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Authors: Flores-Alvarez, John Li, Ladd, Brenton, Velez-Azañero,

Armando, de Mola, Ursula Loret, and Bonser, Stephen

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# Using Knowledge of Plant Persistence Traits to Optimize Strategies for Post-Mine Ecological Restoration on the Peruvian Altiplano

John Li Flores-Alvarez<sup>1,3</sup>, Brenton Ladd<sup>1,2</sup>\*, Armando Velez-Azañero<sup>1</sup>, Ursula Loret de Mola<sup>1</sup>, and Stephen Bonser<sup>2</sup> \* Corresponding author: bladd@cientifica.edu.pe

- School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney, NSW 2052, Australia
- Biotechnology, Wageningen University, P.O. Box 9101, 6700 HB Wageningen, the Netherlands

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The Peruvian Altiplano is an important center of mining activity. As a key hotspot for endemic biodiversity in Peru, it also has conservation value. Thus, ecological restoration strategies need to be developed to ensure

that mining impacts on these biodiversity values are minimized. As a step toward this larger objective, we measured the composition of the soil seed bank on the Peruvian Altiplano and compared it with the standing vegetation in different plant communities. We calculated evolved persistence traits to evaluate the suitability of topsoil removal, storage, and redeployment as an ecological restoration strategy for the Peruvian Altiplano. The comparison demonstrates low similarities (Sorensen Index for sites A: 0.15, B:0.27, C:0.11,

and D:0.12) between the seed bank and the standing vegetation. The flora of the germinable soil seed bank is dominated by ephemeral species. Meanwhile, the standing flora contains a higher percentage of perennial species, many of which invest in the capacity to persist via resprouting from underground storage organs. It is clear that the well-established restoration technique of topsoil removal, storage, and redeployment needs to be tested and will probably need modification to be an effective strategy for the Peruvian Altiplano. The required modifications are relatively straightforward and would ensure that this low-cost and effective restoration technique could be applied to good effect on the Peruvian Altiplano.

**Keywords:** Resprouting; ecological restoration; topsoil removal; topsoil storage; topsoil redeployment; seed bank; Peru.

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# Introduction

The Peruvian economy is growing at a rapid 3.9% per year (Global Economic Prospects 2016), and this growth rate has been maintained over the last 12 years. Between 2005 and 2012 the gross domestic product grew at a rate of 7%, while the national investment rate increased from 17% to 26%, mainly because of investment in mining activities (Charles and Zegarra 2014). Mining activities in the Peruvian Andes are mostly related to copper and gold mining/extraction (Wang and Miao 2016; Fraser 2018). Mining in Peru is important because it inflates the tax base, creating possibilities for the development of new infrastructure and thus for lifting some of the most disadvantaged segments of the population out of poverty. However, mining also generates negative environmental impacts. Peru has created new laws to safeguard against these potential impacts (eg a law to protect the upper reaches of water catchments, Law 30640 of the Peruvian

National Government, approved in August 2017), and mining companies are investing heavily in restoration efforts (eg https://www.blacktogreen.com/nosotros-2/ nuestras-referencias/mina-los-frailes/).

Mining in Peru is concentrated on the Andean plateau (the Altiplano). The Altiplano is well known not only for mining activities (Loayza and Rigolini 2016), but also for its conservation values, being a "biodiversity hotspot" and the largest extension of tropical alpine habitat on the planet (Myers et al 2000). Further, Peru's mountain ecosystems provide a range of ecosystem services, especially water-related ecosystem services (Chapin et al 1997; Vásquez et al 2014) and are home to a range of endemic species of high conservation value (Myers et al 2000; Brooks et al 2002; Gareca et al 2010).

