Acacia saligna's soil legacy effects persist up to 10 years after clearing: Implications for ecological restoration

MLUNGELE M. NSIKANI,[1]* ANA NOVOA,[1,2,3] BRIAN W. VAN WILGEN,[1]
JAN-HENDRIK KEET[1] AND MIRIJAM GAERTNER[1,4,5]

[1]*Centre for Invasion Biology, Department of Botany and Zoology, Stellenbosch University, Private Bag X1, Matieland 7602 (Email: mllue06@gmail.com),* [2]*Invasive Species Programme, South African National Biodiversity Institute, Kirstenbosch Research Centre, Claremont, South Africa,* [3]*Department of Invasion Ecology, Institute of Botany, The Czech Academy of Sciences, Průhonice, Czech Republic,* [4]*Invasive Species Unit (ISU), Environmental Resource Management Department (ERMD), City of Cape Town, Westlake Conservation Office, Tokai, South Africa, and* [5]*Nürtingen-Geislingen University of Applied Sciences (HFWU), Nürtingen, Germany*

Abstract To reduce the negative impacts of invasive plants, management interventions such as control or eradication are usually necessary. It is often assumed that the impacts of invasive plants will diminish immediately after such interventions. However, in some cases the invader can have legacy effects in the soil that might persist for long periods, preventing the natural restoration of the areas managed. Therefore, to achieve the re-establishment of a functional native ecosystem it is important to understand for how long such legacies can persist in the soil. This paper explores this issue, using *Acacia saligna* in South Africa as case study. We collected soil samples in invaded, non-invaded and previously invaded sites (representing 2, 6 and 10 years after clearing) and analysed the levels of pH, carbon, nitrogen, available phosphorus, ammonium, nitrate and electrical conductivity. We also analysed enzyme activities (β-1,4-glucosidase, urease and acid phosphatase). *Acacia saligna* invasion alters overall soil characteristics but specifically raises pH by 0.6–1.8. Moreover, soil characteristics (e.g. pH) are not restored to natural conditions after control (soil legacy effects persist up to 10 years after clearing). Furthermore, *A. saligna* control elevates soil NO_3^- levels and these can remain higher than in invaded (1.55–6.67 mg kg^{-1}) and non-invaded (2.16–4.35 mg kg^{-1}) sites up to 10 years after clearing. Elevated NO_3^- often facilitates secondary invasion and/or weedy native species dominance which may hinder the restoration of functional native ecosystems. Therefore, strategies to manage areas previously invaded by *A. saligna* should take into account the removal of litter from the target invader, secondary invaders and weedy native species.

Key words: ecological impact, enzyme activities, invader control, legacy effects, restoration.

INTRODUCTION

Alien plant invasions are a global problem arising from the widespread translocation of plant species beyond their natural ranges to new areas where some of them become invasive (Richardson *et al.* 2011). Some invasive plants transform the ecosystems they invade (*sensu* Richardson *et al.* 2000; Ortega & Pearson 2005) leading to negative impacts on native biodiversity, ecosystem function and ecosystem services (Vitousek *et al.* 1997; Pyšek & Richardson 2010). To reduce these negative ecological impacts, management interventions such as control or eradication are often necessary (Pickart *et al.* 1998; Malcolm *et al.* 2008; Moran & Hoffmann 2012; Akamatsu *et al.* 2014).

It is often assumed that the negative impacts of invasive plant species will diminish immediately after such interventions (Wittenberg & Cock 2005).

However, this is not always the case. For example, the invader can leave legacy effects in the soil (e.g. altered soil chemical or physical characteristics, or altered soil microbial communities) that might persist after its removal thereby preventing the natural restoration of the area (*sensu* Corbin & D'Antonio 2004; Marchante *et al.* 2009). In such cases, removal of the invader alone may not be sufficient to return the ecosystem to its original state (Macdonald 2004; Marchante *et al.* 2009) – for example legacy effects in dune soils after the removal of the invader *Carpobrotus edulis* can facilitate the establishment of opportunistic species that out-compete native dune species, preventing their re-establishment (Novoa *et al.* 2013).

If soil legacy effects persist after removal of the invader, they can (i) facilitate re-invasion by the same invader or secondary invasion (Yelenik *et al.* 2004; Malcolm *et al.* 2008; González-Muñoz *et al.* 2012); (ii) prevent native plant species recovery and re-establishment (Maron & Jefferies 1999) and (iii) be self-reinforcing and appear to promote an 'alternative

stable state' which is difficult to successfully restore to pre-invasion conditions (Marchante *et al.* 2008; Suding *et al.* 2013; Gaertner *et al.* 2014).

