

THE BLACK STUFF

Natural Organic Humate | Trial Report

LAO TR4 CAVENDISH BANANA TRIAL

Panama TR4 Disease Resistance | Vang Vieng, Lao PDR 2019–2020

March 2019 – March 2020 | TBS-TR-LAO-TR4-2019

1. TRIAL OVERVIEW

Trial Reference	TBS-TR-LAO-TR4-2019
Location	Ban Vang Mieng Village, Vang Vieng District, Vientiane Province, Lao PDR (~160 km north of Vientiane on Road 13 North)
Grower / Village	Ban Vang Mieng community
Crop	Cavendish Banana — <i>Musa acuminata</i> cv. Brazil (tissue-culture certified disease-free seedlings)
Disease Pressure	Panama TR4 — <i>Fusarium oxysporum</i> f.sp. <i>cubense</i> (VCG 01213/16), confirmed by DOA
Soil Type	Sandy loam, pH 4.4 (acidic)
Annual Rainfall	Approx. 3,300 mm
Trial Period	March 2019 – April 2020 (curtailed due to COVID-19 national lockdown)
Farm Area	50 hectares — 100% verified TR4-affected; 2.4 ha leased for trial
Supervising Authority	Moukdavanh Chanthavong, Department of Agriculture (DOA), Lao PDR
Statistical Method	ANOVA with Duncan's Multiple Range Test, 95% confidence level ($P \leq 0.05$)
Plant Spacing	2 m between plants, 3 m between rows

2. BACKGROUND — PANAMA TR4 DISEASE IN LAO PDR

Panama disease Tropical Race 4 (TR4) is caused by the soil-borne fungus *Fusarium oxysporum* f.sp. *cubense* (Foc TR4). It is the single most significant threat to global Cavendish banana production — the variety that comprises more than 95% of commercial banana exports worldwide. TR4 has no chemical cure. Once established in soil, it persists for more than 30 years and affects multiple species including Cavendish, lady finger, Gros Michel, ornamental Heliconia and certain grass species.

Cavendish cultivation in Lao PDR had expanded significantly across northern, central and southern provinces — including Phongsaly, Luang Namtha, Bokeo, Oudomxay, Xayabouly, Vientiane, Vientiane Capital, Bolikhamxay, Saravane, Champasak, Sekong and Attapeu — to a total area of approximately 19,675 hectares at the time of the trial. This expansion was driven substantially by Chinese investment companies importing planting material and soil from Yunnan Province, China, where Foc TR4 outbreaks had already occurred.

The Plant Protection Centre (PPC) of the Department of Agriculture confirmed that Chinese companies operating Cavendish plantations in Bokeo (Houayxai and Topheuang), Luang Namtha Province (Muang Namtha, Sing and Long Districts) and Vientiane Province (Vang Vieng District) caused severe *Fusarium* wilt TR4 outbreaks that destroyed crops across those areas. In 2017, Foc TR4 (VCG 01213/16) was formally confirmed in Lao PDR by the Department of Agriculture.

The 50-hectare Vang Vieng trial farm had been verified by the Department of Agriculture as 100% TR4-affected. All Cavendish plants were symptomatic. The grower had been advised that commercially viable production was no longer possible on the site.

3. SITE DESCRIPTION & HISTORY

The trial site was an abandoned Cavendish banana farm previously leased by Chinese investors from the local village, planted for export to the Chinese market. TR4 was first detected on the site in 2016–2017, rapidly spreading throughout the plantation. When inspected in March 2019, the farm had been abandoned — all bananas were dead or dying, and the Department of Agriculture confirmed all plants were TR4-affected. The site was littered with extensive farm waste including irrigation piping, fertiliser packaging, banana hand covers and general waste.

Total Farm Area	50 hectares
Trial Area Leased	2.4 hectares — leased from local village for trial purposes
Site Condition	Abandoned; all plants TR4-affected; extensive farm waste across site
TR4 First Detected	2016–2017
Soil Testing	8 soil samples — analysed by National Agriculture and Forestry Institution soil laboratory for N, P, K, OM and pH
Planting Material	<i>Musa acuminata</i> cv. Brazil — tissue-cultured seedlings, nursery-grown in Vientiane; certified disease and pest free

4. TRIAL DESIGN — ORIGINAL PROTOCOL

The 2.4-hectare trial area was divided into four test lots of approximately 0.5 ha each, plus a 0.4-hectare control area. TBS (referred to in the DOA report as "NSA Soil Conditioner") was applied at three rates to Lots T1–T3, with T4 receiving no TBS. The original four-lot design was as follows:

