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**UNIVERSITY OF
MPUMALANGA**

Exploring cancer bush-microbial interaction and its implication on sustainable commercialization

by

Thobile Mkhwanazi

Authors: Thobile Mkhwanazi, Zakheleni Dube, Terence Suinyuy, Abby Masenya



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Cancer bush

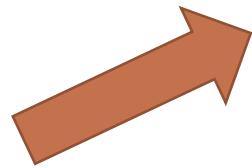
- Cancer bush (*Sutherlandia frutescens*) is a medicinal legume native to Southern Africa (Aboyade et al., 2014).
- Grows in nutrient-poor soils; microbial interactions enhance survival and growth (Fu, 2012).
- Forms symbiotic relationships with rhizosphere microbes (Shengepallu et al., 2018).
- Nutrient acquisition.



Cancer bush - microbe interaction

Nitrogen fixation

Rhizosphere region



Nodules

Roots

Bacterial root nodules on cancer bush

Problem

- Limited research on rhizosphere interaction (Plant-bacteria interaction).
- **PGPR's**

Cancer bush adoption is a Challenge



Then what?

Research

- Role > Microbes



Advantage

Improve cultivation

Commercialization

Promote sustainable agriculture





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Objective

1. To identify and characterize root nodulating bacteria in cancer bush rhizosphere.

Method

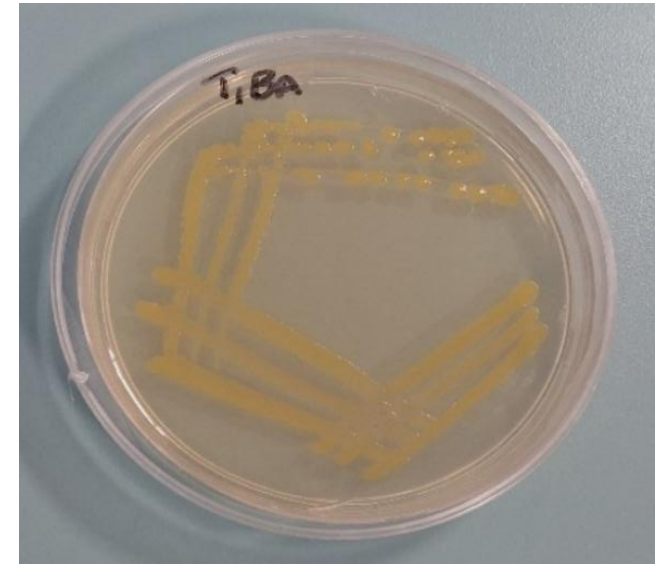
Fig 1: Bacterial isolation



Active nodules



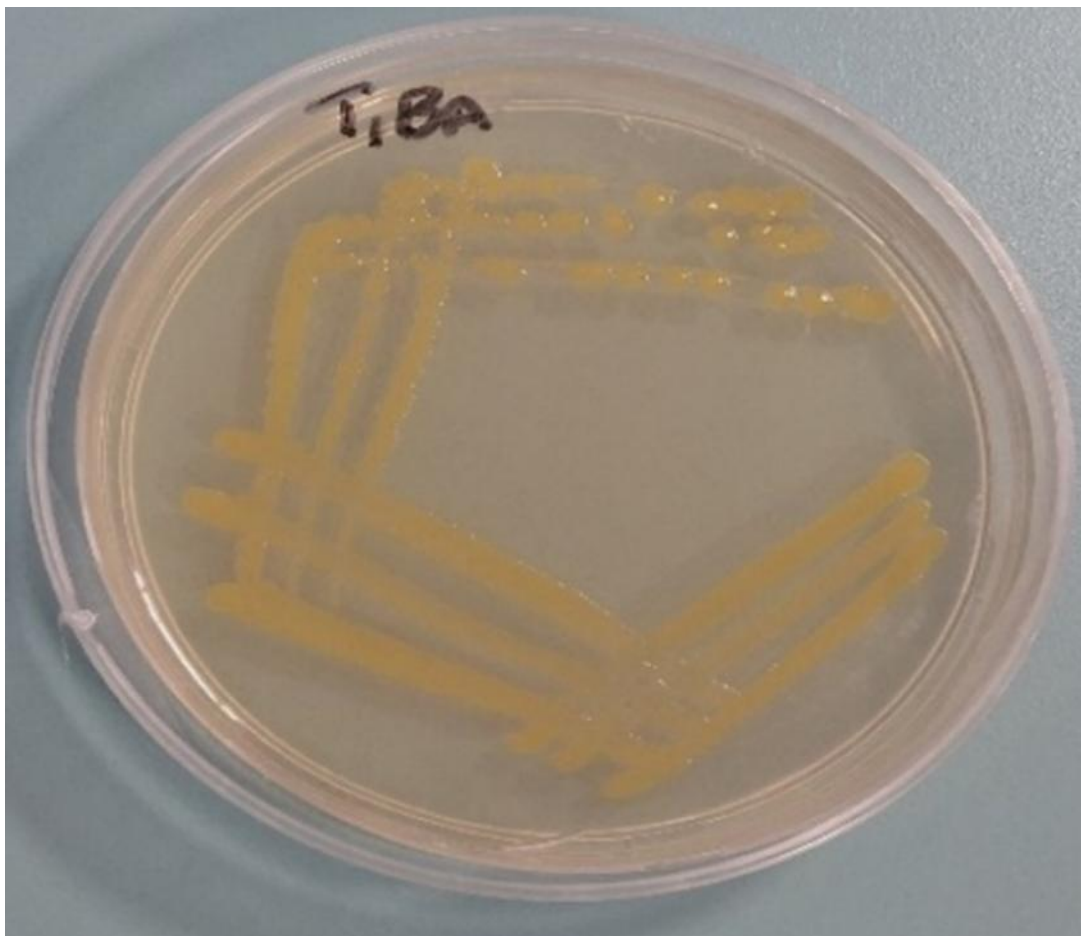
Mixed culture



Pure colony

Method cont'd

2. Morphological identification
 - Colony morphology [shape, color, size, margin & surface]
3. Molecular identification
 - DNA extraction & amplification (Inqaba Biotech Pty. Ltd. South Africa)
 - Target gene: 16s rRNA
 - Primers: 16S -27F and 16S- 1492R
 - Sequence analysis
 - Diversity indices: Shannon-Wiener and Pielou's evenness.



Yellow, Round, Entire & Raised colonies



