than the other treatments (Table 1); the samples from treatment 3 and 4 followed, which contain calcium carbonate and yeast extract, respectively. X_0 is the mean reduction time of the pH.

The $pH_{initial}$ of the natural yogurt was significantly higher than the other treatments. It can be assumed that this value was because of interpolation of the parameters of the Boltzmann equation where a slope with great pH reduction speed of this treatment is included; and which compared to the other treatments probably does not have alkalinizing agents that can affect the inferior and superior limits of the kinetics. In the case of the pH_{final}, the highest values were for the treatment inoculated with freeze-dried yogurt, probably due to some effect of the inoculate and because of the way in which it is prepared

Table 1. Parameters of the pH reduction kinetics based on the Boltzman function.

Experimental treatments	Initial pH	Final pH	X_0	d_x
1) SN, without additives	6.6a	3.9a	6.3cd	1.9b
2) Glycerol	6.7a	3.9ab	6.8d	2.1b
3) Calcium carbonate	6.8a	4.0bc	5.9c	2.3b
4) Yeast extract	6.9a	3.9ab	5.8c	2.2b
5) Glycerol and Calcium carbonate	6.8a	4.0c	6.2cd	2.1b
6) Glycerol, Calcium carbonate and Yeast extract	6.7a	4.0c	6.8d	2.0b
7) Natural yoghurt (Unflavored and Unsweetened)	8.1b	4.0bc	1.3a	1.8b
8) Starter cultures for yoghrt (freeze dried).	7.0a	4.4d	2.6b	0.8a

for its functionality. It is unknown whether it has slightly neutralizing encapsulated agents. The dehydrated cultures that contain yeast extract and calcium carbonate are associated with a fast fermentation activity of the milk ($p \leq 0.05$).

The FTIR spectral data of the yogurt extract samples elaborated with fresh yogurt inoculates and from one of the dehydrated yogurt samples (Treatment 4) are shown in Figure 5. The characteristic peaks of the yogurt sample manufactured with fresh cultures are similar to those of yogurts with dehydrated inoculates, and no difference is reflected in qualitative quality between the yogurt samples from the two cultures, since the spectra were similar.

Several absorption peaks of the infrared electromagnetic radiation were identified in both spectral graphs, which are characteristic in many fermented dairy products (Rodríguez-Saona *et al.*, 2006, 2017). Two peaks are characteristic of lactose at 1046 and 1086 (Rodríguez-Saona *et al.*, 2017), a spectral absorption peak at 1640 cm^{-1} has been reported with an amide I belonging to the union of carboxyl and amino groups of amino acids, which in the case of this study could be derived from casein, which similar to many other proteins has been reported to have spectral peaks around 1650, 1550 and 1250 cm^{-1} due to amide I, amide II and III (Hewavitharana and van Brakel, 1997; Papadopoulou *et al.*, 2021; Derrick *et al.*, 2000).

Authors such as Derrick *et al.* (2000) observed that the peaks of highest infrared absorption of casein samples are from stretching of C-H groups at 3100-2800 C-H, stretching of C=O groups at 1660-1600, flexion of C-N-H groups at 1565-1500, and flexion of C-H groups at 1480-1300. They are also very common in the samples extracted from cheeses (Subramanian and Rodriguez-Saona, 2009) and from yogurt in this study.

Mixtures of α-casein and β-lactoglobulin have also been reported at 1650 cm^{-1} (Susi, 1972) that are united through SH groups, with the resulting proteins from the milk treatment for yogurt. Other authors also say that it is a wave length where there

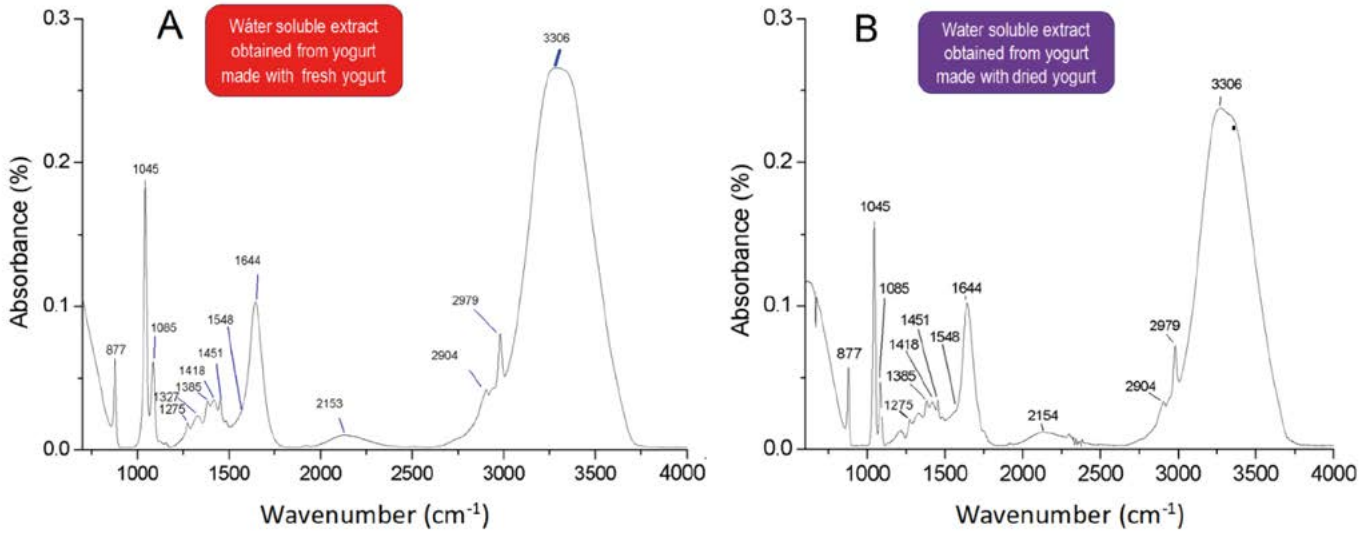

Figure 5. FTIR spectra of yogurt samples manufactured with fresh (A) and dehydrated (B) yogurt inoculates.

is water, and carboxyl acids from organic acids (Fagan, 2014) and water (O-H), which could be lactic acid. More recently, Karadeniz *et al.* (2021) analyzed different samples of exopolysaccharides, commonly produced by *Streptococcus thermophilus* by FTIR spectroscopy. The example showed a wide band in around 3287 cm^{-1} attributed to the stretching vibration of hydroxyl groups from carbohydrates and C-H stretching around 2925 cm^{-1}. The band of 1638 cm^{-1} is associated with the traction vibration of C=O and corresponds to the characteristic absorption of the polysaccharides. All the spectra showed typical bands of polysaccharides. All the spectra showed typical bands of polysaccharides near 1026,9 cm^{-1} within the region of digital signature (1200-950 cm^{-1}). This is novel about convection dehydrated bacteria, their fermentative activity and the assessment of yogurt fermentation products by FTIR. It is possible for this methodology to be used to dehydrate other inoculates based on lactic bacteria, but their effectiveness would have to be evaluated experimentally. The equipment used is of relatively easy access for any laboratory.

CONCLUSIONS

Dehydration with the additives used and vacuum desiccators allows obtaining dehydrated inoculates that can be used to make yogurt with spectral characteristics similar to when inoculation is done with fresh culture, with the disadvantage of a longer fermentation time.

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Bibliometric analysis of scientific research about agroecological strategies

Santillán-Fernández, Alberto[1*]; Vargas-Díaz, Arely A.[1]; Flota-Bañuelos, Carolina[1]; Fraire-Cordero, Silvia[1]; Rosales-Martínez, Verónica[1]; Bautista-Ortega, Jaime[2]

[1] IxM CONAHCyT y Colegio de Postgraduados Campus Campeche. Sihochac, Champotón, Campeche, México. C. P. 24450.
[2] Colegio de Postgraduados Campus Campeche. Sihochac, Champotón, Campeche, México. C.P. 24450.
* Correspondence: santillan.alberto@colpos.mx

ABSTRACT

Objective: To analyze the spatio-temporal evolution of scientific production on the topic of agroecological strategies in the world, through text mining (bibliometrics) and coauthorship networks to determine gaps in research.

Design/methodology/approach: To compile the texts, the database of open-access articles from Web of Science was reviewed. The texts were collected in September 2021, and texts available until August 2021 were considered. The keyword used in the search was *strategies agroecological* identifying it in the titles and keywords of the publications.

Results: In the databases, 463 scientific texts were collected from bibliographic references of Web of Science within the period of 1987 to 2021. Of these, 90.93% (421) were concentrated in six countries: USA (34.77%, 161), Netherlands (19.22%, 89), England (15.55%, 72), Switzerland (12.53%, 58), France (4.54%, 21) and Germany (4.32%, 20).

Limitations on study/implications: Most of the studies have been centered on techniques for agricultural sustainability, so topics associated with the creation of early climate information systems and disaster prevention and mitigation projects are presented as an opportunity for the development of more research.

Findings/conclusions: The study shows that in developing countries such as those in Latin America, where Mexico, Argentina, Chile and Brazil are located, there is scarce research development in topics of agroecological strategies, leaving a wide margin to generate knowledge in the region.

Keywords: Author network, bibliometrics, scientific article, spatio-temporal, analysis, text mining.

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INTRODUCTION

Climate change and food security are currently two priority problems for society as a whole. The variations in temperature and rainfall have made food production increasingly unstable as a response to prolonged droughts or unpredictable flooding. In the presence of this panorama, agricultural production modes face the great challenge of producing without deteriorating the environment, in a sustainable manner. Altieri and Nicholls (2007) consider that the migration from current production systems where the use of chemicals that exhaust natural resources is prioritized, to production systems where natural resources are regenerated, can only be navigated through agroecological techniques where the use of chemicals is minimized.

Among the agroecological strategies that have traditionally been used to reduce agricultural vulnerability in the presence of climate variability, there are the following: crop diversification, maintenance of local genetic diversity, animal integration, addition of organic matter to the soil, water harvest, among others. Today, these innovations are the most stable link for communities vulnerable to climate variations to be able to apply resilient

agricultural production systems, while public policies to reduce the risks of traditional agricultural systems and to promote the creation of early climate information systems are designed, as well as disaster prevention and mitigation projects (Altieri and Nicholls, 2017).

To understand the advance in research of a sector in particular and to propose new strategies for its consolidation, bibliometric studies are a reliable tool that allows studying and analyzing the evolution of scientific activity through its publications (Cañas-Guerrero *et al.*, 2013). The publication of a scientific study is the most effective way of transmitting knowledge acquired from research, and its visibility is important for the researchers themselves, for the institutions where they work, and for the organizations that finance the research (Sanz-Valero and Wanden-Berghe, 2017). The growth of scientific production in recent decades and its indexing in automatized bibliographic databases have potentiated the use of bibliometrics and the generation of indicators to measure the results of scientific and technological activities (Allen *et al.*, 2009).

Bibliometric studies based on published scientific articles allow generating indicators and mathematical models to characterize the development and evolution of the frequency and quality of the publications (Malesios and Arabatzis, 2012). Under this context, the objective of this study was to analyze the spatio-temporal evolution of scientific production on the topic of agroecological strategies in the world, through text mining (bibliometrics) and coauthorship networks to determine the relevance of the topic and to spatially locate the countries that have developed more research on agroecological strategies.

MATERIALS AND METHODS

Origin of the information

To gather the texts, the open-access database of journal articles from Web of Science was used (CONACYT, 2021). The texts were collected in September 2021, and the texts available until August 2021 were considered. The keyword used in the search was *strategies agroecological* identifying it in the titles and keywords of the publications. The keyword was only used in English because it is considered that it is more likely for texts in English to be cited, which allows capturing most of the relevant publications (Leipold, 2014).

Bibliometric indicators

The variables analyzed from each of the texts published were: *country of journal editing*, *language of publication*, and *name of the journal*, which served to determine the profile of the journals that published studies related to the topic of *agroecological strategies*; and the spatial representation of the countries with the highest number of publications. The variables *first author* and *collaborators* served to understand the network of stakeholders involved in the research; the variable *year* to locate the information on a timeline. The *title*, *abstract* and *keywords* was used to categorize the topic approached by the publication according to the classification of Web of Science; and, finally, the impact of the publications was determined with *number of citations*.

The capture of variables for the bibliometric analysis was carried out in a worksheet. The original language of each of the texts was respected. During the capture of all the information, some records were standardized because the information available in the

texts was sometimes incomplete or had variants (Aguado-López *et al.*, 2009). In addition, special characters were eliminated or changed, such as: ñ (for n), accents, superindices, subindices, ®, ©, among others, to ease the analysis.

Analysis with text mining

With the help of the RcmdrPlugin.temis complement of the statistical R software (Bouchet-Valat and Bastin, 2013), the number of texts and bibliographic citations was obtained by: year, journal, research topic, and country where the study was published.

Network analysis

The interactions between the first authors and collaborators were analyzed with the Sci2tool software (Börner, 2011), with the aim of understanding the constancy in the researcher's work; that is, to evaluate if the author has published only in one year or else if they have published constantly throughout time, which gives an idea of their consolidation in the topic of *agroecological strategies*. The syntax used in the Sci2tool software was *Extract bipartite Network*, and the Gephi software was used for its visualization (Bastian *et al.*, 2009). Finally, the spatial representation of the number of articles per country where the research was published was made in the geographic package ARGIS® (ESRI, 2015).

RESULTS AND DISCUSSION

Spatio-temporal evolution

From 1987 to 2021 a total of 463 scientific texts were published in the databases for bibliographic references of the Web of Science (CONACYT, 2021), where the object of study was the agroecological strategies; this scientific production originated 29896 bibliographic citations (Figure 1). From the 463 texts, 414 (89.42%) were scientific articles, and 49 (10.58%) bibliographic reviews. The first recorded study dates from the year 1987,

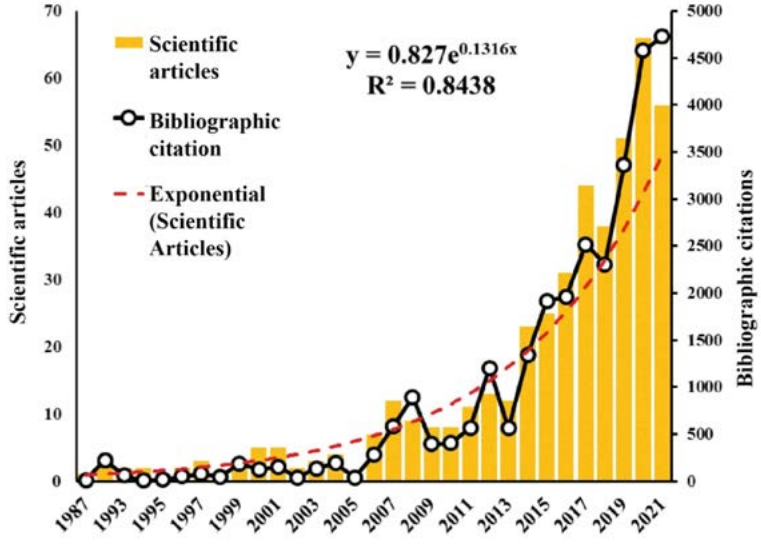

Figure 1. Temporal evolution of the scientific production and bibliographic citations in the topic of agroecological strategies in the world from 1987 to 2021.

although there has been a growing production for the topic of agroecological strategies since the year 2010.

The period of highest productivity was from 2010 to 2021 with 84.02% of the total (389 texts), which contribtued to an exponential trend in the increase of publications (R^2=0.8438). The most cited studies were the ones published in the period 2014-2021 which in total represented 23625 bibliographic citations (79.02% of the total). The exponential trend in the publications denotes, according to Altieri and Nicholls (2017), the relevance that the topic of agroecological strategies currently has in the scientific community, as an alternative to adapt to the present climate variations.

According to the country where scientific texts were published, the 463 works were originated in 27 countries. Of these, 90.93% (421) were concentrated in six countries: USA (34.77%, 161), Netherlands (19.22%, 89), England (15.55%, 72), Switzerland (12.53%, 58), France (4.54%, 21) and Germany (4.32%, 20) (Figure 2). Figure 2 also shows that most of the research developed around the topic of agroecological strategies has happened in USA and European countries with consolidated economies.

According to Gersbach and Schneider (2015), the economic development of a country is directly related to the quality of the research it performs; countries with consolidated economies invest more in their research centers, which allows for greater technological development, compared to underdeveloped economies such as the Latin American, where investment in research is lower. Therefore, agricultural competitiveness in a country is directly related to the quality of the research conducted in the sector.

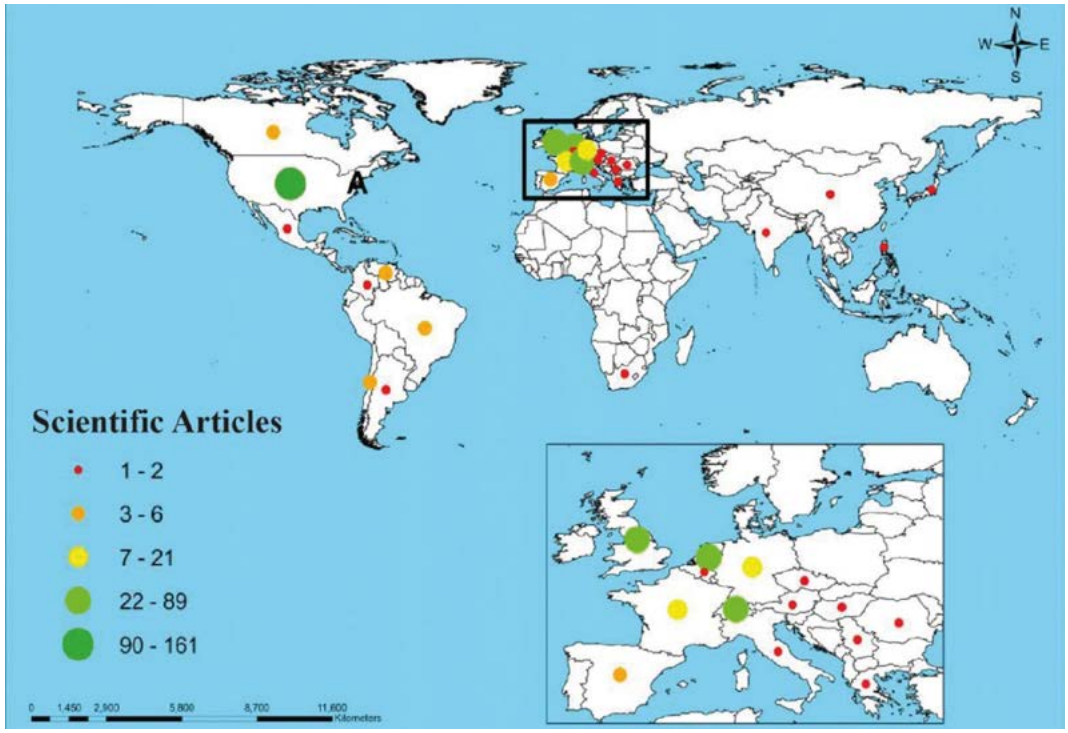

Figure 2. Spatial location of scientific production in the topic of agroecological strategies in the world from 1987 to 2021.

Bibliometric indicators

Of the texts, 97.19% (450 texts) were published in English, 1.30% (6) in Spanish, 0.86% (4) in French, 0.43% (2) in Portuguese, and 0.22% (1) in German. According to Li and Zhao (2015), English is the language adopted as universal by the scientific community, which is why the publications in English have a greater probability of dissemination among the international community. Table 1 shows that the sectors where the most research has been developed are those related with the topics of Agriculture (185 texts, 39.96%) and Ecology (125, 27.00%); this agrees with what was reported by Altieri and Nicholls (2017) who found that among the main agroecological strategies that have been implemented, there are the following: diversification of crops, maintenance of local genetic diversity, animal integration, addition of organic matter to the soil, and water harvest.

However, Altieri and Nicholls (2013) highlight the importance of developing research in topics such as the creation of early climate information systems, disaster prevention and mitigation projects, which as Table 1 shows are incipient topics: Geography (8 texts, 1.73%), Geology (4, 0.86%) and Meteorology (2, 0.43%); therefore, they represent an area of opportunity for the development of more research in these sectors.

The 463 studies analyzed were published in 250 scientific journals. A total of 115 articles, 24.84%, and 8127 bibliographic citations, 27.18%, were concentrated in 10 journals with impact factors higher than 3 (Table 2). Among these 10 main journals, 3 are edited in America, specifically in USA and 7 in Europe (Switzerland 5, United Kingdom 1, and Netherlands 1), where the main editorial houses Elsevier (4), Springer (3) and MDPI (2) coordinate the works for publication; according to Santillán-Fernández *et al.* (2021), this

Table 1. Sectors of research where scientific texts have been published with the topic of agroecological strategies in the world from 1987 to 2021.

Sector	Scientific Articles		Bibliographic Citations	
	Number	%	Number	%
Agriculture	185	39.96	11256	37.65
Ecology	125	27.00	8949	29.93
Entomology	26	5.62	1528	5.11
Botany	20	4.32	1287	4.30
Food	19	4.10	1263	4.22
Biotechnology	17	3.67	960	3.21
Livestock	17	3.67	966	3.23
Anthropology	11	2.38	684	2.29
Biodiversity	10	2.16	640	2.14
Rural Development	8	1.73	558	1.87
Geography	8	1.73	535	1.79
Medicine	5	1.08	294	0.98
Molecular Biology	4	0.86	530	1.77
Geology	4	0.86	250	0.84
Forest	2	0.43	70	0.23
Meteorology	2	0.43	126	0.42
Total	463	100.00	29896	100.00

Table 2. Bibliometric indicators of the main journals that published scientific articles about agroecological strategies in the world from 1987 to 2021.

Journal	Articles		Bibliographic Citations		Journal Information		
	Number	%	Number	%	Factor JCR	Editorial	Country of publication
Agroecol Sust Food	26	5.62	1479	4.95	3.039	Taylor and Francis Group	United Kingdom
Sustainability-Basel	23	4.97	1924	6.44	3.251	MDPI	Switzerland
Agr Ecosyst Environ	13	2.81	717	2.40	5.567	Elsevier	US
Agron Sustain Dev	13	2.81	1590	5.32	5.832	Springer	Switzerland
Agr Hum Values	11	2.38	737	2.47	3.295	Springer	Switzerland
Agr Syst	7	1.51	434	1.45	5.37	Elsevier	US
Agroforest Syst	6	1.30	312	1.04	2.549	Springer	Switzerland
Agronomy-Basel	6	1.30	356	1.19	3.417	MDPI	Switzerland
Eur J Agron	5	1.08	265	0.89	5.124	Elsevier	Netherlands
Int J Food Microbiol	5	1.08	313	1.05	5.277	Elsevier	US
Other (240)	**348**	**75.16**	**21769**	**72.82**			
Total (250)	**463**	**100**	**29896**	**100**			

helps to maintain the impact of the publications by increasing the probability of reaching a higher number of users.

Coauthorship network

In the 463 texts analyzed, 429 different first authors were found, and between first author and coauthors there was a total of 1942 different individuals. The network of authors and coauthors (Figure 3) was made up of 1942 nodes (authors) and 1708 aspects

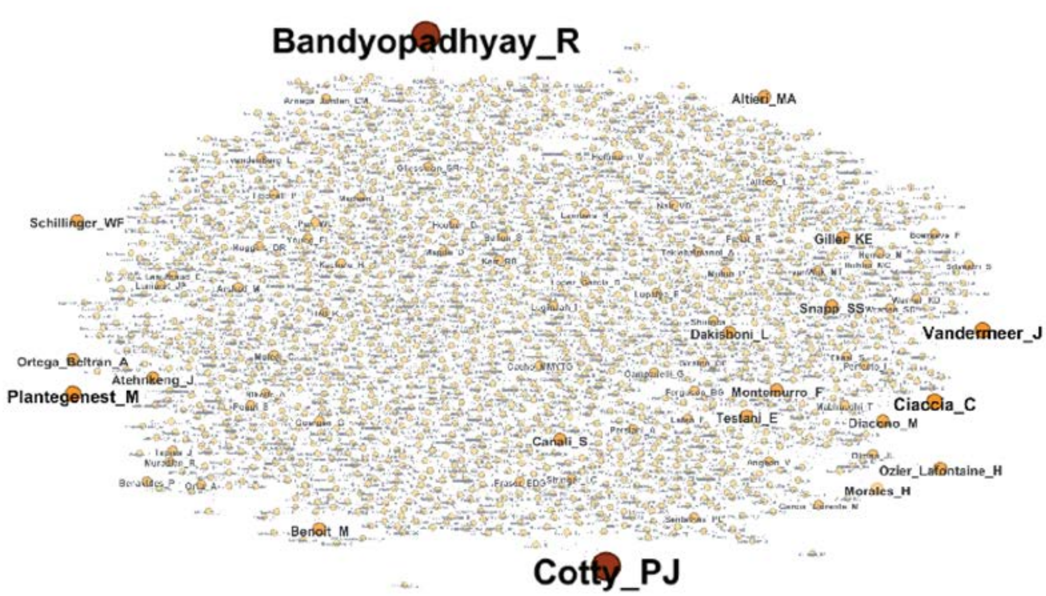

Figure 3. Network of authors and coauthors in the world who have published scientific articles about agroecological strategies from 1987 to 2021. The size of the node corresponds to their productivity.

(links). The links in a coauthorship network analysis are important because it is through them that an author can reach certain ideas, knowledge and information that is socially distant from him (Granovetter, 1973). The main authors who developed research in the area of agroecological strategies from 1987 to 2021 were Cotty_PJ (10 texts) from the University of Arizona (USA), and Bandyopadhyay_R (9) from the International Institute of Tropical Agriculture (Nigeria); both authors have focused their studies on the agricultural sector.

The density of the network had a value of 0.002, which implies that for the topic of agroecological strategies there is not much collaboration between the authors. The density is an indicator in the analysis of coauthorship networks that implies that the nodes interact (are linked) between one another; mathematically, it is a value within the interval [0 to 1], and the closer to 1, the interaction in the network is greater (Aguilar-Gallegos *et al.*, 2016). The low connection of the authors in the research network was evident since a coauthorship mean of 3.83 and a mode (49) of one author per text were found; 98 texts presented more than 6 authors, with extreme values (3 texts) of more than 24 authors.

CONCLUSIONS

The exponential growth that the publication of scientific articles has sustained in topics of agroecological strategies since the year 2010 is a reflection of the importance the topic has acquired among the scientific community as a way of generating knowledge, with the aim of seeking strategies to sustain and even increase the current production of foods, without exhausting natural resources and adapting the modes of production to the present and future climate variations. However, most of the studies have been centered on techniques for agricultural sustainability such as diversification of crops, maintenance of local genetic diversity, animal integration, addition of organic matter to the soil, and water harvest; this leaves a gap in the generation of knowledge in topics such as the creation of early climate information systems and disaster prevention and mitigation projects. Therefore, these areas represent an opportunity for a greater development of research.

The study also found that consolidated economies (USA and European countries) have promoted the development of research about the topic of agroecological strategies to strengthen food quality, while in developing countries such as the Latin American, where Mexico, Argentina, Chile and Brazil are located, it seems that the priority is food security, leaving a wide margin for the development of research in the region.