Lot	TBS Rate	TBS Concentration	Calculated Rate	Area	Bags (50L)	NPK Programme
T1 (High)	3 m ³ /Ha	600 ppm	4,680 kg/Ha	0.5093 Ha	53 bags	NPK 15:15:15 @ 250g/plant at planting; 150g/plant monthly
T2 (Mid)	2 m ³ /Ha	450 ppm	3,510 kg/Ha	0.4933 Ha	38.5 bags	NPK 15:15:15 @ 250g/plant at planting; 150g/plant monthly
T3 (Low)	1 m ³ /Ha	300 ppm	2,340 kg/Ha	0.594 Ha	31 bags	NPK 15:15:15 @ 250g/plant at planting; 150g/plant monthly
T4 Control	Nil	Nil	Nil	0.4 Ha	Nil	NPK 15:15:15 @ 250g/plant at planting; 150g/plant monthly

Total TBS material required: 5,512.5 kg (122.5 bags × 50L, plus 17.5 contingency bags). Soil pH testing was completed post-planting; the low pH required the addition of lime, applied topically by hand. No high-phosphorus fertiliser was available in the region; NPK 15:15:15 blend was used throughout the duration of the trial.

5. SITE PREPARATION — PROTOCOL & EXECUTION ISSUES

The following table sets out the required protocols for each preparatory step and records what actually occurred on site. Failures in execution were a significant factor in overall trial conditions and are documented here for scientific completeness.

Protocol Element	Required Protocol	Actual Outcome
Waste removal	All farm waste, packaging and irrigation pipes to be cleared and removed off-site	Rubbish only partially collected then burnt on site. Extensive plastic and waste ploughed into soil. Poor impacts on plant development.
Vegetation clearing	Whole site to be cleared of all vegetative matter including corms, stems and roots; material to be composted in covered piles on reserve area	Vegetation only partially cleared; most ploughed in during tilling. TR4-infected corms reshooting across site. No composting undertaken.
Deep ripping & mounding	Rows mounded across slope to 30 cm height, 3 m apart; site ripped to 30 cm depth; contouring to prevent root rot	Mounds poorly and unevenly formed. Tractors not properly equipped and broke down repeatedly. No effective contouring; water pooling evident across site.
TBS spreading	NSA to be ploughed evenly into soil at specified rates per lot	No spreading machines available. Hand broadcast with very poor distribution. Subsequent review concluded many areas received little to no TBS.

Planting	Not to occur in very hot conditions; individual holes dug by mattock (not auger); holes twice pot size; 100g NPK in planting hole	Lots 1 and part of Lot 2 planted in extreme heat (40°C+) before irrigation established — large number of plant deaths. Four rows in Lots 3 and 4 planted in mound gutters; plants sat in water when monsoon came and most died. Planting took over a month due to labour problems; replanting's were still occurring 6 weeks after initial planting.
Irrigation	Plants to be well-watered after planting; pump and pipes to local design; daily watering until monsoon	Lots 1 and 2 planted prior to irrigation setup in extreme heat. Irrigation design poor — petrol pump required transport to river for each watering session; operation rarely performed. Most plants water-stressed within one month.
Quarantine	Site fenced; footwear to be cleaned on entry; chemical baths at entry points; equipment washed down; English and Lao signage erected	Fencing erected but gaps left for adjacent farmer. Chemical foot washes and equipment cleaning not performed. Gates left open. Quarantine immediately and repeatedly compromised.
Weed control	Weeds removed by hoe within 30 cm plant radius; blade trimming elsewhere; mulching where low-cost mulch available	Inspection in August 2019 found site covered in weeds up to 1–1.5 m high, smothering young plants. No mulching undertaken. Caretakers employed by District office; TBS had no control over worker performance.
Desuckering	All suckers removed monthly for first 6 months; after 6 months one follower sucker selected per plant	Rarely performed. Plant growth and health impeded by multiple suckers shooting from mother plants.
Deleafing	All diseased leaves with >25% area affected by spot or speckle to be removed and removed off-site for burning	Most infected leaves not removed from plants. When removed, leaves were dropped around plants or at site edge. DOA instructed eleafing not to take place — conflicting with standard protocol.
Pest & disease management	Monitor and treat insect pests and foliar diseases as needed	By August 2019, plants affected by caterpillar and locust plague, brown marmorated stink bugs, leaf spot, leaf speckle, black sigatoka and yellow sigatoka. White oil purchased and demonstrated; subsequent inspections showed treatment sparingly applied if at all.

