

Effect of Amino Acids, Fulvic Acid & CPPA on the Breakdown of Soil Glyphosate Residue through the use of Fast Blast®

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Abstract

Glyphosate is commonly known for its use in broad-spectrum systemic herbicides in the agricultural industry to knock-off weeds. This has led to the multinational adoption of Glyphosate and different derived salts, making it the leading herbicide choice amongst farmers due to its effectiveness. However, debate over its biological and environmental effects have been frequently discussed, especially the issue around Glyphosate residue remaining in soil for long periods of time which negatively impacts soil biology & natural eco systems. Australian studies have shown that the 2 most commonly detected chemical residues in Australian grain cropping soils are Glyphosate and its primary metabolite, Aminomethylphosphonic Acid (AMPA) (Van Zwieten, 2016). It was detected that 50% and 75% of grain cropping soils detected Glyphosate and AMPA residues respectively (Van Zwieten, 2016). These residues bind tightly to soil particles which result in extended break-down rates. Soil micro-organisms are able to further break down residues in the soil, however there are conflicting studies suggesting that Glyphosate has a negative impact on soil microbe communities. The aim of this trial is to assess the increased breakdown rate of Glyphosate and AMPA residues in soil treated with a unique liquid blend (Fast Blast®) consisting of Amino Acids (AA), Fulvic Acid (FA) & CPPA (complex polymeric polyhydroxy acid). Soil cores 15cm deep were taken from control and treated soil every 10 days for 4 consecutive rounds and externally analysed for both Glyphosate and AMPA residues. It was determined that there was a statically significant decrease ($P < 0.05$) in the average level of Glyphosate residue in soil treated with AA, FA and CPPA compared to the control. There was also a steady decrease in the percentage difference in relation to the maximum Glyphosate residue recording on treated soil compared to the control. Furthermore, there was a percentage decrease of 55% in Glyphosate treated with the blend, compared to the control.

Keywords: Glyphosate, herbicide, weed control, soil residue, Amino Acid, Fulvic Acid, CPPA, Glyphosate breakdown

1. Introduction

Weed control is an important aspect of any agricultural production system, as weeds compete with the primary crop for water, light and nutrients resulting in a negative impact on the yield of the primary crop grown. Due to this competition, weeds

need to be removed and controlled to reduce the stress inflicted on primary crop. One of the main chemicals that is used to eradicate weeds in agricultural production systems is Glyphosate [N-(phosphonomethyl)-glycine]. Glyphosate is a broad-spectrum herbicide used worldwide to eliminate weeds in agriculture. Over 500 glyphosate products have been registered in Australia including the very

popular product – Roundup. Due to Glyphosates quick an effective kill, it has been adopted by many Australians for over 40 years (Glyphosate, 2019).

Glyphosate is branded as a group M herbicide meaning that it inhibits the production of the plant-based enzyme 5-enolpyruvyl shikimate-3 phosphate (EPSP) synthase. This enzyme plays a role in the synthesis of 3 key amino acids (phenylalanine, tyrosine and tryptophan) which are needed to create certain proteins required for plant growth (Kanissery et al., 2019).

Glyphosate is widely used by farmers, gardeners and other land managers to effectively control weeds without needing to use older, traditional methods of weed control through ploughing or tilling soil which reduces soil structure and increases soil erosion. Glyphosate is also effective at knocking-down invasive and noxious weeds which improves crop health and reduces competition with the primary growing crop for water and nutrients.

However, over the past 10 years there has been ongoing investigations globally into Glyphosate's safety concerns and associated negative consequences to the environment. In Australia, 18 different weeds species have developed Glyphosate resistance resulting in the use of higher Glyphosate concentrations to combat the excessive weed growth (Herbicide Resistant Weeds, 2021). In terms of soil health, it has been observed that Glyphosate has an affinity to bind tightly to soil particles and therefore accumulates in the top-soil layers leading to numerous environmental risks (Kanissery et al., 2019). Processes such as surface run-off, drift and vertical transport in soil may transport the Glyphosate into ground water and surface water and it has been confirmed that Glyphosate was detected in groundwater which can affect flora and fauna in streams, rivers, wetland and then ocean. This is even more damaging when taking into consideration the extensive usage of Glyphosate which may pose hazards to the ecological environment (Kanissery et al., 2019).

