Ecological Indicators 121 (2021) 107174

Contents lists available at ScienceDirect

Ecological Indicators

journal homepage: www.elsevier.com/locate/ecolind

A protocol for health assessment of cacti populations: A case study from Northwestern Argentina

María Florencia Barbarich [a,b], Florencia Otegui [b,c,d], Alejandro Saint Esteven [e,f], Ignacio M. Soto [b,e,f], Laura Varone [b,g,*]

[a] Laboratorio de Arquitecturas Andinas y Construcción con Tierra, Universidad Nacional de Jujuy, Argentina
[b] Consejo Nacional de Investigaciones Científicas y Técnicas, Argentina
[c] Departamento de Biodiversidad y Biología Experimental. Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Argentina
[d] Instituto de Micología y Botánica, CONICET, Buenos Aires, Argentina
[e] Departamento de Ecología, Genética y Evolución. Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Argentina
[f] Instituto de Ecología, Genética y Evolución de Buenos Aires- CONICET, Argentina
[g] Fundación para el Estudio de Especies Invasivas, Hurlingham, Buenos Aires, Argentina

ARTICLE INFO

Keywords:
Columnar cacti
Health status
MRT
Deterioration
Conservation

ABSTRACT

Argentina is considered megadiverse for Cactaceae, and Jujuy province, with a high proportion of cacti endemism, is a critical region for their preservation. Cacti deterioration is mainly associated with habitat degradation, agricultural frontier advance, urbanization, illegal collection and trade. Of the three species of columnar cacti within *Trichocereus* genus that are present, *Trichocereus atacamensis* represents a valuable species for local communities. In the last decades, detrimental effects on *T. atacamensis* populations increased despite the reduction in the use of timber for construction or handicrafts. Knowledge on the health of cacti populations is fundamental for conservation and resource management, therefore the objective of the present study was to develop a health status classification criterion for cacti in Quebrada de Humahuaca, Jujuy. A generic method was elaborated to assess the health of cacti populations to facilitate and speed-up the decision-making process for conservation purposes. The evaluation of factors involved in the deterioration is very complex. We designed a method considering aspects that the local residents identified as the most relevant: a) presence of air plant (genus *Tillandsia*), b) larvae of *Cactoblastis bucyrus* and c) signs of rot. Band transects were sampled in patches of *T. atacamensis*, surveying these aspects for each individual. A Multivariate Regression Tree analysis (MRT) was conducted. According to this clustering, an accessible assessment protocol with a 5-level health status was established to classify the cactus as optimal, suboptimal, compromised, vulnerable or critical. The general health status of the patches was highly variable. This MRT analysis is potentially applicable to any columnar cacti population and to establish the basis of a decision protocol for health status assessment. It is evident that the situation of *T. atacamensis* transcends as a local problem and exceeds the presence of pathogens suggesting a multifactorial causality that needs special attention.

1. Introduction

Cactaceae is an important plant family endemic to the Americas which includes around 200 genera and 1600–2500 species (Kiesling, 1975; Anderson, 2001; Barthlott and Hunt, 1993; Gibson and Nobel, 1986). Its distribution ranges from southern Canada (Speirs, 1982), south to Patagonia in Chile and Argentina (Kiesling, 1988). Argentina is considered a megadiverse country for Cactaceae, a priority center to undertake conservation actions, along with Mexico, Brazil and Bolivia (Ortega-Baes and Godínez-Alvarez, 2006; Ortega-Baes et al., 2010). There are about 36 genera and between 200 and 300 species in Argentina, 50% of which are endemic (Kiesling 1975, Ortega-Baes et al., 2010). The northwest province of Jujuy has the second highest rate of cacti endemism of all Argentine provinces; it is a critical region for Cactaceae preservation (Ortega-Baes et al., 2010, 2015). Several species of cacti have been listed as threatened and have been incorporated in the CITES (2007) (Convention on International Trade in Endangered Species of Wild Fauna and Flora) as a species at risk of extinction (Oldfield,

* Corresponding author at: FuEDEI (Fundación Para el Estudio de Especies Invasivas), Bolívar 1559, Hurlingham (1686), Buenos Aires, Argentina.
E-mail address: lauvarone@fuedei.org (L. Varone).

https://doi.org/10.1016/j.ecolind.2020.107174
Received 2 March 2020; Received in revised form 13 October 2020; Accepted 9 November 2020