Acacia saligna (Labill.) H.L Wendl. (Fabaceae) was introduced in various parts of the world for a wide range of commercial, subsistence and ornamental uses (Wilson *et al.* 2011). It is currently invasive in parts of Algeria, Chile, Cyprus, Israel, Italy, Kenya, Morocco, Portugal, South Africa and Spain (Thompson *et al.* 2015), covering an estimated area of 600 000 ha worldwide (Griffin *et al.* 2011). In South Africa, *A. saligna* covers approximately 53 000 ha (Van Wilgen *et al.* 2011). In the invaded ecosystems, *A. saligna* alters soil characteristics – mainly through nitrogen fixation, litter production (Musil & Midgley 1990; Witkowski 1991; Musil 1993; Yelenik *et al.* 2004) and changes in soil microbial community structure, diversity and function (Crisóstomo *et al.* 2013). In fact, the invasion of *A. saligna* in South Africa has resulted in the development of extensive woodlands dominated by the alien species and a corresponding decrease in native plant diversity (Le Maitre *et al.* 2011; Van Wilgen *et al.* 2011; Gaertner *et al.* 2012).

There are numerous efforts to globally control *A. saligna* invasions (Le Maitre *et al.* 2011) and some of them are successful in reducing its distribution (Van Wilgen *et al.* 2011). For example, a fire of moderate intensity is often applied after clearing acacias to (i) consume the litter – *A. saligna*'s litter can supplement soil nitrogen and/or form a physical barrier to re-establishing native seedlings (Richardson & Kluge 2008), and (ii) reduce the amount of *A. saligna*'s seeds in the seed bank – by destroying the seeds or triggering mass germination (Milton & Hall 1981). However, most efforts to restore controlled areas fail because the re-introduced native plant species do not establish (Gaertner *et al.* 2012). This might be due to secondary invasion – that is an increase in the abundance of non-target alien plant species that establish after clearing the target invader (Pearson *et al.* 2016), and/or weedy native species dominance – that is an increase in the abundance of native species that are not typically distributed in the area and are often found dominating disturbed environments (M. M. Nsikani, unpublished data, 2016). Furthermore, the failure of such restoration efforts has been associated with strong inhibition caused by soil legacy effects (Macdonald 2004; Gaertner *et al.* 2012). However, limited attention has been given to understanding the soil legacy effects of *A. saligna* (or invasive Australian acacias in general) after clearing (see Yelenik *et al.* 2004; Marchante *et al.* 2009). This paper explores the issue of persistent soil legacy effects, using invasive *A. saligna* in South Africa as case study. Overall, we set out to (i) determine how soil characteristics differ between cleared, invaded and non-invaded sites with emphasis on time after clearing, and (ii) understand the implications that the differences in soils can have for ecological restoration.

MATERIALS AND METHODS

Study sites

The study was conducted in the Cape Flats Sand Fynbos (CFSF) of the Western Cape Province, South Africa – a main hotspot for *A. saligna* invasion in the South African fynbos. The climate in the CFSF is Mediterranean with hot dry summers and cool wet winters, and the soils are broadly classified as well-drained aeolian acidic sands characterized by low nitrogen (Rebelo *et al.* 2006). Native CFSF vegetation consists of evergreen shrub-lands dominated by a mixture of ericoid and proteoid shrubs and restioid (aphyllous graminoid) growth forms (Rebelo *et al.* 2006). Cape Flats Sand Fynbos is a critically endangered vegetation type with less than 1% of the total historical area statutorily conserved and it is the most transformed of the sand fynbos types occurring in the Greater Cape Floristic Region (Rebelo *et al.* 2006).

We selected three study sites in the CFSF, namely, Blaauwberg Nature Reserve (33°46′5.16″S; 18°27′10.08″E), Youngsfield (34°0′30.30″S; 18°29′16.20″E) and Penhill (33°59′0.39″S; 18°43′37.74″E). Youngsfield and Penhill had similar soil types, whereas Blaauwberg Nature Reserve differed slightly (Table 1). The three study sites received different amounts of annual precipitation (i.e. between 361 and 1018 mm; Table 1) which can affect some soil characteristics such as electrical conductivity (Rhoades *et al.* 1976; Sudduth *et al.* 2005).

In each study site, we delimited three areas: non-invaded, densely invaded and cleared of invasive *A. saligna* and left to recover naturally. Invaded areas were densely invaded (75–100% cover) and had been invaded for a 'long time' (>20 years; see Marchante *et al.* (2008) for definition/classification). Each site had experienced at least one fire in the last 10 years, with the time since the last fire and soil sampling for this study ranging from 4 to 7 years (Table 1). The time between clearing and sampling was between 2 and 10 years, and the cleared areas received between one and seven follow-up treatments, in which re-emerging invader seedlings were removed through weeding or herbicides (Table 1). Only one cleared area (Penhill) was burnt immediately after clearing. Cleared areas were dominated by secondary invaders such as *Briza maxima* L., and weedy native species such as *Ehrharta calycina* Sm. (M. M. Nsikani, unpublished data, 2016). Non-invaded areas were characterized by mature native fynbos plant communities, were free from *A. saligna* invasion and we believe that they represent the environmental characteristics of densely invaded sites prior to invasion. Non-invaded areas were kept free from *A. saligna* invasion through management and not as a result of different environmental characteristics. The study sites varied in size from 1.5 to 9.5 km^2.