Glyphosate is popular because it is immovable in the soil due to it binding with soil particles and will reside in the first few centimetres of the surface soil. However, after more research, the half-life of Glyphosate is believed to be longer than previously believed. Once glyphosate is applied to the soil it will take 140 days for it to break down to its half-life and

then will take another 2 years for it to completely be broken down in the soil (Stojanović, et al., 2017). This residue of Glyphosate in the soil can cause environmental pollution to both the soil but also water bodies due to leaching of the herbicide out the soils. In 2015, it was concluded that Glyphosate was “probably carcinogenic to humans” by the International Agency for Research on Cancer (WHO), World Health Organisation, 2015). Over the past few years, research has proven that even though glyphosate may be effective and efficient at eradicating weeds, there may be a lot of environmental and health issues that occur with a slow break down.

When researching the major route of glyphosate degradation in soil, it was found that it is mainly done through the microbial degradation process, meaning that soil microbe organisms such as bacteria and fungi are able to convert Glyphosate into aminomethylphosphonic acid (AMPA) – the metabolite of Glyphosate - depending on microbe health and environmental conditions. It was correlated that Glyphosate breakdown is related to the abundance of *Pseudomonas* bacteria in the soil. However, when repeated applications or long-term use of Glyphosate can reduce the abundance of Glyphosate mineralizing microorganisms (Kanissery et al., 2019). Therefore, it is important to maintain microorganism health.

Fast Blast® is a patented product developed by Dual Chelate Fertilizer Pty Ltd that is formulated to increase plant uptake of both foliar and soil applied products. Fast Blast® consists of 32.8% Biologically Active Organic Molecules (BAOM) also known as CPPA, 48.2% Organically Derived Amino Acids and 8.2% other Organic Acids such as Fulvic Acid. As Fast Blast® contains a multitude of organic molecules, amino acids and bio-stimulants, it is also great for soil and the microbial colonies that are found in the soil. Furthermore, it is a product that is formulated to increase the breakdown of glyphosate residual in the soil. The aim of this trial is to assess the increased breakdown rate of Glyphosate and AMPA residues in soil treated with a unique liquid blend (Fast Blast®) consisting of Amino Acids (AA), Fulvic Acid (FA) & CPPA (complex polymeric polyhydroxy acid).

The aim of this trial is to assess the increased breakdown rate of Glyphosate and monitor AMPA residues in soil treated with a unique liquid biological blend (Fast Blast®) consisting of Amino Acids (AA), Fulvic Acid (FA) & CPPA (complex polymeric polyhydroxy acid). This was a preliminary trial to see

the effects that these biological compounds have on glyphosate residue breakdown. In future trials, a more in-depth analysis will be done to better measure the relationship with biological compounds, soil microbes and herbicide residues

2. Objectives

The specific objectives of this study were:

1. To examine the glyphosate breakdown rate in the soil with Fast Blast®.
2. To examine the time it takes to significantly lower the Glyphosate residue in the soil with Fast Blast®.
3. To understand and demonstrate the benefits of Fast Blast®.

3. Materials and Methods

3.1 Site Selection and Trial Design

The trial was conducted in Robinvale, Victoria. There were two different sites, one was selected in an area where there was even soil and then area was divided into treatment plots of 1m² by 1m² with a buffer zone of 30cm between the different treatment plots. The second site was in the glasshouse, 6 pots with the dimensions of 25cm by 15cm were set up with a 30cm buffering zone between the pot treatments. The treatment plots and pots were arranged in a Randomised Complete Block design to avoid bias.

Table 1: Treatment application rates.

Treatments	Application Rates
Roundup Ultra MAX® + Fast Blast® Treatment	50mL Roundup Ultra MAX®/5L of water + 5mL Fast Blast®/5L of water
Roundup Ultra MAX® Control	50mL Roundup Ultra MAX®/5L of water

A backpack sprayer (Piston pump system 15L) with a 250mL/1m² water rate, was used to spray both the plots and pots to make sure there was an even spray coverage of Roundup® and Fast Blast®.

