## Keywords

Alien species; Colobanthus quitensis; Deschampsia antarctica; human disturbance; Poa annua; tourists.

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#### Abstract

Because of its harsh environmental conditions and remoteness, Antarctica is often considered to be at low risk of plant invasion. However, an increasing number of reports have shown the presence and spread of non-native plants in Antarctica; it is therefore important to study which factors control the invasion process in this ecosystem. Here, we assessed the role of different human activities on the presence and abundance of the invasive Poa annua. In addition, we performed a reciprocal transplant experiment in the field, and a manipulative experiment of germination with $P$. аппиа and the natives Colobanthus quitensis and Deschampsia antarctica, in order to unravel the effects of physical soil disturbance on the establishment and survival of $P$. annиa. We found a positive correlation between abundance of $P$. annиa and level of soil disturbance, and that survival of $P$. annua was $33 \%$ higher in sites with disturbed soil than non-disturbed. Finally, we found that disturbance conditions increased germination for P. аппиа, whereas for native species germination in experimentally disturbed soil was either unchanged or reduced compared to undisturbed soil. Our results indicate that human activities that modify abiotic soil characteristics could play an important role in the abundance of this invasive species. If the current patterns of human activities are maintained in Antarctica, the establishment success and spread of $P$. annиa could increase, negatively affecting native flora.


Considered one of the harshest environments worldwide, Antarctica is characterized by low temperatures, high solar radiation, strong winds, a short growing season and limited water and nutrient availability (Robinson et al. 2003; Wasley et al. 2006). Antarctica has been considered less susceptible to biological invasions than other ecosystems due to its severe environmental conditions, isolation and exceptionally low species diversity (Convey 2003; Hughes et al. 2010). Nevertheless, a number of studies have documented the presence of alien species in maritime Antarctica and on the mainland (LewisSmith 1996; Frenot et al. 2005; Chwedorzewska 2008). Many of the alien species reported in these studies are
low in abundance and are restricted to disturbed sites (Chwedorzewska 2008, 2009; Chwedorzewska \& Korczak 2010; Molina-Montenegro et al. 2012; Cuba-Díaz et al. 2013). The non-native species are mainly found in the vicinity of scientific bases and tourist landing areas, suggesting that their presence and abundance are facilitated by human activities (Chwedorzewska \& Korczak 2010; Chown et al. 2012; Molina-Montenegro et al. 2012).

Human travel is occurring at an unprecedented level across the globe and is becoming a major vector for the transfer of propagules, such as seeds, eggs and spores, of species alien to the Antarctica (e.g., Frenot et al. 2005;

Lityńska-Zając et al. 2012). Introduction routes of alien organisms are largely associated with the movement of people and cargo from scientific programmes and to a lesser extent tourist operations (Frenot et al. 2005; Tin et al. 2009; Chown et al. 2012; Molina-Montenegro et al. 2012). All cargo, personal luggage, clothes and equipment of people visiting Antarctic stations can be a potentially contaminated by alien propagules. Several studies have examined the relationship between the number of alien species established in sub-Antarctica and Antarctica and the number of human visitors (e.g., Chown et al. 1998; Lee $\mathcal{E}$ Chown 2009a, 2009b). In spite of the constant increase in tourist visits mainly in the Antarctic Peninsula region, in many ways research stations could be considered to have a greater impact on the terrestrial environment. About 40000 people, mostly tourists, visit the Antarctic coast every year, whereas Antarctica has a summer population of about 4000 researchers from different countries (Frenot et al. 2005; IAATO 2013). Scientific expeditions remain in one place much longer than tourists, affecting the environment around the station continuously, and bring in large quantities of supplies and equipment (which may be contaminated with soil and propagules). Scientific activity is most intense during summer melts, when vegetation is developing and seabirds and mammals are reproducing (e.g., Chwedorzewska $\mathcal{E}$ Korczak 2010; Chown et al. 2012). The construction of buildings and other infrastructure at Antarctic stations disturbs the soil, physically and chemically, creating areas around each station that could be particularly vulnerable to human impact (Chwedorzewska \& Korczak 2010). Keeping stations supplied and maintaining infrastructure require the use of heavy vehicles, typically concentrated in a very limited area, which contributes to potential soil damage. Most stations are built in ice-free areas, which have the most favourable microclimates for the establishment of plants in the Antarctic.

