Microbial functional diversity of a shrubland soil experimentally burned and treated with two post-fire stabilization techniques (straw mulch and seeding)

M.T. Fontúrbel (1), J.A. Vega (1), E. Jiménez (1), C. Fernández (1), A. Barreiro (2), A. Lombao (2), A. Martín (2), T. Carballas (2), M. Díaz-Raviña (2*)

- (1) Centro de Investigación Forestal de Lourizán, Consellería de Medio Rural, Apdo 127, 3680, Pontevedra (Spain)
- (2) Instituto de Investigaciones Agrobiológicas de Galicia (IIAG-CSIC), Apartado 122, 15780, Santiago de Compostela (Spain)
- * Corresponding author: mdiazr@iiag.csic.es

Keywords

Biolog Ecoplates Experimental fire Metabolic profiling Mulching Seeding

Abstract

The study examined the effect of two post-fire stabilization treatments (seeding and mulching) on microbial diversity of a shrubland area of Galicia after an experimental fire. The soil was a Leptosol developed over granite with a slope of 38-54% and the soil microbial functional diversity was assessed using Biolog substrate utilization EcoPlates (Biolog Inc., Hayward, CA, USA). Soil samples were taken from the A horizon (0-5 cm depth) at different sampling times over one year after the experimental fire. The results indicated that immediately after the fire there were significant differences in the categorized substrate utilization pattern between the microbial communities of the burnt soil treatments and the corresponding unburnt control. The burned soils exhibited significant higher values for the utilization of carboxylic acids, amino acids, carbohydrates and phenolic compounds, suggesting that the microbial community in the burned soils could be favoured by the increase in available C and nutrients following the experimental fire. These changes in the categorized substrate utilization pattern were attenuated with time; thus, one year after the fire, similar values for utilization of different C sources were observed for all unburned and burnt soils. With respect to post-fire treatments only the mulching showed an effect on the C utilization pattern.

1 Introduction

The NW of Spain and the North of Portugal are the European zones most affected by forest wildfires. To minimize the impact of fire on the affected ecosystems, several techniques of soil stabilization and rehabilitation such as the application of different types of mulch, seeding and erosion barriers can be implemented. Recently, the effectiveness of these techniques to reduce soil erosion has been showed (Fernández et al., 2011). However,

information about their effects on soil properties, especially on microbial properties is scarce.

2 OBJECTIVE

The aim of this study is to assess the effect of two post-fire stabilization treatments (seeding and mulching) on microbial diversity (community level physiological profiles by Biolog Ecoplates) of a shrubland area of Galicia after an experimental fire.

Table 1. . Categorized substrates utilization pattern by microbial communities from the different soil treatments at different times (1, 90, 180 and 365 days) after the fire and application of the stabilisation treatments. Substrates: CA, carboxylic acids; AMN, amines/amides; AAC, amino acids; CH, carbohydrates; PHE, phenolic compounds; POL, polymers. Treatments: U, unburned soil; B, burned soil; B+S, burned soil plus seeding; B+M, burnt soil plus straw addition. For the same time and substrate, different letters denote significant differences (p<0.05).

