

# Effects of AM fungi on root biomechanical properties

## Effets des champignons AM sur les propriétés mécaniques des racines

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**ABSTRACT:** Arbuscular mycorrhizal (AM) fungi play a pivotal role in promoting plant growth by enhancing root biomass, cellulose content, and biomechanical properties of plant roots. The present study investigates the potential of AM fungi in conjunction with Bermuda grass (*Cynodon dactylon* L.) to optimize soil bioengineering methods. These environmentally friendly approaches effectively mitigate soil erosion by facilitating root adhesion to the soil matrix and reinforcing both mechanical and hydraulic properties by root biomechanical properties. The study focuses on assessing the impact of AM fungi on distinct growth and decay periods induced by controlled burning and herbicide treatments. Various parameters, including root biomass, cellulose content, and mechanical properties such as root tensile strength and root young's modulus, are measured at different temporal intervals. The findings elucidate that the presence of AM fungi significantly augments plant root biomass and root tensile strength relative to the absence of AM fungi. However, during the decay period, the influence of AM fungi on root tensile strength can increase, but it is not statistically significant. Nonetheless, these properties are notably higher in the presence of AM fungi compared to their absence, suggesting that AM fungi enhance these traits predominantly during the growth phase. The outcomes of this study underscore the profound positive effects of AM fungi on plant root properties and provide valuable insights into their potential implementation in soil bioengineering.

**RÉSUMÉ:** Les champignons mycorrhiziens arbusculaires (AM) jouent un rôle central dans la promotion de la croissance des plantes en améliorant la biomasse des racines, la teneur en cellulose et les propriétés biomécaniques des racines des plantes. La présente étude examine le potentiel des champignons AM en conjonction avec l'herbe des Bermudes (*Cynodon dactylon* L.) pour optimiser les méthodes de bio-ingénierie des sols. Ces approches respectueuses de l'environnement atténuent efficacement l'érosion des sols en facilitant l'adhésion des racines à la matrice du sol et en renforçant les propriétés mécaniques et hydrauliques grâce aux propriétés biomécaniques des racines. L'étude se concentre sur l'évaluation de l'impact des champignons AM sur des périodes distinctes de croissance et de décomposition induites par des traitements de brûlage contrôlé et d'herbicides. Divers paramètres, notamment la biomasse racinaire, la teneur en cellulose et les propriétés mécaniques telles que la résistance à la traction des racines et le module de jeunesse des racines, sont mesurés à différents intervalles temporels. Les résultats montrent que la présence de champignons AM augmente considérablement la biomasse des racines des plantes et leur résistance à la traction par rapport à l'absence de champignons AM. Cependant, pendant la période de décomposition, l'influence des champignons AM sur la résistance à la traction des racines peut augmenter, mais elle n'est pas statistiquement significative. Néanmoins, ces propriétés sont nettement plus élevées en présence de champignons AM qu'en absence, ce qui suggère que les champignons AM améliorent ces caractéristiques principalement pendant la phase de croissance. Les résultats de cette étude soulignent les profonds effets positifs des champignons AM sur les propriétés des racines des plantes et fournissent des informations précieuses sur leur mise en œuvre potentielle dans la bio-ingénierie des sols.

**Keywords:** AM fungi; root biomechanics; growth; decay.

## 1 INTRODUCTION

In vegetated soil, Plant roots play a pivotal role in conferring stability to shallow slopes and preventing

erosion has been underscored in previous studies (Kamchoom and Leung 2018; Ng et al. 2016). A comprehensive understanding of the biomechanical

properties of plant roots is deemed imperative for devising strategies to mitigate soil instability, particularly in regions prone to natural disasters such as landslides and erosion (Apriyono et al. 2022; Stokes et al. 2014). Consistent with this perspective, prior investigations have delved into the biomechanical properties of plant roots, aiming to elucidate their role in mitigating erosion and failures of shallow slopes. However, divergent findings have surfaced in studies exploring the period of plant root decay (Kamchoom et al. 2021; Zhu et al. 2020). It has been observed that during this phase, these biomechanical properties tend to exhibit diminished values, potentially contributing to a detrimental impact on soil stability.

During the process of plant root decay, a symbiotic association with mycorrhizal fungi can exert a significant influence, eliciting positive effects on plant roots (Basyal and Emery 2021). Specifically, Arbuscular Mycorrhizal (AM) fungi facilitate enhanced nutrient absorption by plant roots. Previous studies have indicated that AM fungi may contribute to an increase in cellulose content and even elevate the tensile strength of plant roots. It is important to note that the influence of AM fungi extends to modifications in soil properties (Chen et al. 2019; Solly et al. 2014). These alterations in soil characteristics, in turn, have implications for changes in the properties of plant roots. Nonetheless, the effect of AM fungi subsequent to decay treatment on root biomechanical properties have not been thoroughly investigated.