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Practices and perceptions of fall armyworm (*Spodoptera frugiperda* Smith) and striped grass looper (*Mocis latipes* Guenée) management in maize

Álvarez-Galicia, Alejandra[1]; Fajardo-Franco, Marja L.[1*]; Aguilar-Tlatelpa, Martin[1]; Hernández-Guzmán, Karina[1]

[1] Universidad Intercultural del Estado de Puebla. Calle Principal a Lipuntahuaca S/N, Lipuntahuaca, Huehuetla, Puebla, C.P. 73475.
* Correspondence: marjaliza.fajardo@uiep.edu.mx

ABSTRACT

Objective: To describe practices and perceptions about the fall armyworm and striped grass looper management in maize crops in La Pahua, Francisco Z. Mena, Puebla.

Design/Methodology/Approach: A descriptive study with probability sampling was performed. From April to June 2021, n=79 semi-structured interviews were conducted.

Results: The management of the fall armyworm and striped grass looper is carried out through the application of synthetic insecticides, mainly chlorpyrifos ethyl (81%). The minimal use of personal protective equipment for its application (75%) reflects the lack of knowledge of the population about health risks in the short, medium, and long term; however, producers are interested in receiving technical advice (56%) and are open to the use of alternatives, such as the use of traps (1%) and plant extracts (66%).

Study Limitations/Implications: This is a specific case study; therefore, the results are limited to descriptive statements about the study area.

Findings/Conclusions: The fall armyworm and striped grass looper are controlled with chemicals formulated with chlorpyrifos-ethyl alone or combined with permethrin. These chemicals are applied without the use of appropriate protective equipment. The population showed interest in incorporating other alternatives to synthetic insecticides, especially plant extracts and traps.

Keywords: Opinion, synthetic insecticides, *Zea mays*, bioinsecticides.

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INTRODUCTION

Mexico is the 8th largest maize producer in the global market with a production of 27,228,242 t (SIAP, 2020). This grain accounts for 89.1% of the cereal volume in the country, with an annual per capita consumption of 331.9 kg (SIAP, 2021b). The State of Puebla ranks 10th nationally with a production of 1,165,996 t of grain, out of which 29,926 t are produced in the region of Huachinango. Within this region, the municipality of Francisco Z. Mena ranks second with 3,238 t (SIAP, 2021b).

This crop is impacted by insect pests (CESAVEP, 2020) —especially the fall armyworm (*Spodoptera frugiperda*) (J. E. Smith) (Lepidoptera: Noctuidae)—, causing 30-70% losses in grain production and, consequently, economic losses for producers (Lugo-García *et al.*, 2017; Martínez-Núñez *et al.*, 2018; SENASICA, 2017).

Crops in the municipality of Francisco Z. Mena have not only been affected by the fall armyworm, but also by the striped grass looper (*Mocis latipes*) (Guenée) (Lepidoptera: Noctuidae). In order to design management strategies to control the pests that affect maize, the producers' perception and knowledge about their environment must be understood, in order to support the local and cultural wealth, as well as the environment care (Cortés-Rodríguez and Venegas-Cardoso, 2011). The objective of the study was to describe the farmer practices and perceptions in the community of La Pahua, Francisco Z. Mena, Puebla about the fall armyworm and striped grass looper management.

MATERIALS AND METHODS

The study was conducted in La Pahua, Francisco Z. Mena, Puebla. According to the Rural Medical Unit, 411 families live in the community and, based on this information, the sample size was estimated for the application of semi-structured interviews (Castañeda-Guerrero et al., 2020).

$$n = \frac{NZ^2_{\alpha/2} p_n q_n}{Nd^2 + Z^2_{\alpha/2} p_n q_n}$$

Where: n: Sample size; N: Population size = 411 families; d: accuracy = (0.10); $Z^2_{\alpha/2}$: reliability = 95% = 1.96; $p_n = 0.5$; $q_n = 0.5$.

A total of n=79 interviews were conducted and each one included three sections: 1) general data, 2) main pest control strategies in maize crops, with emphasis on the fall armyworm and striped grass looper, and 3) knowledge about plants with potential as insecticides. The taxonomic identification of the aforementioned plant species with potential bioinsecticide properties was based on their botanical characteristics. Subsequently, herborized specimens were transferred to the XAL herbarium of INECOL (Institute of Ecology) where their identity was corroborated. The information collected was systematized in a database and then analyzed by descriptive statistics.

RESULTS AND DISCUSSION

Characteristics of maize producers

Interviewees were between 20 and 80 years old. Ninety-five percent were men and 5% were women. The predominant language was Spanish (95%), while 5% said they spoke Totonac. While 95% of the interviewees were farmers, 5% are homemakers who also work the land. Regarding their education level, 6% are illiterate, 5% have not completed elementary school, 40% have completed elementary school, 29% have completed junior high school, and 20% have completed high school.

Management of the fall armyworm and striped grass looper

Maize pest management in the study area is done by chemical control. Accordingly, the insecticide mostly used in the La Pahua community to control the fall armyworm and the striped grass looper is Foley® (81%), while the less used are Lorsban® (10%) and Foley Rey® (9%). The latter is applied with a 10-20 ml dose dissolved in 20 L of water (capacity of a backpack sprayer), every 15 days during the first 30 days of growth of the maize seedling and once a month thereafter.

Most farmers either do not wear personal protective equipment when handling or applying insecticides (68%) (Figure 1a) or the equipment they wear is incomplete —*i.e.*, it is limited to only gloves, masks or goggles—, mainly due to lack of economic resources (25%) (Figure 1b). The use of sprinklers in poor condition was also identified. FAO has reported that 99% of agricultural workers are exposed to acute intoxications by direct and indirect contact and by the lack of appropriate protective equipment for handling and application (Guzmán-Plazola *et al.*, 2016). Farmers in the community even mix pesticides (*e.g.*, chlorpyrifos-ethyl with permethrin), assuming that this increases their potential.

Risks of poisoning are latent in these practices. The handling of insecticides can have harmful effects on health, especially on the respiratory tract, digestive system, skin, and eyes —either as a consequence of potential spills on the skin, accidental splashes on the face, or inhalation—, causing symptoms such as vomit, nausea, weakness, headaches, drowsiness, and behavioral alterations, among others (Nava-Pérez *et al.*, 2012). Medium and long-term health repercussions may include cancer, Parkinson's disease, Hodgkin's disease, Alzheimer's disease, endocrine alterations (sterility, diabetes, immunosuppression, kidney impairment, etc.), and hepatotoxic and mutagenic effects, as well as neurocognitive problems (Nava-Pérez *et al.*, 2012) (Díaz-Vallejo *et al.*, 2021).

Likewise, a lack of training was identified: 99% of the interviewees mentioned that they do not receive technical advice for pest management (Figure 1c). They are also unaware about the toxicity and health damage caused by agrochemicals and, just like in other rural regions of Mexico, they have a low perception of risk (62%) (Figure 1d and Figure 1c) (Herrera-Moreno *et al.*, 2018; Ordoñez-Beltrán *et al.*, 2019; Gómez-González, 2017).

Therefore, the following measures should be applied: promoting the use of personal protective equipment; training and advice for the proper handling of pesticides; and raising awareness about the environmental and human health damages caused by the improper handling of agrochemicals.

In addition, 9% of the interviewees declared that they are willing to incorporate other alternatives for the control of the fall armyworm and the striped grass looper (Figure 2a) and 64% are also willing to use other management alternatives, such as traps and plant extracts (Figure 2b).

Given these results, it is relevant to motivate and make the population aware about the integration of environmentally friendly alternatives into their pest control practices (*e.g.*, the use of plant extracts), due to their safer and low-cost bioactives (Rodríguez-Montero *et al.*, 2020). In addition, this type of biopesticides is characterized by metabolites that quickly degrade in the environment. About 1,600 plant species with insecticidal, attractant, and repellent properties have been reported (Cerna-Chávez *et al.*, 2020).

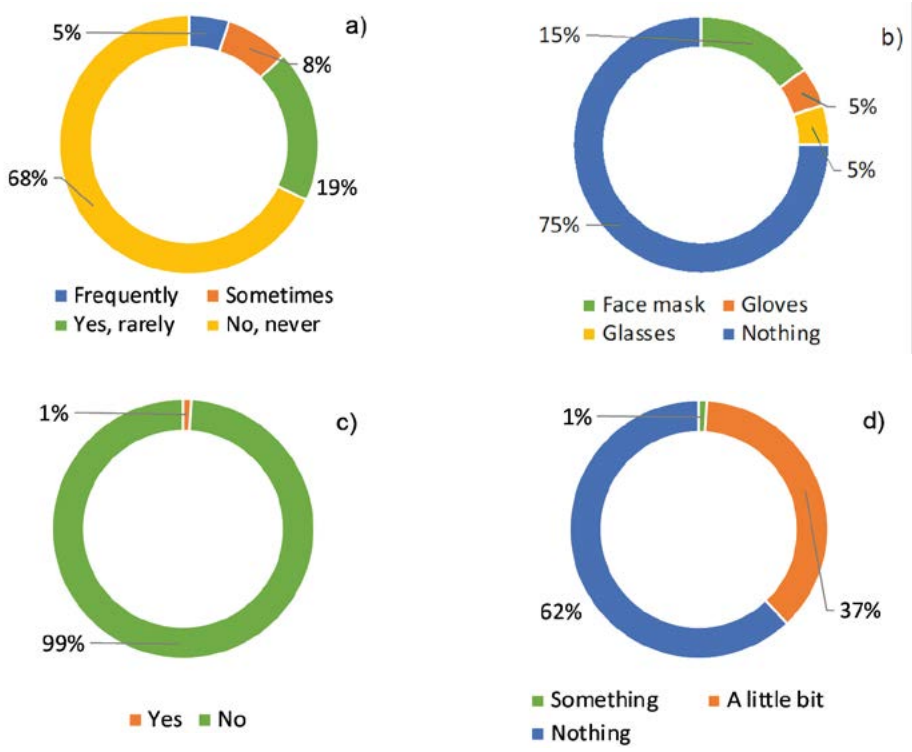

Figure 1. Characteristics and opinions on the control of the fall armyworm and striped grass looper in La Pahua, Francisco Z. Mena. a) Use of protective equipment, b) Type of protective equipment, c) Technical advice, d) Knowledge about health impacts of the use of synthetic insecticides.

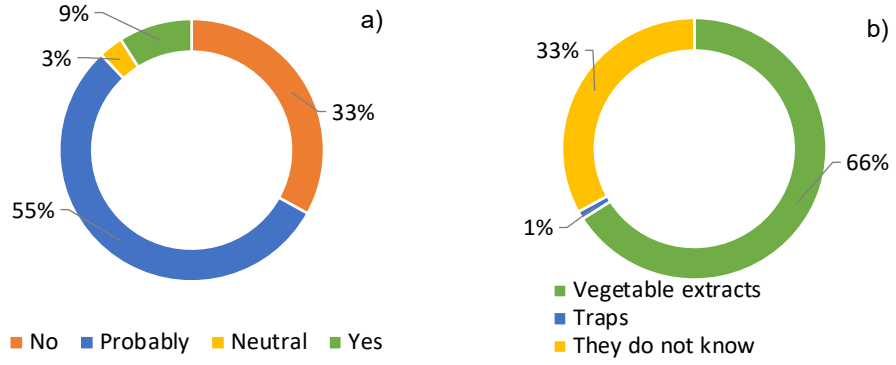

Figure 2. Opinions of maize producers from Francisco Z. Mena about alternatives for the management of fall armyworn and striped grass looper. a) Willingness to incorporate management alternatives in their practices, b) Alternatives that could be incorporated.

Local lore about plants with insecticidal potential

Regarding their knowledge of plants with pest control potential, 81% of the participants mentioned that they do not know plants with insecticidal effect, while 19% mentioned knowing little about the matter; therefore, recovering this type of knowledge could make a significant contribution to a greener agricultural production, which involves less health risks to producers and the general population.

The interviewees consider that some of the plants that grow in La Pahua could be used to make insecticides from plant extracts, including the following: castor oil bean (*Ricinus communis* L.) from the Euphorbiaceae family, Angel's trumpets (*Datura metel* L.) from the Solanaceae family, arrowhead vine (*Syngonium neglectum*) from the Araceae family, parthenium weed (*Parthenium hysterophorus* L.) from the Asteraceae family, cacahuananche (*Gliricidia sepium*) from the Fabaceae family, Aztec marigold (*Tagetes erecta* L.) from the Asteraceae family, and neem (*Azadirachta indica*) from the Meliaceae family (Figure 3).

P. hysterophorus and *G. sepium*, as well as *S. neglectum*, *D. metel*, *R. communis*, *T. erecta*, and *A. indica* have been documented to have insecticidal properties. They have been used to control insect pests and, in some cases, Lepidoptera (Al-Snafi, 2017; Aragón-García *et al.*, 2015; Gahukar, 2014; Ortiz-García *et al.*, 2018; Rodríguez-Rodríguez *et al.*, 2016; UNAM, 2022; Villate-Gómez *et al.*, 2008).

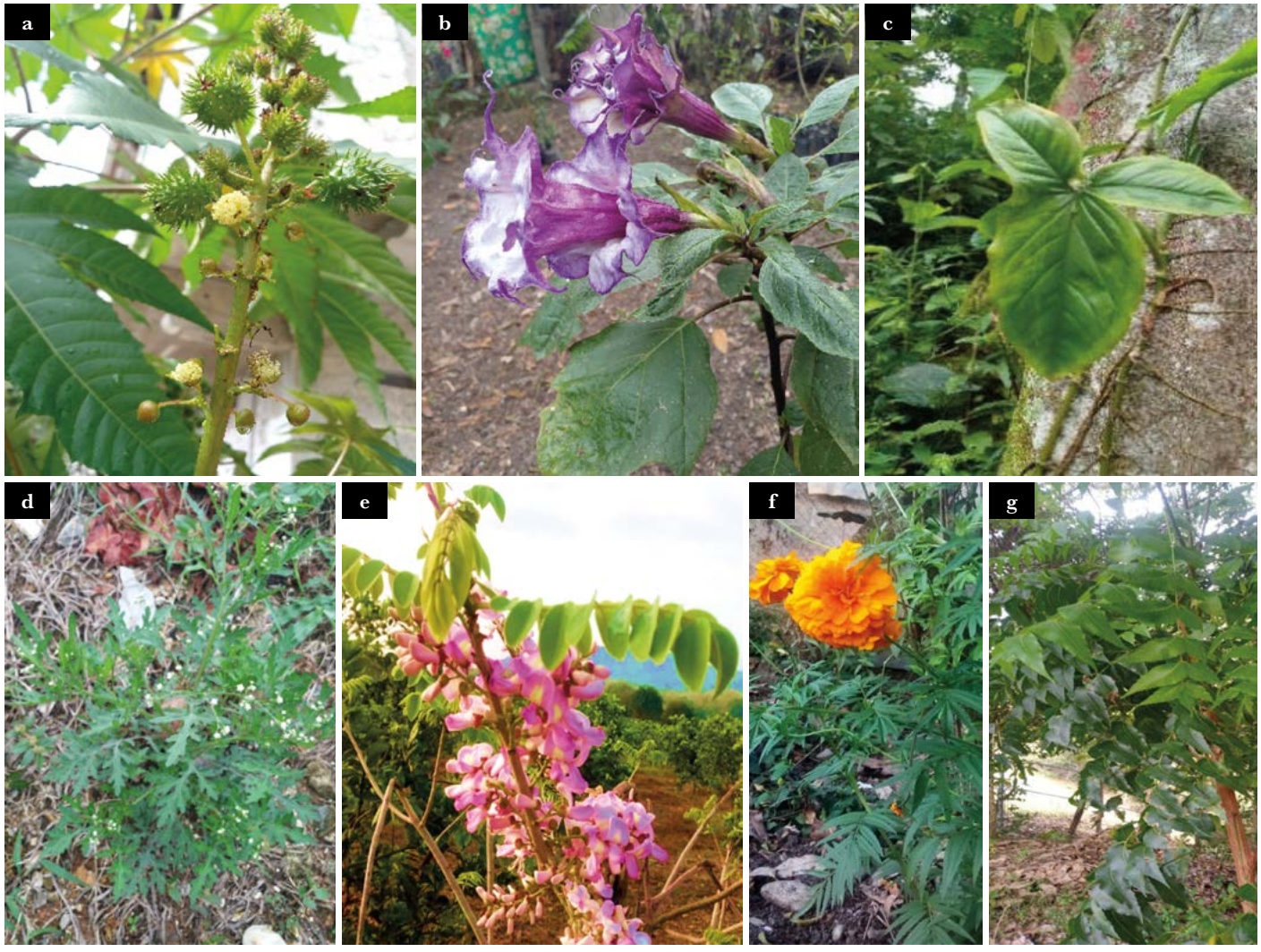

Figure 3. Plants with insecticidal potential according to the inhabitants of La Pahua, Francisco Z. Mena community a) castor oil bean (*R. communis*), b) angel's trumpets (*Datura metel*), c) arrowhead vine (*Syngonium neglectum*), Gliricidia (*Gliricidia sepium*), d) parthenium weed (*Parthenium hysterophorus*), e) and f) Aztec marigold (*Tagetes erecta*), and g) neem (*Azadirachta indica*) (personal file).

CONCLUSIONS

Maize producers from La Pahua, Francisco Z. Mena community, use synthetic insecticides formulated with chlorpyrifos-ethyl (alone or combined with permethrin) to control fall armyworm and striped grass looper in their crops; however, the minimal use of protective equipment reflects their lack of knowledge about the short, medium, and long-term health damage that these chemicals can have. However, the population is open to incorporate the use of plant extracts and traps into pest management practices, including the use of bioinsecticides made from local plant species.

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Estimation of the water footprint in the production of beef from European cattle in Mexico

Trujillo-Nava, Andrea[1]; **Valdivia-Alcalá, Ramón**[1*]; **Hernández-Ortiz, Juan**[1]; **López-Santiago, Marco A.**[1]

[1] Universidad Autónoma Chapingo. Texcoco, Estado de México, México. C. P. 56230.
* Correspondence: ramvaldi@gmail.com

ABSTRACT

Objective: To determine the water footprint of beef from Charolais cattle subjected to stable production and an established diet.

Design/Methodology/Approach: The water footprint was estimated using the methodology proposed by Hoekstra, in which the water footprints of the ingredients of the feed consumed are added to the total volume of water that the animal drank during its life.

Results: The estimated water footprint for beef in this research was 2,972.4 liters per kg, including the blue and green water footprint.

Study Limitations/Implications: The calculation of the gray water footprint is not included, although it is an indicator of the specific zone.

Findings/Conclusions: There is a difference between the water footprint obtained in this study and the footprint reported in the references, perhaps as a result, among other reasons, of the differences in diet and breed of the animals studied.

Keywords: beef, intensive production, European cattle, water footprint.

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INTRODUCTION

The production and consumption of beef in the world has increased in recent years (FAO, 2021), as well as the concern of consumers about the deterioration of the environment and water resources.

Mexico is the 6th producer of beef worldwide, with the states of Veracruz, Jalisco, San Luis Potosí, and Sinaloa as the main producers (SIAP, 2020). According to

Image by JackieLou DL at Pixabay

SEMARNAT (2018), agriculture and livestock account for 76% of the consumptive use of water in Mexico.

There is concern about climate change and its effects —including the increase in temperature, changes in the climate, and floods—, which have consequences for future generations, as well as potential health impacts (Oltra *et al.*, 2009). This situation has motivated researchers to identify indicators that monitor the effect of human activities on natural resources and the environment.

In this context, indicators of environmental impact and of the use of natural resources have been developed for the production of meat products. One of these is the water footprint (WF). WF is defined as an "indicator that shows the human appropriation of water resources. It refers to the total volume of fresh water used to produce something, whether it was incorporated into the product, evapotranspired by a crop, returned to another basin, or used by a body of water to assimilate the pollutant load" (Vázquez del Mercado and Lambarri, 2017). There are three types of WF: blue, green, and gray.

The water footprint has been estimated for several products, including: forage crops and their productive efficiency (Ríos *et al.*, 2015); the production of sugarcane cultivation (Cossio *et al.*, 2019; Garay *et al.*, 2022); bovine milk in Mexico (Flores *et al.*, 2017; Navarrete *et al.*, 2019); onion and tomato under irrigation conditions (Peñaloza *et al.*, 2020); the slaughter of already fattened cattle (Zambrano *et al.*, 2018); and various products of animal origin produced in different parts of the world (Esquivel and Salgado, 2020; Gerbens *et al.*, 2013).

Hanemann (2006) points out that water has an economic value, which may or may not be equal to its price; therefore, determining the water footprint is an useful way to know how much water is used in a production process. With that purpose in mind, the water footprint of beef from Charolais cattle subject to stable production and an established diet was determined.

MATERIALS AND METHODS

The state of Querétaro, Mexico, is the eighth largest beef producer nationwide. It produces 34,426.27 tons of beef with a production value of $2,503,438 pesos. The state has 18 municipalities, out of which the main producers are Ezequiel Montes, Querétaro, and San Juan del Rio (SIAP, 2022). This research used data from the municipality of Ezequiel Montes, given its importance for livestock production. This municipality is part of the Administrative Hydrological Region (RHA) IX Golfo Norte (Municipality of Ezequiel Montes, 2020). This region has medium water stress, an average rainfall of 855.3 mm per year, and excellent water quality overall (SEMARNAT, 2018).

An intensive beef production system was the subject of this study. The young bulls are placed in a confined area until they reach an optimum market weight; with this purpose in mind, they are provided a diet designed for weight gain (Callejas *et al.*, 2017). The breed chosen for the study was Charolais, which has the best productive behavior in relation to daily weight gains (Parra *et al.*, 2011; Bautista *et al.*, 2019).

The beef production and cattle fattening data were obtained from SIAP (2022); meanwhile, the forage ingredients and water consumption data were taken from information

for Mexico and are assumed to be homogeneous for the entire region (Rios *et al.*, 2015). The methodology applied to determine the WF of cattle in fattening was proposed by Mekonnen and Hoekstra (2012) and modified for this study.

First, the expression of the final product (FP) per animal is calculated as follows:

$$FP = \frac{P}{Pop}$$

Where P is the total annual production of beef in the state (kg/year) and Pop is the total population of cattle in the state.

Feed conversion efficiencies (FCE) are then estimated:

$$FCE = \frac{FC}{FP}$$

Where FC is the feed consumption per head (kg of dry mass/year/animal) and FP is the final product produced per head (kg of product/year/animal).

Then the total amount of feed consumed (Feed) is calculated:

$$Feed[n] = FCE \times P$$

Where $Feed$ is the total amount of feed consumed (ton/year) in the state, FCE is the feed conversion efficiency (kg of dry mass of feed/kg of product), and P the total amount of beef product (ton/year) in the state.

Subsequently, the WF of the feed consumed is determined with the following formula:

$$WF_{feed} = \frac{\sum_{p=1}^{n}\left(feed[n] \times WF_{prod}[n]\right)}{Pop}$$

Where $feed$ represents the annual amount of feed ingredient [n] consumed (ton/year), WF_{prod} [n] is the WF of feed ingredient [n] (m^3/ton), and Pop is the number of animals slaughtered per year in the state.

The WF of an animal is expressed as proposed by Mekonnen and Hoekstra (2012):

$$WF = WF_{feed} + WF_{water} + WF_{serv}$$

Where WF_{feed} is the water footprint of the feed consumed, WF_{water} is the total water that the animal drank during its life, and WF_{serv} is the water used in the farm service and corresponds to the gray water footprint (which was not considered for this study). The water footprint of an animal and its three components can be expressed in terms of m^3/

year/animal. When they are added over the lifespan of the animal, they are expressed as m^3 $animal^{-1}$.

RESULTS AND DISCUSSION

During the 120-days fattening period, the animals are constantly offered a sufficient volume of fresh and clean water. The volume that animals drink depends on the temperature, the environment, their body weight, and the dry matter content of the feed (NRC, 1996). The average temperature in the municipality of Ezequiel Montes was 16.7 °C. Meanwhile, the average weight gains for each period were the following: 316.8 kg at reception; 352.5 kg at the beginning, and 445.5 kg at finishing.

The average daily consumption per head of cattle in fattening was 38.9 L. Additionally, the total water consumed per head of cattle (4,664.4 L) was determined for the total days of fattening.

One feed formula was considered for each stage of fattening. All three formulas had the same ingredients and variation in the amounts administered. The amount of feed consumed by cattle in fattening depends on the weight of the animal and its weight gain. Table 1 shows the feed consumptions calculated considering the average weights and average weight gains.

The animals consume the calculated kilograms of feed per day, plus the barley straw that is added directly to the feeder every day. Table 2 shows the total feed consumption per period; these data were used to calculate the amount of ingredients consumed in each period, taking into account the formula used in each one.

The total feed consumption for each period and the quantities of ingredients that make up the formulas were used to calculate the total consumption of ingredients for each fattening period (Table 2). During the entire fattening period, 88,460.6 t of feed were consumed, plus 9,492.1 t of barley straw, giving a grand total of 97,952.7 t of feed for the total number of heads of cattle. The water footprint was determined for the following ingredients used in the formulation: corn silage (forage corn), corn stubble (corn for grain and stubble), sorghum (sorghum for grain and stubble), soybean (soybean for grain), alfalfa hay (alfalfa for fodder), and barley straw (barley for grain and straw). Table 3 shows the results obtained in this research. The water footprint of the beef is 2,972.4 L kg^{-1}.

Mekonnen and Hoekstra (2012) report that the global average of WF for beef in extensive, mixed, and intensive production systems is 15,400 L kg^{-1}, while the global

Table 1. Information on weight and food consumption

Periods	Average weight (kg)	Average daily gain (kg))	Consumption of dry food per day (kg)	Dry matter (%)	Food/day (kg)
Receive	316.8	1.6	7.818	73.19	10.682
Start	352.5	1.8	7.773	78.61	9.889
Ending	445.5	1.9	9.182	81.2	11.308

Source: Table developed by the authors.

Table 2. Food consumption by period and total. Source: self made.

Fattening periods	Duration of the period (days)	Feed consumption/ day/head (kg)	Feed consumption per period and total (t)
Receive	21	10.682	15,101.40
Start	21	9.889	13,980.30
Ending	78	11.308	59,378.80
Total			88,460.50

Table 3. Water footprint per kg of Charolais breed beef. Source: self made.

Total weight gain per head (kg)	Average daily gain (kg)	Total feed intake per head (kg)	Feed conversion (kg)	Feed conversion Carcass weight per head (kg)	Live weight (kg)	Carcass yield Meat (%)	Water Footprint ($L\ kg^{-1}$)
220	1.833	1455.31	6.615	322.4	520	62	2,972.4

average in intensive production systems is 10,244 $L\ kg^{-1}$. For their part, Esquivel and Salgado (2020) determined that the average WF for production in an intensive system in the United States is 4,552 $L\ kg^{-1}$; this result is closer to that obtained in this research. For references purposes, in the Comarca Lagunera region of Mexico, the WF is 13,570 $L\ kg^{-1}$ (Navarrete *et al.*, 2019).

The WF has a geographical and temporal component, which, in the case of beef production, is closely related to the diet. A better diet, a better choice of fattening breed with more productive efficiency, and the climate affect the magnitude of WF. A lower WF allows allocating water for other uses. Taking into account that the economic value of water is generally represented as monetary units, an efficient use of water increases overall productivity (Garay *et al.*, 2022).

CONCLUSIONS

This research contributes to understanding the issue of water footprint in production of beef. The volume of water necessary to produce one kilogram of beef was estimated and it matches the estimate of the blue and green WF. The gray water footprint must be incorporated into the analysis. This fact explains the lower magnitude of the WF with respect to that obtained for stabled cattle in the United States. The context of each research must also be incorporated in the analysis, including the following and other differences: the conditions of the study area, feed cultivation practices, difference in diets, weight gain, feed consumption, and animal carcass yield.