Recent reviews have identified several pivotal factors in determining the establishment and spread of non-native invasive species (e.g., Richardson et al. 2000; Dietz $\mathcal{E}$ Edwards 2006; Theoharides $\mathcal{E}$ Dukes 2007). For instance, while the native community and abiotic features of the invaded habitat are important barriers for invasion (Richardson et al. 2000), disturbance is considered one of the main drivers of plant invasions (Hobbs \& Humphries 1995; Londsale 1999). In Antarctic habitats, disturbances can be caused by natural geomorphological processes such as landslides, cryogenic soil movement (Convey 1996), by deglaciation and by animals, such as seals (Scott $\mathcal{E}$ Kirkpatrick 2005; Hausmann et al. 2013). The majority of non-native species reported in the Antarctica has been found in disturbed sites surrounding scientific bases
(Olech 1996; Chwedorzewska $\mathcal{E}$ Korczak 2010; MolinaMontenegro et al. 2012; Cuba-Díaz et al. 2013) or in pioneer zones (Olech $\mathcal{E}$ Chwedorzewska 2011). Nevertheless, the establishment of alien vascular plants has recently also been reported in natural habitats (Olech $\delta$ Chwedorzewska 2011).

One of the most ubiquitous plant species in the world, Poa annua is widely established in the sub-Antarctic and maritime Antarctica (see Hughes et al. 2010). The wide distribution of $P$. annua may result from its dispersal mechanism and physiological tolerance, mainly in areas subject to human activities or in pioneer zones (Frenot et al. 1997) and in those with disturbance and manuring from animal aggregations (Walton 1975). This alien species is considerate an invasive species in Antarctica, with negative impacts on the biomass and physiological performance of native vascular species, with which it competes for resources and/or space (Molina-Montenegro et al. 2012). In this study we assess different variables related to human activities with the abundance and the successful establishment of the invasive plant species P. annua in the Antarctic Peninsula, and South Shetland Islands. We also analyse the survival percentage of $P$. annua growing in sites with different kinds of soil disturbance. Finally, we evaluate the germination percentage of the alien P. annua and the native plants Colobanthus quitensis and Deschampsia antarctica subjected experimentally to different levels of soil disturbance.

## Materials and methods

We visited 25 different sites on Antarctic islands and the Antarctic Peninsula during the Chilean Scientific Antarctic Expeditions of 2007/08 and 2009/10-the austral summer seasons-in order to search for populations of Poa annua (see Molina-Montenegro et al. 2012). In each site we visually surveyed the presence of $P$. annua around every building and point of landing (Fig. l). Each plant individual was labelled in order to avoid recording the same individuals twice, and its distance to the closest buildings was recorded. Out of all sites visited we found individuals of $P$. annua at six of them. The plant was recorded at one point at each of five sites, whereas at the Polish Arctowski Antarctic Station it was recorded at two points.

In order to find potential drivers of P. annиa establishment, we performed a regression analysis using the abundance of this species and (i) mean number of tourists landed in each point where $P$. annua has been recorded, (ii) mean number of scientists and operators landed in each point where P. annua has been recorded, (iii) distance to the main building of each station and (iv) level of soil disturbance. We used the time in years from


Fig. 1 Poa annua individual growing at Arctowski Station, King George Island, South Shetland Islands, Antarctica. (Photograph by Marco A. Molina-Montenegro, February 2010).
the last time when any building was constructed or renovated at each station as a proxy for soil disturbance. Drawing from the database of the International Association of Antarctica Tour Operators, we considered the mean number of visits (tourists and non-tourists) landed during the 2007/08 and 2009/10 summer seasons-the growing seasons that coincided with our fieldwork.

