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Fungal-induced water repellency in sand

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FUNGAL-INDUCED WATER REPELLENCY IN SAND

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ABSTRACT

Water infiltration into granular soils and the associated pore water pressure increase and reduction in shear strength can trigger landslides, instability of vertical cuts and failure of retaining walls. Water repellent soils can reduce infiltration to maintain soil suction. Recent research has demonstrated the creation of synthetic water repellent soils using chemical methods. This paper investigates a biological treatment for creating water repellent sand via the growth of the fungus *Pleurotus ostreatus*. Water repellency was assessed using: (i) water drop penetration test, (ii) molarity of ethanol drop test and (iii) modified sessile drop method with contact angle (θ) determination via image analysis. Fungal-induced water repellency was found to be 'extreme' (θ >110°) up to 4 weeks and 'severe' (θ > 105°) up to12 weeks, even with no further supply of moisture or nutrients. A water repellent layer was formed and maintained in saturated conditions, which is difficult to achieve using chemical methods.

Keywords: *Water repellency; Ground improvement; Bio geotechnics; Fungi;*

1.1 Introduction

Water repellent soils, defined as having a liquid-solid contact angle $\theta > 90°$, can delay, reduce or prevent infiltration, lower evaporation and prevent capillary rise (e.g. Letey, *et al.,* 1962; Brandt, 1969; DeBano, 1981; Doerr *et al.*, 2000; Wang *et al.*, 2000; Rye and Smettem, 2017). As such they have been proposed to minimise infiltration into slopes, to divert water away from expansive soils, to improve drainage of sports surfaces, and to create barriers for use in landfill caps, or canal/trench liners (Dell'Avanzi *et al.*, 2010; Bardet *et al.*, 2011; Lourenço *et al.*, 2017). Water infiltration into granular soils and the associated pore water pressure increase and reduction in shear strength is a well-known mechanism which can trigger landslides, instability in vertical cuts and failure of retaining walls (Stanier and Tarantino, 2013; Scotto di Santolo *et al.*, 2017; Balzano *et al.,* 2019).

Recent research has demonstrated the creation of synthetic water repellent soils using chemically-based agents to coat soil grains including wax (Bardet *et al.*, 2011, 2014); silane compounds (Bachmann *et al.*, 2000; Bauters *et al.*, 2000; Daniels and Hourani, 2009; Byun et al., 2011; Lourenço *et al.*, 2015; Chan and Lourenço, 2016; Ng and Lourenço, 2016; Keatts *et al*., 2018) polytetrafluorethylene (PTFE) (Dell'Avanzi *et al.*, 2010; Lee *et al.*, 2015) and organic acids (Komatsu *et al.*, 2012; González-Peñaloza, 2013; Wijewardana *et al.*, 2015). The ability to induce water repellency in soils *in-situ* via chemical treatment may be limited by (i) the hazardous effects of some of the chemicals to humans and the environment, e.g. dimethyldichlorosilane (DMDCS) reacts with water to produce hydrogen chloride fumes (Chan and Lourenço, 2016) (ii) the conditions required for treatment, e.g. wax-coated soils require heating to high temperatures (Bardet, *et al.*, 2014), (iii) the influence of organic matter and (iv) the influence of residual water content; silanes are sensitive to residual water and organic matter in soils, requiring increased amounts of chemical compounds to be added with increasing water content (Chan and Lourenço, 2016; Ng and Lourenço, 2016). Naturally

occurring water repellency in soils is typically associated with increased soil erosion, as a result of reduced infiltration and increased surface runoff (e.g. Doerr *et al.*, 2006); similar behaviour has also been observed in chemically-induced water repellent soils (Zheng *et al.*, 2017, 2019).

This paper proposes that water repellency in granular soil could be engineered by biological treatment *in-situ* with fungi in order to reduce water infiltration (e.g. in slopes) and thus maintain soil suction and strength. Filamentous fungi grow via the formation of hyphae, multi-cellular tube-like structures, which also serve as distribution channels for the protein compounds they secrete. Most filamentous fungi secrete hydrophobins, proteins which have both hydrophilic and hydrophobic ends (amphipathic), enabling the fungi to hydrophobise wettable surfaces and vice-versa (Wessels, 1996, 2000; Wösten, 2001). A network of hyphae, known as the mycelium, is created as a filamentous fungus grows. This along with the exudates, results in the physical enmeshment of soil particles and an overall enhanced resistance to wind or water erosion (Vogelsang *et al.*, 2004; Tisdall *et al.*, 2012; Mardhiah *et al.*, 2016).

The aim of this paper is to assess the level of water repellency that can be induced in sterile sand treated with *Pleurotus ostreatus,* a non-pathogenic, non-parasitic, saprotrophic, filamentous fungus. Specifically, the objectives are to:

(1) Assess the degree and persistence of water repellency induced over a 12-week growth period (Experiment 1).
(2) Investigate the influence of initial degree of saturation (Experiment 2).
(3) Investigate the influence of disruption of the mycelium network (Experiment 3).

1.2 Materials and Methods

1.2.1 Experimental design

Three experiments were conducted to address the objectives, the details of each are presented in Table 1.

Table 1: Experimental design: treatment conditions and testing methods

Experiment	Fungal Inoculation Method	S_r (%)	L_c (%)	Temperature (°C)	Relative Humidity (%)	Growth duration (weeks)	Mixing	Test method
1	Spore/ hyphal suspension	15.3	5.5	25	40	1 2 4 8 12	No	WDPT MEDT Modified SDM
2	Colonised beech wood	4.2 7.2 15.2 24.2 34.3 100	4.8	25	100	1	No	WDPT
3	Spawn grains	15.3	5.5	25	40	3 (total)	On days 6, 9, 11, 13, 15, 17, 19	WDPT MEDT Modified SDM

S_r= degree of saturation; for the treated specimens in Exp 1 the liquid was made of the fungal suspension, while in the control specimens the liquid was deionised water.
L_c = lignocel content in total solids
WDPT = water drop penetration time test
MEDT = molarity of ethanol droplet test
SDM = sessile drop method

1.2.2 Preparation of specimens

A fine sand, with particle size ranging from 0.075 – 0.425 mm, (C_u = 1.7), was mixed with lignocellulose, natural wood fibres sized 0.5mm – 1mm (J. Rettenmaier & Söhne GmbH). The lignocellulose provided the carbon source for fungal growth, and was sterilised by autoclaving at 121°C for 21 minutes.

In Experiment 1, the spore/hyphal suspension of *P. ostreatus* (strain M 2191) was prepared by mixing 10 g of spawn (mycelium grown on millet grains) in 100 mL of deionised water and

shaking vigorously for 10 mins, then placing on a shaker at 150 rpm for 20 mins, followed by another 5 mins of vigorous shaking by hand to facilitate detachment of fungal spores and hyphae from the grains into the water. Using a 2 mm mesh sized sieve, the grains were removed. The resulting filtrate formed the liquid content of the fungal treated specimens. In the control specimens water was added in place of the spore/hyphal suspension.

