BioAg (New Zealand) Fertiliser & Stimulant Trials 2016 - 2018

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Executive Summary

The effect of BioAg biological applications on pasture production was assessed in Southland and Canterbury, New Zealand, over the 2016-17 and 2017-18 growing seasons.

Soil and Seed soil bio-stimulant, followed by application of Root and Shoot foliar bio-stimulant, was applied with calcium nitrate, urea and fish hydrolysate to dryland sheep and beef ryegrass-clover pasture near Mataura and irrigated dairy pasture near Rakaia.

In Southland BioAg applications significantly increased pasture yield in the 2016-17 growing season ($P < 0.05$ to $P < 0.01$).

Calcium nitrate increased average pasture production by 43%, showing N limited yield, and fish hydrolysate by 49%. Addition of Soil & Seed/Roots & Shoots increased production by a further 19% and 15%.

In the 2017 – 2018 growing season Roots & Shoots increased production by 12% and calcium nitrate by 11%, and Roots & Shoots with urea by 33%, though these were not statistically significant.

Similar response trends occurred in Canterbury in both growing season, though these were not statistically significant.

In 2016 – 2017 calcium nitrate increased yield by 8% and with Roots & shoots by a further 3%. Fish hydrolysate lifted production by 8% and by 15% with Roots and Shoots.

In 2017 – 2018 Roots & Shoots increased production by 28% and calcium nitrate by14%, and a higher application rate of urea by 54%.

These increases with Soil & Seed / Roots and Shoots are not attributable to direct nutrient supply and suggest that BioAg applications may stimulate soil microbial functioning and increase nutrient or micronutrient availability.

Contents

1. Introduction

BioAg PTY Ltd. represented in NZ by BioAg Ltd is a leading Australasian producer of natural microbial formulations for biological farming programmes.

Development and testing of microbial cultures to match specific requirements of plant growth commenced in 1983 in the United States. Commercial production of Australian products began in 1999 based on combining propriety microbial cultures with sugars, seaweed concentrates, fish hydrolysate and other components during aerobic fermentation. The resulting bio-stimulants contain a range of metabolised vitamins, amino acids, proteins, minerals and growth promoters which act both as bio-stimulants and for element supply. Applied as liquid formulations they are claimed to stimulate soil microbial activity, enhance nutrient availability and uptake and to significantly improve crop and pasture growth[1, 2].

AgScience was commissioned to independently assess the effect of BioAg fertilisers on pasture production in New Zealand. Field trials in Southland and Canterbury commenced during the 2016 - 2017 growing season and continued during the following season.

1.1 Trial Sites

The Southland trial site at Te Tipua, near Mataura, is typical of ryegrass- white clover pastures used for sheep and beef production (Figure 1). The Canterbury trial site at Dorrie, near Rakaia, comprises typical centre pivot irrigated, high fertility ryegrass clover dairy pasture (Figure 2). Both trials were integrated with companion Southern Humate trials at these sites.

Figure 1. Southland, Te Tipua trial site, showing companion humate urea trial plots. BioAg plots located in cut unfertilised rows running from left to right

[1] Barton, A. 2016. BioAg Company Overview. (https://morellofert.com.au/wp-content/uploads/2015/08/Trial-data.pdf)

[2] BioAg. Better soils. Better crops. Better stock. 2019. (https://www.bioag.co.nz).

Figure 2. Canterbury, Dorrie trial site, shallow soil phase. BioAg plots between tapelines.

1.2 Soil Morphology

Southland soils are Waimumu silt loam and Templeton silt and sandy loams at Canterbury. Additional site and soil information is given in previous reports[3]. Both trial sites were precision soil mapped to determine possible soil phase effects (Figure 3).

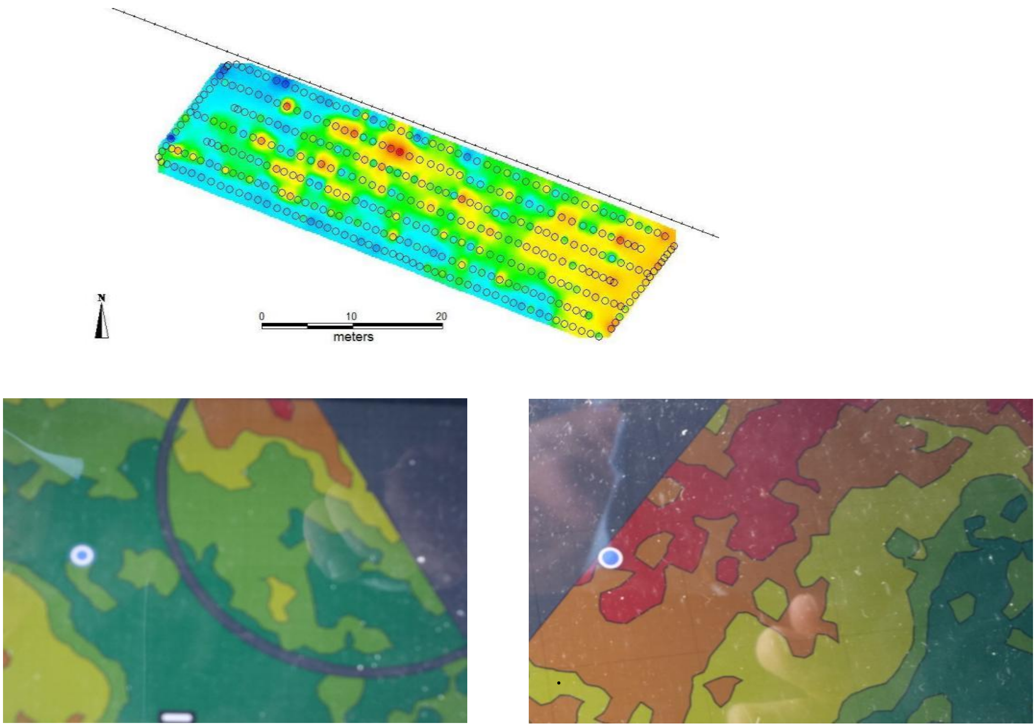


Figure 3. Lower: Southland: shallow (light green) to deep (red) soil phases and trial site.
Upper: Canterbury: shallow (light green) to deep (red) soil phase and trial sites (blue dot).

[3] Espie, P.R. 2014. Southern Humate Trial Establishment October 2014. AgScience Contract Report, November 2014, 12 pp;
Espie, P.R. 2014. Rotokaia Farm Ltd. Environmental Evaluation Soil Phase Assessment. AgScience Contract Report, September 2014. 13 pp.

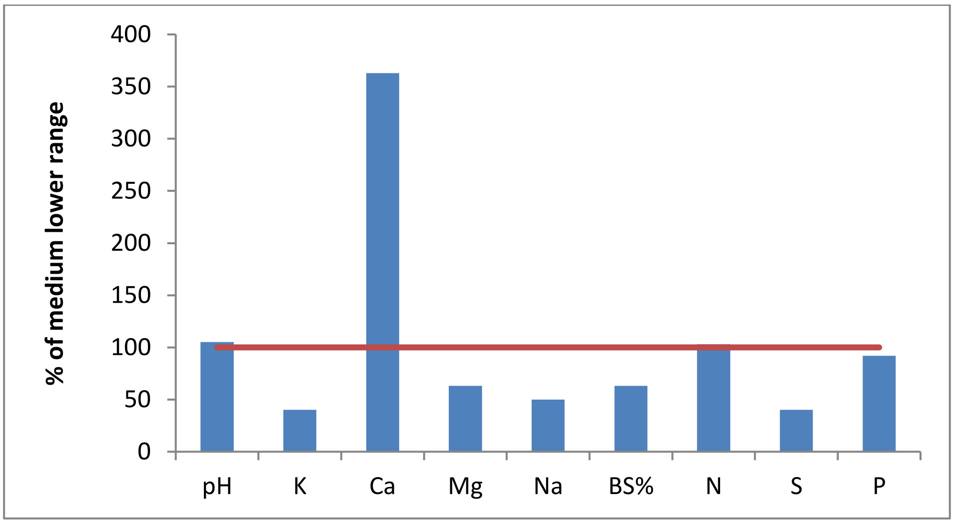
Soil phase profiles were very similar in Southland but differed in Canterbury due to depth to subsoil gravels (Figure 4). Therefore BioAg stimulants were tested separately on the shallow and deep soil phases in Canterbury.

Figure 4. Upper : Southland . Left typical soil profile, depth 70 cm. Right: upper topsoil to 30 cm
Lower: Canterbury. Left: deep phase, gravel depth 64 cm. Right: shallow, gravel 30 cm.

1.3 Topsoil Chemistry

Topsoil chemistry was measured before trial commencement. In Southland the whole trial site was assessed in October 2014 from two 2.5 x 15 cm soil cores were sampled from forty regularly spaced plots (Figure 1). These were bulked, mixed and subsampled for standard nutrient analysis by Hills Laboratories, New Zealand.

Soil pH was 6.1 (medium) with low phosphorus (P) and sulphur (S) levels. Available nitrogen (N) and calcium (Ca) levels were medium or high, due to previous lime and fertiliser application, but potassium (K) magnesium (Mg) and sodium (Na) levels and percentage base saturation (BS%) were all low. Molybdenum was the only trace element with lower than average levels (Figure 5, Appendix 5.1).



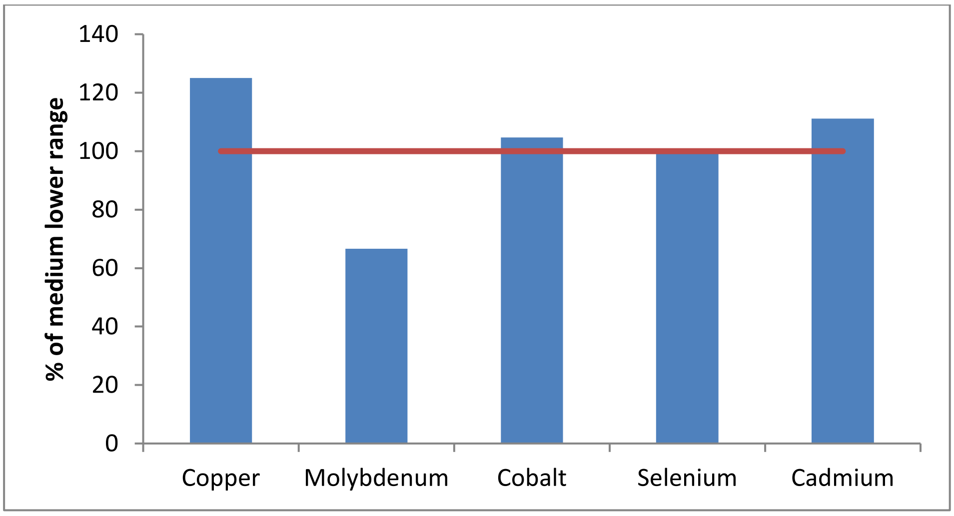
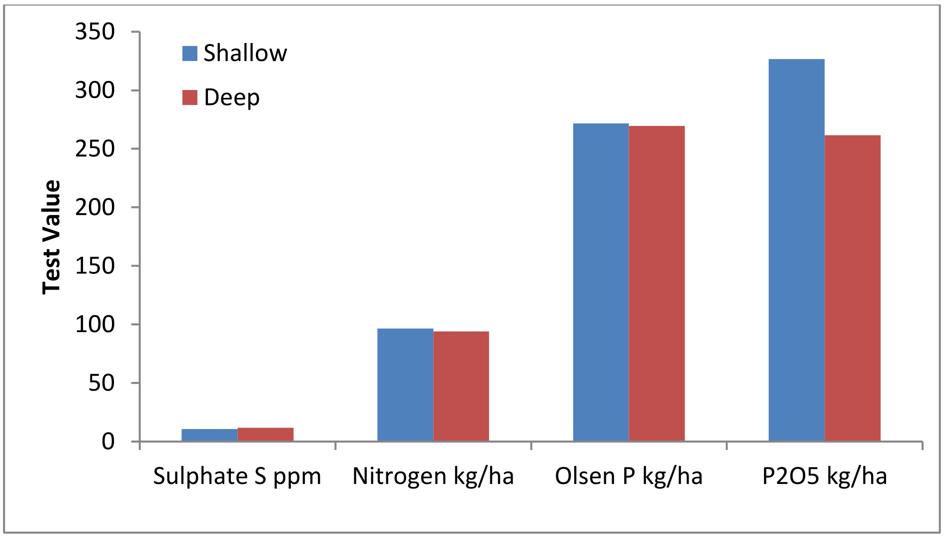


Figure 5. Upper. Topsoil chemistry. Test values as a percentage of the lower limit of the medium range for each value.
Lower. Topsoil trace element Mehlich 3 values as a percentage of the lower limit of the medium range for each value.

Topsoil chemistry in Canterbury was measured in November 2016 from ten 2.5 x 10 cm soil cores, sampled twice, from the shallow and deep soil phase trial areas. Samples were bulked, mixed and subsamples taken for standard nutrient analysis by Perry Analytical Laboratories, USA.

Soil pH was 6.8 (high) on the shallow soil phase and 6.4 (medium) on the deep phase with similar nitrogen and Olsen phosphorus levels between the phases. Sulphate sulphur levels were low and total elemental P, reported as phosphorous oxide P_2O_5, was slightly higher on the shallow phase (Figure 6). When scaled according to the Hills Laboratory lower threshold values for adequate fertility (medium range) only organic matter (OM) levels were slightly below this limit. Note that the high magnesium and sodium percentage levels reflect their much smaller soil concentrations, hence greater variability in calculating the percentage difference (Figure 6, Appendix 5.2). Total soil potassium levels are low and magnesium levels are high relative to Kinsey-Albrecht Perry laboratory recommended levels (Figure 7).



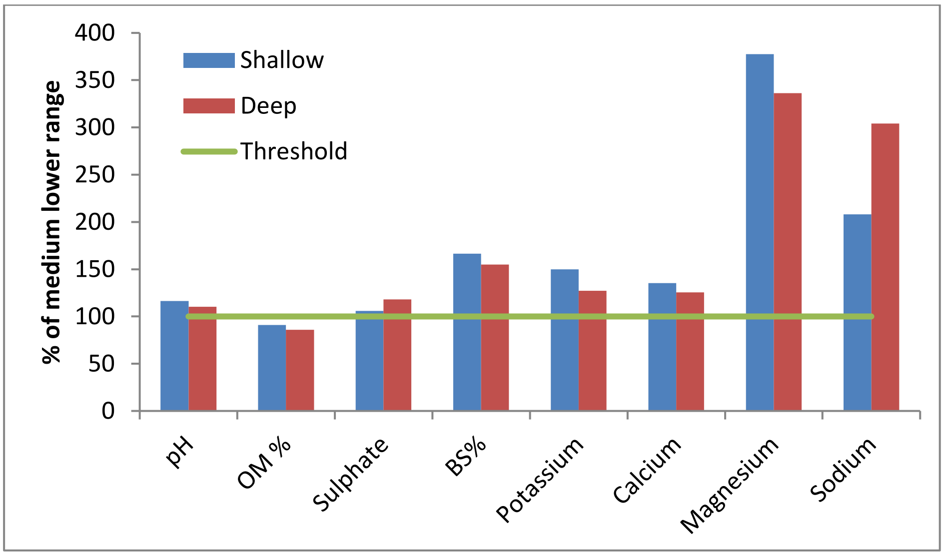


Figure 6. Total and relative soil test values as a percentage of the lower limit of the medium range for each value on shallow and deep soil phases.

