African Journal of Biotechnology Vol. 11(47), pp. 10726-10735, 12 June, 2012
Available online at http://www.academicjournals.org/AJB
DOI:10.5897/AJB11.2111
ISSN 1684-5315 ©2012 Academic Journals

*Full Length Research Paper*

# Inoculation effects of two South African cyanobacteria strains on aggregate stability of a silt loam soil

**Mfundo P. Maqubela[1], Pardon Muchaonyerwa[2]* and Pearson N.S. Mnkeni[1]**

[1]Department of Agronomy, University of Fort Hare, Private Bag X 1314, Alice, 5700, South Africa.
[2]School of Agricultural, Earth and Environmental Sciences, University of KwaZulu-Natal, Private Bag X01, Scottsville 3209, South Africa.

Accepted 7 May, 2012

**Two South African cyanobacteria strains (coded 3g and 7e) of the genus *Nostoc* were evaluated for improvement of the aggregate stability of a silty loam soil with low organic C content and compared with *Nostoc* strain 9v isolated from a Tanzanian soil. The soil was either cropped with maize or non-cropped and inoculated with the three strains in a glasshouse. After 42 days, the aggregate stability based on mean weight diameter (MWD) and fragment size distribution were determined by fast wetting, wet stirring and slow wetting methods. Inoculation of the soil with strains 3g and 7e improved the soil's MWD and increased its proportion of large aggregates, particularly in the cropped soil. The opposite was the case for aggregates in soils inoculated with the reference strain 9v. Strain 3g resulted in greater improvement of MWD estimated by fast wetting, while strains 7e and 9v improved aggregate stability estimated by wet stirring. Improvement of aggregate stability was more related to exocellular polysaccharide (EPS) content than organic C. The results suggest that indigenous strains with high potential for EPS production could improve the soil structural stability of degraded soils in South Africa.**

**Key words:** Aggregate stability, exocellular polysaccharides, indigenous cyanobacteria, mean weight diameter.

## INTRODUCTION

Low aggregate stability of most soils in the Eastern Cape Province is caused by low organic C and high silt content making them susceptible to surface crusting, runoff and erosion during rainfall or irrigation events. Soil management strategies that improve organic matter and aggregate stability could significantly improve the productivity of such soils. Cyanobacteria have been reported to improve aggregate stability through enrichment of soil with organic matter, improvement of biological activity and secretion of exocellular polysaccharides (EPS) (Marshall et al., 1996) and to reduce erosion (Mager and Thomas, 2011). The organic matter and EPS, which dominantly consists of polysaccharides, act as binding agents of soil particles (Tisdall and Oades, 1982; Belnap and Gardner, 1993; Malam Issa et al., 2001). Cyanobacteria have the ability to grow in nearly all environments and to adapt to changes in environmental conditions (Mager and Thomas, 2011).

Inoculation of soil with cyanobacteria is increasingly receiving attention as a potential, simple and low-cost method for restoring the productivity of degraded lands. Improved nitrogen status was observed in some the USA soils (Belnap et al., 2001) and aggregate stability was reported in soils from Niger (Malam Issa et al., 2001), which could also be applicable to South African soils under similar semi-arid conditions. However, there is no literature indicating the long term effects of inoculation of soils with cyanobacteria on soil physical properties. Soil biological crusts with cyanobacteria as the main component, have been reported to be resistant to water and wind erosion (Mager and Thomas, 2011). A laboratory incubation study utilizing cyanobacteria strain

*Corresponding author. E-mail: pmuchaonyerwa@ufh.ac.za. Tel: +27 40 602 2290. Fax: +27 86 628 2813.

**Abbreviations: SEM,** Scanning electron microscopy; **EPS,** exocellular polysaccharides; **FSD,** fragment size distribution; **MWD,** mean weight diameter; **FW,** fast wetting; **WS,** wet stirring; **SW,** slow wetting.

**Table 1.** Exocellular polysaccharide (EPS) production and nitrogen fixation potential of cyanobacteria strains 3g, 7e and 9v.

| Cyanobacteria strain | EPS production | Nitrogen fixation (nmol $C_2H_4$/µg chl/h) |
|---|---|---|
| 3g | +++ | 4.7 |
| 7e | + | 16.1 |
| 9v | +++ | 14.1 |

+ and +++ represent relative abundance of EPS as observed by the India ink method.

9v from Tanzania showed improved stability of aggregates of a soil from Guquka in the Eastern Cape Province, within a few weeks after inoculation (Malam Issa et al., 2007). These results were also confirmed in a subsequent glasshouse study which showed that the strain (9v) improved the aggregate stability of Guquka and Hertzog soils (Maqubela et al., 2009). However, the presence of growing plants minimized the extent of aggregate stability improvement (Maqubela et al., 2009).

The study reported herein investigated the effects of two indigenous *Nostoc* cyanobacteria strains with ability to produce EPS and fix dinitrogen, and coded as 3g and 7e with *Nostoc* strain 9v isolated from Tanzania as a reference strain, on the MWD of a degraded soil in the Eastern Cape Province, South Africa, with and without cropping.

## MATERIALS AND METHODS

### Cyanobacteria and soil used

Two cyanobacteria strains isolated from South African soil and one from a Tanzania using methods described by Rippka et al. (1979) were used in the study, and their characteristics are shown in Table 1. Strains 3g and 7e were isolated from soils in Hertzog and Qunu, respectively in the Eastern Cape, South Africa, while strain 9v (the reference) was isolated from a soil in Mkindo, Morogoro region, Tanzania.