In Experiment 2 Colonised beech wood inocula were prepared according to the method described in Donnelly and Boddy (1998). Each specimen was inoculated by placing a cube of beech wood colonised by *P. ostreatus* on the soil surface in the middle of the petri-dish.

In Experiment 3, specimens were treated by placing grains of spawn of *P. ostreatus* directly on to the specimen surface. To determine if fungal induced soil water repellency persists when the mycelium networks are disrupted, (which may occur in the field due to human or animal actions), specimens were thoroughly mixed using a spatula 6 days after inoculation. WDPT tests were performed on Day 6 prior to and immediately after mixing. WDPT tests were carried out again on Day 7. Specimens were then mixed every 48 hours and tested on Days 12 and 20.

1.2.3 Assessment of water repellency

The procedures followed in assessing WR for the respective experiments are presented in Table 2. A modified Sessile Drop Method was used as the standard method involves sampling using double-sided adhesive and this was considered too invasive, disturbing the arrangement of the fungal mycelium on the sand grains.

Table 2: Methods and procedures for the assessment of water repellency

Method	Procedure	References
Water drop penetration time (WDPT) test	Time recorded for a water droplet (vol. 10 µL) released from a height of 5 mm above the specimen surface, to infiltrate the soil. Repeated 5 times per specimen.	Letey (1969); Doerr (1998)
Molarity of Ethanol Droplet test (MEDT)	Droplets of ethanol solution (vol. 50µL) released from a height of 5 mm above the specimen surface. Solutions of ethanol at increasing concentrations from 0 – 6 mol L^{-1} (at increments of 0.2 mol L^{-1}) were used. Repeated 3 times per specimen. The 90° liquid surface tension of the infiltrating droplet ($\gamma_{90°}$) and subsequently the contact angle (θ) were determined using the equations given by Carillo *et al.*, (1999) and Leelamanie *et al.*, (2008).	Doerr, (1998); Carrillo *et al.*, (1999); Roy and McGill, (2002); Leelamanie *et al*, (2008); Moody and Schlossberg (2010)
Modified Sessile Drop Method (SDM) with CA determination via image analysis	Timelapse images recorded of water droplets (vol. 10 µL) released on to bulk specimen surface at 1s intervals using a 1000X-USB-Microscope-Camera. 3 measurements per specimen. The contact angle was estimated using the Low Bond Axisymmetric Drop Shape Analysis (LBADSA) plugin in *ImageJ* software.	Bachmann *et al.* (2000, 2003); Stalder *et al.* (2010)

The relationship between the level of water repellency, water penetration time, and equivalent contact angles for sands is shown in Table 3.

Table 3: Relationship between WDPT, level of water repellency and equivalent contact angles for sands (After Bisdom, Dekker and Schoute, 1993; Dekker and Ritsema, 1994; Doerr et al., 2006; Liu et al., 2012)

Level of water repellency	Water drop penetration time (s)	Equivalent contact angle θ for sands (°)
Hydrophilic	<5	0
Slight	6 – 60	0 – 80
Moderate	61 – 600	80 – 110
Severe	601 – 3600	
Extreme	>3600	>110

1.3 Results

Experiment 1: Persistence of water repellency with time

After 1 week of growth water droplets remained on the surface of fungal treated sand beyond 24 hours and formed CAs ~118°. Whereas for the corresponding untreated control specimen water droplets infiltrated immediately within ≤2 s meaning that CAs could not be accurately

measured. Figures 1a and 1b show an untreated specimen on Day 0 and after 12 weeks incubation. In contrast 1c, d, e show water droplets on a treated specimen after 12 weeks of fungal growth. Figure 2 and Table 4 show the evolution of CAs and WDPT, with growth duration. The CAs and WDPT indicate that the fungal treated specimens are extreme to severely water repellent up to 8 weeks after inoculation and although both reduce with increasing growth duration, after 12 weeks the fungal treated sand remains severely water repellent. Even under these harsh environmental conditions where no additional moisture or nutrients were supplied to the sterile sand specimens after initial treatment, water repellency persisted for the entire 12 weeks investigated.

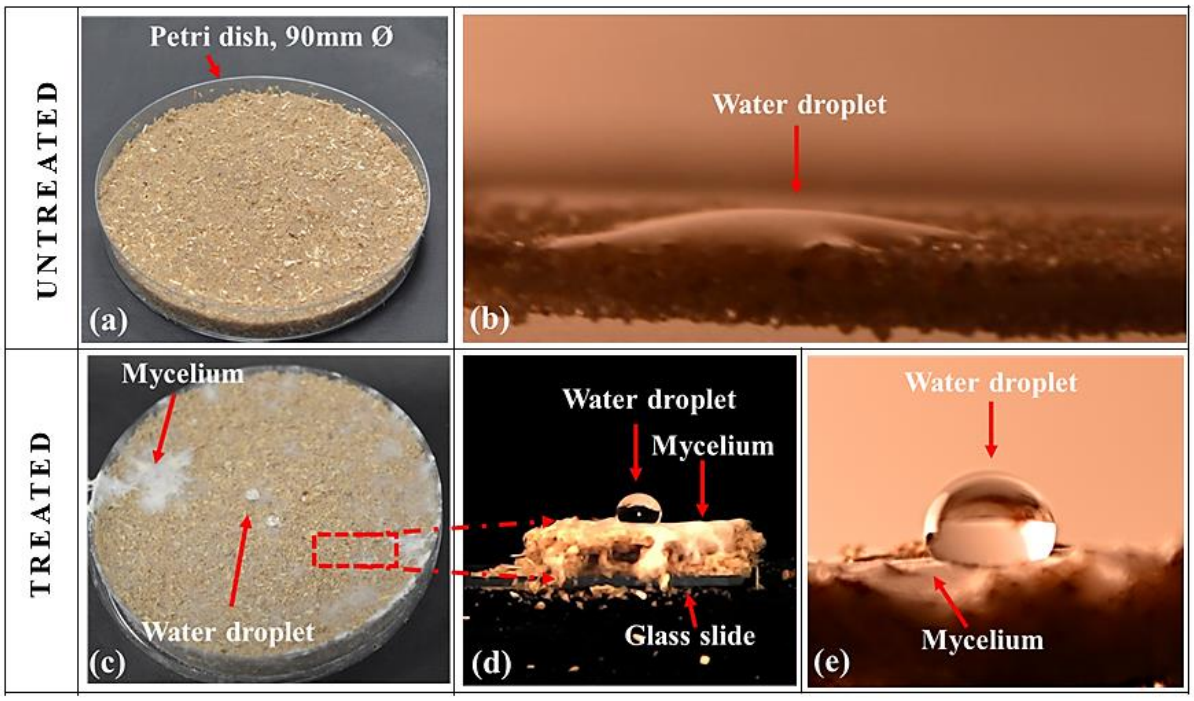

Figure 1: Untreated specimens: (a) on Day 0 and (b) after 12 weeks incubation. Water droplets on treated specimens after a growth period of 12 weeks: (c), (d) and (e); Images captured from the modified SDM set up for the treated specimen after a growth period of 12 weeks.