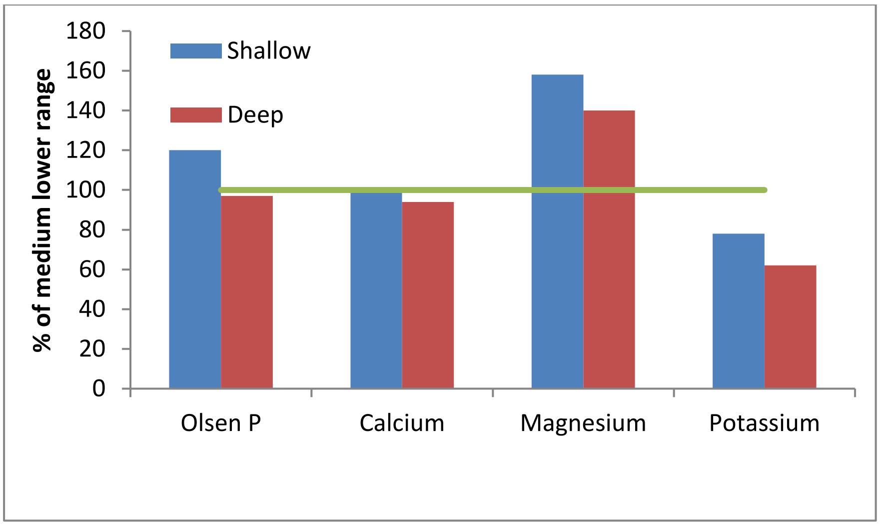


Figure 7. Soil test values as a percentage of the desired Kinsey – Albrecht level.

Trace elements were reasonably similar between soil phases. Iron and manganese were the most abundant followed by zinc with lower levels of copper boron and cobalt (Figure 8). .

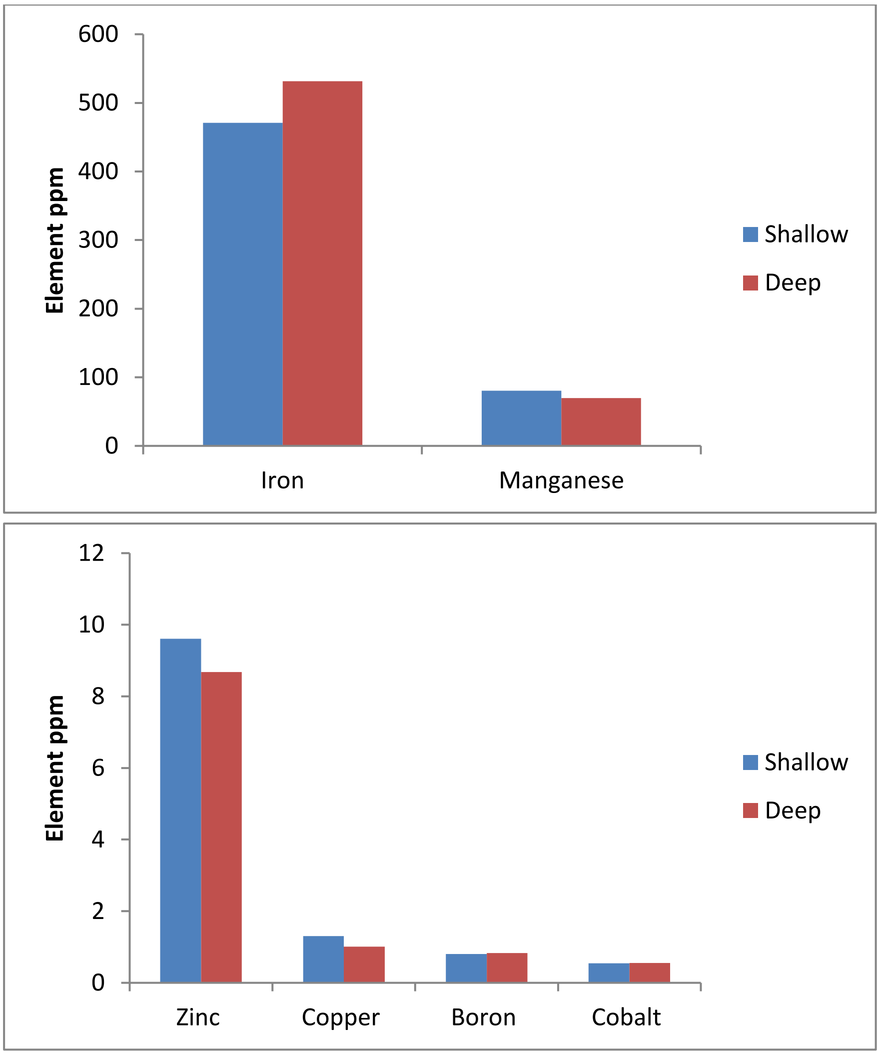


Figure 8. Soil phase micro-nutrient levels.

Topsoil chemistry was re-measured at the end of the trials. In Southland five unfertilised plots were each sampled by two 2.5 x 15 cm soil cores in November 2019 and analysed as previously by Hills Laboratories.

Olsen P and sulphate were the only macronutrients lower than the Hills laboratory optimum threshold (Figure 9). Cobalt, as in the 2014 micronutrients, was lower than average with a weak, but not strong extractant, and molybdenum declined to lower than average (Figure 10).

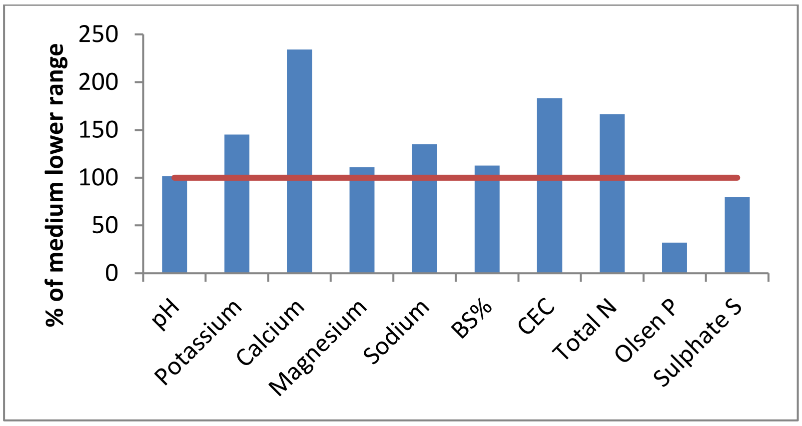


Figure 9. Soil macronutrient test values as a percentage of the lower optimum range threshold.

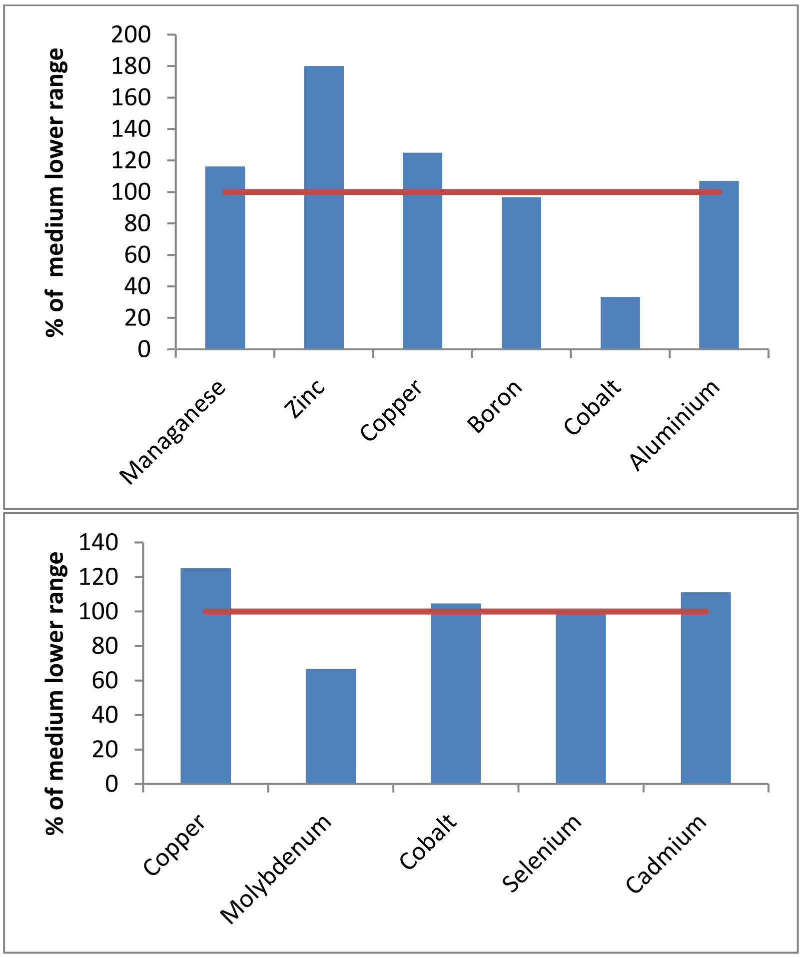


Figure 10. Soil micronutrient test values as a percentage of the lower optimum range threshold.

Topsoil chemistry in Canterbury was re measured on the shallow soil phase in August 2018 from four unfertilised plots by five 2.5 x 10 cm soil cores and analysed as previously by Hills Laboratories with carbon and nitrogen analyses by Eurofins Laboratories.

Soil pH dropped slightly to 6.4 (medium) and Olsen P also was lower (Figure 11).

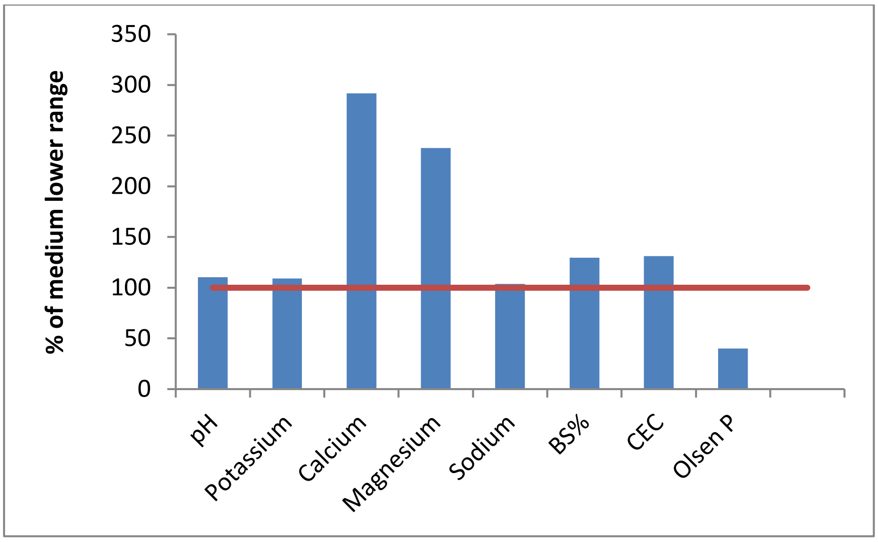


Figure 11. Canterbury topsoil chemistry, shallow soil phase, August 2018.

Soil carbon and nitrogen levels (Table 1) were optimal for N but low for carbon and therefore for the C: N ratio (Figure 12).

Table 1. Canterbury soil carbon and nitrogen levels.

Assay	Count	Average	Std. Error
Hot Water Carbon mg/kg	4	20	1.4
Organic Carbon mg/kg	4	1,473	94.2
Total Carbon %	4	3.4	0.05
Total Nitrogen %	4	0.33	0.005
C : N Ratio	4	10	0.000

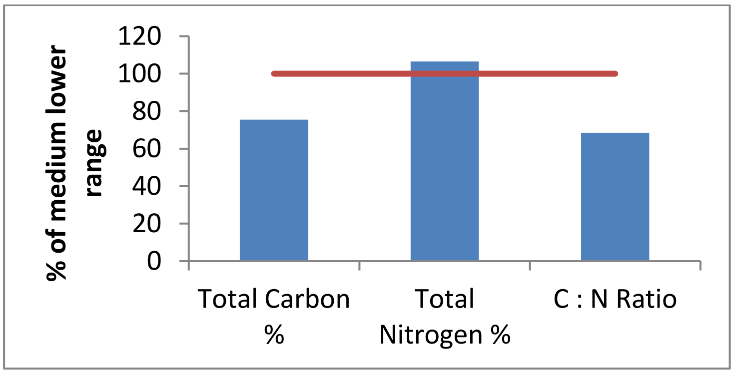


Figure 12. Canterbury topsoil C and N chemistry, shallow soil phase, August 2018.

1.4 Topsoil Microbiology

Soil microbiology in Southland was determined by visual microscopy from bulked 2.5 x 10 cm cores sampled in October 2014, February and March 2015 at the Soil Food Web laboratory, New Zealand. Soil microbiology at Canterbury was assessed from similar soil cores extracted in November 2018 by Microbial Laboratories Australia.

Total bacterial and fungal biomass at Southland decreased from spring to autumn due to summer drought, but microbial activity recovered with early autumn rainfall (Figure 13).

In Canterbury total biomass was lower than in Southland and fungi contributed the majority of soil microbial biomass (Figure 14). Duplicate assays indicate sample analytical variability.

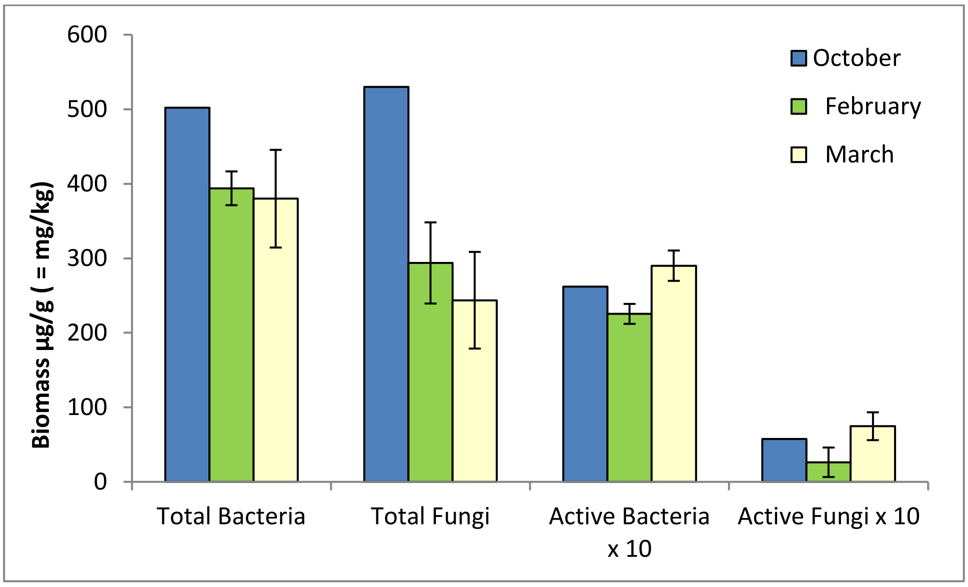


Figure 13. Southland total microbial biomass and activity.

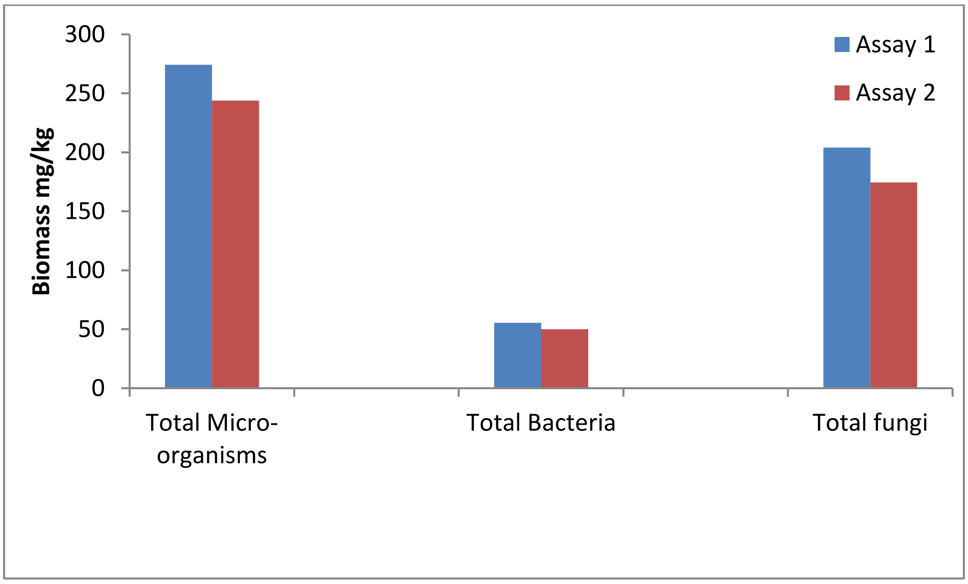


Figure 14. Canterbury total microbial biomass.