Soil collection

Soil samples were collected in the cleared, invaded and non-invaded area of each study site. In each cleared,

Table 1. History of study sites including the mean annual precipitation; years after initial clearing; years since last fire; whether the cleared site was burnt after initial *Acacia saligna* clearing; number of follow-up *A. saligna* clearing treatments and soil type

Study site	Mean annual precipitation (mm)	Years after initial clearing	Years since fire	Post clearing burn?	Number of follow-up treatments	Soil type (*sensu* http://bgisviewer.sanbi.org)
Blaauwberg Nature Reserve	361	2	4	No	1	Greyish sandy soil
Youngsfield	1018	6	6	No	4	Soils with a sandy texture, leached and with sub-surface accumulation of organic matter and aluminium with/without oxides, either deep or on hard or weathering rock
Penhill	556	10	7	Yes	7	Soils with a sandy texture, leached and with sub-surface accumulation of organic matter and aluminium with/without oxides, either deep or on hard or weathering rock

invaded or non-invaded area, we chose five random sampling points (0.5 × 5 m; $n = 45$). In June 2015, five soil sub-samples (approximately 600 g each) were taken from the top 10 cm at each sampling point after removing the organic horizon material. All five sub-samples per sampling point were bulked and homogenized to make a representative sample.

Soil analysis

Approximately 500 g of soil per sample were taken to Bemlab Pty Ltd (Somerset West, Western Cape) and analysed for pH, % carbon (C), % nitrogen (N), available phosphorus (Bray II; AP), ammonium (NH_4^+), nitrate (NO_3^-) and electrical conductivity (EC). PH (1.0 mol L^{-1} KCl), EC and AP were analysed according to the methods given by the Non-affiliated Soil Analysis Work Committee (1990). C and N were determined through total combustion using a Leco Truspec CHN analyser. NH_4^+ and NO_3^- were extracted from soil with 1 N KCl and determined colorimetrically on a SEAL AutoAnalyzer 3 (*sensu* AgriLASA 2004).

Enzyme analysis

Soil microbial communities release extracellular enzymes that allow them to access energy and nutrients present in complex substrates and initiate the processes of decomposition and nutrient mineralization (Allison & Vitousek 2005). Enzyme activities can be used to understand the key processes linking soil microbial populations and nutrient dynamics (Sinsabaugh & Moorhead 1994), and understanding the effects of plant invasions (e.g. Vilà *et al.* 2011; Novoa *et al.* 2013; Souza-Alonso *et al.* 2015). In this study, we analysed the activities of the enzymes β-1,4-glucosidase (E.C. 3.2.1.21; glucosidase); urease (E.C. 3.5.1.5); and

acid phosphatase (E.C. 3.1.3.2; phosphatase) responsible for the cycles of C, N and P respectively.

Approximately 100 g of soil per sample were taken to the molecular ecology lab (Department of Botany and Zoology, Stellenbosch University) and analysed for enzyme activities. The substrates used for the glucosidase, urease and phosphatase assays were *p*-nitrophenol (*p*NP)-β-D glucopyranoside, urea and *p*NP-phosphate respectively. The Modified Universal Buffer (20.14 mmol L^{-1} Tris-hydroxymethyl aminomethane; 17.3 mmol L^{-1} Malic acid; 14.6 mmol L^{-1} Citric acid; 20.3 mmol L^{-1} Boric acid and 1 mol L^{-1} NaOH) was used to make the substrates. There were six analytical replicates and six sample controls of each treatment. For the analysis of glucosidase, urease and phosphatase, the methods followed were those described by Novoa *et al.* (2014).

Statistical analyses

The data were analysed using R version 3.3.0 (R Development Core Team 2016). All soil characteristics and enzyme activities were compared separately – that is only within each study site to avoid unexplainable variation in soils resulting from differences in soil type and micro-climates between study areas. For testing the effect of invasion status (invaded, cleared and non-invaded) on overall soil nutrient composition in each study site, we conducted a principal components analysis (PCA) on the correlation matrix of the soil nutrient variables using the *base* package. After inspection of PCA plots, we conducted a Permutation Multivariate Analysis of Variance (PERMANOVA; Anderson 2001) with 9999 permutations using Euclidean distance (to correspond to the PCA distance) with the function *adonis* from the *vegan* package (Oksanen *et al.* 2014) as an additional test. We first standardized all the variables to mean zero and unit variance and then included invasion status as predictor variable. For testing the effect of invasion status on the

individual soil nutrient levels and enzyme activities in each study site, we conducted a one-way Analysis of Variance (ANOVA) and log transformed the data where necessary to accomplish the normality assumption of ANOVA. Significant mean differences were separated with Tukey's HSD test using *multcomp* package (Hothorn *et al.* 2008).

