

Stenotrophomonas maltophilia, A Commensal of Importance to Biotechnology

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Stenotrophomonas maltophilia (Sm) is endowed with immense prowess that can be exploited beneficially in Agriculture, Nutrition, Medicine, Biodegradation, Bioremediation and Phytoremediation. The bacterium possesses multitudinous extracellular proteins and enzymes lined by inherent and acquired mechanisms and/or genes which are primarily responsible for adaptation and survival in its niche. Accessibility to the versatility and synthetic dynasty embedded in the bacterium is however threatened by ease of contamination with toxic product(s) of the same bacterium and the bacterial implication in life threatening multidrug resistant infections promoted by the presence of resistance genes. High level technology and expertise, with collaboration by scientists from all wards of life is advocated to safely harness the biotechnological potentials of the organism at an economically viable magnitude.

KeyWords: *Stenotrophomonas maltophilia*, Biotechnology,
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Stenotrophomonas maltophilia is a common ubiquitous commensal¹ that is readily isolated from water, soil, sewage and regularly on plant or within plant's rhizosphere where they play key roles in biogeochemical cycling of nitrogen, sulphur and other important elements. Though it has been implicated as opportunistic pathogens^{2,3} and as true pathogens⁴⁻⁶ due to their roles as aetiologies of life threatening infections^{7,8}, yet their beneficial roles in their niche vis-a-vis their importance in biotechnological advancement cannot be underestimated⁹⁻¹¹. This article reviews the potentials of *S. maltophilia* in their niche and in biotechnological advancement, the inherent

genes predicating the bacterium's attributes including antibiotic resistance genes and other selective properties.

***Stenotrophomonas maltophilia* (Sm) in an Ecological Niche: Adaptability and Resilience**

Stenotrophomonas maltophilia is a Gram negative commensal bacterium found in myriads of habitats where it occupies a vital niche ranging from terrestrial to aquatic habitats¹² including the irrigation solutions used in hospitals¹³. While attempting to assess the safety of drinking water in the process of treatment until delivery, Newcombe *et al.*¹⁴ detected *Stenotrophomonas maltophilia* among other opportunistic pathogens in International Space Station by qPCR. Some other studies have revealed that the bacterium is notable among the rhizosphere bacterial inhabitants¹⁵⁻¹⁷. Its fitness in this domain is dependent on the environment. A peculiarly related example is the

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resistant mutants of the bacterium that produce the multidrug efflux pump SmeDEF excessively^{18,19} but less competitive in a slime mold infection model. Meanwhile, *Stenotrophomonas maltophilia* generally has immense adaptability to its natural environment, even in the face of unsuitable stress or at conditions below optimal. As already known, plants secrete a diverse class of polyphenolic compounds called flavonoids to ascertain interaction between microorganisms and the plant²⁰. This compounds poses a challenge for less adaptable bacteria as they have been proven to have antimicrobial activities against extended spectrum beta lactamase producers²¹⁻²⁴, thus creating additional stress for rhizospheric bacteria including *Stenotrophomonas maltophilia* to combat. *Stenotrophomonas maltophilia* however withstands this stress and stands among the most successful rhizosphere bacteria.

It has been equally reported that the development of association with plants built up by *Stenotrophomonas maltophilia* encourages their abilities to survive in soils that are deplete of nutrients¹⁵. This association is facilitated by a number of anatomical structures like flagella²⁵ with which the bacteria moves in response to chemo-attraction from the root; pilli, fimbriae and biofilm for adhesion and adaptation against adverse chemicals and ions²⁶. The bacterium obtains nutriment and shelter in this mutual association just as its extracellular proteins (enzymes) expel or exhibit lethal effect e.g. lipases, chitinases, nucleases, elastases and proteases²⁷, exterminating the root borers within the rhizosphere and leave the plants protected. Extracellular polymers produced by *Stenotrophomonas maltophilia* therefore appears to have potentials as possible source of antibiotics, even if it demands the incorporation of halogen moieties to reduce toxicity and enhance potency as does fluorine in fluoroquinolone²⁸.