The composition of the major bacterial and eukaryote groups at Canterbury are shown in Figure 15. The biomass from all groups is ranked as good compared to reference levels for agricultural soils by Microbial Laboratories Australia.

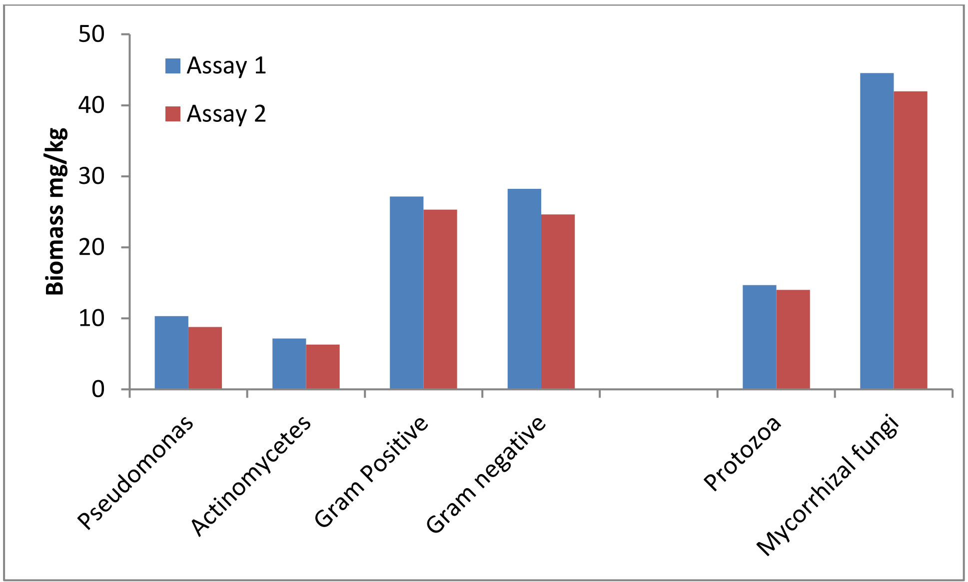


Figure 15. Left: bacterial biomass; Right: eukaryote biomass

Other site information is summarised in the initial trial establishment reports[4,5].

1.5 Experimental Design

In Southland, the experimental design for the 2016 - 2017 season was a randomised block with five blocks, five fertiliser/stimulant applications and five replications per fertiliser treatment. Fertilisers and stimulants were applied in twenty five 2 m^2 (1 x 2 m) plots separated by 0.5 or 1 m buffer strips with five nil fertiliser plots 10 m^2 (2 x 5 m) from companion Southern Humate trials, totalling 30 plots. Initial fertiliser & stimulant applications of Soil and Seed and fertilisers on the 23rd December 2016 were: nil application, calcium nitrate at 20 kg/ha, calcium nitrate plus Soil and Seed at 8 l/ha, Bio Marinus Fish Hydrolysate at 20 l/ha, and Fish Hydrolysate plus Soil and Seed at 8 l/ha).

Production was assessed by harvesting by rotary mower to 5-6 cm pasture height, herbage double weighed ± 0.5 g and sub samples taken from every plot for laboratory dry matter determination. Summer (16th February) and autumn (18th April 2017) production was measured.

[4] Espie, P.R. 2014. Southern Humate Trial Establishment October 2014. AgScience Contract Report, November 2014, 12 pp.

[5] Espie, P.R. 2017. Rotokaia Biological and Synthetic Fertiliser Trials Establishment and Summer Assessments. AgScience Contract Report, February 2014, 13 pp.

The trial was expanded in the 2017 – 2018 season to examine how different rates of BioAg Soil and Seed stimulant (S&S) affected production and also how BioAg stimulants interacted with urea. The experimental design was eight blocks, the same five fertilisers as previously but now with three replicates of 3, 6 and 9 l/ha of BioAg S&S giving 9 plots of S&S alone, nine plots of S&S plus calcium nitrate at 20 kg/ha, nine plots of S&S plus Bio Marinus Fish Hydrolysate at 20 l/ha, and eleven nil application plots, 5 plots of calcium nitrate at 15 kg/ha, totalling 47 plots. All plots were lightly pre-mown to a uniform 6 cm height in spring (11th October) and fertilisers were spray applied on the 11th October 2017, 25th November with 5 additional plots of S&S at 6 l/ha plus 60 kg/ha urea and on the 9th March 2018.

Steven Haswell (BioAg) and I visually scored pasture and clover growth a month after fertiliser application on the 16th November 2017 in five main classes (1 = low, light 3 = high dense, with two intermediate sub-classes), then the trial was mowed. It was assessed by electrical capacitance (GrassMaster Pro probe) and mowing in summer (6th December) and autumn (9th and 27th March 2018).

In Canterbury a randomised block design was used, stratified by the two soil phases, with four blocks, six fertilisers and four replications per fertiliser treatment per phase in both seasons. Fertilisers and stimulants were applied in twenty-four 10 m^2 (2 x 5 m) plots separated by 1 m buffer strips on each of the shallow and deep soil phases. An additional eight nil fertiliser and four urea plots (10 m^2) were available from a synchronous surrounding Southern Humate companion trial on each phase.

Soil and Seed soil stimulant and fertilisers were applied on the 9 December 2016 on the deep soil phase and on the 17th December on the shallow soil phase. Treatments consisted of: no fertiliser application; urea at 30 kg/ha, calcium nitrate at 20 kg/ha, calcium nitrate plus BioAg Soil and Seed bio-stimulant at 8 l/ha, Soil and Seed bio-stimulant alone at 8 l/ha, Soil and Seed bio-stimulant at 8 l/ha plus fish hydrolysate at 20 l/ha. Soil and Seed was applied as a folia application at 8 ml per plot and fish hydrolysate at 20 ml per plot in 150 ml of water. Fertilisers were reapplied in March 2017 with BioAg Root and Shoots foliar bio-stimulant application used subsequently in place of Soil and Seed.

In the 2017 -2018 season the trial was mown to a uniform height in spring (10th October 2017) and the same fertilisers were applied on the 29th October 2017 (Soil and Seed 8 l/ha), replaced subsequently by Roots & Shoots at 1.5 l/ha on the 13th November, 23rd December, 9th February and 15th March 2018. Urea was increased to 50 kg/ha in March (leaching trial).

Seasonal pasture production was determined in the same way as at Southland. Pasture growth was visually scored a month after fertiliser application on the 11th January 2017 in five classes (1 = low, 5 = high) and by electrical capacitance (GrassMaster Pro probe) on 27th October, 14th November, 29th November, 22nd December 2017, 7th February and 14th March 2018. Pasture was double cut with rotary mowers on 30 November, 14th March and 17th April 2018. Herbage was weighed ± 2.5 g, sub-sampled for dry matter determination, and returned to plots (Figure 16). The statistical package R was used for production analysis.

Figure 16. Canterbury trial site.
Upper: Pasture production assessment by visual scoring, shallow soil phase
Lower: Pasture assessment by mowing, deep soil phase. Initial high height cut followed by a second cut then harvested herbage evenly returned after weighing.

Results

2.1 Southland pasture production 2016 - 2017

In February 2017, two months after base/initial fertiliser/stimulant application (55 days), calcium nitrate increased pasture production by 53% compared with unfertilised pasture (Figure 17). Roots and Shoots addition to calcium nitrate increased yield by 30 % compared with calcium nitrate to 83% more than unfertilised pasture. Fish Hydrolysate increased yield by 58% and of Soil and Seed addition increased yield by a further 26% (Figure 17).

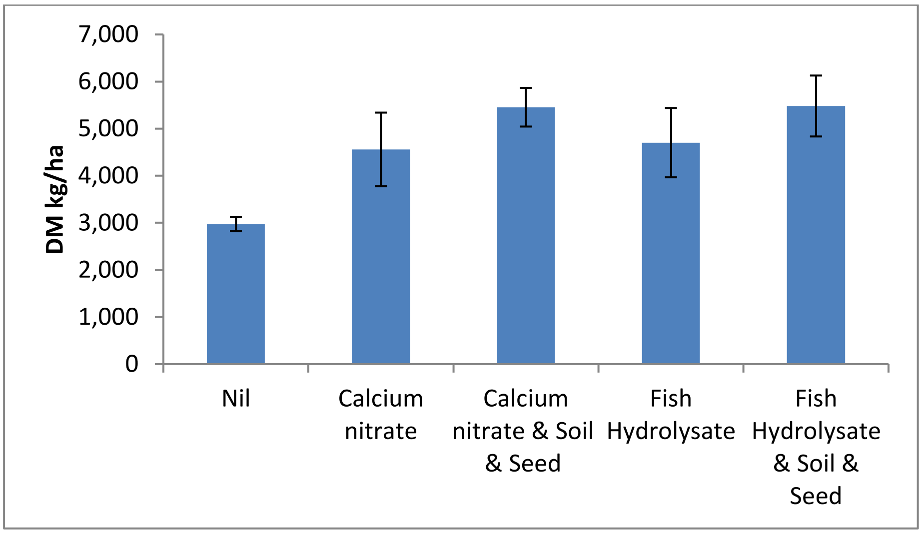


Figure 17. Fertiliser effect on pasture yield ± standard error of the mean (SEM).

In April 2017, four months after fertiliser/stimulant application (116 days), these production responses persisted, though they were understandably smaller (Figure 18).
Calcium nitrate increased pasture production by 33% and addition of BioAg stimulant by a further 8%. Hydrolysate increased yield by 39% and addition of BioAg stimulant by a further 3%.

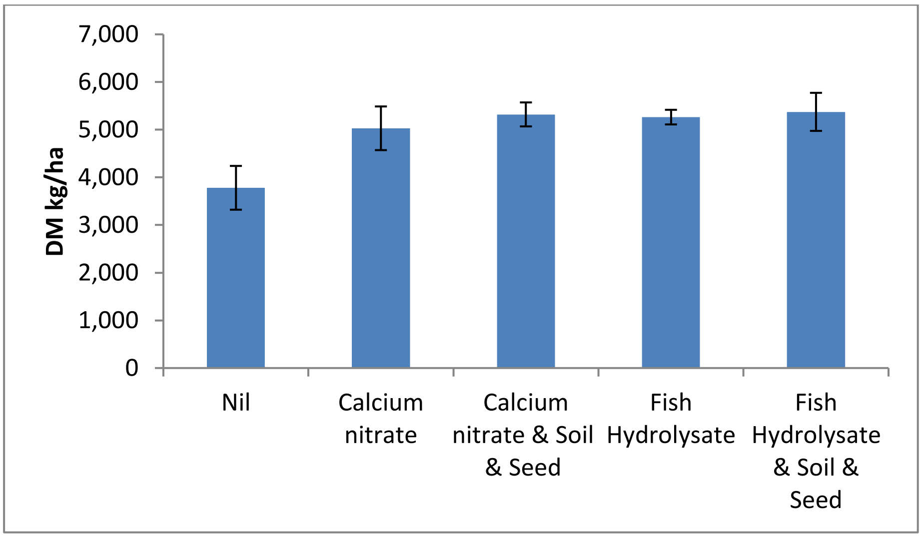


Figure 18. Fertiliser effect on pasture yield ± SEM.

The average effect of BioAg stimulants on pasture production in the 2016 – 2017 season in Southland, expressed as the percentage increase compared with unfertilised pasture, varied between 14.5% and 19% (Table 2).

Fertiliser and stimulant application significantly affected yield ($P < 0.001$). Compared with nil fertiliser, calcium nitrate ($P<0.05$), calcium nitrate plus BioAg R&S ($P<0.01$), fish ($P < 0.001$).and fish plus BioAg R&S ($P < 0.001$) all significantly increased yield.

Table 2. Fertiliser effect on pasture production relative to unfertilised pasture 2016 - 2017.

Fertiliser	% Increase	% Increase due to Soil & Seed
Nil	0	
Calcium nitrate	43	
Calcium Nitrate & BioAg	62	19
Fish Hydrolysate	48.5	
Fish Hydrolysate & BioAg	63	14.5

2.2 Southland pasture production 2017 – 2018

The vigour of spring growth in the 2017 – 2018 growing season is shown by the regrowth in November from pre- mowing before trial assessment on the 11th October (Figure 19).

Figure 19. Spring growth on BioAg trial plots (centre) seen in he strip on the right hand side of the plot. Herbage returned after visual assessment, 16 November 2017.

Soil and Seed bio-stimulant increased pasture growth by 70% and clover cover by 50% more than the lowest yielding fish hydrolysate plots (Figure 20). Soil and Seed alone notably gave similar pasture production as when combined with 30 kg/ha urea and increased production by 18% when combined with 20 kg/ha calcium nitrate compared with 50 kg/ha calcium nitrate. The response to calcium nitrate und urea shows nitrogen deficiency limited spring growth and therefore suggests that Soil & Seed may also affect N supply.

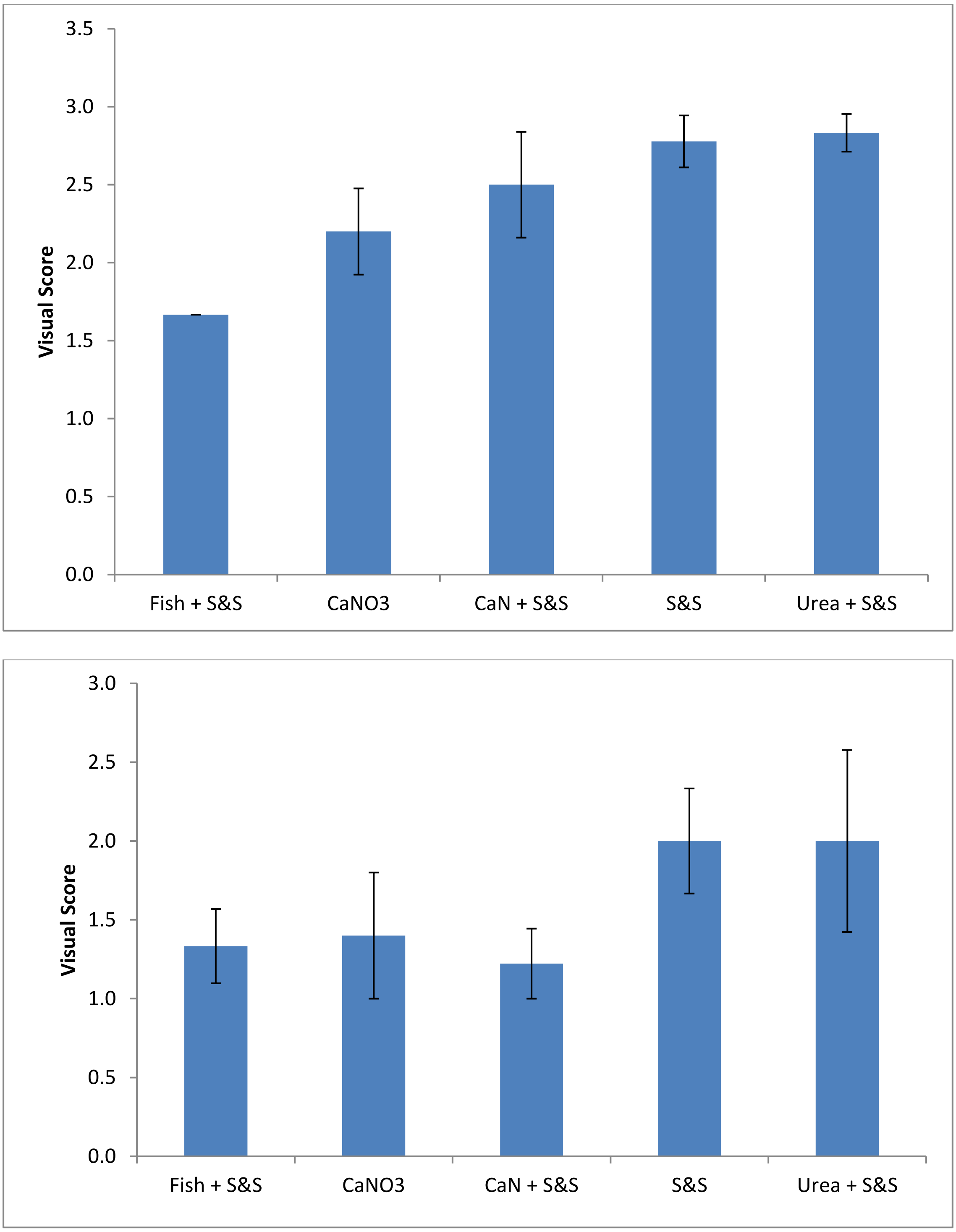


Figure 20. Fertiliser effect on spring pasture yield (upper) and Clover cover (lower) ± SEM.