RESULTS

Overall soil characteristics between invaded, cleared and non-invaded areas

The PCAs conducted per study site were based on five replicates per invasion status and they separated the three invasion statuses (Fig. 1). For Blaauwberg Nature Reserve, the first two principal components explained 67% of variation (PC1 = 45%; PC2 = 22%) in the dataset. PC1 separated the cleared from the invaded and non-invaded areas, whereas PC2 separated the non-invaded from the invaded and cleared areas. Variables that contributed the most to PC1 were NO_3^-, AP and EC, and for PC2 were pH and NH_4^+. For Youngsfield, the first two principal components explained 63% of the variation (PC1 = 36%; PC2 = 27%) in the dataset. The invaded, cleared and non-invaded areas were different to each other in terms of PC1 and PC2. Variables that contributed the most to PC1 were N, NO_3^- and AP, and for PC2 were C and pH. For Penhill, the first two principal components explained 62% of the variation (PC1 = 39%; PC2 = 23%) in the dataset. PC1 separated the three areas from each other. Variables that contributed the most to PC1 were NO_3^-, pH and AP, and for PC2 were C and NH_4^+. PERMANOVA results showed that the overall soil characteristics between all cleared sites and their reference invaded and non-invaded sites were significantly different (Table 2).
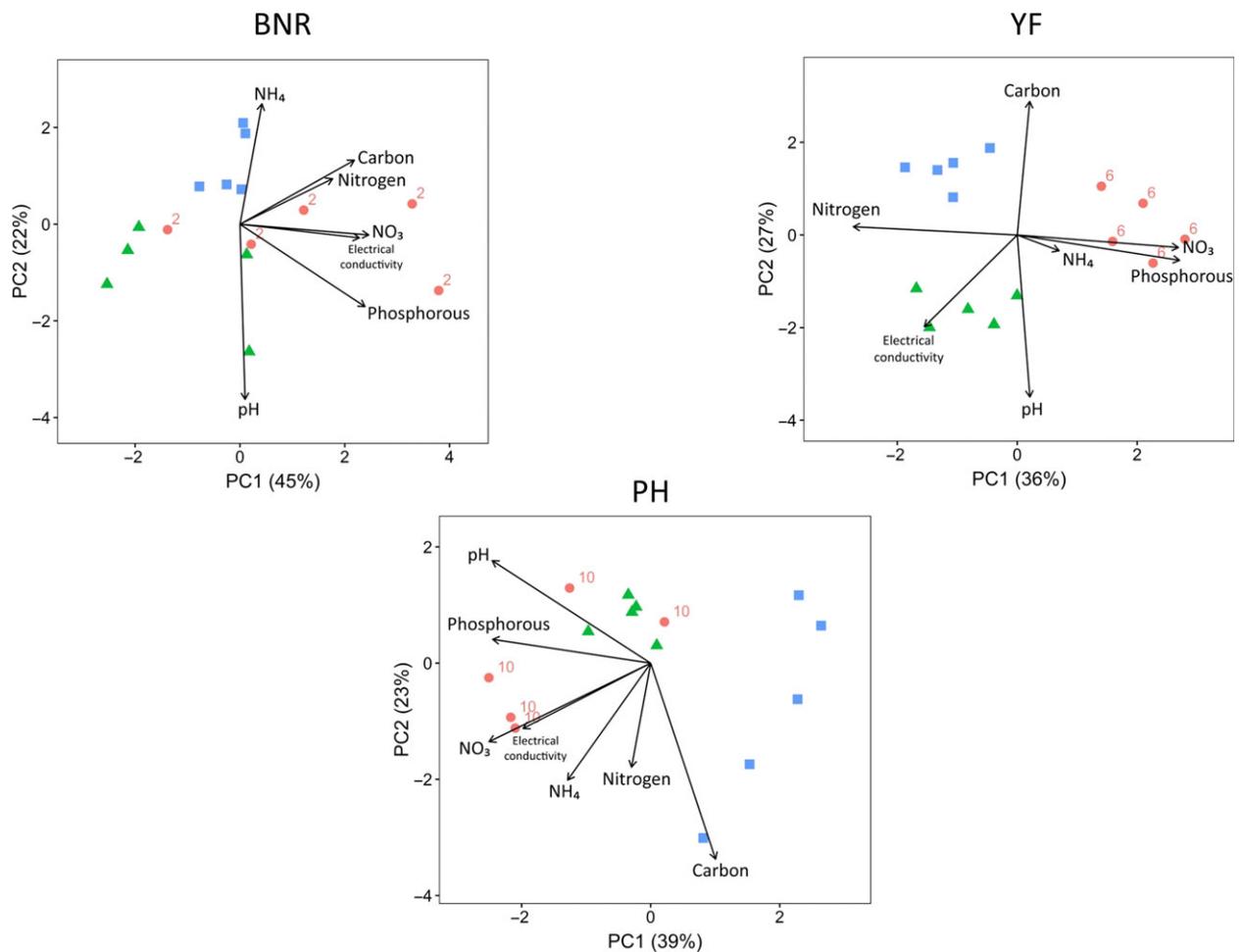