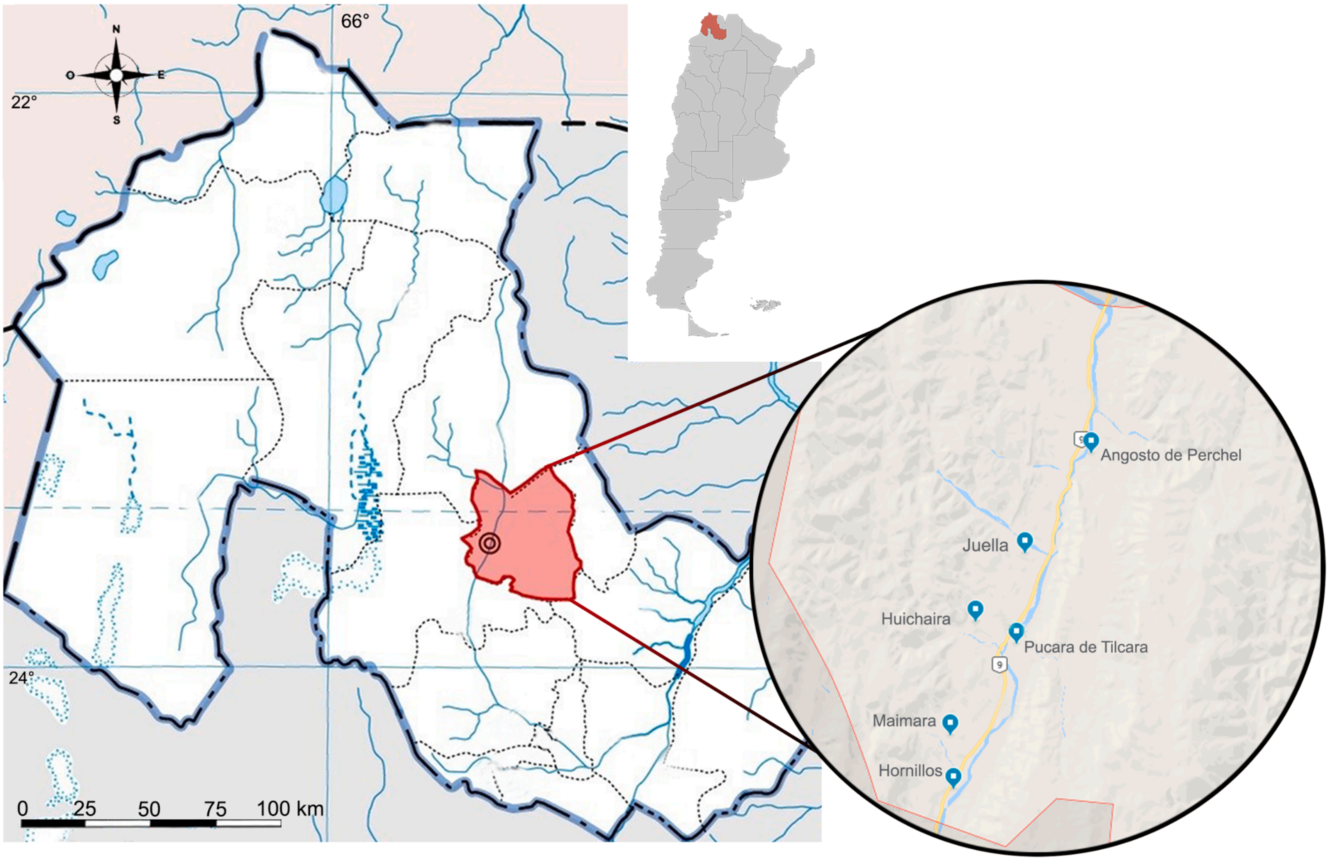

Fig. 1. Research area at Tilcara Department, Jujuy Province, Argentina. The 6 patches (forests) of *Trichocereus atacamensis* studied were located throughout the Quebrada de Humahuaca.

1997; Ortega-Baes et al., 2010). Their deterioration is associated with habitat degradation, agricultural activities, urbanization, mining, collection and illegal trade for ornamental purposes, and potentially climate change (Ortega-Baes et al., 2010).

Trichocereus is a widely distributed Cactaceae genus in northwestern Argentina. It is a tree-like, decumbent, tree-climbing and creeping species, with three iconic species of columnar habit of considerable size: *Trichocereus atacamensis* (Phil.) Backeb., *T. terscheckii* (Parm. ex Pfeiff.) Britton & Rose, and *T. tarijensis* (Vaupel) H. Friedrich & G. D. Rowley (Kiesling, 1978). *Trichocereus atacamensis* is endemic to the Prepuna region and is commonly named "cardón" by the local people ("cardones" is plural); where the term "cardonal" is used to name a patch of cardones or forest.

Trichocereus atacamensis is a valuable species for communities in the region of Quebrada de Humahuaca (Jujuy Province). Cacti have multiple uses and symbolic roles for local residents, who have become aware of the decrease in the health, and the vulnerability, of this emblematic plant in the last two decades (Barbarich and Suárez, 2018). UNESCO declared this region a World Natural and Cultural Heritage in 2003, prompting an increase of tourism that propelled reclassification of land use and changes in local economy (Troncoso, 2009). The Quebrada de Humahuaca experienced an increase in human population growth, followed by higher waste disposal, and intensification of agriculture practices, including the use of agrochemicals (Troncoso, 2009; INDEC, 2010; Reboratti et al., 2003; Salleras, 2011). In addition, Evans et al. (1994) reported surface damage to several cacti of Northwestern Argentina, including a hardening of the epidermis evidenced by the change of color of the skin from green to brown (probably linked to a process of senescence), increased epicuticular wax deposits, and a loss or blockage of the stomata. Detrimental effects on *T. atacamensis* individuals have also been reported by human disturbance despite the reduction in their use as timber for construction or handicrafts (Pinto and Moscoso, 2004).

The evaluation of human, climatic, and biological factors involved in the detriment of *T. atacamensis* is complex. However, previous studies highlighted three factors mentioned by local residents as the most relevant associated with the poor health of *T. atacamensis* (Barbarich and Suárez, 2018): a) surface coverage by epiphytic plants (genus *Tillandsia*), b) feeding by larvae of *Cactoblastis bucyrus* Dyar (Lepidoptera: Pyralidae), and c) signs of internal plant rot associated with changes of color, injuries, presence of other insects, etc. Although epiphytic *Tillandsia* spp. (Bromeliaceae) seem to form a commensalism relationship (Begon et al., 2006), the *Tillandsia* spp. plants are affecting *T. atacamensis* ecophysiology by competing for light and creating a microclimate favorable to other organisms (Flores-Palacios, 2016; Stanton et al., 2014; Callaway et al., 2002) which could be detrimental. *Cactoblastis bucyrus* is a cactophagous lepidopteran specific to *Trichocereus* (Arce de Hamity and Neder de Román, 1999). Mated females lay eggs in a structure called an "egg stick" that contains between 70 and 90 eggs, that hatch simultaneously and larvae penetrate gregariously into the plant. Larvae complete their development building galleries as they feed on soft interior tissues, and discard their feces outside the plant. Last instar larvae leave the plant, spin a cocoon in ground debris and pupate (Arce de Hamity and Neder de Román, 1999). While the congeneric species *C. cactorum* has been widely studied and is known both as an *Opuntia* pest where the genus is native (Varone et al., 2014, 2019), as well as a biological control agent where *Opuntia* are invasive (Winston et al. 2014), little is known about *C. bucyrus* attacking *T. atacamensis,* both native species to Argentina. Finally, widespread rot injuries have not previously been reported in *T. atacamensis,* but work related to culturing this plant mentioned bacterial and fungal pathogens as leading factors of different types of rot, including a 'dry rot'

Table 1
Traits of the six *Trichocereus atacamensis* study sites.

Patch	Hornillos	Maimara	Huichaira	Pucara de tilcara	Juella	Angosto de perchel
Elevation at SW corner (m.a.s.l)	2388	2389	2589	2560	2545	2588
Area (ha)	27.4847	14.1698	0.8344	5.3909	3.1276	3.0448
Perimeter length (m)	2269	1659	680	1000	717	734
Number of transects	10	6	6	5	3	5
Environmental features	Located on Rio Grande margin. Active open-air dump present over a decade	Protected environment by topographical characteristics. Located on an alluvial ejection cone.	Peri urban site located in a transversal ravine to the Quebrada de Humahuaca. Fragmented by roads, animal pens and houses.	Inside an exploited archeological site, on a sedimentary relief of an alluvial terrace, south of the Rio Grande and Rio Huasamayo confluence.	Plants are limited and site occupied by roads, a cemetery and homes. Located in a transverse ravine to Quebrada de Humahuaca.	Access restricted because location is on communal lands. Occurs in the narrowest section of the Quebrada de Humahuaca, in a repaired environment.