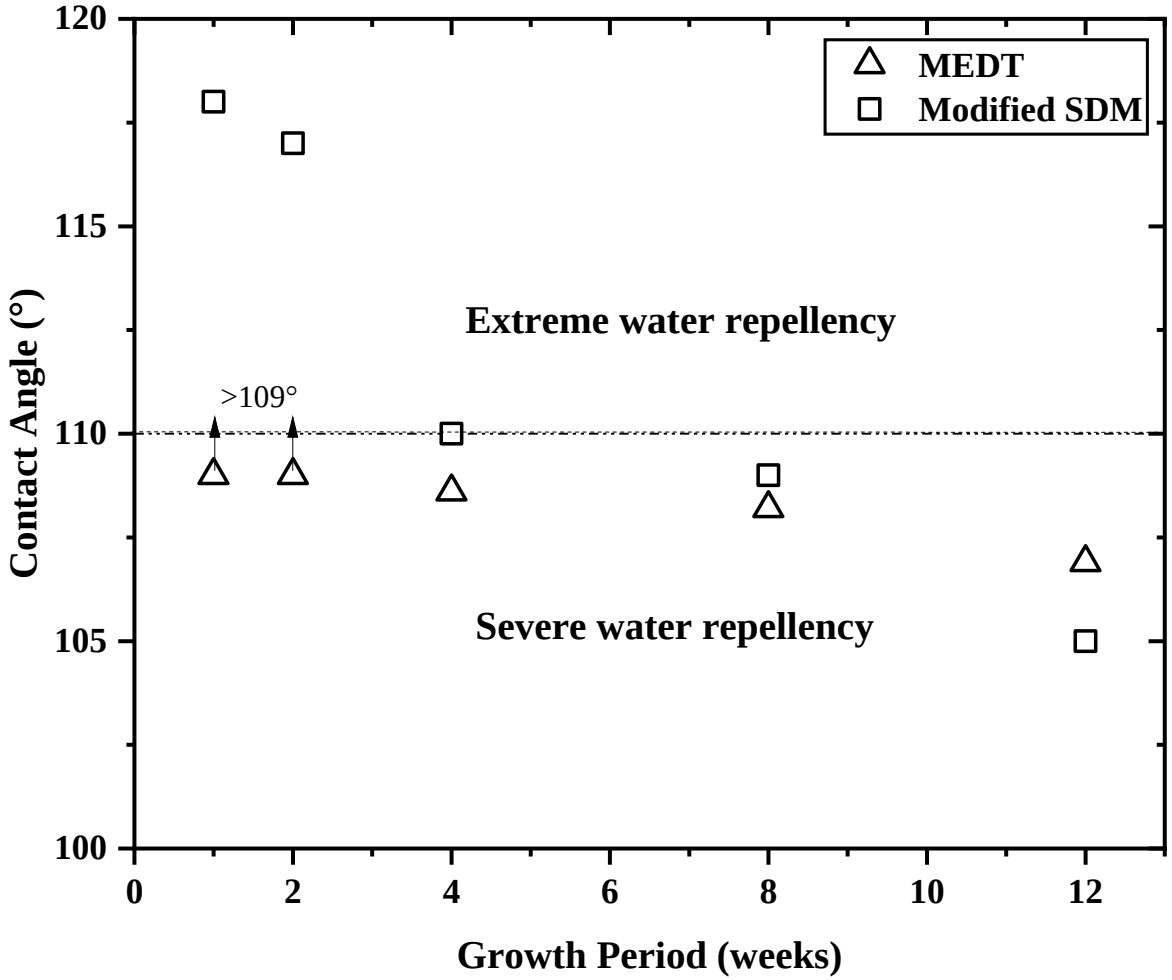

Figure 2: Evolution of contact angle with growth duration. Levels of water repellency are shown based on classification presented in Table 2 .

Table 4: Water drop penetration time evolution with growth duration

Growth period (weeks)	Water Drop Penetration Time (WDPT)	Classification of WR
1	>24hrs	Extreme
2	>1hr	Extreme
4	>1hr	Extreme
8	3400s	Severe
12	607s	Severe

Experiment 2: Influence of initial degree of saturation (S_r)

Water repellency was consistently extreme (with WDPT >3600s) as S_r increased from 4.2 to 15.2% but decreased to 'severe' and 'moderate' levels as S_r increased from 24.2 to 100%

(Figure 3). It is worth noting that water repellency was not lost even at saturation (Figure 3). At saturation the fungal mycelium formed a hydrophobic layer at the water-air interface, rather than growing directly around the sand grains (Fig. 4). It was observed that irrespective of initial S_r, *P. ostreatus* induced WR wherever fungal mycelium was visible. This is a significant finding, given that for most chemical methods of inducing water repellency (wax, DMDCS, stearic acid), the soils should initially be in an dry state (Leelamanie and Karube, 2007; Bardet, Jesmani and Jabbari, 2014; Chan and Lourenço, 2016).

