



Impact of Intercropping and Biofertilisers in a 2-Year Field Trial in North-Central Victoria

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Executive Summary

Intercropping, the growing of two or more species simultaneously within the same field, is an agronomic practice that can potentially increase crop productivity and deliver multiple ecosystem benefits compared to monoculture crops.

To date, results from global meta-analyses from intercropping trials indicate that:

- Intercropping improves land use efficiency, i.e. less land needed to grow the same or more agricultural products (Tang et al. 2021)
- Intercropping results in more efficient use and uptake of phosphorus (Tang et al., 2022
- Cereal-legume intercrops can increase yield stability by 25% compared to cereal alone (33 studies, Raseduzzaman and Jensen, 2017)
- Intercrops reduced weed biomass by 58% compared to monocultures due to increased plant density and soil coverage (Gu et al., 2021)
- Intercrops support more significant populations of predatory insects, leading to reduced pests and the need for pesticides (Rakotomalala et al., 2023)
- Intercropping increased soil carbon (SOC) sequestration by 17.75% compared to monocropping. (Li et al. 2024, 33 papers covering seven countries and 35 crops). The SOC increase was primarily located in the top 20 cm, and Total N, C/N ratio, Microbial biomass C, and Microbial biomass N were positively correlated with increases in SOC.

This suggests that the practice of intercropping, especially the inclusion of legumes, leads to increased inputs of N and greater microbial populations, an integral source material for soil organic matter and carbon in soil (Miltner et al., 2012).

Many trials have been conducted in Australia in the last decades.

A review of the published literature indicates that:

- Across multiple studies in the southern wheat belt, Fletcher et al. 2016 found that:
 - 'Peaola' (canola-field pea intercrops) increased yield by 50% compared to monocultures in 70% of the 34 trials analysed.
 - Cereal-legume intercrops outperformed monocultures 64% of the time across 22 trials.
 - Intercropping cereal varieties together gave no statistical difference in yield compared to monocultures across 113 studies.



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Project Details

Title	The effects of intercropping and biofertilisers in a 2-year field trial in North-Central Victoria
Author	Dr. Adam O'Toole, SoilCQuest
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Introduction

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- Yield benefits and increased land use efficiency are not always observed (variable results) (Khanal et al. 2021)
- Positive yield results were found with intercropping of faba bean/wheat and pea/canola and are economically viable when intercropping results in a Land Equivalent Ratio (LER) >1.1* (Mitchell et al. 2021).

In summary, the weight of evidence from Australian research suggests that intercropping leads to improved agricultural productivity in most cases. However, results are not guaranteed, and multiple factors (rainfall and soil moisture status at germination, sow timing, knowledge and expertise, seed/C3/C4 combinations used, machinery, pests and diseases) may influence individual results. Intercropping has yet to be widely adopted in broadacre agricultural systems in Australia, where monocultures and rotation are the standard practice.

Khanal et al.(2021) identified some of the barriers that currently limit adoption, including:

- Greater complexity and demand for more skill and knowledge to implement intercropping successfully
- Possible increased machinery costs related to sowing and harvesting
- Increased postharvest costs related to grain separation from the different species grown together

SoilCQuest believes that greater adoption of sustainable farming practices is more likely when farmers can see demonstrated results on commercial farms so that both agronomic and economic elements can be considered. SoilCQuest conducts its research primarily on commercial farms, so we collaborated with farmer Grant Sims on this project.

Grant operates a mixed cropping and grazing enterprise in Lockington, Victoria. For the last 15 years, the farm has implemented several innovative and sustainable practices, including multi-species cover crops and on-farm production of biofertilisers. Grant has improved his profitability by producing custom biofertilisers, which can be applied as foliar sprays at specific periods in the growing cycle to provide plants with specifically required nutrients and stimulate microbes to support improved nutrient cycling and delivery.



^{*}A LER of 1.1 means that 10% more crop yield was produced with intercrops than if monocultures were grown on the same land area. If LER=1, there is no additional yield benefit from intercropping. If LER<1, intercropping is reducing yields compared to monoculture cropping. Contrary to expectations, greater water use alone was not a significant predictive indicator for observed yield benefits with intercrops (i.e., multiple factors were at play) in a 2019-2020 field trial at two locations in Victoria (Wallace et al. 2021).