Mass production of selected cyanobacteria strains was done by the batch culturing technique in the laboratory and outdoors. The medium used to grow cyanobacteria was BG $11_0$, which was composed of (g $L^{-1}$): $K_2$ $HPO_4.3H_2O$ (0.04); $MgSO_4.7H_2O$ (0.075); $CaCl_2.2H_2O$ (0.036); $Na_2CO_3$ (0.02); citric acid (0.006); ferric ammonium citrate (0.006); EDTA $Na_2$ (disodium – magnesium salt) (0.001). One litre of BG $11_0$ was mixed with 1 ml of trace metal mix which was composed of (g $L^{-1}$): $H_3BO_3$ (2.86); $MnCl_2.4H_2O$ (1.81); $ZnSO_4.7H_2O$ (0.222); $NaMoO_4.2H_2O$ (0.39); $CuSO_4.5H_2O$ (0.079); $CoCl_2.6H_2O$ (0.0494). Temperatures were maintained between 25 and 30°C by the use of heaters, and light intensity was controlled between 10 to 30 µmol photons $m^{-2}$ $s^{-1}$ by increasing the number of fluorescent tube light bulbs from one up to four as the concentration of cyanobacteria in BG $11_0$ increased. Small samples of selected strains were first cultured in small flasks and then transferred to increasingly bigger containers (50, 100, 250, 1000 and 2000 ml conical flasks, 25 L glass bottles and finally 200 L motorized-paddled outdoor ponds) whenever the cyanobacteria dry matter concentration reached 1 g/L. The dry matter was determined weekly by filtering 25 ml of cyanobacteria through pre-weighed filter paper. The filter paper with cyanobacteria was then dried at 60°C to a constant weight, after which it was weighed to determine its mass with cyanobacteria.

The Hertzog soil was used in this study because it had low aggregate stability and was the source of strain 3g. The soil was collected in bulk from the site, air dried and sieved through an 8 mm mesh sieve and used in the pot study in the glasshouse. It was classified as Typic Haplustalf (Soil Survey Staff, 1975) with a pH of 7.46, and C and N contents of 0.77 and 0.07%, respectively. The pH was determined at a soil: water ratio of 1: 2.5, whereas soil C and N were determined using a LECO C and N analyser (LECO Corporation, 2003).

### Glass house experiment

The experiment was carried out using 36 cm diameter pots with 25 cm depth, filled with 15 kg of soil. It was laid out as a factorial experiment with two cropping levels (cropped and non-cropped) and four inoculation treatments (3g, 7e, 9v (reference strain), and non-inoculated control). Treatments were arranged in a completely randomized block design with five replications. Potassium phosphate (40 mg P/kg and 54 mg K/kg) was applied to each pot before the seeds were planted to provide both P and K. Five maize seeds were sown in the cropped treatments and thinned to two plants per pot at the two leaf stage. In inoculated treatments, suspensions of cyanobacteria strains were uniformly poured on the surfaces of potted soil at a rate of 6 g $m^{-2}$ (60 kg/ha) (dry matter basis). Watering was done with distilled water as necessary to compensate for water loss due to evapotranspiration. Temperature in the glasshouse was maintained at an average of 25°C during the day and 10°C at night. The glasshouse had sensors that were programmed to switch heaters on when the temperature fell below 10°C, and a wet wall and fan when temperatures were above 25°C.

Soil sampling for aggregate stability measurements was done after harvesting the maize plants at six weeks after the crop was established. The sampling was done by gently scooping the top 0 to 5 mm depth, which is the layer mostly affected by surface applied organic materials and greatly influences infiltration and runoff of water. The soil samples were placed into rigid containers to avoid breakage of the aggregates. The micro-morphological characteristics of the selected samples were investigated with a scanning electron microscope (SEM) model QUANTA 200 Phillips. Exocellular polysaccharides (EPS) / sugars were analysed by extracting 10 g of soil with 30 ml of 0.5 *M* NaOH and then hydrolysing the extract overnight in 5 ml of 12 *M* $H_2SO_4$. Samples were then centrifuged at a relative centrifugal force of 15557 × g for 30 min using the Eppendorf 5810 centrifuge and the concentration of the extracted EPS was determined spectrophotometrically using the phenol-sulphuric acid method (Dubois et al., 1956). A blank which did not contain the phenol was also prepared.

Air dry soil aggregates larger than 3 mm and smaller than 5 mm after sieving were dried at 40°C for 48 h. Aggregate stability measurements were performed in triplicate using 5 g soil aggregates according to the method of Le Bissonnais (1996). Samples were then subjected to fast wetting, slow wetting, or mechanical breakdown treatments according to Attou et al. (1998).

Fast wetting tests forces related to entrapped air within the aggregates and differential swelling of clays (Chenu et al., 2000). It was done by gently immersing soil aggregates in 50 ml distilled water for 10 min. The water was drawn off with a pipette leaving behind the slaked aggregates. Slow wetting tests the stability of aggregates under low moisture conditions such as those subjected to moderate rains. It measures aggregate stability under conditions in which air entrapment and differential swelling are minimized. This was performed by placing aggregates on a filter paper maintained at a matric potential of -0.3 kPa. After 30 min, the residual aggregates were collected. The mechanical breakdown method tests the cohesion of soil aggregates independently of slaking by air entrapment and the effect of differential swelling. Air was removed from the aggregates before energy was applied, by immersing soil aggregates into 50 ml ethanol. After 10 min, the ethanol was drawn by pipette and aggregates were transferred to another flask with 50 ml of deionised water. The flask was filled with 200 ml of distilled water, stoppered and agitated end over end 20 times and left to stand for 30 min to allow coarse particles to settle. Suspended material and excess water was removed with a pipette and the residual aggregates collected.

Following each test method, the residual aggregates were transferred to a 50 µM sieve immersed in ethanol. The aggregates which were retained on the sieve were transferred to evaporation dishes and dried at 40°C for 24 h. The fragment size distribution (FSD) was measured by dry-sieving the aggregates with a set of six sieves of 2, 1, 0.5, 0.2, 0.1 and 0.05 mm in diameter. The weight of aggregates collected on each sieve was determined and expressed as a percentage of the initial sample dry mass. Aggregate stability was described using the resulting fragment size distribution in the seven granulometric classes and the mean weight diameter (MWD) calculated as follows:

$$MWD = \frac{\sum_{1}^{7} \bar{x}_i w_i}{100}$$

Where, $\bar{x}_i$ is the mean inter-sieve size and $w_i$ is the percentage particles left on each sieve.

