

Advances in biology through agronomy, aquaculture, coastal and environmental sciences

Leandris Argentel Martínez
Ofelda Peñuelas Rubio

Editors



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Ofelda Peñuelas Rubio**

**Advances in biology through
agronomy, aquaculture, coastal and
environmental sciences**



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Prologue

Advances in biology through agronomy, aquaculture, coastal and environmental sciences is an electronic book, edited by Pantanal Editora, based on the compilation of research papers where the authors of the different chapters have used highly current scientific methodologies and research equipment.

The biological sciences as the main object of research in agriculture, aquaculture, coastal and environmental sciences generate every day an understanding of knowledge that allows raising the scientific level of society as part of universal access to knowledge.

This book mainly addresses issues related to the use of plants extracts as sustainable alternatives for biocontrol of pests and bacterial diseases. It also brings together information on viruses and other diseases in aquatic organisms. In addition, studies of mangroves structure and their contribution to carbon sinks in experimental sites in northwestern Mexico are presented. Finally, an analysis on educational strategies for environmental education based on plant biology is carried out.

Editors appreciate the participation of the authors who have come from higher education institutions and research centers of great scientific prestige in Mexico. The majority of them are members of the National Research System of CONACyT, Mexico.

The authors


Summary


Prologue	4
Chapter 1	6
Phytotoxicity of hydroalcoholic extracts of <i>Parkinsonia aculeata</i> L. sp. Pl., in tomato plants. Polyphenol and flavonoid content	6
Chapter 2	17
Evaluation of the antioxidant and antimicrobial activity of hydroalcoholic extracts of <i>Larrea tridentata</i> leaves	17
Chapter 3	27
Morphological characterization of creole populations of ancho pepper of San Luis de la Paz, Guanajuato, Mexico	27
Chapter 4	37
Extraction of carotenoids present in the byproducts of bell pepper (<i>Capsicum annuum</i> L.) using the solvent method assisted with ultrasonic pulses	37
Chapter 5	46
Intracellular Holosporaceae pathogen intensifies the susceptibility of shrimp (<i>Litopenaeus vannamei</i>) to the white spot syndrome virus (WSSV): a preliminary approach	46
Chapter 6	53
Yellow head syndrome virus, a latent problematic for western aquaculture. A review	53
Chapter 7	69
Probiotic effects in tilapia <i>Oreochromis niloticus</i> culture based on growth performance, survival and water quality.....	69
Chapter 8	80
Structure and carbon stock in relation to the biomass of <i>Nymphaea elegans</i> and <i>Sagittaria longiloba</i> in three temporary lagoons in the arid northwest of Mexico	80
Chapter 9	92
Plant galleries as a strategy for environmental education in México	92
Index	104
About the editors	105


Morphological characterization of creole populations of ancho pepper of San Luis de la Paz, Guanajuato, Mexico

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ABSTRACT

The objective of this research was to carry out a morphological characterization of four creole populations of ancho pepper (*Capsicum annuum* L.) from the municipality of San Luis de la Paz, Guanajuato. The research was conducted under greenhouse conditions at Inifap-Cebaj in 2015 and 2016. A completely randomized design with 4 populations and two replications was used, considering 18 plants per replication as indicated in the National System for Seed Inspection and Certification (SNICS) descriptors manual. Plant height (PA), color by anthocyanins at the node level (CAN), leaf length (LH), leaf width (AH), leaf pubescence (PH), fruit diameter (DF), fruit length (LF), fruit length-to-width ratio (RLAF), fruit peduncle cavity (CPD), fruit peduncle cavity depth (PCPF), fruit peduncle thickness (GPF), fruit peduncle length (LPF), fruit pericarp thickness (GPF), flowering initiation time (TIF) and ripening time (TM) were recorded. The joint principal component analysis across years showed that the first two components explained 74,79% of the total morphological variability. On the other hand, the variables with the greatest explanatory capacity for CP1 were AH, LH and LF, and in CP2, GPF, LPF and CAN stood out. These results indicate that leaf and fruit traits are the most important components for the characterization of ancho pepper populations.

Keywords: *Capsicum annuum*, creole populations, morphological characterization.