Objectives

In 2019-2020, SoilCQuest collaborated with Grant Sims and Kalyx (trial manager) to scientifically validate the intercropping and biofertiliser application in a field experiment conducted on Grant's farm.

The objectives were:

- 1. To measure whether soil carbon changes in an intercrop vs monoculture crop over a 2-year rotation
- 2. To evaluate the effect of biofertiliser (vermicast + guano + farm-made biofertiliser) on soil carbon and crop yields compared to synthetic fertiliser (MAP and urea) as a basal fertiliser over a 2-year rotation
- 3. This study evaluates the economic outcomes of intercropping combined with farm-made biofertilisers compared to monoculture practice with modest mineral fertiliser application.





Methods

Field site

A small-plot field experiment was carried out at Pine Grove, Victoria (-36.173163, 144.442796), (Fig. 1), in the winter growing season of 2022 and 2023. The soil texture is a silty clay loam Sodosol.

Kalyx Australia Pty Ltd conducted the plot establishment, sowing, fertilisation, harvesting and pest management.



Fig. 1. Canola 02.06.2022 (left) Canola and vetch 22.07.22 (middle), sowing on June 1, 2023 (right).



Experimental design

Plots were arranged in two blocks with a buffer strip between each block. (Fig. 2). The experimental design consisted of 6 treatments and five replicates, as described in Tables 1 and 2 for 2022 and 2023, respectively.

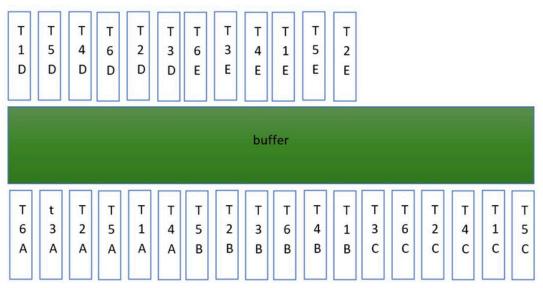


Fig. 2. Field trial layout. T=Treatment, Numbers=Treatment nr., Letters=replicates.

Treatment	Сгор	Fertiliser
T1	Canola and Vetch	Biofertilisers
T2	Canola and Faba Bean	Biofertilisers
T3	Canola	Biofertilisers
T4	Canola and Vetch Mineral fertilis	
T5	Canola and Faba Bean	Mineral fertiliser
Т6	Canola	Mineral fertiliser

Table 1. Experimental design 2022

Table 2. Experimental design 2023

Treatment	Сгор	Fertiliser
T1	Wheat 'Scepter' and Pea	Biofertilisers
T2	Pea	Biofertilisers
Т3	Wheat 'Scepter'	Biofertilisers
Τ4	Wheat 'Scepter' and Pea	Mineral fertiliser
T5	Pea	Mineral fertiliser
Т6	Wheat 'Scepter	Mineral fertiliser

*The Appendix provides more details on the crop sowing rates and the types and doses of fertiliser.



Description of fertilisers used in the study

Grant Sims supplied seeds, minerals, and biofertilisers. The biofertilisers used at sowing were 30 L Liquid Inject (30 L/ha), Guano: 40kg/ha, and Vermicast 2 kg/ha (produced by Nutrisoil*). The mineral fertilisers used at sowing were MAP at 100 kg ha-1 and 50 kg urea-1. No top dressing of nitrogen was applied during the season.

Sowing date, temperature and precipitation

In 2022, Canola, vetch, and faba bean sowing occurred on 27/4/2022. In 2023, the owing of wheat and field peas occurred on 1/6/2024. Precipitation in 2022 was 556 mm and 428 mm in 2023. Notably, nearly half the rainfall that fell in 2022 occurred in October (Fig. 3).