Fig. 1. Principal Component Analyses of the soil characteristics in each study area. Dots = cleared sites; triangles = invaded sites; and squares = non-invaded sites. BNR, Blaauwberg Nature Reserve; PH, Penhill; YF, Youngsfield. [Colour figure can be viewed at wileyonlinelibrary.com]

Table 2. PERMANOVA results for each study area based on soil characteristics and invasion status of sites within the areas

Site name	Factor	d.f.	Sums of squares	Mean squares	F model	R^2	P
Blaauwberg Nature Reserve	Invasion status	2	34.676	17.338	3.2856	0.35384	0.0019**
	Residuals	12	63.324	5.277		0.64616	
	Total	14	98	1			
Youngsfield	Invasion status	2	54.441	27.22	7.4988	0.55552	<0.001***
	Residuals	12	43.559	3.63		0.44448	
	Total	14	98	1			
Penhill	Invasion status	2	42.854	21.427	4.6626	0.43728	<0.001***
	Residuals	12	55.146	4.5955		0.56272	
	Total	14	98	1			

Significance indicated in bold as: **$P < 0.01$; ***$P < 0.001$.

Individual soil nutrient levels and enzyme activities, pH and EC between cleared, invaded and non-invaded sites

We believe that prior to invasion, each densely invaded area had similar soil characteristics to its reference non-invaded area. Of the soil chemistry characteristics investigated, invasion significantly elevated pH levels by 0.6–1.8, and lowered C content by 75% at Youngsfield and Penhill (Fig. 2). The remaining characteristics were unaffected. Clearing of invaded sites elevated NO_3^- levels and these remained higher than the invaded (1.55–6.67 mg kg^{-1}) and non-invaded (2.16–4.35 mg kg^{-1}) areas up to 10 years after clearing (Fig. 2). In addition, the cleared area (6 years after clearing) at Youngsfield, had lower N and EC levels (88% and 49% respectively) but higher (252%) AP. The cleared area (10 years after clearing) at Penhill also had higher (136%) AP. After clearing, NO_3^- (2.16–4.35 mg kg^{-1}) and pH (0.88–1.76) levels remained higher up to 10 years after clearing (Fig. 2). Furthermore, the cleared areas at Youngsfield and Penhill had higher levels of AP, NO_3^- and pH, whereas the cleared area at Youngsfield had
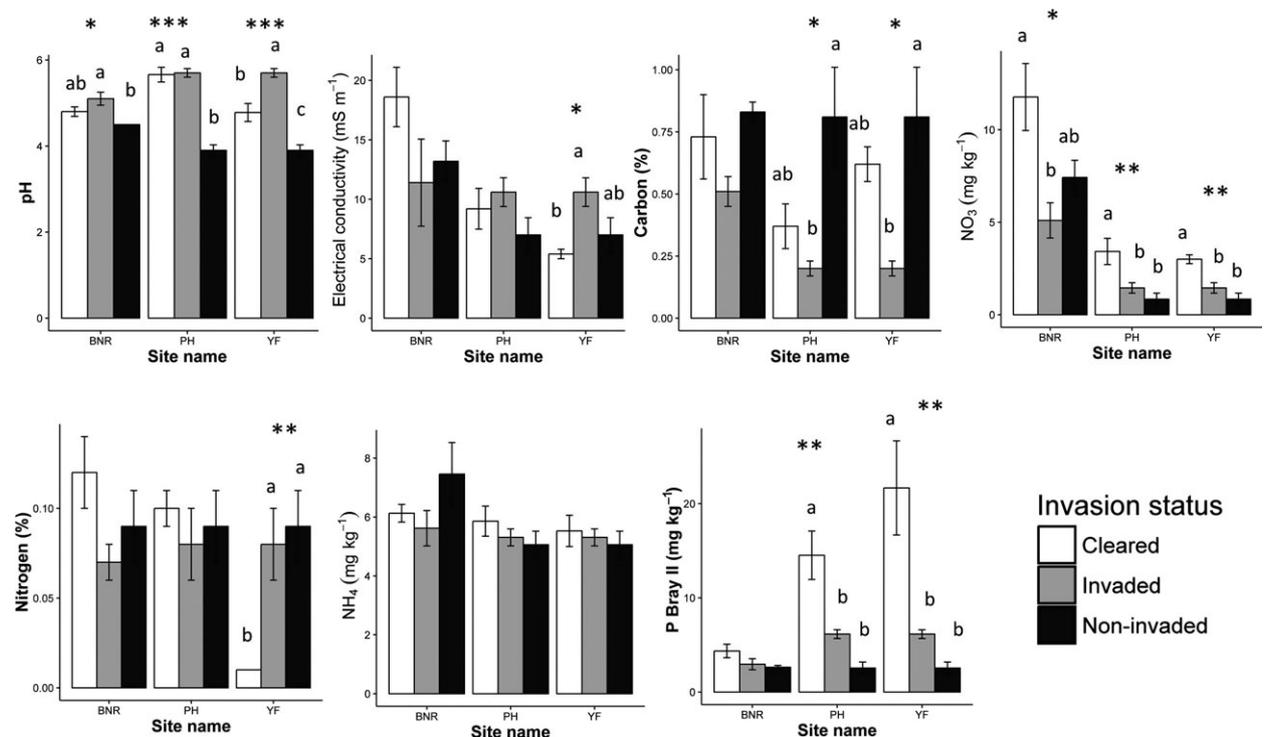