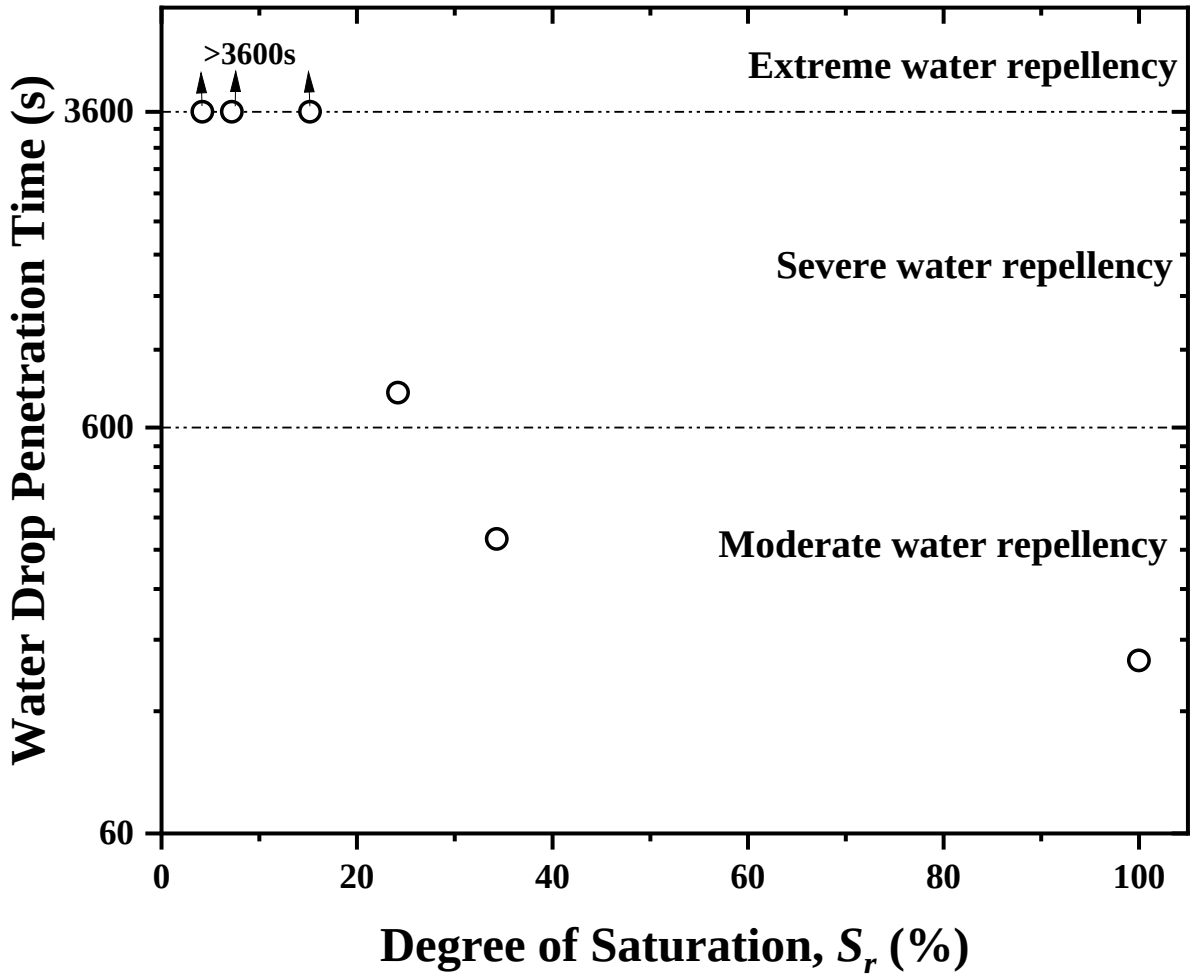

Figure 3: Influence of initial degree of saturation on water drop penetration time. Levels of water repellency are shown based on classification presented in Table 2.

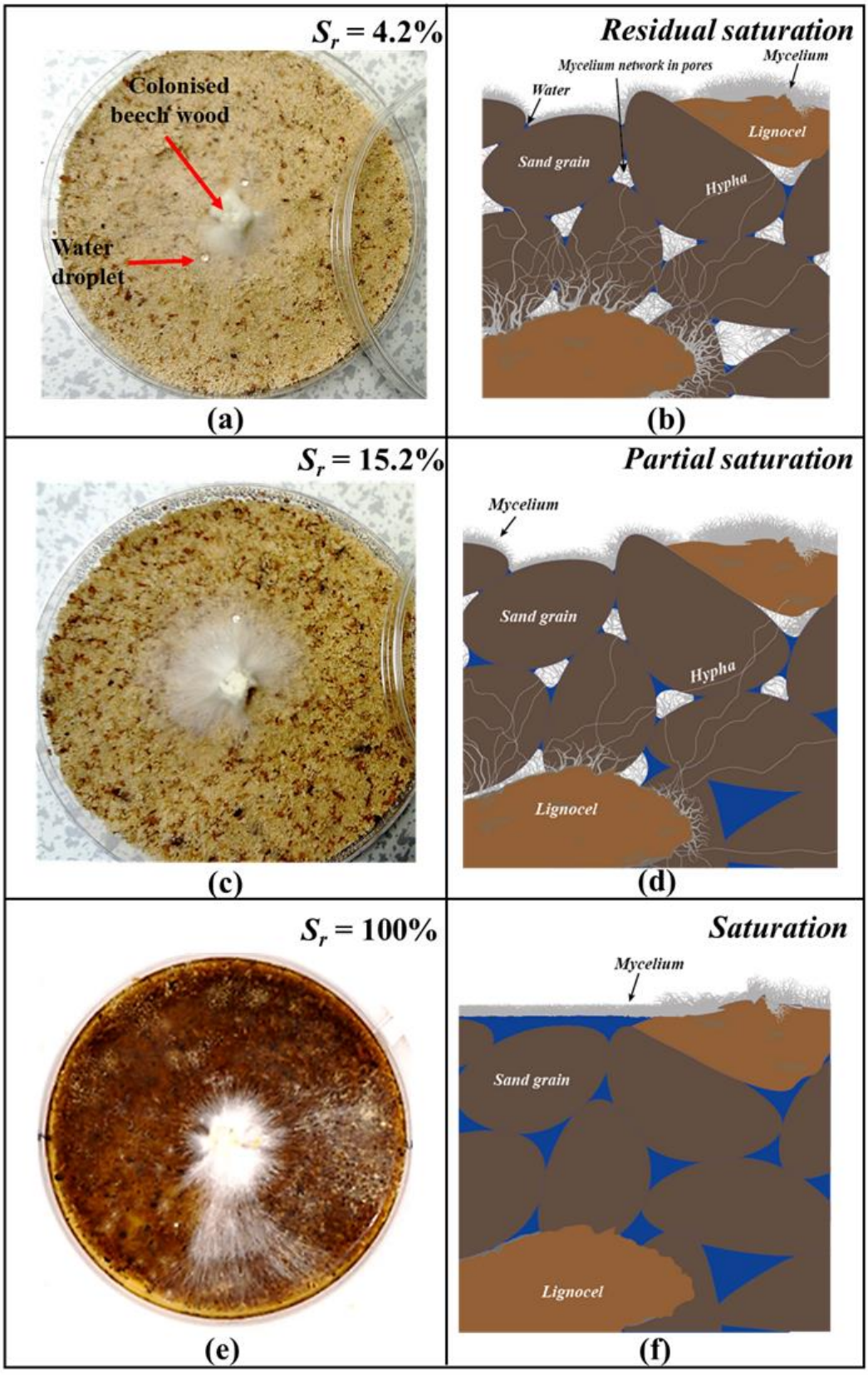

Figure 4: Photographs and schematics showing formation of hydrophobic layers at different saturation states. At residual saturation (a &b) mycelium grows within pore space and on the surface of the specimen. At partial saturation (c & d) mycelium grows within reduced pore space and on the surface of the specimen. At saturation (e & f) mycelium forms only on the surface of the specimen.

Experiment 3: Influence of the disruption of the mycelium network

For the fungal treated specimens after 6 days of growth, the water droplets did not infiltrate even after 24 hrs and formed CAs of ~110° (Fig 5a & b). However, immediately after mixing the soil, the WDPT was < 5s. After 24hrs, slight-moderate water repellency was regained. Thereafter the specimens were mixed every 48 hrs (dashed vertical lines in Figure 5). It is evident that despite regular mixing of the sand specimens WR was regained, from slight hydrophobicity (WDPT = 134s) on Day 7 to severe hydrophobicity on Day 12 (WDPT 1565 s; CA ~100°) and finally to extreme hydrophobicity (WDPT 3400s; CA 115°) on Day 20.

Although immediately after mixing on Day 6, water repellency had been lost, these results suggest that 24hrs is sufficient for the fungal treated specimens to restore some level of hydrophobic behaviour, without any further introduction of fungal inoculation or nutrients. Two mechanisms are suggested for the persistence of WR after mycelium disruption: (i) the repeated mixing redistributes the hydrophobins, such that they progressively coat grain surfaces. This may explain why WR remains, and is extreme even when there is no obvious mass of fungal mycelium remaining on the sand surface after repeated mixing (Figure 6). The specimen has transitioned from one with a hydrophobic surface layer to a sand with hydrophobised grains. (ii) Filamentous fungi are naturally self-healing, such that if hyphal cells become damaged, septal pores can be plugged using Woronin bodies or septal pore caps, preventing loss of cytoplasmic fluids and ensuring continuous hyphal growth (Jedd and Pieuchot, 2012).

