

Water Status of *Polylepis* Forests Related to Altitudinal Gradient, Microclimate Variables and Slope

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ABSTRACT

The *Polylepis* forests in the central Andes of Peru remain in hostile environments due to their location above 4000 meters of altitude. They are home to a great biodiversity with a high level of endemism and are extremely vulnerable to climate change and human pressure. Variations in rainfall and temperature have been affecting plant health. These aspects have led to the analysis of the physiological response of plants through water stress and NDVI, in dry periods and related to altitudinal gradients and slope, of five forests located in the regions of Junin and Lima, where the species *Polylepis rodolfo vasquezii*, *P. canoi* and *P. flavilpila* are found. Seven 15 x 21 m plots and 10 sub-plots were established, distributed in the lower, middle and upper parts of the forest. The water potential of leaflets was measured by a Scholander pressure pump in a Pascale unit, microclimate variations by means of installed soil and air humidity and temperature sensors; the NDVI by means of multispectral images captured by an unmanned airborne vehicle. Water stress was different among species and changed according to the altitudinal gradient, soil hydrological variation and temperature. Leaflet size adaptation related to stress changes and a high correlation of NDVI with plant stress status were observed.

Keywords: microclimate, NDVI, *Polylepis*, water status.

INTRODUCTION

The *Polylepis* forests are considered high Andean ecosystems that are home to great biodiversity with a high level of endemism (Zutta *et al.*, 2012), are extremely vulnerable to climate change and human pressure (Esperon & Barradas, 2016). It is the dominant tree genus whose distribution is restricted along the Andes Mountain Range above 3500 m from Venezuela to central Argentina (Kessler and Schmidt-Lebuhn, 2006; Zutta *et al.*, 2012; Zutta and Rundel, 2017), establish favorable microsites for its development, forming an ecological ecotone due to freezing, drying and winds that restrict its development in high Andean ecosystems (Braun, 1997). These forests are strongly threatened,

either by timber extraction, cattle grazing, and the burning of adjacent pastures (Kessler *et al.*, 2014). The genus *Polylepis* is conditioned to very hostile territories and markedly seasonal climates; they have adapted to spaces where sub-zero temperatures occur and to critical periods of drought (Azócar *et al.*, 2007). Studying the adaptation of *Polylepis* forests to adverse environmental conditions, and predicting the physiological response of plants to these factors through the analysis of water stress in relation to the altitudinal gradient, is a concern of researchers for the recovery and restoration of degraded forests.

Water in the plant usually constitutes 80 to 95% performing functions in the transfer of molecules between cells (Moreno, 2009) through the

conductive vessels of the plant (Bidwell, 1979), a function that is affected when transpiration exceeds the rate of water assimilation (Kar, 2011); and it is aggravated by a weak correlation that exists between soil moisture and plant development. Water uptake by roots is not an autonomous process, but depends largely on the rate of water loss during the transpiration process, and water deficits can develop even in moist soils (Arroyo, 2015).

The water status of plants can also be affected by climate, specifically in the Andes by the microclimate, generated by the topography and the presence of other species known as the wetting effect that improve the quality of the microsite (Armas *et al.*, 2008). When the transition from the wet to the dry period occurs, they generate a decrease in leaf water potential and to preserve the soil-vegetation-atmosphere relationship, the plants decrease their osmotic potential and thus mitigate water loss and their tolerance to stress (Morgan, 1984); (Aranda *et al.*, 2005). Water availability is a major constraint to plant growth in dry and seasonally dry regions, as in much of the Andean subtropical and tropical range (Sparacino *et al.*, 2020). The movement of water bodies is a consequence of the intensity of the gradient, and modulated by the hydraulic conductivity of the soil (Hernández & Medina, 2012; Aranda *et al.*, 2005).

The water within the conductive vessels is transferred from areas with greater potential to areas with lesser potential (Lira, 1994). Water stress is the result of considerable tension within the plant, if it exceeds its own plasticity it damages enzymatic processes causing its mortality, however some plants have developed special means to survive in extreme conditions due to lack of water availability, acquiring resistance capacities (Bidwell, 1979).