3.2 Quantitative analysis: Residual content of glyphosate in the soil

For the residual testing, a soil core (15cm) was taken from a plot located in a clear area of the vineyard and then a soil core (15cm) was taken from the matching pot in the glasshouse (i.e. plot 1 soil core was mixed with pot 1 soil core). The plots and pots were categorised into 2 different treatments:

1. Roundup Ultra MAX®
2. Roundup Ultra MAX® + Fast Blast®
3. Roundup Ultra MAX®
4. Roundup Ultra MAX® + Fast Blast®
5. Roundup Ultra MAX®
6. Roundup Ultra MAX® + Fast Blast®

Once soil cores had been taken, the treatments were mixed, meaning 1, 3 and 5 were mixed together and 2, 4 and 6 samples were combined. Combining the soil samples allowed for an overall reading to be obtained. The trial was done over a 9-week period and during this time, a total of 5 soil samples were taken from each trial plot and pot. The 5 soil samples were taken on day 4, day 14, day 26, day 40, day 53 and then express posted to ACS Laboratories, VIC, Australia to analyse the glyphosate residue in the soil.

4. Results and Discussion

The results from this trial show that with the addition of Fast Blast® + Roundup Ultra Max®, there is a decrease in the amount of residual glyphosate left in the soil. Over the 53-day period, Roundup Ultra Max® + Fast Blast® consistently showed less Glyphosate in the soil compared to Roundup Ultra Max® on its own. This has been proven to be statistically significant at the 95% confidence level which is highlighted in table 2 showing a P-value of 0.0342. This shows that the residual Glyphosate values obtained from the Fast Blast® treated soil are significantly lower than those without initial additions of Fast Blast®.

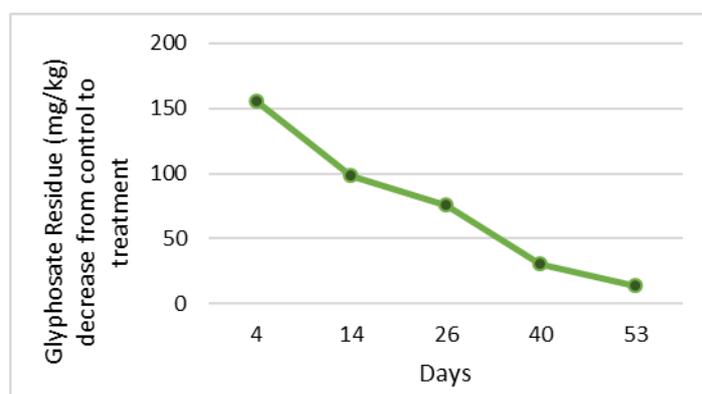


Figure 1: Percentage difference in relation to the maximum Glyphosate residue recording on treated soil compared to the control.

Table 2: Analysis of Glyphosate residue of different plots and pots. P-vale was calculated at the 0.05 significance level.

		Roundup Ultra MAX + Fast Blast	Roundup Ultra MAX
Glyphosate Residue (mg/kg)	Mean	217.6	481.6
	Std. Deviation	96.7	207.5
	Std. Error of Mean	43.2	92.8
	95% confidence interval	97.6 – 337.6	223.9 – 739.3
	P value (Control vs Treated)	0.0342	

When looking at figure 1, the graph shows that there is a continuous decrease in residual glyphosate in the soil when comparing Roundup Ultra Max[®] to Roundup Ultra Max[®] + Fast Blast[®]. This decrease shows that Fast Blast[®] effectively assists in the breakdown of Glyphosate residue during the 53 day period.

At day 53, there was a decrease of 44% in the residual content of glyphosate found in the soil when Fast Blast[®] was applied with Roundup Ultra MAX[®], which confirms that Fast Blast[®] facilitated in the increased the breakdown of glyphosate in the soil. After careful assessments of the results from this study, the additions of Fast Blast[®] with glyphosate herbicides can increase the rate of glyphosate breakdown in the soil.

After analysing the results, it is hypothesised that the significant decrease in glyphosate observed in soil treated with Fast Blast was due to the increase microbial activity breaking down the Glyphosate compounds into less threatening forms such as the AMPA, acetyl-glyphosate and sarcosine. As stated earlier, Amino Acids, CPPA and Fulvic acid all contain useful carbon and nitrogen which is utilised by soil microorganisms causing their population numbers to rise and metabolise the Glyphosate.