## 2 MATERIALS AND METHODS

An investigation was conducted on the roots of *Cynodon dactylon*, commonly known as Bermuda grass, an herbaceous species. Bermuda grass is frequently employed in soil erosion control due to its ability to develop fibrous root systems in shallow soils (Faucette et al. 2006; Ng et al. 2014). The soil selected for this study is lateritic soil, a type commonly encountered in engineering projects for landscaping purposes in tropical and subtropical regions, such as southern China and Thailand. This soil has been characterized as clayey sand. To ensure its suitability for the research, the collected soil underwent an initial sanitation process at 120 °C for 3 hours, followed by an additional two days of oven-drying at 60°C to inactivate AM fungi in situ. Subsequently, the dry soil was sieved through a 2 mm sieve, and the AM fungi were sourced from a local supplier.

To promote optimal root development, Bermuda grass seeds were germinated in nutrient-rich vermicompost one month prior to transplantation into

the soil specimens. AM fungi inoculation was introduced by preparing a mixture comprising 1 kg of soil and 25 g of dry AM fungi inoculum. The soil-AM fungal inoculum underwent compaction within polyvinyl chloride cylinders (90 mm in diameter, 130 mm in depth) using the moist tamping method, resulting in a dry density of 1520 kg/m<sup>3</sup>. Following the compaction of the soil column, a 15 mm thick layer of vermicompost was applied to the upper surface.

To investigate dynamic changes in root, two distinct experimental groups were established: Soil with AM fungi (denoted as M) and Soil without AM fungi (denoted as NM). Each experimental group comprised 30 samples, meticulously maintained in a controlled glasshouse environment. Within each group, 9 samples were specifically selected for the detailed examination of root growth impact. The grass underwent cultivation for durations of 60, 120, and 180 days, collectively referred to as growth treatments. (For each treatment, a standard reference and two replicates were employed). Following 180 days of grass growth, the remaining 21 specimens underwent two distinct decay treatments: herbicide treatment and burning treatment. In the burning treatment, the entire aboveground biomass, encompassing shoots and leaves, was subjected to a 15-minute incineration using a gas torch, followed by a 3-hour smouldering period to facilitate root decay. 12 specimens were retained in the glasshouse for durations of 30, 60, 120, 180, and 360 days after burning treatment. For the herbicide treatment, two doses of Propanil (N-[3,4-dichlorophenyl] propanamide) were applied as an irrigation solution with a concentration of 36% w/v. Subsequently, 9 specimens underwent detailed assessments of root decay after intervals of 15, 30, and 60 days following the herbicide treatment.

### 2.1 Measurement of Root biomechanical properties

Each soil specimen containing roots was rinsed gently with running tap water and was subsequently sieved through sieves No.4 and No.10 to obtain root samples. The roots retained on the sieve were carefully collected. The saturated weight of the roots was determined by immersing them in distilled water for a period exceeding 12 hours. For tensile testing in each treatment, approximately 30 roots were chosen, ensuring the selection of root segments without any tortuosity or tissue loss. These root segments were then subjected to a uniaxial tensile test using a universal testing frame, employing a constant extension rate of 0.1 mm/s. Root segments that failed at or near the clamp margins were excluded from analysis. The root young's modulus was subsequently calculated from

stress-strain relationships derived from the tensile strength test.

## 2.2 Measurement of Cellulose content

The chemical composition of roots, specifically the content of lignin and cellulose, was assessed on the residual portion of each root segment using the method described by Leavitt and Danzer (1993). In general, the procedure entailed the removal of lignin polymers subsequent to the elimination of organic compounds from the root material. This process resulted in the classification of the residual constituent as a compound composed of cellulose.

## 3 RESULTS

### 3.1 Effects of AM fungi on root biomechanical properties

During the phase of normal growth, observed in the NM condition (Soil without AM fungi), there was an 18% increase in mean tensile strength from day 60 to 120. In the condition of M (Soil with AM fungi), a modest increase of 4% was noted from day 60 to 120, followed by a more substantial increase of 23% from day 120 to 180 (Figure 1a). Young's modulus demonstrated minimal impact from AM fungal inoculation at 60 and 120 days but exhibited a notable improvement of 35% at 180 days compared to the NM condition (Figure 1b).

During the decay phase, a significant decrease in mean tensile strength was observed, particularly in condition of decay due to herbicide treatment. At 60 days after decay (day 240), the mean tensile strength for NM and M was 14.9 MPa and 19.5 MPa, respectively, for herbicide treatment, and 27.6 MPa and 28.8 MPa, respectively, for burning treatment. After 180 days of burning-induced decay (day 360), AM fungi conditions displayed a clear increase in mean tensile strength, with values of 16.6 MPa for NM and 23.9 MPa for M (Figure 1a), representing a 44% increase. Regarding Young's modulus, after 60 days of decay due to herbicides (day 240), values for NM and M were 146 MPa and 160 MPa, respectively, while for decay due to burning, the values were 252 MPa and 277 MPa, respectively. A comparison of these values indicated that M condition was approximately 11-18% higher than NM condition under both decay phase.