Fig. 2. One-way ANOVA results of the different soil nutrients, pH and electrical conductivity in the different *Acacia saligna* invasion statuses in each study site. Mean values of each soil nutrient in each study area with the same letter are not significantly different. Significance indicated in bold as: *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$. BNR, Blaauwberg Nature Reserve; PH, Penhill; YF, Youngsfield.
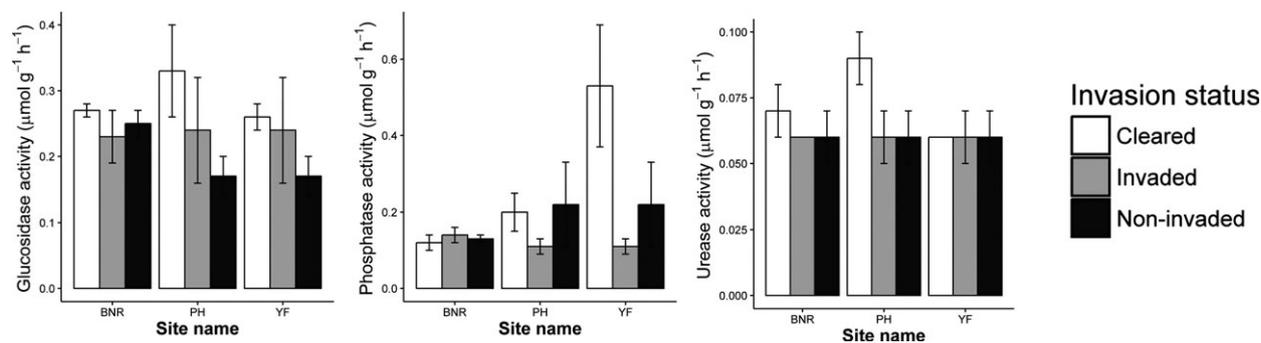
Fig. 3. One-way ANOVA results of the different enzyme activities in the different *Acacia saligna* invasion statuses in each study area. BNR, Blaauwberg Nature Reserve; PH, Penhill; YF, Youngsfield.

lower (89%) N content. There were no significant differences in the activity of glucosidase, phosphatase and urease among all cleared sites and their reference invaded and non-invaded sites (Fig. 3).

DISCUSSION

Impacts of *Acacia saligna* on soil characteristics

Our results showed significant differences in pH and carbon levels between non-invaded and invaded areas. These observations support previous studies which showed that a significant proportion of invasive alien species, particularly N_2-fixing species, alter soil characteristics and processes in the areas they invade (Vitousek 1990; Stock *et al.* 1995; Gordon 1998; Maron & Jeffries 2001; Marchante *et al.* 2008; Von Holle *et al.* 2013).

First, our results suggest that invasion by *A. saligna* increases soil pH levels (makes them more basic). Native South African fynbos is characterized by acidic soils (Rebelo *et al.* 2006). The decomposition of the large amounts of litter that *A. saligna* adds to the invaded soils might cause the observed increase on such low pH levels (Ehrenfeld 2003, 2004; Raizada *et al.* 2008). Various mechanisms might explain why litter decomposition can increase the soil pH – for example release of NH_3 from the decomposing litter or depletion of protons in the soil through oxidation of organic anions (Noble *et al.* 1996). Increases in pH have been generally observed with both N_2-fixing (e.g. *A. saligna* in the South African fynbos; Gaertner *et al.* 2011; Mostert *et al.* 2016) and non-N_2-fixing alien plant invaders (e.g. *Berberis thunbergii* and *Microstegium vimineum* in New Jersey (USA); *Parthenium hysterophorus* in grassland communities of central Nepal; Timsina *et al.* 2011). Second, C content was lower in the invaded areas at Youngsfield and Penhill in comparison to non-invaded areas.