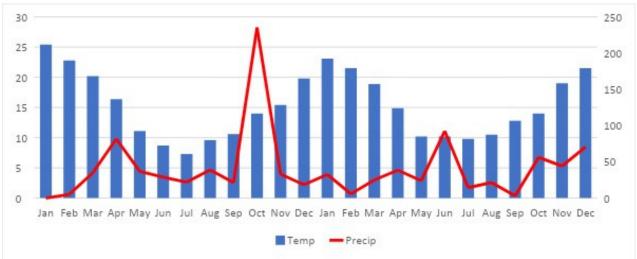


Fig. 2. Temperature and precipitation in 2022 and 2023, recorded at the Pine Grove Fire Station, approximately 1 km from the field site.



Biofertiliser

The biofertiliser used is a custom-made liquid fertiliser brewed by Grant Sims in large on-farm tanks (Fig. 4). It consists of a proprietary mix of organic and inorganic ingredients, which are lacto-fermented. While many farmers are brewing their own bioferments on-farm, their exact recipes may differ based on materials used and experience. A general guide has been published, indicating the types of materials to use and the process of how to do it (Hardwick, 2021). Grant Sims gives a general introduction to his philosophy of farming with cover-crops and biofertilisers in the following video: Integrating bio-fertilisers, cover crops and animals to improve soil health | Down Under Covers, <u>youtube.com/watch?v=JqGjR_FcLel</u>



Fig. 4. Tanks used for fermenting different blends of biofertilisers on Grant Sim's farm (left), and image of the liquid biofertiliser with high humic content injected into the sowing furrow (right). Image: Grant Sims, <u>downundercovers.com</u>

*https://nutrisoil.com.au/nutrisoil-worm-castings



Measurements taken

Table 3 lists the measurements taken on the field in 2022 and 2023.

Year/Month	Method/Activity	Measurements taken/data collected
2022 (June) Baseline soil samples		Soil C (0-10cm, 10-20, 20-30 and 30-60 cm) and bulk density. Baseline soil sampling in June 2022 showed that bulk density was uniform across the experimental site, with the two blocks recording an average bulk density of 1.1 g cm-3
2022 and 2023	Plant establishment count	Nr of plants m-2 (2 subsamples taken per plot)
2022	Leaf tissue sampling	July – 10 leaf samples per plot, aggregated to analyse per treatment (50 leaf samples)
2022 and 2023	Yield	Grain (kg/plot converted to kg/ha) Seed weight separated between grain and legume (only in 2023)
2022 (Dec)	Post-harvest soil sampling	Soil C (0-10cm, 10-20, 20-30 and 30-60 cm) and bulk density
2023 (Dec)	Post-harvest soil sampling	Soil C and bulk density at 0-10 cm

Table 3. Methods and measurements taken throughout field trial

Statistical analysis

Yield results were analysed using a linear mixed model using the Lme4 package in R (R Core Team (2024). Fixed factors were Treatment and fertiliser type (biofertiliser vs mineral fertiliser), and the block was taken as a random factor. ANOVA of model results was done using the 'emmeans' package.



Results and Discussion

Yield

In 2022, yield was not significantly different between the individual 6 treatments. However, the grouped mineral fertilised treatments were substantially higher (p=0.04) than the biofertiliser treatments (Table 4). In 2023, pea alone had a poor establishment. Intercropping performed slightly better regarding land use efficiency with mineral fertiliser (LER= 1.04) than biofertilisers (0.94). However, for intercropping to return a positive economic return for farmers, Mitchell et al. 2021 calculated from intercropping field trials in Rutherglen, VIC, that the intercrop LER would need to be greater than 1.1. A Land Equivalent Ratio > 1 indicates that intercrops lead to more productivity than mono-crops per unit of land used. An LER<1 means that intercrops are performing worse than mono-crops.