To assess the effect of different soil-patch types on establishment, we performed a transplant experiment with adult individuals of $P$. annиa collected close to the coastline (within 30-40 m to the sea). This experiment was conducted in a study area near Arctowski Station. All plants used ranged in height from 5 to 7 cm and appeared healthy. Each individual plant was excavated together with the soil around the roots (ca. 500 g ) and kept wellwatered in a plastic box under natural conditions of light and temperature for 12 h until the transplant. Plant status was visually assessed just before the transplant (plants showing foliar and/or root damage were excluded). We considered two soil-patch types for the experiment; the first type was in the bare ground between the resident vegetation (natural) and the second was in the disturbed soil surrounding the scientific stations. We selected three patches by each soil-type, with distances of 23,41 and 56 m among each pair of sites (natural-disturbed). At each patch, 10 P. annиa indivi-
duals were planted within each of three soil-patches of each type ( $n=30$ individuals per soil-type). Individuals were planted at least 5 cm apart from each other. Transplants were carried out during the 2010/11 growing season and seedling survival was evaluated after six weeks. Additionally, we compared nitrogen availability and soil moisture between five non-disturbed and five disturbed patches. A soil sample of 10 cm depth (ca. 100 g ) was taken in each patch. This depth was selected is this the average depth of the root zone of P. annua in the study. Soil samples were stored in sealed plastic bags and sent for analyses to determine the concentration of nitrate ( $\mathrm{NO}_{3}^{-}$; Robarge et al. 1983) and ammonium $\left(\mathrm{NH}_{4}^{+}\right.$; Longeri et al. 1979). Soil moisture (soil matric potential) was measured in five non-disturbed patches and five disturbed patches during the mid-growth season (January 2010). At each sampling point, a 2725 series Jet Fill Tensiometer (Soil Moisture Equipment Corp., Santa Barbara, CA, US) was dug into the soil to a depth of 10 cm . Tensiometers were placed at mid-day and, after a stabilization period of 2 h , the soil matric potential was recorded. At the end of experiment, all surviving individuals were removed from the site and destroyed.

Finally, we performed a manipulative experiment to assess the effect of soil disturbance on germination of Pou annua, Colobanthus quitensis and Deschampsia antarctica. We collected seeds from the inflorescence of the previous year's adult healthy-looking plants growing in tussocks in ice-free zones close to the coastline. At the commencement of this experiment three seeds of each species were tested to assess viability. This was done by cutting the seed in half on the equatorial axis with a razor blade after 24 h of immersion in tetrazolium chloride. All seeds were viable.

Each experimental unit was a plastic pot ( $50 \times 40 \times 30$ cm ) filled with soil extracted from the study site. Each experimental unit consisted 10 seeds of $P$. annиa or C. quitensis or $D$. antarctica ( $n=6$ pots per species; total $n=18$ pots). Seeds were buried $4-5 \mathrm{~cm}$ deep in a plastic pot, as this was the maximum depth at which P. annua seeds were found at the study site. Pots were subjected to a $20 / 4 \mathrm{~h}$ light/dark photoperiod at $4^{\circ} \mathrm{C}$ and were watered with 100 ml of water every four days. To determine the effect of soil disturbance on germination, half the pots of each species was subjected to manual disturbance in the top $4-5 \mathrm{~cm}$ of soil every three days until emergence of the first seedling (total $n=9$ pots). After the first germination, manual disturbance ceased in order to avoid seedling damage, but the recording of germination continued. The remaining pots-the controls-were undisturbed. Disturbance was deemed similar in intensity and effects to that of footfall and light vehicle traffic.

At the conclusion of the experiment, which ran for three months, we recorded the number of emerged seedlings.