One of the indicators that allow quantifying the water status of plants are the water potential values with negative figures in Mpa (Klepper, 1968) and the normalized difference vegetation index NDVI (Trueba, 2017). The NDVI is one of the most widely used vegetation indices for ecosystem monitoring (Tucker, 1979) and vegetation vigor (Aguilar *et al.*, 2012). The scale used to determine the water status of the vegetation goes from -1 to 1, the value zero means the beginning of plant absence (Romero, 2016). On the other hand, the altitude is an important factor

that influences the normal functioning of the flora due to sudden changes in solar radiation, temperature, intense winds, accompanied by a decrease in CO₂ and O₂, (Esperón & Barradas, 2016). The temperature intervenes directly in the physiological processes of the plant by varying the rate of transpiration and the anabolic processes (Gonzalez, Pastenes, & Horton, 2001), depending on the dynamism of the water in the soil, as a result of the intensity of the gradient, modulated by the hydraulic conductivity of the soil (Hernández & Medina, 2012). However, some authors mention that all plants have acclimatization mechanisms (Kessler *et al.*, 2014), such as the Andean flora that modify the angle of insertion of the leaves to the stem, to avoid a drastic increase in temperature within the plant (Morales *et al.*, 2004). During the transpiration process, the plants act as a conduit between the soil and the atmosphere, in order to regulate the opening of the stomata (Arroyo, 2015), generating water tension in the plant due to the retention forces of the soil and the attraction of water by the atmosphere (Luna *et al.*, 2012).

Results from some research on *Polylepis* trees revealed that plants respond to the effect of altitude through morphological changes, decreased transpiration and lower rate of photosynthesis, to counteract low temperature and water stress (Macek *et al.*, 2009). In the Piedras Blancas moor in Venezuela, leaf water potential decreased twice as much as normal during the transition from the wet to the dry period, finding a leaf water potential of -1.3 MPa during the early afternoon and -2.2 Mpa at midday (Radae *et al.*, 1996). Azócar *et al.* (2007) in studies of temporary measurement of leaf water and osmotic potential in *Polylepis*, *P. sericea* and *P. tarapacana* found a decrease in leaf water potential in the dry season. Values above -16 Mpa or more represent a severe degree of stress, affecting the plant with decreased photosynthesis and incipient plasmolysis process at cellular level (Lira, 1994).

In this context of knowledge, the question arose: How does the water status of the *Polylepis* forests relate to the altitudinal gradient, slope and microclimatic variables? To find the answers, the objective was to determine the water potential and the NDVI as a function of the altitudinal gradient, slope and microclimatic factors of the *Polylepis* forests in the central sierra of Peru.

MATERIAL AND METHODS

Study area

The scope of the study includes the *Polylepis* forests located in the rural communities of Curimarca (district of Molinos, province of Jauja), Mariamoya and Pomamanta (district of Comas, province of Concepción), Laraos (district of Laraos, province of Yauyos) belonging to the region of Junín and Lima. The forests are distributed between altitudes of 3900 to 4600 m, composed of three species: *P. Rodolfo-vasquezii*, *P.canoi* and *P.flavipila* (Figure 1, Table 1).

The rural community of Curimarca is located in the sub-basin of the Curimarca river that flows into the Perené basin (ANA, 2016). The Mariamoya and Pomamanta communities are located in the Perené and Laraos basins in the Laraos River sub-basin (INRENA, 2003). These spatial areas have an average annual precipitation between 715 and 775 mm, maximum average temperature between 14.6°C and 20°C and minimum

temperature between 0.8°C and 2.0°C, with marked differences between the rainy and dry seasons during the year. The three species studied are located in the following life zones: *P. rodolfo-vasquezii* (semi-arid subalpine steppe), *P.canoi* (semi-arid montane shrubland) and *P.flavipila* (subtropical humid subalpine shrubland) (Aybar and Lavado, 2017).