5. Future Research

Further research is to commence with trials already in the pipeline to solidify the outcomes observed in this trial. Future plans are to replicate this trial again and

include an analysis of the different pools of soil biota and also measure the amount of CO² produced by microorganisms in the soil from the mineralization of Glyphosate with the use of Fast Blast against a control.

6. References

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Appendix 1. Raw data from trial

Table 1: The results obtained from ACS laboratories Australia measuring the amount of Glyphosate and AMPA (Aminomethylphosphonic acid) in the soil in glyphosate treated soil and glyphosate + Fast Blast soil.

Day	Roundup Ultra Max [®] + Fast Blast [®]	Roundup Ultra Max [®]	Roundup Ultra Max [®] + Fast Blast [®]	Roundup Ultra Max [®]
	Glyphosate (mg/kg)		(AMPA mg/kg)	
4	155	660	18	37
14	120	190	79	83
26	328	424	59	56
40	170	430	29	35
53	315	704	43	26

Appendix 2. Statistical analysis of results

Values given are mean + standard deviation. P values <0.05 was considered to be significant

Parameter	Treatment		P- Value	Significance	% increase
	Roundup Ultra Max [®] + Fast Blast [®]	Roundup Ultra Max [®]			
Glyphosate Residue (mg/kg)	217.6 ± 96.7	481.6 ± 207.5	0.03	yes	264



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6 June 2019

Report No. ACS1920261

Penny Nesbitt
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Robinvale 3549

REPORT OF ANALYSIS

Date of Sample Receipt: 17th May 2019
Number of Samples Received: 2

Table 1: Results / (mg/kg)

Sample ID	Lab No.	Glyphosate	AMPA
Roundup Day 4	20261-1	660	37
Roundup fast blast day 4	20261-2	155	18

Method: ACS-TM-AM-029

Yours faithfully,
ACS Laboratories (Australia)

Barry Blythman
Analytical Chemist



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28 June 2019

Report No. ACS1920386

Penny Nesbitt
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REPORT OF ANALYSIS

Date of Sample Receipt: 31st May 2019
Number of Samples Received: 2

Table 1: Results / (mg/kg)

Sample ID	Lab No.	Glyphosate	AMPA
Control – Roundup only Day 14	20386-1	190	83
Round up + fast blast – Day 14	20386-2	120	79

Method: ACS-TM-AM-029

Yours faithfully,
ACS Laboratories (Australia)

Barry Blythman
Analytical Chemist



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28 June 2019

Report No. ACS1920507

Penny Nesbitt
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REPORT OF ANALYSIS

Date of Sample Receipt: 13th June 2019
Number of Samples Received: 2

Table 1: Results / (mg/kg)

Sample ID	Lab No.	Glyphosate	AMPA
Roundup – Day 26	20507-1	424	56
Roundup + fastblast – Day 26	20507-2	328	59

Method: ACS-TM-AM-029

Yours faithfully,
ACS Laboratories (Australia)

Barry Blythman
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19 July 2019

Report No. ACS1920737

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REPORT OF ANALYSIS

Date of Sample Receipt: 8th July 2019
Number of Samples Received: 2

Table 1: Results / (mg/kg)

Sample ID	Lab No.	Glyphosate	AMPA
Day 26 – Roundup	20737-1	430	35
Day 26 – Roundup + fast blast	20737-2	170	29

Method: ACS-TM-AM-029

Yours faithfully,
ACS Laboratories (Australia)

Handwritten signature of Barry Blythman in black ink.

Barry Blythman
Analytical Chemist



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23 July 2019

Report No. ACS1920814

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REPORT OF ANALYSIS

Date of Sample Receipt: 16th July 2019
 Number of Samples Received: 2

Table 1: Results / (mg/kg)

Sample ID	Lab No.	Glyphosate	AMPA
Roundup day 53	20814-1	407	26
Roundup + fast blast day 53	20814-2	315	43

Method: ACS-TM-AM-029

Yours faithfully,
ACS Laboratories (Australia)

Barry Blythman
Analytical Chemist