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Colegio de Postgraduados

Response of sugarcane (*Saccharum officinarum*) to organic fertilizer in northern Belize

Tun, Sergio J.[1]; Interián-Ku, Victor M.[1]; Cázares-Sánchez, Esmeralda[2]; Sosa-Madariaga, Jaime D.[2]; Hernández-Rodríguez, Gustavo[1*]

[1] Tecnológico Nacional de México, Campus Instituto Tecnológico de la Zona Maya, Carretera Chetumal-Escárcega, Km 21.5, Ejido Juan Sarabia, Othón P. Blanco, Quintana Roo, México, C.P. 77965.
* Correspondence: interku2002@yahoo.com.mx

ABSTRACT

Objective: To provide an ecological alternative for sugarcane producers in northern Belize evaluating the response of the crop to organic fertilizer (bovine biol and bokashi).

Design/Methodology/Approach: Ten treatments with different doses and combinations of bokashi and bovine manure-based biol were applied in a crop of the sugarcane variety B79-474 in the northern region of Belize.

Results: The combination of 3 t ha^{-1} of bokashi applied to the soil and 2% biol applied to the leaves recorded the best sugarcane yields, with an average weight of 2.0 kg per processable stalk.

Study Limitations/Implications: Bokashi production involves the use of large amounts of manure, which are difficult to transport and handle with simple tools.

Findings/Conclusions: The sugarcane variety B79-474 had a positive response to organic fertilizer (average yield: 2.0 kg per stalk and 330 t ha^{-1} of cane), applying 3 t ha^{-1} of bokashi to the soil and 2% biol to the leaves.

Keywords: Sugarcane, organic agriculture, organic fertilizers, sustainable development.

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INTRODUCTION

The production of sugarcane is of great importance in the world. Sugarcane is grown on 25.4 million hectares, distributed in more than 130 countries, with an average yield of 80 t ha^{-1} (FAOSTAT, 2013). The stalk of this crop is considered an agricultural fruit since it distributes and stores sugar. It consists approximately of 75% water and is made up of nodes and internodes, whose length, diameter, shape, and color differ, depending on the variety (SAGARPA, 2017).

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Sugarcane production is the main agricultural industry of Belize, making an important contribution to the agricultural sector, national income, and foreign exchange. It accounts for 7.8% of the gross domestic product (GDP), 10% of employment, and 6% of the currency income (Statistical Institute of Belize, 2015). However, the sugarcane production system is unsustainable, as a result of the environmental deterioration caused by the inefficient application of fertilizers and herbicides that have directly affected soil and water quality and caused a nutritional imbalance in plants (Chi *et al.*, 2017, 2020).

Other factors that affect this activity in Belize are the decrease in world demand for sugar and the competition from larger producers. Therefore, other markets have been explored, such as the United States of America, the Caribbean Community (CARICOM), and the fair trade market (Morris *et al.*, 2017).

Within the agronomic field, one of the alternatives to solve the nutritional problem and contain the rise in the prices of synthetic fertilizers is the use of organic fertilizers, which can be produced locally with vegetable and animal by-products. In addition, they have shown a positive impact on crop yield, because they enrich the soil with microorganisms and nutrients and likewise contribute to environmental conservation (Velasco-Velasco, 2018).

These fertilizers include bokashi ("fermented organic matter"), which was developed by Japanese scientists. Its use activates and increases the number of microorganisms in the soil; it also improves its physical properties, chemical characteristics, and organic matter (Ramos-Agüero and Terry-Alfonso, 2014). For its part, biol is one of the products obtained from the anaerobic fermentation of organic matter (*e.g.*, animal manure); its production dates back to the year 1900, when the first biodigester for the production of biogas was built in India (Aguilar and Botero-Botero, 2016). It is currently used to fertilize the leaves of crops, due to its mineral and organic molecule content which favors plant development. Cowo-Cruz (2022) recommends the use of agricultural organic fertilizers in northern Belize. Currently, the use of this type of fertilizer is non-existent in that region of the country. This recommendation is particularly pressing, given the well-known risks to human health involved in the use of agrochemicals.

In this context, the objective of the present work was to evaluate the response of sugarcane cultivation to the application of organic fertilizer to the soil and the leaves, as an economic and ecological production alternative for sugarcane producers in northern Belize.

MATERIALS AND METHODS

The study was carried out in the village of Louisville, Corozal district, in northern Belize. The region is located at 5 m.a.s.l, has Vertisols and Inceptisols, its climate is tropical with summer rainfall, and has an annual rainfall of 1,500 mm. The average temperature is 25.9 °C (National Meteorological Service of Belize, 2014).

Manure production

To produce biol, the following ingredients were used: 53 kg of fresh bovine manure, 110 L of chlorine-free water, 7 L of molasses, 1.3 L of whole milk, and 267 g of yeast. These

ingredients were mixed in a 200-L dark plastic container, which was hermetically sealed. Previously, a gas outlet was adapted on the lid, using a 1.5-m long and ½" diameter plastic hose. The other end of the hose was fitted in a 2-L plastic bottle filled with water, which served as a seal to prevent the entry of air into the container and guarantee the anaerobic fermentation process. The biol was applied to the crop two and a half months after it was manufactured.

To produce bokashi, 425 kg of bovine manure, 25 kg of green leaves, 25 kg of dry leaves, 2.5 kg of powdered charcoal, and 2.5 kg of stove ash were first mixed. Afterward, that mix was hydrated with a solution made with 2.5 kg of molasses, 300 g of yeast, and 100 liters of lukewarm water. The entire mixture was turned over every three days and the process ended after approximately 45 days when the temperature of the fertilizer equaled the environmental temperature (35°C on average). This fertilizer was used two months after its manufacture, to enable the mineralization process to conclude properly.

Land preparation

First, the weeds were manually eliminated with a machete, then the land was leveled plowing once with a tractor, followed by subsoiling to loosen the soil, and a harrowing pass to break clods or large soil particles. To establish the experimental plot, furrowing was done every 1.5 m × 30 m. In total, a treatment was established in each of the 10 furrows.

Cane variety used and sown

The Sugar Industry Research and Development Institute (SIRDI) recommends the use of the B79-474 variety for this area of Belize. Segments of 1 m in length, 1 to 2 cm in diameter, 7 to 8 months old, and with good vigor and development were used for planting. The sowing was carried out in February 2021.

Weed, pest, and disease control

The weeds were removed manually with a machete when their size exceeded 10 cm, to control them without the use of herbicides. Regarding pests, there are two major animals that have a negative impact on sugarcane production in northern Belize: the froghopper (*Aeneolamia* spp.) and the stem borer (*Diatraea* spp.) (Chaves Solera, 2012). However, they were completely eradicated with three applications of biol (with doses according to the analyzed treatments) and 227 grams of the biological control (*Metarhizium anisopliae*) plus 20 mL of *Dipax per* L.

Treatments

Ten treatments were evaluated in this research, applying bokashi organic fertilizers to the soil and bovine manure biol to the leaves. In addition, the synthetic chemical fertilizer most frequently used in the area (15-4-28 + 2Ca and 1.36Mg) was used as a control (Table 1).

Three months after the sugarcane sowing, the bokashi was applied as follows: a) 50% to the soil in the form of a stream, opening a channel in the furrow 10 cm away from the stalk, at a 5-cm deep, which was later covered with earth; and b) the remaining 50% was

Table 1. Treatments evaluated in sugarcane cultivation in northern Belize.

Treatment	Bocashi		Biol in water (%)	Fertilizer kg ha^{-1}
	t ha^{-1}	kg tra^{-1}		
T1	0	0	0	Control without fertilization
T2	0	0	0	250
T3	2	9.1	0	
T4	3	13.6	0	
T5	4	18.2	0	
T6	0	0	1	
T7	0	0	2	
T8	0	0	3	
T9	2	9.1	1	
T10	3	13.6	2	

applied 30 days after the first application. The amount applied per 30-m furrow (Table 1) was calculated as follows: ((Dose per ha in kg/66 furrows) /100 m per furrow) * 30 m of one furrow.

Based on the dose indicated for each treatment, biol was applied to the leaves every 30 days, before 9:00 a.m. (Table 1). A 20-L backpack sprayer was used and the nozzle was directed towards the underside of the leaves. The first application was carried out when the first leaves reached 30 to 40 cm long. In total, three applications were made.

Variables evaluated in stalks

Once the crop cycle was over (11 months after sowing), 20 processable stalks were randomly selected from the planting furrow. The green tip (between sections 8 and 10) and the dry leaves were removed from each stalk and the following elements were recorded: the diameter in the middle part (with a digital vernier), the length (with a tape measure), and the weight of the stalk (with a digital scale), as well as the weight of the dry leaves and the tip (with a digital scale). Additionally, 20 non-processable stalks were randomly selected and their total length and weight were also recorded.

Maturity index in processable stalks

A portable refractometer was used to measure °Brix in the first and last third of the stalk. Based on these data, the maturity index (MI) was calculated according to the methodology proposed by Viswa-Nath and Kasinath (1935), with the following formula:

$$Maturity\ Index\ (MI)\ \frac{°\text{Bx}\ of\ the\ upper\ internode\ of\ the\ stalk}{°\text{Bx}\ of\ the\ lower\ internode\ of\ the\ stalk}$$

Values close to 1 show greater maturity and values close to zero show less maturity.

Yield estimation and ratio of processable/non-processable stalks

To determine the number of processable and non-processable stalks, a 2-m border was left at the beginning of each row and three 1-m long sampling plots were established 5 m apart from each other. To estimate the yield, the average weight of the processable stalks was determined (eliminating dry leaves and the tip) and multiplied by the average number of stalks per linear meter, per 100 meters, and per 66 rows. To determine the processable:non-processable stalks ratio, the number of processable stalks was divided by the number of non-processable stalks.

Experimental design and statistical analysis

The experimental design was completely randomized, with 10 treatments and 20 repetitions, considering one plant as the experimental unit. The resulting data was subjected to an analysis of variance and a means comparison, using Tukey's test and the SAS version 9.0 statistical package (Statistical Analysis System).

RESULTS AND DISCUSSION

The measured variables recorded highly significant differences ($P \leq 0.001$) among the treatments, with the exception of stalk length which only registered significant differences ($P \leq 0.005$), as indicated in Table 2.

The stalk diameter, stalk weight, maturity index, and weight of dry leaves in processable stalks variables had a positive effect in combination with the application of bokashi and biol to the soil and the leaves, respectively. Treatment 10 (3 t of bokashi and 2% of biol) showed the highest average values (Tukey, $P \leq 0.05$). It is important to highlight that the separate use of these fertilizers did not have an obvious effect on the crop (T3-T8).

With respect to the weight of the stalk tip, the T10 recorded a lower average value, indicating that this treatment tends to increase the quality of the stalk (length, diameter, and °Brix) and to reduce the number of stalks that do not meet the grinding requirements, although all treatments had similar lengths (Table 3). Specifically, the stalk length has a statistically equal behavior, although the stalk diameter and maturity index variables are separated —with T10 recording the highest average values. According to Table 3, it is possible to distinguish the effect of the fertilizers: on the one hand, by themselves, bokashi and biol do not increase the average value of the variables in the plant, but, on the other hand, at high doses, their combination potentiates the effect (3 t of bokashi and 2% of biol per hectare).

Table 2. Mean squares of the analysis of variance for the variables measured in sugarcane to which organic fertilizers were applied in northern Belize.

SV	DF	SD	SL	SW	MI	NDL	WDL	TW	PW	PL
		Processable stems							Unprocessable stems	
Tre	9	120^{***}	0.2885^{*}	2.4^{***}	128.9^{***}	50.4^{**}	9214^{***}	0.0152^{**}	0.3849^{***}	0.7403^{***}
Error	190	14.9	0.1280	0.3200	17.9	19	1753	0.0056	0.1108	0.2057

SV=Source of variation; Tre=Treatment; DF=Degrees of freedom; *=≤0.05; **≤0.01; ***=≤0.001; ns=not significant; SD=Stalk diameter; SL=Stalk length; SW=Stalk weight; MI=Maturity Index; NDL=Number of dry leaves; WDL=Weight of dry leaves; TW=tip weight; PW=Plant Weight; PL=Plant length.

Table 3. Average treatment values and the response variables of sugarcane to organic fertilizers in northern Belize.

T	N	SD (mm)	SL (m)	SW (kg)	MI (%)	NDL	WDL (g)	TW (kg)	PW (g)	PL (m)
T1	20	27.2 bcd	1.77 a	1.66 abc	81.3 c	14.4 ab	93.5 bc	0.1805 ab	0.31 ab	1.23 abc
T2	20	24.4 d	1.73 a	1.23 bcd	87.1 ab	14.7 ab	111 ab	0.2504 a	0.18 b	1.25 abc
T3	20	23.8 d	1.70 a	0.99 d	85.2 bc	11.4 b	65.6 c	0.2121 ab	0.64 a	1.09 bc
T4	20	24.3 d	1.71 a	1.18 bcd	87.7 ab	10.8 b	62 c	0.2117 ab	0.15 b	1.05 bc
T5	20	28.9 ab	1.90 a	1.73 ab	88.0 ab	12.3 ab	89.9 bc	0.1736 b	0.30 b	1.47 ab
T6	20	26.6 bcd	1.78 a	1.27 bcd	89.0 ab	12.4 ab	97.7 abc	0.1894 ab	0.34 ab	1.23 abc
T7	20	28.8 abc	2.02 a	1.43 bcd	89.4 ab	13.2 ab	92.3 bc	0.2044 ab	0.38 ab	0.91 c
T8	20	24.9 cd	1.98 a	1.55 bcd	88.9 ab	12.9 ab	93.5 bc	0.1879 ab	0.41 ab	1.57 a
T9	20	25.5 bcd	1.88 a	1.12 cd	88.2 ab	12.9 ab	78.9 bc	0.1589 b	0.33 ab	1.15 abc
T10	20	31.2 a	1.98 a	2.14 a	89.9 a	16.2 a	136.4 a	0.1617 b	0.23 b	1.17 abc
HSD		3.91	0.3623	0.5728	4.2946	4.4434	42.4	0.0758	0.337	0.4592

T=Treatment; N=Number of repetitions after treatment; SD=Stalk diameter; SL=Stalk length; SW=Stalk weight; MI=Maturity Index; NDL=Number of dry leaves; WDL=Weight of dry leaves; TW=Tip Weight; PW=Plant weight; PL=Plant length; HSD=Honestly significant difference. Values with the same letters between rows are not statistically different (Tukey, $P \leq 0.05$).

According to the yield estimation, treatment 10 obtained the best results (330 t ha^{-1}), followed by 1 and 5, showing that the combination of high doses of organic fertilizers tends to achieve a higher yield (Table 4) than that reported by the sugar industry of Belize (54 to 64 t ha^{-1} on average). Meanwhile, during the 2020-2021 production cycle, Mexico reported 64 t ha^{-1} (Chaves Solera, 2012; Comité Nacional para el Desarrollo Sustentable de la Caña de Azúcar (CONADESUCA), 2022). For its part, the Inter-American Development Bank (2017) indicates that Belizean sugarcane has low quality and productivity (average yield: 42 t ha^{-1}) which is 50% lower than countries such as Guatemala and Nicaragua, which usually produce 100 t ha^{-1}. The ratio of processable:non-processable stalks indicates

Table 4. Estimated yield per hectare of sugarcane and processable:non-processable stalk ratio in northern Belize.

T	ASLM	AWS	ER (t ha^{-1})	PS	NPS	RP/NPS
T1	18	1.8	213	35	26	1.35
T2	17	1.5	168	33	20	1.65
T3	15	1.2	118	29	13	2.23
T4	16	1.6	169	30	13	2.31
T5	19	1.7	213	36	19	1.89
T6	17	1.7	190	33	17	1.94
T7	20	1.5	198	38	12	3.17
T8	18	1.5	178	35	19	1.84
T9	16	1.3	137	31	11	2.82
T10	25	2.0	330	49	29	1.69

T=Treatment; ASLM=Average stalks per linear meter; AWS=Average weight of stalks; ER=Estimated return; PS=processable stalks; NPS=non-processable stalks; RP/NPS=Ratio of processable: non-processable stalks.

that treatments 7, 9, 4, and 3 had a greater number of processable stalks for each non-processable stalks; however, their estimated yields are lower than those of T10, suggesting that these variables are independent from each other.

This work can demonstrate the high quality of organic fertilizers, since they even exceed the average yield of sugarcane harvest in Belize and they can replace synthetic chemical fertilizers and reduce production costs.

Similar studies confirm the findings of this work. For example, García-Peña *et al.* (2020) used bokashi and biol in *Cucurbita argyrosperma* and exceeded the average domestic yield by 50%, obtaining 4.48 t ha^{-1} of fresh fruit and 1.76 t ha^{-1} of dry seed. Likewise, Favorito *et al.* (2019) showed that the application of bokashi in corn cultivation increased grain yield by 20% compared to the other treatments. For their part, González *et al.* (2015) observed higher increases in stalk length, stalk diameter, and yield, applying 30% Supermagro (biol) to a watermelon crop. Meanwhile, Ferreira *et al.* (2013) recorded yields of 10 t ha^{-1} as a result of the application of 1,000 g m^{-2} of bokashi to broccoli cultivation. Finally, Diniz *et al.* (2008) recorded 12.5 t in the same species with the application of 25 t ha^{-1} of this fertilizer. All of this proves the importance, and above all the effect, of these organic fertilizers for the productivity of cultivated species.

Other studies show that the use of organic fertilizers in crops improves the physical, chemical, and biological properties of the soil. For example, Matos-Pech *et al.* (2022) used cover crops with the *Vigna unguiculata* L., *Phaseolus lunatus* L., and *Mucuna pruriens* L. species and registered a positive improvement in organic matter (3.6-3.9%), electrical conductivity, pH, macronutrient content (N, P, K, Ca, Mg, and Na), micronutrient content (B, Fe, Mn, Cu, Zn), and C:N ratio, as well as the sum of exchangeable bases (meq/100 g). For their part, Cervantes-Vázquez *et al.* (2022) found the same improvements in soil where watermelon was grown, as a result of the application of 6 to 9 t ha^{-1} of vermicompost combined with bovine manure as fertilizer. Similarly, Orozco Corral *et al.* (2016) record that applying 6 t ha^{-1} of vermicompost lead to an increase of $>10\%$ in water storage capacity, 83% in cation exchange capacity, $>24\%$ in organic matter, and $>113\%$ in microbial biomass. Likewise, Gashua *et al.* (2022) and Pérez *et al.* (2008) carried out a chemical characterization of the bokashi fertilizer and found the following mineral elements: Mo, N, P, K, Ca, Mg, Mn, Fe, Zn, Na, Cd, Cr, Cu, Ni, and Pb.

The chemical and nutritional improvements of the soil and crop yields are mainly based on the solubilizing action of microorganisms present in organic fertilizers on mineral elements. A few examples follow:

1) the biological fixation of N is caused by the action of free-living organisms or in symbiosis with the roots, as is the case of bacteria of the following genera: *Azospirillum*, *Enterobacter*, *Klebsiella*, *Pseudomonas*, *Burkholderia*, *Rhizobium*, *Ensifer*, *Bradyrhizobium*, *Azorhizobium*, and *Mesorhizobium* (Berrada and Fikri-Benbrahim, 2014; Dhayalan and Karuppasamy, 2021; Estrada-de los Santos *et al.*, 2001; Jarvis *et al.*, 1997).
2) P fixation is the result of the action of several bacteria and fungi. The bacteria include: *Erwinia*, *Pseudomonas*, *Bacillus*, *Rhizobium*, *Klebsiella*, *Burkholderia*, *Serratia*,

Achromobacter, *Agrobacterium*, *Micrococcus*, *Aerobacter*, *Flavobacterium*, *Enterobacter*, *Arthrobacter*, *Rhodobacter*, and *Pantotea*. For their part, fungi include: *Aspergillus*, *Penicillium*, *Trichoderma*, and *Fusarium* (Awasthi *et al.*, 2011; Jones and Oburger, 2011; Khan *et al.*, 2010).

3) Finally, K fixation is also carried out by both bacteria and fungi. The former includes *Bacillus mucilaginosus*, *B. Edaphicus*, *B. Circum*, *Arthrobacter* spp., *Burkholderia*, *Acidithiobacillus ferrooxidans*, *Enterobacter homaechei*, *Paenibacillus glucanolyticus*, *Paenibacillus frequentans*, *Cladosporium*, and *Aminobacter*. The latter includes: *Aspergillus terreus*, *Aspergillus niger*, *Rhizobium*, and *Pseudomonas* (Meena *et al.*, 2014; Pattnaik *et al.*, 2021; Prabina *et al.*, 2022; Upadhyay *et al.*, 2022).

CONCLUSIONS

T10 (3 tons per hectare of bokashi applied to the soil + 2% bovine biol diluted in water applied to the leaves) was the best treatment, with an average weight of 2.0 kg per stalk, a total weight of 330 t ha^{-1}, and a maturity index of 89, which exceeds all treatments. In addition, the use of bovine biol as a spotted fly (*Aeneolamia varia*) repellent decreases the population of this pest in the plantation, although it does not eradicate it.

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Seed germination of four amaranth species (*Amaranthus* spp.)

Ramírez-Bautista, Marco Antonio[1]; Chiquini-Medina, Ricardo Antonio[1]; Pech-May, Nelson Jesús[1]; Castellot-Pedraza, Vicente[1]; Lee-Borges, Brígido Manuel[1]; Escalante-Poot, José Rubén[1]; Ganzo-Guerrero, Wilbert Inocente[1]; Gonzalez-Lazo, Edith[3]; Villarino-Valdivieso, Ariel-Miguel[1]; Marín-Quintero, Manuel[1]; Novelo-Salazar, Rafael Adrián[2]; Dzib-Castillo, Benito Bernardo[1*]

[1] Tecnologico Nacional de México, Instituto Tecnológico de Chiná, Calle 11 entre 22 y 28, Colonia Centro Chiná, Campeche, México, C.P. 24520.
[2] Centro de Estudios Tecnológicos del mar No.2, Carretera Campeche-Hampolol, Col. Palmas, Campeche, México, C.P. 24027.
[3] Tecnologico Nacional de México, Instituto Tecnológico Superior de Escárcega, calle 85 S/N entre 10-B, Col. Unidad Esfuerzo y Trabajo No.1, Escárcega, Campeche, C.P. 24350.
* Correspondence: bernadzib@yahoo.es

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ABSTRACT

Amaranthus (*Amaranthus* spp.) is a species of great importance that benefits human and animal nutrition; therefore, its reproduction must be based on useful information obtained from rigorous experiments. Consequently, the aim of this work was to determine the germination of 20 accessions from four *Amaranthus* spp. The work was carried out at the Instituto Tecnológico de Chiná, Campeche, Mexico, using seeds from Africa, Asia, Greece, South America, the US, and Mexico, donated by The North Central Regional Plant Introduction Station (NCRPIS), Iowa State University. After they were weighed and measured, the seeds were placed in Petri dishes and kept in the dark inside a germination chamber, at 27 °C and with a 54% relative humidity. Germinated seeds were counted and removed every 24 hours. The analyses were carried out using the ANOVA test in order to identify weight, length, and germination differences between accessions. The results showed no statistical differences in seed length, neither between species nor accessions; however, there were statistical differences in the weight, both between species and between accessions. A. hypochondriacus from India recorded the highest weight (0.00093±0.000075 g). Regarding germination, there were statistical differences between the various evaluation periods (24 and 48 hours): the highest germination was recorded by *A. hypochondriacus* and *A cruentus*. Therefore, the following conclusion was reached: seed germination is different between species and accessions.

Keywords: *Amaranthus* spp., germination, amaranth, adaptation, accessions.

INTRODUCTION

Amaranthus (*Amaranthus*) is a dicotyledon pseudocereal (Amaranthacea), known for its nutritional value. Its structure does not contain gluten and therefore can be used to develop

Image by Birgit Böllinger at Pixabay

food formulas for coeliac patients (Pagamunici *et al.*, 2014). Plants from this genus can be easily grown under water scarcity and high temperature environmental conditions (Silva *et al.*, 2019; Zhang *et al.*, 2019).

The Amaranthacea family has a cosmopolitan distribution: its 183 genera and 2,050 to 2,500 species can be found in arid, saline, and disturbed environments (Stevens, 2001). Although it is native to Mesoamerica and is adapted to regions located from 0 to 2,600 m.a.s.l., in recent times it has been introduced in milieux beyond its original adaptation range (Espitia *et al.*, 2021).

To understand the reproduction processes of seed species, their germplasm must be studied first. During the seed formation process, most species undergo changes first in the dessication state and later in the latency, until the point of germination (Legaria-Solano *et al.*, 2000). This germination process involves rehydration, the use of reserves, and the development of synthetical structures that will enable the seedling to take on an autothropic existence mode (King, 1991).

Water absorption is one of the processes that enable the start of germination. Amaranthus seeds absorb the maximum rate of water between 0 and 12 h, when water contains no salts; during this period, seeds absorb 50% of the water, increasing their weight in the same ratio. At 48 h, their weight increases by 60%, with just a 10% water absorption. Meanwhile, between 48 and 60 h, the increase reaches 93%, equivalent to an $\leq$30% absorption. Finally, seeds that were hydrated with water containing a -1.6 MPa concentration of NaCl were able to absorb 35% of water by 60 h (Legaria-Solano *et al.*, 2000).

Consequently, further studies must be carried out to understand every factor involved in germination. Although studies have been carried out on this subject, additional research is still required to determine and understand this process for each species and consequently to increase the efficiency of their reproduction. The environmental adaptation and survival characteristics are highly likely to change from one population to the next, particularly if the environmental conditions are significantly different between the places in which the populations live (Silvertown and Charlesworth, 2001).

The germination process is influenced both by the physiological characteristics of the seed itself and its environmental conditions, promoting a sequence of metabolic activities which result in the development of the embryonic axis (Bewley *et al.*, 2012).

This characteristic grows in importance when it is considered as a major adaptation trait that shows the high intraspecific phenotypic variability resulting from seed latency (Christal *et al.*, 1998). In most cases, the differences between the latency levels of the populations have been attributed to their production or germination conditions (Allen and Meyer 2002; Lacerda *et al.*, 2004). However, several studies have proven that different populations from several species can have similar latency levels and seedling emergence patterns, regardless of the diverse environments in which they are originated (Grundy *et al.*, 2003). Therefore, the objective of this work was to determine the morphological characteristics and the germination behaviour under lab conditions of seeds from 20 accessions from four *Amaranthus* spp. with different origin.

MATERIALS AND METHODS

Study area

The work was carried out in 2019, at the facilities of the Tecnológico Nacional de México, Campus Chiná, Campeche, Mexico. Campus Chiná is located at 18° 50' 11" N and 90° 24' 12" W and an altitude of 20 m.a.s.l. The weather is Aw1, with a 1,138 mm of annual rainfall and a mean annual temperature of 26.8 °C.

Morphological characterization of the seeds

The seeds used in this study belonged to four species, from which 20 accessions were taken (Table 1). They were donated by The North Central Regional Plant Introduction Station, Iowa State University (NCRPIS). A total of n=20 seeds was randomly selected to determine seed morphology. Their weight and length were measured with a CY 304® analytical balance (ACZET) and a graduated scale microscope, respectively.

Germination test

For the germination test, n=20 seeds from each of the 20 Amaranthus accessions were used. The seeds were placed on blue blotting paper and moistened with water in 5"×5¼" plastic boxes. Subsequently, they were established in a germination chamber, in total darkness. at 27 °C, and with a 54% relative humidity ((http://www.ars.usda.gov/mwa/ames/

Table 1. Accessions, species, and origin of the *Amaranthus* spp. seeds evaluated.