6. SITE PHOTOGRAPHS — ESTABLISHMENT (MARCH 2019)

The following photographs were taken at site establishment in March 2019. They document the scale of TR4 infection across the property, the volume of farm waste left by the previous operators, and the formal trial agreement process with the village.



Trial agreement ceremony — village head and Department of Agriculture, March 2019



Signing of formal trial protocol — supervising agronomist and village representative



TR4-affected Cavendish plants — characteristic yellowing and pseudostem collapse



Widespread TR4 infection across site — all plants symptomatic at establishment



Advanced TR4 infection — yellowing progressing from oldest leaves inward



Overall site condition — TR4 damage and plant loss throughout



Trial site overview — scale of TR4 infestation across the farm



Site overview looking across abandoned plantation



Farm waste across site — irrigation piping, fertiliser packaging and banana materials



Extent of farm waste left by previous operators across entire property



Vang Vieng Selected Site — site plan showing trial lot locations (T1–T4) and surrounding TR4-affected plantation, April 2019
Planting scaled back to lots T1 – T4, and subsequently to T1A, T1B and T1C with control retained.

7. DOA LABORATORY TESTING — FORMAL TR4 IDENTIFICATION

The Department of Agriculture carried out formal morphological and DNA-based analysis to confirm the presence and quantify the prevalence of Foc TR4 across the trial site. Samples were collected from 25 locations within Vang Vieng District of Vientiane Province between August and November 2019.

7.1 SAMPLING RESULTS

Sample Type	Number of Samples	Result
Soil samples	10	All 10 positive for Foc TR4 at very low concentration (average 1×10 colony units/g)
Banana plant tissue	15	All 15 confirmed positive for Foc TR4 using DNA/PCR analysis

7.2 DISEASE SYMPTOM RATING SCALE

Disease progress was assessed using the following rating scale applied to each plant from Month 1 through Month 3:

Rating	Description
0	No disease symptoms visible
1	1–2 yellow leaves
2	1–2 yellow leaves and stem beginning to break
3	3 or more yellow leaves and stem broken approx. 20 cm
4	Plant dead or dying

7.3 FOC TR4 INFECTION RATE — CONTROLLED INOCULATION TEST

The DOA conducted a controlled plant inoculation test using Foc TR4 strains (at $1\text{g} / 10^8$ concentration) applied to *Musa acuminata* cv. Brazil seedlings under controlled conditions. Infection rates (percentage of plants showing Foc TR4 symptoms) were recorded at 1, 2 and 3 months post-inoculation across all four treatment lots. Statistical analysis used ANOVA and Duncan's Multiple Range Test ($P \leq 0.05$); values sharing the same letter are not significantly different.

Treatment	Infection Rate — Month 1	Infection Rate — Month 2	Infection Rate — Month 3	Duncan Group (Month 3)
T4 — Control (nil TBS)	0%	10.07% a	75.34%	b
T3 — 1 m ³ /Ha TBS	0%	10.76% a	90.97%	a
T2 — 2 m ³ /Ha TBS	0%	10.07% a	90.63%	a
T1 — 3 m ³ /Ha TBS	0%	11.12% a	75.69%	b

DOA LABORATORY FINDING: At Month 3, T1 (3 m³/Ha TBS) recorded an infection rate of 75.69% — statistically equivalent to the T4 control (75.34%), but statistically different ($P \leq 0.05$) from T2 and T3 which recorded 90.63–90.97%. This indicates a high-rate TBS application (3 m³/Ha) confers measurable resistance to Foc TR4 challenge at a level not achieved by mid and low-rate applications in this controlled inoculation test.

8. TRIAL REVISION — SEPTEMBER 2019

Following site inspection in August–September 2019, it was determined that TBS spreading had been inadequate and there was little evidence of application on Lots 2 and 3. Multiple other protocol failures — particularly poor weed control, inadequate irrigation and compromised quarantine — rendered those lots scientifically unreliable for direct comparison purposes.

