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Control measures for a recent invasion of *Hieracium pilosella* in Southern Patagonian rangelands

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Summary

Plant invasions have important ecological impacts on biodiversity, the functioning of ecosystems and economic sustainability. In this study, we evaluated the effects of four control measures (pasture sown + fertiliser, fertiliser and selective/non-selective herbicide applications) in two different grazing conditions (grazed and ungrazed) during a recent invasion of the exotic herb *Hieracium pilosella* in northern grasslands of Tierra del Fuego Island in Southern Patagonia, Argentina. As response variables, we measured the cover of the invasive species, the dominant growth forms of other plant species, litter and bare soil at patch scales (m^2) during two consecutive growing seasons. The effects of fertilisation depended on the grazing conditions; *H. pilosella* cover decreased by more than 92% and was replaced by dicotyledonous herbs in the ungrazed/fertilised subplots, while it exhibited no decrease in the grazed/fertilised subplots after the second growing season. Both herbicides (selective and non-selective) reduced *H. pilosella* cover by *c.* 63% compared with the untreated subplots independently of grazing. However, the non-selective herbicide application resulted in an increase in bare soil and litter cover in the treated grazed and ungrazed subplots respectively. In contrast, such effects were not observed with the selective broad-leaved herbicide application. A control strategy based on the local application of selective herbicides and/or NP fertilisers, in conjunction with a transient ban on sheep grazing, reduced the invader's cover in the short term and at a local scale and also reduced the cover of bare soil through the restoration of native vegetation. An economic assessment of this strategy supported the profitability of these control measures.

Keywords: mouse-ear hawkweed, disturbance, grassland, pasture, plant invasion, rangeland management, resource availability, sheep grazing.

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Introduction

Invasions by plant species represent a significant threat to biodiversity conservation (Diaz *et al.*, 2006), the functioning of ecosystems (Vilà *et al.*, 2010) and economic sustainability (Mack *et al.*, 2000) for natural systems around the world. Thus, the study of changes in the abundance and distribution of invasive plants in novel habitats and the identification of the environmental factors involved are fundamental to elucidate causes and minimise consequences of plant invasions (Drake *et al.*, 1989). However, it is equally important to assess and develop control tools, which are particularly important during the early stages of invasions.

Grasslands are ecosystems particularly prone to invasion, owing to anthropogenic disturbances associated

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with crop cultivation and livestock grazing (Hobbs & Huenekke, 1992; Buckland *et al*., 2001). Grazing can promote the introduction and expansion of exotic plants, because of selective feeding behaviour, changes in the balance of biotic interactions between native and exotic species, soil disturbance such as trampling and increases in the dispersal range of plants by herbivores (Bellingham & Coomes, 2003). Moreover, soil tillage for crop cultivation or different grazing management practices (e.g. sown pastures) may facilitate the colonisation of invasive weed species, owing to the fact that they disturb soil (Hobbs & Huenekke, 1992; Walker *et al*., 2005). Invasive weeds in rangelands generally reduce forage yield and quality, increase management costs and impair animal performance (DiTomaso, 2000).

The perennial herb *Hieracium pilosella* L. (mouse-ear hawkweed, Asteraceae) is a grassland species native to Europe and West Asia (Bishop & Davy, 1994). This species is highly competitive and rapidly spreads by forming mono-specific patches (i.e. carpets) through vegetative growth (Moen & Meurk, 2001). This species has invaded grasslands in New Zealand (Allan, 1924), the USA (Voss & Böhlke, 1978), Chile (Domínguez, 2004) and, recently, Tierra del Fuego Island in southern Argentina (Livraghi *et al*., 1998). The main economic impact of this weed species in novel habitats is a decrease in the forage biomass and secondary productivity, caused by its replacement of native and palatable species of grasslands (Makepeace, 1985a,b; Makepeace *et al*., 1985; Johnstone *et al*., 1999). *Hieracium pilosella* is generally avoided by sheep, owing to its prostrate growth, highly pubescent leaves and high concentrations of secondary metabolites (Makepeace *et al*., 1985; Bishop & Davy, 1994).