Worth noting also is the resilience of this Gram negative rod, that resulted in its adaptation in diverse habitats and biomes across the globe²⁹. Botes *et al.*³⁰ reported the survival of *Stenotrophomonas maltophilia* in South African antimony mine which is an environment that had been heavily impacted with high dosage of arsenic occasioned by refining activities. The resilience to withstand the effect of this supposedly adverse

chemical in large concentrations (10 mmol l⁻¹ arsenite and 20 mmol l⁻¹ arsenate) might have informed the authors' reference to the observation as "hyper-resistance to arsenic". This adaptation extends to human and non human animals as a habitat for *Stenotrophomonas maltophilia*. For instance, Bollet *et al.*¹ noted that in France, the frequency of isolating *Stenotrophomonas maltophilia* from clinical samples has been on the increase since 1987, at time of the emergence of imipenem, the parenteral carbapenem with high pharmacokinetic profile. This adaptive attribute is due to its high resistance profile and unique physio-pathological attributes with which it avoids the non specific anatomical barrier of the immune system³¹. Also, *Stenotrophomonas maltophilia* exhibits mono-cultural growth and multiplies widely utilizing the accessible nutrients in the phyllosphere environment. To achieve this, it alters the cuticle of leaf surface to which it attaches e.g. in *Hedera helix* and *Prunus laurocerasus*³² to increase the availability of water and dissolved compounds. *Stenotrophomonas maltophilia* also maintains good ecological relationships with other phyllospheric bacteria. Its effective alteration of plants cuticle is benefited to the epiphytic bacteria in the environment³³. Previous study also revealed that *Stenotrophomonas maltophilia* strain BP1 and *Pseudomonas syringae* TLP2dell jointly exhibit a high level of coexistence with "respect" for each other, despite the seeming preponderance of *Stenotrophomonas maltophilia*³⁴.

Biotechnological Importance of *Stenotrophomonas maltophilia* as agent in biological control

Another dimension to the importance of *Stenotrophomonas maltophilia* is its role in biological control. An example of such role is found in the biological control of *Bipolaris sorokiniana* on Tall Fescue by *Stenotrophomonas maltophilia* Strain C3³⁵. In Zhang-Yuen's research, strain C3 was observed in growth chamber experiment to prevent the germination of conidial on the surfaces of leaves. When compared with non-treated control, noticeable reduction in lesion and infected area by *B. sorokiniana* was observed by Zhang and Yuen³⁵ which was proportional to the dosage used. This has been explained to have been predicated by the production of chitinase which

prevent the conidial germination⁹. The lytic activity affected by the enzymes produced by this bacterium is a notable mechanism the organism explores for biocontrol^{10,36}. Enzyme system that brought about the lytic activities have caught the attention of researchers on biocontrol agents especially those that are active in disrupting fungi cell wall e.g. chitinase⁹. Hence, *Stenotrophomonas maltophilia* is an important rhizosphere bacteria which can be explored for agricultural improvement against fungal infection³⁷.

Attributes of *Stenotrophomonas maltophilia* that can be explored in biocontrol arsenal include ease of colonization of rhizosphere, production of antimicrobial compounds and extracellular proteins (enzymes)^{9,38,39}, some of which have been discussed earlier in this review. Notable examples in this respect are four isolates designated as PD3531, PD3532, PD3533 and PD3534 which suppressed potato brown rot caused by *Ralstonia solanacearum* in Egyptian clay soil³⁷ and the *Stenotrophomonas maltophilia* strain 34S1 that was identified as a biocontrol against the fungus *Magnaporthe poae* which is an agent for patch disease of Kentucky bluegrass (*Poa pratensis*)^{40,41}, though more research input is advocated in this biocontrol activities to further define the roles of the participating agents.

Stenotrophomonas maltophilia in biogeochemical cycling

Stenotrophomonas maltophilia has been implicated in the biogeochemical cycling of vital elements like Nitrogen, Sulphur, Phosphorus and a number of others. Dungan *et al.*⁴² reported the transformation of selenate and selenite by the bacterium; the attribute which made the author suggests the role of this bacterium in entire biogeochemical cycling. Since their research focussed on Agricultural pond sediment containing selenium, their conclusion affirmed the bacterium's importance, not only for nutrient cycling, but also in bioremediation⁴². In the meantime, the roles of the bacterium amidst other bacteria in nitrogen fixation have been observed by Park *et al.*⁴³ in Korea. The study was conducted in high inorganic fertilizer impacted soil within the rhizospheres of rice, maize and wheat, and the bacterium was observed to exhibit appreciable potentials for nitrogen fixation. Other studies have also confirmed *Stenotrophomonas maltophilia* as

a good solubilizers of phosphate and phytate, and as biological fertilizer⁴⁴⁻⁴⁶. Mineralization of phytate in the biosphere with phytases produced by the soil microorganisms including *Stenotrophomonas maltophilia* stands as a formidable process of phosphorus recycling⁴⁶. This attribute is of particular interest to crop breeders and soil scientists as the bacterial solubilisation will not only recycle nutrients but also convert them from various forms (e.g. tricalcium phosphate in phosphate min)⁴⁷ into utilizable form by plants.