INTRODUCTION

Mesoamerica and Mexico, as an important part of this region, are recognized as centers of origin and/or domestication of agricultural crops of worldwide importance. In Mexico, pepper (*Capsicum*

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annuum L.) is one of the most important horticultural species due to the value of its production and the high labor demand it generates. It is cultivated in almost all the states of the republic, from sea level altitudes up to 2500 m, and because it is its center of origin, a great diversity of types and varieties have been generated, which constitutes a valuable resource for its genetic improvement (Laborde; Pozo, 1984; Hernández-Pérez et al., 2011). In this regard, MacNeish (1964) and Hernández-Verdugo et al. (2012) state that *Capsicum* spp. was one of the first plants domesticated in the Americas.

Aguilar-Rincón et al. (2010) report that ancho pepper is grown in the states of Zacatecas, Durango, San Luis Potosí, Guanajuato and to a lesser extent in the state of Puebla. In green, it is used in the preparation of stuffed pepper or rajas. However, most of it is consumed dry as a condiment in the preparation of marinades or moles. Despite the great genetic and phenotypic diversity of *C. annum* in Mexico, the regional variants of great economic and social importance are little recognized at the national level.

Knowledge of morphological variation and their geographic distribution patterns is of considerable interest for understanding the evolution of plant species and working on their conservation (Solís-Neffa, 2010). Among the geographic factors that influence the differentiation of populations are climate, latitude and altitude. Climate is considered one of the main factors affecting the distribution and variation of plant species because it can act directly on the fisiological processes of growth and reproduction or indirectly through ecological interactions, such as competition for resources. Several studies have shown that precipitation and temperature influence geographic patterns of morphological variation (Hernández-Verdugo et al., 2012).

Morphological or phenotypic markers have traditionally been used to differentiate varieties (Tapia et al., 2005; Adugna et al., 2006; Piña-Escutia et al., 2010). In this regard, Hernández-Villarreal (2013) proposed that morphological characterization of plant genetic resources is the determination of a set of characters through the use of defined descriptors that allow taxonomic differentiation of plants and concluded that characterization is the first step in crop improvement and conservation programs.

In 2002, the International Union for the Protection of New Varieties of Plants (UPOV) recommended that to measure variability, it is necessary to use discriminatory descriptors and to establish the experiment with a minimum of five plants per accession in homogeneous lots in two replications, thus obtaining better and greater information in the statistical analysis. When performing the characterization, reliable morphological variables should be used to discriminate, thus allowing differentiation between groups. These variables are already established in the so-called “technical guides for varietal description”. In this regard, Villota-Cerón et al. (2012) conducted a morphological characterization study of 68 *Capsicum* introductions and selected promising introductions to increase the varietal supply of this genus. On the other hand, Santiago-Luna et al. (2016) mention that with respect to phenotypic diversity patterns, differences were determined between the populations of Santa María Tonameca and Santo Domingo de Morelos. The latter were highly variable in the characters evaluated.

Three groups of phenotypic diversity were determined, in plant, fruit and those associated with yield per plant.

On the other hand, Elizondo-Cabalceta and Monge-Pérez (2017) conducted a morphological characterization of 15 bell pepper genotypes with square- or rectangular-shaped fruits grown under a greenhouse; they found 5 variables at the qualitative and 8 quantitative levels. The data showed wide variability in plant height, leaf area, stem diameter, stem length, fruit width, fruit length, fruit length/fruit width ratio and fruit wall thickness. They also stated that this information is useful for producers in the process of genotype selection in their production system, according to the market niche of interest, and that the morphological characterization of genotypes is an activity that allows the selection of the most promising varieties of a crop for subsequent use in breeding programs.

In the Macro Vegetable Network in 2016, it is mentioned that a total of 1,226 accessions have been characterized, where 50% refers to morphological characters and 27% corresponds to agronomic evaluation. Therefore, the main results of the chile crop characterization projects stand out, with 715 accessions identified for fresh and dry yields, earliness, fruit quality and size, resistance to pathogens, pigments, colorants and capsaicin. Based on the above, the objective of the present research was to carry out the phenotypic description of four creole populations of ancho pepper from San Luis de la Paz, Guanajuato, under controlled greenhouse conditions.

MATERIALS AND METHODS

The research was carried out in a glass chapel greenhouse belonging to the National Institute of Forestry, Agricultural and Livestock Research, Bajío Experimental Field (INIFAP-CEBAJ), located at km 6 of the federal highway Celaya-San Miguel de Allende, in Celaya, Guanajuato, located at 20° 34' North Latitude and 100° 50' West Longitude and an altitude of 1765 m (Google maps). The climate is semiwarm with an average annual rainfall of 400 to 700 mm.