In New Zealand, *H. pilosella* gradually expanded its geographic distribution over a long period (1920–1963) but remained locally rare. The spread of *H. pilosella* in New Zealand was attributed to the presence of overgrazed pastures and fescue grasslands (Connor, 1992; Rose *et al*., 1998; Johnstone *et al*., 1999). However, the cause of the expansion remains controversial, as other studies have found no strong connection between the invader's expansion and grazing (Meurk *et al*., 2002; Rose & Frampton, 2007) or disturbance (Walker *et al*., 2005).

In Tierra del Fuego Island (Argentina), the current invaded region is dominated by fescue grasslands (*Festuca gracillima* Hooker f.) and scrublands (*Chiliotrichum diffusum* (Forster f.) O. Kunze) (Livraghi *et al*., 1998). It is an area that has been subject to high grazing levels by sheep for the past 100 years (Anchorena *et al*., 2001). *Hieracium pilosella* was first reported in 1993, and in the following years, its cover increased rapidly at particular sites and spread across the entire grassland region (*c*. 5000 km^2). At present, despite the low regional cover of the species ($<2\%$), the frequency across the whole grassland region is quite high (60%), with several foci or hotspots where the invader has achieved high cover (10–70%; Cipriotti *et al*., 2010). These foci encompass areas of *c*. 10–100 ha, where several *H. pilosella* patches (1–20 m diameter) commonly replace native species and form a network in the inter-tussock or inter-shrub spaces. Native short graminoids (e.g. *Poa pratensis* L., *P. spiciformis* (Steud.) Hauman & Parodi, *Hordeum pubiflorum* Hook., *Deschampsia flexuosa* L. (Trin.), *Bromus catharticus* Vahl, *Festuca magellanica* Lam, *Rytidosperma virescens* (E.Desv.) Nicora, among many others) and naturalised vegetation (e.g. *Trifolium repens* L.) growing in the interspaces are highly diverse and palatable (Posse *et al*., 1996) and thus fundamental for biodiversity conservation and livestock management.

The *H. pilosella* invasion in Tierra del Fuego rangelands, a system largely dependent upon natural pastures for its economic sustainability (Anchorena *et al*., 2001), is expected to have a substantial economic impact in the long term, as has occurred in New Zealand (Rose *et al*., 1998; Meurk *et al*., 2002). Additionally, the main ecosystem impacts of the *H. pilosella* invasion on the Fuegian steppe may be related to changes in nutrient cycling, owing to the increased decomposition rates of the invader's litter (Braun, 2009). The local changes in the soil biogeochemistry promote the self-perpetuation of *H. pilosella* and lead to a loss of biodiversity (Boswell & Espie, 1998).

Appropriate measures to contain and control weed invasions should promote the regeneration of native species, alter the disturbance regime and/or modify the availability of resources among the species within the recipient community (Firn *et al*., 2008). Unfortunately, most of the control methods used in New Zealand were generally assessed at late stages of the *H. pilosella* invasion. There is thus a significant gap in our current knowledge about the effectiveness of control measures and the role of grazing during early stages of this plant invasion for cold temperate grasslands. Although it has spread regionally, the invasion of *H. pilosella* in the Argentinean portion of Tierra del Fuego is still at an early stage (Cipriotti *et al*., 2010), making this the ideal opportunity to implement policies to mitigate the effects of the invasion and prevent it from worsening (Radosevich *et al*., 2003). In this study, we evaluated how different control measures mitigated the effects of the *H. pilosella* invasion under two different grazing regimes. Our final goal was to identify a strategy to constrain the current invasion of *H. pilosella*, one which would take into account the role of grazing role and restore the natural vegetation communities, at least in the short term. We hypothesised that during the early stages of invasion, grazing would interact with control measures in different ways, modifying the response of the invasive species and limiting the restoration of vegetation communities.