#### Statistical analysis

The results of the fragment classes were also grouped into macro (>0.2 mm in diameter) and micro (<0.2 mm in diameter) aggregates. Statistical analysis of data obtained was done using GenStat – Release 4.2, Discovery edition 2. (2005), while means separation was done using least significant differences LSD at $p < 0.05$.

## RESULTS

### Polysaccharide content and micro-morphological characteristics of soils

Soil inoculation with all three cyanobacteria strains increased EPS levels both in cropped and non-cropped soils, except in the cropped soil inoculated with strain 7e, which had EPS levels comparable to the control. In the cropped soils, the EPS content followed the order 3g = 9v >7e = control, whereas it was 9v = 3g > 7e > control in the non-cropped soils (Table 2). Thus, strains 3g and 9v were associated with high EPS contents. Scanning electron microscope observations revealed micro-morphological characteristics of EPS and intertwined cyanobacteria filaments covering surfaces of soil particles inoculated with strain 3g (Figure 1a and c), but no EPS and filaments were observed on the surfaces of non-inoculated soils (Figure 1b and d).

### Aggregate stability

The MWD of aggregates in the non-inoculated control as determined by three test methods were generally higher in the non-cropped than cropped soils (Table 2). The MWD of the non-inoculated and non-cropped soils were 0.44, 0.53 and 1.04 as determined by fast wetting, wet stirring and slow wetting methods, respectively (Table 2). Inoculation with the three cyanobacteria strains significantly increased the MWD of the soil aggregates (Table 2). The MWD were increased by inoculation with all three strains (3g, 7e and 9v) in both cropped and non-cropped soils when all three test methods were used, except that the non-cropped 3g treatment was similar to the controls when the wet stirring method was used (Table 2). In cropped soils, the MWD determined by the fast wetting method were in the order 3g > 9v = 7e, whereas the order was 9v = 7e > 3g in non-cropped soils.

The cropped soil inoculated with strain 3g had a greater proportion of macro-aggregates than micro-aggregates, whereas in soil inoculated with strains 7e and 9v, and the control, micro-aggregates constituted the greater proportion based on the fast wetting test method (Figure 2a). Non-cropped soils followed the opposite trend with smaller proportions of macro-aggregates than micro-aggregates in the 3g inoculated soil, whereas higher proportions of macro-aggregates were observed in the other inoculation treatments. There were greater proportions of larger aggregates (>0.5 mm) and lower proportions of smaller soil aggregates (< 0.2 mm) in the cropped soils inoculated with 3g when compared to the other treatments (Figure 3a), but in the non-cropped soils; inoculation with strains 7e and 9v resulted in a greater proportions of larger aggregates (>0.5mm) (Figure 3b).

When the wet stirring method was used, MWD followed the order 7e > 3g > 9v in cropped soil, and 9v > 7e > 3g in the non-cropped soil. Soils inoculated with all three strains had greater proportions of macro-aggregates than micro-aggregates, whereas proportions were similar in the control irrespective of cropping. The proportion of macro-aggregates was also similar to that of micro-aggregates in the non-cropped 3g inoculated soils (Figure 2b). The proportion of larger aggregates in cropped soil was higher when inoculated with all three strains (3g, 7e and 9v) (Figure 3c), whereas in non-cropped soils, inoculation with strain 9v resulted in a higher proportions of larger aggregates (Figure 3d).

**Table 2.** Cropping and inoculation treatment effects on soil organic carbon, exocellular polysaccharides (EPS) and mean weight diameter (MWD).

| Cropping | Inoculation | EPS (mg/g) | Soil C (%) | $MWD_{FW\#}$ | $MWD_{WS}$ | $MWD_{SW}$ |
|---|---|---|---|---|---|---|
| | | | | (mm) | | |
| Non-cropped | Control | $2.0^{d\S}$ | 0.70 | $0.44^{d}$ | $0.53^{e}$ | $1.04^{c}$ |
| | 3g | $3.9^{a}$ | 0.77 | $0.49^{c}$ | $0.53^{e}$ | $1.11^{b}$ |
| | 7e | $2.4^{c}$ | 0.81 | $0.53^{b}$ | $0.59^{d}$ | $1.11^{b}$ |
| | 9v | $4.0^{a}$ | 0.81 | $0.55^{b}$ | $0.72^{a}$ | $1.24^{a}$ |
| Cropped | Control | $2.0^{d}$ | 0.53 | $0.33^{e}$ | $0.51^{e}$ | $0.80^{d}$ |
| | 3g | $3.2^{b}$ | 0.55 | $0.61^{a}$ | $0.68^{b}$ | $1.06^{c}$ |
| | 7e | $2.3^{cd}$ | 0.57 | $0.54^{b}$ | $0.72^{a}$ | $1.13^{b}$ |
| | 9v | $3.1^{b}$ | 0.53 | $0.53^{b}$ | $0.64^{c}$ | $1.20^{a}$ |
| Probability of > F | | | | | | |
| Source of variation | | | Soil C | $MWD_{FW}$ | $MWD_{WS}$ | $MWD_{SW}$ |
| Inoculation | | | $0.613^{ns}$ | $0.001^{***}$ | 0.001*** | 0.001*** |
| Cropping | | | 0.001*** | $0.770^{ns}$ | 0.001*** | 0.001*** |
| Inoculation * Cropping | | | $0.809^{ns}$ | 0.001*** | 0.001*** | 0.001*** |

[+]Average of MWD as determined by FW, WS, and SW methods. [#]FW stands for fast wetting, WS for wet stirring, and SW for slow wetting tests. [§]Means in each column followed by the same letter are not significantly different at P< 0.05 according the LSD test.