2022		
Biofertilised	Mean (t ha-1)	SE±
T1: Canola Vetch	1.53	±0.29
T2: Canola Faba	1.41	±0.29
T3: Canola	1.42	±0.25
Mineral-fertilised		
T4: Canola-Vetch	1.61	±0.24
T5: Canola-Faba	1.78	±0.18
T6: Canola	1.89	±0.18
Treatment (Not sig.)		p>0.5
Fertilisation		p=0.04

2023			
Biofertilised	Mean (t ha -1)	SE±	LER*
T1: Wheat-Pea	2.34	±0.34	0.94
T2: Pea	0.41	±0.14	
T3: Wheat	2.61	±0.19	
Mineral-fertilised			
T4: Wheat-Pea	3.05	±0.29	1.04
T5: Pea	0.40	±0.12	
T6: Wheat	3.04	±0.24	
Treatment***	p<0.001		
Fertilisation*	p=0.40		

*We were not able to calculate LER in 2022, because there was no monocrop of the legume grown separately in 2022. Both an intercrop and a monocrop of each species must be grown to calculate LER (mistake made during Experimental Design)



Considering that our trial did not receive top dressing, it was not surprising that in both years, the yield achieved was well below the district average (GRDC cereal variety trials). In our trial in 2022, the yield of monocropped Canola (1.89 t ha-1) was 38% less than that achieved at canola variety trials conducted nearby at Diggora (3.07 t ha-1) (GRDC, 2022).

In our trial in 2023, the yield of monocropped Field Pea (0.4 t ha-1) was about a third of the average yield from field pea variety trials in Deniliquin in 2023 (1.53 t ha-1) (GRDC, 2023a). We inspected the sowing dates at both sites for both years, and there was a delay of no more than one week at our site compared to Diggora. Grant Sims commented, however, that the Diggora site has better soils and historically outperforms yields in Lockington/Pine Grove.

The trial managers at Kalyx reported that the low yield in the field peas in 2023 could be partly explained by chemical drift (or presumably broadleaf herbicide) from a neighbouring paddock, which set back pea growth and establishment early in the season. Similarly, our yield of monocropped wheat in 2023 (3 t ha-1) was under half of that achieved at nearby variety trials at Diggora (6.31 t ha-1) (GRDC, 2023b).

Our analysis of plant tissue showed that tissue phosphorus (P) was significantly lower in the biofertiliser treatments (supplied by Guano and Vermicast) compared to MAP, which may have contributed to the lower yield in the biofertiliser treatments.

We should note that Grant Sims' farming approach involves low inputs of mineral fertilisers and a supply of required crop nutrients via stimulation of soil biology and targeted foliar applications tailored to the plants' needs (as dictated by plant tissue and sap tests). Grant states, "What is important for the farmer is not how much total yield is achieved but the gross margin [profitability] of the system after input cost savings have been accounted for".



Economic Analysis

Contrary to expectations, mono-cropped canola with mineral fertiliser had the highest gross profit/ha with \$916/ha (Fig. 5). The lowest performer was the Canola-Faba intercrop on biofertiliser (\$679/ha). Input costs were up to 70% lower in the biofertiliser treatments (T1-T3) than in mineral fertiliser treatments (T4-T6). However, only Canola-Vetch with biofertilisers in 2022 achieved the same gross profit when grown with biofertilisers compared to mineral fertilisers. Canola-Faba intercrop and Canola mono-crop grown with biofertilisers were 10 and 18% lower in profit than their mineral fertiliser counterparts. A full description of the numbers used to calculate gross profit is given in the appendix in Table S4.

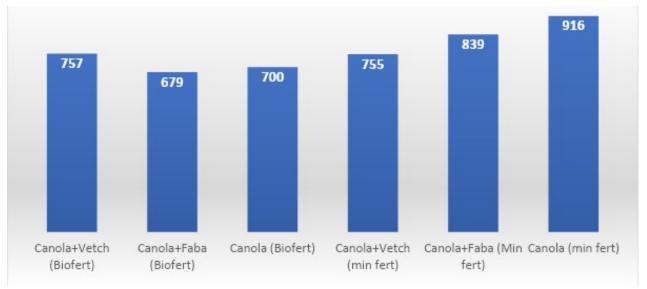


Fig. 5. Gross profit per ha in 2022 from the different intercropping x biofertiliser treatments.