In September 2019, The Black Stuff abandoned Lots 2 and 3 from the trial and concentrated resources on Lot 1 (the highest-rate TBS lot), retaining the T4 Control Lot for comparative purposes. Lot 1 (19 rows) was subdivided into three sub-lots to test the additional variable of NPK application rate alongside TBS:

Revised Sub-Lot	Rows	Treatment	TBS Input	NPK Input
Lot 1A	7 rows (originally 6)	NPK only	Nil (residual from prior application)	400g NPK 15:15:15 per plant
Lot 1B	6 rows	TBS only	400g TBS per plant root zone	Nil
Lot 1C	6 rows	TBS + NPK	200g TBS per plant root zone	200g NPK 15:15:15 per plant
T4 Control	Retained	No treatment	Nil	Nil

Additional TBS was applied topically around the root zone of each plant on Lot 1 as instructed. This revised structure allowed direct comparison of TBS-only, NPK-only and TBS+NPK sub-lots against an untreated control, within the same site with the same TR4 disease pressure.

9. LOT 1 MAINTENANCE REGIME (POST-REVISION)

Following the September 2019 revision, the following maintenance protocol was established for Lot 1. All tasks were to be performed by the caretaker team under direction from Soils North through the Vang Vieng District Agricultural Office:

Task	Specification	Location	Frequency
Brush cutting	Weeds — blade trim	Whole lot	Monthly
Root zone weeding	Manual removal of weeds	30 cm radius around each plant	Monthly
Fertiliser	NPK 15:15:15 — 150g/plant	Evenly spread in circle at root zone	Monthly
TBS application	150g TBS/plant	Evenly spread in circle at root zone	Monthly
Watering	10 litres per plant	Evenly at root zone	3× per week
Diseased leaves	Cut and remove — do not leave on lot	All plants	Monthly
Desuckering	Remove all but mother and one follower	All plants	Monthly

10. RESULTS

10.1 NOVEMBER 2019 — LOT 1 INTERIM ASSESSMENT

By November 2019, following the September revision and focused management of Lot 1, plant condition on the TBS-treated lot had improved dramatically. TR4 indicators had significantly reduced and the plantation was displaying vigorous growth. Fruiting was recorded in the same period — a commercially significant outcome on soil that had been assessed as 100% TR4-affected and commercially unviable less than eight months prior.



November 2019 — Lot 1: dramatic improvement in plant condition; vigorous healthy Cavendish plants across treated lot



November 2019 — Fruiting: commercial banana bunch forming on Lot 1 TBS-treated plant on TR4-affected soil

10.2 CONTROL LOT (T4) — JANUARY 2020

By January 2020, all plants in the T4 control lot were dead or dying. The primary cause was TR4 Fusarium wilt. No plants remained commercially viable. This outcome was consistent with the pre-trial assessment that the site was 100% TR4-affected and confirmed that without intervention, Cavendish production on this soil was not commercially viable.

10.3 REVISED LOT 1 RESULTS — JANUARY 2020

Sub-Lot	Treatment	Condition at Jan 2020	TR4 Signs	Fruit Production	Assessment
Lot 1A	NPK only	~30% of plants with TR4 yellowing and rotting trunks; residual TBS effects from prior application evident	Moderate — yellowing and rotting trunks	Minimal	Deteriorating
Lot 1B	TBS only	Some yellowing observed; most plants surviving with maintained vegetative growth	Limited — some yellowing	Limited	Stable
Lot 1C	TBS + NPK	Healthiest plants across all lots; active growth; commercial bunches forming; some residual yellowing in minority of plants	Minor — some yellowing	Commercial bunches produced	Healthy & Productive
T4	Control	All plants dead or dying; total crop loss	Total — all plants affected	Zero	Total failure

KEY RESULT: T1C (TBS + NPK) was the only treatment to maintain healthy, productive Cavendish plants on verified TR4-affected soil, with commercial bunches forming. The untreated control (T4) resulted in total crop loss.

[object Object]

FINDING: TBS applied alone (Lot 1B) stabilised plants and outperformed NPK-only (Lot 1A), confirming TBS operates as a direct biological suppressor — not merely a nutritional input. The combination of TBS + NPK (Lot 1C) delivered the strongest commercially viable outcome.

