Microbial polysaccharides as carbohydrate sinks in sugarcane

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Plant metabolism can produce an enormous variety of molecules and storage polymers. Most research on complex carbohydrates for sustainable energy and material production has focused on starch. Sugarcane, a member of the grass family, is a more efficient source of bioenergy as it uses C4 photosynthesis more effectively to produce sucrose as a storage molecule, which can be directly fermented. Sugarcane is likely to provide the means for producing a wide range of designer storage products for varying end uses, such as biofuels. We aim to increase biomass production in sugarcane by co-producing high-value products, such as biopolymers. Constructs were generated to allow expression of levansucrases from *Lactobacillus sanfranciscensis* TMW 1.392 and *L. reuteri* LTH5448, as well as a reuteransucrase from *L. reuteri* ATCC 55730. Unlike the majority of plant polysaccharides, microbial exopolysaccharides are water soluble. Transgenic lines were therefore screened for the accumulation of water soluble polysaccharides. Our results demonstrated that sugarcane can be manipulated to store foreign carbohydrate by introducing bacterial glycosyltransferase genes.

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Indigenous *Hypocalyptus*, *Podalyria*, *Cyclopia* and *Virgilia* species are nodulated by diverse beta-rhizobia

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Certain Gram-negative bacteria or so-called "rhizobia" are involved in a nitrogen-fixing symbiosis with leguminous plants, which results in the formation of root nodules. Although it was initially thought that this capability is restricted to the members of the Alpha-Proteobacteria (alpha-rhizobia), it is now known that this symbiosis can also be established by bacteria from the Beta-Proteobacteria (beta-rhizobia). For most part, beta-rhizobia are typically found in association with legumes from the subfamily Mimosoideae. There are however, two notable exceptions in southern Africa - the nodulation of *Cyclopia* (tribe Podalyrieae) and *Aspalathus* species (tribe Crotalaria), by species of *Burkholderia*. The aim of this study was to determine the extent to which legumes indigenous to southern Africa are nodulated by members of the beta-rhizobia. For this purpose, we focused on species in the genera *Podalyria*, *Virgilia* and *Cyclopia* that are all members of the tribe Podalyrieae and the genus *Hypocalyptus*, which was originally thought to form part of this tribe. The root-nodule bacteria associated with these plants were isolated and identified using 16S rRNA and *recA* gene sequence analyses. Our results indicated that the majority of the rhizobia examined represented diverse members of the Beta-Proteobacteria genus *Burkholderia*. There were, however, no association among the bacteria in terms of host or geographic origin, in contrast to the situation encountered with *Bradyrhizobium* species. Some of the bacteria appeared to be conspecific with known beta-rhizobia. For example, some of the isolates associated with *Hypocalyptus* and *Cyclopia* were closely related and highly similar to *B. tuberum*. There were also isolate clusters which did not group with any described *Burkholderia* species possibly indicating novel species. Overall, these data therefore suggest that nodulation of legumes by the beta-rhizobia is much more wide spread than initially anticipated.

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Induction of the defence responses and resistance to wheat leaf rust by plant extracts

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The potential of two natural bio-stimulants (*ComCat*® and the seed suspension of *Lupinus albus*; SS) and two extracts with antifungal activity (*Tulbaghia violacea* and *Agapanthus africanus*) to induce defence responses and resistance to wheat leaf rust was investigated. Foliar application of the different plant extracts on susceptible and resistant wheat plants under conditions of infection and non-infection revealed that only the *A. africanus* treatment increased the *in vitro* activities of the apoplastic PR-proteins significantly. Furthermore, RT-PCR indicated that treatment with the *A. africanus* extract is responsible for induction of the *PR* genes. The active compound that act as an elicitor in the mechanism of the defence reaction of wheat against leaf rust infection was isolated and identified as a saponin. From the results obtained in this study it is clear that the *A. africanus* extract have a great potential in controlling phytopathogens in crop. Moreover, the *A. africanus* extract inhibited spore germination confirming the potential of the extract to control wheat leaf rust when applied either correctively or preventatively.

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Primary assessment of *Thecaphora* anther-smut infection of CFR *Oxalis*

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