Increasing the application rate of Soil and Seed increased pasture production and possibly clover cover (Figure 21).

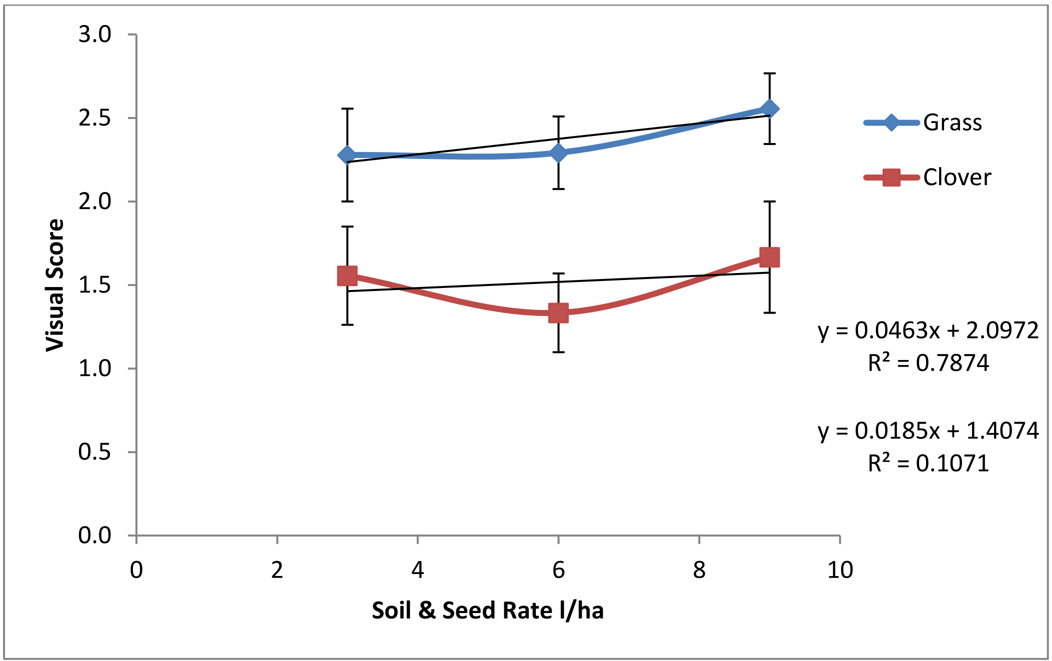


Figure 21. Effect of Soil and Seed application rate on pasture and clover production ± SEM.

In early summer, on the 6th December 2017, 20 days since the previous harvest, Root & Shoots and urea increased production by 38% from unfertilised pasture (Figure 22). Root & Shoots alone increased production by 16% and gave a 1% further increase when added to calcium nitrate. Calcium nitrate alone increased yield by 25% and R&S with fish hydrolysate by 10%.

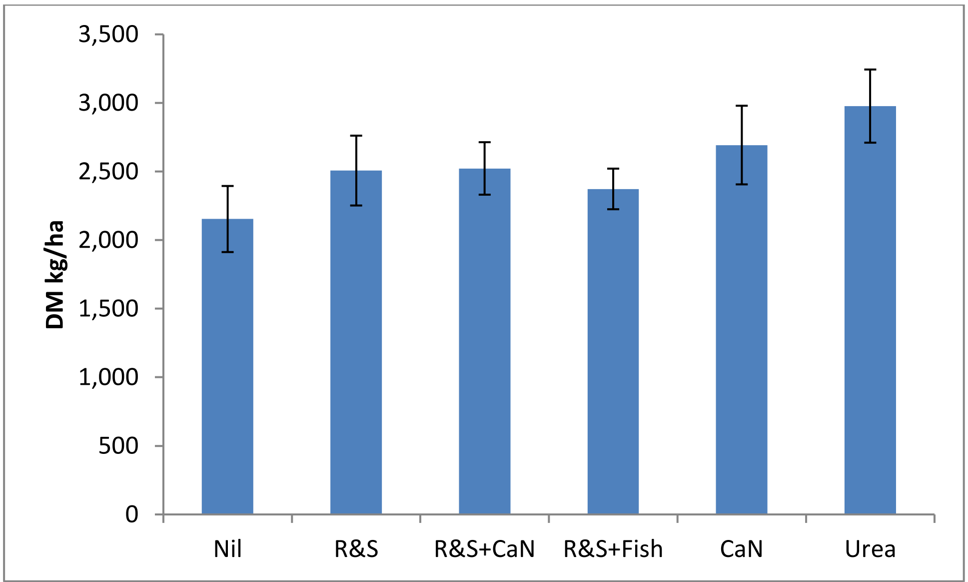


Figure 22. Effect of Roots and Shoots application rate on pasture production ± SEM.

BioAg stimulants with calcium nitrate appeared to increase production at the highest S&S base application rate but not with fish hydrolysate (Figure 23). The low replication of three plots per individual fertiliser rate and experimental variability limit more definitive analysis of individual fertiliser responses (Figure 23).

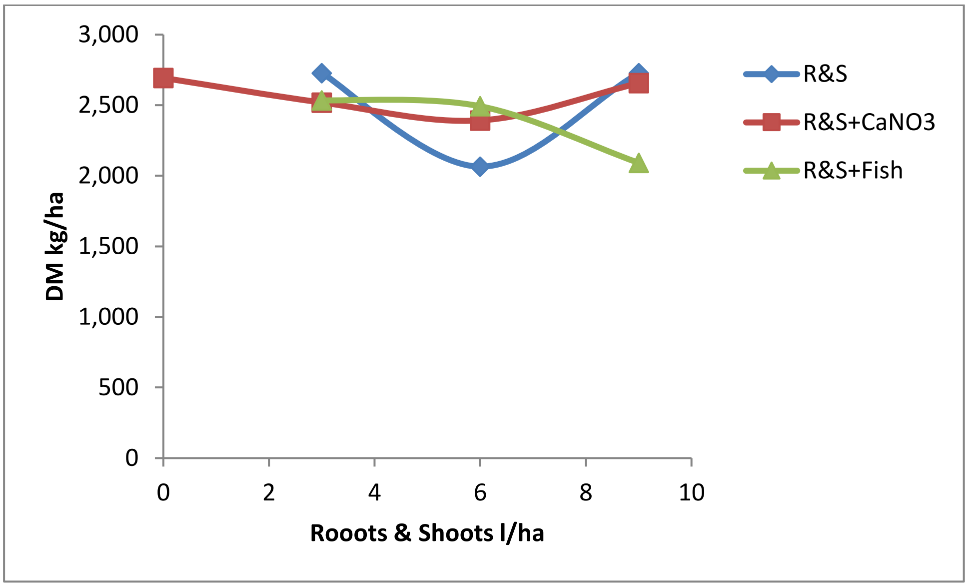


Figure 23. Pasture response to Roots and Shoots application rates effect, 6th December.

In autumn, March 2018, urea and calcium nitrate gave the greatest increase in production, up to 21% greater than unfertilised pasture, while Roots and Shoots did not lift production (Figure 24).

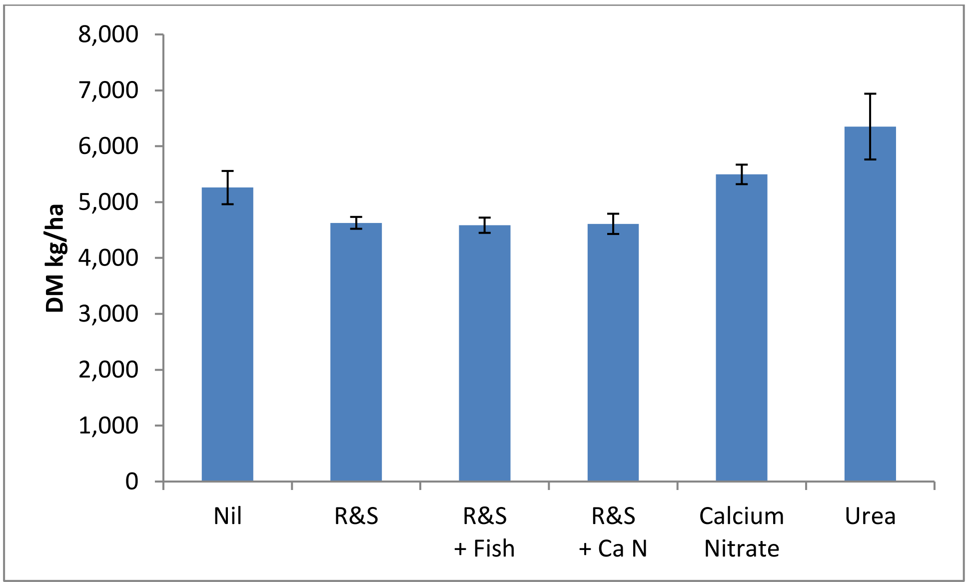
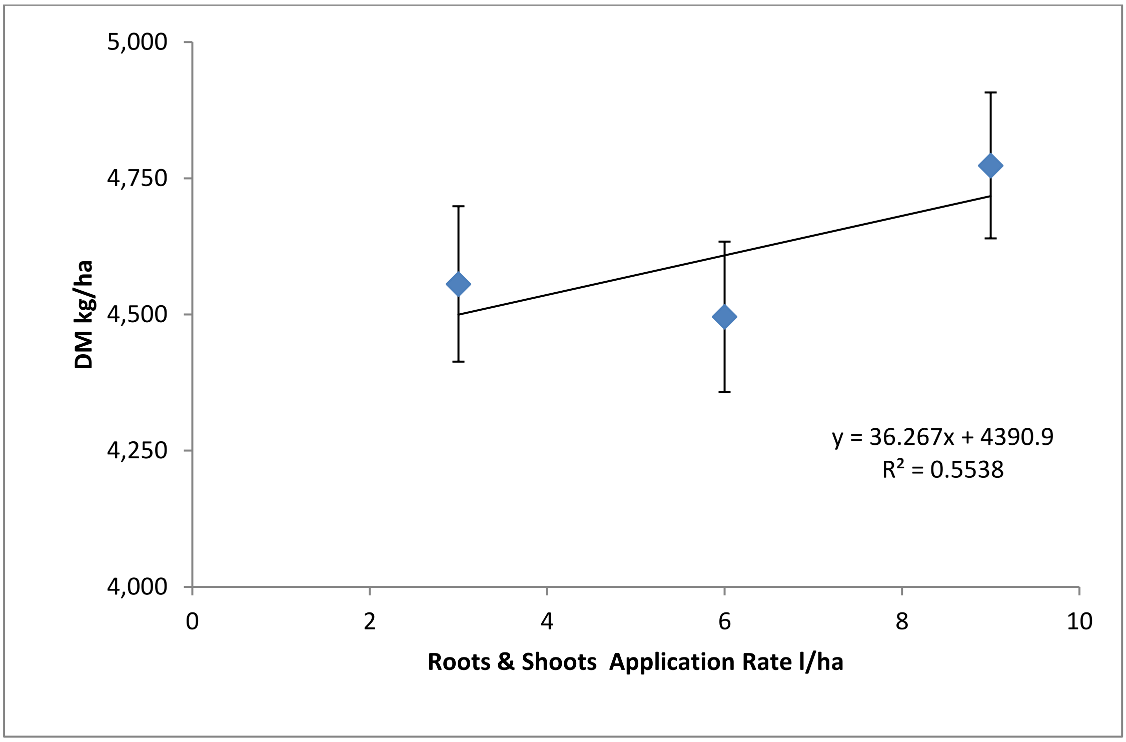


Figure 24. Effect of fertiliser application on pasture production ± SEM.

Roots and Shoots lifted production only at the highest S&S base application rate when averaged across all applications (Figure 25). Roots & Shoots with calcium nitrate and with fish hydrolysate appeared to increase production at the highest S&S application rate but not Roots & Shoots alone.



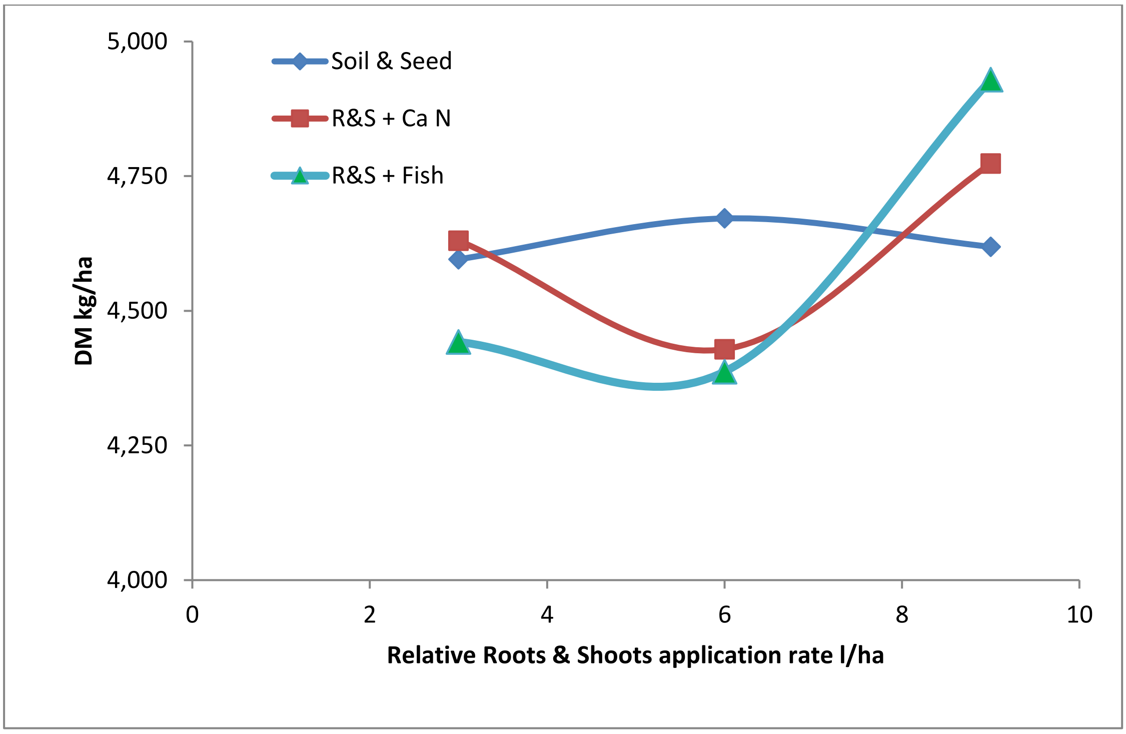


Figure 25. Effect of Roots & Shoots application rate on pasture production ± SEM.

Over the 2017 – 2018 growing season urea with Soil and Seed gave the greatest production, a 33% increase from unfertilised pasture (Table 3). Calcium nitrate increased production by 11% and Roots and Shoots by 12%, but no fertiliser effect was significant (P = 0.457).

Table 3. Effect of fertiliser on percentage difference in production relative to unfertilised pasture 2017 – 2018 growing season.