In an effort to safeguard these conservation values, mining companies are implementing an ecological restoration strategy of topsoil removal, storage, and redeployment, a technique that is commonly used in

Australia as a successful and low-cost method for postmine restoration (Rokich et al 2000; Koch 2007). However, the existence of a suitable seed bank that could contribute to successful ecological restoration using the topsoil removal and redeployment method has not been verified on the Peruvian Altiplano. The potential for achieving successful ecological restoration using topsoil removal and redeployment should depend heavily on the persistence traits of both the standing flora and the germinable soil seed bank (Vesk and Westoby 2004; Koch 2007; Fowler et al 2015). As we still lack basic information about the persistence traits of Peruvian flora, generating such information would be important to optimize ecological restoration strategies (Gondard et al 2003; Suding et al 2008; Ostertag et al 2015; wa Ilunga et al 2015; Díaz et al 2016).

Topsoil removal, storage, and redeployment (hereafter simply defined as topsoil redeployment) could, for example, produce undesirable effects if the soil seed bank is dominated by exotic species (Chambers et al 1994; Patzelt et al 2001; Holl 2002; Skowronek et al 2014). Persistence traits expressed by native species could also be an important factor for the success (or failure) of restoration efforts using topsoil redeployment (Fowler et al 2015). For example, perennial species, including resprouting species, may not invest heavily in the production of seeds. In such cases, the success of ecological restoration through topsoil redeployment is likely to be limited (Koch 2007). Further, even when the main form of persistence for a plant community is via the production of seeds, the possibilities for achieving successful restoration using topsoil redeployment will vary from species to species. For example, ephemeral species that complete their life cycles within a single growing season often produce a large number of small seeds with thin seed coats, which are easily dispersed by wind (Vesk and Westoby 2004). Seeds that possess thin seed coats are unlikely to persist for a significant length of time within a topsoil storage pile (Abdul-Kareem and McRae 1984; Golos and Dixon 2014).

Persistence traits tend to evolve in response to ecological factors such as disturbances that can remove standing plant biomass from ecosystems and increase mortality in plant populations (Vesk and Westoby 2004; Perez-Harguindeguy et al 2013). Resprouting is a key persistence trait in perennial herbaceous and woody plant species, where resprouting species reserve resources and meristems in protected tissue (underground or within stems) and survive disturbance events by activating these reserves post-disturbance (Bond and Midgley 2001). Resprouting capacity is a persistence trait that can define the persistence and success across habitats. Observing persistence traits has proven useful in understanding variation in ecological strategies (Lavorel and Garnier 2002). In addition, the physical environment (pronounced seasonality, high radiation, and cold dry winters) may also have led to the evolution of a perennial flora that is heavily reliant on the ability to resprout from underground storage organs such as rhizomes and tubers as a means of persistence (Vesk and Westoby 2004; Lütz 2010; Rada et al 2011; Neuner 2014; Reich 2014).

Surveys conducted in Andean ecosystems have shown that it seems plausible that the flora of the Peruvian Altiplano possesses persistence traits that would present significant challenges for any restoration strategy that relies upon topsoil redeployment (Arroyo et al 1999; Cano et al 2010; Cano et al 2011). First, these ecosystems were already heavily degraded before the start of mining, due to millennia of human impact (Kessler 2002). The human impacts (overgrazing, indiscriminate use of fire to promote the growth of new pasture) may have favored the dominance of ephemeral species or alternatively, the evolution of a perennial strategy such as resprouting from underground storage organs (Bond and Midgley 2001; Vesk and Westoby 2004; Gurvich et al 2005; Fernández et al 2010). In both cases, it seems reasonable to hypothesize that the potential to restore using topsoil redeployment would be limited because both thin-coated ephemeral seeds and underground resprouting organs would likely putrefy in a topsoil storage pile.

This research focuses on the semi-natural ecosystems used as rangeland in the region. These ecosystems are important because they account for the majority of the Altiplano; they are used by the human population for livestock production and they need to be restored after mining. Here we provide a first assessment of the persistence traits of the flora of the Peruvian Andes, in both the soil seed bank and the standing flora, focusing specifically on traits related to persistence/reproduction. We specifically hypothesized that (1) the soil seed bank would be dominated by ephemeral weedy species and (2) the standing flora would be dominated by perennial species including species that persist via resprouting from underground storage organs.