Materials and methods

Study site

The Fuegian steppe covers about 5000 km^2 from the Strait of Magellan to approximately 54°S, at the northern end of Tierra del Fuego Island. It is the southernmost portion of the Magellanic steppe (Hueck & Seibert, 1972), a rare example of cold temperate oceanic grasslands in South America. In a regional context, the area can be viewed as a transition between the arid Patagonia and the humid sub-Antarctic climates (Collantes *et al.*, 1999). At Río Grande, a city on the Atlantic coast (53° 47′ 27″ S, 67° 42′ 46″ W), the mean temperature during the coldest month (July) is 0°C and then rises to 10°C during the warmest month (January). Precipitation is evenly distributed throughout the year and decreases from the SW (400 mm per year at the south of Río Grande city) to the NE (260 mm per year at Cabo Espíritu Santo) away from the Andes range, creating a rain shadow (Collantes *et al.*, 1999). There are noticeable water balance deficits in December and January. The vegetation has been classified as that of a humid grass steppe (Collantes *et al.*, 2005). Highlands are covered by a tussock steppe of *Festuca gracillima* Hooker f. that blends into a scrub of *Chiliotrichum diffusum* (Forster f.) O. Kunze or into dwarf shrub heaths of *Empetrum rubrum* Vahl ex Willd, while lowlands are covered by wetland communities dominated by hygrophilous vegetation. Following European settlement during the 19th century, land use has been dominated by sheep farming for wool and meat (Martinic, 1982).

Field experiment

In October 2005, we set up a field experiment at the Cullen Ranch in the north of Fuegian steppe (52° 55′ S, 68° 33′ W) in a large paddock (*c.* 1220 ha). In the same paddock, shrubs had been mechanically removed, and forage species (e.g. *Festuca arundinacea* Schreb., *Dactylis glomerata* L., *Trifolium repens* L.) had been sown in the 1980s to improve the quality of the pasture. We selected six homogeneous areas of 1 ha (plots) inside the paddock with a large number of *c.* 2 m diameter *H. pilosella* patches. We buried long steel tubes (1.6 m) in the four corners of each plot to easily identify them during fieldwork. Three of the six delimited areas were selected at random, and a permanent fence 1.4 m high was constructed with five wires to exclude livestock from the selected plots. We considered the three enclosed areas the 'ungrazed' plots, while the three adjacent unfenced areas were the 'grazed' plots. Before setting up our experiment, all six field plots (the grazed and ungrazed) had a similar stocking rate. In addition, all plots had a similar terrain slope, initial vegetation cover and composition, including *H. pilosella* cover (*c.* 35%). Sørensen's similarity index (Sørensen, 1957) based on vegetation census was higher than 70% for all possible pairwise comparisons of vegetation among the plots.

We set up five subplots (5 × 5 m) randomly with representative vegetation on all six plots but made sure that each had at least one large *H. pilosella* patch (diameter *c.* 2 m) in the centre, resulting in a total of 30 subplots. Each subplot was delimited by burying a wood marker (40 cm) in each corner. In addition, each subplot was identified by a number and colour label on the south-western wood marker to indicate its respective treatment. We left a minimum distance of 10 m as corridors between subplots. From the grazing exclusion plots, we left also a minimum distance of 2 m between the fence and the subplots to avoid any grazing by sheep through the fence. To characterise the initial invasion of the subplots before the treatments, we measured the cover of the *H. pilosella* and that of the main plant growth forms (i.e. grasses, shrubs, dwarf shrubs and dicotyledonous forbs) on two permanent transects according to Cingolani *et al.* (2005). In addition, we took photographs and drew maps with the locations and shape of the *H. pilosella* patches in each subplot. The largest *H. pilosella* patch (two orthogonal diameters) on each subplot was measured, and a permanent quadrat (12 × 12 cm) was set inside at the patch's centre to count ramets and stolons. This quadrat was also delimited by burying four wood markers (25 cm) at the corners.