No	Accesión	Especie	Planta	Procedencia
1	PI 667162	*Cruentus*	RRC 638	México
2	PI 604567	*Hybridus*	Mapes 830	Puebla, Mex.
3	PI 482048	*Hybridus*	TGR 540	Zimbawe
4	PI 490347	*Hybridus*	J&T 137	Burkina Faso
5	PI 500249	*Hybridus*	ZM 1845	Zambia
6	PI 526228	*Hybridus*	AMM 384	Zimbawe
7	PI 605351	*Hybridus*	RRC 847	Grecia
8	PI 511721	*Hypochondriacus*	HH 93	México
9	PI 604577	*Hypochondriacus*	Mapes 847	Puebla, Mex.
1	PI 6652860	*Cruentus*	RRC 1266	Venezuela
1	PI 490607	*Caudatus*	HH 54	Bolivia
1	PI 482051	*Cruentus*	TGR 603	Zimbawe
1	PI 494777	*Cruentus*	ZFA 3653	Zambia
1	PI 527570	*Cruentus*	IZ 166	Rwanda
1	PI 566897	*Cruentus*	Kerala Red	kerala, India
1	PI 628784	*Cruentus*	RRC 1139	Puebla, Mex.
1	PI 500267	*Cruentus*	ZM 2309	Zambia
1	PI 647848	*Cruentus*	RRC 548	California, USA
	PI 658729	*Cruentus*	RRC 844	Rep. Cen. Africana
2	PI 615696	*Hypochondriacus*	Annapurna	India

ncrpis). They were counted every 24 h and germinated seeds with >0.5-mm long radicles were removed.

Data analysis

The germination percentage was calculated based on the number of seeds placed in the Petri dishes and the number of germinated seeds. On the one hand, an analysis of variance (ANOVA) was carried out to identify differences between seed weight; on the other hand, a regression analysis was used to develop an equation that estimated the number of seeds per kilogram (Di Rienzo, 2020).

RESULTS AND DISCUSSION

The statistical analysis showed no difference regarding the length variable. However, the average length value was 25.77±20.55 μm, while the shortest seed length was recorded by *A. caudatus* (17.00±7.0 μm), followed by *A. hybridus* (20.67±13 μm) and *A. cruentus* (25.73±19 μm). Meanwhile *A. hypochondriacus* had the longest seeds (41.22±32 μm). Regarding their origin, the longest seeds belonged to *A. hypochondriacus* from the state of Puebla, Mexico (62.00±11 μm) (Figures 1 and 2).

Statistical differences were recorded between species (P=0.0046), as well as between origins (P<0.0001), regarding the seed weight variable, which fluctuated between 0.00015 and 0.00093 g. *A. hypochondriacus* from India had the heaviest seeds (0.00093±0.000075 g), while the seeds of *A. cruentus* from Mexico City recorded the lowest weight (0.00015±0.000092) (Figures 3 and 4). The abovementioned results are different from the values reported by Nieto (1990), who recorded seeds that weighed between 0.0001 and 0.0003 g and were 1 to 1.5 mm long.

The results do not differ from the findings of several studies in which seed weight were compared. For example, Ramírez-Sánchez (2006) recorded a seed weight of 0.00076 g and 0.0006-0.0008 g for *A. caudatus* and *A. hypochondriacus*, respectively. For his part, Spehar (2003) recorded a seed weight of 0.0007 g and 0.0003-0.0004 g for *A. cruentus* and *A. hybridus*, respectively. The individual seed weight of *A. cruentus* recorded

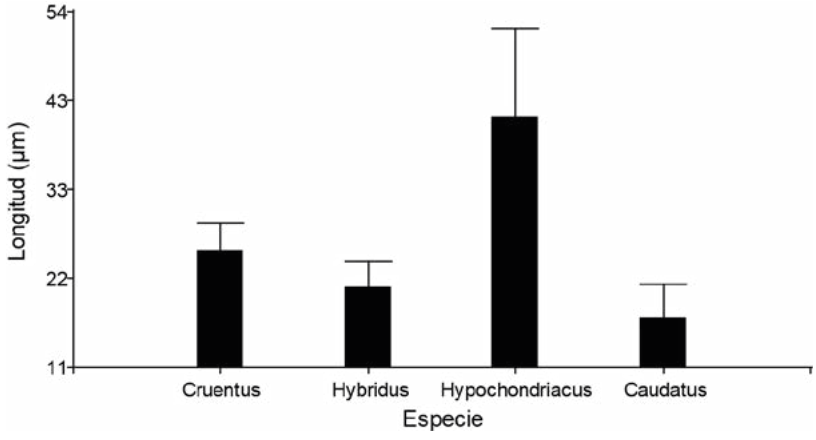

Figure 1. Seed length from several *Amaranthus* spp.

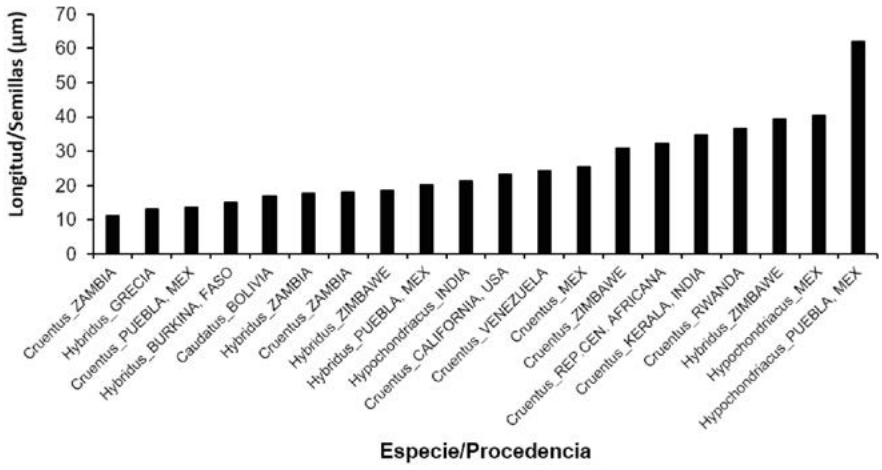

Figure 2. Seed length of several *Amaranthus* spp. from several origins.

the highest variation. Variations are mainly the result of weather, photoperiod, and sowing method.

An equation based on the seed weight data was applied to estimate the amount of seeds that can be obtained per kilogram, through a potential regression analysis (Figure 5) (Equation 1).

$$Seeds/kg = 10297619.34e^{-3270.17\,(X)} \quad R^2=0.90 \qquad \text{(Equation 1)}$$

No statistical differences were found between species and origin regarding seed germination; however, when speed germination was compared throughout time, statistical differences were recorded between each of the species studied. *A. cruentus* (P>0.00001) and *A. hypochondriacus* (P=0.0394) recorded the highest germination at 24 and 48 h. For its part, *A. hybridus* (P=0.0012) had the highest germination at 48 h. Finally, *A. caudatus* was the only species that did not record statistical differences regarding germination throughout time.

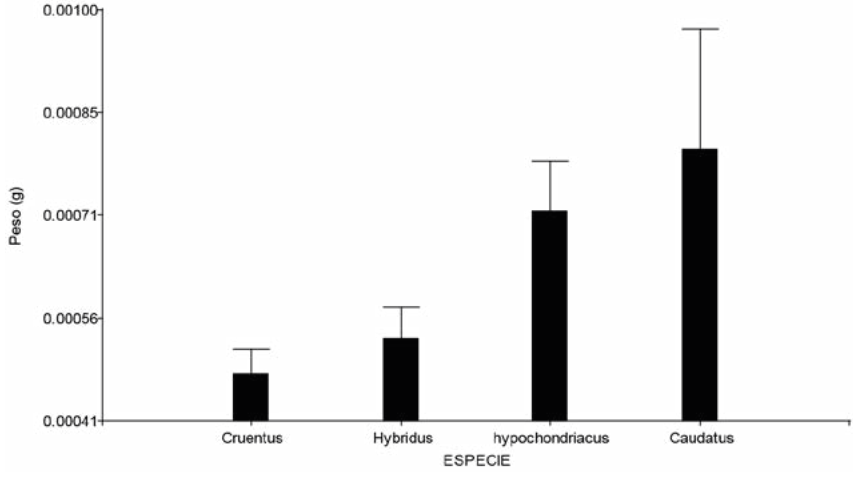

Figure 3. Average seed weight of four *Amaranthus* spp.

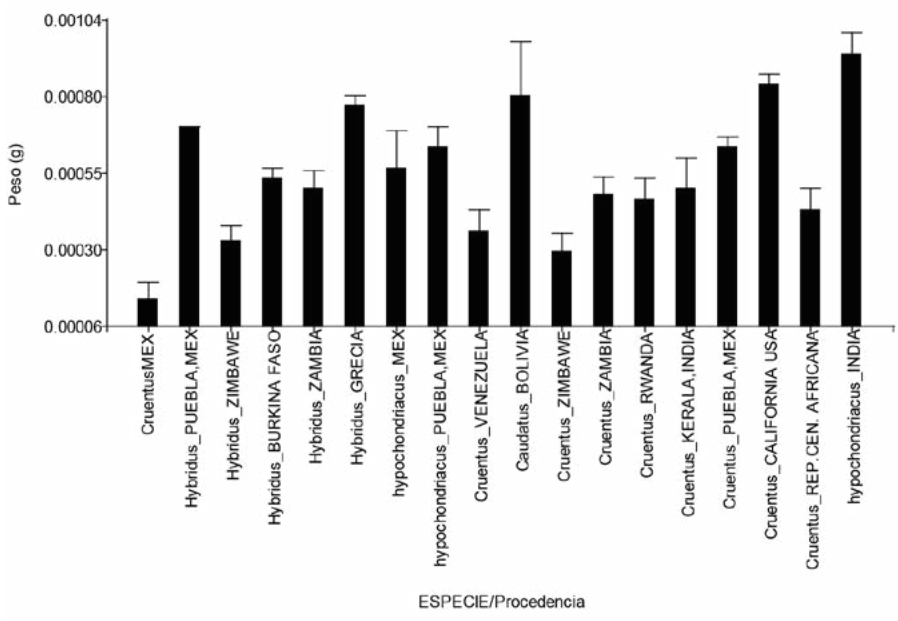

Figure 4. Seed weight of four *Amaranthus* spp. from several origins.

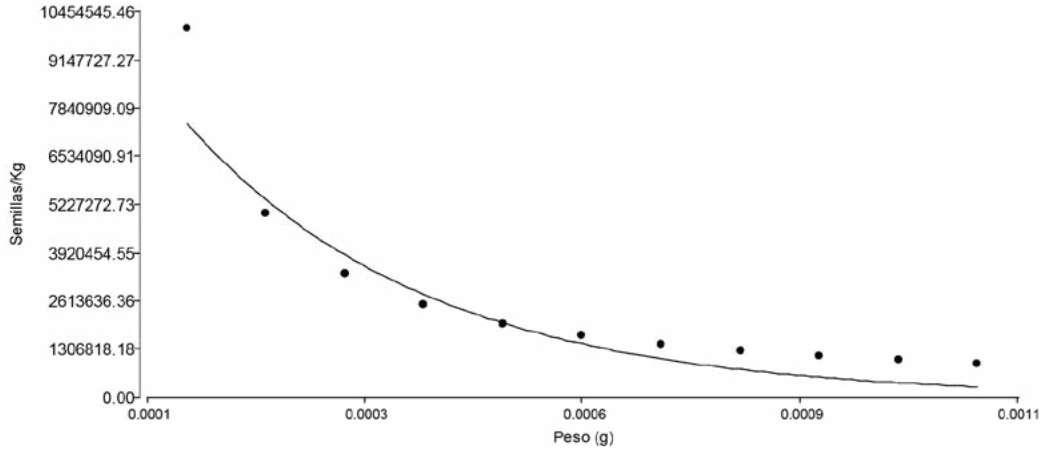

Figure 5. Estimation of the number of seeds per kilogram.

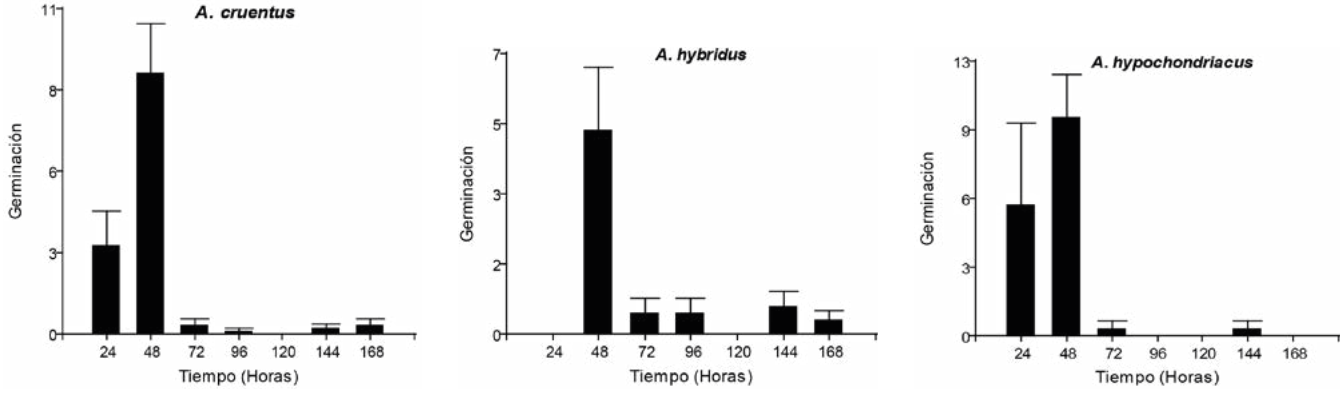

Figure 6. Germination behaviour of the seeds of various species of the *Amaranthus* genus throughout time (after sowing).

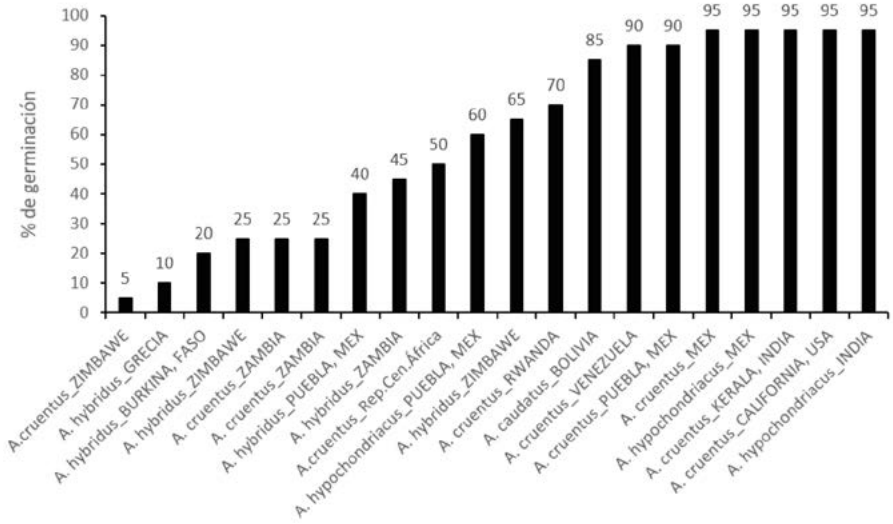

Figure 7. Germination percentage of *Amaranthus* spp. seeds from various origins.

Meanwhile, the highest germination percentages were obtained by A. hypochondriacus from Mexico and India (95%), as well as by A. cruentus from Mexico, California (USA), and Kerala (India) (95%) (Figure 7).

CONCLUSIONS

Regarding their morphology, *A. hypochondriacus* recorded the longest and heaviest seeds. The humidity and temperature conditions under which the seeds were established did not have any effect on the germination of the various species, only on the germination speed of each species. *A. cruentus*, *A. hybridus*, and *A. hypochondriacus* recorded the highest germination between 24 and 48 h. Meanwhile, *A. caudatus* had the same germination throughout the evaluated period.

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Usage of agricultural inputs in sugarcane producers of the ejido Jaeros, State of Veracruz, Mexico

López-Escudero, Roberto J.[1]; Torres-Tadeo, César, M.[1]; Inurreta-Aguirre, Héctor, D.[2]; Lango-Reynoso, Verónica[1]; López-Romero, Gustavo[1*]

[1] Colegio de Postgraduados Campus Veracruz. Carretera Xalapa-Veracruz km 88.5, Tepetates, Manlio Fabio Altamirano, Veracruz. México. C. P. 91690.
[2] Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias. Km. 4.5 Carretera Toluca-Zitácuaro, Vialidad Adolfo López Mateos, Zinacantepec. Estado de México. C. P. 21350.
* Correspondence: gustavolr@colpos.mx

ABSTRACT

Objective: in this research, we assessed the level of organic inputs usage in contrast to chemical inputs in sugarcane producers at the ejido Jareros, Veracruz, in the municipality of Ursulo Galvan.

Design/methodology/approach: Through surveys, a descriptive statistical analysis was conducted on the main socioeconomic aspects of the sugarcane producers in the ejido and an analysis of the means significance of the level of usage of agricultural inputs (chemical/organic) divided into five variables: pesticides, fungicides, acaricides, fertilizers and weed control.

Results: The significance value of the five variables analyzed was: pesticides (p=0.1774); fungicides (P=0.2090); acaricides (0.3625); fertilizers (P=0.0005) and weed control (P=1.0000).

Findings/conclusions: Based on the significance values and mean difference of the five assessed variables, it is concluded that the excessive use of nitrogenous fertilizers is the factor with the greatest potential to negatively impact the environmental and edaphic deterioration of arable soils in the Jareros ejido, State of Veracruz, Mexico.

Keywords: sugar cane, chemical inputs, organic inputs, analysis.

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INTRODUCTION

Worldwide, the production of sugarcane (*Saccharum* spp.) is of great importance. It is cultivated in more than 130 countries on a 25.4 million hectares area, with an average yield of 80 t/ha (FAOSTAT, 2013). It is the ninth in importance regarding the value of production (56 billion dollars), but the first crop by the quantity of produced raw material (1800 million tons per year) (FAOSTAT, 2013).

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Water and soil pollution, soil erosion, and climate change are phenomena related to the agricultural practices that exist in Mexican agroecosystems, including those applied in sugarcane production. Although in Mexico, the sugar industry is one of the most important, there is evidence of the negative impact that sugarcane production causes on the geophysical environment of the soils due to the excessive use of agrochemicals of synthetic origin (Saavedra and Vargas, 2000). Likewise, Rodríguez and collaborators (2007) noted that the soil impoverishment and the annual decrease in the sugarcane agricultural yield relate to nutrient extraction and export, and the continuous loss of organic matter (Rodríguez *et al.*, 2007).

According to Bravo (2015), contamination due to agrochemicals, pesticides, and synthetic inputs in sugarcane areas not only disturbs the environment but also affects human health, causing poisoning by absorption of chemical agents during cultivation, such as insecticides and other agrochemicals application.

Research with long-term experiments in the experimental network of the National Sugarcane Research Institute (INICA) demonstrated that the monoculture of sugarcane contributes to the gradual degradation of soils, with a marked decrease of organic matter content due to inadequate management methods, progressively accentuating when the layer of crop residues is eliminated by burning or other cultural practices, including excessive agrochemicals usage (Pablos *et al.*, 2007).

Cabrera and Zuaznábar (2010) state that sugarcane favors the loss of organic carbon in soils due to the excessive use of nitrogenous fertilizers, the extraction generated by obtaining the stem of the plant, and the burning of harvest residues carried out when the economically important product is cut for harvest.

Jareros is a marginalized rural community from the Úrsulo Galván municipality, it has an ejido of sugarcane producers made up of cooperative members registered in the La Gloria and El Modelo sugar mills, which monopolize the ejido's production. However, due to its location, there is no diagnosis of the phenomenon.

Based on the above and considering the subject's relevance, the objective of this research is to generate a quantitative analysis to compare the degree of organic inputs usage in contrast to chemical inputs, in sugarcane producers from the ejido Jareros, State of Veracruz.

MATERIALS AND METHODS

Study area

The ejido Jareros locates in the Úrsulo Galván municipality, in the State of Veracruz, Mexico. It is geographically located between the coordinates, −96.474722 Longitude and 19.443333 Latitude, and has 516 inhabitants. It has an average maximum temperature of 27 °C and a minimum of 22 °C and an annual 250 millimeters average rainfall. Its main crops are malanga, corn, and sugar cane (INEGI, 2021).

Selection of the study sample

Considering that there is a census of sugarcane ejidatarios that make up the Jareros ejido, the following statistical equation was implemented to determine the sample size (n=17) of a fixed population character (Table 1).

Table 1. Equation to determine the sample size in fixed populations.

$$n = N * Z^{2}_{\alpha} * p * q / e^{2} * (N-1) + Z^{2}_{\alpha} * p * q$$

$$n = 20 * (1.96)^{2}_{\alpha} * (0.5) * (0.5) / (9.44)^{2}_{\alpha} * (20-1) + (1.96)^{2}_{\alpha} * (0.5) * (0.5)$$

$$n = \mathbf{17}$$

n: sample size, *N*: size of the population or universe, *Z*: statistical parameter depending on the level of confidence, *e*: maximum accepted estimation error, *p*: probability of the studied event occurring, *q*: $(1-p)$ probability that the studied event will not occur (Martínez, 2012; Pérez, 2005; Scheaffer *et al*., 2007).

Survey implementation

Once the value of "n" was generated, the survey was built considering 17 variables to identify socioeconomic aspects (Table 2) and management of agricultural inputs (Table 3), where the Likert scale (Guil, 2006) was used to generate an analysis. Comparison between the agronomic management of a chemical and organic nature that is given to the cultivation of sugar cane. 17 questionnaires were applied during XXXX.

Results analysis

To accept or reject the null hypothesis (Ho) of the study variables, the information obtained was statistically analyzed using the R studio® Software (R core team, 2011),

Table 2. Socioeconomic variables analyzed among the sugarcane producers from the Jareros ejido, State of Veracruz, México.

Variable	Variable type	Indicator and measurement scale
Gender	Nominal Qualitative	Male Female
Age	Discrete quantitative	Numeric
Education level	Discrete quantitative	Years of study
Number of household members	Discrete quantitative	Number of Members
Sugarcane planted Area	Discrete quantitative	Number of hectares

Table 3. Analysis variables on the use of agricultural inputs in sugarcane producers from the Ejido Jareros, State of Veracruz, México.

Variable	Variable type	Indicator and measurement scale
Implementation of insecticides (Organic - Chemical)	Quantitative discrete	Organic: 1 - 5* Chemical: 1 - 5*
Implementation of fungicides (Organic - Chemical)	Quantitative discrete	Organic: 1 - 5* Chemical: 1 - 5*
Implementation of acaricides (Organic - Chemical)	Quantitative discrete	Organic: 1 - 5* Chemical: 1 - 5*
Implementation of fertilizers (Organic - Chemical)	Quantitative discrete	Organic: 1 - 5* Chemical: 1 - 5*
Weed control (Organic or cultural – Chemical)	Quantitative discrete	Organic: 1 - 5* Chemical: 1 - 5*

*: Likert scale 1=not used, 2=used little, 3=used them in the same proportion as organic/chemical inputs, 4 = frequently used, 5 = always used.

applying descriptive statistics to the socioeconomic variables and the Lilliefors test (Wilks, 2011) to the variables obtained by Likert scale, to determine the type of data distribution and choose an appropriate method for comparison of means.

RESULTS AND DISCUSSION

One hundred percent of the ejidatarios of Jareros are male, with an average age of 37 years and 13 years of studies (Figure 1), 4 members families and a 4 (ha^{-1}) sugar cane plantation (Figure 2).

After analyzing the results of the Likert scales of the two factors (Chemical/organic) from the five study variables (Pests; fungi; mites; fertilization; cleaning of weeds), for the analysis of variance, a non-parametric Mann-Whitney U test was implemented (Turcios, 2015), given the abnormal distribution of the data, obtaining values of $P<0.05$.

Figure 3 shows the graphic behavior of the means analyzed under the Mann-Whitney U statistical method (Turcios, 2015), as well as the significance values obtained for the study variables of the two contrasted factors.

According to Figure 3, the variable with the greatest significant difference was that of the use of fertilizers ($P=0.0005$). Based on the above, and considering the agronomic management shown in Table 4, described by the producers; using the SWAT model

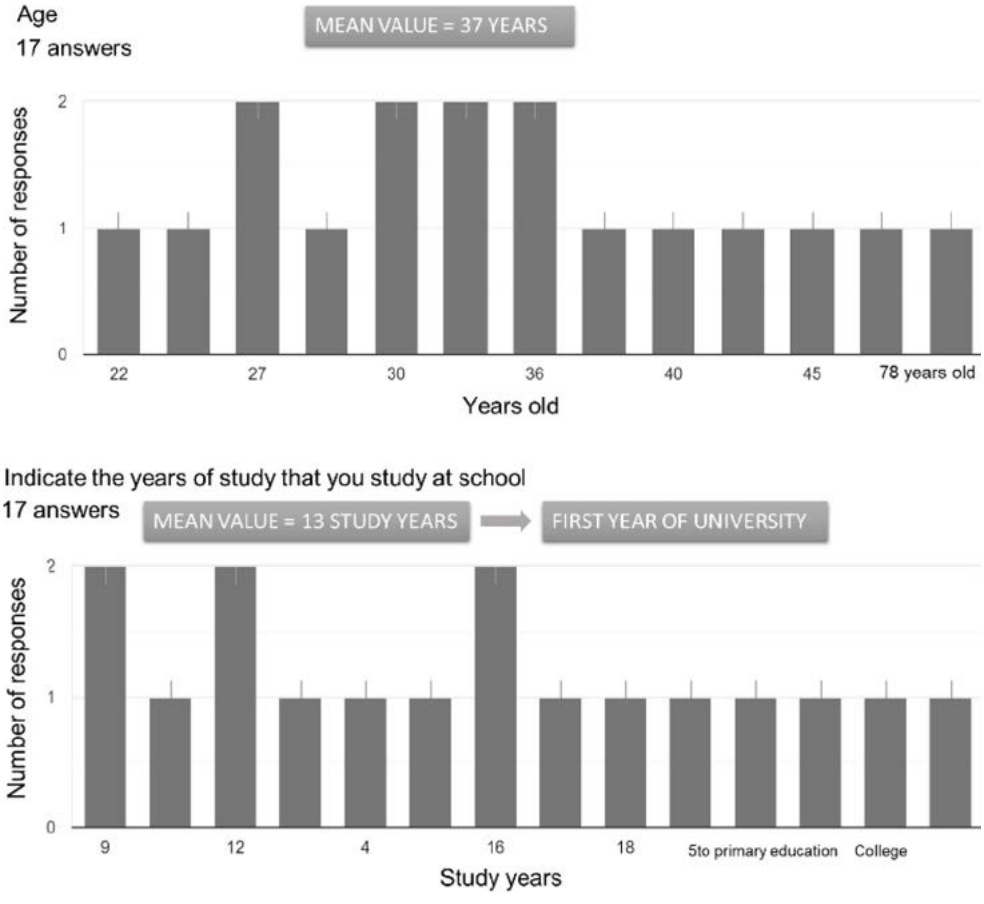

Figure 1. Age and years of studies of sugarcane producers from the ejido Jareros, State of Veracruz, México.