### **Methods**

The measurements were made in a landscape reserve known as Nor Yauyos Cochas located in the central Andes of Peru, 4000 meters above sea level close to mining operations. The flora in these ecosystems is dominated by the *Compositae* and *Poaceae* families, as well as woodlands dominated by small trees of the *Rosaceae* family (*Polylepis*) (INRENA 2006). These plant communities are classified as swamps (Sites A, C), roquedales (Site B), and cryoturbated (Site D). Sites B and D were located on hillsides, and these ecosystems commonly contain a great variety of species (Cano et al 2011; Gonzáles 2015). However, while we possess taxonomic information of the flora in the region there is a notable absence of information on the flora's persistence traits (Cano et al 2010, 2011; Kahn et al 2016). The locations sampled in this study, as is the case for the

entire Peruvian Andes, are used by local people for livestock production (Gutiérrez 2011).

Four sampling locations were selected during the first weeks of the spring season (September-December 2015), at the start of the rainy season on the Altiplano. The geographic coordinates of the 4 sites are as follows: (12°19′38.40″S, 75°46′52.10″W [site A]; 12°19′37.90″S, 75°46′49.96″W [site B]; 12°22′41.20″S, 75°44′42.85″W [site C]; 12°22′35.12″S, 75°44′41.11″W [site D]). At these 4 locations, 20 soil samples (replicates) were collected at each location (a total of 80 soil cores) using a metal tube  $(diameter = 10.5 \text{ cm}, area = 86.6 \text{ cm}^2, depth of excavation})$ = 5 cm) to standardize soil volumes, in order to develop the seed bank germination survey. We chose 5 cm as the sampling depth because previous studies have shown that very few seeds persist/occur below this depth (Koch et al 1996; Rokich et al 2000; Fowler et al 2015), and 4 control samples with sterile soil were used to control the effect of possible growth of undesirable weeds not from the seed bank. In addition, we conducted comprehensive botanical surveys of the standing flora at the same 80 locations from which soil samples were collected, using quadrats of 2 meters by 2 meters. Plants registered in these 4 m<sup>2</sup> quadrats that could not be identified in the field were collected as voucher specimens and photographed for identification in the lab (Macbride 1936). Likewise, comparisons were made with the San Marcos Herbarium (USM) of the Natural History Museum in Lima, Peru.

Soil samples were transported in Ziploc bags to the Universidad Cientifica del Sur's greenhouse in Lima, where they underwent 2 weeks of cold stratification in a refrigerator (4 °C) to mimic the cold winters typical in the Peruvian Andes. After this, soil seed bank samples were allocated to pots arranged in a randomized complete block experimental design on 15 October 2015, watered 4 times a week, and the newly emerged seedlings were counted on the same days that the pots received water. Once the emerged seedlings had grown for 1 to 3 months and their identification became possible, they were harvested for further identification.

The resulting data were used to compare the similarity of the flora in the soil seed bank with the flora in the standing vegetation using the Sørenson index (Sørensen 1948):

$$\emptyset = 2c/(a+b) \tag{1}$$

where  $\emptyset$  = Sørensen index, a = Number of species in the field, b = Number of species in the seed bank, c = Number of species found in both sites.

The Sørenson index was calculated for each of the 4 sites independently, as well as using a dataset that combined the data from the 4 sites (*Supplemental material*, Table S1: http://dx.doi.org/10.1659/MRD-JOURNAL-D-17-00088.S1). We also characterized species persistence traits in both the standing vegetation and in the soil seed bank using the classification scheme developed by Vesk and

Westoby (2004). We classified plants as ephemeral (annual) or perennial. Ephemeral species usually germinate and die during a single season (i.e., spring, summer) and may even complete 2 generations in a year, while perennial species persist in the ecosystem for more than 1 year. The species in this survey were also classified species as either seeders or resprouters (after Vesk and Westoby 2004). We also classified species that resprout from underground storage organs as perennials, and we ascertained the presence or absence of perennial tubers by excavation in the field (Figure 1).