Red, irregular, Serrate/Filamentous

Fig 2: Morphological analysis.

Molecular analysis

Table 1: Homology of isolates with NCBI GenBank sequences						
Isolate	Nearest BLAST search	Family	Accession number	% Similarities	γ E-value	Function(s)
T2CB	Rhizobium petrolearium	Rhizobiaceae	JX042461.1	84.86	0.000	N-fixing
T1AA	Bacillus licheniformis	Bacillaceae	MN013952.1	81.38	0.000	N-fixing
M4BA	Enterobacter absuriae	Enterobacteriaceae	CP134636.1	83.13	0.000	N-fixing
M7BA	Serratia marcescens	Yersiniaceae	CP055161.1	99.66	0.000	N-fixing
T1BA	Kosakonia cowanii	Proctobacteriaceae	CP035129.1	80.58	2e-143	N-fixing
M4CA	Cellulosimicrobium cullulans	Promicromonosporaceae	OP990691.1	80.77	6e-109	N-fixing
M7CA	Leucobacter chromiiresistens	Microbacteriaceae	MT533900.1	87.37	1e-174	N-fixing
M6CA	Sphingobacterium multivorum	Sphingobacteriaceae	CP068088.1	82.65	2e-153	N-fixing
M8AB	Stenotrophomonas maltophilia	Xanthomonadaceae	OQ940482.1	83.32	0.000	N-fixing
T1CB	Stenotrophomonas geniculate	Xanthomonadaceae	OR117356.1	93.97	0.000	N-fixing
T2BA	Stenotrophomonas pavanii	Xanthomonadaceae	MN030333.1	91.26	0.000	N-fixing
T2CA	Alcaligenes faecalis	Alcaligenaceae	OQ028682.1	85.78	4e-115	N-fixing
M8AA	Stenotrophomonas maltophilia	Xanthomonadaceae	MN09019.1	86.01	0.000	N-fixing
M5B2	Stenotrophomonas maltophilia	Xanthomonadaceae	CP040439.1	89.61	0.000	N-fixing
M3A	Serratia marcescens	Yersiniaceae	CP055161.1	99.66	0.000	N-fixing
T1A	Enterobacter bugandensis	Enterobacteriaceae	CP097255.1	82.51	0.000	N-fixing
T2B1	Micrococcus yunnanensis	Micrococcaceae	KT44390.1	87.60	0.000	N-fixing
T5A2	Bacillus sp.	Bacillaceae	MW272534.1	86.74	0.000	N-fixing
T4B41	Lysinibacillus sphaericus	Caryophanaceae	FJ528593.1	78.33	2e-151	N-fixing

γ E-values = Lower (stronger) E-value (≤ 0) = Significant alignments; Higher (weaker) E-value (> 0) = Alignment might be a random event.