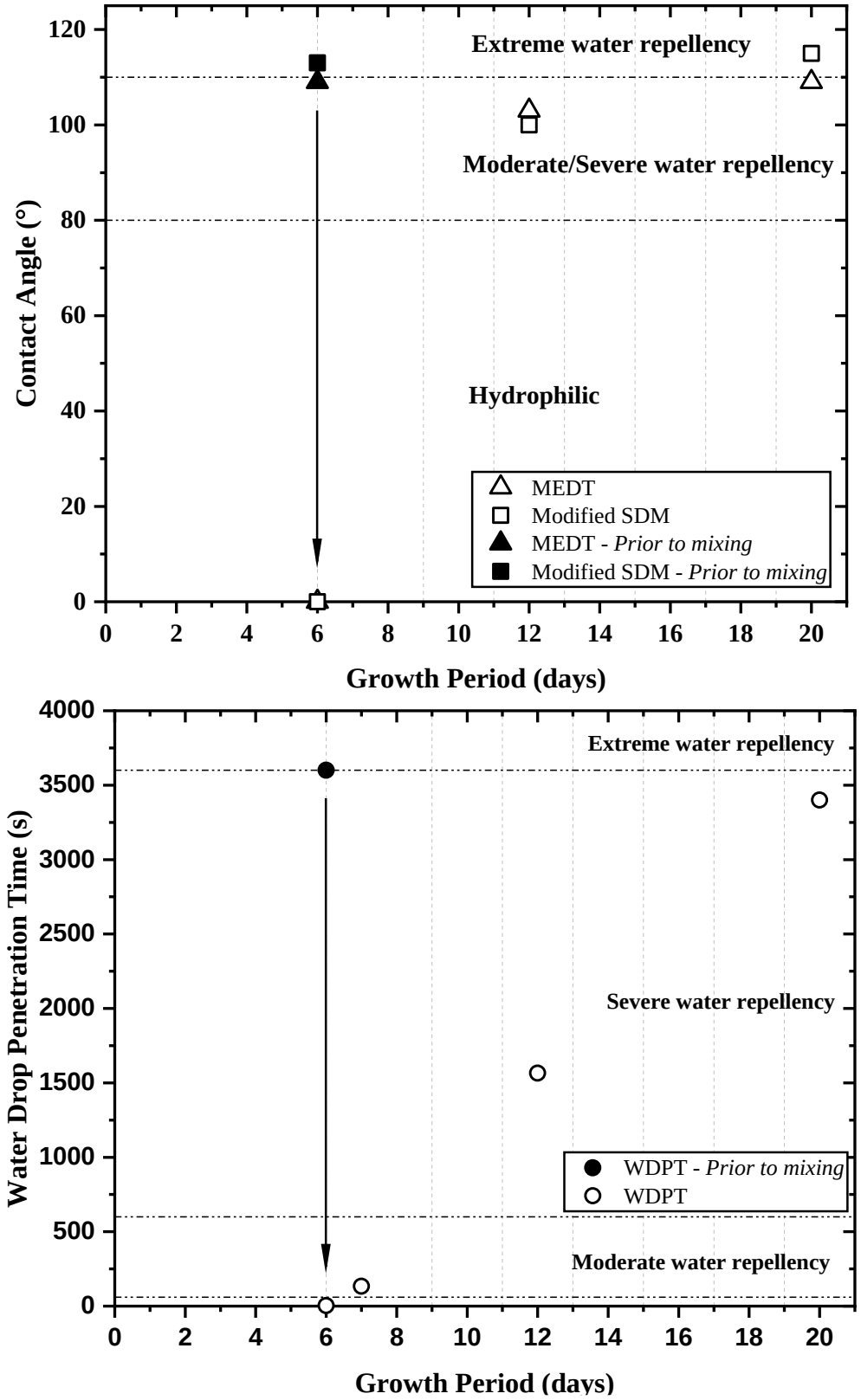

Figure 5: Assessment of water repellency for treated soil specimens subjected to mixing every 48hrs to disrupt fungal growth up to 20 days: (a) Contact angle and (b) Water drop penetration time. The dashed vertical lines indicate days on which specimen. The dashed vertical lines indicate days on which specimens were mixed. The arrows show that the initially extreme WR exhibited by specimens (black symbols) was completely lost immediately after mixing on the 6th day.

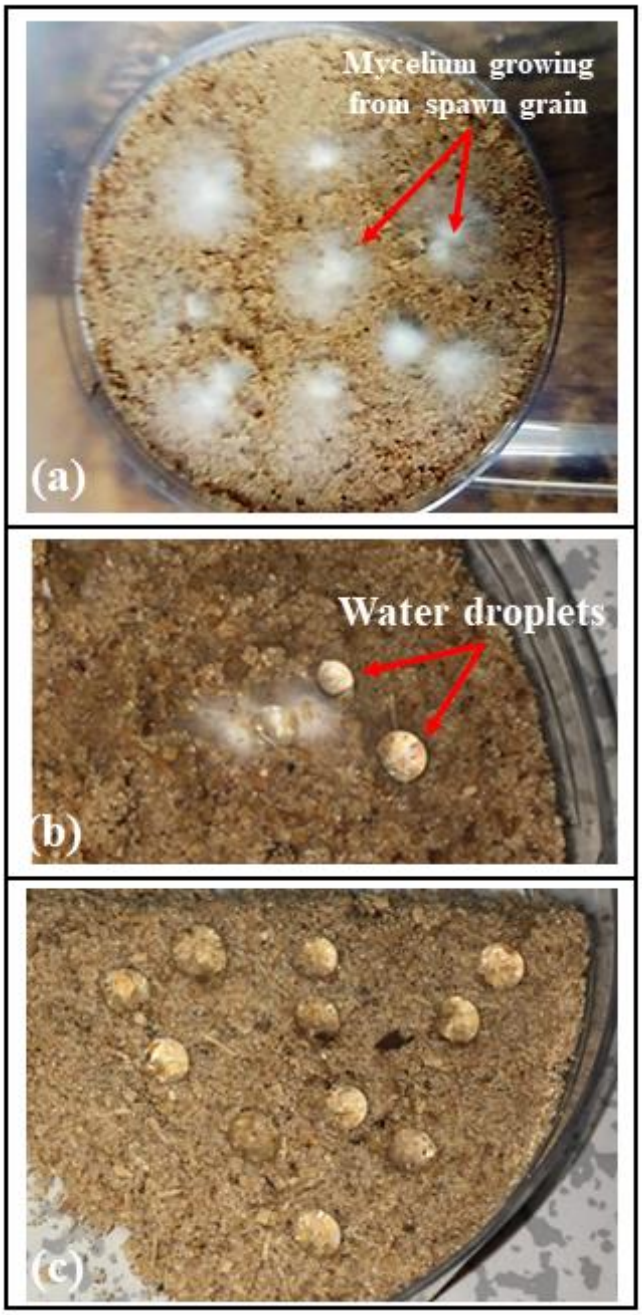

Figure 6: Photographs illustrating that mycelium (i.e. fungal hyphae) become less visible with increased mixing (a) specimen on the 6th day before testing (b) water droplet on specimen on the 6th day immediately before mixing (c) several water droplets on specimen with no visible mycelium on the 20th day after being subjected to 48-hourly mixing.