### **Results**

A total of 870 seeds germinated from the Yauyos seed bank, which emerged from 71 soil samples. No seeds germinated from the remaining 9 samples. The seed bank was composed mainly of *Compositae* (28%) and *Poaceae* (14%) species ranging from 0 to 104 individuals (0 to 12,010 seeds/m²), on average 10.9 seeds (1258 seeds/m²) per pot (see Figure 2 and *Supplemental material*, Table S1: http://dx.doi.org/10.1659/MRD-JOURNAL-D-17-00088.S1).

The seed bank of swamp soil samples was dominated by the ephemeral species Cotula mexicana (DC.) Cabrera (Sites A and C). In contrast, only a small number of individuals of this species emerged from soil samples collected on the hillsides (Sites B and D). Cotula mexicana (DC.) Cabrera was also observed in the standing vegetation in the field but only at the swamp sites. Other abundant species included Trifolium amabile Kunth., and Oxalis brasiliensis G. Lodd on hillsides. The most important species recorded, in terms of conservation value, was Polylepis subsericans J.F.Macbr. However, only 3 specimens were recorded in the seed bank. The Sørenson index showed a low similarity (Site A: 0.15, B: 0.27, C: 0.11, 0.12) between the flora in the soil seed bank and the standing vegetation for all 4 locations (Supplemental material, Table S1: http://dx.doi.org/10.1659/MRD-JOURNAL-D-17-00088. S1), and an overall comparison shows that only 14.5% of species occurred in both the standing vegetation and the seed bank. This lack of similarity was apparent also when data from all 4 sites were analyzed as one data set (Sørenson index = 0.25).

Plants that germinated from the soil seed bank were mainly ephemeral species (Supplemental material, Table S2: http://dx.doi.org/10.1659/MRD-JOURNAL-D-17-00088.S1) that germinate rapidly and grow fast during the first few weeks after germination, such as Cotula mexicana (DC.) Cabrera, which was mainly observed in location A (Figures 2 and 3). On the other hand, most species encountered in the field during the botanical surveys were perennial species, many of which use resprouting as their main persistence strategy (Supplemental material, Table S2: http://dx.doi.org/10.1659/MRD-JOURNAL-D-17-00088.S1; Figures 1, 3). Our excavations in the field showed that many perennial species develop taproots and woody

FIGURE 1 (A) Observation in the field suggests that resprouting capacity from underground storage organs is common on the Peruvian Altiplano. (B) A species from the Nototriche genus (Malvaceae family), in which resprouting is an ancient trait for long-term persistence. To casual inspection, these plants look like insignificant annual herbs. However, when excavated it becomes apparent that these plants may be very old (perhaps 800 years old) and that they are heavily reliant on resprouting from underground storage organs for their persistence. (C) The surface of a recently created topsoil storage pile for a new mine on the Peruvian Altiplano. This photograph shows that resprouting can occur when topsoil is redeployed immediately, but only in the superficial layer of the pile. Rhizomes buried deep within the topsoil pile will putrefy after a year or two. (Photos by Brenton Ladd)



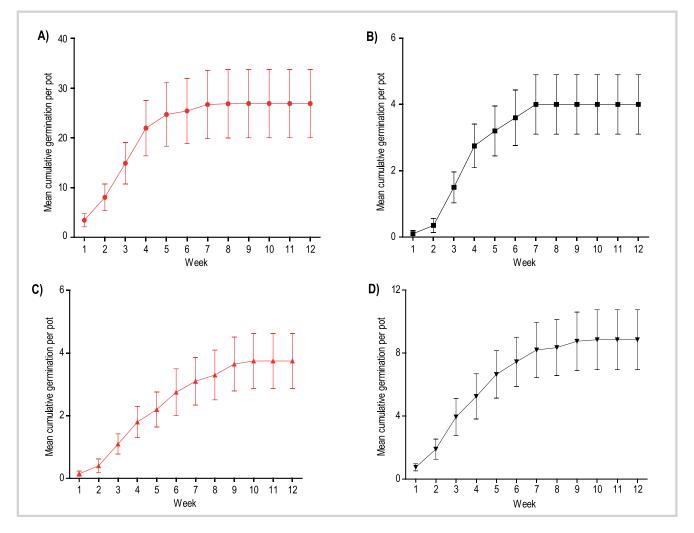
rhizomes buried underground, which reveals an adaptation to harsh conditions such as pronounced seasonality.