Moreover, when the slow wetting method was used, MWD followed the order 9v > 7e > 3g in cropped and 9v > 7e = 3g in the non-cropped soils. The macro-aggregates constituted greater proportions of the soil in all inoculation treatments than micro-aggregates irrespective of cropping (Figure 2c). There were higher proportions of larger aggregates (>0.5 mm) in the cropped soil inoculated with the three strains (3g, 7e and 9v) (Figure 3e), but in non-cropped soils only strains 3g and 9v resulted in a higher proportions of larger aggregates (>0.5 mm) (Figure 3f). The relationship between EPS and MWD of soils inoculated with the three strains was stronger with fast wetting ($R^2$= 0.659) followed by slow wetting ($R^2$=0.468), but was weak with wet stirring ($R^2$ = 0.284) (Figure 4). Soil C and MWD relationship in soil inoculated with the three cyanobacteria strains was weak for all three aggregate stability test methods (Figure 5).

## DISCUSSION

The aggregate stability of Hertzog soil as determined by fast wetting, wet stirring and slow wetting methods could respectively be classified as very unstable, unstable and partly stable according to criteria suggested by Le Bissonnais (1996). Therefore, Hertzog soil had low aggregate stability which could be attributed to its high fine sand and silt, and low organic C contents. The observed improvement in MWD (aggregate stability) of the soil following inoculation with the three cyanobacteria strains seems to have largely been due to the gluing effect of excreted polysaccharides since the MWD was more strongly associated with EPS than with soil C. Therefore, changes in organic matter content *per se*, due to inoculation, seem to have had limited effect on aggregation. The contrasting effects of the two indigenous strains 3g and 7e on aggregate stability are consistent with their relative EPS production potential, whereby strain 3g was a better producer of EPS than strain 7e. However, in addition to the gluing effects of EPS, observed improvements in aggregation following inoculation with the strains could also have been aided by the hydrophobic properties that EPS impart on soil aggregates. The imparted hydrophobicity retards the release of entrapped air (Kidron et al., 1999) as cited by (Nisha et al., 2007) and thus breakdown of soil aggregates. This is supported by the stronger relationship between EPS and MWD determined using the fast wetting method, which essentially estimates the stability of aggregates against failure due to entrapped air caused by fast wetting. These results are consistent with findings of earlier studies on microbiotic soil crusts (Belnap and Gardner, 1993; Malam Issa et al., 1999, 2001, 2007) or in soils inoculated with cyanobacteria (Malam Issa et al., 2007) which also partly attributed improvements in the aggregation of inoculated soils to increases in EPS that caused changes in the micro morphological characteristics of the aggregates.

Whereas the 3g strain was isolated from the test soil, which was unstable, the observed increase in aggregate stability could be explained by the large amount of the strain through inoculation compared to naturally occurring amounts in the soil. The response therefore suggests that