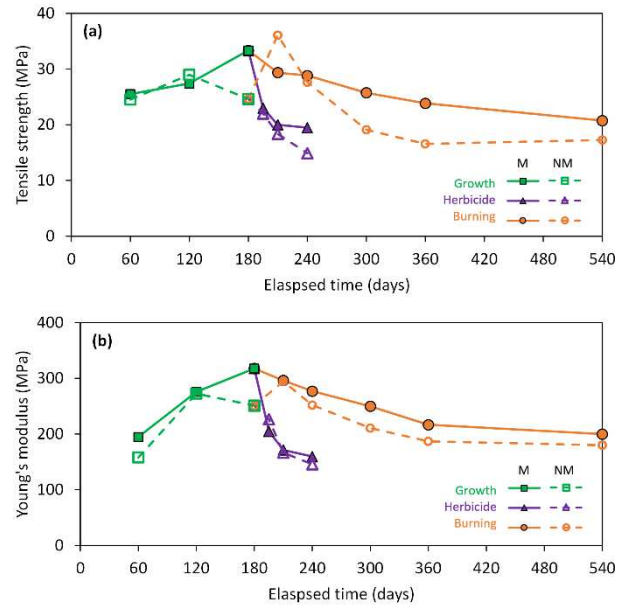


Figure 1. Variation in (a) Tensile strength and (b) Young's modulus of root with and without AM fungi.

### 3.2 Effects of AM fungi on root biomass and cellulose content

In the M condition, both cellulose content and root biomass exhibited higher values compared to the NM condition at each recorded time point. Throughout the growth period, cellulose content demonstrated an increasing trend. However, between days 120 and 180, a slight decrease was observed in the NM condition (Figure 2a), corresponding with the concurrent reduction in tensile strength and Young's modulus during that phase (Figure 1). During the decay phase, a decline in cellulose content and root biomass was noted, with a more pronounced reduction in the condition of herbicide-induced decay. In the presence of mycorrhizal (M), this decline in cellulose content and root biomass was mitigated, particularly in instances of decay due to herbicide (Figure 2c and d). Notably, in condition of burning treatment, although cellulose content decreased, the rate of decline was slower (Figure 2e). Root biomass also experienced a decrease, but the reduction was less pronounced (Figure 2f). The presence of mycorrhizal further attenuated the rate of decline, particularly when compared to herbicide-induced decay.

## 4 CONCLUSIONS

This study involved the inoculation of Arbuscular Mycorrhizal (AM) fungi to Bermuda grass, a species recognized for its resilience in revegetation stress. The primary aim was to investigate the influence of AM fungi on root biomechanical properties relevant to soil reinforcement. Preceding decay processes, AM fungi

contributed to an increase in root biomass, providing more initial root materials. In general, roots inoculated with AM fungi exhibited higher cellulose contents compared to without AM fungi. Although a positive correlation between cellulose content and root tensile strength was evident. AM fungi contributed to roots consistently demonstrated significantly higher cellulose content, tensile strength, and Young's modulus than without AM fungi. These observations were particularly pronounced during normal growth and following the burning treatment, except for the herbicide treatment, indicating that herbicide application influenced plant biochemical or physiological processes, accelerating decomposition. Furthermore, certain plant specimens managed to withstand burning while retaining intact root systems. While no significant difference in the in-situ decay rate of roots was observed between with and without AM fungi, the application of AM fungi resulted in noteworthy enhancements in root biomass, cellulose content, tensile strength, and Young's modulus. Our findings suggest that inoculating grass-rooted soils with AM fungi is an effective method for preserving their physical stability.

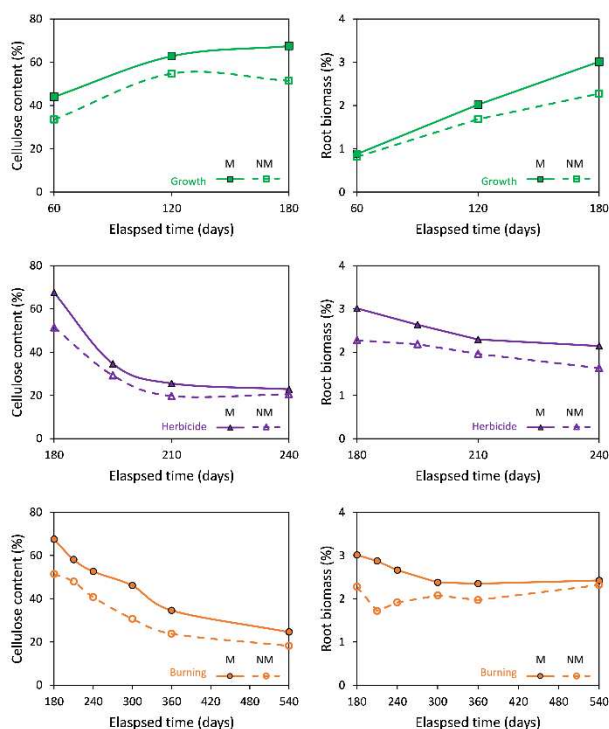


Figure 2. Variation in (a,c,e) Cellulose content and (b,d,f) Root biomass of root with and without AM fungi.

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