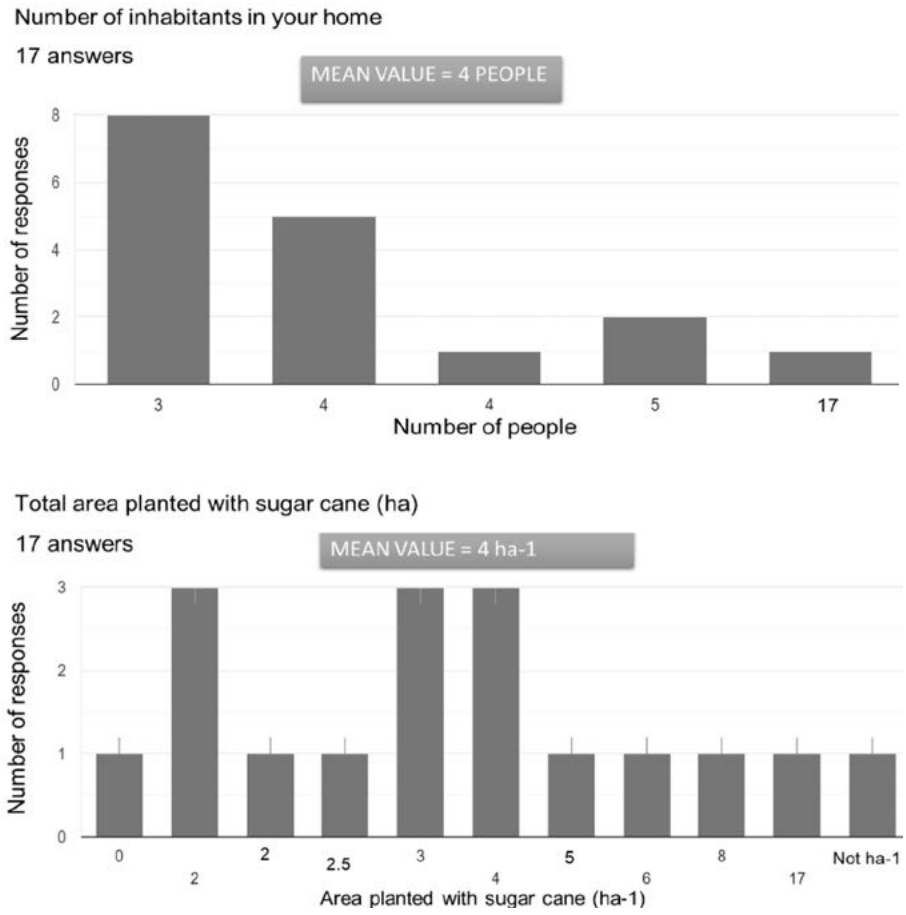

Figure 2. Number of family members and area planted with sugarcane by producers from the ejido Jareros, State of Veracruz, México.

(Arnold *et al.*, 1998; Neitsch *et al.*, 2005; Narasimhan *et al.*, 2005; Garg *et al.*, 2011; Du *et al.*, 2006; Akhavan *et al.*, 2010; Guzmán *et al.*, 2004) and implementing the methodology by Inurreta *et al.* (2013), the potential crop yield in the Úrsulo Galván municipality was simulated, including the area of study under two fertilization scenarios (100% and 70% nitrogen application) during the 2011 - 2020 period and later, through the Dobermann equation (2005), shown in Table 5, the efficiency of the nitrogen fertilization use (PFPN) was also analyzed. The obtained results are shown in Table 6.

Table 4. Sugarcane management introduced to the model under rainfed conditions.

Activity	Year	Operation	Input or activity	Date
Crop establishment	1	Planting	Sugarcane	July 1st
Fertilization	1	1st Fertilization	36-92-00 NPK (kg ha^{-1})	July 30th
	1	2nd Fertilization	218-00-108NPK (kg ha^{-1})	September 30th
Harvest	2	Harvest	Harvest and kill operation	October 30th

NPK: nitrogen, phosphorus, potassium. Source: consensus with ejidatarios from Jareros, State of Veracruz, México.

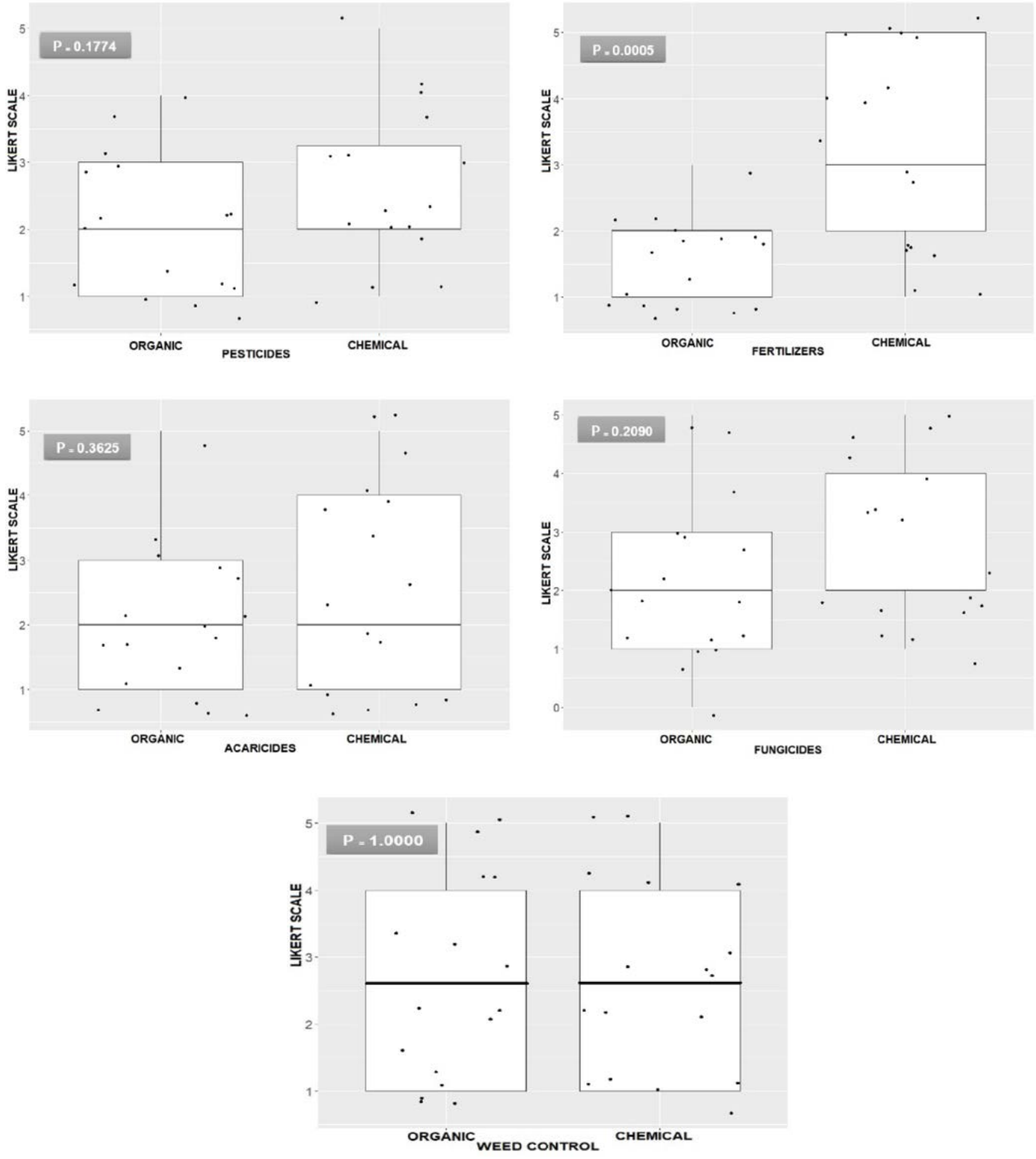

Figure 3. Means analysis of variance from the research developed in the ejido Jareros, State of Veracruz, México.

Table 5. Equation for calculating the nitrogen efficiency.

$PFP_N = Y_N / F_N$
PFP_N = Efficiency in the use of nitrogen; Y_N = Yield of the crop under the application of N (t ha^{-1}); F_N = Amount of N applied (Kg/ha^{-1}) (Doberman, 2005).

Table 6. Nitrogen efficiency in sugarcane under 100% and 70% dose of nitrogen application at ejido Jareros, State of Veracruz, México.

F_N Factor	Mean Y_N	Maximum Y_N	PFP_N Value
Optimum fertilization at 100% (254 kg N/ha^{-1})	42.5 t ha^{-1} of sugar cane	70.2 t ha^{-1} of sugar cane	PFP_N (Mean)=0.17 PFP_N (Maximum)=0.27
Fertilization at 70% (178 kg N/ha^{-1})	40.1 t ha^{-1} of sugar cane	68.1 t ha^{-1} of sugar cane	PFP_N (Mean)=0.22 PFP_N (Maximun)=0.38

PFP_N = Efficiency in the use of nitrogen; Y_N = Yield of the crop under the application of N (t ha^{-1}); F_N = Amount of N applied (kg ha^{-1}) (Doberman, 2005).

Table 6 shows that even by decreasing the nitrogen fertilization dose to 70% it is possible to obtain mean and maximum crop yield values like those from the 100% fertilization scenario, the application dose of 178 kg N/ being even more efficient. has^{-1}.

CONCLUSIONS

All the interviewed ejidatarios were men, with 13 years of studies and 4 hectares per producer area, and 37 years age average. Regarding the level of chemical and organic inputs usage, a balanced trend of both inputs was shown in terms of their application for pests, fungi, and termites' control and traditional cleaning management. However, regarding crop fertilization, it should be noted that the use of chemical fertilizers is much higher than that of organic fertilizers. Therefore, it is recommended to carry out training campaigns in the ejido Jareros to raise awareness among producers about the use of other complementary technologies to chemical fertilization, to reduce the environmental and agricultural degradation of the sugarcane agroecosystems in the ejido.

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Community perspective of the fishing activity in El Arenal, Acapulco, Mexico

Cardona-Castaño, Juan C.[1*]; Rodríguez-Alviso, Columba[1]; Maganda-Ramírez, María C.[2]; Villerías-Salinas, Salvador[3]; Brito-Carmona, Rosa M.[1]; Aparicio-López, José L.[1]

[1] Universidad Autónoma de Guerrero, Centro de Ciencias de Desarrollo Regional. Municipio de Acapulco, Guerrero, México. C.P. 39640.
[2] Instituto de Ecología, A. C. Municipio de Xalapa-Enríquez, Veracruz, México. C.P. 91073.
[3] Universidad Autónoma de Guerrero, Centro de Investigación y Posgrado en Estudios Socioterritoriales. Municipio de Acapulco, Guerrero, México. C.P. 39390.
* Correspondence: columbaalviso@uagro.mx

ABSTRACT

For many communities settled on the coastline of coastal lagoons, fishing is an important activity because of the income it generates.

Objective: To analyze the community perspective of the fishing activity in the community of El Arenal, in Guerrero, Mexico.

Methodology: The research is a mixed case study and its development consisted in seven stages, which were: description of the area where field immersion was carried out and identification of socioenvironmental problems, dialogues with the fishermen, information processing, data analysis through descriptive statistics, and deductive analysis of the discourse.

Results: Overfishing is evident, and the breach of closed season and high levels of capture in live weight which was 1275 t in 2022. The 37 cooperatives do not have trade channels and brokers are in charge of trading the product. At the socioenvironmental level, the community of El Arenal does not have residue collection programs or for lagoon or mangrove care.

Conclusions: The production model based on overfishing exhausts the natural resource because there is no planning for the fishing resource and it does not respect the closed season law, resulting in socioenvironmental problems for the community, overfishing, contamination of the lagoon, and social marginalization.

Keywords: fishing activity; rural community; socioeconomic structure; Laguna de Tres Palos; socioenvironmental problems.

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INTRODUCTION

The fishing activity is an important economic sector because it is part of food security at the global level and in 2021 there was a production of 188 million tons (FAO, 2022). When it comes to the Mexican context, it occupied the 13th position of countries with most fishing activity and contributed 2% of the world production, which makes it a country with strong economic development in this economic environment (FAO, 2020). The fishing activity in Guerrero, Mexico, is in the third branch of highest development and economic growth; during 2021 there was capture in live weight of 1.8 t, where Acapulco contributed 49.83% (CONAPESCA, 2020). However, this economic activity faced intense difficulties when the

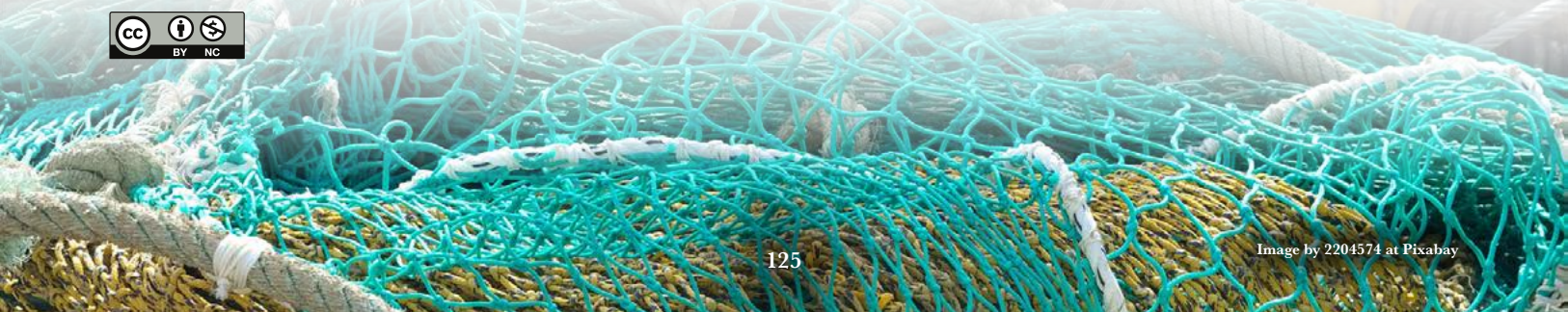

Image by 2204574 at Pixabay

fishing activity decreased considerably, which resulted in economic impacts in the rural sector in national, state and local contexts (Sandoval *et al.*, 2020; Secretaría de Agricultura y Desarrollo Rural, 2021). Fishing is an important source of income for 280 households and 385 fishermen in El Arenal, although the social marginalization rate continues to be high because there are no social programs to help fishermen. The fishing activity has decreased due to overfishing, the breach of closed season laws, the contamination of the ecosystem and deforestation of the mangrove. In addition, there is low economic diversification and null financial strengthening.

The cooperatives do not have trade channels and the appearance of intermediaries ("brokers") has caused inequality in commercialization of the product, which prevents the community from having a sustainable fishing production system. Therefore, government strategies are necessary for the welfare of the community and the fishermen. The government must develop social programs that contribute to the wellbeing of fishermen, and which strengthen and diversify local economy to minimize the inequalities in commercialization of the product and to ensure a sustainable fishing productive system. In addition, it is necessary to control and plan the fishing activity better to avoid the breach of the closed season law and the appropriation of the Laguna de Tres Palos. At the same time, it is important to take steps to reduce overfishing and deforestation of mangroves. These actions are fundamental for the sustainable development of the fishing community of El Arenal.

The perspective of the community of El Arenal through this study prevails because it makes a significant contribution about the fishing activity and shows the socioenvironmental state of an activity of the primary sector, which provides sustenance to rural households far from tourism as a dominant economic matrix in the city of Acapulco. In addition, it documents a community socioenvironmental reality and questions the lack of community and government connections within rural and territorial development of the fishing activity.

Theoretical discussion

Economic structure

The economic structure is the dynamic set of the relationship of productive forces that are empowered to promote development at the social level (Tamames and Rueda, 2014; Ceballos and De anda, 2021). This interaction between society and economy forms new market niches that lead to overconsumption, exhaustion of natural goods, and cause a decline in rural development because production is low scale and originates a deep scarcity of natural resources in rural communities (Gutiérrez and Glückler, 2022; Richmond and Casali, 2022; Zhang *et al.*, 2022). Therefore, in the case of fishing communities, scarcity is an economic problem generated by the production model, the socioenvironmental or extraction problems, and the susceptibility of market dynamics (Debaere & Kapral, 2021; Kuhl, 2021).

Fishing activity

The fishing activity is important because it offered 174 thousand direct jobs in 2021 in the rural population in Mexico and contributed to the decrease in unemployment by 2.7%

(SADER, 2021 A; SADER, 2021 B). It also contributed to the nation's food security and sovereignty and is part of the basic economy of the coastal states in Mexico (CONAPESCA, 2020). According to SADER (2019), in Mexico the total capture of the fishing sector is approximately 2 million tons (T), where a large amount is extracted from lagoons, which have a fundamental role within production and capture since they are strategic and economically productive ecosystems. However, fishing production in Mexico presents overfishing and generates strong socioenvironmental pressures on the coastal lagoons that end up affecting local and rural communities (Cruz *et al.*, 2021; SADER, 2022). In this context, paradoxically, the fishing resource allows the growth of local economies; rural communities have taken advantage of the economic niche to obtain monetary income, recognizing that in some zones next to lagoons, lakes and the sea there is overfishing which reduces the abundance of the resource (García *et al.*, 2021).

During recent years, Mexico has shown a sustained growth in the fishing activity (capture and production); however, during 2017 and 2018 there was a decrease, and even so, fishing contributed 35 billion pesos MX, a historical value (SEDER, 2017). All in all, the fishing sector has experienced a favorable significant growth (Table 1) and it has been consolidated as an important economic branch for rural and economic sustainability of the country, despite the socioenvironmental problems that underlie this economic activity (SEGOB, 2020; Cortés, 2021; SADER, 2022).

According to SEGOB (2022), the state of Guerrero had a fishing capture of 1.7 million tons in 2021. This figure marked the trend of a rising primary sector, which drives the local development of the communities that have fishing as the basis of their economy. Based on this, the authors Villerías *et al.* (2016), Herrera *et al.* (2016), and Villerías (2021) explain that fishing is an economic sector driven by tourism, which guarantees rural development. However, the touristic economic spill has led to socioeconomic vulnerability due to inequality in commercialization of the product and, consequently, low income. Therefore, the activity does not ensure the social wellbeing of the fishermen in addition to the socioenvironmental damage to fishing ecosystems with economic value (Uc, 2018; Villería *et al.*, 2017).

In accordance with this, the study analyzes the community perspective of the fishing activity in the community of El Arenal.

MATERIALS AND METHODS

The case study

This research is framed by a mixed approach, which allows an integral analysis of qualitative and quantitative data and approaching the phenomenon's complexity. The decision was made for a case study design that leads to an in-depth understanding of a

Table 1. Growth of the fishing activity (2013-2022).

Year	2013	2014	2015	2016	2017	2018	2019	2020	2021	2022
Growth (%)	4.50%	21.40%	17.90%	1.90%	−4.90%	−5.00%	5.40%	1.60%	7.50%	15%

Source: Prepared by the authors with data from SEGOB (2020), Cortés (2021), and SADER (2022).

problem or phenomenon and involves epistemological amplification (Buttazzoni, 2022). The research had seven phases: a) description of the study area; b) field immersion; c) review of institutional databases and geo-portals; d) visits and visual inspection; e) informal dialogue; f) information processing; and g) information analysis; and it is a case study located in the rural sector of the city of Acapulco. The community of El Arenal was selected because of the research precedents achieved by Juárez *et al.* (2019), which make a prevailing recommendation to research socioenvironmental problems within the fishing activity as an economic axis of the communities adjacent to Laguna de Tres Palos.

Description of the study area

El Arenal is part of the rural zone of the city of Acapulco (16° 48' 43.85" N and 99° 41' 47.50" W) and it is an annex of the community of Cacahuatepec (Figure 1). At the population level, the community of El Arenal (Acapulco, Guerrero) appears for the first time in the census of 1960 performed by the National Statistics Direction with 76 inhabitants (INEGI, 2020), recorded as a ranchería; the growth of the community has

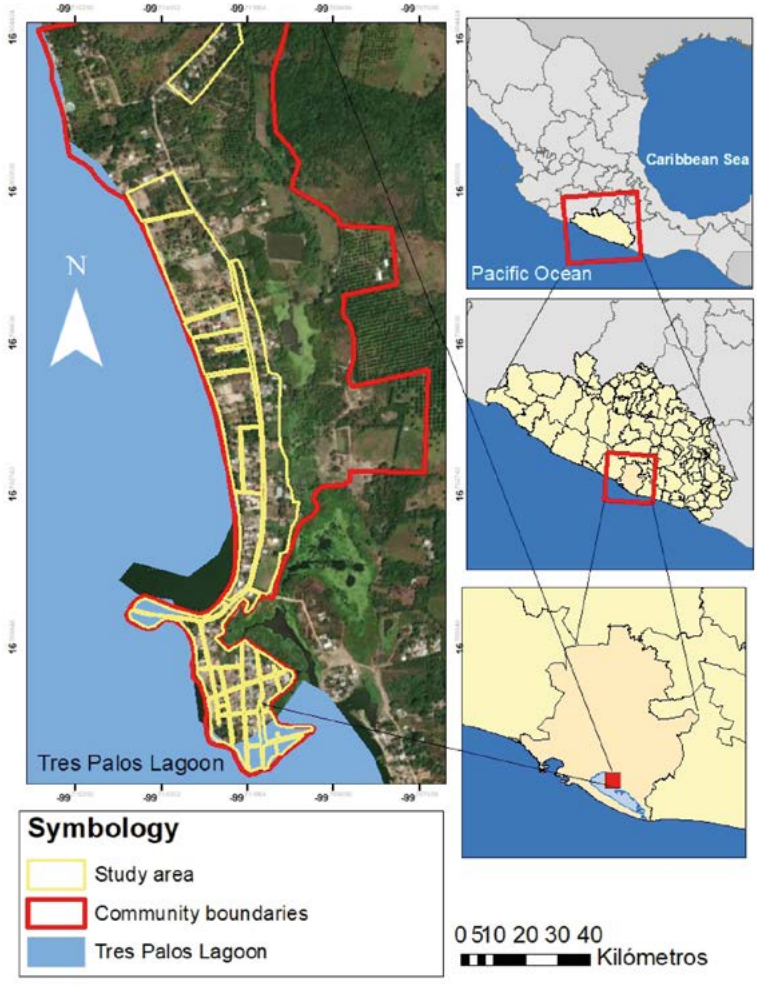

Figure 1. Study area, community of El Arenal, Guerrero, Mexico.
Source: Prepared by the authors with data from INEGI and Google Earth.

been exponential and currently there are 1037 inhabitants with a population growth rate of 92% and a base of young population between 10 and 40 years old, which indicates a significant productive strength (INEGI, 2020).

Regarding the biophysical system, the study area is found in the sub-basin of the La Sabana River and Laguna de Tres Palos; it has an average temperature of 27.4 °C (Celsius) and an average annual rainfall of 107.4 millimeters (mm) with a rainfall system divided into two seasons (Google Earth, 2022), which is important for the reproductive cycles of fish and the tasks for fishermen; it is an important ecosystem for territorial development because of the offer of ecosystem services and its key use, exploitation and conservation for the economic and territorial development of the communities that are part of the sub-basin. Indirectly, El Arenal is in continuous relationship with the La Sabana River and directly with Laguna de Tres Palos, and the latter ecosystem provides the raw material (fish) that is the economic basis of the place.

Stages in the research

The research was divided into seven stages that range from the compilation of information to obtaining and analyzing empirical data, which allowed responding to the objective of the study and satisfying the thematic interest about the socioenvironmental state of the fishing activity in the community of El Arenal.

A) Description of the study area: it consisted in the review of authors (Juárez *et al.*, 2019; Galán *et al.*, 2020; Casarrubias, 2021) who have studied the subsequent zones to Laguna de Tres Palos or the communities adjacent to the Laguna de Tres Palos, including El Arenal. The review managed to evidence the need to research the community of El Arenal.

B) The field immersion consisted in four visits to the study area to carry out the prior recognition to build a communication bridge between the local stakeholders (fishermen) and the researchers. This resulted in five visits to the study area to begin the dialogue with these local stakeholders with the aim of obtaining permission to work with the community.

C) The review of institutional databases and geo-portals, was important because some socioenvironmental and economic problems were detected that currently persist in the community and which affect the territory and the Laguna de Tres Palos. Information about the fishing production, felling of the mangrove and economic problems of the community were obtained within the analysis of geo-portals; this information was contrasted with the field immersion. However, four in-person visits to government institutions were necessary to request information, which were to the following: National Statistics and Geography Institute (*Instituto Nacional de Estadística y Geografía*, INGEI); National Commission for Aquaculture and Fishing (*Comisión Nacional de Acuicultura y Pesca*, CONAPESCA), and National Council for Evaluation of the Social Development Policy (*Consejo Nacional de Evaluación de la Política de Desarrollo Social*, CONEVAL).

D) The visits and visual inspection of the places where socioenvironmental problems are manifested were the fourth step of this research. This consisted in various field visits with the fishermen of the community, as well as a guided visit to the Laguna de Tres Palos with a local stakeholder. The visual inspection was key in the detection of socioenvironmental problems and the knowledge of species with economic value, the fishing techniques and arts, as well as the commercialization of the product inside and outside the community.

E) Dialogues with the fishermen in the community: the amplification and the knowledge of the fishing economic dynamics were clarified through the knowledge from local stakeholders. At this time of the study, the institutional information was contrasted.

F) Information processing: the quantitative information was analyzed through the Excel software of the Microsoft Office package with descriptive statistics, and the qualitative data with deductive categories.

G) Information analysis: the information was analyzed through Excel and Atlas-Ti of the socioenvironmental and economic aspects of the fishing activity: the quantitative analysis was done with independent variables and the qualitative data through discourse analysis.

RESULTS AND DISCUSSION

Approaching the local stakeholders

Once the study area was described, approaching the fishermen began in order to understand the socioeconomic dynamics through a structured interview, although it was not enough for the quandary, the commercialization of the production, and the analysis of economic units in the community of study (cooperatives). The local stakeholders considered the following about the fishing economy:

Local stakeholder 1:

"The economy of the community of El Arenal is dependent on fishing and the environmental problems have made us vulnerable because of this, since there is almost no more fish, and when we fish, we cannot eat and there is no income to sustain the households; it is very complicated to have a job without guarantees and the backing programs do not reach this community to face the scarcity".

One of the worries that emerged from the dialogues with the stakeholders is the dependency on fishing and the socioenvironmental inequality as basic resource of the local economy and the economic dependency on a natural resource. This economic circumstance is the result of rural productive systems with low diversification and little attention from the institutions; the participants of this research feel economic vulnerability at the local level because the environmental factors of the lagoon sometimes decrease the production of fish and this affects the monetary resource of the fishermen.

On the other hand, the stakeholder describes recognition of the vulnerability that emerges from the absence of the productive model which sustains the community, leading to suggest the following: socioenvironmental inequalities due to the shortage and lack

of recognition of the role of "Fisherman", lack of strength of fishing cooperatives, and little institutional help which creates deep inequalities in the community with the city of Acapulco (center).

Local stakeholder 2:

"The fishing production decreases when the bar between the lagoon and the sea is closed, since water from the sea that could contribute nutrients to the lagoon does not enter; the drought makes the body of water decrease and young fish are lost because oxygen in the water drops, and in sum, we are left without products to sell and eat".

Therefore, a mono-productive economy through fishing dominates in the community, which represents a monetary value and is not only a natural resource that also has an ecological value, but rather a relational value (fisherman-lagoon) that transcends a meaning of welfare and the possibility of satisfaction and sustenance. The community has moved away from other modes of production, they have forged a productive system and human capital for fishing, and thus the loss of the tasks means moving away from welfare.