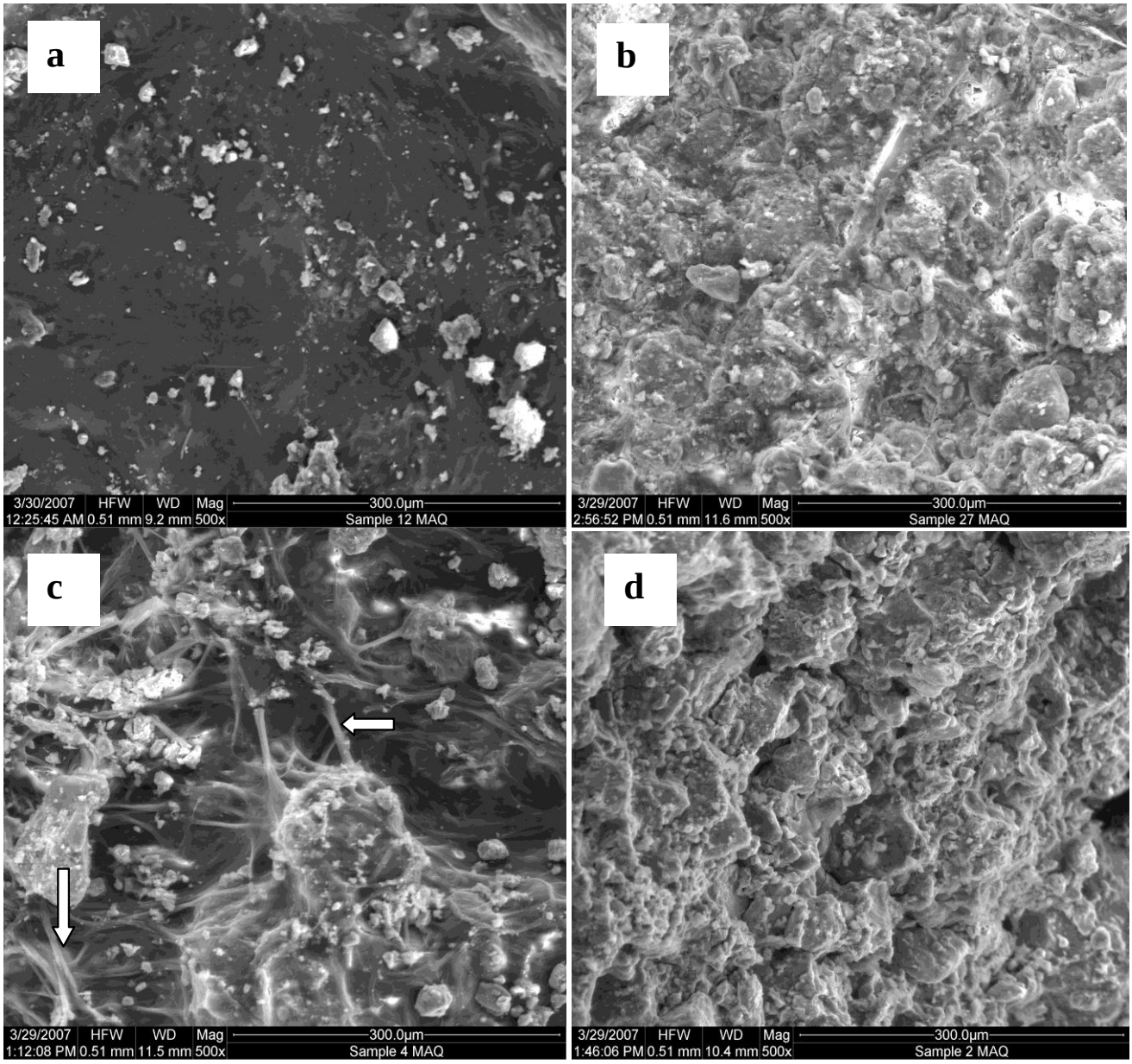


**Figure 1.** (a-b) Surfaces of non-cropped soils: (a) massive surface coating of exocellular polysaccharides (EPS) on soil inoculated with cyanobacteria strain 3g, and (b) soil particles on non-inoculated (control) soil. c-d, Surfaces of cropped soils: (c) filaments (arrows show some) and EPS coating binding soil particles and fragments in soil inoculated with strain 3g, and (d) soil mineral particles with no observed filaments or EPS on the surfaces of non-inoculated (control) soil.

culturing cyanobacteria in ponds to produce large biomass, which will then be applied to the soil surface, could be a viable option to improve the benefits of cyanobacteria on degraded soils in semi-arid environments. However, the feasibility of this approach needs to be tested, taking into consideration the possible development of some toxic species that could harm the plants (Ibelings and Havens, 2008).

Strain 7e significantly improved aggregate stability, as estimated by the three different methods, under cropped and non-cropped conditions far more than could be attributed to its EPS production which was only slightly higher than what was observed in the non inoculated control. Therefore, other mechanisms must have played a greater role in the improved aggregate stability associated with this strain. One possible explanation is its relatively greater impact on soil organic matter both under cropped and non-cropped conditions. However, the enmeshing effects of its filaments in stabilizing aggregates, though not examined in this study, could not be ruled out as their aggregate formation effects were demonstrated in this study for strain 3g (Figure 1c).

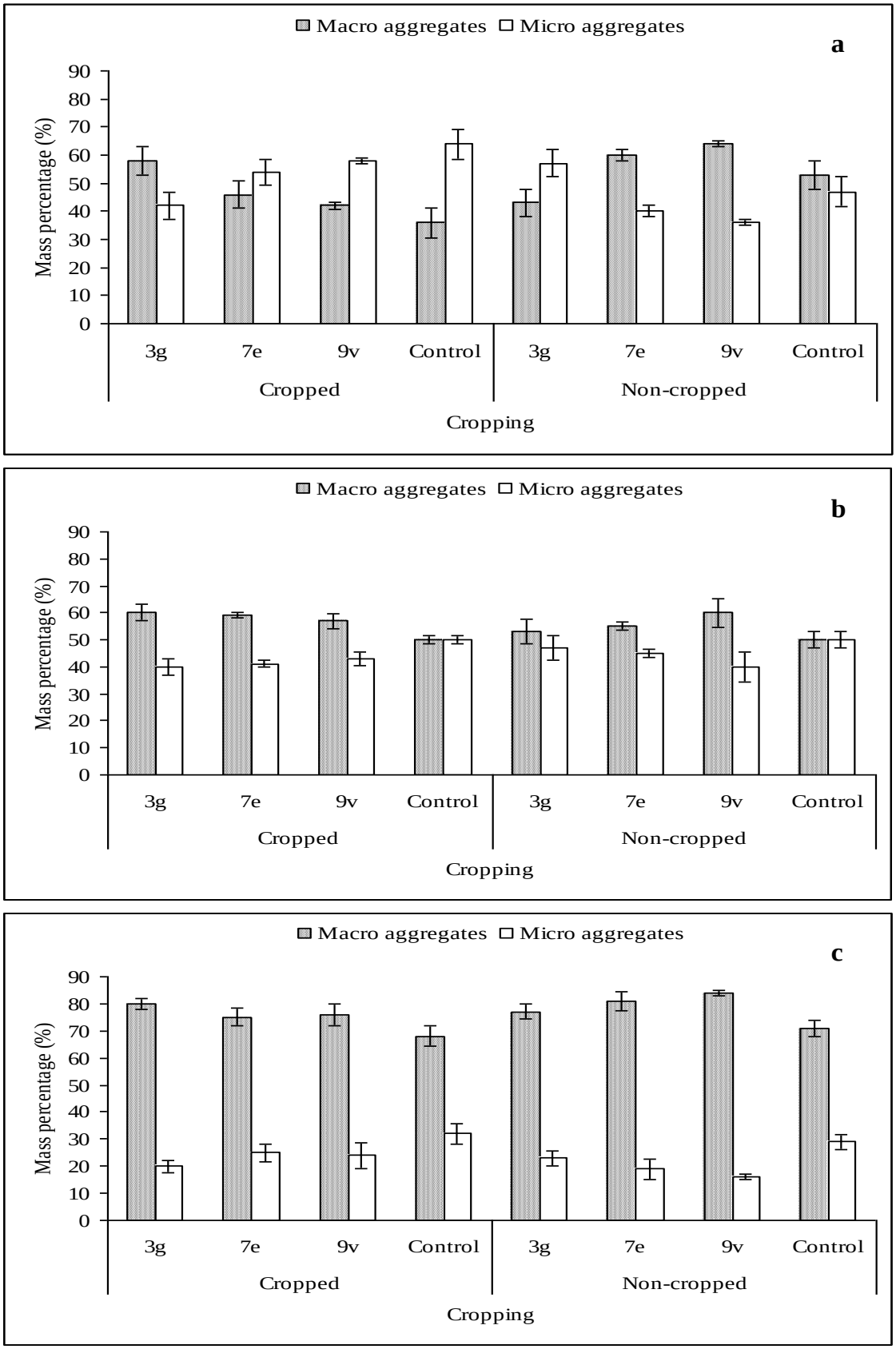


**Figure 2.** Macro aggregates and micro aggregates from cropped and non-cropped soils treated with different strains of cyanobacteria as determined by (a) fast wetting, (b) wet stirring and (c) slow wetting test methods.