Data Collection

In order to quantify the diurnal water potential, NDVI, humidity and temperature (soil and air) of forests of the genus *Polylepis*, forests larger than two hectares were selected, in which seven plots of 15 m x 21 m and 10 sub-plots distributed in the low, medium and high part of the forest were established, whose delimitation was made with the help of metric winches, GPS and a compass for the orientation of the plots. In each subplot, two individuals were selected and four branches were collected from each tree (Urribarri *et al.*, 1996), from each branch leaflets were

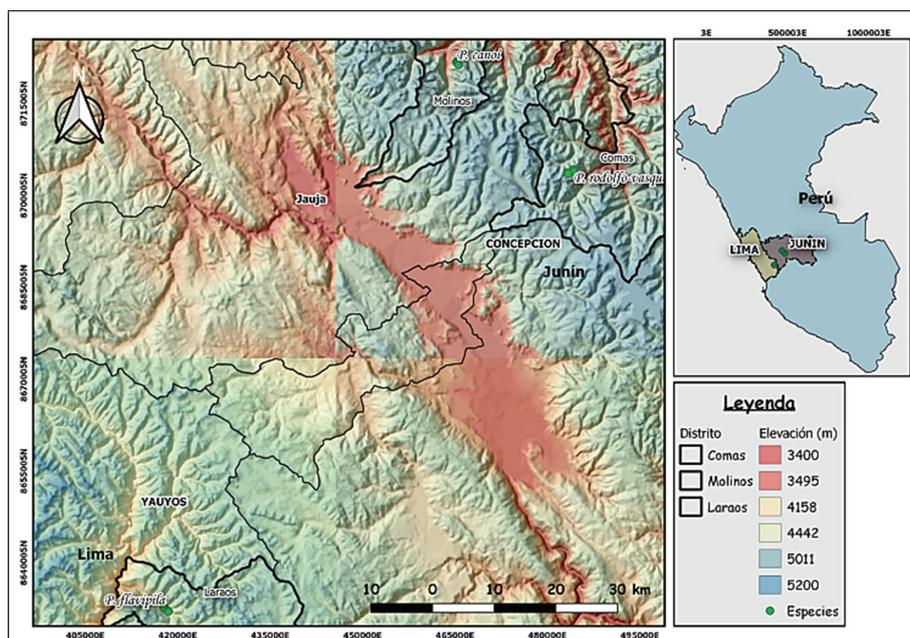


Figure 1. Map of the location of the study area and the three endemic species of *Polylepis*: (a) *P. rodolfo-vasquezii*; (b) *Polylepis canoi*; (c) *P. flavipila*.

Table 1. Distribution of *Polylepis* forests according to rural community

Region	Province	District	Forests	Sector	X	Y	<i>Polylepis species</i>
Junín	Concepción	Comas	Pomamanta	Paracshopampa	484194	8704078	<i>P. Rodolfo- vasquezii</i>
Junín	Concepción	Comas	Maria Moya	Riciscancha	484135	8704058	<i>P. Rodolfo- vasquezii</i>
Junín	Jauja	Molinos	Curimarca	Jucha	466584	8723206	<i>P.canoi</i>
Lima	Yauyos	Laraos	Laraos	Shutco Canyon	416642	8635513	<i>P. flavipila</i>

selected that were exposed to light without any symptoms of disease or evident damage.

To determine the water potential of the species under study, a Scholander type pressure pump was used in a Pascals unit (Mpa) (Calderon, 2013). Readings were taken at midday at intervals of 1–2 h and along the altitudinal gradient (Rada *et al.*, 1999; Azócar *et al.*, 2007). Four branches of the same tree were chosen according to the cardinal points of each two individuals of 1.5–4 m., then four leaflets were collected from the apical part of 5 cm \pm 1.25 cm in average length in each altitudinal gradient (Rada *et al.*, 1999, Urribarrí *et al.*, 1996). The selected branches were placed in a darkened ziploc bag during 5 minutes, then the cut was made with the help of a knife, sealing it to stop the transpiration or dehydration at the time of cutting. Then the sample was introduced into the pressure chamber, adding pressure with gaseous nitrogen at ½ bar per second until the water meniscus was visualized with the help of the 10x magnifying glass to record the data from the pressure meter; each data obtained was assigned its respective code according to the methodology followed by Calderon (2013).