Soil Carbon Analysis

Results from the three soil sampling campaigns showed that soil carbon concentration increased from approximately 1.65% in June 2022 to 1.8% in December 2022 (Fig. 6). This concentration declined in the final measurement on December 23 to below the baseline level of 1.55%. Notably, the seasonal differences in soil carbon are far greater than those between the six treatments. These seasonal differences are likely due to natural variation in soil carbon that occurs over an annual cycle. This phenomenon is well documented by Scheidung et al. (2017), who observed a coefficient of variation of 20% in soil organic carbon (SOC) when measuring monthly over a year.

Inter-laboratory variation may also be a contributing factor, as three separate labs analyzed each of these three measurements. It is noteworthy that soil carbon concentrations varied by an average of 9% at the treatment level at baseline soil measurements and 8% at the final measurement in December 2023. This variation suggests that to detect changes in SOC stocks due to a practice change, SOC would need to increase by a minimum of 10%, and ideally by more than 20%, to make more confident assertions about linking practice changes to soil carbon increases. Additionally, even within one plot (measuring 12 m × 1.5 m), samples taken within a few meters of one another varied by an average of 10%.

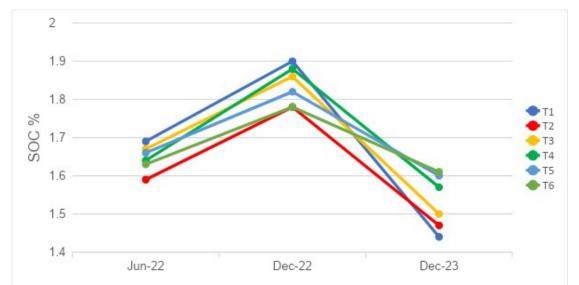


Fig. 6. Soil Organic Carbon (%) concentration at 0-10cm soil depth from samples taken at the field trial's start, middle, and end.



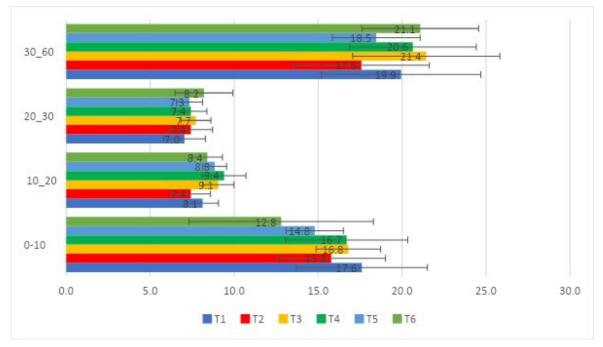


Fig. 7. Post-harvest (Dec. 22) soil organic carbon stocks (t C ha-1) at four depth increments from 0-60cm depth.

Another finding from the study was there was considerable soil carbon found in the subsoil (30-60 cm depth) (Fig. 7), which made up 38% of the carbon found in the 0-60 cm depth (Fig.8). As soil carbon is mainly derived from plant roots (Rasse et al. 2005), this illustrates the legacy effect of plant roots growing deep into the soil, and contributing to the SOC pool. At the 30-60 cm depth, soil carbon is significantly protected from the higher microbial decomposition rates at the 0-30 cm depth. Cultivating multiple species in intercrops may provide opportunities to exploit more significant volumes of the soil due to the different rooting depths between species (Homulle et al., 2022).

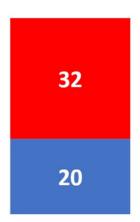


Fig. 8. Distribution of SOC (t ha-1) at 0-30cm (red) and 30-60cm (blue) from samples taken Dec. 22.



Conclusions and Reflections

Contrary to expectations and the majority of intercropping trials undertaken in Australia, this 2-year small-plot trial in North Central Victoria found limited productivity and economic benefits from Canola-Vetch, Canola-Faba, and Wheat-Pea intercropping during the 2022-2023 seasons compared to monocropping Canola and Wheat.

Despite significantly reduced input costs from using farm-made biofertilisers, the decreased yield could not compensate for the reduced revenue due to yield reductions, except for the biofertiliser Wheat-Pea intercrops in 2023.