Fertiliser	% Increase
Nil	0
Roots & Shoots	12
R&S + Fish Hydrolysate	0
R&S + Calcium nitrate	1
Calcium nitrate	11
S&S + Urea	33

Pasture growth rate in the 2017 - 2018 season appeared to be related to rainfall, particularly with the rise in production in autumn (Figure 28).

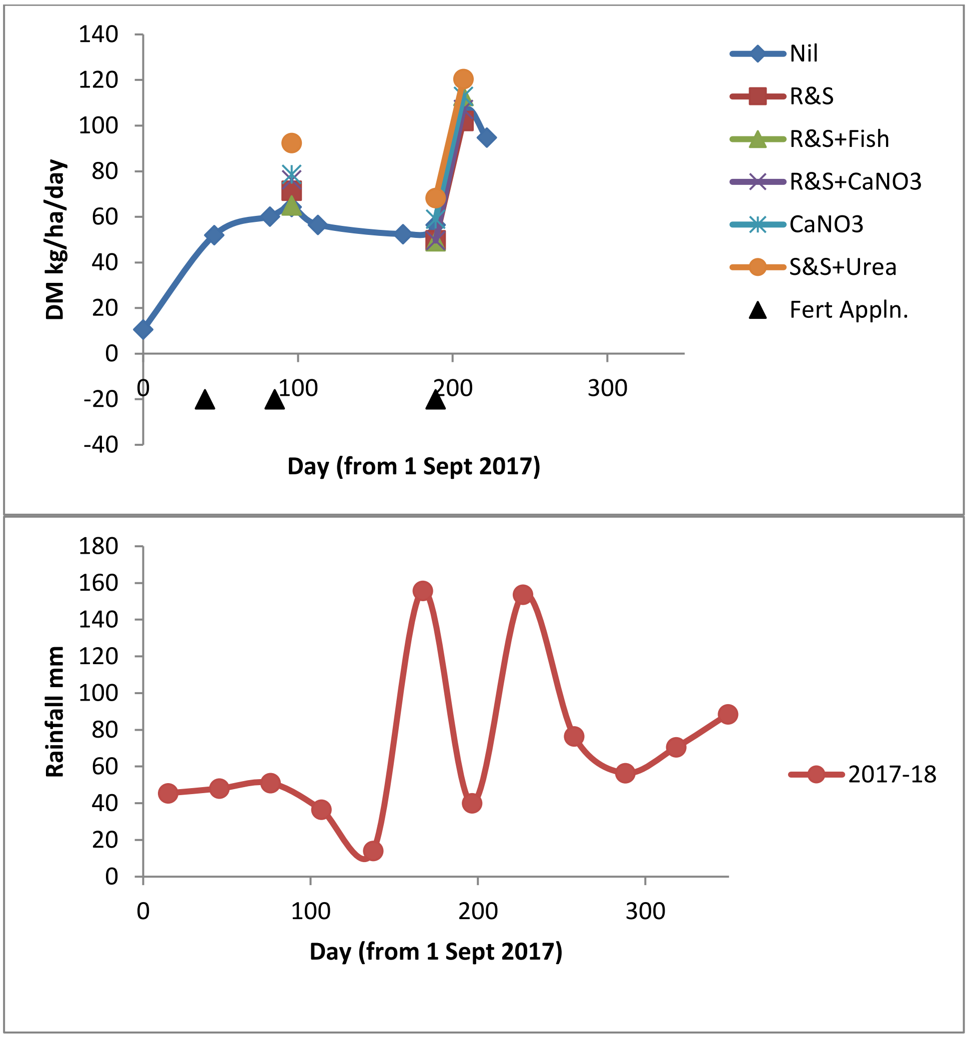


Figure 26. Fertiliser effect on seasonal growth rate (upper) and rainfall (lower). Additional nil fertiliser responses included from the companion Southern Humate trial.

2.3 Canterbury pasture production 2016 - 2017

In January 2017 visual ranking of pasture growth on the deep soil phase using a 1 to 5 (low - high) scale showed urea gave the greatest response (Figures 26, 27). Calcium nitrate increased pasture production by 11% and addition of Soil and Seed yield by a further 11%. Hydrolysate increased yield by 11% with no further gain by addition of Soil and Seed.

Figure 26. Canterbury deep soil phase pasture growth.

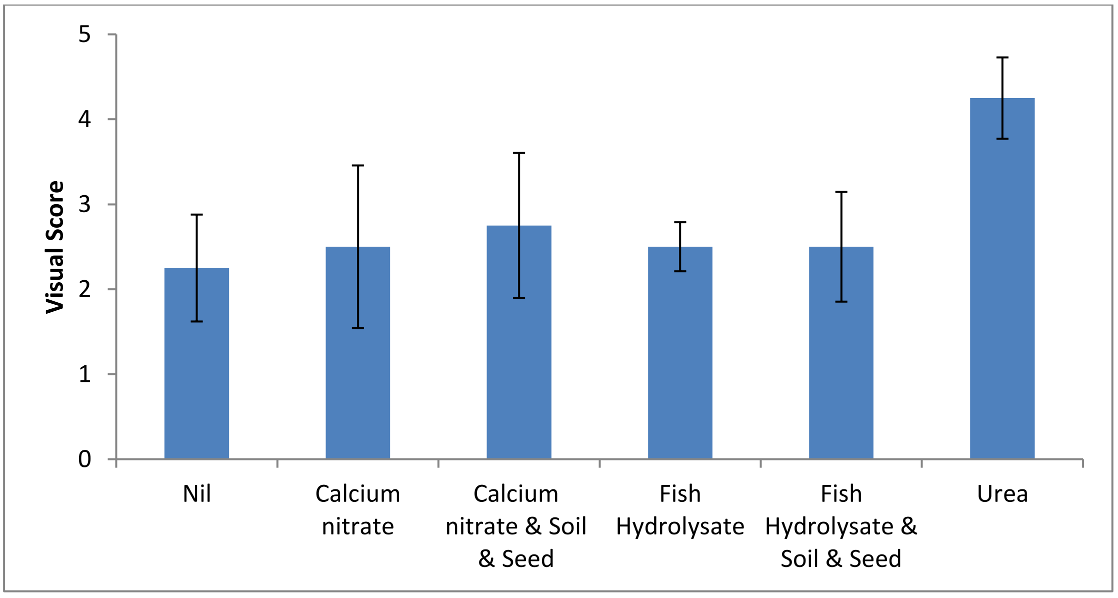


Figure27. Fertiliser effect on pasture yield 13 January 2017 ± SEM.

When pasture response is assessed on the basis of units of N supplied, not accounting for possible field volatilisation losses, calcium nitrate gave greater N production efficiency (Figure 28). The response to urea in Figures 29 and in all subsequent graphs is related to the higher application rate and N content of urea compared with calcium nitrate.

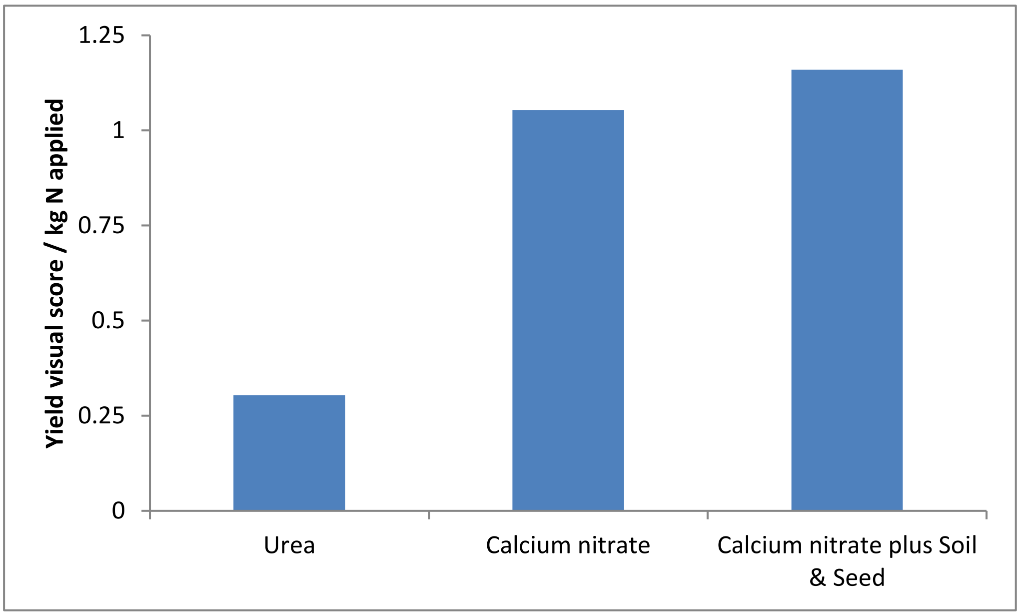


Figure 28. Pasture response per unit of fertiliser N supplied.

In February, cows broke into the shallow phase site, on the deep phase calcium nitrate increased production by 17% and addition of BioAg stimulants yield by a further 32%. Hydrolysate increased yield by 36% by a further 14% with BioAg stimulants (Figure 29).

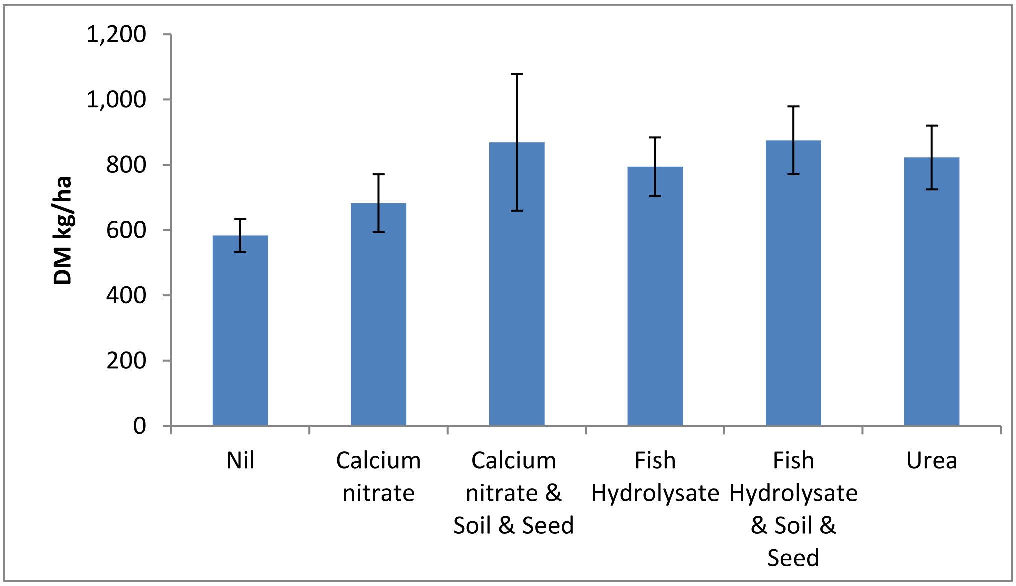


Figure 27. Fertiliser effect on pasture yield deep soil phase ± SEM.

By the 16th March the difference between treatments was far smaller (Figure 30). Calcium nitrate increased yield by 6%, which was very similar to urea at 7%. Addition of BioAg stimulants had no further effect. Fish hydrolysate yield was 4% lower than unfertilised pasture but addition of BioAg stimulants increased yield by 24%.

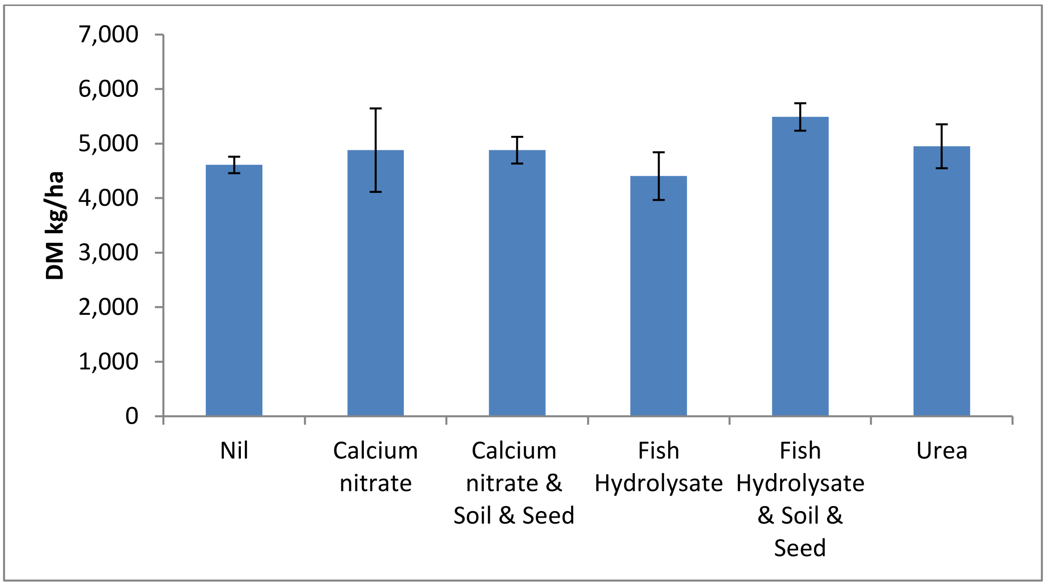


Figure 30. Fertiliser effect on pasture yield deep soil phase ± SEM.

By early winter, in May, calcium nitrate increased yield by 10% and with Soil and Seed by 11% (Figure 31). Hydrolysate increased yield by 17% and by a 15% increase with Soil and Seed. Urea increased yield by 23%.

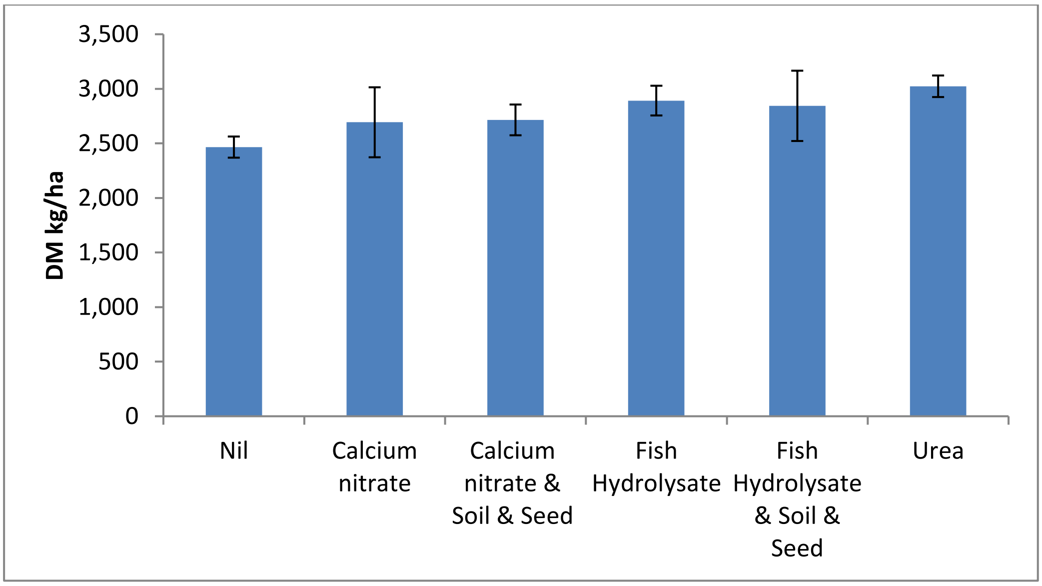


Figure 31. Fertiliser effect on pasture yield ± SEM.

The shallow soil phase was not measured in this trial due to unintended grazing immediately after initial fertiliser application in December 2016. Nevertheless a small selection of 12 plots was also harvested on the shallow soil phase in May to see if there were any indications of long term fertiliser effects (Figure 32). Calcium nitrate increased yield by 12%, hydrolysate by 6% and hydrolysate plus Soil & Seed by 18%. Urea raised production by 42%.