To characterize the microclimate conditions in the forest, humidity and temperature sensors were installed for both air and soil, two meters above the soil surface and 10 cm deep in the ground respectively (Toivonen, 2014). The NDVI was measured using RPAS equipment with a visual sensor that allowed remote takeoffs and landings. Flights of up to 20 minutes were planned, with average runs of 500 m (Romero, 2016). A multispectral camera composed of two elements was used, the multispectral lenses which have spectral sensors or filters and a solar sensor which recorded the intensity of light (ACG Drone, 2016).

Data analysis

The data distribution fitting test was performed, using R Project's *riskDistributions* statistical package, which provides a set of functions for distribution estimation or distribution testing (Belgorodski *et al.*, 2017). In addition, principal component analysis (PCA) was performed with R Project's *factoextra* statistical package for the observation of possibly correlated variables and building predictive models. In addition, the non-parametric U-Mann-Whitney and Kruskal-Wallis tests were used to compare the physiological parameters, water potentials and NDVI of the *Polylepis*.

RESULTS AND DISCUSSION

Water potential (Mpa) in *Polylepis* forests in relation to the altitudinal gradient

The water potential of the leaf (Ψ_L) during the dry season, measured at midday, was different in each *Polylepis* species forest. A progressive decrease was observed as the tree population declined from the top line of the trees to the start line of the tree population. *P. flavipila* showed lower values of (Ψ_L) near the tree line of -1.83 Mpa at 4147 m with moderate water stress levels and a minimum value of -0.90 Mpa at altitudes of 3915 m with mild water stress levels; however, *P. rodolfo-vasquezii* populations were found at higher altitudes and exhibited a maximum value of -1.50 Mpa at 4438 m presenting a moderate water stress level and a minimum value of -0.75 Mpa at 4174 m indicating that they had no water stress problems. Populations of *P. canoi* were found at lower altitudes and in more humid soils, the (Ψ_L) at higher elevation (3914 m) was -1.24 Mpa resulting in a mild water stress level and at the lower elevation (3843 m) the (Ψ_L) was -0.65 Mpa ranking without water stress problems (Figure 2a). Significant differences were detected in water potentials according to species ($p < 0.05$), *P. flavipila* and *P. rodolfo vasquezii* showed low water stress values with moderate stress levels at altitudes above 4100 m, which confirmed that the species come from different populations.

The differences observed in the leaf water potential of the *Polylepis* forests studied are due to the thermal conditions of their habitat and the water status of the plants (García *et al.*, 2004); this is confirmed by the results obtained, *P. flavipila* grows in drier places, therefore it tends to tolerate better extreme climates, on the contrary, *P. canoi* showed variable behaviors, they were found in humid areas and in areas with low availability of soil moisture, in higher elevations where environmental conditions were more rugged, they showed decrease in the values of water status in areas with low availability of soil moisture. The three species were not affected by water stress, because these species have numerous and long roots that allow them to absorb available water at greater depth (Bucci *et al.*, 2011). On the other hand, the trees of the genus *Polylepis* showed different responses to the altitude gradient, presenting different levels of water stress significantly higher in species near the tree boundary line (Macek et

al, 2009), all of which can be explained by the greater stomatic control that *Polylepis* individuals exert when presenting smaller leaflets (Azócar *et al.*, 2007), and by the genetically determined functional adaptation to high altitude and cold conditions (Toivonen, 2014). This behavior was demonstrated in *P. flavipila*, which presented greater water potential in places with high temperatures at midday and soil with low humidity; these results have allowed rejecting the hypothesis of homogeneous behavior of the *Polylepis* species with respect to water potential. In the light of the findings, it is possible to establish the criterion that the different *Polylepis* species are specifically adapted to the particular conditions of temperature, humidity and altitudinal gradient. The high and negative correlation between the water potential and the altitudinal gradient obtained for the plants in each forest reflects the great capacity of adaptation of the *Polylepis* individuals to the extreme climate changes that occur at high altitudes in the central sierra of Peru and Latin America, due to their presence from Venezuela to the south of Chile (Castro and Flores, 2015).