## Statistical analysis

To investigate the relationship between the abundance of $P$. annиa individuals and the different human activities in the South Shetland Islands and the Antarctic Peninsula, we used standard Pearson correlations. A one-way ANOVA was performed to compare the nitrogen content, water availability and survival percentage of $P$. annиa individuals growing in soil patches with different disturbance type. To evaluate the effect of disturbance on the germination percentage in both natives and alien plant species, we conducted a two-way ANOVA. For all two-way ANOVAs, the assumptions of normality and homogeneity of variances were tested using the ShapiroWilks and Bartlett tests, respectively (Zar 1999).

## Results

During the 2007/08 and 2009/10 growing season, the presence and abundance of $P$. annиа at six different sites was recorded out of a total of 25 sites in the South Shetland Islands (one site at Deception Island and two sites at

Arctowski Station) and the Antarctic Peninsula (Bernardo O'Higgins Station, Gabriel González Videla Station, and Almirante Brown Station). Regression analysis did not show a relation between P. аппиа abundance and the mean number of tourists or non-tourists landed at each scientific station (Fig. 2a, b). However, the distance of $P$. annua individuals to the main station building showed a positive but not significant correlation (Fig. 2c), and the abundance of $P$. annua was positively and significantly correlated with the number of years elapsed since any building was constructed or renovated (Fig. 2d).

In the transplant experiment, the survivorship of P. annиa individuals was significantly higher ( $F_{1,8}=6.11$; $P=0.039$ ) in sites with soil disturbance than without disturbance (Fig. 3). Soil moisture was not different ( $F_{1,8}=28.52 ; P=0.09$ ) between disturbed and nondisturbed patches of soil $(-25 \pm 4$ and $-30 \pm 6 \mathrm{MPa}$ disturbed and non-disturbed, respectively). In contrast, the available nitrogen content of the disturbed soil was significantly higher ( $F_{1,8}=231.21 ; P<0.01$ ) than in nondisturbed conditions ( $7.56 \pm 1.2$ and $3.98 \pm 1.6 \mathrm{mg} \cdot \mathrm{K}^{-1}$ disturbed and non-disturbed, respectively).

Overall, the percentage of germination was significantly increased under the disturbance treatment ( $F_{1,24}=563.33$; $P<0.001$ ), but this trend was not similar for all species


Fig. 2 Analysis of correlations between abundance of Poa annua recorded at different sites and (a) annual mean number of tourists landed, (b) annual mean number of scientists landed, (c) distance to main building of each site, and (d) time in years since last time that a building was built or renovated in each site. We considerate in this analysis all sites containing some P. annua individuals: General Bernardo O'Higgins Station in northern Graham Land, Almirante Brown Station at Paradise Bay, Gabriel González Videla Station at Paradise Bay, two sites at Henry Arctowski Station at King George Island, and Deception Island.


Fig. 3 Survival percentage (mean $\pm 2 E E$ ) of Poa annua individuals growing in soil patches without and with disturbance.
$\left(F_{2,24}=364.25 ; ~ P<0.001\right)$. For $P$. annua the germination increased with disturbance, for $C$. quitensis germination decreased significantly under disturbance while for D. antarctica the germination percentage was not different between the disturbance and control treatments (Fig. 4).

## Discussion

In the 21 st century, the number of visitors to the Antarctica and the scale of human influence have increased dramatically, mainly due to tourism (IAATO 2013). There has been concern about the potential impact of visitors on the Antarctic environment (Culik $\mathcal{E}$ Wilson


Fig. 4 Germination percentage (mean $\pm$ SD) of the natives Colobanthus quitensis and Deschampsia antarctica and the alien Poa annua subjected to manipulative experiments without and with soil disturbance. Asterisks indicate significant differences (a posteriori Tukey test $\alpha=0.05)$.

1995; Chown et al. 2012). Several studies have aimed to detect the effects of this visitor pressure (Chwedorzewska \& Korczak 2010).