The phytases produced by *Stenotrophomonas maltophilia* which are important in phosphate conversion have other numerous biotechnological applications. They reduce phytate contents in animal feed as well as human food and improve phosphorus' availability^{48,49}. The enzymes are peculiarly incorporated into feeds of farm animals including poultry, swine, and fish diets, amino acids, and energy. *Stenotrophomonas maltophilia* ability to produce this enzyme in large deposit can serve as additional benefit similar to the cases of *Bacillus* sp.⁵⁰, *Raoultella* sp.⁵¹, *Citrobacter braakii*⁵².

Stenotrophomonas maltophilia in Biodegradation and Bioremediation

The impacts of *Stenotrophomonas maltophilia* in various forms of degradation processes stand as an indispensable prowess in nature's self cleansing dynamics¹¹. Recalcitrants of various forms with tendency to choke up some low forms of life in various habitats are easily degraded by *Stenotrophomonas maltophilia*. Studies in laboratory scale and their subsequent applications in larger scales showed these remarkable decomposition properties. These attributes have been utilized in many quarters of human endeavour and may be applied in the removal of clogging that obstruct water filtrations in water treatment plants⁵³. Biofilm produced by *Stenotrophomonas maltophilia* has been employed to biodegrade branched anionic surfactants from activated sludge by Farzaneh *et al.*¹ just as the bacterium has equally been recognized as a potent agent in bioremediation. Aromatic industrial emissions like toluene, xylene, benzene and ethylbenzene have been degraded using the *Stenotrophomonas maltophilia* strain T3-c⁵³⁻⁵⁵. *Stenotrophomonas maltophilia* strain M1 degrades methomyl, an oxime carbamates which

though used extensively for the control of insects and nematode, is a toxic xenobiotics that disrupt the balance in the ecosystem killing vital primary consumers⁵⁶. According to Mohammed⁵⁶, this plasmid based degradation exhibited by this organism was discovered through multiphase-coupled mass spectrometry, and this bioremediation potential promises to be of colossal advantage as one or more pesticides often detected in 95% of surface water systems' samples in USA could be treated with *Stenotrophomonas maltophilia*. Guan *et al.*⁵⁷ observed the degradation of aflatoxin B1 by *Stenotrophomonas maltophilia* 35-3 with highest degradation index of 0.84 at slightly alkaline pH (pH of 8). This enzymatic degradation has great industrial application. Also, many studies have reported the degrading potentials of various strains of *Stenotrophomonas maltophilia* with huge successes^{16,58-60}.

In the same vein, this important role in bioremediation encompasses heavy metals removal and phyto-remediation. Vallini *et al.*⁶¹ reported selenium precipitation by *Bacillus mycoides* and *Stenotrophomonas maltophilia*. Antonioli *et al.*⁶² report also confirmed this, as the researchers observed that *Stenotrophomonas maltophilia* strain SeITE02 can detoxify a selenite contaminated environmental matrix aerobically, reducing selenite to selenium. This attribute is also true for other heavy metals. Non-viable cells of *Stenotrophomonas maltophilia* can be utilized with higher effectiveness to remove Cu (II) from aqueous solutions than a viable one⁶³. This makes *Stenotrophomonas maltophilia* a rear bacterium that is beneficial both as viable cell culture as well as its cell-free extracts or non viable cells. The use of non viable cells can stand as a unique dimension with less demand for cell maintenance in the management of wastewater to eliminate or at least, reduce the heavy metals especially discharged copper wastewater in the environment. Also, *Stenotrophomonas maltophilia* have been observed to play active roles in phyto-remediation of crude oil impacted soil. This attribute was closely connected with their nitrogen fixing potentials as all the bacteria isolated from the plants rhizosphere and used for the study were equally phyllospheric nitrogen-fixing (diazotrophic) bacteria, *Stenotrophomonas maltophilia* inclusive⁶⁴.