A split-plot design was used for the statistical analyses of the field experiment, where four treatments and the untreated control were assigned randomly to the subplots after the initial randomisation of the grazing condition (plots). The treatments were the following: (i) untreated control, (ii) pasture sown (10:5:5 kg ha^{-1}, *Dactylis glomerata:Trifolium repens:Trifolium pratense*) + NP fertilisation with phosphate ammoniac (200 kg ha^{-1} $PO_4H[NH_4]_2$, 18-46-0), (iii) NP fertilisation with phosphate ammoniac (200 kg ha^{-1} $PO_4H[NH_4]_2$, 18-46-0), (iv) selective broad-leaved herbicide application [2,4 DB ester (93% a.e.) at 1.5 L ha^{-1}] and (v) non-selective herbicide application [Glyphosate© (48% a.e.) at 1 L ha^{-1}]. These control measures were selected because there is evidence that they are capable of containing the invasion of *H. pilosella* in similar grasslands (Scott, 1993; Woodman *et al.*, 1997; Norton *et al.*, 2006). The fertilisation and pasture sown treatments are aimed at improving native or naturalised vegetation, while herbicide applications focus on removing the invasive species. The grass–legume forage species mixture selected for the experiment is used frequently across the region to improve rangeland grazing quality. In addition, the *Trifolium* species are effective competitors against *H. pilosella* (Scott & Sutherland, 1993). Response variables were the cover of the invasive species, dominant growth forms (Cingolani *et al*., 2005), litter and bare soil, all of which were measured on the subplots at the end of the two consecutive growing seasons (in February 2005–06 and 2006–07). All cover measurements were taken using the line intercept method along two orthogonal transects. Additionally, from small quadrats (12 × 12 cm) installed in the centre of *H. pilosella* patches, we quantified the number of rosettes, stolons and head flowers of the species on the same dates and subplots.

The herbicide applications were carried out using a portable backpack sprayer with a tapered-edge flat spray nozzle (spray angles 15° to 110°) that produced medium-sized drops (2–4 mm). In addition, the sprayer had a plastic spray shield (50 × 12 cm) around the nozzle to avoid herbicide drift. The herbicide applications were carried out before midday to avoid the high temperatures in the afternoon. Before sowing, we harrowed the top soil layer (5–10 cm) with a disc harrow and then added the seeds. Fertilisers and seeds were applied by broadcast application covering the whole subplot area in a uniform way. Seed mass was set according to an initial seed density of *c*. 1000 seeds m^{-2} for the grass species and 300 seeds m^{-2} for each clover species. Seeds of both legume species used were inoculated and pelleted in advance with a commercial product (Ribol©) to facilitate manipulation. Ammonium phosphate fertiliser was applied as medium-sized (1–2 mm) solid crystalline grains. At the beginning of the second growing season (October 2006), the fertilisers and the herbicides were applied again to the respective subplots following the same protocol of the previous year.

Statistical analysis

The experiment was based on a split-plot design with a whole plot used to determine the grazing effects (two levels: grazed and ungrazed) and the subplots used for control measures (five levels according to the treatments). We used an ANOVA model for split-plot designs with subplots nested within each plot, with grazing and control measure as the fixed effects and error as the random effect. In addition, we included a covariate term to the statistical model to take into account the possible initial differences in abundance of *H. pilosella* among subplots. Given that it was not our goal to study interannual variation and we were not able to do so with only 2 years of data, we performed two separate analyses for each growing season (years 2006 and 2007). Percentage and count variables (e.g. number of ramets or stolons) were previously transformed as the log(x) of the original variable (x) to restore normality. When we detected statistically significant effects for the main factors or terms of interaction, the *post hoc* comparisons of the different treatments were made using Fisher's LSD tests with a significance level (alpha) of 5%.

Results

The overall results showed that the NP fertilisations caused a reduction dependent on grazing, while the application of both herbicides (selective and non-selective) had an effect independent of grazing on *H. pilosella* abundance (Fig. 1). The *H. pilosella* cover in fertilised/ungrazed subplots (with or without added forage seeds) decreased by at least 70% with respect to the untreated control during the first year, while no weed cover decrease was observed in the fertilised/grazed subplots (Fig. 1A). Moreover, at the end of the second year, the cover of *H. pilosella* in the fertilised/ungrazed subplots was lower than 3%, representing a decrease of around 92% in the initial mean value of the paddock (ca. 35%; Fig. 1D). In addition, the measurements on the permanent quadrats showed that after the second growing season, the decrease in the *H. pilosella* cover was associated with a similar decrease in its number of ramets (Fig. 1E) and stolons (Fig. 1F). This decrease in *H. pilosella* cover in the fertilised/ungrazed subplots was accompanied by an increase in the dicotyledonous herbs' cover (Fig. 2B), triggered mostly by the white clover (*Trifolium repens*) response (91 ± 3.2% of the total cover of dicotyledonous herbs).