## **Discussion**

The density of the seed bank reported here is quite similar to that of other alpine regions and to values measured for a number of restoration projects (Funes et al 2003; Koch 2007). Furthermore, the diversity found in the botanical survey in the field is comparable to other botanical surveys from the Peruvian Andes in other locations (Cavieres 1999; Chanco et al 2006; Cano et al 2011; Gonzáles 2015). However, the Sørenson similarity index

(0.25) demonstrates that the standing vegetation is distinct from flora found in the soil seed bank (Supplemental material, Tables S1 and S2: http://dx.doi.org/10.1659/MRD-JOURNAL-D-17-00088.S1) and that the seed bank is dominated by ephemeral (annual) species, confirming the results of Arroyo et al (1999) in the Andean ecosystems of Chile (Sørensen index = 0.22), where no perennial species were found in the soil seed bank. In contrast, in the field, the standing vegetation is dominated by perennial species—either woody species (trees, shrubs) or species that resprout from perennial underground storage organs such as tubers and rhizomes. These perennial species recorded in the field have likewise been recorded in previous botanical surveys conducted in the region

FIGURE 2 Total number of cumulative germinates from the alpine soil seed bank over time (mean values). At week 12 the germination values were (A:26.9, B:4, C:3.75, D:8.85) and the average was 10.9 seeds per pot. Swamp sites have a red connecting line and hillsides black ones. The swamp site where mean number of germinates per soil sample was over 25 reflects a high abundance of the annual ephemeral *Cotula mexicana* (DC.) Cabrera. Letters A, B, C, and D correspond to the location described in the methods. Bars indicate the standard deviation of the data.



(Teillier 1998). Although *Polylepis subsericans* J.F.Macbr. was registered in the seed bank, its abundance was limited to only 3 individuals from 80 soil samples. This low density implies that restoration of key species must follow propagation methods other than topsoil removal storage and redeployment (Ruiz-Jaen and Mitchell Aide 2005; Fowler et al 2015).

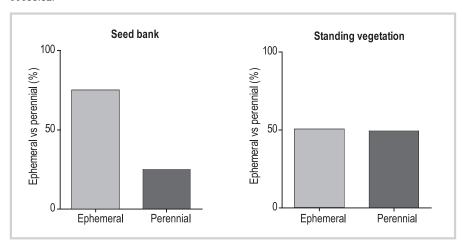
The presence of species that persist mainly via seed production recorded in the field but absent in the seed bank could be attributed to several factors. Research in Andean ecosystems has shown that plants may have limited recruitment due to the production of nonviable seed (Enrico et al 2004). The lack of viability of the seed has been related to water stress, constant frost, and a lack of nutrients such as nitrogen (Enrico et al 2004; Kessler 2006), which all lead to the production of sterile seeds (Renison et al 2004; Seltmann et al 2007). This is a possible explanation for the fact that we recorded only 3

individual *Polylepis* in the germination trial, which would not be enough to support a restoration project using soil redeployment as the main method.

Quite apart from issues related to seed viability the results demonstrate clearly that the standing flora and the soil seed bank differ because the standing flora is composed of a significant number of perennial species that invest in the ability to resprout from underground storage organs (Figure 1). With this high investment in longevity via resprouting the production of large numbers of seeds that could persist in a soil seed bank is not necessary or even possible due to energetic constraints (Thompson 1978, 1985; Archibold 1984; Guariguata and Azocar 1988; Chapin et al 1997; Cavieres 1999).