■ Tubatse



□ Makgupheng

Summary of Results: Objective One

Isolates: 30 isolate in winter, 70 isolate in summer; 19 molecularly characterized.

Genera Identified:

- *Cellulosimicrobium*, *Sphingobacterium*, *Rhizobium*, *Bacillus*, *Micrococcus*, *Lysinibacillus*, *Alcaligenes*, *Stenotrophomonas*, *Enterobacter*, *Leucobacter*, *Serratia*, *Kosakonia*.

Dominant Species:

- *Stenotrophomonas* spp. (both sites).
- *Bacillus* (Tubatse), *Serratia* (Makgupheng).

Diversity Indices:

- Shannon-Wiener (H'): Tubatse (2.04 winter; 2.00 summer), Makgupheng (2.40 winter; 1.61 summer).

Functional Traits:

- Nitrogen fixation: Tubatse (94% isolates positive), Makgupheng (90%).

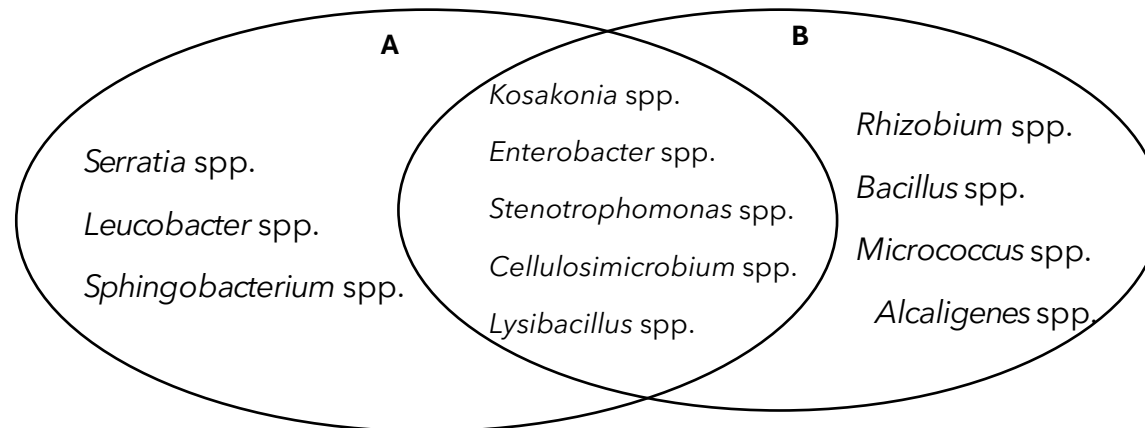


Fig 3: Venn diagram of species found in the two sites and shared species among the Makgupheng (A) and Tubatse (B)



Discussion

- *Rhizobium* sp. in soybean and cowpea (Ndusha, 2011).
- Species such as *Bacillus* sp., *Enterobacter* sp., *Sphingomonas* sp., *Alcaligenes* sp. and *Leucobacter* sp. (Singh *et al.*, 2015).
- *Bacillus* sp., *Alcaligenes* sp., *Enterobacter* sp. isolated from *Ocimum sanctum* (Tulsi) (Singh *et al.*, 2025).
- *Bacillus* sp. identified on *Matricaria chamomilla*, *Calendula officinalis* and *Solanum distichum*.

Discussion cont'd

- Dual nodule occupancy:
 - *Rhizobium* sp. (T2CB) + *Stenotrophomonas* sp. (T2BA) + *Alcaligenes* sp. (T2CA)
 - *Serratia* sp. (M7BA) + *Leucobacter* sp. (M7CA)
 - *Enterobacter* sp. (M4BA) + *Cellulosimicrobium* sp. (M4CA)
 - *Bacillus* sp. (T1AA) + *Kosakonia* sp. (T1BA)
- Dual nodule occupancy in pea (Bourin *et al.*, 2018), soybean (Wongdee *et al.*, 2021; Nguyen *et al.*, 2010).