Both herbicides reduced *H. pilosella* cover by *c*. 63% compared with the untreated control values, independently of grazing and to an extent similar to the fertilised/ungrazed subplots (Fig. 1). However, herbicides differed in terms of their effects on the community and ecosystem after the 2 years (Fig. 2). Bare soil cover increased more than five times on the grazed subplots treated with the non-selective herbicide, compared with the same treatment on the ungrazed subplots (Fig. 2D). In contrast, the subplots treated with the selective broad-leaved herbicide exhibited a significant increase in the grass cover (Fig. 2A) and almost no bare soil cover (Fig. 2D), regardless of whether there was grazing on the site. On the other hand, the litter cover in ungrazed subplots treated with the non-selective herbicide was high (88.3%) and it increased by *c*. 59%

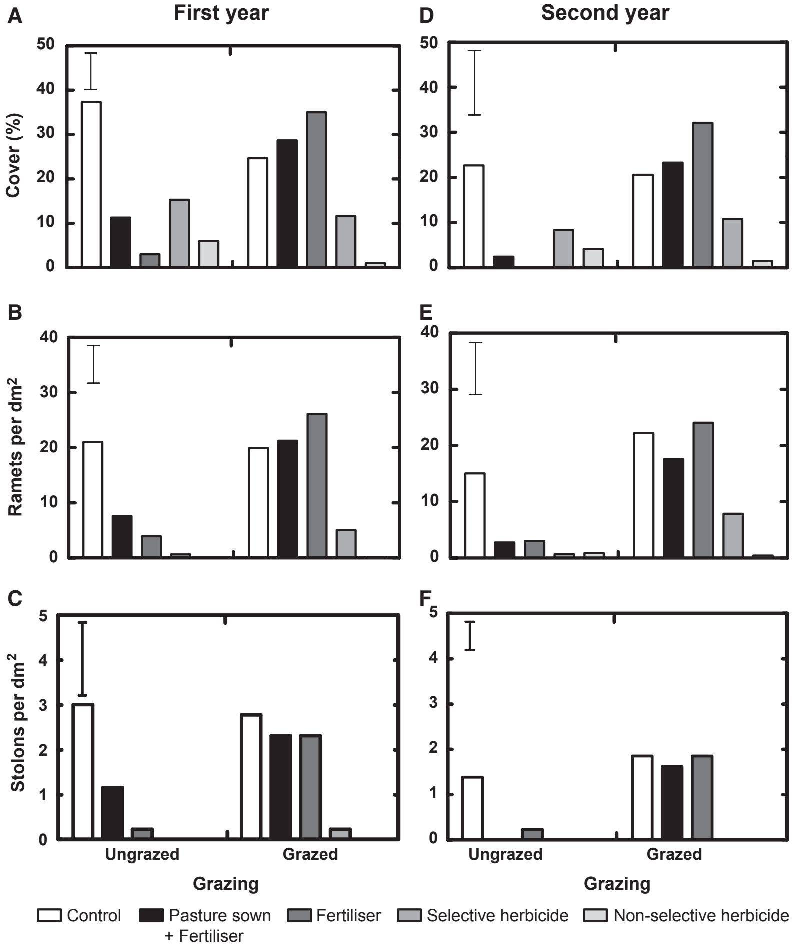

Fig. 1 Percentage cover (A, D) and number of ramets (B, E) and stolons (C, F) of *Hieracium pilosella* under five treatments in ungrazed and grazed plots after the first (A, B, C) and second (D, E, F) year of the experiment. Wide bars indicate means, while vertical narrow bar in the upper left corner of each panel indicates LSD (5%). dm^2, decimeter square.