Genetic basis for the attributes of *Stenotrophomonas maltophilia*

Observed beneficial attributes of *Stenotrophomonas maltophilia* are orchestrated by inherent and acquired repository of genes^{18,41,60}, of which phenotypic expressions are primarily important for the survival of the bacterium in the natural environment. In terms of relatedness of some strains expressing these "wonders", Rocco *et al.*⁶⁵, observed that the chromosomes of *Stenotrophomonas maltophilia* K279a and R551-3 strains bear same GC content (67%), but different in length, i.e. K279a DNA has the length 4,851,126 bp while R551-3 DNA has 4,573,969 bp. This author further reported higher potential gene products in K279a than in R551-3. Meanwhile, the clusters of type I pili genome are distributed in a unique manner throughout the bacterial gene sequence. This may be interpreted as similar colonization strategies by *Stenotrophomonas maltophilia* in plants and animals. *Stenotrophomonas maltophilia* carries a number of biosynthetic genes for lipopolysaccharide and/or exopolysaccharide which include rmlA, rmlC and xanB⁶⁶. The phenotypic expression of the genes in producing lipopolysaccharide, of course is imperative in cell function, cell integrity and bacterial adaptation, bringing about resistance to antibiotics and neutral detergents^{67,68}. Some clusters of these genes (including the antimicrobial resistant genes) might have been transferred from other bacteria, even those belonging to distant species¹⁸. This is possible as many *Stenotrophomonas maltophilia* strains have been found to have identical BOX-PCR patterns with some endophytic isolates and acquire gene for the earlier observed beneficial attributes by horizontal transfer (HGT). A good example is trans-conjugation that has been observed in *Stenotrophomonas maltophilia* and *Enterobacter* sp¹⁷.

With regards to antibiotic resistance genes, Alonso *et al.*¹⁸ showed that a *Stenotrophomonas maltophilia* strain acquired a cluster of genes coding for antibiotic and heavy metal resistance from Gram positive bacteria. This was equally observed by Ojo *et al.*⁶⁹ in *Stenotrophomonas maltophilia* and two other Gram negative rod bacteria. *Stenotrophomonas maltophilia* is equally viewed as a reservoir for disseminating the resistance gene to other Gram

negative rods. For example, Gordon and Wareham⁷⁰ following their observation of considerable diversity within plasmid-borne quinolone resistance gene, Smqnr alleles in *Stenotrophomonas maltophilia*, suggested that the bacterium might be a reservoir for the spread of quinolone resistant factor to the Enterobacteriaceae family. Some schools of thought believe that commensal bacteria including *Stenotrophomonas maltophilia* are reservoir of antibiotic resistance genes^{19,71}, out of which some which are antibiotic producers are “developers” of antibiotic resistance and transfer same to pathogenic species by HGT. The presence of such large antibiotic resistance determinants and/or genes in soil actinomycetes was quoted as evidence to support their assertions^{72,73}.

For quinolone resistance, Hernandez *et al.*⁷⁴ noted that quite a number of “contributors” include protein protecting target sites, enzymes that modify fluoroquinolone and efflux pumps contribute immensely. Zhao and Drlica⁷⁵ and Drlica⁷⁶ showed mutation as the applicable yardstick to determine quinolone resistance measure. Quinolone resistance can be transferable being plasmid-borne. Fear correlation exists between the plasmid-borne quinolone resistance gene, Smqnr allele and quinolones resistance phenotype among the *Stenotrophomonas maltophilia* isolates^{19,77}. Hence, the reservoir for quinolone-resistance genes and the risk of patients’ compliance to antibiotics regimen in inducible antibiotic resistance in non-clinical environments is of immense interest to the clinical epidemiologist.

CONCLUSION

A careful consideration of the adaptation, ‘prowess’ and multitudinous applications of this bacterium reveal the inherent benefits and challenges. The attending challenges like concomitant production of toxic products and growing trend of pathogenicity should be tackled through a multidisciplinary approach. This step becomes imperative due to the active role the organism plays in nitrogen fixation, biodegradation, biological control, bioremediation and high potentials for use as a source of novel enzymatic activities in biotechnology. Also, how widespread the role of the bacterium is with regards to acting

as reservoir of antibiotic resistance genes especially in underdeveloped countries should be of interest and is a subject of on-going investigation in our group.

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