Our initial intention in conducting this study was to scientifically document the positive results achieved by Grant Sims on his farm using multi-species crops and low-cost, farm-made biofertilisers. In hindsight, our approach to running the trial with small plots needed revision to replicate the results Grant had achieved in his commercial practice. For example, one of the critical benefits of biofertiliser use is the observed reduction in pest incidence. However, this benefit may not be obtainable in a small plot trial where mineral fertiliser and biofertiliser applications are co-located nearby, allowing pests to enter the site and potentially damage both treatments.

For this reason, we decided to prematurely shorten this field trial from 4 years to 2 years and instead invest our energy and resources into a full paddock trial where Grant, as the farmer, will have a more direct role in implementing his ideas on a larger scale. This project, called Multi-Graze, is the next initiative SoilCQuest will be conducting with Grant Sims from 2024 to 2027. On a positive note, we gained valuable insights into the variability of soil organic carbon (SOC) concentrations when measured at three different time points over two years. These insights will help guide future experiments regarding the frequency of measurements and the trial duration needed to observe changes in SOC concentrations due to new practices.



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Appendix

	D Treatment Seeding Rate Fertilisation - Fertilisation Rate In season fertilisation						
ID.	Description (Year 1)	Seeding Rate	Sowing	Fertilisation Rate	(pending confirmation from Kalyx on actual amounts applied as top dressing)		
T1	Canola + Vetch	C: 2kg./ha V: 5kg/ha	Bio Fert Guano VermiCast	BF: Liquid Inject (Amount?) Guano: 40kg./ha VC: 2kg./ha	Foliars: Same as adjacent paddock granular ammonium sulphate: 65kg/ha. (season dep.) UREA 2/3 applications of 20kg./ha		
Т2	Canola + Faba Beans	C: 2kg./ha FB: 20kg/ha	Bio Fert Guano VermiCast	BF: Liquid Inject (Amount?) Guano: 40kg./ha VC: 2kg./ha	Foliars: Same as adjacent paddock granular ammonium sulphate: 65kg/ha. (season dep.) UREA 2/3 applications of 20kg./ha		
тз	Canola	C: 2kg/ha	Bio Fert Guano VermiCast	BF: Liquid Inject (Amount?) Guano: 40kg./ha VC: 2kg./ha	Foliars: Same as adjacent paddock Granan: 65kg/ha. (season dep.) UREA 2/3 applications of 20kg./ha		
T4	Canola + Vetch	C: 2kg./ha V: 5kg/ha	МАР	MAP: 70kg./ha	UREA: 2 applications (pre-rain) of 150kg/ha. (season dep.)		
Т5	Canola + Faba Beans	C: 2kg./ha FB: 20kg/ha	МАР	MAP: 70kg./ha	UREA: 2 applications (pre-rain) of 150kg/ha. (season dep.)		
Т6	Canola	C: 2kg/ha	MAP	MAP: 70kg./ha	UREA: 2 applications (pre-rain) of 150kg/ha. (season dep.)		