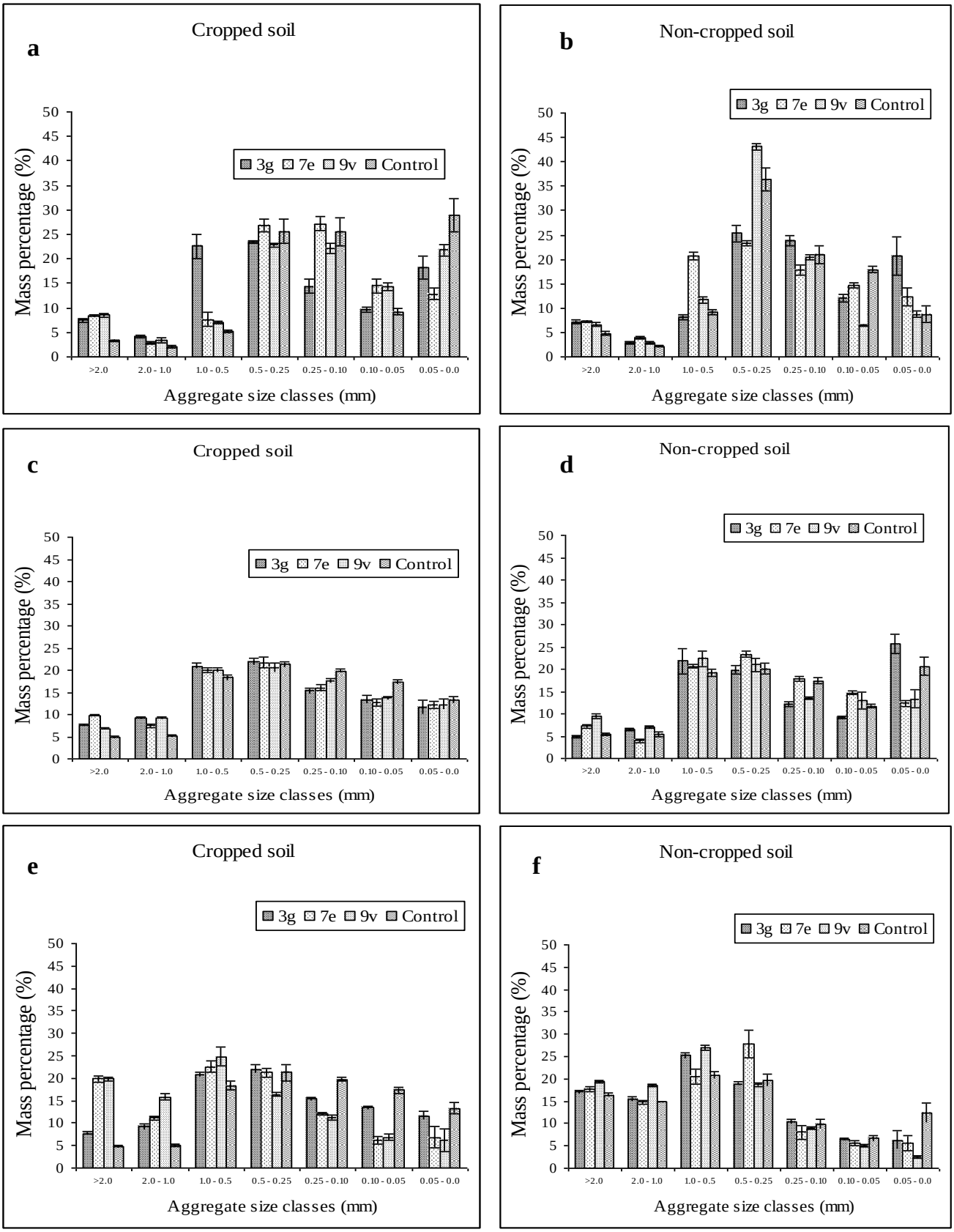


**Figure 3.** Fragment size distribution (FSD) of cropped and non-cropped Hertzog soil inoculated with three strains of cyanobacteria as determined by fast wetting (a and b), wet stirring (c and d) and slow wetting methods (e and f).