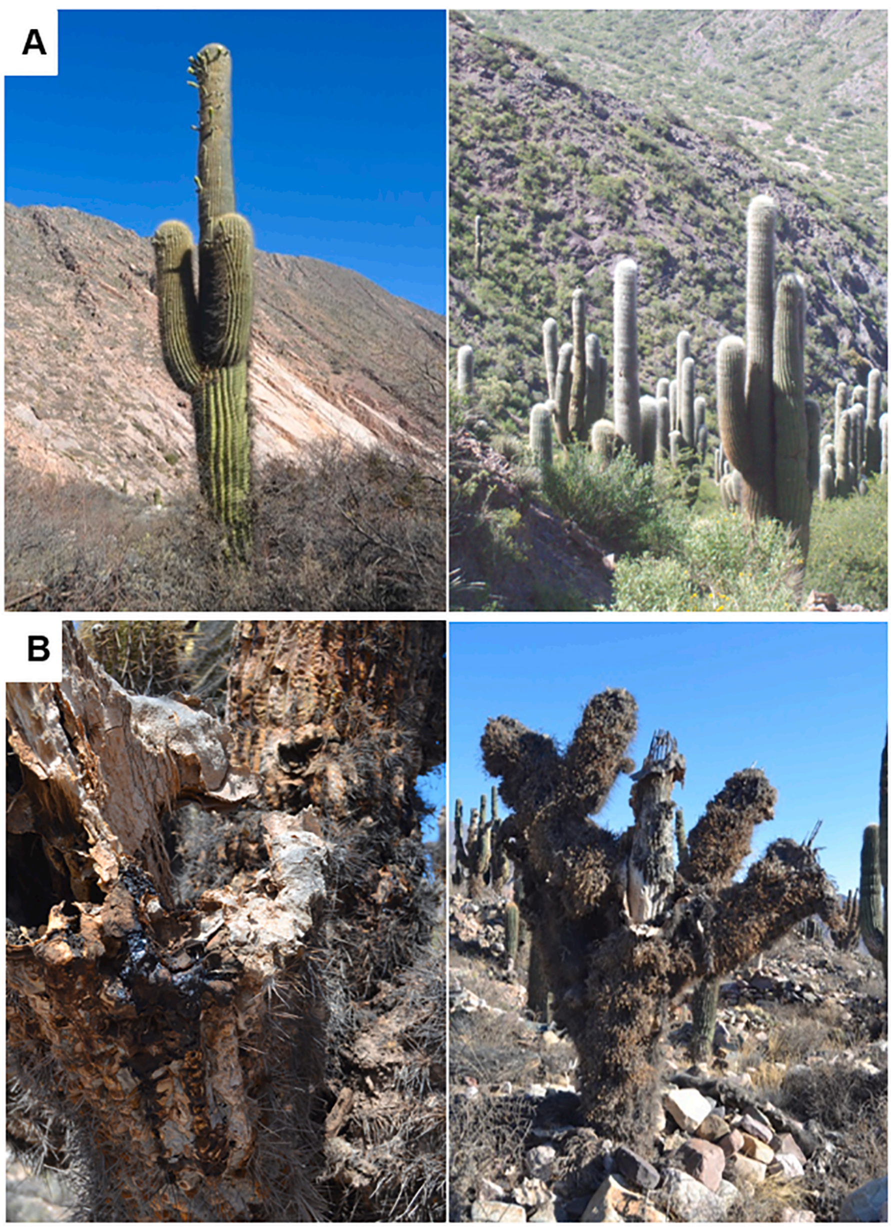

Fig. 2. *Trichocereus atacamensis* A) without signs of damage, and B) showing brownish rotten tissues critical at the Quebrada de Humahuaca research area (Photos: MF Barbarich).

distinguished by a complete dehydration and drying of the standing plant, and a "soft rot" distinguished as a process of degradation of all tissues (Arredondo Gómez, 2002).

Knowledge on the populations' health status of *T. atacamensis* is fundamental to develop conservation and resource management tactics, and is required to identify factors to take medium and long-term actions. Many authors have highlighted the botanical importance of the species by documenting reproductive characteristics, distribution and association with other species (Cazón et al., 2002; de Viana, 1996; de Viana et al., 1990, 2001; Font and Picca, 2001) yet health assessment of the species has not been studied. The objective of this study was to develop a classification criterion for the health status of *T. atacamensis* in Quebrada de Humahuaca, province of Jujuy, Argentina, considering the three factors that the inhabitants of the region suggested as the main causes of the deterioration. Furthermore, a generic method to elaborate a suitable protocol for health assessment of cacti populations is proposed to facilitate and speed-up the decision-making process for conservation purposes.

2. Materials and methods

2.1. Study area

Tilcara is one of the 16 departments of Jujuy Province, located in northwestern Argentina (Fig. 1), and covers an area of 1485 km^2, including a part of the Quebrada de Humahuaca, a basin north–south tectonic depression. The climate is characteristic of high altitude tropical deserts, cold and dry: rainfall of 80 to 400 mm per year, with wide variations of daily temperatures from 0 to 30 °C, the annual rainfall and precipitation means are 140 mm and 12,5°C respectively (Buitrago and Larrán, 1994; Buitrago, 2000). It belongs to the Prepuna phytogeographical province (Chaco Domain), commonly named *Quebrada*, with a vegetation dominated by xerophytic shrubs and an important presence of Bromeliaceae and Cactaceae (Cabrera, 1976). *Trichocereus atacamensis* is distributed throughout the *Quebrada* but have clear areas of clustering with variable density of individuals. For this study, six accessible patches of *T. atacamensis* were selected in the Tilcara department (Table 1). These patches were located along the banks of the Río Grande river and were named according to the name of their closest settlement (Fig. 1). Elevation, area, perimeter length and environmental characteristics were determined for each site (Table 1).

2.2. Sampling

A systematic sampling by parallel band transects was conducted, tracing west-to-east, 100 m long and 10 m wide. The first band transect was positioned 50 m north to the southwestern-most extreme limit of the *T. atacamensis* forest, and each subsequent transect was located every 50 m in a northern direction until the whole area was covered. Therefore, the total number of transects depended on the total area of the forest (Table 1).

The number of individuals present per band transect was recorded to estimate the abundance and density of *T. atacamensis* in each forest. To ensure correct identification of the species, only specimens equal to, or taller than, 50 cm were considered. The health status was assessed by sampling 5 individuals in each band transect, while walking along the central axis in a west to east direction and systematically selecting the closest individual every 20 m. If the number of individuals in a band transect was equal to, or<5, all the plants were sampled. Three health proxies were recorded for each sampled plant.

2.2.1. Coverage of Tillandsia air plants (Ap)

Four variables were measured by dividing the surface of each specimen in quarters according to cardinal orientation (N, E, S and, W), and the surface covered by *Tillandsia* was eyeball determined, always by the same researcher. This allowed to evaluate if there was a preferred orientation. Each cardinal face was assigned to one of five levels according to the percentage of *Tillandsia* spp. coverage on the surface of the *T. atacamensis* individuals: "Ap 1" = absence of air plant, "Ap 2" = 1 to 25%, "Ap 3" = 26 to 50%, "Ap 4" = 51 to 75%, or "Ap 5" = 76 to 100%.