Differential normalized vegetation index (NDVI) in *Polylepis* forests related to the altitudinal gradient

The NDVI for the dry season recorded at midday were significantly different among the *Polylepis* forests ($P < 0.05$) (Figure 2b), these decreased progressively according to their distribution from the tree line to the lower limit of the

tree population. *P. flavipila* showed lower NDVI with an index of 0.34 at an altitudinal gradient of 4147 m (near the tree line), with moderate water stress and an index of 0.43 at 3915 m with mild water stress. In populations of *P. rodolfo-vasquezii* the NDVI was 0.39 at elevations of 4438 m in forest 1 showing moderate water stress, forest 2 registered a NDVI of 0.47 presenting mild stress, due to higher soil moisture; at lower altitudes the NDVI was 0.60 to 4174 m in forest 1 and in forest 2 the index was 0.58 to 4416 m not presenting stress problems. *P. canoi*, recorded a NDVI of 0.43 to 3914 m resulting in mild water stress and at lower elevations the index was 0.69 to 3843 m which indicates no problem of water stress, this result obtained could be attributed to these populations are located at lower altitudes with wetter soils compared to other populations.

The NDVI values calculated for the three *Polylepis* species varied between 0.3 and 0.7, and are within the range established by Rosemary (2016). Indices with high values showed plant vigor at lower altitudes (forest onset). The strong negative correlation of the NDVI with respect to the altitudinal gradient is supported by the variation in soil moisture and air temperature, which changes according to altitude and slope orientation. A limitation in the capture of multispectral images were those areas with higher elevations where the topographic relief presents slopes with strong inclination, the sampling plots had differences in altitudinal location, this situation hindered the use of automatic flight plans of the drone phantom 4 PRO, and manual flights were

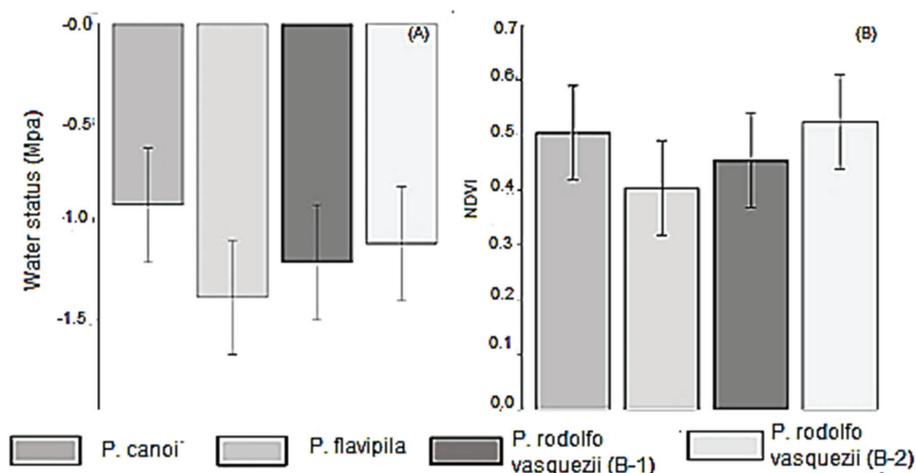


Figure 2. Water status (a) and NDVI (b) in *Polylepis* forests of *P. canoi*, *P. Rodolfo-vaquezii* and *P. flavipila* populations, in the dry season There is a significant difference ($P < 0.05$) between species.

performed over each specific space of the plot; the data obtained was processed for each subplot giving a rather limited point cloud for the construction of orthophotos. Unmanned aerial vehicles in mountainous conditions, fly over small spaces and at an altitudinal scale, with very short flight times due to the variation of the weather during the day and the presence of strong winds mainly in the afternoon. The high and negative correlation between water potential and NDVI shows that altitude has an inverse effect on water potential and NDVI, which indicates changes in water status in the *Polylepis* forest, therefore, forests that are located over areas at higher altitudes the soil moisture decreases (Bucci *et al.*, 2011)

Correlation between water potential and NDVI index in *Polylepis* forests with the altitudinal gradient

Figure 3a shows the dispersion of trees with respect to water potential and the altitudinal gradient and their association between forests. It is observed that the variation in altitude is associated with the variation in leaf water potential, and according to Spearman's correlation analysis a significant, strong and negative correlation is observed ($p < 0.001$), indicating that at higher elevations the water potential of the trees is lower.