Socioenvironmental situation

Regarding the social situation of the community, local stakeholder 3 suggested the following:

"We feel that Acapulco has forgotten us, the social problems of the city's growth reach us daily in the form of trash. The social problem lies in that we feel excluded from the opportunities of Acapulco's tourism model, there is a lot of poverty in the community and a lack of opportunities; when we don't fish, we don't have anything to take home and the local government does not support us. On the other hand, there is the garbage issue, because socially Acapulco does not have much environmental awareness, and this is reflected every day in the residues that reach the beaches on the lagoon and inside the mangrove forest. The mangrove is vital because that is where important species that we fish have a hiding place when we don't fish them; for example, with popoyote we have less income and this makes the problem of residues not only an environmental issue but also a social one because the community does not have residue collection by the local government; the river brings the trash from the high part and the government does not clean it".

Likewise, the social situation together with the institutional neglect has derived into a community with a high degree of marginalization (High DM) (Figure 2), and this leads to the assertion that the "institutional neglect", the scarce management of the municipal, state and federal governments, and the low social cohesion of fishermen are part of the socioenvironmental quandary that still has not been solved. This is how the fishermen explain it:

"Institutional neglect, Acapulco hoards the public services and does not answer for the welfare of rural communities that are part of a productive system not included in the city's development".

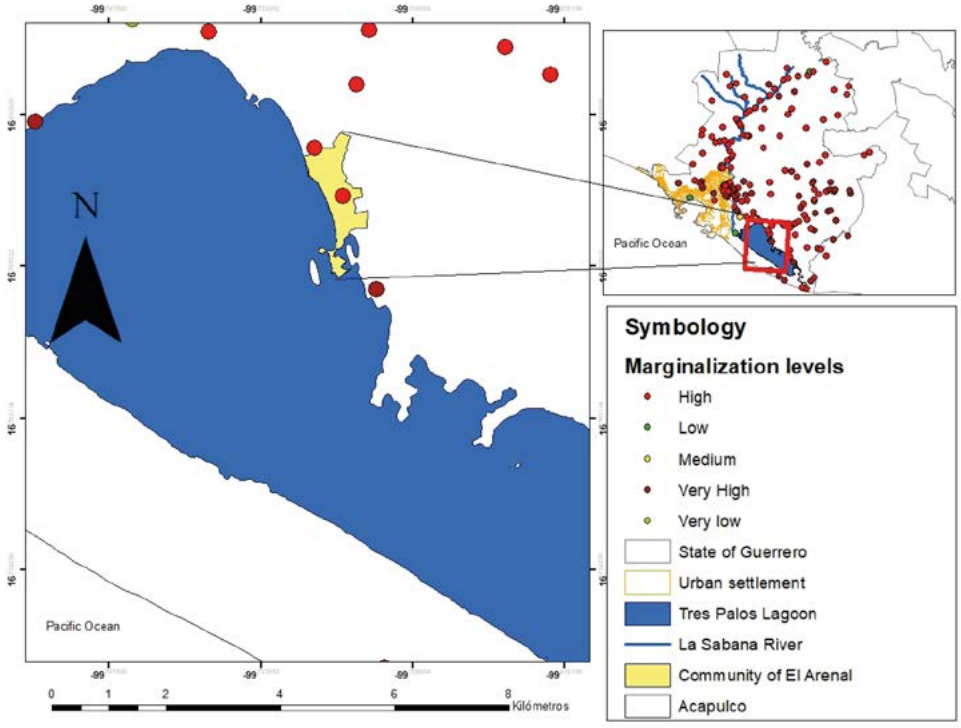

Figure 2. Marginalization Index.
Source: Prepared by the authors based on data from the National Council of Social Development Policy Evaluation (*Consejo Nacional de Evaluación de la Política de Desarrollo Social*, CONEVAL).

Therefore, there is evidence of a structural social problem in the DM of the municipality of Acapulco de Juárez; the analysis centers its attention in the communities adjacent to the Laguna de Tres Palos, and it detects that all the communities settled in that area have high marginalization indices. Broadening the spectrum of marginalization, the rural zone of the municipality presents the same characteristic, and it also explains how the economic structure of the communities that depend on the lagoon, even El Arenal, are far from the economic motor of tourism in the city; therefore, they have wagered on fishing and extraction activities for sustenance.

Socioeconomic problems of the community of El Arenal

The abundance of the fishing resource is associated to two factors, mainly the rainy season and the opening of the bar that separates the lagoon from the Pacific Ocean. The first causes an exchange of nutrients with the ocean that stabilizes the salinity of the lagoon and the second removes the nutrients from the bottom and scarcity of oxygen. If these main factors do not happen, they cause an economic problem induced by environmental causes of which there is no control. However, there are variables that constitute an economic problem from fishing and are manageable: contamination of the lagoon by the La Sabana River and overexploitation of the fishing resource.

The problem of fishing in the community of El Arenal lies in overexploitation, breach of closed season, and a rural economy of low development and diversification, as well as

prioritization of the exploitation of an unrenewable resource, exhausting it and leading to a local economic crisis, which explains why the community of El Arenal has deep socioenvironmental problems that are evidenced by inequality and marginalization with a young workforce.

Economic activity

The primary sector is the main sustenance of the community of El Arenal, with a productive system of the fishing exploitation of Laguna de Tres Palos. According to one of the founders of the community, fishing has been the daily productive activity of communities adjacent to the lagoon and it has developed since the decade of the 1960s, when there was a significant economic potential, and the state of Guerrero was supplied from there. However, the appearance of exogenous factors influenced the exploitation of fishing and defined the productive system of artisanal type due to the following causes: the price determined by the market and the contamination of the lagoon. Also, although this phenomenon would not be spoken about until years later, the real estate growth of the city of Acapulco between the 1980s and the 2000s, the tourism economy, the opening of the bar that separates the lagoon from the sea, and the insecurity for fishermen.

This mix of socioenvironmental problems detonated a strong crisis at the economic level in the community from the 2000s until today; the price of production was regulated by the market of offer and demand of the fishing resource and by the high levels of competition; in some moments it made the price of the fish decrease and profits were not generated. On the other hand, it should be clear that the contamination of the Laguna de Tres Palos is consequence of the real estate development of Acapulco, from the La Sabana River, which discharges domestic and industrial residual waters and causes the loss of species with economic importance for the fishermen (Melgar *et al.*, 2021).

The urban growth of the city of Acapulco is a factor that aggravated the stability of many ecosystems like the lagoon systems, the pelagic and inter-oceanic zones and even the Laguna de Tres Palos, and it is well-known that the La Sabana River nourishes the lagoon; it also discharges contaminated water and solid residues that affect the ecosystem, all product of the urbanization that developed in the high zone of the sub-basin and the growth of irregular settlements, which do not have drinking water and a drainage system, whose option is the direct discharge into the river.

The economic tourism model of the city of Acapulco did not integrate the rural zones; instead, it generated an imbalance in the socioeconomic matrix that made a "center" (city) and a "periphery" (rural) emerge. The first responds to a capitalist model and the second to a primary model where fishing and agriculture are part of the livelihood system. On the other hand, the opening of the bar is important because it favors the entry of fish and crustacean species, and the input of marine species that are exploited by the fishermen; however, the bar that separates the ecosystem from the sea many times does not open naturally and there are also political and economic interests, since opening it would imply a mobilization of the municipality's financial and technological resources and many times the local government is not willing to commit to expenses. Lastly, insecurity started in 2010, power groups stirred the territory and

the community of El Arenal was affected, since it had a local economy that was based on the service sector, fishing and agriculture before, which was reduced and created dependence exclusively on the fishing sector.

Commercialization of fish

Fishing production is distributed through dealers called "brokers" which are people who have monetary, logistic and commercial resources to purchase and resell the raw material in the local, state and even national markets. They act independently as a bridge between the fisherman and the market system, substituting the function of fishing cooperatives which, in theory, should buy the production from the fishermen. The brokers become important figures within the fishing activity because they guarantee commercial dynamism at the local level, and they are included in the productive chain. The fishermen have a good opinion of intermediaries, as explained by the Key Stakeholder 5:

> *"The brokers are important because the cooperatives do not buy the raw material; here in the community we do not have income to trade fish with our own means at the state level or throughout Mexico, so we need them, since they guarantee the exit of the fish".*

Regarding the sale price of the fish, the free market determines the price; however, the "brokers" have the power to impose quite reduced prices, placing the fishermen at a disadvantage, since they obtain low profits from their work. On the other hand, the broker resells the raw material in the local and national markets at a much higher price than they acquired.

Factors of production (lagoon territory)

Laguna de Tres Palos is a means of production for the community of El Arenal, where the fish is extracted for commercial profit. This common economic zone of approximately 57 square kilometers has led to territorialization of the lagoon, with communities that establish social, political and productive links with the consumption and exploitation of the fishing resource. It has generated a social value of the ecosystem. These fishing areas are identified by knowledge of the fishermen, although other communities can also extract the fish. This equality and mobility make the lagoon a common good for everyone, despite the fixed zones where the communities acquire the resource. The link between these communities is manifested through economic and cultural means. The Laguna de Tres Palos is an economic zone of great importance for the community of El Arenal, since it provides resources such as fish for consumption and commercialization. The various communities have established economic, social and cultural relationships based on the use and exploitation of these resources. This has generated a social value of the ecosystem, in addition to ensuring that all the members of the community have equality or access to the resource. Although there are fixed zones for fishing where members of the community extract the fish, the lagoon is a common good for everyone. These relationships between the communities have been established based on the mobility of the resources, ensuring for all to have equal access.

Based on this, the territorialization and the appropriation by communities adjacent to the Laguna de Tres Palos became a central axis of the local economy. In this sense, a means of production was developed and specialized human capital (fishermen) emerged for the exploitation and extraction of fish offered by the ecosystem. This relationship between human capital and fish extraction resulted in a subsistence model at the community level. The fish has become the main input and the motor of the economy because it materialized in monetary income for the households that live at the expense of the lagoon.

The visits to the study area allowed the identification of a considerable productive force; the fishermen practiced their trade and took advantage of the fishing resource from the lagoon with its fishing arts (*atarraya*, *trasmayo* and string), and the points for fishing appropriation where the fish is extracted were revealed (Figure 3). In dialogue with the Local Stakeholder 1, he described that: *"We know that the lagoon belongs to everyone, but each community has its fishing points; I have been making this journey since I was a child, and we find a lot of fish in these points".*

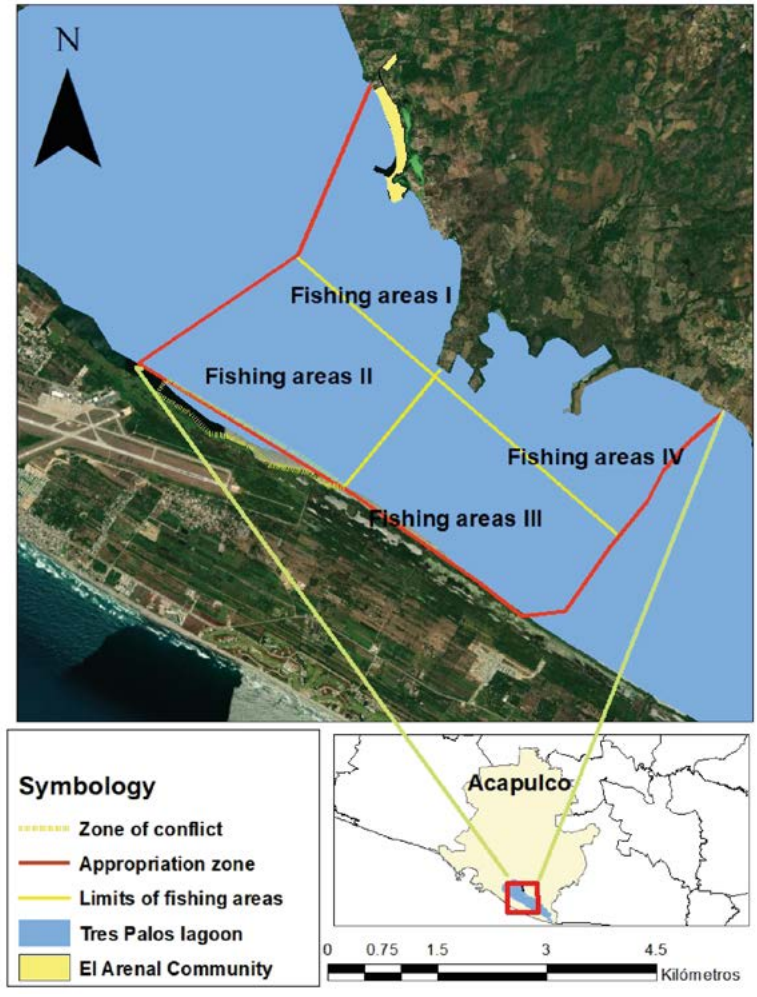

Figure 3. Appropriation of the Laguna de Tres Palos and strategic zones for fishing.
Source: Prepared by the authors with data obtained from the visit by boat at Laguna de Tres Palos with the fishermen.

Fishing cooperatives

In the community of El Arenal there are 37 fishing cooperatives and they group 385 fishermen, which in theory were created to purchase the production from fishermen and to carry out the productive process within the market for consumption (Figure 4). On the other hand, the function of the cooperatives changed, and now they are devoted to managing monetary resources from the state to strengthen and support fishermen. According to Hernández *et al.* (2021), the cooperatives have a social and organizational function: the dynamization of local productive processes. However, in the community of El Arenal there was a change in the social function of the cooperatives, which did not solve the socioeconomic needs and the socioenvironmental problems —such as those from the community—, and in fact there was no evidence of consolidated community cooperative movement in matters of decision making and management of fishing problems, which resulted in a disorganization of the productive model that would solve many of the problems already described, which are disadvantages in face of other communities that have secured the cooperative movement as a mechanism for organization and strengthening.

Regarding the capture in live weight and the commercialization value, it was analyzed during the period of 2017-2022; there was a growth rate of capture of 13.4% and a variation of average price of 21.2% (Tables 2, 3 and Figure 5). The production has been limited and has been impacted for some years by socioenvironmental problems, for example:

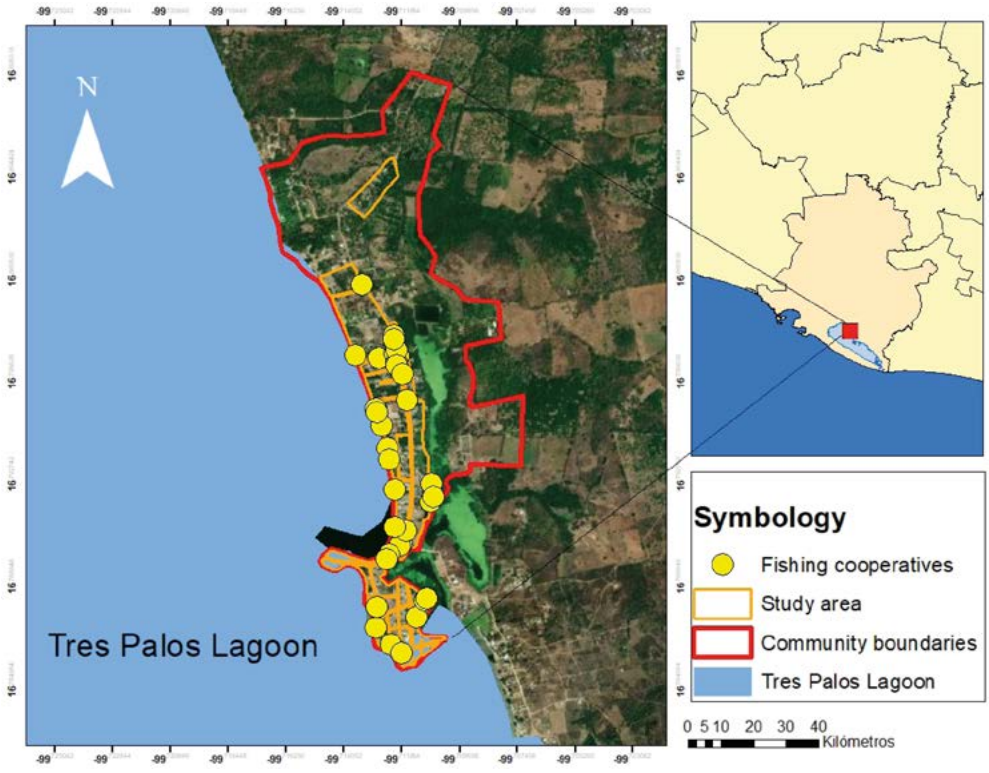

Figure 4. Fishing cooperatives of the community of El Arenal.
Source: Prepared by the authors with data from the Statistical Directory of Economic Units (*Directorio Estadístico de Unidades Económicas*, DENUE).

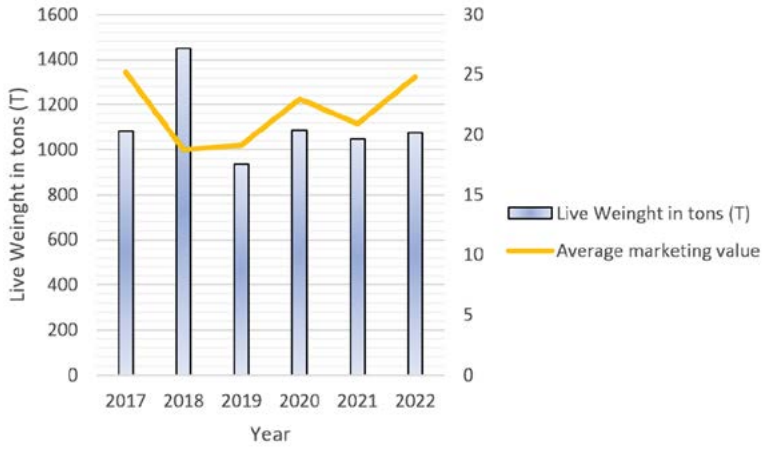

Figure 5. Live weight capture and commercialization price (2017-2022).
Source: Prepared by the authors with data from CONAPESCA.

opening of the bar that separates the lagoon from the sea, contamination of residual waters of domestic and industrial origin that are dragged by the La Sabana River, cutting the mangrove, and intensive fishing.

The fishing resource is pressured because of overfishing by all the communities adjacent to the Laguna de Tres Palos; if this rhythm continues, a future exhaustion of the resource is possible, and approximately 10 rural communities live from the fishing activity offered by the Laguna de Tres Palos. This entails an impact on the environmental capital and the ecosystem services, the lack of regulation and control of overfishing would lead to profound changes in the ecosystem and by default in the human settlements that have fishing as a basis of economy.

Table 2. Species with economic value for fishing.

Common name	Scientific name
Bagre	*Ictalurus punctatus*
Camarón blanco	*Litopenaeus vannamei*
Chacal	*Macrobrachium tenellum*
Charal	*Lile stolifera*
Cuatete	*Ariopsis guatemalensis*
Huarache	*Paralichthys woolmani*
Langosta verde	*Panulirus gracilis*
Langostino de agua dulce	*Macrobrachium rosenbergii*
Lisa	*Mugil curema*
Pargo	*Lutjanus guttatus*
Popoyote	*Dormitator latifrons*
Robalo	*Centropomus nigrescens*
Tilapia	*Oreochromis niloticus*

Source: Prepared by the authors with data from CONAPESCA.

Table 3. Capture of species of economic importance in live weight and commercialization value (2017-2022).

Year	2017		2018		2019		2020		2021		2022	
Species	**LW (T)**	**ASP *Kg**	**LW**	**ASP *Kg**	**LW**	**ASP *Kg**	**LW**	**ASP *Kg**	**LW**	**ASP *Kg**	**LW**	**ASP *Kg**
Bagre	0	0	0	0	12.7	19.8	3.3	20.4	0	0	9.9	20
Bandera	0	0	306.3	16	277.1	16.7	102	18.6	96.8	20.2	89.2	20.4
Carpa	3.8	40	0.63	14	1.7	12	0	0	3.3	25	3	25
Chacal	12.5	43.5	0	0	0	0	0	0	0	0	0	0
Charal	2.5	37	19.9	12	26.3	10.5	278.5	14.2	675.4	15	696.7	13.4
Charra	5.9	15	8.1	13	0	0	400	15	0	0	0	0
Cuatete	276.4	15	0	0	0	0	0	0	0	0	0	0
Huarache	0	0	0	0	0.85	15	0	0	0	0	0	0
Langosta	0	0	0	0	0	0	0.08	36	0	0	0	0
Langostino	2.3	47.2	22.1	45	73.1	42.5	38.5	40	56.4	33.7	56.9	32.4
Lisa	325.7	14	477.8	14	233.7	16.4	118.7	15.5	93.7	17	93.9	17.3
Mojarra	0	0	311.3	21	309.4	24.3	116.7	25.5	99.1	24.2	106.1	24.5
Pargo	0	0	0	0	0	0	0	0	0	0	1.1	20
Pijolin	0.155	15	0	0	0	0	0	0	0	0	0	0
Popoyote	197	14	304.4	15	0	0	28.5	12	23.3	12.1	17.9	12.4
Tijereta	0	0	0	0	0	0	0.215	25.3	0	0	0.36	27.5
Tilapia	255.8	17	0	0	0	0	0	0	0	0	0	0
Robalo	0.068	19	0	0	1.7	15	0	0	1	20	0.44	60

Source: Prepared with data from CONAPESCA. (LW = Live Weight; ASP = Average Sale Price; kg (kilograms) and t (tons).

Economic matrix

The economic matrix was validated with field visits and informal dialogue with the tenants; the fishing activity has a representation of 66% and it is the predominant sector; 17.8% corresponds to agricultural activities. It is important to emphasize that productive systems of backyard agriculture are small-scale, which function as alternative model when there is scarcity of fishing; for example, there are watermelon, legumes and transitory crops that serve as sustenance for the households. The commercial activity has a representation of 8.9% and this sector is not as developed; they are shops and tortilla selling; and lastly, the livestock production sector which has a representation of 7.1% and is not a very developed sector at the local level. These activities have a strong impact on the rural sector and the human needs of the study area. The community of El Arenal has several problems at the economic level: the low diversification of the primary sector and a mono-productive matrix (Figure 6). The development of new forms of social organization is proposed as strategy to protect and conserve the fishing resource, the increase in backyard agriculture, and the strengthening of community links with institutions of power for an optimal consolidation of fishing cooperatives and the minimization of the exhaustion of natural resources.

According to Josephraj *et al.* (2022), in fishing zones of Colombia there was evidence of a strong marginalization, the rural communities have few access paths, and this made the fishermen sell the fishing product to intermediaries at a low cost. This reality is not far from

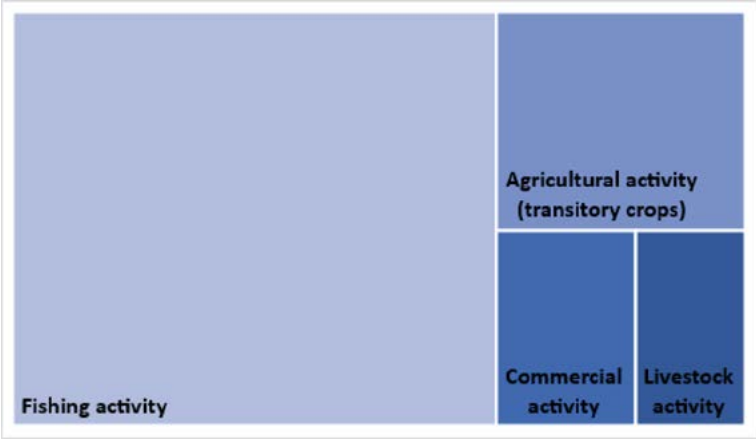

Figure 6. Economic matrix. Source: Prepared by the authors with data from the Statistical Directory of Economic Units (*Directorio Estadístico de Unidades Económicas* (DENUE), field observation of economic units, and informal dialogue with key stakeholders.

this study, where a strong marginalization has been detected due to the lack of integration of the community of El Arenal with the city of Acapulco, which has eased the emergence of intermediaries or the so-called "brokers" that purchase the fishing product at a low cost and sell it to the local markets. According to Kimani *et al.* (2020), this problem of brokers is a consequence resulting from the lack of guarantees from local governments. However, the commercialization of fishing and the sustainability of households from the communities is achieved, even with low income for the fishermen.

This is related with the function of fishing cooperatives in the community of El Arenal. In theory, these associations have to guarantee the purchase and the commercialization of the fishing product and the search for market niches that ensure the exits of the stock of the fishing product. Likewise, in the study area the fishing activity does not have a positive impact at the local and regional level because communities lack monetary resources, inter-institutional management, and organization between cooperatives. However, in the studies by Samian *et al.* (2022) and Salomón (2023), they found that fishing cooperatives have a great socioeconomic impact and guarantee the purchase and sale of the fishing product to fishermen. The fishing cooperatives contribute to the regulation of prices and manage support programs to reduce the vulnerability of the social capital; the cooperative movement has been important to stop the appearance of monopolies and to avoid overfishing and the intermediaries that place fishermen at an economic disadvantage.

On the other hand, the socioeconomic problem of coastal fishing in El Arenal is the lack of management and planning of the fishing resource and the lack of programs for the mitigation of overfishing, in addition to the breach of the closed season that generates a strong pressure on important species for the social, economic and ecological scope (Juárez *et al.*, 2019; Sandoval *et al.*, 2020). The socioeconomic problem of the fishing activity is similar and has been detected by Makwinja *et al.* (2021), where the Molombe Lake (Malawi) was affected from overfishing, the lack of imposition of closed seasons and of community leadership to stop overfishing and the recovery of lagoon resources also,

causing a loss of social capital and a deceleration in the regional and local development of communities.

Fishing is linked to a territorial conception that allows fishermen from the community of El Arenal the legitimate right to take advantage of the resources from the Laguna de Tres Palos. With time, this lagoon has been territorialized and has become a common economic zone for the subsistence of the community. This appropriation of the lagoon has also generated social, political, and productive links related to the consumption and use of the fishing resource. This has also created a social value of the ecosystem thanks to the knowledge of fishermen about the fishing areas, ensuring that they all have equal access to it. However, for Song and Soliman (2019) and Lechuga *et al*. (2021), the right to fishing territory leads to a process of governance and conceptualizing the relationship of the individual with the productive territory is a challenge in terms of the fishing exploitation and the recognition of rights of fishermen on the use of the space. The productive territories for the use and exploitation of fishing still have problems in terms of the recognition, the legitimation of the fishing task, and the role that fishermen play within the local economies. Likewise, it opens the window to few socioeconomic guarantees on those territories that became productive for fishing and the sustainability of the communities.

On the other hand, Raycraft (2019) and Castillo and Mariano (2020) found similarities. The fishing activity led to processes of governance at the community level because the participation of fishermen, the management and the knowledge about the ecosystem for exploitation have contributed to a territorial configuration of the link and the social recognition of an ecosystem that provides welfare. However, this study has not reached an analysis of the presence of governance of fishing in the communities adjacent to the Laguna de Tres Palos; if the socioeconomic development is linked to territorialization of that lagoon and perhaps there are possible leaderships that serve or act as an instrument for scenarios of fishing governance, and which allow greater guarantees within the economic process and the institutional stakeholders involved in local economies.