Rot injuries (Ri). The observations of rot injuries were conducted at each cardinal orientation, and also by subdividing the individual vertically in three even sections: "low", "middle" and "high". Consequently, 12 measurements were obtained from each specimen. Rot injuries were considered when clear signs of decaying tissues such as browning, softening and dripping were observed. Both dry and soft rot were considered (Fig. 2). The surface of rot injuries was estimated and assigned to one of five levels: "Ri 1" = absence of rot injuries, "Ri 2" = 1to 25%, "Ri 3" = 26 to 50%, "Ri 4" = 51 to 75%, or "Ri 5" = 76 to 100%.

Table 2
Trichocereus atacamensis abundance and density at the six study sites. Means and confidence intervals (CI) are shown for each study site.

Cardonal	Abundance			Density m^{-2}		
	CI low	Mean	CI high	CI low	Mean	CI high
Hornillos	460.92	714.60	1035.35	16.77	26.00	37.67
Maimara	1211.66	1629.53	2700.06	85.51	115.00	190.55
Pucara de Tilcara	613.11	797.85	989.55	117.44	148.00	183.56
Angosto de Perchel	276.56	438.45	747.07	90.83	144.00	245.36
Juella	155.56	239.78	354.96	49.74	76.77	113.49
Huichaira	20.08	38.94	69.41	24.07	46.67	83.19

2.2.2. Cactoblastis bucyrus damage (Cb)

Damage from *C. bucyrus* was determined by observation and each individual plant was assigned to one of six insect categories: "Cb 1" = absence of damage, "Cb 2" = presence of attack signs of undetermined age, "Cb 3" = presence of egg sticks and/or dry frass, "Cb 4" = presence of egg sticks and small amount of fresh frass, "Cb 5" = presence of feeding galleries and high amounts of fresh frass, or "Cb 6" = presence of feeding galleries, high amounts of fresh frass and direct observation of larvae.

2.3. Data analysis

Sub-population density, abundance estimates and confidence intervals were calculated for each *T. atacamensis* forest (Krebs, 1999). The independence of the 17 variables measured to assess the cactus health (four from coverage of air plant, twelve from rot injuries and one from *C. bucyrus* damage) was determined by an ANOVA test. The correlation of the new defined variables was analyzed with Pearson index ($p < 0.05$). In addition, cluster analysis was performed considering the new combined variables with the Multivariate Regression Tree method (MRT) (De'Ath, 2002), an extension of the Classification and Regression Trees to multivariate response data (Legendre and Legendre, 2012). The MRT method allowed an index construction by the ordination of a data set through the recursive partitioning of a quantitative variable under the control of a set of quantitative or categorical explanatory variables. The MRT method has no limitations in the number of possible categories a data set can be partitioned, although the final size of the tree has an effect in the predictive error of the method. The relevance of this effect was evaluated by the cross validated relative error test (CVRE) (De'Ath, 2002; Legendre and Legendre, 2012). According to the clustering, and to elaborate an accessible assessment protocol, a 5-level health status of the individual cactus was established. The MRT and CVRE analyses were performed using the "MVpart" package in R environment (De'ath, 2014; R Core Team, 2018).

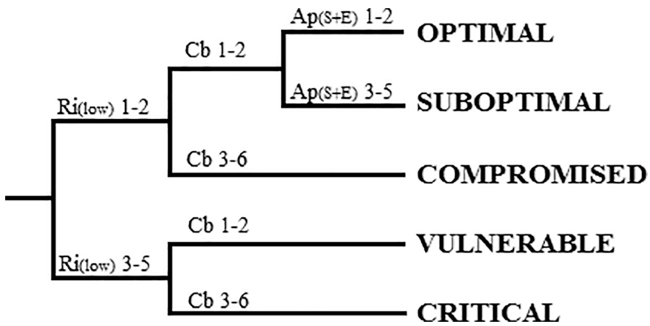

Fig. 3. Health status categories of *Trichocereus atacamensis* determined by a pruned multivariate regression tree (MRT). The MRT contained five leaves which were obtained in the analysis using five variables for the partitions. The decision rule used in the partition of the levels was placed above each leaf. The three variables included in the output designed by the analysis were rot injuries occurring low on the plant (Ri L), damage from the insect *Cactoblastis bucyrus* (Cb), and surface coverage by bromeliads on the south and east cardinal face of the plant (Ap S-E). (Error: 0.341; CV Error: 0.373; SE: 0.0319).

3. Results

The abundance and density of individuals in each *T. atacamensis* forest was heterogeneous (Table 2). The forest that presented the highest abundance was Maimara, almost double of the second most abundant, Pucara de Tilcara. The latter also showed the highest density. In contrast, the Huichaira forest had the lowest abundance, while the lowest density was estimated at Hornillos.

A total of 137 *T. atacamensis* individuals were sampled in the 6 locations and measured for their health proxies. Estimation of health status of cacti based on coverage of air plants, rot injuries and *C. bucyrus* damage resulted in 17 measured variables that were summarized according to their codependence, namely: coverage of air plant on the area facing north and west (Ap N-W), or on the south and east faces (Ap S-E); percentage of rot injuries at the high and medium level (Ri H-M), or at the lower level (Ri L); and presence of *C. bucyrus* (Cb). The paired correlations between the variables showed that Ap S-E was correlated with Ri H-M ($Ps = 0.22$, $p < 0.05$), and Ap N-W was correlated with Ri L ($Ps = 0.22$, $p < 0.05$). The two variables of rot injuries (Ri H-M and Ri L) correlated with Cb ($Ps = 0.46$ and $Ps = 0.24$, respectively, both $p < 0.05$). The mean coverage of air plant for all specimens studied was 2.16 $\pm$ 0.37% for the N-W cardinal face, and 23.04 $\pm$ 1.46% for the S-E cardinal face. Also, the variable Ap S-E correlated with Cb ($Ps = 0.29$, $p < 0.05$).

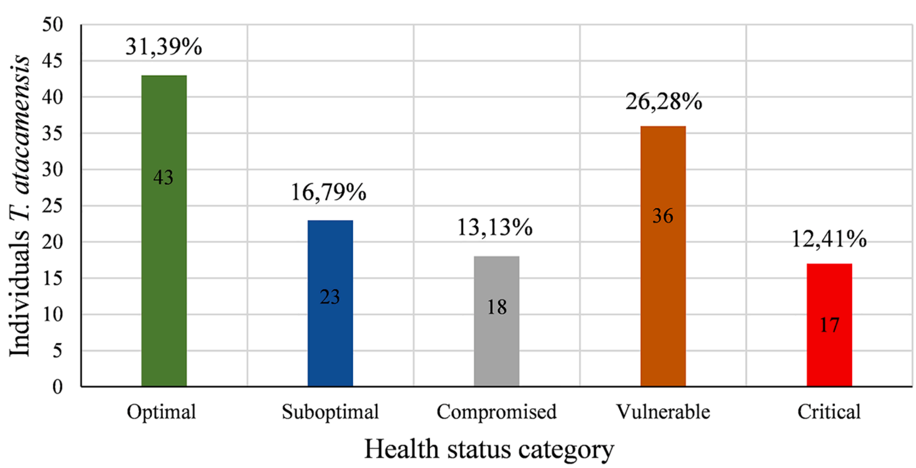

Fig. 4. Distribution percentage of the 137 *Trichocereus atacamensis* in the five health status categories in Tilcara Department, Jujuy Province, Argentina.