We did not find a positive correlation between Poa annua abundance and the mean number of tourists or scientists visiting the Antarctic. However, a positive relationship between the abundance of $P$. annиa and the time since a base underwent construction suggests that this kind of human activity influences the establishment phase of this species. We believe that human activity associated with research stations enhances soil nutrients and modifies the soil matrix, and therefore facilitates the germination and establishment of $P$. annua. Several studies have shown that cargo and vehicles transported to Antarctic research stations contain propagules of many plants (Whinam et al. 2005; Lee $\delta$ Chown 2009b; Chown et al. 2012). Hughes et al. (2010) highlighted that research station construction also facilitates the transport of non-native seeds. We found that time since construction influences the presence of $P$. annua, most likely because more time allows for the establishment and population expansion of the plant. In Antarctica, ice-free areas near the coast are rather scarce and are hotspots for local fauna and flora (Convey 1996) as well as humans (Chown et al. 2012). High traffic around research stations creates a high level of soil disturbance, which can facilitate the establishment of exotic plants (Chwedorzewska 2008; Cuba-Díaz et al. 2013), as our experiments support. In stressful environments such as those found in the Antarctic terrestrial ecosystems, it has been predicted that alien species are unlikely to colonize and extend their range of distribution unless some process relaxes the environmental constraints (Dullinger et al. 2003; Pauchard et al. 2009).

It is widely assumed that soil disturbance enhances resource availability (e.g., Burke \& Grime 1996; Davis et al. 2000), and some have examined the effect of disturbances in cold environments and shown increased nutrients, or higher accessibility, following disturbance (Stanton et al. 1994; Grieve 2000). We have elsewhere shown that alien species take advantage of high resource availability (e.g., nutrient or water), improving their ecophysiological performance (Molina-Montenegro et al. 2011). In the study reported here, we found that disturbed soils had higher levels of nutrients, as well as germination and survival percentage of $P$. annua, suggesting that disturbance by humans may generate microhabitats more favourable to P. annua than to native plants. Alternatively, the enhanced establishment and survival of $P$. annиa in disturbed areas could result from non-random spatial distribution due to wind. In cold environments some plant
species show greater establishment and survival percentages when sheltered from the desiccating effect of wind (Resler et al. 2005). In Antarctica P. annua has been shown to become established at sites sheltered from wind, such as alongside buildings (Olech 1996). More surveys should be conducted to further explain the spatial distribution and performance of $P$. annua in Antarctica.

Since it was first recorded in the mid-1980s (Olech 1996), P. annua is the only introduced plant species to have established permanent scattered populations in the Shetland Islands and on the Antarctic Peninsula (Frenot et al. 2005; Chwedorzewska 2008, 2009; MolinaMontenegro et al. 2012). The species has shown a steady, constant increase in abundance and number of new populations (Molina-Montenegro et al. 2012). Having spread to alpine ecosystems (Johnston \& Pickering 2001), northern Europe (Gederaas et al. 2012) and Sub-Antarctic islands (Frenot et al. 2005), P. аппиа is "generalist" invasive species that tolerates stressful environments and takes advantage of favourable conditions, e.g., enhanced nitrogen availability in disturbed soils.

Surprisingly, nowadays there is no international policy to control the introduction of non-native species to Antarctica. Our study provides evidence that Antarctic environments could be severely impacted in the near future by such introductions and in the long-term they may have unsuspected consequences on Antarctic diversity. Although, some efforts have been made to accomplish this goal, they are still insufficient to mitigate the negative impacts of human activities on Antarctic biodiversity (see Hughes \& Convey 2014).

In summary, $P$. annua is positively correlated with human activities and its survival and germination is enhanced in soil disturbance conditions. We conclude that human impacts, mainly associated with research activities, may have negative effects on the native species but positively influence the abundance and spread of the invasive P. annиa on the Antarctic continent.

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