CONCLUSIONS

The production model based on fishing generated overfishing at the Laguna de Tres Palos. The communities adjacent to the lagoon would be the ones most affected, even El Arenal, given that the primary economy has low diversification. On the other hand, fish production is in an economic scheme where "brokers" play an important role in the commercial dynamics. The low function of the cooperatives to fulfill the role of facilitators between the commercial relation of the fisherman and the local market has been minimal. All in all, one of the socioeconomic problems is the few guarantees that the community has in times of scarcity. On the other hand, institutional neglect is an element that contributes to the problem due to the lack of support programs, as well as "temporary employment". In addition to this situation, there is centralism of Acapulco and scarce attention to the rural communities that was evidenced in the high marginalization that displaces the possibility of accessing basic services and overcoming human needs (education, employment, health, and water).

On the other hand, within the production factors, something that stands out is the knowledge of locals about the loss of biodiversity from overfishing at the Laguna de Tres Palos; the local stakeholders who participated in this research considered that the excessive exploitation without respecting closed seasons ultimately determines the interaction with the ecosystem and the attainment of a future for the product; this is explained by the relationship of economic dependency on a resource and a local economy that has low diversification, sustained by fishing cooperatives.

Fish reproduction and fishing is mediated by the opening of the bar which favors the exchange of sediments and salinity, and the influx of marine fish with economic value, as well as climatic factors and rainfall that increase the abundance of species. Community participants express a social and cultural rootedness with the lagoon, which is perceived as a common economic zone that favors the sustainability of the households.

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Colegio de Postgraduados

Livestock in the sustainability of changing livelihoods in high mountain communities in a Natural Protected Area in Central Mexico

Martínez-Hernández, J.[1]; **Colín-Navarro, V.**[1]; **Rosa-García, R.**[2]; **Albarrán-Portillo, B.**[3]; **Valdés-Piña, María G.**[4]; **Arriaga-Jordán, C. M.**[1]; **Estrada-Flores, J. G.**[1]

[1] Universidad Autónoma del Estado de México, Instituto de Ciencias Agropecuarias y Rurales (ICAR), Univ, Campus UAEM El Cerrillo, C.P. 50090, Toluca, Estado de México, México.
[2] Servicio Regional de Investigación y Desarrollo Agroalimentario (SERIDA), Carretera a Oviedo s/n, 33300 Villaviciosa, Asturias, España.
[3] Universidad Autónoma del Estado de México, Centro Universitario UAEM-Temascaltepec, Carretera Toluca-Tejupilco, km. 67.5, Temascaltepec, C.P.51300, Estado de México, México.
[4] Universidad Autónoma del Estado de México, Facultad de Planeación Urbana y Regional, Uni Mariano Matamoros s/n, Toluca, C.P. 50130, Estado de México, México.
* Correspondence: jgestradaf@uaemex.mx

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ABSTRACT

Objective: Livelihoods of communities within the high mountain Natural Protected Area for Flora and Fauna 'Nevado de Toluca' (NPAFFNT) in the central highlands of Mexico have changed due to statuary limitations established in management program respect to agro-silvopastoral activities. In these communities, the small scale farmers who still rely on livestock, mainly sheep production. There is no knowledge on the sustainability, strengths and weaknesses of these systems and the influence of seasons in this area of marked bioclimatic seasonality.

Methodology: The IDEA method to assess sustainability was applied in a small village within NPAFFNT in the dry as in the rainy season.

Results: Sustainability is limited by the economic scale while their main strengths were in the agroecological scale in both seasons. Indicators show deficiencies in farmer education, lack of technical assistance, poor local infrastructures, lack of access to institutional support programmes, and the revision of norms for the natural protected area.

Conclusions and implications: Results are useful for improving the sustainability of these systems that may improve que quality of life of high-mountain communities and provide ecosystemic services relevant for society. Diversification of income sources are of needed given the limitations that the new statute create on livestock and agricultural activities. It is advisable to increase the educational level of the inhabitants and farmers receive specialized technical training in agricultural and sustainable livestock production.

Keywords: Sustainability; seasonality; IDEA method; high mountain systems; indicators.

Image by Sarah Mc Evoy at Pixabay

INTRODUCTION

The future of society is jeopardized by patterns of consumption and exploitation of natural resources that threaten the diversity and resilience of human communities and the ecosystems they inhabit. Therefore, there is a need to redirect production systems and livelihoods towards more sustainable strategies (FAO, 2018).

The relationships between the agricultural field and the environment in mountainous territories are usually approached from the point of view that the negative effects that may occur due to these practices affect natural resources as little as possible, allowing sustainable use to be given to these on the part of the population, especially those who are dedicated to livestock, taking charge of giving an adequate use to the grasslands that are destined for grazing to protect the biodiversity in this type of areas (Lasseur, 2005).

Mountain agricultural systems are characterized by their biological diversity, in addition, they are considered as forms of family livelihood conditioned mainly by the scarcity of arable land and the difficulties of access to such territories (Martínez y Rosser, 2012; Kohler Y Romeo, 2014); these are generally dynamic and imbalances occur over time, given by the interactions that pass between each of its components (Calvente, 2007); allowing them to be sustainable but not stable, since they are in a construction process that is based on past experiences and future projections (Fuentes y Marchant, 2016). Under those high mountain conditions, some crops and livestock rearing constitute the basis for the livelihoods and subsistence of those communities (Lasanta, 2010), who at the same time exert a strong activity that shapes rural landscapes (González *et al.*, 2019). Among mountain territories, there is a special interest in the development sustainable livestock strategies for those systems located in protected areas, because there the objectives for the use of natural resources must be strongly aligned to the parallel efforts of environmental conservation. A mountain natural protected zone with singular characteristics is the Natural Protected Area for Flora and Fauna 'Nevado de Toluca' (NPAFFNT) in the central highlands of Mexico, since it not only holds an important natural heritage, but has moved from the status of National Park decreed in 1936 to that of a protected natural area in 2013 (CONANP, 2013), with the objective of conciliating the socio-economic interests of communities living within the area, and the environmental protection objective (Lasanta,2010; González *et al.*, 2019).

The volcano 'Nevado de Toluca' (Snowed Mountain of Toluca) or *Xinantecatl* in the native nahuatl language, is the fourth highest peak in Mexico at an altitude of 4,660 m, and the protected area comprises 53,590 ha of which 1,941 ha are the totally protected nucleus and 51,649 ha comprise the buffer zone, where 16 small rural villages are located with a population of 5,297 people live (CONANP, 2016).

A source of knowledge on the current situation and the evolution of the natural and cultural heritage are the local communities since they possess the ecological knowledge of their surroundings from their experience and adaptation to optimize the use of natural resources. This knowledge enables the inhabitants of the area to valorise changes from a holistic perspective attending socioeconomic and environmental components, as well as identifying causes for those changes and possible consequences (CONANP, 2016).

However, local ecological knowledge needs to be considered in actual times has received little attention by the scientific community, in spite of its potential to implement strategies better adapted to local conditions (Santana, 2012) and better valued by communities who inhabit those areas (Caro-Borrero *et al.*, 2017).

Several authors have analyzed the main factors that may jeopardize the sustainability of livestock rearing in Mexico (Fadul-Pacheco *et al.*, 2013; Salas-Reyes *et al.*, 2015; Vences *et al.*, 2015; Prospero *et al.*, 2017) and in NPAFFNT (Granados *et al.*, 2018; Santana, 2012; Santana-Medina *et al.*, 2013; Endara *et al.*, 2012).

However, available information on sustainable strategies to solve the challenges in this protected natural area is scarce, and not from a holistic assessment of current productive systems considering environmental, socioeconomic, and cultural factors including governance; and that integrates both scientific as well as local ecological knowledge.

The NPAFFNT represents a challenge for human settlements that have lived within the area for around a hundred years. The area was initially established as a national park in 1936, and although formally there should be no human settlements within the park, many small villages founded before the decree of national park remained. Their livelihoods were linked to a silvopastoral system based mainly on sheep rearing and some cattle that used to be grazed in the forest and in openings in the forest, complemented with crop residues during the dry season.

However, in 2013 the statute of the area changed and became a Natural Protected Area of Flora and Fauna, that enables human settlements within the buffer zone, and several productive activities as cropping, livestock rearing, and forestry, although with a strictly enforced new statute limiting many of these activities. Grazing has been banned in most of the forested area, severely limiting the traditional activities of the silvopastoral systems.

The objective was to assess the sustainability of the farms in a community located within the NPAFFNT that live under the new statute, in order to identify strengths and weaknesses, and if these are limited by season of the year. The objective was met by an integrated analysis of information from a battery of sustainability indicators together with that derived from de local ecological knowledge of the people that inhabit the area.

MATERIALS AND METHODS

Study area

Work was undertaken in the village of Agua Blanca (19° 04' 45" N and 99° 50' 25" W) in the municipality of Zinacantepec in the State of Mexico in the central highlands of the country. Altitude is 3200 m, and the village is located in the buffer zone, at 6.5 km from the nucleus protected area. Predominant climate is sub-humid semi-cold, with mean annual rainfall of 1300 mm and mean annual temperature of 12 °C (INEGI, 2018). The town is integrated with 17 family units. The grasslands have some grasses such as *Vulpia myuros* (L.) C. C. Gmel, *Nassella mucronata* (Kunth) Pohl, *Trisetum spicatum* (L.) K. Richt and *Muhlenbergia* sp.; there are also some species like *Astragalus tolucanus* Rob. & Seaton, *Cuphea procumbens* Ort., *Prunella vulgaris* L., *Salvia reptans* Jacq., *Loranthus greenm* (Jack) and *Senecio angulifolius* DC. (Martínez *et al.*, 2018).

Collection of information

Collection of information followed the case study methodology (Yin, 2018) in terms of agrosilvopastoral activities. The assessment of sustainability was from December 2017 to November 2018. All 17 families were invited to participate in the study. All households kept animals. All of them kept sheep as the main livestock species in these high mountain systems, and most keep poultry and some also kept cattle. Eventually, only 13 households accepted to participate in the study during the whole year. Despite new restrictions brought about by the new statute as a natural protected area, families keep relying on livestock, mainly sheep, as the basis for their livelihoods.

Farmers do not keep any records, so information was collected through monthly visits when semi-structured questionnaires were applied to collect detailed information of the family activities; which included the sowing of seasonal crops, such as oat (*Avena sativa* L.) and wild edible plants (*Chenopodium* spp. and *Amaranthus* spp.), sheep farming is practiced in extensive production systems, made up of common areas, paddocks and farmland. Another important activity is the collection of wild mushrooms in the forest in the rainy season, which are marketed in local tianguis, firewood is also collected which is used to cook their food. Another way to obtain income is that some heads of families are dedicated to construction worker or work as day laborers inside or outside the community.

Sustainability assessment

The IDEA method (Indicateurs de durabilité des exploitations agricoles) version 3.0 was used. The method enables the integral assessment of the sustainability at farm level through 42 indicators grouped in 10 components that comprise the three scales of sustainability: (agro-ecological, socio territorial, and economic (Zahm *et al.*, 2008; Vilain *et al.*, 2008).

The IDEA method establishes a scoring scale that goes from 0 to 100 to evaluate the sustainability of each the indicators that comprise it; the scale with the lowest score identified as the limiting scale for the overall sustainability scales (Vilain *et al.*, 2008). The method had to be adapted to local conditions before it could be applied, due to the difference between the original context of French agroecosystems and the study area (NPAFFNT) as a protected natural area (Table 1).

The study area does have a marked bioclimatic seasonality, with well-defined rainy (May to October) and dry (November to April) seasons (Figure 1), that may determine how farming systems function and the timing of activities. Therefore, information was collected and analysed for each season.

Statistical analysis

For analysis of scores of agroecological, socio-territorial and economic scale, the indicators were separated and grouped. Subsequently, the indicators were selected for each of criteria. The results of rainy and dry conditions for each of the criteria were analyzed through a chi square test ($P<0.05$).

Table 1. Summary of modified indicators to assess sustainability in Agua Blanca.

Scale	Component		Indicators
Agroecological	Organization of space	A9	Assessed according to the management plan for Natural Protected Areas (CONANP, 2016).
	Farming Practice	A15	The score was changed to 0-6 points, penalizing self-diagnostics and the application of treatments without veterinary advice, as well as lack of records.
	Quality of products and the land	B1	It was following state norms (40), with 3 pints if protected denomination of origin or green label.
Socio-territorial	Employment and services	B8	It was considered as services to the community (common work, road and water spring maintenance)
	Ethics and human development	B14	It was scored according to schooling of farmer. 0 = no schooling, 1 = primary education, 2 = secondary education, 3 = high school, and 4 = Children with university education.
Economic	Viability	C1	Assessed by Benefit/Cost Ratio (BCE) (22), and not in terms of minimum wage.

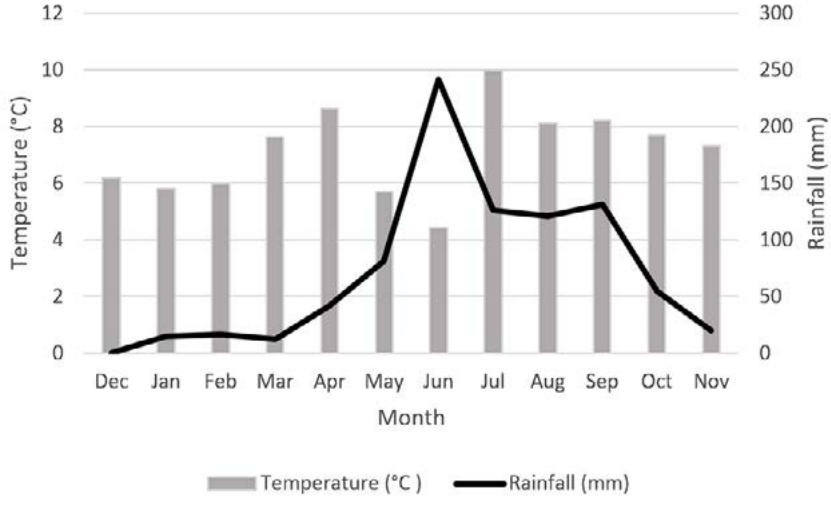

Figure 1. Temperature (°C) and rainfall (mm) since December 2017 to November 2018 (Fuente: CONAGUA, 2015).

RESULTS AND DISCUSSION

Although the total area for the village of Agua Blanca is 2,820 ha of pine (mainly *Pinus hartwegii*) - fir (*Abies religiosa*) forest, that used to be grazed by flocks and herds of the community, the area destined by the new statute for NPAFFNT to the village of Agua Blanca is only 48.5 ha for houses, crops, and livestock rearing with strict norms in place limiting the access of the community livestock to areas outside the village. Therefore, farms are very small, from 0.6 to 6.1 ha where 61% of land is destined for cropping maize and faba beans (*Vicia faba*) for self-consumption, and oats (and straws from maize and faba beans) as forage for livestock.

Grazing has been banned from most of the forest areas in the remaining 2,771.5 ha, so that the traditional silvopastoral management has been severely limited. Families can undertake approved activities in the forest area, mainly the collection and sale of wild mushrooms that are an important source of complementary income in the rainy season (Martínez *et al.*, 2019); but the ban on grazing has meant a need to reorganize their activities, although livestock rearing is still the basis of their livelihoods.

Table 2 shows the animal inventories of the 13 participating farms, who raised a total of 241 sheep, 30 cattle, and 156 diverse poultry (chickens, turkeys and one farm had quails). Sheep is the basis of the livestock system, although two farms sold their sheep during the study to meet economic needs. One farm also had one goat, and one farm kept one pig, but these species are not common in these high mountain systems.

The sheep are grazed during the day in pastures close to the family unit, in the dry season they are grazed inside the forest, an average of four and up to 7 hours a day. They are housed overnight in pens that are made with material from the region. Sometimes supplements such as tequesquite (mineral salts) or oatmeal are provided, in dry season, if they have economic resources to acquire them. Families in Agua Blanca have had to reorganise their livelihoods since they are now unable to graze their livestock outside the village boundaries imposed by the new statute.

The overall sustainability assessment for the three scales of the 13 farms is shown in Table 3 for both seasons. In the dry season, the economic scale with a score of 49 points determines the sustainability, having a large seasonal variation since the score for the economical scale was 74 in the rainy season. It is the socio-territorial scale that limits sustainability in the rainy season with a score of 59 points. The agroecological scale had the highest scores similar in both seasons with 79 in the dry season, and 82 points in the rainy season (Table 3).

Table 4 shown the scores obtained for each of the indicators belonging to the agroecological scale. It was possible to observe the strengths in the agricultural management practices that the inhabitants have, which are stable in both seasons, this was measured

Table 2. Animal inventories for 13 participating farms.

	Farm													
Species	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**Total**
Sheep	6	35	21	13	60	2	0	24	50	0	6	4	20	241
Cattle	0	0	0	1	11	0	0	0	7	0	0	0	11	30
Poultry	1	7	12	7	15	16	2	0	7	5	13	51	20	156

Table 3. Sustainability score by three scales per season.

Scale	**Season**		**Maximum possible score**
	Dry	**Rainy**	
Agroecological	79	82	100
Socio-territorial	60	59	100
Economic	49	74	100

Table 4. Scores for the agroecological scale of the locality Agua Blanca per season.

		Score		Maximum possible score	P<0.05
		Dry	Rainy		
Diversity	**Sub-total**	**25**	**25**	**48**	**7.81***
Diversity of annual and temporary crops	A1	2	2	14	
Diversity of perennial crops	A2	6	6	14	
Animal Diversity	A3	14	14	14	
Valorization and conservation of genetic resources	A4	3	3	6	
Organization of space	**Sub-total**	**21**	**24**	**43**	**12.59***
Crop rotation	A5	0	3	8	
Dimension of fields	A6	6	6	6	
Management of organic waste	A7	3	3	5	
Ecological buffer zones	A8	7	7	12	
Contribution to environmental issues of the territory	A9	1	1	4	
Improvement of the space	A10	2	2	5	
Fodder area management	A11	2	3	3	
Farming practices	**Sub-total**	**33**	**33**	**46**	**12.59***
Fertilization	A12	6	8	8	
Manure management	A13	3	3	3	
Pesticides	A14	11	10	13	
Veterinary products	A15	0	0	3	
Soil resource protection	A16	5	5	5	
Water resource management	A17	4	4	4	
Energy independence	A18	4	3	10	
Total		**79**	**82**	**137**	

through fertilization, which presented a slight increase in the rainy season, and the use of pesticides that is also bigger at the same season. The water management as a natural resource and the protection given to the soil to prevent its loss due to erosion and the management of manure in the production unit (indicators A12 to A18).

At the same scale, low values were found for the diversity of crops (A1 and A2) since oats and some edible plants are planted only in some production units. Crop rotation does not occur in the locality, nor does the veterinary management of livestock (A9 and A15), which is within this scale because it is the producers themselves who take care of their animals and sometimes, they self-medicate their animals. The statistical analysis showed significant differences between seasons ($p<0.05$).

The results obtained from the indicators of the socio-territorial scale are presented in Table 5. According to the quality of the products and the land, very homogeneous values were presented between seasons, importance is given to the use and management of manure, access to the spaces that make up the territory and the involvement of the inhabitants in the different social issues of the community (B1 to B5) which gave a total of

22 points at the time of rainy season and 21 points in the dry season; however, they only represent 60% of the total maximum points to be obtained in these indicators of the IDEA method. Among these indicators are areas for improvement which are related to manure management and accessibility to all spaces belonging to the locality that may be important for the inhabitants since they can be a way of obtaining extra income for the family.

Regarding the indicators related to employment and services in the locality (B6 to BII) where reference is made above all to the trade of products that takes place between localities (tianguis), to collective work, which gives way to self-employment and the improvement of sheep farming, sustainability in each of the production units and the way in which the inhabitants value the natural resources they have. The use of these resources is mainly intended for the houses building, (B18) although these are generally with dirt floors and have very few services, which is similar in the houses of almost the entire town.

In relation to indicators related to ethics and human development, scores were around 50% of maximum possible scores, although with important differences among indicators and between seasons, observing a 14% decrease in scores in the rainy season. Variations ranged from high scores for isolation (B17) to a null score for intensity of work (B15) and training and education (B14). The indicator on reliance of purchased concentrates (B12) also showed a large inter-seasonal variation with a higher dependence of purchased feeds in the dry season and lower in the rainy season, due to the low availability of forage in the forest in the dry season.

There are two indicators that may clearly be improved: animal welfare (B13) and training and education (B14). Significative differences were observed ($P<0.05$) among the criteria of the socio-territorial scale.

Incomes in the economic scale were from the sale of livestock (mainly sheep), manure (in the dry season), and wild mushrooms (in the rainy season). Some families have started to provide services to tourists, as eco-tourism is promoted as means to generate incomes and wellbeing in these poor mountain communities. One family rented a place to spend the night, and one family prepared and sold meals to visitors, although this activity is very incipient as Agua Blanca is away from the main areas visited by tourists and hikers.

Indicators for the economic scale showed large variation between seasons (Table 6). Indicators for economic viability were particularly low in the dry season (only 12 of 30 possible points), that increase in the rainy season (19 points). The indicator for transmissibility (C5) is slightly higher with 13 out of 20 possible points. Significative differences were observed ($P<0.05$) among the criteria of the economic scale.

Arriaga-Jordán *et al.* (2005) documented the livestock agrodiversity of small-holder *campesino* farms on hill-slope indigenous agricultural communities of the highlands, and the importance that livestock rearing has in the survival of these families. Hence the importance of evaluation in a high mountain area as in this case where sustainability dependent on the season, limited strongly by the economic scale (mainly in the dry season), with the socio-territorial scale limiting sustainability in the rainy season. The strengths of the system are in the agroecological scale.

These results are like those obtained in specialized market oriented small-scale dairy systems in temperate agricultural valleys (Fadul-Pacheco *et al.*, 2013; Prospero *et al.*, 2017),

Table 5. Scores for the socio-territorial scale of the locality Agua Blanca.

		Score		Maximum possible score	P<0.05
		Dry	Rainy		
Quality of the products and the land	**Sub-total**	**22**	**21**	**34**	**9.49***
Quality of manure	B1	7	7	10	
Enhancement of buildings and landscape heritage	B2	6	5	8	
Inorganic waste management	B3	2	2	5	
Space accessibility	B4	2	2	5	
Social involvement	B5	5	5	6	
Employment and services	**Sub-total**	**27**	**27**	**36**	**11.07***
Short trade	B6	7	7	7	
Autonomy and enhancement of local resources	B7	8	8	10	
Services, multi-activities	B8	5	5	5	
Contribution to employment	B9	3	3	6	
Collective work	B10	1	1	5	
Probable farm sustainability	B11	3	3	3	
Ethics and human development	**Sub-total**	**11**	**11**	**39**	**12.59***
Dependence on commercial concentrates	B12	0	1	10	
Animal welfare	B13	1	1	3	
Training-education	B14	2	1	6	
Labour intensity	B15	0	0	7	
Quality of life	B16	4	4	6	
Isolation	B17	3	3	3	
Quality of buildings	B18	1	1	4	
Total		**60**	**59**	**109**	

Table 6. Scores for the economic scale of the locality Agua Blanca.

		Score		Maximum possible score	P<0.05
		Dry	Rainy		
Viability	**Sub-total**	**12**	**19**	**30**	**3.84***
Economic viability	C1	2	9	20	
Economic specialization rate	C2	10	10	10	
Independence	**Sub-total**	**19**	**19**	**25**	**3.84***
Financial autonomy	C3	12	12	15	
Sensibility to government subsidies	C4	7	7	10	
Transferability and efficiency	**Sub-total**	**18**	**36**	**45**	**3.84***
Transferability	C5	13	13	20	
Efficiency of the productive process	C6	5	23	25	
Total		**49**	**74**	**100**	

as well as in dual-purpose silvopastoral cattle systems in sub-tropical areas (Salas-Reyes *et al.*, 2015; Vences *et al.*, 2015).

Scores obtained in the agroecological and socio-territorial scales in this study are higher than findings by Fadul-Pacheco *et al.* (Fadul-Pacheco *et al.*, 2013) in small-scale dairy systems in temperate agricultural valleys, or by Vences-Pérez *et al.* (2015). Higher agroecological scores herein reported may be since the studied farms are within a natural forested area in a silvopastoral system based on mainly sheep production that makes use of the natural vegetation of the area. Ross *et al.* (2016) found that resource economy and the ecological impacts of sheep grazing, showing that sustainability boundaries are most likely to be exceeded in fragile environments.

Currently, there is great interest in agrosilvopastoral systems that include livestock in ecosystems that integrate crops, grasses, shrubs and trees that offer a range of resources, and at the same time provide important environmental services (Godinho *et al.*, 2018). Agrosilvopastoral management enable multifunctional and more productive systems than if the activities they include were independent as in monocultures (Albarrán-Portillo *et al.*, 2019). Despite these benefits, it should be considered that this study was carried out within a protected natural area and agrosilvopastoral activity is restricted to limited areas.

Studies with sheep production in Morocco confirm higher sustainability scores for extensive and agro-silvopastoral systems, that showed higher strengths in the agroecological scale and the use of local breeds, as well as weakness in the economic scale (Araba and Boughalmi, 2016).

The high socre in animal diversity (A3) is because, although sheep is the dominant species, there are other domestic animal species in the farms as cattle, poulty (with one farm raising Japanese quails), (Table 2). Multispecies livestock systems enable a more efficient use of natural resources, particularly in comples agro-silvopastoral systems characterised by a range of varied resources (from crop residues to forest biomass) that may be valorised by the diverse selection of diets and grazing behaviour of different species (Fraser and Rosa, 2018).

In this study, the different stratified use of areas was observed in the height of grazed vegetation, such that sheep grazed the lower strata, while cattle the higher stratum. In the area, sheep consume at least six different plant species (Esquivel y Estrada, 2014).

It is the agrobiodiversity of both plant and animal species linked to agrosilvopastoral systems that may contribute to food self-sufficiency and productive diversification in farms that need to improve on the economic scale for which management options must be developed (Ochoa *et al.*, 2009), for example improving feeding systems and animal health.

Low scores for the cropping indicators (A1, A2, and A3) reflect the little crop production in the studied village, where families till an average of 0.11 ha of a limited number of plant species with low yields (Piñar, 2001; Méndez, 2021). Inter-season variability may be due to the presence and intensity of frosts, as well as by rainfall which is dependent on geographic latitude (Herrera *et al.*, 2015), which limits the possibility of crops in the dry season. Only one farmer sowed oats (*Avena sativa*) as forage, and faba beans (*Vicia faba*) for self consumption in the dry season, whilst in the rainy season most farmers sowed oats for

forage, and maize and faba beans for their family. Straw and residueds from these latter two are fed to livestock.

The indicator for the contribution to the environmental challenges (A9) is determined by the statute on the use and management of the protected area, in terms of approved activities in the two zones in the area according to the management plan of NPAFFNT (CONANP, 2016).

As mentioned, the area for agricultural activities is now severly limited to the 48.5 ha of the village, with most of the other land set off-limits to grazing in order to protect the forest since work previous to the change of statute from national park to natural protected area identified grazing and uncrontrolled fires (many due to burning grassland to promote regrowth) as two factors, together with illegal loging, that were hampering the renewal and conservation of the forests in the area (Endara *et al.*, 2012).

Now farmers have to face for the seasonality of available feed resources, and the low productivity of forages, particularly in the dry season (Martínez *et al.*, 2018), coupled with the very low land endowment per farm (1.3 ha on average). Extensive grazing in the forest is now banned and authorities promote confined rearing of their livestock even though these families have subsisted and come from a grazing tradition. According to the current statute, confining livestock could increase the risk of environmental damages from a high livestock density.