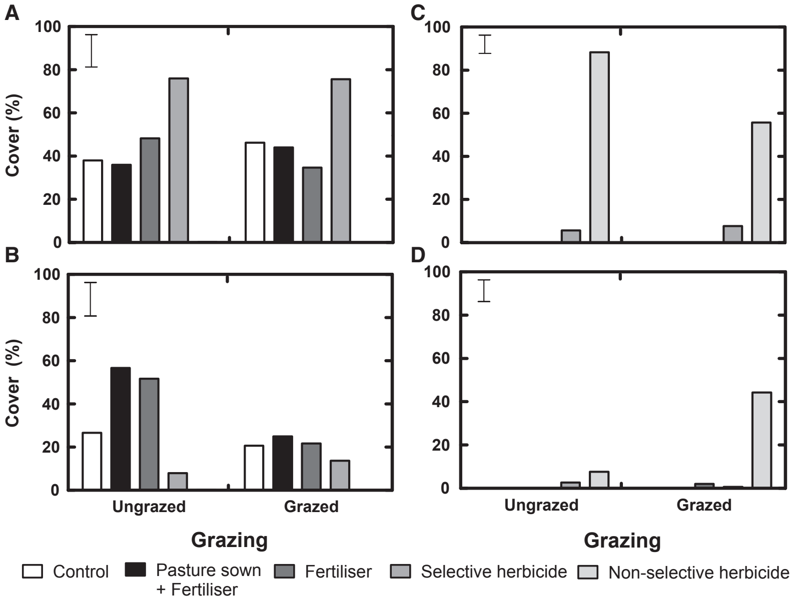

Fig. 2 Percentage cover of grasses (A), dicotyledonous herbs (B), litter (C) and bare soil (D) under five treatments in ungrazed and grazed plots after the second year of experiment (2007). Wide bars indicate means, while vertical narrow bar in the upper left corner of each panel indicates LSD (5%).

with respect to grazed subplots, while it was very low (5–8%) and showed no differences in terms of grazing for the subplots treated with the selective herbicide (Fig. 2C).

Discussion

Increases in the nutrient availability (NP additions) and grazing exclusion enhanced native and naturalised vegetation growth and resulted in a decrease in the *H. pilosella* cover of the invaded plots. These results corroborate our original hypothesis about the control measure and the interactive effects of grazing, but they contradict previous studies on the ability of *H. pilosella* to capture high-resource pulses (Boswell & Espie, 1998). Although similar control measures on the invasion of *H. pilosella* have been described for other grasslands of the world (Scott, 1993; Woodman *et al.*, 1997), interactive effects between these common control measures (e.g. fertiliser, herbicide applications, sown pastures, etc.) and sheep grazing have not been reported for this species in the past (Scott, 1993; Meurk *et al.*, 2002; Rose & Frampton, 2007).

Field experiments on control measures for this exotic species in New Zealand (e.g. management, chemical and biological control) under different grazing regimes have been thoroughly documented. In most cases, the results indicated that the cover of *H. pilosella* was decreased in the short and the long term through intensive pasture management, using fertilisers, herbicides and sown forage seeds (Scott, 1993; Scott & Sutherland, 1993; Norton *et al.*, 2006). However, the decrease in *H. pilosella* cover achieved through pasture management was mostly independent of grazing (Scott, 1993; Meurk *et al.*, 2002; Walker *et al.*, 2005; Rose & Frampton, 2007).

The interactive responses between control measure and grazing found in this study may be related to the invasion stage of *H. pilosella* in this ecosystem and to the role of plant competition, which varies depending on grazing. The invasion of *H. pilosella* in the Fuegian steppe is relatively recent (< 15 years, Livraghi *et al.*, 1998) in comparison with this species' invasion in New Zealand or USA grasslands (> 50 years; Voss & Böhlke, 1978; Connor, 1992). At the early stages of the invasion, when the weed cover is low and there is a matrix of interspaces occupied mainly by highly palatable plant species (Posse *et al.*, 1996), grazing may reduce the intensity of plant competition with the matrix species. Therefore, the application of fertilisers and grazing exclusion may promote the growth of established competitors (e.g. native or naturalised dicotyledonous or grass species from the inter-tussock matrix), resulting in a decrease in *H. pilosella* patch growth. However, at the advanced invasion stages (i.e. big carpet-like patches of *H. pilosella,* where matrix species are almost absent), grazing exclusion may not favour any competitor species at the local scale.