The fact that many of the plant species on the Peruvian Altiplano resprout from underground storage organs should come as no surprise. Resprouting is an adaptation to strong annual cycles of climatic variation: it

**FIGURE 3** Percentage ephemerals versus perennials in the soil seed bank and standing vegetation across the 4 sites; see also *Supplemental material*, Table S2: http://dx.doi.org/10.1659/MRD-JOURNAL-D-17-00088.S1.



makes sense to retreat underground to avoid harsh winters. Furthermore, germination may also be limited in the high Andes because of extreme radiation, constant frost, and low water availability (Robberecht et al 1980; Enrico et al 2004; Kessler 2006). Resprouters store nutrients and sugar underground which they use to quickly occupy gaps when conditions become favorable. This is a common evolutionary strategy where growing seasons are short (Bond and Midgley 2001). Tubers such as the potato which evolved in the Andes resprout when temperatures rise above 5°C and remain dormant when temperatures are below 3°C (Vreugdenhil et al 2011). This avoids the destruction of meristems by low temperatures and may allow an individual to persist for hundreds of years (Figure 1). Resprouting is also a commonly observed reproductive strategy when herbivores are abundant (Rosenthal and Kotanen 1994; Díaz et al 2001; Fornoni et al 2003a; Ickes et al 2003). Plant tissues that generate new growth (meristems) are energetically expensive to produce and placing them underground means that they are protected (Howell et al 2011). In the Peruvian highlands, there are harsh winters as well as overgrazing (Brack-Egg and Mendiola-Vargas 2010), which may have resulted in the evolution of a flora that is heavily dependent on resprouting as a means of persistence.

The low relative abundance of perennial species with the ability to resprout from underground storage organs in the soil seed bank provides evidence that the topsoil removal, storage, and redeployment technique of ecological restoration used to such good effect in Western Australia is unlikely to function well in the Peruvian highlands without modification, as recommended by Koch (2007) for dry forests in Australia. The fact that the seed bank is dominated by annuals adapted to wind dispersal demonstrates that most species in the seed bank will have thin seed coats. Thin seed coats are ideal for wind dispersal and colonizing disturbed habitats, but they are

less than ideal for persisting within a topsoil storage pile where the seeds are likely to putrefy after a year or two (Abdul-Kareem and McRae 1984; Fowler et al 2015). Therefore, complementary techniques such as nursery salvage and propagation of individual plants in greenhouse facilities will also be required (Koch 2007; Fowler et al 2015).

### Conclusion

The results of the study illustrate that without modification, topsoil removal, storage, and redeployment is unlikely to achieve the restoration of the perennial species that dominate the standing flora in the region on the Peruvian Altiplano. Further research and data collection are needed. It is highly likely that the storage effect and seed dormancy (Chesson 2000) is important for the maintenance of diversity in these high Andean ecosystems, and this should be assessed in future studies.

The tubers and rhizomes that many plants in the Andean flora use to persist in the environment likely will die after a period of time if placed in a topsoil storage pile. Likewise, the ephemeral-type seeds with thin seed coats that seem to dominate the soil seed bank are unlikely to persist for the length of time that a typical mine is active. The problem could be resolved in a number of ways. For instance, if excavated topsoil is redeployed immediately, it is possible that many of the root fragments will resprout and the genetic resource will be preserved (Figure 1). The direct and immediate redeployment of topsoil could be achieved by moving excavated topsoil to already degraded sites (e.g., old mines), which are abundant on the Peruvian Altiplano (Brack-Egg and Mendiola-Vargas 2010), and so direct and immediate redeployment of topsoil is possible. This would help to compensate the negative effects that mine pits, tailings dams, etc., have on biodiversity.

Topsoil removal, storage, and redeployment is an important low-cost method and is indispensable for providing a substrate for revegetating post-mining. However, it will be essential to complement this

restoration strategy with additional actions to ensure the preservation of the significant component of the Andean flora that is reliant on resprouting for persistence.

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# Supplemental material

**TABLE S1** Data and calculations of the Sørenson index across 4 sites in the Peruvian Andes (swamps [sites A, C], *roquedales* [site B], and cryoturbated [Site D]) for the standing vegetation vs the soil seed bank.

**TABLE S2** Species list of plants observed in the standing vegetation and in the soil seed bank in the Peruvian Andes.

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