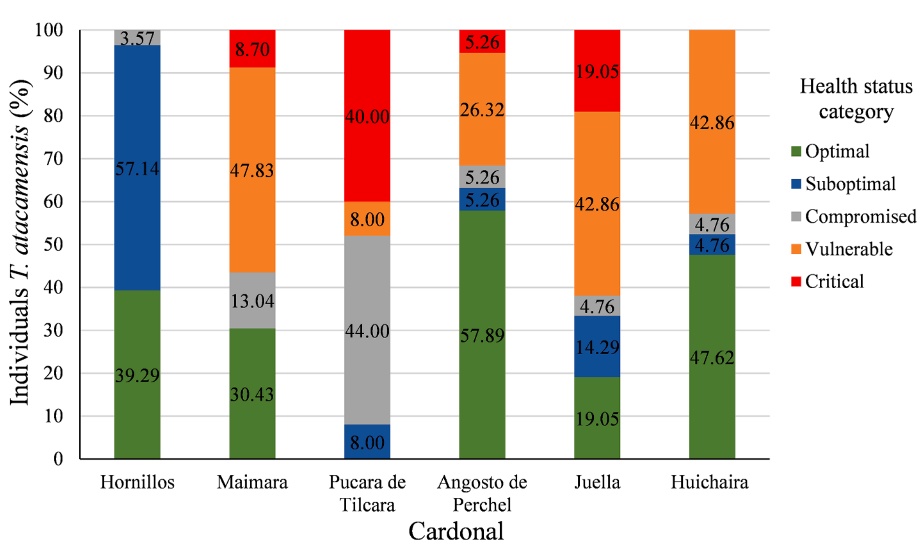

Fig. 5. Individuals of *Trichocereus atacamensis* in the 5 health status categories defined for the six patches in the Tilcara Department, Jujuy Province, Argentina.

The MRT cluster analysis produced a 3-step decision tree based on the level of rot injuries in the lower section, *C. bucyrus* damage, and the presence of air plant in the south and east faces, which was nested within the lowest levels of *C. bucyrus* damage (Fig. 3). Middle and upper rot injuries and the north and west air plant coverage were discarded by the analysis. According to the composition of each leaf in the tree, 5 hierarchical health status categories were defined (Fig. 3). Specimens in "optimal" and "suboptimal" categories presented the lowest values for the Ri L and *C. bucyrus* presence, and they differed in the Ap S/E coverage, being in optimal condition when the cacti were covered with the least amount of air plants. The middle category, named "compromised", included the lowest values of low rot injuries and highest values of *C. bucyrus.* Finally, the categories with the highest values of low rot injuries were deemed "vulnerable" and "critical", where vulnerable had relatively the lowest infestation of *C. bucyrus* and critical had the highest infestation.

The general distribution of the 137 specimens in the health status categories showed the highest number of individuals in optimal and vulnerable condition (Fig. 4). In addition, the general health status of the individuals was highly variable and each forest was analyzed based on the proportion of individuals in each health category (Fig. 5). Cacti at the site Pucara de Tilcara were in the worst health condition with the majority of plants identified as critical or vulnerable; none were in optimal condition. Pucara de Tilcara contained the largest number of dead specimens that had fallen. Hornillos and Huichaira did not contain individuals in critical state, while this category was widely represented in Pucara de Tilcara, where none was classified as optimal. Additionally, the individuals in suboptimal conditions were mostly found in Hornillos. In the case of Maimara, vulnerable health status was the most common. All the patches showed individuals in compromised status in different proportions.

4. Discussion

Our study highlights the health status of *T. atacamensis* in a region of northwestern Argentina. Among the many variables that could be linked to the health status of these columnar cacti, we focused on three critical issues identified by local inhabitants: rotting of plants, surface coverage by bromeliads, and attack by a host specific internal feeding insect (Barbarich and Suárez, 2018; Barbarich, 2019). While the percentage of rot was similar on all cardinal faces of a plant, the highest percentages occurred in the lowest section. The increased moisture levels in the soil and around the base of the plant would support colonization and growth of rot-associated fungi and bacteria. Despite not having been studied, it is important to highlight that the existence of open garbage dumps in the area could be a source of new pathogens, or their growth in abundance. The higher percentage of bromeliad coverage on the south and east faces reported by the inhabitants (Barbarich and Suárez, 2018) is supported by our survey. This finding could be related with the humidity-laden winds that rise upslope along the Quebrada de Humahuaca valley from the south, or differential sun exposure that confers photosynthetic advantages, as previously reported (Sanger and Kirkpatrick, 2015; Buitrago and Larrán, 1994).

All dead and fallen cacti found during the surveys were completely covered by bromeliads, suggesting that full plant coverage by these epiphytic plants might lead to death. The invasion of this plants could be due to the increased human activity producing quantity of atmospheric dust, which provides nutrients to plants. However, further research would be required to clarify any potential cause-effect relationship. The correlation found between bromeliad coverage on the S-E cardinal faces, the rot in the high-medium level and the presence of *C. bucyrus* may be driven by the habits of this insect. Female moths may oviposit preferentially in younger cactus tissues that are easier for larvae to penetrate and become established. Bromeliad coverage might also favor the establishment of *C. bucyrus* either because adults and egg sticks find refuge, or because physiological variations are produced that favored larval development. Although it was suggested that *C. bucyrus* was responsible for triggering infections of rot (Arce de Hamity and Neder de Román, 1999), the direct linkage of these factors has not been studied and we found evidence that browning could occur without the presence of *C. bucyrus* (and *vice versa*). This is the first systematic survey of the presence of *C. bucyrus* attacking *T. atacamentsis,* so the effect on survival is still unknown and further research is needed.

Pucara de Tilcara was the *T. atacamensis* forest with the worst health status, with high numbers of rotten cacti. In an ethnobotanical survey, the local residents identified two causes of death, the "natural" death that involves mostly dry and dead-standing plants, but they also acknowledged that in the last two decades it has become more common to see plants fall in the ground with signs of rot (Barbarich and Suárez, 2018). The unfavorable condition of Pucara de Tilcara may be related to the permanent presence of people, since it is a popular archaeological tourist site, which could drive away birds that predate on *C. bucyrus* larvae. Also, the proximity to open-pit garbage and the impossibility of removal of dead cacti because it is an enclosed, protected and sacred archaeological site, both could be acting as a source of pathogens. Despite specimens at archeological sites being higher and older (Halloy, 2008), the higher proportion of dead cacti at this site could be related to ageing, however this does not explain the general increase in dead and damaged cacti reported by the inhabitants (Barbarich and Suárez, 2018).