This is probably a result of previous fire events in the study sites (Table 1) that most likely burnt off the soil organic material that was otherwise high in C content. Soil C content was also reported lower in in Portuguese coastal dunes invaded by *Acacia longifolia* that experienced fire compared to non-invaded areas that had not been burnt (Marchante *et al.* 2008). Generally, soil C content may decrease after one or more fire events (Carreira *et al.* 1994; Paul *et al.* 2002) and the intensity of the fire can affect the amount of C lost (Paul *et al.* 2002).

However, we found no differences in N or AP between invaded and non-invaded areas. Previous studies have shown elevated N levels associated with the invasion of N_2-fixing plants (Vitousek & Walker 1989; Stock *et al.* 1995; Pickart *et al.* 1998; Malcolm *et al.* 2008; Marchante *et al.* 2008; González-Muñoz *et al.* 2012; Grove *et al.* 2015). Surprisingly, our results show no significant differences in N between invaded and non-invaded sites. The reason behind this might be that the reference non-invaded areas of our study showed higher N levels than expected (see Musil 1993 and Yelenik *et al.* 2004). We also observed non-significant differences in nitrogen and phosphorus availability (i.e. NH_4^+; NO_3^-; AP) between invaded and non-invaded areas. These observations support previous findings on *A. saligna* and *Acacia cyclops* invasions (Stock *et al.* 1995; Gaertner *et al.* 2011). The magnitude of the effect of *Acacia* invasions on nitrogen or phosphorus availability may differ depending on the leaf chemistry of the *Acacia* spp. and/or the soil properties of the study site (Stock *et al.* 1995).

Legacy effects of *Acacia saligna* on soil characteristics

Numerous studies have observed persistence of altered soil characteristics for long periods after clearing invasive alien N_2-fixing species – for example

more than 4 years in the pine–oak systems (USA) cleared of *Robinia pseudoacacia* (Malcolm *et al.* 2008) and Portuguese coastal dunes cleared of *A. longifolia* (Marchante *et al.* 2009). Our results show that pH levels can either be recovered after *A. saligna* is controlled – that is in the cleared sites at Blaauwberg Nature Reserve and Youngsfield, pH levels were intermediate to the invaded and non-invaded sites – but they can also remain similar to invaded sites – that is in the cleared site at Penhill they were similar to the invaded site. These observations suggest that the impact of *A. saligna* on pH can be profound and persist up to 10 years after clearing. Altered pH levels often exert negative impacts on fungal and bacterial compositions (Bååth & Anderson 2003), enzyme activities (Dick *et al.* 1988) and availability of some nutrients such as manganese and zinc (Sims 1986).

Moreover, we found a low N 6 years after clearing compared to the reference invaded and non-invaded areas. The differences are most likely a result of high N losses in the cleared area (Stock & Lewis 1986; Witkowski 1991; Malcolm *et al.* 2008; Jovanovic *et al.* 2009; Staska *et al.* 2014). It is worth noting that low N in the cleared area compared to the invaded and non-invaded areas were not standard across all study sites. All our study sites were located on coarse-textured soils which are known to have a lower water holding capacity and a greater potential to lose N from organic matter and NO_3^- through leaching (Stock & Lewis 1986; Witkowski 1991). Therefore, we suspect that the lower water holding capacity in combination with high precipitation and uptake by secondary invaders and weedy native species (personal observation) in the cleared area resulted in higher N losses compared to the reference invaded and non-invaded areas.

Our results suggest that clearing invasive *A. saligna* increases NO_3^- levels, and these changes persist up to 10 years after clearing. Such changes might be a result of a stimulation of the nitrification process with clearing – that is the initial effect of clearing the invader allows the sun to reach the ground previously covered by the invader canopy, which increases temperatures in the soil stimulating the nitrification process (Yelenik *et al.* 2004). We suspect that with time (2, 6 and 10 years after clearing) some of the NO_3^- may be lost through leaching and taken up by secondary invaders and/or weedy native species (Stock & Lewis 1986; Jovanovic *et al.* 2009; Gaertner *et al.* 2011; Staska *et al.* 2014). However, we believe that NO_3^- levels are often high after initial clearing such that the losses may not be high enough to have an overall effect. We also suspect that organic matter from *A. saligna* cleared and left on site after follow-up treatments, secondary invaders and/or weedy native species in

these sites (personal observation) may contribute to maintaining elevated NO_3^- levels in the cleared sites.