1.4 Discussion

Fungal treatment has promise in applications which have previously considered the use of chemically hydrophobised soils as fill materials, moreover it has potential to be used to induce water repellency in soils *in-situ*, which is currently only possible with some of the chemical

177 techniques reported in the literature (e.g. organosilanes, Daniels and Hourani, 2009). Treatment
178 of granular soils in slopes/hillsides where failures are triggered in response to wetting could be
179 carried out via fungal treatment to reduce infiltration, thereby maintaining higher levels of
180 suction and associated strength, without enhancing soil erosion.

181 Fungal treatment has a number of characteristics which makes it attractive for *in-situ* treatment:
182 (i) fungi can grow to massive sizes, a single organism in N. America is ~150,000m^2 (Smith *et*
183 *al.,* 1992), (ii) fungi can be incredibly long-lived with individuals dated as ~1,500years old,
184 (Smith *et al.,* 1992), (iii) fungal treatment would be inexpensive and could be carried out in a
185 similar manner to hydroseeding, (iv) water repellency can be induced irrespective of initial
186 saturation, (v) fungi can survive and even thrive in nutrient poor environments (Sterflinger,
187 2000; Gorbushina, 2007; Cantrell *et al.*, 2011) and (vi) fungal hyphae and mycelium networks
188 are self-healing (Müller *et al.*, 1998; Plamann, 2009; Jedd and Pieuchot, 2012; Tegelaar and
189 Wösten, 2017).

190 Although water repellency has been assessed here as a first step using contact angles, it should
191 be noted that intrusion of water into fungal treated soils is also a function of the surface tension,
192 pore size and geometry, all of which may be altered by the presence of fungal biomass, and the
193 water pressure applied (Washburn, 1921). Further research is needed to understand these
194 changes alongside a detailed investigation of the hydraulic behaviour (including determination
195 of the water-entry pressure required for breakthrough, hydraulic conductivity) following fungal
196 treatment in order to understand its potential for reducing infiltration into granular soils. Indeed,
197 for other applications, this should be investigated for a range of soil types as fungal activity
198 may enhance the formation of clay aggregates, increase clay porosity (Chenu, 1989), and align
199 clay particles along hyphae (Rilig and Mummey, 2006) with the potential for the development
200 of preferential flow paths.

While some fungal organisms are known to be very long lived in the natural environment with sufficient nutrient availability, (Smith et al., 1992), the survival of *P. ostreatus* in extreme environmental conditions (e.g. wetting-drying cycles) remains to be investigated. Although some studies have reported that fungal biochemical exudates (such as glomalin produced by arbuscular mycorrhizal fungi) may persist in the soil long after decomposition of fungal hyphae (Rillig and Mummey, 2006), it is unclear at present for how long residual water repellency could be sustained.

This study has presented the hydrophobic effects due to the introduction of a single fungal species: *P. ostreatus*. Yet it should be noted that there are 99,000 known fungal species (Carris et al., 2012). Indeed *P. ostreatus* is a saprotrophic fungus which digests non-living organic matter, i.e. wood, as such it would not be suitable for deployment near wooden structures due to the potential for decay. It is anticipated that many other fungal species could also induce similar water repellent behaviour in soils to that presented herein. The selection of a species should also consider if there is any potential for damage to other infrastructure on-site under low nutrient conditions. Furthermore, the success of bioaugmentation strategies (i.e. the introduction of a single species) *in situ* can be limited by a decline in microbial population after injection due to environmental conditions but primarily as a result of biotic effects e.g. predation and/or competition by native microbial communities (Van Veen et al., 1997). Biostimulation of native microorganisms may be a more promising strategy in the long-term.

1.5 Conclusion

This study proposes for the first time that water repellent sand could be created for ground engineering applications using fungal treatment. Water repellency was induced in sterile sands via the growth of *P. ostreatus*. The findings of this study are:

1. Fungal-induced WR was found to be extreme (CAs ≥ 110°) for up to 4 weeks, and severe for up to 12 weeks (CAs ≥105°) in a resource-depleting environment.
2. WR induced by *P. ostreatus* reduces with increasing initial saturation. However, a hydrophobic layer with moderate water repellency can still be created even in fully saturated conditions.
3. Disruption of fungal mycelium networks in sand results in a loss of WR but this was shown to re-establish within 24 hours.

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Data Statement

Data associated with this publication are openly available from the University of Strathclyde KnowledgeBase at https://doi.org/10.15129/1671ac61-507a-4fc9-a215-370c4178c8c5.

References

Bachmann, J. *et al.* (2003) 'Extended methodology for determining wetting properties of porous media', *Water Resources Research*, 39(12), pp. 1–14. doi: 10.1029/2003WR002143.

Bachmann, J., Ellies, A. and Hartge, K. H. (2000) 'Development and application of a new sessile drop contact angle method to assess soil water repellency', *Journal of Hydrology*, 231–232, pp. 66–75. doi: 10.1016/S0022-1694(00)00184-0.

Balzano, B., Tarantino, A. and Ridley, A. (2019) 'Preliminary analysis on the impacts of the rhizosphere on occurrence of rainfall-induced shallow landslides', *Landslides*, 16(10), pp. 1885–1901. doi: 10.1007/s10346-019-01197-5.

Bardet, J. P., Jesmani, M. and Jabbari, N. (2011) 'Effects of compaction on shear strength of wax-coated sandy soils', *Electronic Journal of Geotechnical Engineering*, 16 D, pp. 451–461.

Bardet, J. P., Jesmani, M. and Jabbari, N. (2014) 'Permeability and compressibility of wax-coated sands', *Geotechnique*, 64(5), pp. 341–350. doi: 10.1680/geot.13.P.118.

Bauters, T. W. J. *et al.* (2000) 'Physics of water repellent soils', *Journal of Hydrology*, 231–232, pp. 233–243. doi: 10.1016/S0022-1694(00)00197-9.

Bisdom, E. B. A., Dekker, L. W. and Schoute, J. F. T. (1993) 'Water repellency of sieve fractions from sandy soils and relationships with organic material and soil structure', *Geoderma*, 56(1), pp. 105–118. doi: https://doi.org/10.1016/0016-7061(93)90103-R.

Brandt, G. H. (1969) 'Water movement in hydrophobic soils', in *Proceedings of the Symposium on Water Repellent Soils. Univ. Calif., Riverside, CA*, pp. 91–115.