The second worst forest was Juella, with a large number of specimens classified in the vulnerable state. This site had less movement of people than Pucara de Tilcara, but it is currently immersed in a population growing zone with increased pressure from tourist activities. The overall health conditions of the cacti at Maimara and Huichaira were better. Maimara was located close to the road and on a shear cone that might confer protection from winds by the surrounding mountains. Both Huichaira and Angosto de Perchel contained the highest number of individuals in optimal or vulnerable conditions and were located farthest from the road. Considering the growth of the region, the urbanization progress, and the presence of an open-pit dump at the entrance to Huichaira, this area should be considered for special protection, because it contained the highest number of juvenile *T. atacamensis*. Field observations showed that in Angosto del Perchel there was a greater number of young plants, followed by Maimara, both sites greatly more noticeable from the rest. Although young replacement plants were not counted in our study, it is important that future studies investigate the renewal rate under natural settings given the low germinability of seeds, their difficulty to establish in the ground (de Viana et al., 1990), and the dependence of juveniles on nurse plants (de Viana et al., 2001a,b).

Our field observations agreed with those made by local inhabitants (Barbarich and Suárez, 2018; Barbarich, 2019) that revealed a high number of *T. atacamensis* in deteriorated conditions. Local people ensured that the measured symptoms were not manifested with such severity some decades ago, and the oldest inhabitants pointed out that most of the *T. atacamensis* forests used to look like the healthy ones of nowadays. Because there is a lack of historical estimates on the health status of *T. atacamensis* populations, the local knowledge was the main source of data, and the current study could be used as a baseline for future estimates. We would be facing a conservation problem since only 48% of the specimens are in optimal or suboptimal condition. In agreement with the Convention on Biological Diversity, only by understanding the human relation with nature we can take helpful steps to conserve biodiversity (Brown, 1994). Particularly, the specimens categorized with the worst health status need to be further studied to determine the organisms that are causing damage and rot injuries and even leading to death.

Several reports from around the world on other columnar cacti have mentioned death or decay of plants from unknown causes (Bashan et al., 1995; Espinosa, 1993; McAuliffe, 1993). In Mexico, an entire population of *Pachycereus pringlei* Britt. & Rose was reported to have a browning or whitening that ended in epidermis rupture, which bears similarity to our

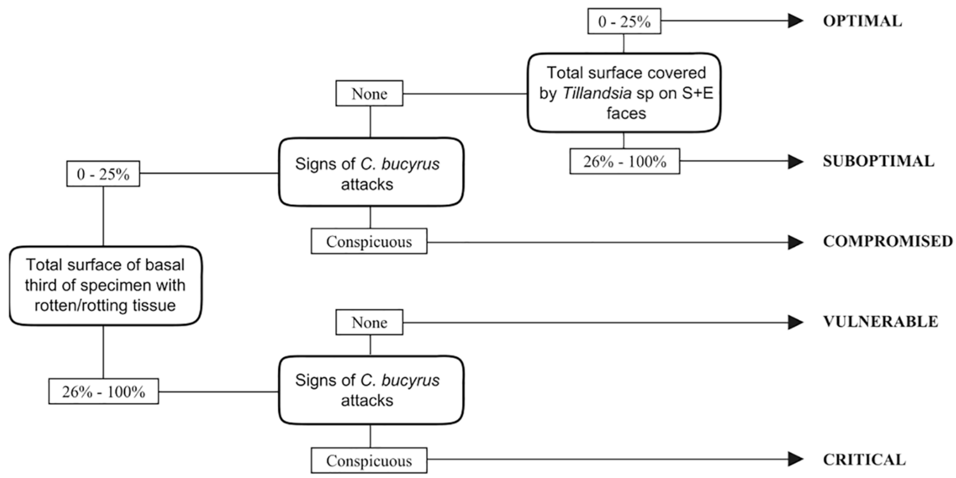

Fig. 6. Flowchart based on MRT output, proposed as an extensive health assessment tool for *Trichocereus atacamensis* populations.

field observations (Bashan et al., 1995) and a similar phenomenon was reported in the Sonoran Desert for four species of columnar cacti (Evans et al., 1994). These reports could be pointing out a widespread, worldwide problem in columnar cacti likely caused by environmental factors such as damage to the ozone layer, pollution, bacterial necrosis, and climate change with long precipitation periods or extreme temperatures (Krantz, 1992; Pacenti, 1993). Given that plant species are often tolerant to common pathogens in their home range, these new (or increased) symptoms over the past decades (Barbarich and Suárez, 2018) could indicate the arrival of new pathogens to the area (Bashan et al., 1995), or changes in environmental conditions. While the present work has not identified the species of pathogens present in *T. atacamensis*, there are at least 28 groups of bacteria and 20 groups of fungi related to necrosis in other cacti (Fogleman and Foster, 1989; Foster and Fogleman, 1993, 1994; Mongiardino Koch et al., 2015) that deserve further study. The form of growth and longevity of *T. atacamensis*, as well as their location in ruins, can serve as indicators of past climatic conditions and land use (Halloy, 2008). Thus, the loss of these columnar cacti would constitute a loss in biodiversity and anthropological information. The importance of *T. atacamensis* populations in northwestern Argentina is emphasized by the cultural value and the relevance of the area being declared as a site of Natural and Cultural Heritage of Humanity.

The MRT analysis proved to be an adequate approach to address the health status of *T. atacamensis* forests at the Quebrada de Humahuaca, and can be used to determine the health of other columnar cacti populations. Furthermore, this methodology could be extended to specimens other than cacti if the ecological sampling were duly adjusted based on the researchers̀s̀ previous knowledge. The pertinence of this method resides in three features. First, it is a robust type of analysis that supports the simultaneous use of numerical, categorical and ordinal variables. This allows the customization of the study to suit the attributes of any particular cacti population. Secondly, MRT methods tolerate missing data. In this study, for instance, the complete coverage of bromeliads on the surface of a few individual plants prevented the ability to identify rotten tissue or *C. bucyrus* damage. This impediment was overcome by the use of MRT analysis, although a mild underestimation of health status in these specimens cannot be ruled out. Thirdly, MRT methods have predictive power because their output is a decision tree that can be used *a posteriori* to assign new elements to the resulting categories of the analysis. A single, systematic field survey followed by an analysis through MRT method can establish the basis of a decision protocol to assess the health status of further individuals. Once this protocol is resolved, future surveys of the population for monitoring are greatly facilitated, given that every new individual observed in the field can be easily characterized. Furthermore, the resulting MRT output can be reformulated in a simple key or flowchart that would also allow its utilization by non-experts, thus setting a customized, straightforward tool for collaboration of institutions and groups that engage in conservation programs (Fig. 6).