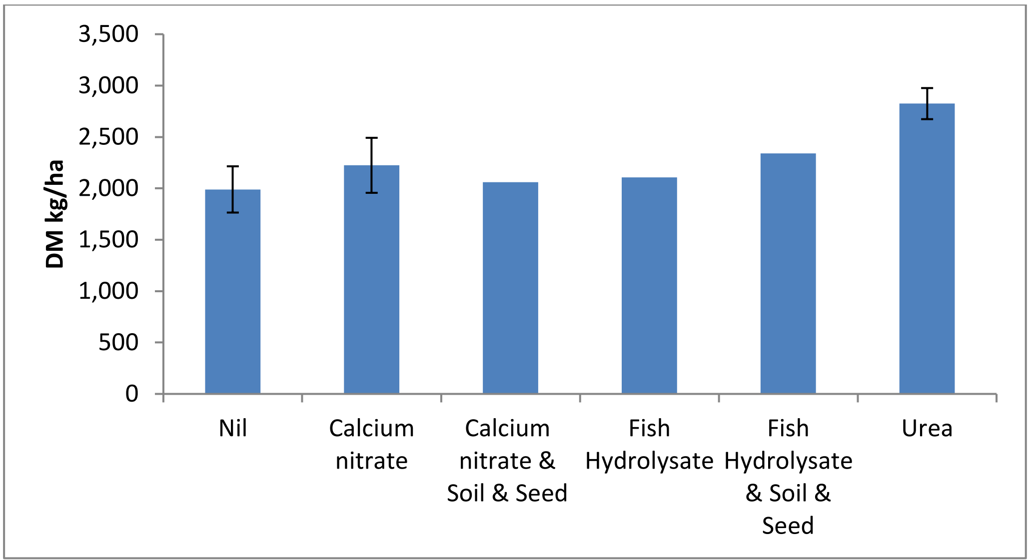


Figure 32. Fertiliser effect on pasture yield ± SEM.

The seasonal trend of fertiliser response over the 2017-18 season is shown relative to production on unfertilised pasture on the deep soil phase (Figure 33). Note that the January values (Day 132) are derived from visual scores, not dry matter determinations, and are presented only to indicate relative differences.

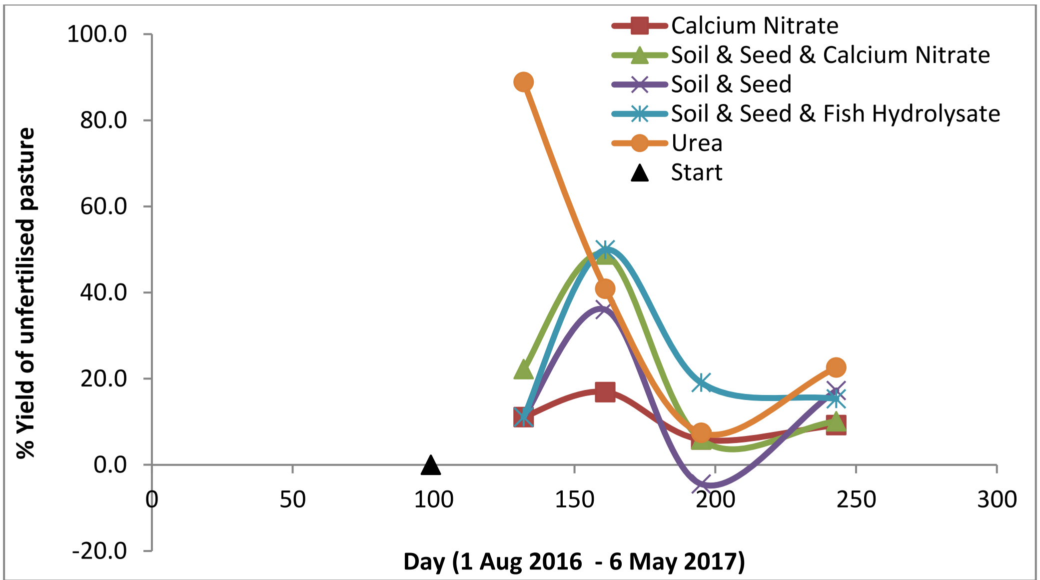


Figure 33. Fertiliser effect on pasture production April 2016 – February 2017 ± SEM.

Seasonal effects of fertiliser on total dry matter production are shown in Table 4.

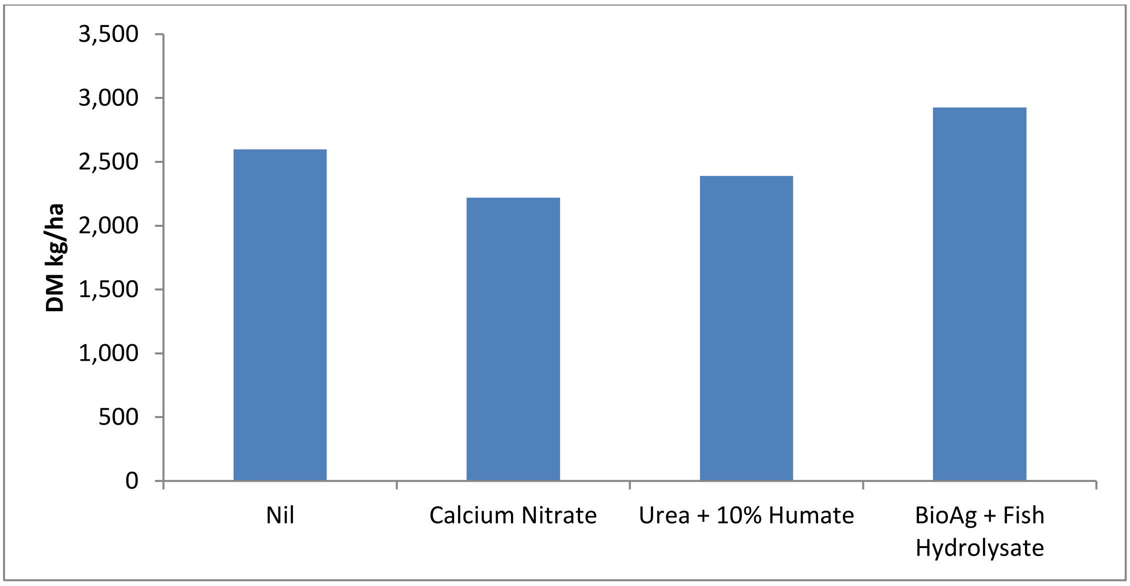
Table 4. Fertiliser effect on pasture production deep soil phase 2016- 2017 growing season.

Fertiliser	% Increase	% Increase due to Soil & Seed
Nil	0	
Calcium Nitrate	7.8	
Calcium Nitrate & Roots & Shoots	10.5	2.7
Fish Hydrolysate	5.6	
Fish Hydrolysate & Roots & Shoots	20.2	14.6
Urea	14.9	

Statistical analysis of the harvested dry matter during both growing seasons showed no significant difference in pasture production between fertiliser applications (ANOVA $P = 0.99$). There was also no statistically significant difference using the combined measured and capacitance estimated DM.

2.4 Canterbury pasture production 2017 - 2018

Pasture growth over winter ranged between 1,806 – 2,906 kg/DM/ ha with BioAg stimulants and fish hydrolysate giving the greatest production (Figure 34a). In spring (27th October 2017), seven months after the final autumn fertiliser application in March, BioAg stimulants increased growth on the shallow soil phase by 20% and by 17.5% when combined with fish hydrolysate (Figure 34b). Other applications were similar to nil fertiliser. The deep soil phase had been grazed by cows and not assessed.



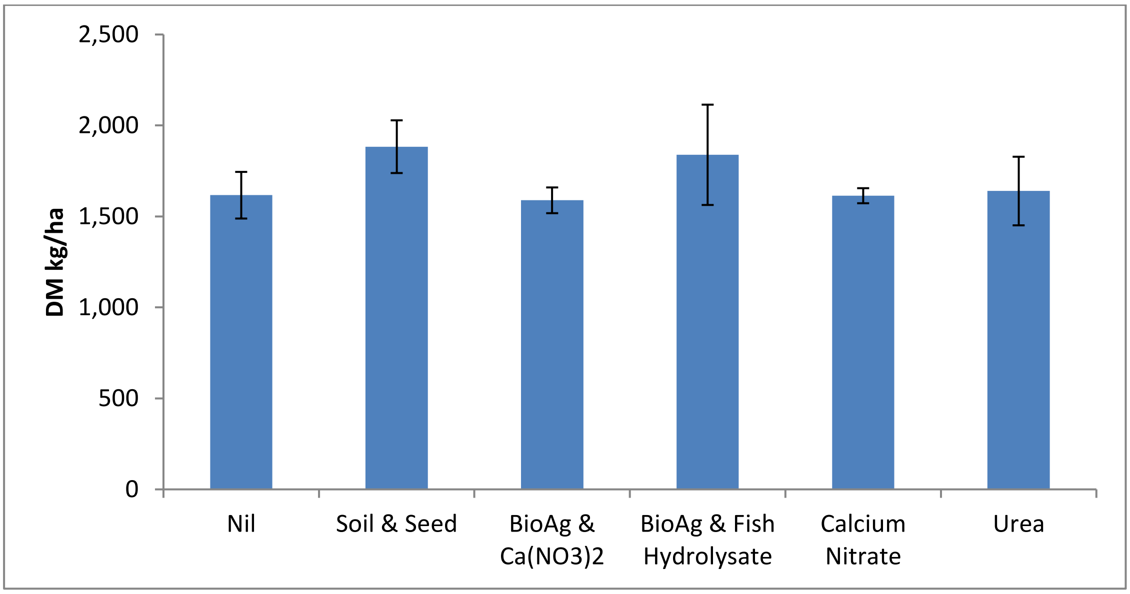


Figure 28. a) Fertiliser effect on winter yield from five 1m^2 samples on the 10th October (upper) and b) in spring on the 27th October (lower), shallow soil phase ± SEM.

Pasture production was estimated two ways: by visual ranking and by an electrical capacitance pasture meter. Visual estimation of spring pasture production was broadly correlated with capacitance estimates of dry matter production, but with wide variability between plots (Figure 35).

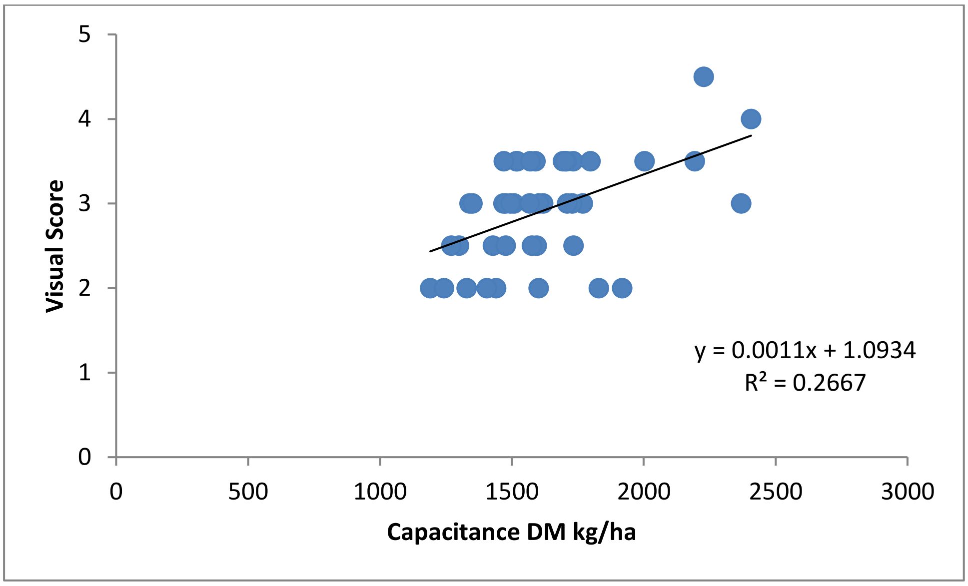


Figure 35. Visual score and electronic capacitance estimated DM, shallow soil phase, 27th October. R^2 statistic shows closeness of fit between 0 – 1 (0 = no relationship, 1 = perfect fit).

In late spring- early summer on the 14th November, 3 weeks after fertiliser application, capacitance and visual estimates of pasture production were more closely related, though still with considerable variability (Figure 36).

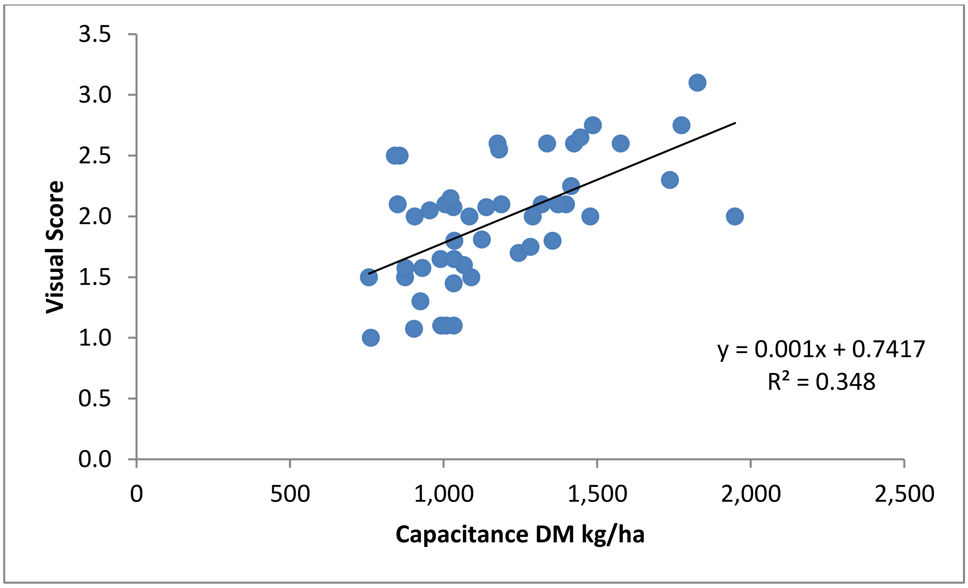
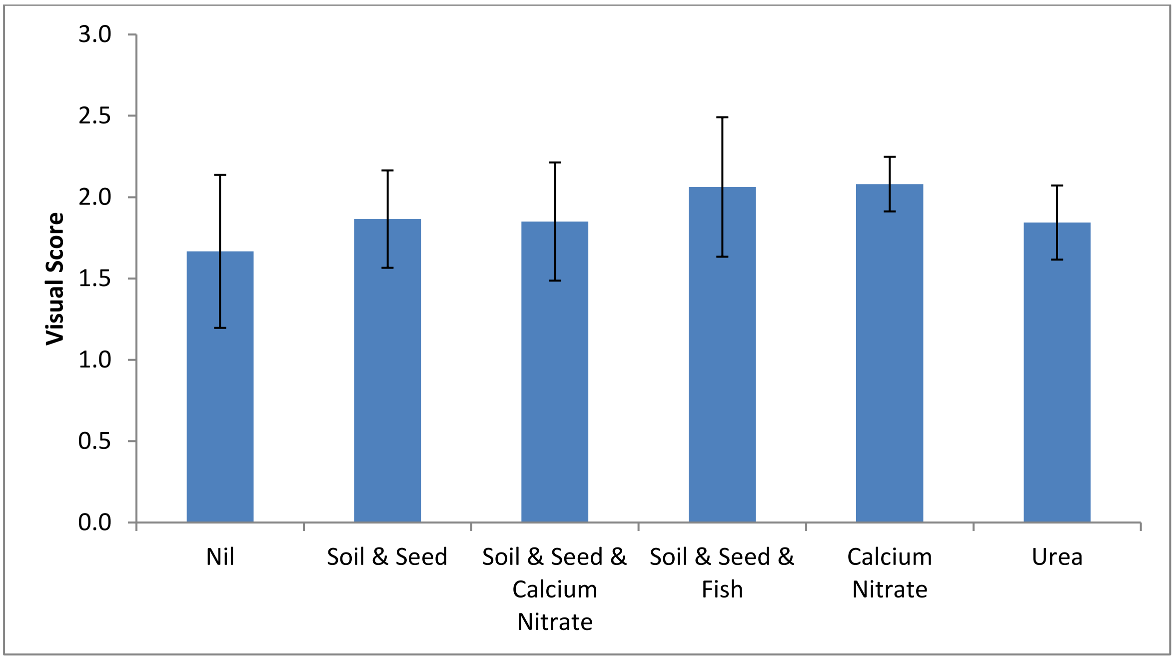


Figure 36. Relationship between visual score and electronic capacitance estimated DM, deep soil phase, 14th November.