The biological material (Table 1) was donated by Ing. José Antonio Morín Prado of the Agrisan company, located at Avenida Juárez No. 306, and the seed hoarder Juan amigo of the Estación de Lourdes community in the municipality of San Luis de la Paz, Guanajuato, Mexico. Both companies are located at 21° 29' North Latitude and 100° 70' West Longitude and an altitude of 1990 m (Google maps).

Sanitary management consisted of seed disinfection for fungal prevention using the fungicide Captan 80 WG at a dose of 150 g/100 kg of seed, which was mixed in a container with 500 ml of water along with the seed. Fifty-cavity plastic planting trays were used, which were disinfected in a 10% chlorine solution. Sterile Peat Moss No. 3 was used for seedling production, depositing one seed per cavity at a depth of 0,5 cm. The trays were placed in a germination chamber at a temperature of 25°C, light irrigation was applied to promote seed germination, and the moist substrate was covered with black plastic to prevent evaporation.

Table 1. Creole populations of ancho pepper. San Luis de Paz, Guanajuato, Mexico.

Name of the population	Type of pepper	Origin of the population	Origin
San Luis 1	Ancho	Creole	Acaparadora Juan amigo
San Luis 2	Ancho	Creole	Acaparadora Juan amigo
Especial	Ancho	Creole	Empresa Agrisan
Esmeralda	Ancho	F ₂ Population	Empresa Agrisan

The experiment was carried out in two cycles; in the first cycle, sowing was performed on 30/03/2015, and in the second cycle, sowing was performed on 18/03/2016. A total of 144 pots of 12 inches were used, which were disinfected in a 10% chlorine solution. The substrate preparation was made with 10% Tezontle, 10% leaf and 80% lama soil and was sterilized with Busan 30 W (2-thiocyanomethylthio benzothiazole) for the prevention of *Phytophthora capsici*. Transplanting was performed when the plants were an average of 11 cm tall and had 6 true leaves.

A completely randomized design with four populations and two replicates was used; each replicate consisted of 18 plants per population.

The fertilization dosage recommended by INIFAP was applied based on the Technical Guide for Pepper Wilt (INIFAP, 2011). This fertilization program is dosed by phenological stages to be applied in irrigation during crop development and up to the first 115 days after transplanting; at transplanting, ammonium sulfate (110 kg ha⁻¹) was applied in a period of 10 days; in development, 10 applications were made in a period of 15 days using potassium nitrate (35 kg ha⁻¹), ammonium sulfate (130 kg ha⁻¹) and technical MAP (30 kg ha⁻¹); for growth, there were 10 applications in a period of 20 days using potassium chloride (70 kg ha⁻¹), calcium nitrate (40 kg ha⁻¹), phosphonitrate (110 kg ha⁻¹) and potassium nitrate (75 kg ha⁻¹); and for fruit set and fruit development, there were 15 applications in a period of 30 days with the fertilizers magnesium nitrate (75 kg ha⁻¹) and calcium nitrate (75 kg ha⁻¹). At transplanting, base fertilization with 40 g YaraMila Complex fertilizer (15-15-15) per pot was applied.

On the other hand, imidacloprid and methylcarbamoil were applied at doses of 15 and 12 ml, respectively, in the irrigation system to control whitefly and red spider mites. In addition, a preventive application of the fungicide Busan 30 W (2-thiocyanomethylthio benzothiazole) was made to control *P. capsici* at a concentration of 50 ppm of commercial product, applied every 20 days after transplanting. Forum (dimatomorph) was also applied simultaneously at a dose of 22,45 ml of the commercial product and Cercobin (thiaphanate methyl) at a dose of 20 ml of the commercial product against *Fusarium* spp. and *R. solani*, applied 18 and 39 days after transplanting.

Harvesting was performed manually for both cycles. The fruits were harvested individually per plant from each experimental unit and were placed in different bags for labeling. On the other hand, imidacloprid and methylcarbamoil were applied at doses of 15 and 12 ml, respectively, in the irrigation system to control whitefly and red spider mites. In addition, a preventive application of the fungicide

Busan 30 W (2-thiocyanomethylthio benzothiazole) was made to control *P. capsici* at a concentration of 50 ppm of commercial product, applied every 20 days after transplanting. Forum (dimatomorph) was also applied simultaneously at a dose of 22.45 ml of the commercial product and Cercobin (thiaphanate methyl) at a dose of 20 ml of the commercial product against *Fusarium* spp. and *R. solani*, applied 18 and 39 days after transplanting. Harvesting was performed manually for both cycles. The fruits were harvested individually per plant from each experimental unit and were placed in different bags for labeling.