5. Conclusions

Likely, the particular situation of the columnar cacti exceeds the presence of bromeliads, *C. bucyrus* and rot injuries, and depends on multiple factors related to the environment. A broad scientific consensus has been reached that human activities have directly modified the composition of the atmosphere and lead to global climate change (Lastra et al., 2008). Each natural population must cope with a particular set of stressing variables depending on the specific environmental and human-related conditions of its location. There must be a continuous evaluation of ecosystem and biodiversity status in order to control deterioration, therefore the search for new and improved tools for health assessment is key. In this scenario, it seems implausible to find a unique evaluation procedure that could fit every population in any given geographical region. However, MRT analysis is an efficient and accessible tool that enables the customization of protocols according to the particularities of the system, data available and crew resources, which can be used in conservation policies. In particular, the current situation with *T. atacamensis* needs special attention, where local campaigns are necessary, but the ultimate solution will only be achieved by global actions to stop environmental deterioration.

CRediT authorship contribution statement

María Florencia Barbarich: Conceptualization, Methodology, Investigation, Writing - original draft. **Florencia Otegui:** Methodology, Investigation, Data curation. **Alejandro Saint Esteven:** Data curation, Software, Formal analysis. **Ignacio M. Soto:** Formal analysis, Supervision, Writing - review & editing, Funding acquisition. **Laura Varone:** Conceptualization, Methodology, Investigation, Writing - review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

We are grateful to Dr. Stephen Hight for reviewing earlier versions of the manuscript, and Arabella Peard for improving the final version. We thank Lic. Carlos Ibarra for collaborating with field trips and sampling efforts, and thanks to Julio Accame for logistic and experimental design support. Research funding was granted to IMS from ANPCyT (PICT 2017-0220) and Buenos Aires University (UBACyT 2018).

References

E.F. Anderson The Cactus Family 2001 Timber Press U.S.A.

Arce de Hamity, M.G., Neder de Román, L.E., 1999. Bioecología de *Cactoblastis bucyrus* (Lepidoptera: Phycitidae), especie dañina al cardón *Trichocereus pasacana* en la Prepuna jujeña (Argentina). Rev. Soc. Entomol. Argent. 58, 23–32.

Arredondo Gómez, A. (2002) Propagación y mantenimiento de cactáceas. Folleto técnico No. 21. Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias, San Luis Potosi, México. 31 pp.

Barbarich, M.F., Suárez, M.E., 2018. Los guardianes silenciosos de la quebrada de Humahuaca: etnobotánica del «cardón» (*Trichocereus atacamensis*, Cactaceae) entre pobladores originarios en el departamento de Tilcara, Jujuy, Argentina. Bonplandia 27, 59–80.

Barbarich, M. F. (2019) Estudio multidisciplinario de los cardones (Trichocereus sp. Cactaceae) de la provincia de Jujuy, Argentina: aspectos etnobotánicos, químicos, nutricionales y sanitarios (phD thesis). Universidad de Buenos Aires. 285 pp.

Barthlott, W. & Hunt, D. (1993) Cactaceae. In Flowering Plants: Dicotyledons (Edited by K. Kubitzki, J.G. Rohwer, and V. Bittrich). 161-197. Springster, Berlin, Heidelberg, Germany.

Bashan, Y., Toledo, G., Holguin, G., 1995. Flat top decay syndrome of the giant cardon cactus (*Pachycereus pringlei*): description and distribution in Baja California Sur, Mexico. Can. J. Bot. 73 (5), 683–692.

Begon, M., Townsend, C.R., Harper, J.L., 2006. Ecology: From Individuals to Ecosystems. Blackwell Publishing, UK.

Brown, K., 1994. Approaches to valuing plant medicines: the economics of culture or the culture of economics? Biodivers. Conserv. 3, 734–750.

Buitrago, G., 2000. El clima de la provincia de Jujuy. de Jujuy, UNJU, S. S.

Buitrago, L.G., Larrán, M.T., 1994. El clima de la Provincia de Jujuy. Facultad de Ciencias Agrarias, Universidad Nacional de Jujuy, Argentina, Cátedra de Climatología y Fenología Agrícola.

Cabrera, A. L. (1976) Regiones fitogeográficas Argentinas. In Enciclopedia Argentina Agrícola y de Jardinería 2nd ed. (1-85). Acme. Buenos Aires, Argentina.

Callaway, R.M., Reinhart, K.O., Moore, G.W., Moore, D.J., Pennings, S.C., 2002. Epiphyte host preferences and host traits: mechanisms for species-specific interactions. Oecologia 132 (2), 221–230.

Cazón, A., de Viana, M.L., Gianello, J.C., 2002. Comparación del efecto fitotóxico de aleloquímicos de *Baccharis boliviensis* (Asteraceae) en la germinación de *Trichocereus pasacana* (Cactaceae). Ecol. Austral 12 (1), 73–78.

Cites, I., 2007. Convención sobre el Comercio Internacional de Especies Amenazadas de Fauna y Flora Silvestres. Apéndice I. Secretaría de Recursos Naturales y Desarrollo Sustentable, Dirección de Fauna y Flora Silvestres. Buenos Aires, Argentina.

de Viana, M., 1996. Distribución espacial de *Trichocereus pasacana* (Cactaceae) en relación al espacio disponible y al banco de semillas. Rev. Biol. Trop. 44 (3), 95–103.

de Viana, M., Acreche, N., Acosta, R., Moraña, L., 1990. Población y asociaciones de *Trichocereus pasacana* (Cactaceace) en Los Cardones. Argentina. Rev. Biol. Trop. 38 (2), 383–386.

de Viana, M.L., Sühring, S., Manly, B.F.J., 2001a. Application of randomization methods to study the association of *Trichocereus pasacana* (Cactaceae) with potential nurse plants. Plant Ecol. 156 (2), 193–197.

de Viana, M.L., Ortega Baes, P., Saravia, M., Badano, E.I., Schlumpberger, B., 2001b. Biología floral y polinizadores de *Trichocereus pasacana* (Cactaceae) en el Parque Nacional los Cardones, Argentina. Rev. Biol. Trop. 49 (1), 279–285.

De'Ath, G., 2002. Multivariate regression trees: a new technique for modeling species-environment relationships. Ecology 83 (4), 1105–1117.

De'ath, G. (2014) mvpart: Multivariate partitioning. R package version 1.6-2. https://CRAN.R-project.org/package=mvpart.

Espinosa, I., 1993. Dying *eulychnias*. Cact. Succ. J. 65, 205–206.

Evans, L.S., McKenna, C., Ginocchio, R., Montenegro, G., Kiesling, R., 1994. Surficial injuries of several cacti of South America. Environ. Exp. Bot. 34 (3), 285–292.

Flores-Palacios, A., 2016. Does structural parasitism by epiphytes exist? A case study between *Tillandsia recurvata* and *Parkinsonia praecox*. Plant Biol. 18 (3), 463–470.