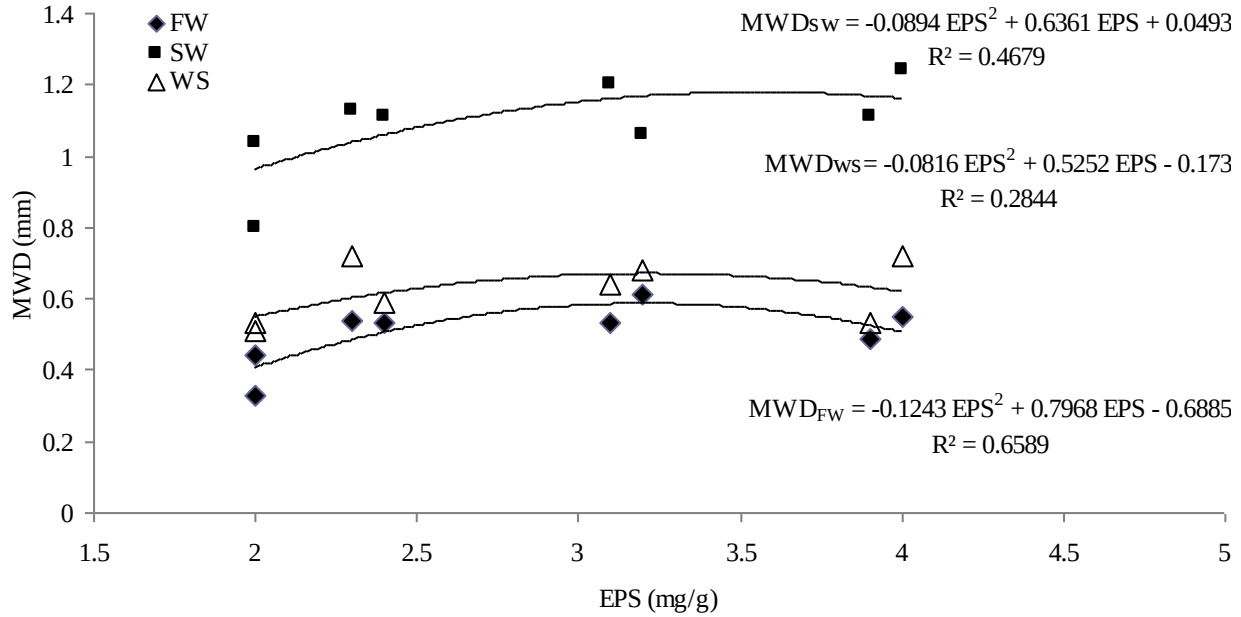


**Figure 4.** Relationship between EPS and mean weight diameter of aggregates from soil inoculated with strains 3g, 7e and 9v. FS, SW and WS represent fast wetting, slow wetting and wet stirring methods, respectively.

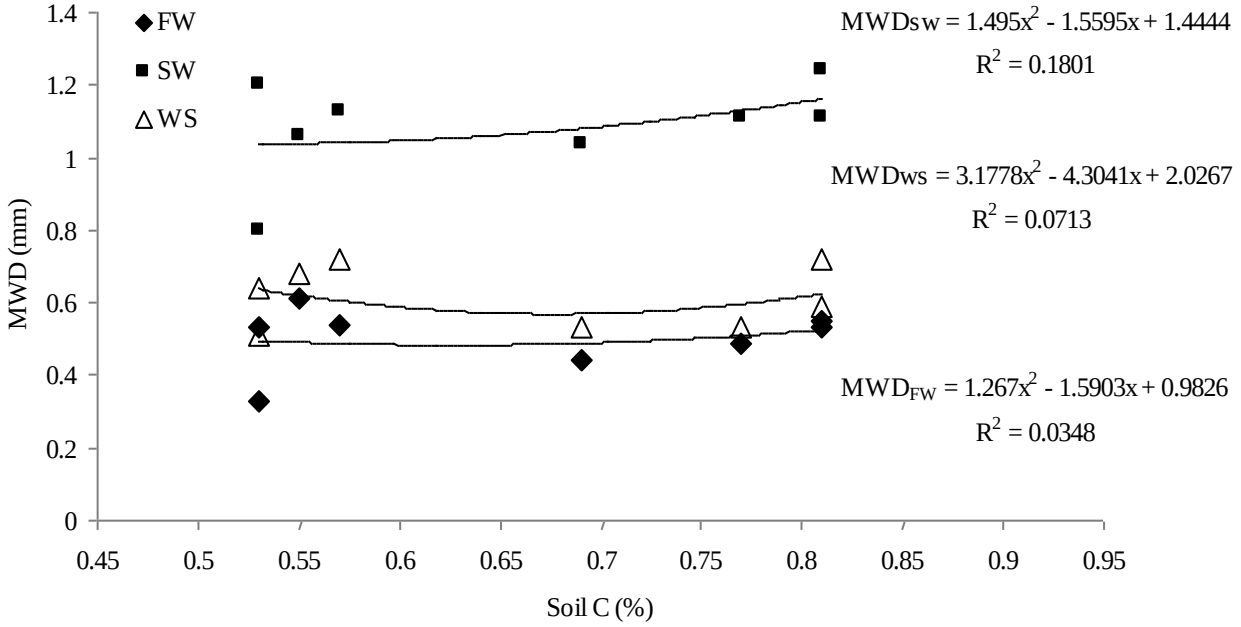


**Figure 5.** Relationship between soil C and mean weight diameter (MWD) of aggregates from soil inoculated with strains 3g, 7e and 9v. FS, SW and WS represent fast wetting, slow wetting and wet stirring methods, respectively. Soil C is represented by the letter 'x' in the formula.

Maqubela et al. (2009) also showed that enmeshing of soil particles by strain 9v cyanobacterial filaments contributed to aggregate stabilization. The magnitude of this effect was unexpected but interesting as the strain 7e was used because of its higher biofertilization than bio-conditioning potential (Table 1), yet it had a profound impact on soil aggregate stability.

Greater improvement in aggregate stability estimated by the fast wetting method in soils inoculated with strain 3g suggest that this strain may be more effective in

reducing breakdown of soil aggregates by raindrop impact. By contrast, greater improvement in aggregate stability estimated by the wet stirring method in soils inoculated with strains 7e and the reference strain 9v indicate that these strains may be more effective in reducing aggregate breakdown by mechanisms of mechanical breakdown (like ploughing) and slow wetting (like gentle rain or irrigation). On average, the improvement in aggregate stability was within unstable and medium stable aggregates with MWD of 0.4 to 0.8 and 0.8 to 1.3 mm, respectively and further studies have to be conducted to determine inoculation methods and experiment length that will result in stable or even very stable aggregates with MWD of 1.3 to 2.0 mm and >2 mm respectively. These observations imply that in order to address most stresses that lead to aggregate breakdown, it may be necessary to inoculate soils with multiple cyanobacteria strains. Therefore, more work is needed to ascertain the complementary effects of strains 3g and 7e, and to identify other indigenous cyanobacteria strains that complement each other in improving soil structural stability.

Nisha et al. (2007) showed that the EPS produced through biofertilization with cyanobacteria provided a substrate for the growth and enhanced activity of heterotrophic microflora such as saprophytes and symbiotics (*Rhizobium*), which in turn produce more EPS, further amplifying its effect on soil structural stability. The effects of indigenous strains 3g and 7e on other soil microflora was not investigated in the present study, but it is an important aspect that needs to be addressed in future studies. Generally, cropping with maize reduced the effectiveness of inoculation with the reference strain 9v in improving aggregate stability of soils, whereas the effect of the indigenous strain 3g and to lesser extent strain 7e was enhanced by cropping. The observed trend for strain 9v is consistent with the results obtained by Maqubela et al. (2009), and could partly be attributed to the observed negative effect of cropping on strain 9v establishment and subsequent lower production of EPS. It could also be attributed to other causes such as those highlighted by Reid et al. (1982) who presented evidence suggesting that poor aggregate stability in soils cropped with maize could be due to the destruction of organic matter – (Fe or Al) – mineral particle linkages when the Fe and/or Al cations that link organic matter to mineral particles are removed by chelating agents released by maize roots into the soil rhizosphere.