Byun, Y., M. K. Tran, T. S. Yun, J. Lee (2012) Strength and stiffness characteristics of unsaturated hydrophobic granular media, Geotechnical Testing Journal, 35(1), 1-8. https://doi.org/10.1520/GTJ103650.

Cantrell, S. A. *et al.* (2011) 'Unusual fungal niches', *Mycologia*, 103(6), pp. 1161–1174. doi: 10.3852/11-108.

Carrillo, M. L. ., Letey, J. and Yates, S. R. (1999) 'Measurement of initial soil-water contact angle of water repellent soils', *Soil Science Society of America Journal*, 63, pp. 433–436.

Carris, L. M., Little, C. R., & Stiles, C. M. (2012). Introduction to fungi. The plant health instructor. https://doi.org/10.1094/PHI-I-2012-0426-01 Available at: http://www.apsnet.org/edcenter/intropp/PathogenGroups/Pages/IntroFungi.aspx.

Chan, C. S. H. and Lourenço, S. D. N. (2016) 'Comparison of three silane compounds to impart water repellency in an industrial sand', *Geotechnique Letters*, 6(4), pp. 263–266. doi: 10.1680/jgele.16.00097.

Chau, H. W. *et al.* (2012) 'Wetting properties of fungi mycelium alter soil infiltration and soil water repellency in a γ-sterilized wettable and repellent soil', *Fungal Biology*. Elsevier Ltd, 116(12), pp. 1212–1218. doi: 10.1016/j.funbio.2012.10.004.

Daniels, J. L. and Hourani, M. S. (2009) 'Soil improvement with organo-Silane', *Advances in Ground Improvement (GSP 188)*, (188), pp. 217–224. doi: 10.1061/41025(338)23.

DeBano, L. F. (1981) 'Water Repellent Soils: a state-of-the-art. General Technical Report PSW-GTR-46', *US Department of Agriculture Forest Service Pacific Southwest Forest and Range Exp. Stn.*, pp. 1–25. doi: 10.2737/PSW-GTR-46.

Dekker, L. W. and Ritsema, C. J. (1994) 'How water moves in a water repellent sandy soil: 1. Potential and actual water repellency', *Water Resources Research*, 30(9), pp. 2507–2517. doi: 10.1029/94WR00749.

Dell'Avanzi, E. *et al.* (2010) 'Potential use of induced soil-water repellency techniques to improve the performance of landfill's alternative final cover systems', in Buzzi, O., Fityus, S., and Sheng, D. (eds) *Unsaturated soils: Experimental studies in unsaturated soils and expansive soils*. Crc Press, Taylor and Francis group, London, pp. 461–466.

Doerr, S. H. (1998) 'On standardizing the "water drop penetration time" and the "molarity of an ethanol droplet" techniques to classify soil hydrophobicity: a case study using medium textured soils', *Earth Surface Processes and Landforms*, 23(7), pp. 663–668. doi: 10.1002/(SICI)1096-9837(199807)23:7<663::AID-ESP909>3.0.CO;2-6.

Doerr, S. H. *et al.* (2006) 'Occurrence, prediction and hydrological effects of water repellency amongst major soil and land-use types in a humid temperate climate', *European Journal of Soil Science*, 57(5), pp. 741–754. doi: 10.1111/j.1365-2389.2006.00818.x.

Doerr, S. H., Shakesby, R. A. and Walsh, R. P. D. (2000) 'Soil water repellency: its causes, characteristics and hydro-geomorphological significance', *Earth-Science Reviews*, 51(1), pp.

33–65. doi: https://doi.org/10.1016/S0012-8252(00)00011-8.

Donnelly, D. and Boddy, L. (1998) 'Developmental and morphological responses of mycelial systems of *Stropharia caerulea* and *Phanerochaete velutina* to soil nutrient enrichment', *New Phytologist*, 138, pp. 519–531. doi: Doi 10.1046/J.1469-8137.1998.00117.X.

González-Peñaloza, F.A., Zavala, L.M., Jordán, A., Bellinfante, N., Bàrcenas-Moreno, G., Mataix-Solera, J., Granged, A.J.P., Granja-Martins, F.M., and Neto-Paixão, H.M. (2013). "Water repellency as conditioned by particle size and drying in hydrophobized sand", Geoderma, Vol., 209-210, 31-40.

Gorbushina, A. A. (2007) 'Life on the rocks', *Environmental Microbiology*, 9(7), pp. 1613–1631. doi: 10.1111/j.1462-2920.2007.01301.x.

Jedd, G. and Pieuchot, L. (2012) 'Multiple modes for gatekeeping at fungal cell-to-cell channels', *Molecular Microbiology*, 86(6), pp. 1291–1294. doi: 10.1111/mmi.12074.

Kallio, J. M., Linder, M. B. and Rouvinen, J. (2007) 'Crystal structures of hydrophobin HFBII in the presence of detergent implicate the formation of fibrils and monolayer films', *Journal of Biological Chemistry*, 282(39), pp. 28733–28739. doi: 10.1074/jbc.M704238200.

Keatts, M.I., Daniels, J.L., Langley, W.G., Pando, M.A. and Ogunro, V.O. (2018) "Apparent Contact Angle and Water Entry Head Measurements for Organo-Silane Modified Sand and Coal Fly Ash", ASCE Journal of Geotechnical and Geoenvironmental Engineering 144 (6) 1-9.

Komatsu, T. *et al.* (2012) 'Characterizing Time-Dependent Contact Angles for Sands Hydrophobized with Oleic and Stearic Acids', *Vadose Zone Journal*, 11(1), p. 0. doi: 10.2136/vzj2011.0055.

Lee, C. *et al.* (2015) 'Water-Entry Pressure and Friction Angle in an Artificially Synthesized Water-Repellent Silty Soil', *Vadose Zone Journal*, 14(4), p. 0. doi: 10.2136/vzj2014.08.0106.

Leelamanie, D. and Karube, J. (2007) 'Effects of organic compounds, water content and clay on the water repellency of a model sandy soil', *Soil Science and Plant Nutrition*, 53(6), pp. 711–719. doi: 10.1111/j.1747-0765.2007.00199.x.

Leelamanie, D., Karube, J. and Yoshida, A. (2008) 'Characterizing water repellency indices: Contact angle and water drop penetration time of hydrophobized sand', *Soil Science and Plant Nutrition*, 54(2), pp. 179–187. doi: 10.1111/j.1747-0765.2007.00232.x.

Letey, J. (1969) 'Measurement of contact angle, water drop penetration time, critical surface energy', in De Bano, L. F. and Letey, J. (eds) *Proceedings of Symposium on Water-Repellent Soils*. Univ. of Calif., Riverside, pp. 43–47.

Letey, J., Osborn, J. and Pelishek, R. E. (1962) 'The influence of the water-solid contact angle on water movement in soil', *International Association of Scientific Hydrology. Bulletin*, 7(3), pp. 75–81. doi: 10.1080/02626666209493272.

Liu, H. *et al.* (2012) 'Artifi cial Hydrophobic Soils and Its Relevance for Soil Water Desorption Curves', *Soil Science Society of America Journal*, 76(2), pp. 342–349. doi: 10.2136/sssaj11.0081.