Capacitance and visual estimates of pasture production on the deep soil phase on the 14th November, given this variability, unsurprisingly showed quite different responses to fertilisers (Figure 37). Both visual and capacitance showed similar production with Soil and

Seed to increases of 13% and 12% respectively, but agreement between the averages for the other fertilisers differed by up to 27%.



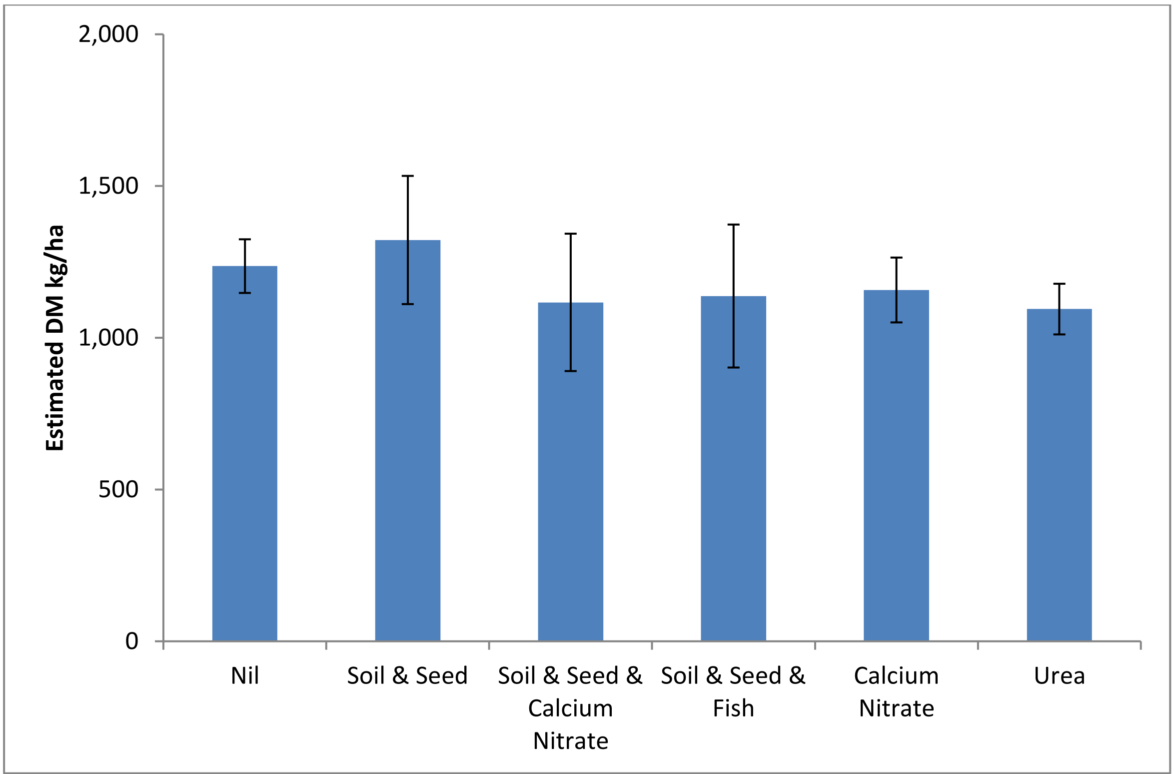


Figure 37. Visual (upper) and electronic capacitance (lower) estimates of fertiliser effect on deep soil phase pasture production ± SEM.

On the shallow soil phase only visual scoring estimated production. There was a 53% pasture response to Soil and Seed, 16% to Soil and Seed plus fish hydrolysate and 15 % to calcium nitrate (Figure 38).

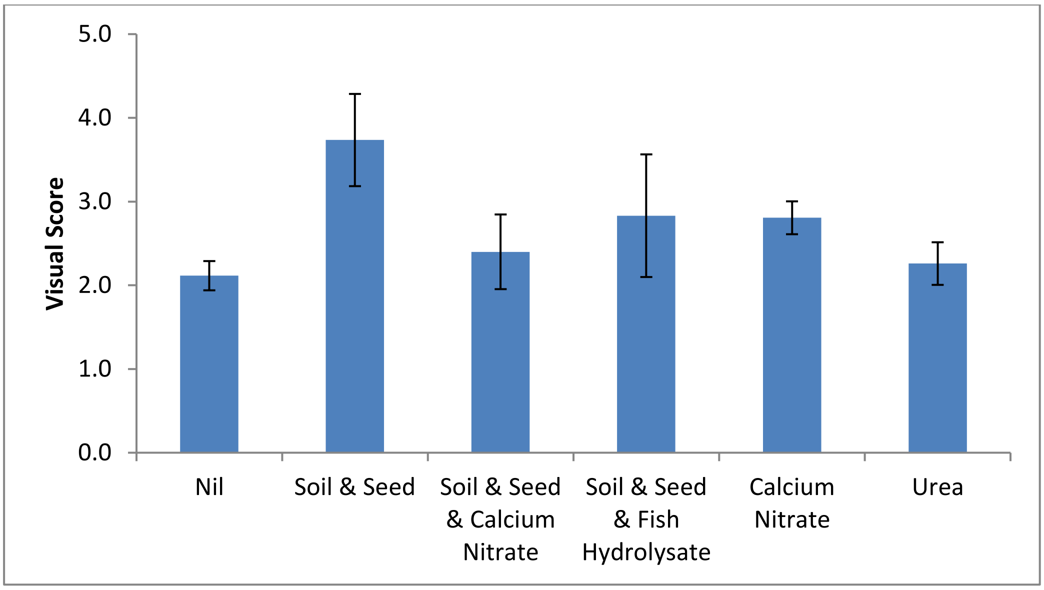


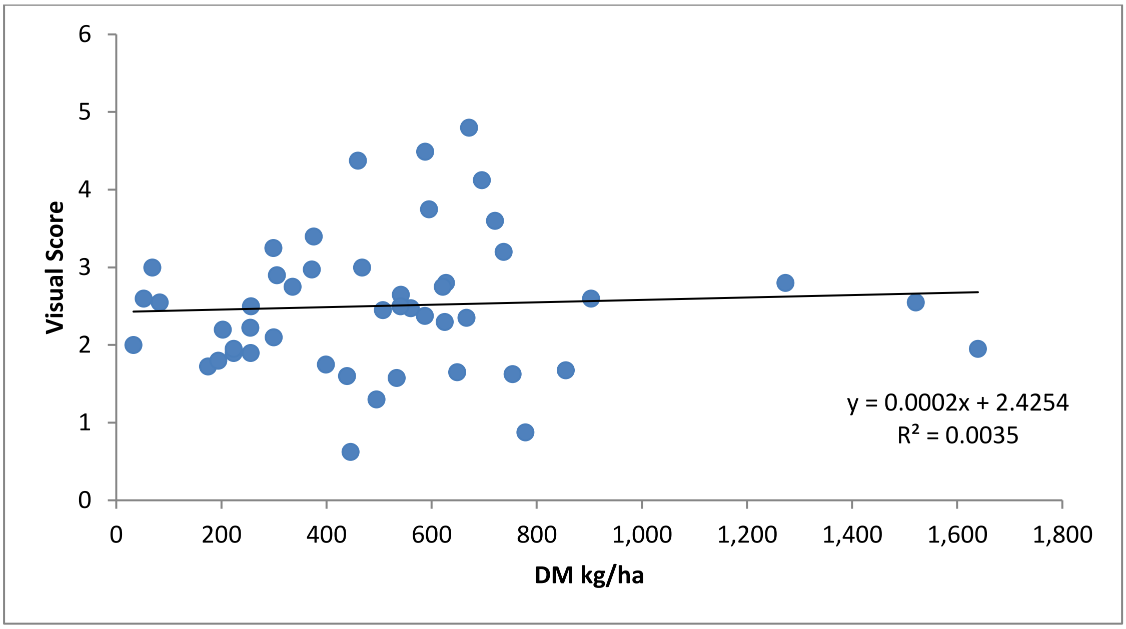
Figure 38. Visual estimates of fertiliser effect on pasture production ± SEM.

Two weeks later, on the 30th November, capacitance and visual measurements were also affected by variable patch production (Figure 39). Visually these were estimated as a percentage of the plot, scored separately, and combined into a total estimate. Twenty capacitance readings, collected in a regular sampling grid pattern, were averaged per plot.

Figure 39. Capacitance measurement of pasture variability, shallow phase, 30th November.

Visual estimates of pasture production on the shallow soil were very poorly related to direct measurements of dry matter production, possibly due to underestimation of production on the highest yielding plots (Figure 40).

Capacitance estimation was better. Capacitance estimated DM correlated reasonably well with measured DM on the deep phase, as shown by the higher statistical R^2 values measuring the closeness of plots to the overall trend, but not on the shallow phase where outliers reduced the fit (Figure 40).



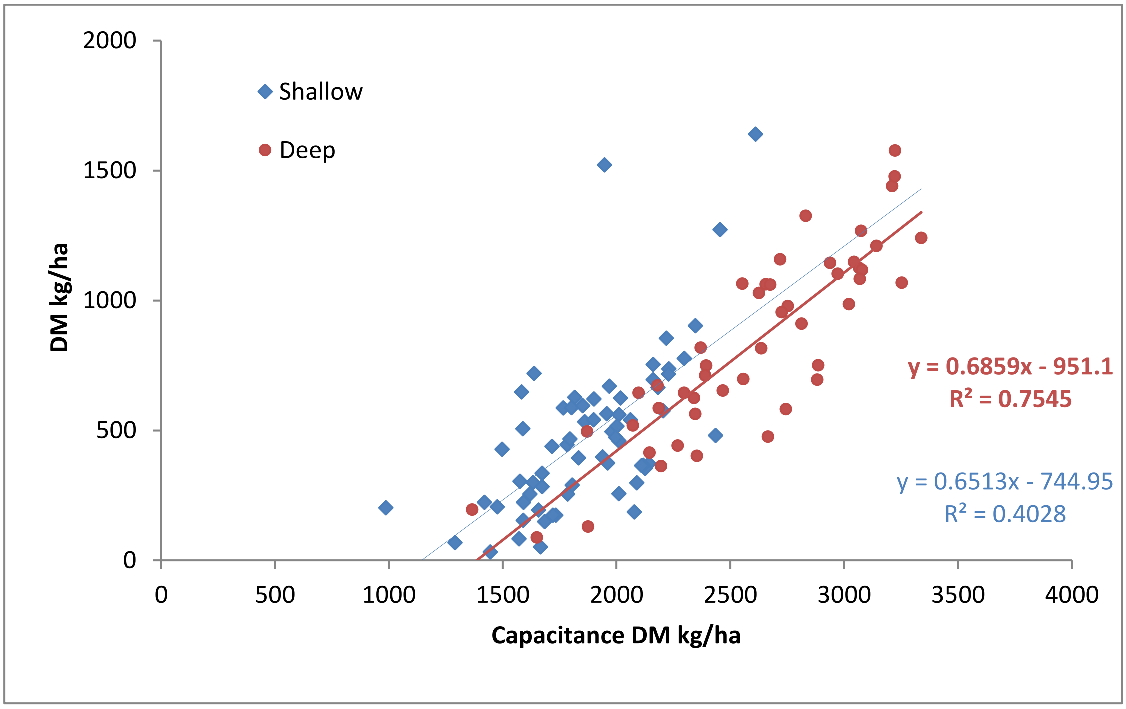
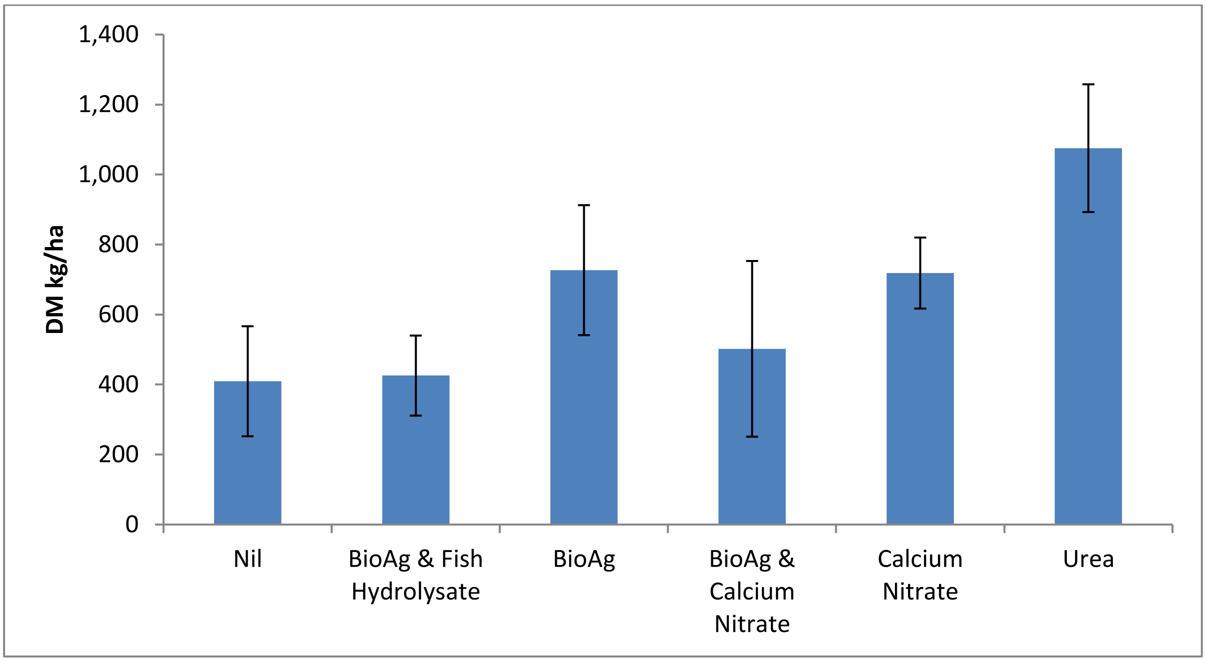


Figure 40. Upper: Visual and capacitance estimation of pasture DM production

By early summer (28th November) urea increased measured DM production on the deep soil phase by 163%, BioAg stimulants by 78% and calcium nitrate by 75% (Figure 41a). On the shallow soil phase urea increased production by 90%, BioAg stimulants by 68%, BioAg stimulants and fish hydrolysate by 65% and calcium nitrate by 45% (Figure 41b).