Fogleman, J.C., Foster, J.L.M., 1989. Microbial colonization of injured cactus tissue (*Stenocereus gummosus*) and its relationship to the ecology of cactophilic *Drosophila mojavensis*. Appl. Environ. Microbiol. 55, 100–105.

Font, F., Picca, P., 2001. Natural hybridization between *Trichocereus atacamensis* (Philippi) Marshall and *Denmoza rhodacantha* (Salm-Dyck) Britton & Rose (Cactaceae). Bradleya 19, 59–66.

Foster, J.L.M., Fogleman, J.C., 1993. Identification and ecology of bacterial communities associated with necroses of three cactus species. Appl. Environ. Microbiol. 59, 1–6.

Foster, J.L.M., Fogleman, J.C., 1994. Bacterial succession in necrotic tissue of agria cactus (*Stenocereus gummosus*). Appl. Environ. Microbiol. 60, 619–625.

A. Gibson P. Nobel The cactus primer 1986 (Harvard Un) Cambridge, UK 286 pp.

Halloy, S., 2008. Crecimiento exponencial y supervivencia del cardón (*Echinopsis atacamensis* subsp. pasacana) en su límite altitudinal (Tucumán, Argentina). Ecología en Bolivia 43 (1), 6–15.

INDEC (Instituto Nacional de Estadísticas y Censos). (2010) Censo Nacional de Población, Hogares y Viviendas 2010: Censo del Bicentenario. Pueblos originarios: región Noroeste Argentino. 1a ed. Ciudad Autónoma de Buenos Aires, Argentina.

Kiesling, R., 1975. Los géneros de Cactaceae de argentina. Bol. Soc. Argen. Bot. XV I (3), 197–227.

Kiesling, R., 1978. El género *Trichocereus* (Cactaceae) I: Las especies de la República Argentina. Darwiniana 21, 263–330.

Kiesling, R. (1988) Cactaceae. En M. Correa (Ed.), Flora Patagónica 5, Dicotiledoneas dialipétalas (Oxalidaceae a Cornaceae), 218-243. Colección Científica Del INTA 8. Buenos Aires, Argentina.

Krantz, M., 1992. The case of the disappearing cactus. Audubon 9, 21–23.

Krebs, C.J., 1999. Ecological Methodology, second ed. Benjamin Cummings, Menlo Park, California, U.S.A., p. 620

Lastra, J.A.S., Carmona, M.L., Mendoza, S.L., 2008. Tendencias del cambio climático global y los eventos extremos asociados. Ra Ximhai 4 (3), 625–634.

Legendre, P. & Legendre, L. (2012) Numerical Ecology, third English ed. Elsevier, Oxford, UK. 1006 pp.

McAuliffe, J. R. (1993) Case study of research, monitoring, and management programs associated with the saguaro cactus (Carnegiea gigantea) at Saguaro National Monument, Arizona (No. 48). National Park Service, Cooperative National Park Resources Studies Unit, School of Renewable Natural Resources, the University of Arizona, U.S.A.

Mongiardino Koch, N., Soto, I.M., Galvagno, M., Hasson, E., Iannone, L., 2015. Biodiversity of cactophilic microorganisms in western Argentina: Community structure and species composition in the necroses of two sympatric cactus hosts. Fungal Ecol. 13, 167–180.

Oldfield, S., 1997. Cactus and succulent plants: status survey and conservation action plan. En IUCN/ SSC cactus and succulent specialist group. (Internatio), Gland, Switzerland, and Cambridge, UK.

Ortega-Baes, P., Godínez-Alvarez, H., 2006. Global diversity and conservation priorities in the Cactaceae. Biodivers. Conserv. 15 (3), 817–827.

Ortega-Baes, P., Sühring, S., Sajama, J., Sotola, E., Alonso-Pedano, M., Bravo, S., Godínez-Alvarez, H., 2010. Diversity and conservation in the cactus family. In: Desert Plants: Biology and Biotechnology. Springer, Berlin, Heidelberg, Germany, pp. 157–173.

Ortega-Baes, P., Godínez-Alvarez, H., Sajama, J., Gorostiague, P., Sühring, S., Galíndez, G., Bravo, S., López-Spahr, D., Alonso-Pedano, M., Lindow-López, L., Barrionuevo, A., Sosa, C., Curti, R.N., Juárez, A., 2015. La familia Cactaceae en Argentina: patrones de diversidad y prioridades políticas para su conservación. Bol. Soc. Argent. Bot. 50, 71–78.

Pacenti, J. (1993) Sentinel of the desert is dying. Santa Barbara Cactus and Succulent Society Newsletter, 93(9).

Pinto, R. & Moscoso, D. (2004) Estudio Poblacional De Echinopsis Atacamensis (Cactaceae) en la Región de Tarapacá (I), Norte de Chile. Chloris Chilensis, 7(2).

R Core Team (2018) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Reboratti, C.E., Arzeno, M., Castro, A.H., 2003. Desarrollo sustentable y estructura agraria en la Quebrada de Humahuaca. Población & sociedad 10 (1), 193–213.

Salleras, L., 2011. Territorio, turismo y desarrollo sustentable en la Quebrada de Humahuaca. Paisaje y naturaleza al servicio de la práctica turística. Estud. Perspec. Tur. 20 (5), 1123–1143.

Sanger, J.C., Kirkpatrick, J.B., 2015. Moss and vascular epiphyte distributions over host tree and elevation gradients in Australian subtropical rainforest. Aust. J. Bot. 63, 696–704.

Speirs, D.C., 1982. The cacti of western Canada: Part 3. Nation. Cact. Succ. J. 37 (2), 53–54.

Stanton, D.E., Huallpa Chávez, J., Villegas, L., Villasante, F., Armesto, J., Hedin, L.O., Horn, H., 2014. Epiphytes improve host plant water use by microenvironment modification. Funct. Ecol. 28 (5), 1274–1283.

Troncoso, C.A., 2009. Patrimonio y redefinición de un lugar turístico: La Quebrada de Humahuaca, provincia de Jujuy, Argentina. Est. Perspect. Tur. 18 (2), 144–160.

Varone, L., Logarzo, G.A., Briano, J.A., Hight, S.D., Carpenter, J.E., 2014. *Cactoblastis cactorum* (Berg) (Lepidoptera: Pyralidae) use of *Opuntia* host species in Argentina. Biol. Invasions 16 (11), 2367–2380.

Varone, L., Aguirre, M.B., Lobos, E., Ruiz, D., Hight, S., Palottini, F., Guala, M., Logarzo, G., 2019. Causes of mortality at different stages of *Cactoblastis cactorum* in the native range. Biocontrol 64 (3), 249–261.

Winston, R.L., Schwarzländer, M., Hinz, H.L., Day, M.D., Cock, M.J.W., Julien, M.H., 2014. Biological Control of Weeds: A World Catalogue of Agents and Their Target Weeds, 5th edn. USDA Forest Service, Morgantown, West Virginia U.S.A, p. 838.