Lourenço, S. D. N. *et al.* (2017) 'Soil wettability in ground engineering: fundamentals, methods, and applications', *Acta Geotechnica*, 13(1). doi: 10.1007/s11440-017-0570-0.

Lourenço, S. D. N., Wang, G.-H. and Kamai, T. (2015) 'Water repellent soils for slope stability', in *Proceedings of the XVI ECSMGE Geotechnical Engineering for Infrastructure and Development*, pp. 1609–1613. doi: 10.1680/ecsmge.60678.

Mardhiah, U. *et al.* (2016) 'Arbuscular mycorrhizal fungal hyphae reduce soil erosion by surface water flow in a greenhouse experiment', *Applied Soil Ecology*. Elsevier B.V., 99, pp. 137–140. doi: 10.1016/j.apsoil.2015.11.027.

Moody, D. R. and Schlossberg, M. J. (2010) 'Soil Water Repellency Index Prediction Using the Molarity of Ethanol Droplet Test', *Vadose Zone Journal*, 9(4), p. 1046. doi: 10.2136/vzj2009.0119.

Müller, W. H. *et al.* (1998) 'Structural differences between two types of basidiomycete septal pore caps', *Microbiology*, 144(7), pp. 1721–1730. doi: 10.1099/00221287-144-7-1721.

Ng, S. H. Y. and Lourenço, S. D. N. (2016) 'Conditions to induce water repellency in soils with dimethyldichlorosilane', *Geotechnique*, 66(5), pp. 441–444. doi: 10.1680/jgeot.15.T.025.

Plamann, M. (2009) 'Cytoplasmic Streaming in Neurospora: Disperse the Plug To Increase the Flow?', *PLoS Genetics*, 5(6), p. e1000526. doi: 10.1371/journal.pgen.1000526.

Rillig, M. C. and Mummey, D. L. (2006) 'Tansley review Mycorrhizas and soil structure', pp. 41–53. doi: 10.1111/j.1469-8137.2006.01750.x.

Roy, J. L. and McGill, W. B. (2002) 'Assessing SOil Water Repellency Using the Molarity of Ethanol Droplet (MED) Test', *Soil Science*, 167(2), pp. 83–97.

Rye, C. F. and Smettem, K. R. J. (2017) 'The effect of water repellent soil surface layers on preferential flow and bare soil evaporation', *Geoderma*. Elsevier B.V., 289, pp. 142–149. doi: 10.1016/j.geoderma.2016.11.032.

Scotto di Santolo, A., Evangelista, L. and Evangelista, A. (2017) 'Gravity retaining walls in unsaturated pyroclastic materials: Observations and numerical modeling', *Rivista Italiana di Geotecnica*, 51(1), pp. 26–42.

Shokri, N., Lehmann, P. and Or, D. (2009) 'Characteristics of evaporation from partially wettable porous media', *Water Resources Research*, 45(2), pp. 1–12. doi: 10.1029/2008WR007185.

Smith, M. L., Bruhn, J. N. and Anderson, J. B. (1992) 'The fungus Armillaria bulbosa is among the largest and oldest living organisms', *Nature*, 356(6368), pp. 428–431. doi: 10.1038/356428a0.

Stalder, A. F. *et al.* (2010) 'Low-bond axisymmetric drop shape analysis for surface tension and contact angle measurements of sessile drops', *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 364(1–3), pp. 72–81. doi: 10.1016/j.colsurfa.2010.04.040.

Stanier, S. A. and Tarantino, A. (2013) 'An approach for predicting the stability of vertical cuts in cohesionless soils above the water table', *Engineering Geology*, 158, pp. 98–108. doi: 10.1016/j.enggeo.2013.03.012.

Sterflinger, K. (2000) 'Fungi as geologic agents', *Geomicrobiology Journal*, 17(2), pp. 97–124. doi: 10.1080/01490450050023791.

Tegelaar, M. and Wösten, H. A. B. (2017) 'Functional distinction of hyphal compartments', *Scientific Reports*, 7(1), pp. 6–11. doi: 10.1038/s41598-017-06422-6.

Tisdall, J. M. *et al.* (2012) 'Stabilisation of soil against wind erosion by six saprotrophic fungi', *Soil Biology and Biochemistry*. Elsevier Ltd, 50, pp. 134–141. doi: 10.1016/j.soilbio.2012.02.035.

Van Veen, J. A., Van Overbeek, L. S., Van Elsas, J. D. (1997) 'Fate and activity of microrganisms introduced into soil, *Microbiological and Molecular Biology Reviews*, Vol. 61, No. 2, p121-135.

Vogelsang, K. M. *et al.* (2004) *The Use of Mycorrhizal Fungi in Erosion Control Applications, Contract Report*. Available at: https://pdfs.semanticscholar.org/3c44/156d03c80e327913c06fac4fa8084d5584d0.pdf?_ga=2.216907669.2012585161.1572648647-240717194.1572648647.

Wang, Z. *et al.* (2003) 'Effects of water repellency on infiltration rate and flow instability', *Soil Water Repellency: Occurrence, Consequences, and Amelioration*, 232, pp. 235–244. doi: 10.1016/B978-0-444-51269-7.50024-2.

Washburn, E. W. (1921) 'The Dynamics of Capillary Flow', *The Physical Review*, 17(3), pp. 273–383.

Wessels, J. G. H. *et al.* (1991) 'The thn mutation of Schizophyllum commune, which suppresses formation of aerial hyphae, affects expression of the Sc3 hydrophobin gene', *Journal of General Microbiology*, 137(10), pp. 2439–2445. doi: 10.1099/00221287-137-10-2439.

Wessels, J. G. H. (1996) 'Hydrophobins: Proteins that Change the Nature of the Fungal Surface', in *Advances in microbial physiology*. Academic Press, pp. 1–45. doi: 10.1016/s0065-2911(08)60154-x.

Wessels, J. G. H. (2000) 'Hydrophobins, unique fungal proteins', *Mycologist*, 14(4), pp. 153–159.

Wijewardana, N. S. *et al.* (2015) 'Characterization of water repellency for hydrophobized grains with different geometries and sizes', *Environmental Earth Sciences*. Springer Berlin Heidelberg, 74(7), pp. 5525–5539. doi: 10.1007/s12665-015-4565-6.

Wösten, H. A. B. (2001) 'Hydrophobins: Multipurpose Proteins', *Annual Review of Microbiology*, 55(1), pp. 625–646. doi: 10.1146/annurev.micro.55.1.625.

Zheng, S. *et al.* (2017) 'Hydrologic behavior of model slopes with synthetic water repellent soils', *Journal of Hydrology*. Elsevier B.V., 554, pp. 582–599. doi: 10.1016/j.jhydrol.2017.09.013.

Zheng, S. *et al.* (2019) 'Erodibility of synthetic water repellent granular materials: Adapting the ground to weather extremes', *Science of the Total Environment*, 689, pp. 398–412. doi: 10.1016/j.scitotenv.2019.06.328.

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