The improved aggregate stability observed under cropping in soils inoculated with the indigenous strains 3g and 7e implies that the effectiveness of the two indigenous strains on aggregate stability improvement may not be affected by cropping with maize, at least. This effect cannot, however, be explained by the organic C or EPS levels as cropping had a negative effect on these parameters. Reid and Goss (1981) reported that roots of maize, tomato and wheat decreased the stability of soil aggregates, while those of perennial ryegrass and lucerne improved aggregate stability. Inoculation soil with cyanobacteria strains 3g and 7e may further enhance aggregate stability of soils on which ryegrass and lucerne are grown. The contrasting effects of cropping on the influence of the reference strain 9v and the two indigenous strains (3g and 7e) on aggregate stability, suggest a need for further studies to investigate the interaction effects of different cyanobacteria strains with different plant species on soil aggregate stability.

## Conclusions

Inoculating Hertzog soil, which had low aggregate stability, with indigenous cyanobacteria strains 3g and 7e improved the soil's MWD and hence aggregate stability and increased its proportion of large aggregates. Moreover, the improvement in aggregate stability was more related to changes in EPS than soil C content. Unlike the reference strain 9v, the two indigenous strains 3g and 7e improved the aggregate stability of the cropped soil to a greater extent than in the non-cropped soil, indicating their compatibility with the maize cropping system. The results of this study indicate that indigenous cyanobacteria strains screened for high EPS production could contribute to the amelioration of the structural stability of physically degraded soils in South Africa.

## REFERENCES

Attou F, Braund A, Le Bissonnais Y (1998). Effect of clay content and silt – clay fabric on stability of artificial aggregates. Eur. J. Soil Sci., 49: 569-577.

Belnap J, Gardner S (1993). Soil microstructure in soils of the Colorado plateau: The role of the cyanobacterium *Microcoleus vaginatus*. Great Basin Nat., 53: 40-47.

Belnap J, Kaltenecker JH, Rosentreter R, Williams J, Leonard S, Eldridge D (2001). Biological Soil crusts: Ecology and Management. Technical Reference 1730-2. U.S. Department of the Interior, Colorado, pp. 29-40.

Chenu C, Le Bissonnais Y, Arrouays D (2000). Organic matter influence on clay wettability and soil aggregate stability. Soil Sci. Soc. Am. J., 64(4): 1479-1486

Dubois M, Gilles KA, Hamilton JK, Rebers PA, Smith F (1956). Colorimetric methods for determination of sugars and related substances. Am. Chem. Soc., 23: 351-359.

Genstat Release 4.24DE (2005). Lawes Agricultural Trust. VSN International, Hemel Hempstead, Hertfordshire, UK.

Ibelings BW, Havens KE (2008). Cyanobacterial toxins: a qualitative meta-analysis of concentrations, dosage and effects in freshwater, estuarine and marine biota. Adv. Exp. Med. Biol., 619: 675-732.

Le Bissonnais Y (1996) Aggregate stability and assessment of soil crustability and erodibility: I. Theory and methodology. Eur. J. Soil Sci., 47: 425-437.

LECO CORPORATION (2003). TRUSPEC C/N Determinator. Instruction Manual. LECO Corporation, 3000 Lakeview Avenue. St Joseph, MI49085, USA.

Mager DM, Thomas AD (2011). Extracellular polysaccharides from cyanobacterial soil crusts: A review of their role in dryland soil processes. J. Arid Environ., 75: 91-97.

Malam Issa O, Défarge C, Le Bissonnais Y, Marin B, Duval O, Bruand A, D'Acqui LP, Nordenberg S, Annerman M (2007). Effects of the

inoculation of cyanobacteria on the microstructure and the structural stability of a tropical soil. Plant Soil, 290: 209-219.
Malam Issa O, Le Bissonnais Y, Défarge C, Trichet J (2001). Role of a cyanobacterial cover on structural stability of sandy soils in the Sahalian part of western Niger. Geo. 101: 15-30.
Malam Issa O, Trichet J, Défarge C, Couté A, Valentin C (1999). Morphology and microstructure of microbiotic soil crust on a tiger bush sequence (Niger, Sahel). Catena, 37: 175-196.
Maqubela MP, Mnkeni PNS, Malam Issa O, Pardo MT, D'Acqui LP (2009). *Nostoc* cyanobacterial inoculation in South African soils enhances soil structure, fertility, and maize growth. Plant Soil, 315: 79-92.
Marshall TJ, Holmes JW, Rose CW (1996). Soil Physics. 3rd ed. Cambridge University Press. U.S.A: pp. 199-228.
Nisha R, Kaushik A, Kaushi CP (2007). Effect of indigenous cyanobacterial application on structural stability and productivity of an organically poor semi-arid soil. Geo. 138: 49-56.
Reid JB, Goss MJ, Robertson PD (1982). Relationship between the decreases in soil stability affected by the growth of maize roots and changes in organically bound iron and aluminium. J. Soil Sci., 33: 397-410.
Rippka R, Derueles J, Waterbury JB, Herdman M, Stainer RY (1979). Generic assignments, strain histories and properties of pure cultures of cyanobacteria**.** J. Gen. Microb. 111: 1.
Soil Survey Staff (1975). Soil Taxonomy – a basic system of soil classification for making and interpreting soil surveys. U.S. Dept. Agric. Washington, Handbook No. 436: U.S. Govt. Printing Office.
Tisdall JM, Oades JM (1982). Organic matter and water – stable aggregates in soils. J. Soil Sci., 33: 141-163.