Forty-seven morphological characteristics were recorded; these were organized by character type and averaged to obtain descriptive statistical values using the technical guide for varietal description of chili developed by the National System for Seed Inspection and Certification (SNICS, 2014).

Of the 47 characters recorded, a mode and mean analysis was carried out to discriminate variables from noninformative characters, obtaining 15 variables with the greatest explanatory capacity for the variability observed among the Creole populations of broad chili (data not shown).

A principal component analysis (PCA) was performed, and the scatter plot of the components, the percentage of variance, the eigenvalues of the variables and the two-dimensional plot of the dispersion of the variables within the components were obtained using the program past (paleontological statistics) ver. 3.15.

RESULTS AND DISCUSSION

In the principal component analysis (PCA) for the pooled data of the 2015-2016 cycles, seven principal components were formed and explained 64.939, 9.857, 8.015, 7.691, 5.633, 3.640 and 0.223%, respectively, of the total morphological variability observed among the studied populations (Table 2).

Table 2. Grouping of components and percentage of variability in ancho pepper materials. Cycles 2015 and 2016.

Principal Component (PC)	Eigenvalues	Percentage of variance
1	9.740	64.939
2	1.478	9.857
3	1.202	8.015
4	1.153	7.691
5	0.844	5.633
6	0.546	3.640
7	0.033	0.223

According to the interpretation and decision making of the data presented in the sedimentation graph (Figure 1), the first two components should be selected since together they explain 74.796% of the total variability of the populations in the two study cycles.

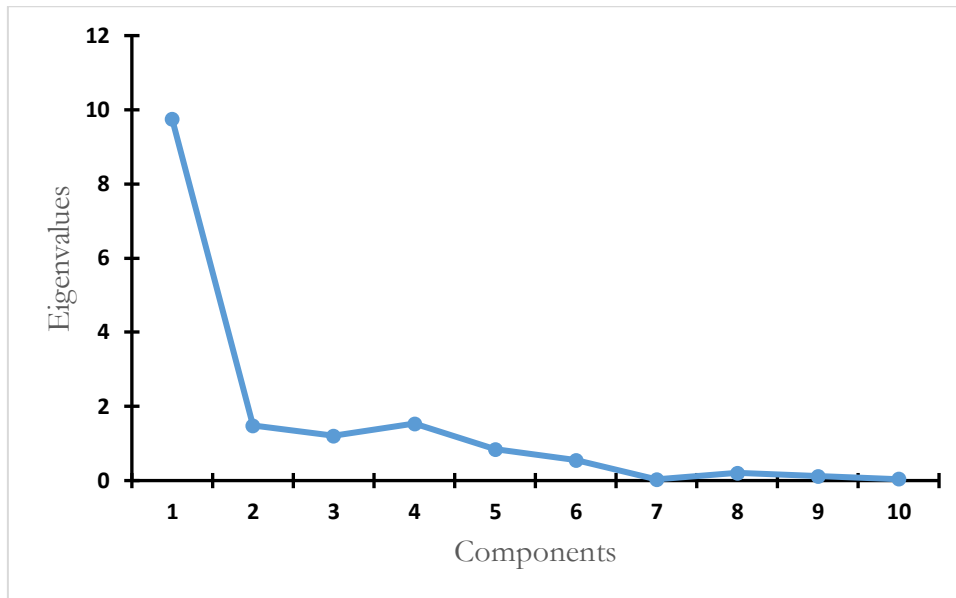


Figure 1. Graph of sedimentation in ancho pepper populations. Cycles 2015 and 2016.

Table 3 shows the variables with the greatest influence for each component; in this sense, the first principal component explained 64.939% of the total variability among the populations and was explained by the width (0.316) and length of leaf (0.313), length (-0.312) and diameter of fruit (0.295), fruit length-width ratio (-0.297), thickness of fruit stalk (-0.296) and time of initiation to flowering (0.293). The second principal component explained 9.857% of the total variability and was related to the variables anthocyanin color at the node level (0.444), fruit stalk length (0.462) and fruit pericarp thickness (0.613). These results agree with those of Ramírez-Novoa et al. (2018), who studied morphological diversity in chili piquín populations from Queretaro and Guanajuato and stated that the characteristics that had the greatest proportion of influence in explaining the total variation among populations were those related to fruits and leaves.