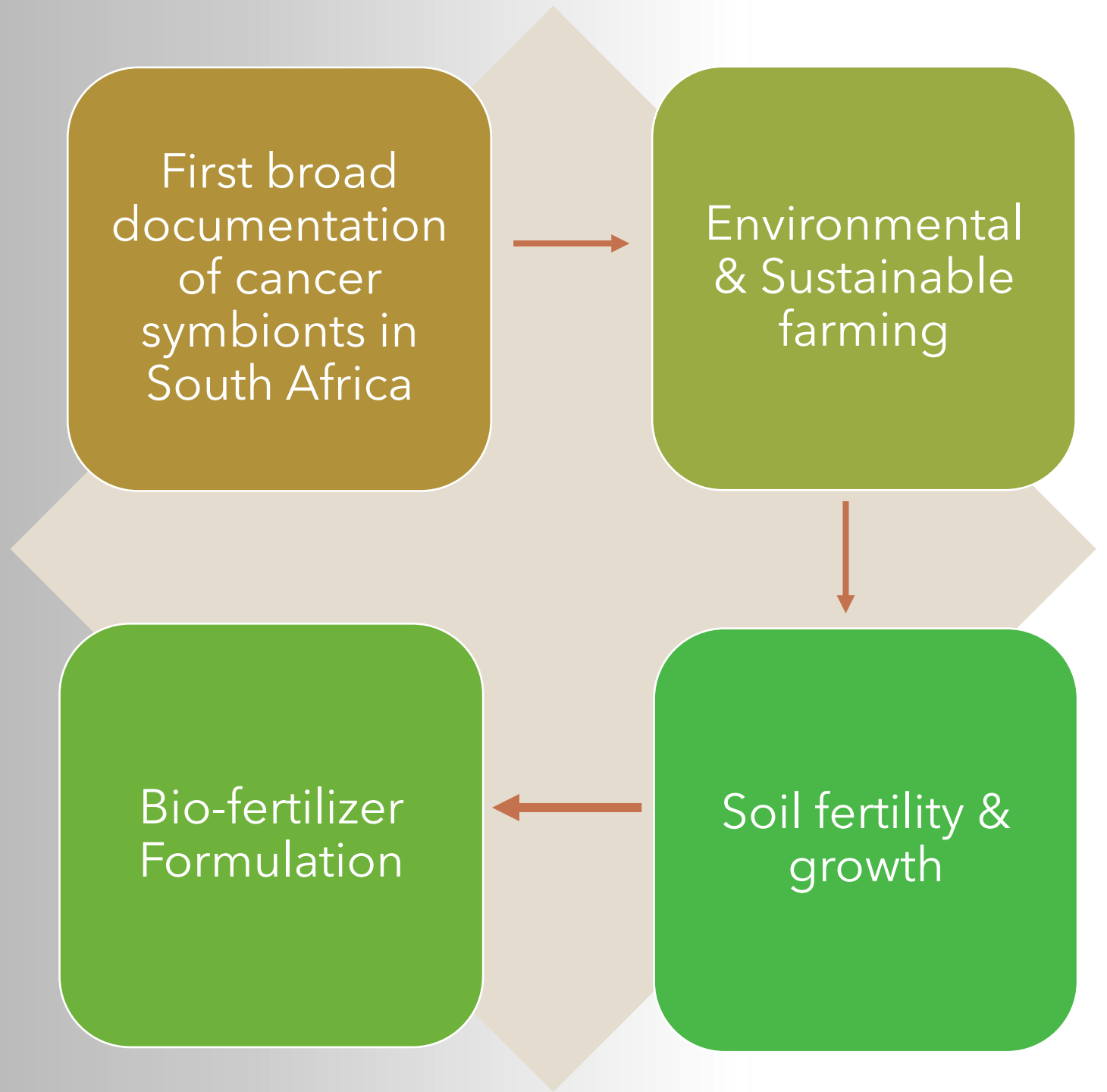
Conclusion

First broad
documentation
of cancer
symbionts in
South Africa

Environmental
& Sustainable
farming

Bio-fertilizer
Formulation

Soil fertility &
growth



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THANK YOU!

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