The statute for the NPAFFNT fails in its soecioeconomic objectives of providing sustainable livelihoods for communities that live within the area by severely limiting the size and growth possibilities of farms. There is no possibility of intensifying livestock rearing under total confinement as proposed, since that would require forage cropping which is not possible given the very small land endowment for crops in each farm, the low education and training level of farmers, and the impossibility to open new lands to cropping. In addition, the farmers do not agree on the confinement of the livestock.

It therefore calls for a revision of the current statute in order to take into consideration the activities and needs of the people in these communties, designing conservation strategies within the social context and the distribution of natural resources in time and space. The need for this revision has been pointed out by other authors (Santana-Medina *et al.*, 2013) that show the dire circumstances faced by the inhabitants in terms of pollution, lack of health services, transport, low levels of community participation, problems with animal health (Méndez, 2021), lack of secondary schools for children, and limited possibilities for income generation or employment.

Recent studies (Granados *et al.*, 2018) have highlighted these circunstances and show that the change in statute from national park to natural protected area have meant little changes in the poverty conditions of the people.

As observed by Santana-Medina *et al.* (Santana-Medina *et al.*, 2013), there is little management of animal health (A15) and veterinary assistance to farmers, although almost 50% vaccinate and 75% de-wormed their livestock. Survival rates of lambs are low (48%) and the incidence of respiratory problems is high 47.5% (Maldonado-Ferrucho *et al.*, 2014). Isolation, difficult access, and poor dirt roads limit the technical assistance farmers get, who treat their livestock with their own resources and from their experience. Animal health

issues linked to poor farm management and preventive strategies in poor communities is a worldwide problem that limits the prospects of a sustainable livestock production.

In regards to the socio-territorial scale, in terms of quality of products and the land, there were low scores in the management of inorganic residues (B3), since villagers just burn or bury their inorganic rubbish. Low scores for accesibility of space (B4) highlights the need to improve roads in the area.

High scores were obtained in the indicator on manure quality (B1), since farmers value sheep manure as the best organic fertiliser compared to manure from other species. This is local knowledge from observation, that coincides with what is established in the State standards for organic soil ammendments, reaffirming the role of traditional local knowledge in the development of sustainable systems (NTE-006-SMA-RS-2006).

Sheep manure is fundamental not only to improve soil fertility and fertilise crops, but for income generation through its sale, within a diversified subsistence economy that combines the sale of seasonal products as manure which is sold in the dry season, and wild mushrooms that are sold in the rainy season, toghether with livestock sales year round.

Among other items related to the conservation of their forests, and set in a participatory community action plan local, villagers mentioned the importance of wild animals for the environment, the provision of firewood for their households, the sale of wild mushrooms as an important source of income, and the collection of moss, sold by some young members of the community in December for Christmas decorations (Santana-Medina *et al.*, 2013).

Another indicator with high scores is related to short trade chains (B6), since the sale of their products is directly to consumers (mushrooms and manure) or to local diners and small roadside restaurants who sell lamb and mutton dishes (Martínez *et al.*, 2019). The obtaines score agrees with reports by Salas-Reyes *et al.* (Salas-Reyes *et al.*, 2015) in dual purpose cattle farms, and related to the low number of links in the trade chain of their produce.

The contribution to employment (B9) and collective work (B10) indicators are areas of improvement. The medium score for contribution to imployment (indicator B9 with 3 points out of 6) relates to the possibility of families to obtain a subsistence livelihood from their small farms and small flocks, although through precarious incomes. As is typical of smallholder farms in Mexico (Posadas-Domínguez *et al.*, 2014), studied farms rely on family labour.

In terms of collective work (B10), there is a tradition in Mexican rural communities to collaborate in work with other members of the community, be it in common tasks (*i.e.* road maintenance) or helping neighbours and family members on specific chores like harvest where help is rotated among participants (Ruíz, 2011; Vizcarra *et al.*, 2013). However, there was a low level of collective work in Agua Blanca, due to strong individualism that limits the potential for the development of productive activities and the capacity of local communities to defend their rights and preserve their cultural heritage (Santana-Medina *et al.*, 2013).

There is a high dependance on purchased concentrates and feeds for livestock (B12) with a strong seasonal variation, with high requirements of external inputs in the dry season. The ban on grazing in the forests has increase the need for purchased feeds.

The low scores in the economic scale, and the strong seasonal variation, are due to low scores in economic viability (C1) and productive efficiency (C6), closely related to the seasonal availability of forage resources. Sheep sales are the main source of income and essential for the family economy as a factor of financial security (Hernández-Valenzuela *et al.*, 2019).

Indicators for economic independence (C3 and C4) did not show seasonal variation, and although high scores according to the IDEA method, they mask the real precarious situation of poverty of households. The indicator for financial autonomy (C3) means farmers do not resort to loans, which makes them economically independent. Reality is that due to marginalisation and poverty, inhabitants of these communities do not meet requirements to be able to opt for loans.

Indicator C4 relates to dependance of government support programmes, but studied farms usually do not have all the legal documentation required (as legal deeds on their farms), which leaves them out of several government support programmes.

In terms of transferability of farms (C5) that relates to the permanence of farms in time, farms are inherited to the children similar to reports by Srour *et al.* (Srour *et al.*, 2009) in Lebannon, with lower scores than those reported by Benidir *et al.* (Benidir *et al.*, 2013) for sheep systems in Algeria probably due to the fact that farms in Agua Blanca do not have important infrastructure or equipement, since farms with high assets may be divided.

A common aspect that was not recorded are share framing agreements that has an effect on transferability. Share farming are oral agreements without any written documents between two persons (Flores, 2016) that is a usual subsistence strategy and means to build social bonds. A person provides livestock and the other one land, labour and inputs, and benefits are shared in equal parts. These shared agreements in some communities of NPAFFNT enable larger incomes from the sale of sheep and manure (Estévez-Moreno *et al.*, 2019).

CONCLUSION

It is concluded that the sustainability of the farms in the study area fluctuates along the year following the availability of natural resources. Although the agroecological scale shows the potential of the locality to make use of natural resources and manage them properly, and the socio-territorial scale shows that there are areas of opportunity, the condition of being in a protected natural area limits the possibilities of improvement that are required locally; because the NPAFFNT management program of itself prohibits the construction of houses with materials other than those available on the site (wood).

It is advisable to increase the educational level of the inhabitants, especially the youngest, and farmers receive specialized technical training in agricultural and livestock production for community memmbers are a starting point to improve the efficiency of farms so that they may also implement more sustainable practices in the use of natural resources.

The weakest scale of sustainability in the studied village is the economic scale both in indicators of economic viability and productive efficiency, as well as in the seasonal fluctuations observed in these indicators due to there is no intensification in sheep production and this activity is carried out as a savings strategy for families.

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CONFLICT OF INTEREST

Authors declare there are no conflicts of interests.

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Colegio de Postgraduados

Canopy rainfall interception by pine (*Pinus hartwegii* Lindl) and oyamel [*Abies religiosa* (Kunth) Schltdl. y Cham.] at the Zoquiapan Experimental Forest Station, Mexico

Bolaños-Sánchez, Claudia[1]; Prado-Hernández, Jorge V.[2*]; Silván-Cárdenas, José L.[3]; Arévalo-Galarza, Gustavo A.[2]

[1] Universidad Autónoma Chapingo, Posgrado en Ingeniería Agrícola y Uso Integral del Agua, Km. 38.5 Carretera México-Texcoco Chapingo, 56230 Texcoco, Estado de México, México.
[2] Universidad Autónoma Chapingo, Departamento de Suelos. Carretera México-Texcoco km 38.5, Chapingo, Texcoco, México, C. P. 56230.
[3] Centro de Investigación en Ciencias de Información Geoespacial (CentroGeo), Contoy 137, Lomas de Padierna,14240 Tlalpan, Ciudad de México, México.
* Correspondence: vpradohdez@gmail.com

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ABSTRACT

Objective: To quantify and characterize the canopy rainfall interception of two dominant forest species of a coniferous forest in Mexico, in order to determine its magnitude and importance in the hydrological cycle and have a source of estimation of interception to improve the accuracy of water balance calculation.

Methodology: Twenty-one rainfall events in the period May-June 2018 were analyzed in two 0.25-ha experimental plots, one with pine (*Pinus hartwegii*) and the other with oyamel (*Abies religiosa*), at the Zoquiapan Experimental Forest Station (EFEZ), located in the Sierra Nevada in the State of Mexico. Rainfall was measured with an automated weather station and interception was recorded by placing collectors under the canopy and collars on the trunks.

Results: The pine tree *P. hartwegii* intercepted 16.37% of the precipitation, 79.86% of the throughfall, and 3.74% of the stemflow. For *A. religiosa*, recorded interception was 24.68%, throughfall 72.42%, and stemflow 2.90%. Precipitation had a linear relationship with both throughfall and stemflow, and an exponential one with canopy rainfall interception.

Implications: The analysis should be extended to other rainy periods to strengthen the study.

Conclusions: The intercepted volume depends on the forest measurement characteristics and leaf area index (LAI) of the species, and the rainfall amount. The fraction of rainfall intercepted is considerable and should be included in hydrological balances.

Keywords: canopy effect, throughfall, rainfall interception, stemflow, water balance.

Image by Bernabe Colohua at Pixabay

INTRODUCTION

Forest ecosystems retain part of the rainfall (rainfall interception) through foliage, branches, leaves and trunks of their trees. A fraction of this captured water returns to the atmosphere as evaporation; the remaining water drains down the canopy structure (throughfall) or the trunk (stemflow), and reaches the ground producing superficial flows through which it finally integrates into the soil as infiltration. This process presents great complexity and is part of the water cycle; its importance lies mainly in the relationship between the effect of the tree cover and the modifications that the latter exerts on water balance (Santiago-Hernández, 2007). Because of this, several authors have conducted studies to identify and account for rainfall intercepted by tree cover. The forest covers of temperate climates and those formed by conifers are the most frequently studied ecosystem types (Návar-Cháidez *et al.*, 2008). According to Besteiro and Rodríguez (2012), interception ranges from 12.2% to 27.2% in temperate forest plantations, agreeing with León *et al.* (2010) and Fan *et al.* (2014) who report 19.0, 22.4 and 22.9%, respectively, for similar species. However, Chen *et al.* (2013), Pérez *et al.* (2015) and Gavazzi *et al.* (2016) differ, reporting 33.2, 29.6 and 33.4% for conifer forests of *Pinus tabulaeformis*, *Pinus pinea*, and *Pinus taeda* L., respectively.

Sadeghi *et al.* (2015) reported variations in rainfall interception between deciduous forests and perennial forests, with 25% and 40%, respectively. For tropical forests, values close to 39% are reported (Crockford and Richardson, 2000). For species of semi-arid ecosystems, percentages vary from 2% to 5% of total rainfall (Oyarzún *et al.*, 1985; Carlyle, 2004); this type of environment is the least studied due to the methodological complexity involved in its measurement and its low annual precipitation.

Although in recent years rainfall interception has become important in hydrological and forest hydrology studies, the increase in studies in different covers and ecosystems is not enough to establish a constant fraction of precipitation as rainfall interception, because the distribution varies according to each cover's specific characteristics and to the zone's particular weather conditions. For this reason, the objective of this study was to estimate canopy rainfall interception, through the quantification of these hydrological process components in the *P. hartwegii* and *A. religiosa* forest species, by using experimental plots and collectors to directly measure rainfall interception. The relationship between throughfall and stemflow, and the volume of canopy rainfall interception as a function of incident precipitation was analyzed. The behavior of the two types of flow and the retention of rainfall by the canopy of both forest species was analyzed based on their forest measurement characteristics, since these species were subjected to the same experimental conditions.

MATERIALS AND METHODS

The experimental area is located in the EFEZ, which has an area of 1638 ha and elevations ranging from 3200 to 3500 m a.s.l. (DICIFO, 2005). It is located in the southeast of the State of Mexico, and enclosed within the orographic system of the Sierra Nevada. The climate is temperate cold with summer rains, and the rainfall ranges from 900 to 1200 mm per year. The main forest species in the EFEZ are *Abies religiosa*, *Pinus hartwegii* and *Alnus firmifolia* (DICIFO, 2005). Two experimental plots of 0.25 ha (50 × 50 m) each were

selected: one for *A. religiosa* and one for *P. hartwegii*; both plots were in the central area of the EFEZ and 600 meters apart.

Characterization of experimental plots

The LAI and forest measurement parameters were obtained from measurements made in 64 *P. hartwegii* and 50 *A. religiosa* randomly selected trees.

Instrumentation for the components of Interception

To obtain throughfall, several 3 L plastic bottles were placed on wooden bases (1.20 m high) and used as collectors. Previously, these collectors were calibrated with a weather station ($P<0.05$). Throughfall was obtained based on the following equation:

$$TH = \frac{vm}{ac} \quad (1)$$

where *TH* is the throughfall (mm); *vm* is the volume captured by each collector (ml); and *ac* is the capturing area of the collector (cm^2).

Several collar-type implements were attached to the trunks in order to measure the stemflow. They were made out of plastic hoses, and attached around the trunks 2.5 times their circumference to form a downwards spiral. The rainfall guttered by these implements was collected in 3 L plastic bottles, and at the end of each event, the volume was measured with a 100 ml plastic test tube. To estimate the depth of the stemflow, the Price and Carlyle (2003) equation was used:

$$FC = \frac{n \times FC(a)}{FA} \quad (2)$$

where *FC* is the stemflow (mm); *n* is the number of tree samples; $FC(a)$ is the mean volume measured from the trees sampled (ml); and *FA* is the basal area of the canopies of n number of trees sampled (m^2).

Gross precipitation and meteorological variables

Twenty-one rainfall events that took place in May and June 2018 were analyzed. That year was very dry: there were few precipitation events and the most representative ones occurred in those months indicated above. The sole condition for these twenty-one events to be classified as such was the absence of rainfall events between them, for at least 6 hours. (Hosseini *et al.*, 2012). The gross rainfall (incident precipitation) (P, mm) was recorded every 10 minutes by a DAVIS® weather station, model Vantage Pro2™ Wireless.

Experimental design

Throughfall was measured with 39 collectors randomly distributed in 13 measurement sites with different coverage characteristics (LAI and forest measurement variables), which are associated with this type of flow (Flores-Ayala *et al.*, 2016). Stemflow was quantified in five trees from each species using one collar per tree (Figures 1a and 1b); the trees selected

had vertical trunks and no bifurcations below 1.3 m. The behavior of the throughfall of the sampling sites was statistically analyzed to show the variation of the canopy characteristics and justify their selection.

Mathematical modeling of rainfall interception

Different empirical mathematical models were tested to obtain throughfall, stemflow, and canopy rainfall retention, as a function of the gross rainfall depth (liquid precipitation rainfall falling on the canopy) of each precipitation event. Linear models were tested as suggested by Leyton *et al.* (1967), and Gash and Morton (1978).

RESULTS AND DISCUSSION

Forest measurement characteristics of the species at plot level

The results showed forest measurements and leaf cover differences between the *P. hartwegii* and *A. religiosa* forests. *P. hartwegii* presented a lesser tree height (H), LAI and Diameter at breast height (DBH), but a greater trunk height (Hf) than *A. religiosa*. On the other hand, *A. religiosa* presented greater dimensions in the higher and lower diameters of its canopies (DM and Dm), providing a greater foliage cover area and lower gap fraction between canopies than *P. hartwegii*. The mean values of H, (DBH), Hf, DM and Dm for *P. hartwegii* were 21.1 m, 15.1 m, 37.6 cm, 7.8 m and 4.6 m, and for *A. religiosa*, they were 23.2 m, 11.6 m, 50.5 cm, 8.4 m and 5.9 m. The LAI values obtained from *P. hartwegii* (2.83) and *A. religiosa* (2.99) corresponded to previously reported ranges for forest species (Peduzzi, 2007; Muzylo *et al.*, 2009; Pérez-Arellano *et al.*, 2015).

Characteristics of rainfall events

The events analyzed are light and moderate rains, according to the CONAGUA criterion (Fan *et al.*, 2014). Approximately 57.2% of the rainfall events corresponded to precipitation of up to 5 mm, 9.5% to 5-10 mm, 23.8% to 10-15 mm, and 9.5% to 15-20 mm.

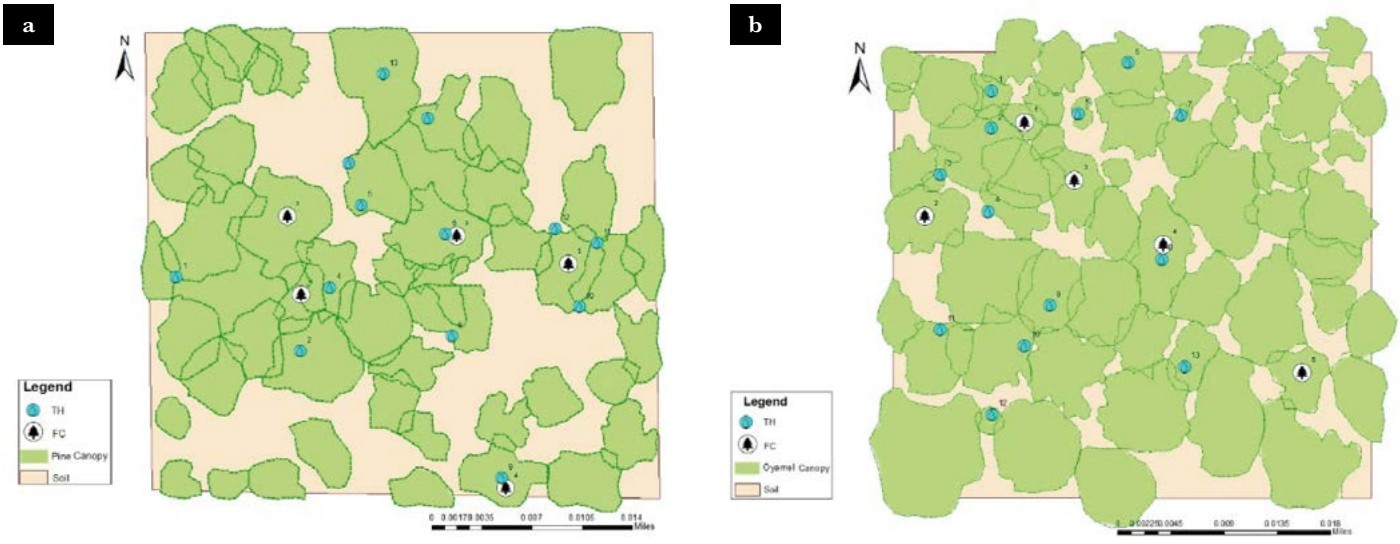

Figure 1. General instrumentation arrangement in the experimental plot for throughfall and stemflow: a) *P. hartwegii*, b) *A. religiosa*.

Rainfall interception and its components

The throughfall of *P. hartwegii* was 100.8 mm, which is 79.86% of the total rainfall depth of the 21 events analyzed (126.2 mm). This percentage was similar to that reported by Santiago-Hernández (2007) (79%) for mountain forests, to that observed by Ghimire *et al.* (2012) (76.2%) for pine forests, and to that found by Pérez *et al.* (2015) (70%) for *Pinus pinea*. However, it was lower than the 89% observed by Gavazzi *et al.* (2016) in *Pinus taedea* L. The differences were attributed to the fact that the rainfall events and the meteorological conditions were different, as well as the characteristics of the trees studied.

The throughfall in *A. religiosa* was lower than that in *P. hartwegii*, with 72.42% of total rainfall. The differences found between the two species were explained by their morphology, LAI and canopy-covered area. *A. religiosa* is a tree with a conical canopy in multi-layers that presents insertion of its leaves in a spiral shape (Arriola-Padilla *et al.*, 2014); this type of structure and geometry allowed retaining more canopy rainfall than *P. hartwegii*, which is a tree with hemispherical round canopy, and lower LAI and foliage thickness values (Tivo-Fernández, 2004).

The throughfall obtained in *A. religiosa* was similar to some of the results reported in species with similar canopy geometry and morphology. Iroume and Huber (2000), and Besteiro and Rodríguez (2012) observed 60% and 60%-75%, respectively. However, Oyarzún *et al.* (1985), Valente *et al.* (1997) and León *et al.* (2010) reported higher percentages of 74%-80%, 82.6% and 81%, respectively.

The stemflow recorded in the *P. hartwegii* plot was 3.74% of total rainfall, agreeing with the results of Ghimire *et al.* (2012) who reported 3.1%. It was higher than what was found by Pérez *et al.* (2015) who reported 0.3%, and it was lower than the 6.7% reported by Santiago-Hernández (2007). The differences among these results can be explained by differences in the morphology of the branches and trunks of the trees.

In the *A. religiosa* plot a stemflow of 2.90% was obtained, a percentage lower than what was obtained in the *P. hartwegii* plot, due to higher water retention by the trunk because its surface is more exposed and rougher. The percentage recorded for *A. religiosa* does not agree with that found in other studies in similar species, and it is attributed to the differences in morphological and phenological characteristics of the trunks of the species (Oyarzún *et al*, 1985; Iroume and Huber, 2000; Léon-Peláez *et al.*, 2010; Besteiro and Rodríguez, 2012).

A linear behavior between throughfall (TH, in mm) and gross precipitation (P, in mm) for both studied species was found (Equation 3 for *P. hartwegii* and Equation 4 for *A. religiosa*), as reported by other authors (Carlyle, 2004; Návar-Cháidez *et al.*, 2008; Carlyle and Gash, 2011; Pérez-Arellano *et al.*, 2015).

$$TH = 0.8452\ P - 0.2795 \quad (3)$$

$$TH = 0.8441\ P - 0.7209 \quad (4)$$

The coefficients of determination obtained were 0.99 and 0.98, and the RMSE values were 0.50 and 0.61 mm for *P. hartwegii* and *A. religiosa*, respectively.

In both species, a positive linear relationship was observed between stemflow (FC, in mm) and gross precipitation (P, in mm); this is a trend similar to those reported in other studies (Carlyle, 2004; Návar-Cháidez *et al.*, 2008; Carlyle and Gash, 2011). Equation 5 corresponds to *P. hartwegii* and equation 6 to *A. religiosa*, with coefficients of determination of 0.77 and 0.86, and RMSE values of 0.11 and 0.10 mm, respectively:

$$FC = 0.0382\ P - 0.0043 \quad (5)$$

$$FC = 0.0398\ P - 0.0646 \quad (6)$$

The analyses of variance conducted on the means of the throughfall depths collected from the rainfall events indicated a highly significant difference ($P<0.05$). Therefore, it is inferred that the chosen sampling sites were suitable since the characteristics of the canopy were both different and representative of the experimental plots.

Canopy rainfall interception

The interception calculated for *P. hartwegii* and *A. religiosa* was 20.66 mm and 31.15 mm, respectively, corresponding to 16.37 and 24.68% of total rainfall. Flores-Ayala *et al.* (2016) reported different total values in a study conducted on the same species as in this study, with 19.20% in *P. hartwegii* and 26.10% in *A. religiosa*. The difference in results between these two studies can be attributed to the fact that Flores-Ayala *et al.* (2016) did not consider stemflow in their calculation of rainfall interception, since it was calculated by subtracting the throughfall from the incident precipitation. If stemflow is not considered, although it apparently has small percentages in large areas with high depth rainfall, this omission can generate important errors in water balances. Previous studies in similar *P. hartwegii* species report values similar to those obtained in this research (Santiago-Hernández, 2007; Ghimire *et al.*, 2012; Pérez-Arellano *et al.*, 2015). For *A. religiosa*, there are similarities with what was reported by Iroume and Huber (2000) and Besteiro and Rodríguez (2012), but there are differences with what was obtained by Oyarzún *et al.* (1985), Valente *et al.* (1997) and León *et al.* (2010). The discrepancies in the rainfall intercepted between species confirm that the redistribution of canopy rainfall is a function of the morphological and structural characteristics of the vegetation type, the characteristics of the rainfall, and the meteorological conditions during the events (Iroume and Huber, 2000).

The relationship between the percentage of rainfall interception and gross rainfall was expressed through a negative exponential function for both plots, with coefficients of determination (R^2) of 0.78 and 0.89, and RMSE values of 4.89 and 7.05 mm for *P. hartwegii* and *A. religiosa*, respectively (Figure 2). Some authors reported a similar relationship between these two components (Oyarzún *et al.*, 1985; Carlyle, 2004; Carlyle and Gash, 2011; Besteiro and Rodríguez, 2012). It was observed that as the amounts of rainfall increases, the rainfall interception decreases, with a fast decline in small amounts and with asymptotic behavior in large amounts, saturating the *P. hartwegii* canopy with smaller precipitation amounts than in the *A. religiosa* canopy. Flores-Ayala *et al.* (2016) found a potential relationship for *A. religiosa* and a logarithmic relationship for *P. hartwegii*,

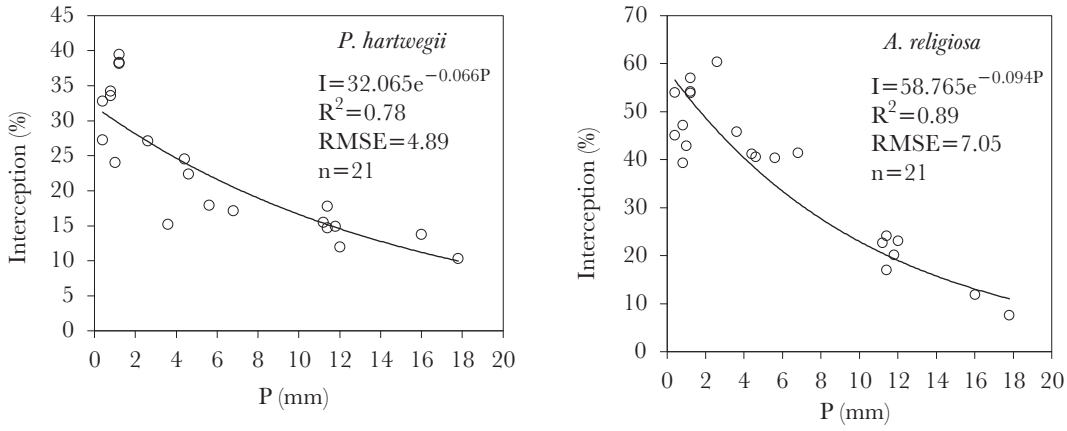

Figure 2. Canopy rainfall interception as a function of gross precipitation on *P. hartwegii* and *A. religiosa*.

with R^2 values much lower than those found in our study, since they obtained 0.58 and 0.42, respectively. In our study, the linear relationships (first-order polynomials) presented values of R^2 equal to 0.89 and 0.78 for *A. religiosa* and *P. hartwegii*, respectively, which are also higher than the values found by Flores-Ayala *et al.* (2016).

CONCLUSIONS

Canopy rainfall interception in the *A. religiosa* plot was higher than that in the *P. hartwegii* plot (24.86% and 16.37%, respectively). The difference is associated with the different characteristics of the canopy in the two species, reflected in the LAI values obtained and in the forest measurement parameters reported, primarily in terms of canopy size.

The *A. religiosa* plot presented canopy density and canopy diameter values higher than those in *P. hartwegii* plot, which led to a lower water contribution through the plant cover toward the ground, since throughfall was 72.42% and 79.86%, respectively. Instead, it generated higher water retention in the trunk, showing a lower stemflow (2.90 and 3.74, respectively), due to its rougher and thicker bark, and greater exposed surface.

Canopy rainfall interception percentage is related to incident precipitation with a negative exponential function, with R^2 greater than 0.78 in both species. Throughfall and stemflow were found to have a linear function with incident precipitation in both species, with R^2 greater than 0.98 and 0.77, respectively.

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