Phosphorus availability was higher 6 and 10 years after clearing in comparison to the reference invaded and non-invaded areas. This might be a result of high amounts of organic matter from *A. saligna* cleared and left on site after follow-up treatments, secondary invaders and/or weedy native species in these sites (personal observation). Organic matter can elevate available phosphorus in two ways (i) decomposition of organic matter can release phosphorus products that can be available in the soil solution (Guppy *et al.* 2005), and/or (ii) competition between the decomposition products of organic matter and P for soil sorption sites can result in phosphorus being more available in the soil solution (Guppy *et al.* 2005).

Finally, the C content in the sites 6 and 10 years after clearing was intermediate to the reference invaded and non-invaded areas suggesting that the C content of the soil was on the path to recovery from the effect of fire events experienced in the sites, most likely with the aid of accumulating secondary invader and/or weedy native species litter (Dumontet *et al.* 1996).

Soil enzyme activities

Extensive evidence from previous studies has shown that soil microbiota is highly responsive to the type of plant species occupying the soil; hence changes in species composition and pH that come with invasion are expected to alter the structure and/or functional properties of soil microbiota (Ehrenfeld 2004; Fierer & Jackson 2006; Lauber *et al.* 2008; Vilà *et al.* 2011). Numerous studies of *A. saligna* invasion have suggested that the species produces large amounts of litter with high N content resulting in more N returning from the above-ground biomass to the soil (Witkowski 1991; Stock *et al.* 1995; Yelenik *et al.* 2004).

In light of current knowledge, we expected differences in the activity of enzymes related to N and C cycling (urease and glucosidase), and P cycling (phosphatase) between invaded, cleared and non-invaded areas; however, we did not observe this. Instead our results conflict with numerous studies that have shown a significant increase in enzyme activity under the invader canopy compared to native stands (Kourtev *et al.* 2002; Allison *et al.* 2006; Caldwell 2006; Marchante *et al.* 2008; Vilà *et al.* 2011). We suspect that enzyme activities in our study were limited by moisture availability (Yelenik *et al.* 2004) which has been shown to limit the decomposition process in the South African fynbos around the time

we sampled for soil enzyme activity (summer; Witkowski 1991). Low moisture content could have resulted from the hot and dry conditions in our study areas (Rebelo *et al.* 2006) and compounded by rapid surface drying caused by the significant percentage of coarse sand in the soils (Witkowski 1991; M. M. Nsikani, unpublished data, 2016). Low soil moisture can strongly limit enzyme activities in the soil (Steinweg *et al.* 2012). To accurately sample for soil enzyme activities in the South African fynbos, it may be advisable for future studies to sample during the hot and dry summer, and the cold and wet winter.

CONCLUSIONS AND IMPLICATIONS FOR ECOLOGICAL RESTORATION

Overall, our findings suggest that invasion of native areas by the alien *A. saligna* (i) changes overall soil characteristics; and (ii) leaves soil legacies that can persist up to 10 years after clearing. Therefore, clearing the invader does not reverse these effects; instead it elevates soil NO_3^- levels. Elevated NO_3^- levels after clearing *A. saligna* often facilitate secondary invaders (Yelenik *et al.* 2004; Pearson *et al.* 2016) and weedy native species that often out-compete native species (Yelenik *et al.* 2004).

To our knowledge, there is no known management strategy that can be successfully used to facilitate the recovery of overall soil characteristics and elevated NO_3^- levels after clearing invasive *A. saligna*. However, we believe that active restoration by planting native species could, over time, return soils to previous conditions. Many species adapted to low N availability (e.g. typical native fynbos species) can (i) immobilize N and slow N cycling using various mechanisms, such as producing relatively recalcitrant and high C:N litter; and (ii) restore ecosystem function, when they successfully re-establish (Perry *et al.* 2010). However, re-establishment of such planted native species after clearing *A. saligna* could be elusive due to competition with secondary invaders and/or weedy native species (Yelenik *et al.* 2004; M. M. Nsikani, unpublished data, 2016). Strategies to reduce soil N availability and their efficacy have been reviewed by Perry *et al.* (2010). We urge practising restoration ecologists to consider them when managing sites previously invaded by *A. saligna* to facilitate the recovery of elevated NO_3^- levels. Moreover, we believe that litter from the target invader, secondary invaders and weedy native species should be removed to aid the recovery of pH and available phosphorus after clearing. Manual removal of invader litter layer using a rake or similar instrument (small invader populations; Pickart *et al.* 1998) or fire (extensive invasions; Richardson & Kluge 2008) can be used to remove the invader litter layer.

We believe that restoration efforts after clearing invasive *A. saligna* should not focus on restoring native soil characteristics because (i) there are very few management strategies that can be implemented and success is not guaranteed, and (ii) altered soil characteristics per se often do not have negative effects on the re-establishment of native species (M. M. Nsikani, unpublished data, 2016). Instead restoration efforts need to focus on conducting multiple successive follow-up treatments to avoid re-invasion by *A. saligna* (Van Wilgen *et al.* 2011), and the removal of secondary invaders and weedy native species (Pearson *et al.* 2016).

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