The correlation between the NDVI and the altitudinal gradient was inverse, very significant and strong ($p < 0.001$), which indicates that at higher altitudes the NDVI index presented lower values (Figure 3b)

NDVI and leaf water status are used to estimate the level of water stress in the plant at a given time, and these two parameters are very dynamic during the day. As soil moisture decreases, leaf water potential becomes more negative and NDVI decreases, resulting in a stressed plant, results consistent with those reported by (Himada *et al.*, 2012), who correlated NDVI and leaf water potential, having found a positive correlation of 0.86. From the evaluation of the water status of *Polylepis* forests, leaf water potential and NDVI at noon in the relic of *P. rodolfo-vasquezii* A and B, *P. canoi* and *P. flavipila* presented a positive correlation (0.73, 0.84, 0.93 and 0.93) respectively; which indicates that there is a direct and high association between the variables studied, and a common characteristic that determines the water status of the vegetation.

Water status as a function of NDVI, slope and microclimate variables of *Polylepis* forests

The results of Spearman's correlation of microclimate variables with water potential and NDVI are presented in Table 2. The correlation between air and soil moisture with NDVI and water potential for *Polylepis* populations in the four rural communities was positive and significant ($p < 0.001$). Populations located in the rural communities of Laraos and Pomacancha showed a high and very significant correlation. Correlations between air and soil temperature with NDVI and water status for the study populations, showed a negative and very significant correlation ($p < 0.001$), the populations located in Laraos, Pomacancha and Maria Moya, registered high and very significant correlations. The correlation of the slope with the NDVI and the water potential did not preserve this homogeneity in the results. A positive correlation was observed in the communities of Maria Moya and Pomacancha and a negative correlation in the communities of Curimarca and Laraos.

The homogeneity of correlations of microclimate variables with water status and NDVI is the result of the high correlation between them (García *et al.*, 2004); however, the differences between moisture and temperature variables that are related in different ways in the four forests evaluated, endorse that the methods and instruments used in the evaluation are appropriate for high Andean *Polylepis* forests. On the other hand, with an increase in temperature, soil moisture decreases and this affects the water status of the plant through increased transpiration (Toivonen, 2014; Miranda *et al.*, 2020). This demonstrates the high effectiveness of the use of unmanned aerial vehicles that can capture multispectral images to represent NDVI values, and monitor the water status of plants, the water status of forests and crops, and for restoration actions (Tito *et al.*, 2020). These results are consistent with the study conducted by Marusig *et al.* (2020) in Northeast Italy, who monitored the water status of trees against the risk of dehydration during dry periods, as well as the results of Shimada *et al.* (2012) who compared two methods to evaluate the relationship between water status and NDVI in crops, and monitored the response of vegetation to precipitation and temperature. Additionally, the mathematical models found are good predictors and useful, because of the significant correlation