| Time (days) | Substrate | Soil treatment | | | |
|-------------|-----------|----------------|--------------------|-------------------|-------------------|
| | | U | В | B+S | B+M |
| 1 | CA | 0.89 ± 0.07 a | 1.39 ± 0.03 b | 1.31 ± 0.07 b | 1.05 ± 0.16 ab |
| | AMN | 0.56 ± 0.28 a | 0.83 ± 0.17 a | 0.66 ± 0.15 a | 0.61 ± 0.23 a |
| | AAC | 1.21 ± 0.03 a | 1.55 ± 0.13 b | 1.64 ± 0.07 b | $1.43 \pm 0.14 b$ |
| | CH | 0.89 ± 0.09 a | 1.51 ± 0.15 b | 1.28 ± 0.16 b | 1.37 ± 0.06 b |
| | PHE | 0.56 ± 0.09 a | 1.12 ± 0.15 b | 1.07 ± 0.11 b | 0.95 ± 0.25 ab |
| | POL | 0.89 ± 0.13 a | 1.02 ± 0.14 a | 1.06 ± 0.11 a | 1.16 ± 0.19 a |
| 90 | CA | 0.94 ± 0.09 a | 1.20 ± 0.11 ab | $1.40 \pm 0.11 b$ | 1.55 ± 0.16 b |
| | AMN | 0.51 ± 0.11 a | 1.10 ± 0.19 b | 1.03 ± 0.12 b | 1.08 ± 0.21 b |
| | AAC | 1.14 ± 0.05 a | 1.28 ± 0.05 a | 1.30 ± 0.07 a | 1.36 ± 0.12 a |
| | CH | 1.12 ± 0.04 a | 1.43 ± 0.08 a | 1.28 ± 0.16 a | 1.60 ± 0.17 a |
| | PHE | 0.72 ± 0.11 a | 0.93 ± 0.09 a | 0.94 ± 0.13 a | 1.11 ± 0.10 a |
| | POL | 1.31 ± 0.14 a | 1.37 ± .021 a | 1.38 ± 0.12 a | 1.48 ± 0.13 a |
| 180 | CA | 0.94 ± 0.16 a | 1.15 ± 0.10 a | 1.28 ± 0.19 a | 1.19 ± 0.08 a |
| | AMN | 0.35 ± 0.04 a | 0.78 ± 0.16 a | $0.62 \pm 0.10 b$ | 1.24 ± 0.11 c |
| | AAC | 1.27 ± 0.09 a | 1.43 ± 0.12 a | 1.24 ± 0.09 a | 1.44 ± 0.02 a |
| | CH | 1.14 ± 0.06 a | 1.08 ± 0.09 a | 1.43 ± 0.21 a | 1.13 ± 0.12 a |
| | PHE | 0.86 ± 0.05 a | 0.81 ± 0.11 a | 0.80 ± 0.06 a | 0.75 ± 0.07 a |
| | POL | 1.00 ± 0.12 a | 1.03 ± 0.09 a | 1.11 ± 0.06 a | 1.37 ± 0.17 a |
| 365 | CA | 0.94 ± 0.09 a | 1.20 ± 0.05 a | 1.22 ± 0.05 a | 1.04 ± 0.14 a |
| | AMN | 0.70 ± 0.10 a | 0.86 ± 0.25 a | 1.01 ± 0.17 a | 0.58 ± 0.16 a |
| | AAC | 1.17 ± 0.07 a | 1.34 ± 0.10 a | 1.27 ± 0.06 a | 1.22 ± 0.09 a |
| | CH | 1.06 ± 0.21 a | 1.17 ± 0.18 a | 1.16 ± 0.16 a | 1.28 ± 0.12 a |
| | PHE | 0.75 ± 0.04 a | 0.91 ± 0.06 a | 0.94 ± 0.17 a | 0.81 ± 0.02 a |
| | POL | 0.87 ± 0.01 a | 1.22 ± 0.26 a | 1.07 ± 0.13 a | 1.19 ± 0.16 a |

3 Materials and methods

The study was conducted in an experimental field located at an altitude of 660 a.s.l. in Cabalar (A Estrada, 42º 38' 58" N; 8º 29' 31" W; N.W. Spain). The soil, a Leptosol developed over granite and with a slope of 38-54%, has a vegetation cover dominated by Ulex europaeus L. and some Pteridium aquilinum (L.) Kuhn., Ulex gallii Planch., Daboecia cantabrica (Huds.) K. Koch and Pseudoarrenhaterum longifolium Rouy. The main characteristics of the A horizon (fraction < 2 mm) of this soil were acid pH (3.7) and high organic matter content $(179~g~C~kg^{-1}, 14.8~g~N~kg^{-1})$. After the experimental fire four treatments were considered by quadruplicate (30 x 10 m plots): unburnt soil (C) as control; b) burnt soil (B); c) burnt soil with 232 g m⁻² of straw mulch (B+M); d) burnt soil with a mixture of seeds at a rate of 45 g m⁻² (Lolium multiflorum, 35%; Trifolium repens, 25%; Dactylis glomerata, 20%; Festuca arundinacea, 10%; Festuca rubra, 5%, Agrostis tenuis, 5%) (B+S). Soil samples were taken

from the A horizon (0-5 cm depth) at different sampling times over one year after the experimental fire.

The soil microbial functional diversity was assessed using Biolog substrate utilization EcoPlates (Biolog Inc., Hayward, CA, USA; Garland & Mills, 1991). Each EcoPlate consists of a 96-well microtiter plate filled with three replicates of 31 different carbon substrates (10 carbohydrates; 7 carboxylic acids; 6 amino acids; 4 polymers; 2 amines/amides and 2 phenolic compounds) and one control with distilled water. Each well is also filled with an indicator substance, the tetrazolium dye that changes the colour to purple with substrate consumption. Differences in the well colour development were recorded as optical density (OD) at 590 nm at 24-h intervals for seven days of incubation and data from day 4 were used for analysis. The analysis were made at different times (1, 90, 180 and 365 days) after the fire and application of the stabilization treatments, and compared with the same measurements made in the respective burned and unburned control soils.