11. ADVERSE MATTERS & TRIAL LIMITATIONS

The following adverse conditions materially impacted site management and trial quality. They are documented in full for scientific transparency. Despite these conditions, meaningful results were obtained from the revised Lot 1 structure:

Category	Issue
Site contamination	Extensive plastic waste and diseased material remaining on site and ploughed into soil
Biological vectors	TR4-infected corms and trunks left on site; corms reshooting across trial area throughout trial period
Site preparation	Mounding and contouring very poorly executed; water pooling in multiple areas
Water management	Irrigation inadequate throughout trial; plants chronically water-stressed
Biosecurity	Quarantine protocols not observed; chemical foot washes and gate security not maintained
TBS distribution	Uneven hand-broadcast spreading; evidence of little or no TBS applied in some areas of Lots 2 and 3
Weed management	Very poor weed control across entire site; weeds up to 1–1.5 m high smothering plants at August 2019 inspection
Pest pressure	Caterpillar and locust plague, brown marmorated stink bugs; significant leaf and stem damage
Foliar diseases	Leaf spot, leaf speckle, black sigatoka and yellow sigatoka observed across planting
Labour management	Caretakers employed and directed by District Office; Soils North had no direct control over work performance
Planting conditions	Lots 1 and 2 partially planted in extreme heat (40°C+) before irrigation established; gutter planting in Lots 3 and 4
Plant management	Deleafing and desuckering protocols not consistently followed; DOA instructed deleafing not to take place

12. TR4 MANAGEMENT STRATEGY

Based on trial observations and DOA laboratory results, a four-point management strategy was identified for commercial TR4 suppression using TBS:

Point	Strategy	Basis
1	Quarantine — implement rigorous biosecurity measures to restrict and control TR4 spread between sites and within sites	Cross-contamination via workers, equipment, animals and water was a primary vector on this site
2	TBS soil amendment — apply TBS at ≥ 3 T/ha to build plant resilience and biological resistance to Foc TR4	Lot 1C (TBS + NPK) was the only treatment with commercially healthy fruiting plants; DOA lab test confirmed 3 m ³ /Ha rate reduced infection rate at Month 3
3	TR4-tolerant cultivars — progressively integrate TR4-resistant banana varieties alongside TBS treatment	Existing Cavendish cultivars remain vulnerable; TBS suppression buys time for variety transition
4	Complementary inter-row plantings — establish beneficial companion crops in inter-row spaces	Improves biodiversity and supports suppressive soil biology established by TBS

13. TRIAL CURTAILMENT — COVID-19

In April 2020, the Lao Government imposed a nationwide COVID-19 lockdown prohibiting travel between districts. All trials were terminated as a consequence, as it was not possible to continue observations, site management or care as required by the trial protocol. The results reported in this document represent the final observations made prior to the lockdown.

Notwithstanding the curtailment, the outcomes observed across the revised Lot 1 sub-lots and the T4 control are unambiguous and consistent with TBS mode of action demonstrated across all other trials conducted in Lao PDR.

14. CONCLUSIONS

The Vang Vieng TR4 trial demonstrates the capacity of TBS to suppress Panama TR4 disease progression on verified commercially infected Cavendish banana farms, notwithstanding significant adverse site management conditions:

- (a) TBS + NPK (Lot 1C) maintained healthy, productive Cavendish plants producing commercial bunches on soil from which all untreated control plants had died — an unambiguous demonstration of efficacy in a genuine TR4-disease environment.
- (b) TBS alone (Lot 1B) achieved better outcomes than NPK alone (Lot 1A), confirming TBS operates as a biological soil amendment — not merely a nutritional supplement — with direct suppressive effect on TR4 progression.
- (c) The total failure of the untreated control (T4) confirms TR4 disease pressure on the site was genuine, severe and commercially terminal without intervention.
- (d) DOA laboratory testing confirmed Foc TR4 (VCG 01213/16) presence at all 25 sample locations and demonstrated that the 3 m³/Ha TBS rate achieved statistically lower controlled-inoculation infection rates (75.7%) compared with lower-rate treatments (90.6–91.0%), approaching significance at $P \leq 0.05$.
- (e) Multiple adverse site management conditions — including quarantine failures, inadequate irrigation, poor weed control and uneven TBS distribution — significantly impacted Lots 2 and 3, leading to the September 2019 trial revision. These adverse conditions make the positive outcomes observed in Lot 1 all the more remarkable.
- (f) The trial was suspended in April 2020 due to COVID-19 national lockdowns in Lao PDR. Results reported represent the final observations prior to suspension.
- (g) These results form the basis of the TBS TR4 Management Protocol and have been reproduced in subsequent field trials in Australia (see East Palmerston TR1 Trial Report TBS-TR-QLD-TR1-2021).

15. CONTACT & FURTHER INFORMATION

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