Particular weather conditions during the course of our studies may have affected the magnitude, although not the direction of the effect of added fertiliser. During the first year of the field experiment, rainfall at the beginning (November 2005) and in the middle (January 2006) of the growing season was a 287% and 157% higher than the respective historic monthly means (Collantes *et al.*, 2005). These high rainfalls may have enhanced the effect of fertilisers applied in October and the consequent growth of plant competitor species, particularly the response of the naturalised white clover among other dicotyledonous herbs. White clover is a highly water-demanding species and also one of the best competitors of *H. pilosella* (Scott & Sutherland, 1993).

Both evaluated herbicides, selective and non-selective (2,4 DB and glyphosate respectively), were quite effective in reducing the *H. pilosella* cover independently of grazing. However, their effects on the community and ecosystem differed and interacted with sheep grazing. Non-selective herbicide application led to a litter increase on ungrazed plots and to an increase in bare soil in the grazed plots after the second year. The litter generated by the application of this herbicide was probably transformed into bare soil through sheep trampling (Posse *et al.*, 2000) or other mechanical effects in grazed plots. By contrast, no litter or bare soil increase was observed with the selective herbicide application, regardless of grazing. Moreover, this herbicide favoured the recovery of inter-tussock palatable grasses on ungrazed plots. Maintenance of plant cover is a very important issue on these sub-humid rangelands to avoid soil erosion, which results in grassland degradation and productivity loss (Anchorena *et al.*, 2001; Collantes *et al.*, 2005). Moreover, the aim of controlling *H. pilosella* invasion should involve bare soil reduction, given that bare soil has been related to the seed recruitment of this invader and its successful colonisation of new areas (Winkler & Stöcklin, 2002). Hence, the application of selective broad-leaved herbicide represents a more soil-conservative approach to control, enabling the growth of native competitors like short grass species.

Hieracium pilosella is an aggressive weed that has caused severe ecological and economic problems in specific regions of New Zealand (Meurk *et al.*, 2002). At the advanced invasion stages, the resident vegetation is completely replaced. In fact, there is a particular concern regarding native flora loss owing to the invasion by European hawkweeds in North America and New Zealand (Wilson & Callihan, 1999). In the Fuegian steppe, the invasion by *H. pilosella* represents the introduction of a life form with very different functional traits compared with the resident species (Braun, 2009). Litter of *H. pilosella* is more labile (i.e. higher nutrient and lower fibre content) and decomposes at much higher rates compared with dominant native litter (e.g. *C. diffusum, P. spiciformis* or *F. gracillima*) (Braun, 2009). Higher decomposition rates could result in ecosystem impacts (e.g. higher CO_2 release to the atmosphere and soil organic matter loss) and in locally higher soil nutrient availability, which may favour *H. pilosella* colonisation owing to its ability to capture pulses of high nutrient concentration. Thus, the invader species favours its own invasion by a positive feedback or 'switch' through changes in soil biogeochemistry, as it has been reported for other plant invasions (Wilson & Agnew, 1992; Boswell & Espie, 1998).

In this scenario, the recent mouse-ear hawkweed invasion in Tierra del Fuego Island challenges rangeland management practices aimed at the restoration of the ecosystem and invasion mitigation. In this study, we studied different control measures that may be effective at achieving both of these aims. The local application of selective herbicides or the combination of NP fertilisations and grazing exclusion may result in a comprehensive control strategy to mitigate the recent *H. pilosella* invasion in the Fuegian steppe in the short term. Moreover, this management proposal is backed up by a recent economic analysis on similar control measures made at farm scale in medium-sized ranches invaded by *H. pilosella*, which showed that the use of herbicides is more cost-effective compared with the NP fertilisations (*c.* 120 vs. 450 US $\$ \text{ha}^{-1}$) owing to the cost of agrochemicals (Cabezas *et al.*, 2011). Finally, the local control measures proposed here should be integrated within regional rangeland management. Because bare soil has been related to invasion patches formation (Cipriotti *et al.*, 2010), it is necessary to promote pasture labour practices that minimise soil disturbance (i.e. minimum tillage or none at all) and quick sowing and vegetation restoration in the case of extensive soil disturbance (e.g. roads or oil pipeline construction).

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