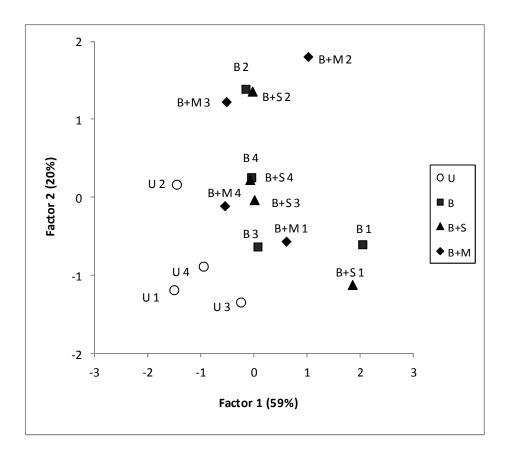


Figure 1. PCA results of the categorized substrates utilization pattern by the microbial communities from the different soil treatments at different times (1, 2, 3 and 4 denote samples collected 1, 90, 180 and 365 days after the fire and application of the stabilisation treatments, respectively). Treatments: U, unburned soil; B, burned soil; B+S, burned soil plus seeding; B+M, burned soil plus application of straw.

In order to evaluate the effect of the different treatments on the microbial use of the six groups of substrates, for each sampling time analysis of variance was applied and in the cases of significant F statistics, the Tukey's minimum significant difference test was used to compare the mean values. The homogeneity of variances and normality of the data were previously checked by the Levene and Shapiro-Wilk tests, respectively. A principal component analysis (PCA) was also carried out on the microbial use of the six groups of substrates using the mean values for each treatment at each sampling time (6 variables, 16 soil samples). The SPSS (2004) statistical package was used to carry out the respective analyses.

4 RESULTS AND CONCLUSIONS

The average utilization of the C sources obtained for all soil treatments at different sampling times are shown in Table 1. The results indicated that immediately after the fire there were significant differences in the categorized substrate utilization pattern between the microbial communities of the burnt soil treatments (B, B+S, B+M) and the corresponding unburnt control. The burned soils exhibited significant higher values for the utilization of carboxylic acids, amino acids, carbohydrates and phenolic compounds, suggesting that the microbial community in the burned soils could be favoured by the increase in available C and nutrients following the experimental fire. These changes in the categorized substrate utilization pattern were attenuated with time. At day 90, the burned soils showed a higher use of carboxylic acids and amines/amides than that of the unburned soils and this increased use of the later substrate was still evident at day 180, particularly in the B+M treatment. However, one year after the fire, similar values for utilization of different C sources were observed for all unburned and burnt soils. Therefore, these results showed a slight positive influence

of prescribed fire at short- and medium term (up to 180 day) but this effect did not last one year. This behaviour can be attributed to the low severity of the experimental fire studied (Fontúrbel et al., 2012), which is coincident with results of other authors showing slight and transitory changes in functional diversity in soil ecosystems following fires (Staddon et al., 1997; D'Ascoli et al., 2005). The principal component analysis (PCA) for the microbial use of substrates revealed that factor 1 and factor 2 explained 79% of the total variance (Figure 1).

The factor 1 (59% of variance) tended to separate the burned treatments (positive values along factor 1) from the unburned treatments (negative values along factor 1). The burned soils were associated with higher levels of use of most categorized substrates than those in the corresponding unburned soils. The factor 2 (20% of variance) tended to separate one sample collected at day 180 (B+M3) and the samples collected 90 days after the fire, which were associated with high level in the use of polymers and amines, from the rest of the samples. For all sampling times, B and B+S samples were grouped together and separated from the B+M treatment, suggesting a greater effect of the mulching treatment on the C utilization pattern than that observed for the seeding treatment.

ACKNOWLEDGEMENTS

This study was supported by the Consellería de Educación y Ordenación Universitaria de la Xunta de Galicia (08MRU002400PR) and by the Ministerio de Ciencia e Innovación (AGL2008-02823), Spain. A. Barreiro and A. Lombao are recipients of FPU grants from Spanish Ministry of Education.

REFERENCES

- D'Ascoli R, Rutigliano FA, De Pascale RA, Gentile A, Virzo De Santo A. 2005. Functional diversity of the microbial community in Mediterranean maquis soils as affected by fires. International Journal of Wildland Fire 14: 355–363.
- Fernández C, Vega JA, Jiménez E, Fonturbel T. 2011. Effectiveness of three post-fire treatments at reducing soil erosion in Galicia (NW Spain). International Journal of Wildland Fire 20: 104-114.
- Fontúrbel MT, Barreiro A, Vega JA, Lombao A, Martín A, Jiménez E, Carballas T,Fernández C. 2012. Effects of an experimental fire and post-fire stabilisation

- treatments on soil microbial communities. Geoderma 191: 51-60.
- Garland JL, Mills AL. 1991. Classification and characterization of heterotrophic microbial communities on the basis of patterns of community-level sole-carbon-source utilization. Applied and Environmental Microbiology 57: 2351–2359.
- Staddon WJ, Duchesne LC, Trevors JT. 1997. Microbial diversity and community structure of postdisturbance forest soils as determined by sole-carbon-source utilization patterns. Microbial Ecology 34: 125-130.