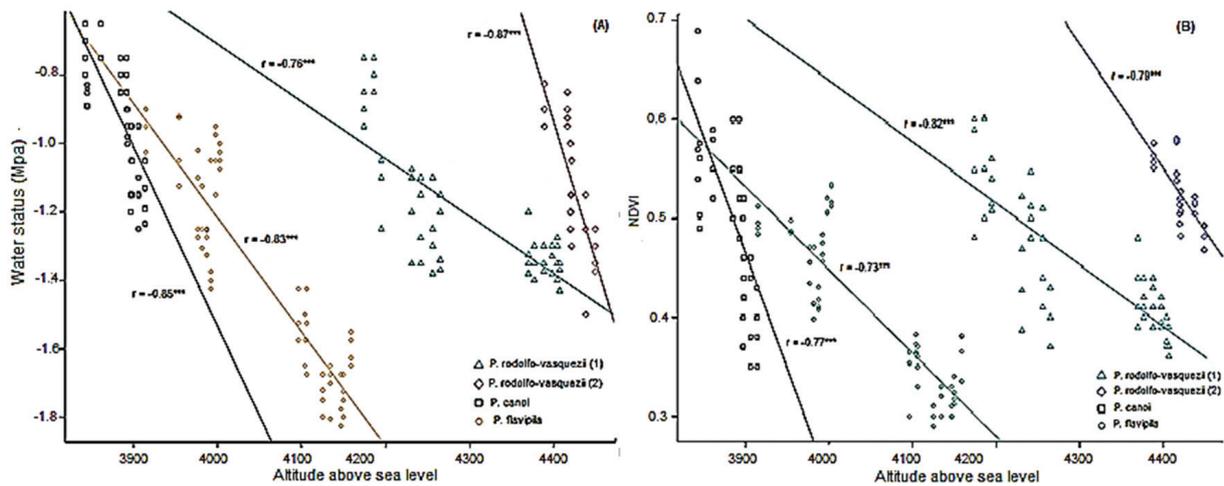


Figure 3. Correlation between leaf water status (a) and NDVI (b) of *P. canoi*, *P. rodolfo-vasquezii* and *P. flavipila* with the altitudinal gradient

Table 2. Correlation of the water status of *Polylepis* and the NDVI in relation to microclimate variables and slope in four rural communities

Rural community	Variables	NDVI	Water status (Mpa)
Maria Moya	Air moisture %	0.43**	0.39**
	Air temperature °C	-0.79***	-0.72***
	Soil moisture %	0.74***	0.69***
	Soil temperature °C	-0.63***	-0.67***
	Vapour pressure density	-0.62***	-0.53***
	Slope %	-0.26*	-0.28*
Pomacancha	Air moisture %	0.81***	0.8***
	Air temperature °C	-0.80***	-0.87***
	Soil moisture %	0.68***	0.83***
	Soil temperature °C	-0.82***	-0.85***
	Vapour pressure density	-0.81***	-0.79***
	Slope %	-0.13*	-0.34**
Curimarca	Air moisture %	0.21*	0.05*
	Air temperature °C	-0.76***	-0.82***
	Soil moisture %	0.79***	0.80***
	Soil temperature °C	-0.41***	-0.55***
	Vapour pressure density	-0.76	-0.71
	Slope %	0.36**	0.49**
Laraos	Air moisture %	0.93***	0.93***
	Air temperature °C	-0.78***	-0.78***
	Soil moisture %	0.88***	0.90***
	Soil temperature °C	-0.84***	-0.77***
	Vapour pressure density	-0.81***	-0.76***
	Slope %	0.13*	0.10*

Correlation is significant at the level *0.05 **0.01 ***0.001 (2 tails).

between variables such as: soil moisture, temperature and air moisture as input variables, which are those that influenced the results of the physiological status of the *Polylepis*.

CONCLUSIONS

The three species of *Polylepis* showed a changing water stress in different altitudinal gradients, the water status varied from mild to moderate associated with soil moisture, individuals showed greater vigor in low areas than in areas near the tree line. The species studied showed morphological changes according to the altitudinal gradient influenced by the hydrological variation of the soil and the temperature, whose response was manifested through the size of their leaflets and the stomatic control in front of the hydric stress, factors like the altitude should be taken into account in the reforestation programs, for a particular species of *Polylepis*. The high correlation of the Normalized Differential Vegetation Index (NDVI) with the water status of *Polylepis* plants will allow the use of unmanned aerial vehicles (drones) in monitoring programs of mountain forests with steep slopes. The significant and differentiated correlation of the slope with the NDVI and the water status is another finding that indicates the particular adaptation of *P. rodolfo-vasquezii* individuals in relation to *P. canoi* and *P. flavipila*, which should also be taken into account in reforestation programs.

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