They are also similar to the results obtained by Toledo-Aguilar (2010), who studied morphological diversity in native varieties of poblano chile and concluded that fruit weight and color are important characteristics in phenotypic diversity. On this topic, Narez-Jiménez et al. (2014), in their study of the in situ morphological diversity of wild chiles in the State of Tabasco, found that CP1 explained 43.61% of the total variation and was explained by fruit variables. Another study reported by Pardey et al. (2006) affirms that variability among chili bell pepper populations is explained by fruit characteristics, plant architecture, flower structure and number of flowers per axil.

Likewise, Medina et al. (2006) found morphological differences, mainly in fruit and foliage, in different populations of the genus *Capsicum*. Similarly, it has been reported that fruit width in chili can vary between 5.9 and 10.2 cm (Dasgan and Abak, 2003; Hutton and Handley, 2007; Moreno-Pérez, et al., 2011). Similarly, Castañón-Nájera et al. (2008) mention that wild morphotypes of chile have fruits of

small length and width and affirm that this difference between morphotypes is probably due to changes produced by domestication.

Table 3. Eigenvalues of the variables in the PCA in the characterization of ancho pepper populations. Cycles 2015 and 2016.

Characteristic	ABV	CP1	CP2
Plant height	AP	0.224	0.179
Anthocyanin color at the knot level	CANN	0.202	0.444
Leaf length	LH	0.313	0.052
Leaf width	AH	0.316	0.010
Leaf pubescence	PH	0.253	-0.266
Fruit length	LF	-0.312	0.013
Fruit diameter	DF	0.295	-0.023
Fruit length/breadth ratio	RLAF	-0.297	-0.025
Peduncular cavity of fruit	CPF	-0.099	0.222
Depth of fruit peduncular cavity	PCPF	0.274	-0.060
Fruit stalk thickness	GPF	-0.296	0.188
Fruit stalk length	LPF	0.202	0.462
Thickness of fruit pericarp	GPF	-0.147	0.613
Time to flowering	TIF	0.293	-0.043
Maturation time	TM	0.228	0.115