ID	Treatment Description (Year 2 - 2023)	Seeding Rate Seeding depth	Fertilisation - sowing	Fertilisation Rate sowing	in season fertilisation *(pending confirmation from Kalyx on actual amounts applied as top dressing)	Plant protection
т1	75% Wheat (specter) + 25% Pea	W: 55 kg./ha, 20mm P: 23kg/ha, 35mm	Bio Fert Guano	BF: Liquid Inject (30 L) Guano: 40kg./ha Vermicast 2 kg/ha	Grant Sims custom-made foliar: Coordinate this closely with Grant Sims, according to plant sap tests at the tillering and flag leaf stage. It can include foliar N and soluble nutrients, which may cause the plant to be deficient.	Pre-emergent herbicides can be used, but no fungicides or insecticides. Grants foliars wi be used instead.
T2	Pea	P: 90kg/ha, 35 mm	Bio Fert Guano	BF: Liquid Inject (30 L) Guano: 40kg./ha Vermicast 2 kg/ha	Grant Sims custom-made foliar: Coordinate this closely with Grant Sims, according to plant sap tests at the tillering and flag leaf stage. It can include foliar N and soluble nutrients, which may cause the plant to be deficient.	Pre-emergent herbicides can be used, but no fungicides or insecticides. Grants foliars wi be used instead.
T3	Wheat	W: 80kg/ha, 20mm	Bio Fert Guano VermiCast	BF: Liquid Inject (30 L) Guano: 40kg./ha Vermicast 2 kg/ha	Grant Sims custom-made foliar: Coordinate this closely with Grant Sims, according to plant sap tests at the tillering and flag leaf stage. The sap tests will determine. It can include foliar N and soluble nutrients, which may cause the plant to be deficient. Consider the use of UAN combined with Grant's micronutrient and chelating agents	Pre-emergent herbicides can be used, but no fungicides or insecticides. Grants foliars wi be used instead.
T4	75% Wheat + 25% Pea	W: 55 kg./ha, 20mm, P: 23kg/ha, 35mm	MAP	MAP: 100kg/ha, placed at 20 mm? 25 kg/ha urea (reducing N to avoid disabling N fixation too much)	50 L/ha Urea Ammonium Nitrate, as foliar spray (timing important)	Use conventional plant protection products as appropriate. Keep notes. Kaly will advise on this.
Τ5	Pea	P:90kg/ha	МАР	MAP: 100kg/ha	No in crop fertilisation	Use conventional plant protection products as appropriate. Keep notes. Kaly will advise on this.
_				Placed at 35mm		
τ6	Wheat (as per standard practice In the district)	W: 80kg/ha	МАР	MAP: 100kg/ha, Urea 50 kg/ha Placed at 35mm	UREA: 2 applications (pre-rain) of 100kg/ha. (season dep.) As per standard practice	Use conventional plant protection products as appropriate. Keep notes. Kaly will advise on this.



Chemical	Concentration
Total N	2.74%
Total P	11.9%
κ	0.16%
Organic C	10%
S	2.4%
Ca	27.5%
Mg	1.9%
Na	0.08%
Mn	22273 mg/kg
Zn	2597 mg/kg
Cu	522 mg/kg
Fe	24399 mg/kg
В	42.4 mg/kg
Co	13.4
Mo	5.35 mg/kg

Table S3. Indicative Chemical Analysis of Guano used in the field trial. Values supplied by the supplier (Soil Management Systems, www.soilms.com.au)

Reference: Soil Management Systems, www.soilms.com.au



Table S4. 2022 Eco	nomic Analysis	for Grant Sims	Trial
		ion on only on the	

	т1	T2	тз	Т4	T5	Т6
	Canola+Vetch	Canola+Faba	Canola	Canola+Vetch	Canola+Faba	Canola
Yield (t ha-1)	1.53	1.41	1.42	1.61	1.78	1.89
5-year av canola price 2020	560	560	560	560	560	560
Revenue	856.8	789.6	795.2	901.6	996.8	1058.4
CanolaSeed amount (kg/ha)	2	2	2	2	2	2
Seed price	15	15	15	15	15	15
Seed cost per ha	30	30	30	30	30	30
Vetch/Faba Seed amount (kg/ha)	5	20		5	20	
Seed price	1	0.8		1	0.8	
Seed cost per ha	5	16		5	16	
MAP amount (kg/ha)				70	70	70
MAP price/tonn				1600	1600	1600
MAP cost per ha				112	112	112
Biofert amount (L)ha)	30	30	30			
Biofert price (\$/L)	1	1	1			
Biofert cost per ha	30	30	30			
0	40	10				
Guano (kg/ha)	40	40	40			
price cost per ha	850 34	850 34	850 34			
	0.1	0.	01			
Vermicompost (kg/ha)	2	2	2			
price	500	500	500			
cost per ha	1	1	1			
Total costs	100	111	95	147	158	142
Reduction in input costs	47	47	47			
% Reduction in input costs	68%	70%	67%			
Gross profit (\$/ha)	757	679	700	755	839	916
% Gross profit compared to counterpart control	0%	-10%	-17%	-18%	-8%	