CP1=principal component 1, CP2=principal component 2

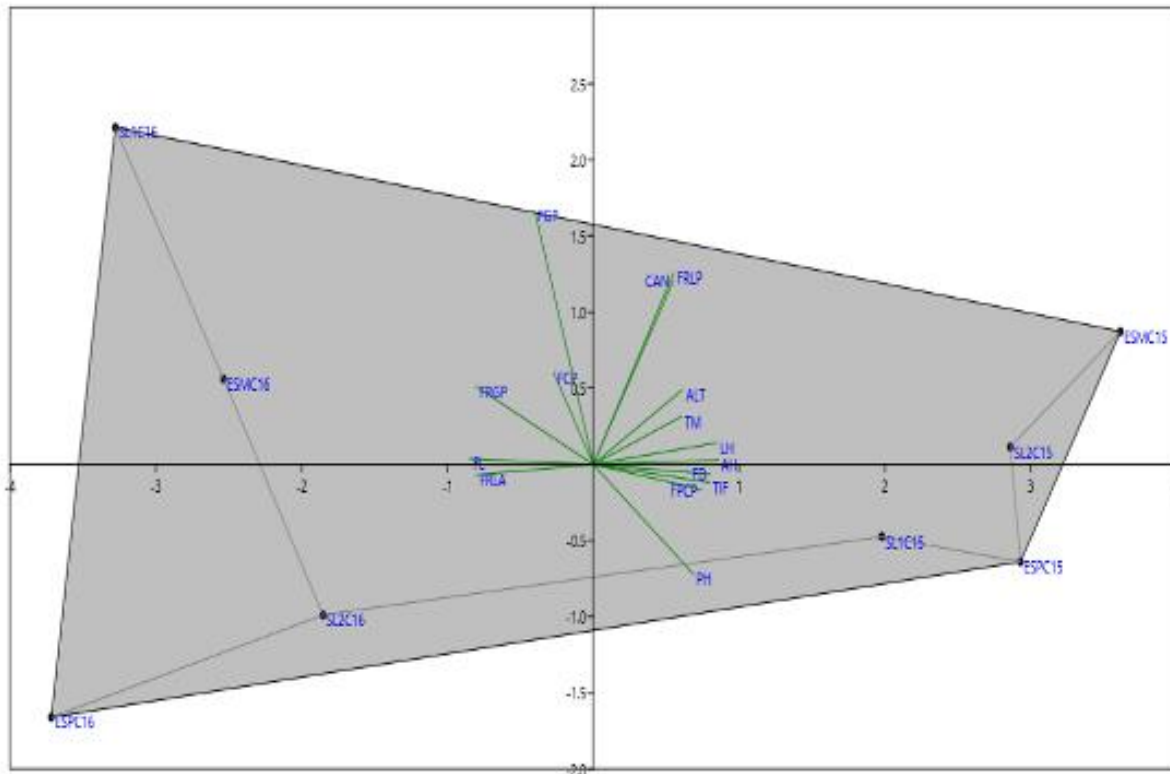


Figure 2. Two-dimensional plot of CP1 and CP2 in ancho pepper populations. Cycles 2015-2016.

Figure 2 shows the formation of two groups on different axes of the two principal components. The first group was formed by the Esmeralda (ESMC15) and San Luis 2 (SL2C15) populations of the

2015 cycle, and the variables positively related to the first axis were anthocyanin color at the node level (CAN), fruit stalk length (FRLP), plant height (ALT), ripening time (TM), leaf length (LH) and leaf width (AH). The second axis shows the Special (ESPC15) and San Luis 1 (SL1C15) populations of the 2015 cycle, explained by the variables TIF, FD, FPCP and PH. The second group is found in the third axis of the two principal components, where the San Luis 1 (SL1C16) and Esmeralda (ESMC16) populations of the 2016 cycle stand out with the variables FGP, FCP, FRGP and FL. The fourth axis is negatively linked, and the Special (ESPC16) and San Luis 2 (SL2C16) populations of the 2016 cycle are found with the variable FRLA. According to López et al. (2016), the predominant shape of the longitudinal section is ongular and angular with a strong transverse undulation, as well as the predominant characteristic of the presence of the peduncular cavity of medium depth in ancho poblano pepper hybrids.

CONCLUSIONS

The CP analysis for the pooled data from the 2015 and 2016 evaluation cycles highlights two principal components that explain 74.79% of the total variability.

The characteristics that contributed most to the total variation among the populations studied were mainly those related to leaves and fruits.

In the 2015 and 2016 cycles, with respect to the two-dimensional plot of CP1 and CP2, two groups were obtained, distributed in 4 axes. On the first axis were the Esmeralda (ESMC15) and San Luis 2 (SL2C15) populations, explained by the fruit, leaf and plant height variables. On the second axis, the Special (ESPC15) and San Luis 1 (SL1C15) populations were found with the flowering variables. The second group, on the third axis, included the populations San Luis 1 (SL1C16) and Esmeralda (ESMC16) explained by the fruit depth variables, and on the fourth axis, the populations Especial (ESPC16) and San Luis 2 (SL2C16) and the fruit length/width ratio variable.

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Index

- A**
Agroecology, 97
antimicrobial, 17, 18, 20, 22, 23
antioxidant, 17, 18, 21, 23
- B**
Biomass, 83, 84, 85
- C**
Capsicum, 27, 28, 32
Carbon, 83, 85, 86, 87
- E**
extracts, 6, 7, 8, 9, 10, 11, 12, 13
- F**
flavonoids, 6, 7, 9, 10, 12, 13
- G**
Growth, 69, 71, 75, 76
- H**
hydroalcoholic extracts, 17
- L**
Larrea tridentata, 17
leaves, 17, 18, 21, 26
Lipids, 42
- M**
Maturation time, 33
mortality, 46, 47, 50
- O**
Oreochromis niloticus, 69, 70
- P**
Parkinsonia, 6, 7, 9, 11, 13
Peduncular cavity of fruit, 33
Probiotics, 69, 70
- S**
shrimp, 46, 47, 48, 49, 50
syndrome, 46, 47, 48
- T**
Two-dimensional plot, 33
- W**
Water, 91
Water quality, 69, 71
Windbreaks, 97, 98

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