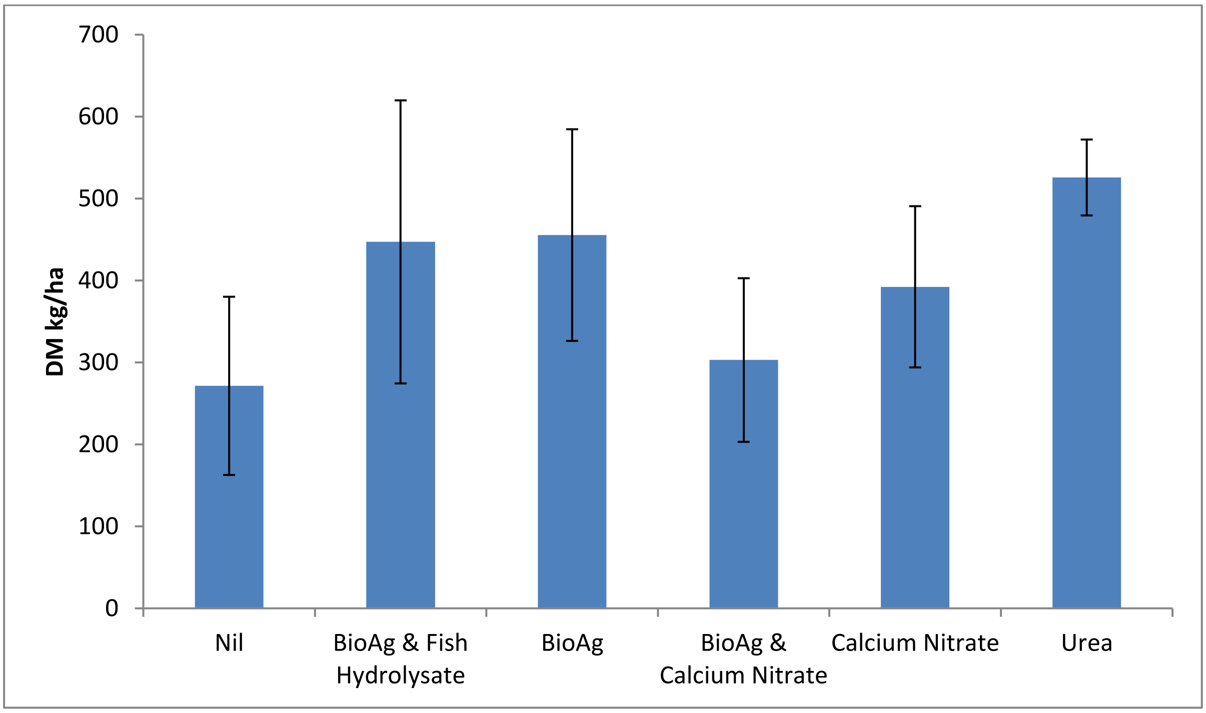


Figure 41. Effect of fertiliser on pasture production on a) deep soil phase (upper) and b) shallow soil phases (lower) ± SEM.

By mid-summer (22 December) there was little difference in capacitance estimates of production between unfertilised pasture and any fertiliser applications on the shallow or deep soil phases (Figure 42). Only urea and calcium nitrate on the deep soil phase gave a small increase, 15% and 4% respectively.

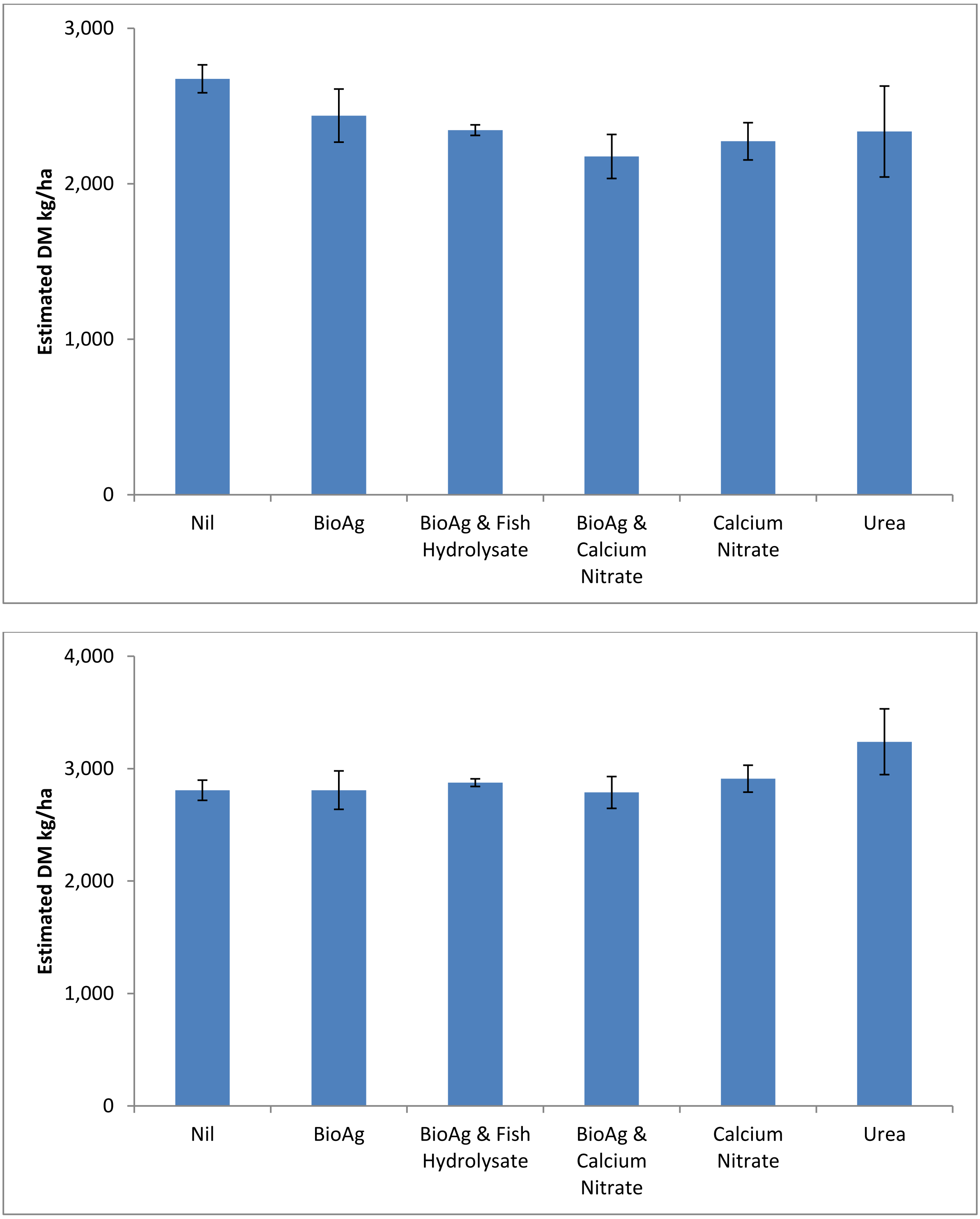
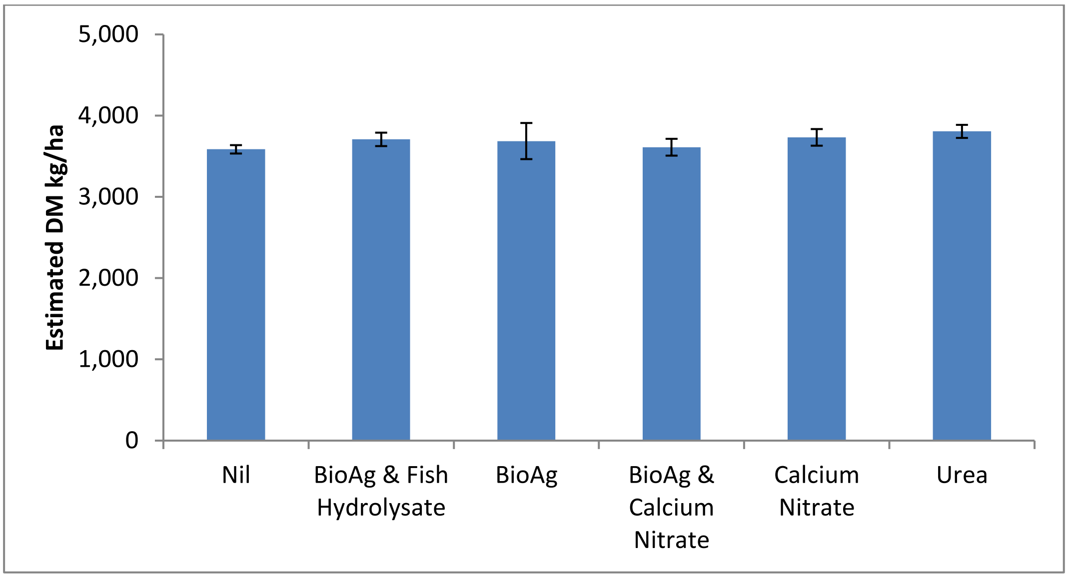


Figure 42. Effect of fertiliser on estimated pasture production on shallow (upper) and deep soil phases (lower) ± SEM.

By late-summer (7 February 2018) there was still very little difference in capacitance estimates of production between unfertilised pasture and any fertiliser applications on the shallow or deep soil phases (Figure 43). Urea, calcium nitrate, BioAg stimulants and BioAg stimulants with fish hydrolysate on the shallow soil phase gave small increases between 3% - 6%.



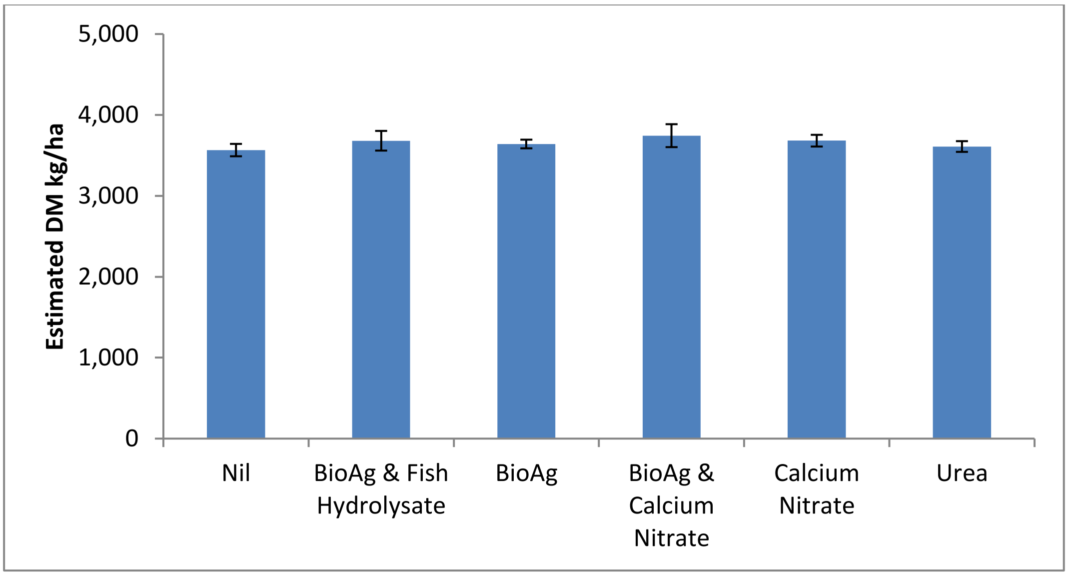


Figure 43. Effect of fertiliser on estimated pasture production on shallow (upper) and deep soil phases (lower) ± SEM.

Cows broke into the deep soil site so only the shallow soil phase was assessed in early Autumn (15 March 2018). Capacitance estimation of pasture production on the shallow soil were only very weakly related to direct measurements of dry matter production, (Figure 44).

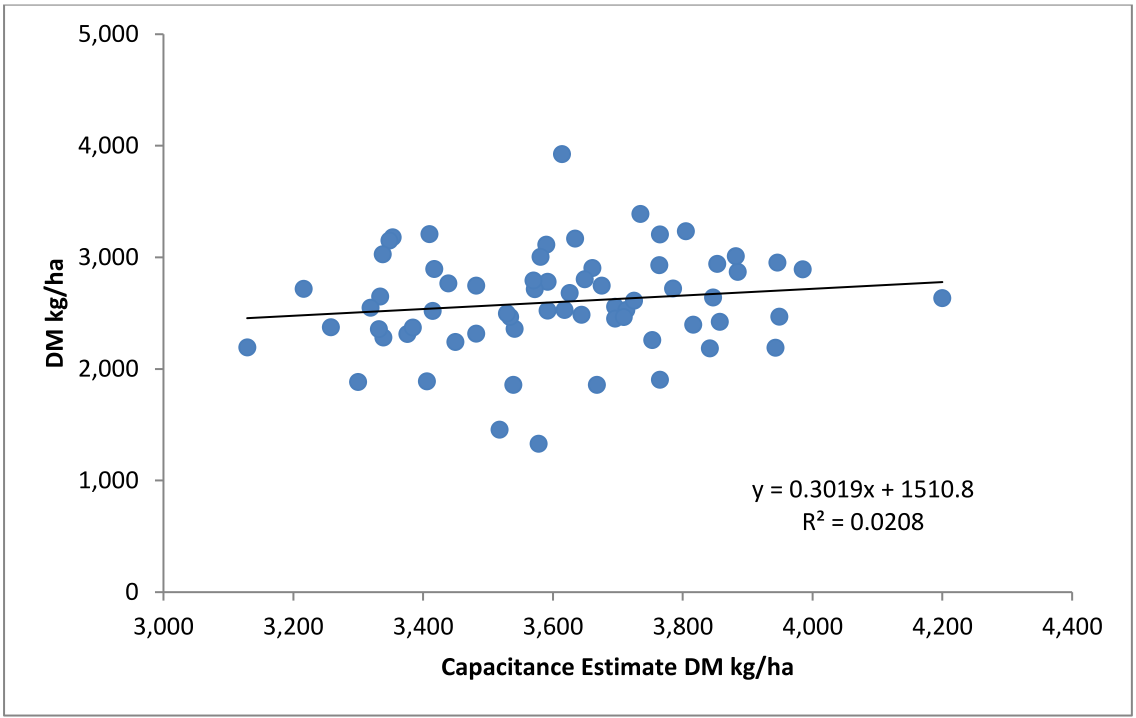


Figure 294. Capacitance estimation and actual pasture DM production.

There was very little difference in dry matter production between unfertilised pasture and any fertiliser applications (Figure 45).

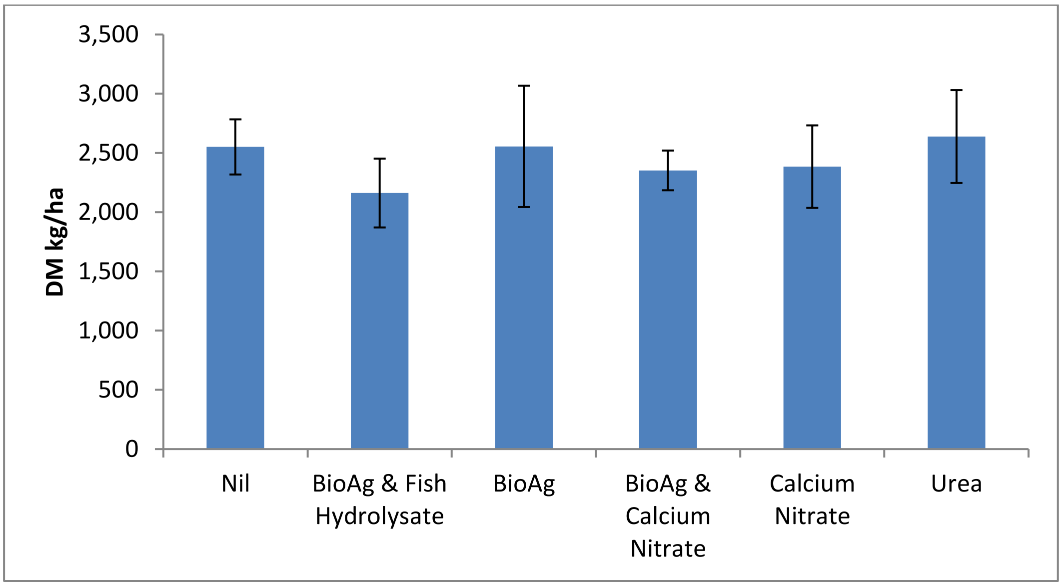


Figure 305. Effect of fertiliser on estimated pasture production on shallow soil phase ± SEM.

In late autumn-early winter on the shallow soil phase urea and BioAg stimulants with calcium nitrate gave the greatest production, though only 4% and 3% above unfertilised pasture (Figure 46). On the deep soil phase urea gave the greatest increase in yield, 37%, then BioAg stimulants by 22% and BioAg stimulants with calcium nitrate aby 18%. BioAg stimulants with fish hydrolysate increased production by 11%.

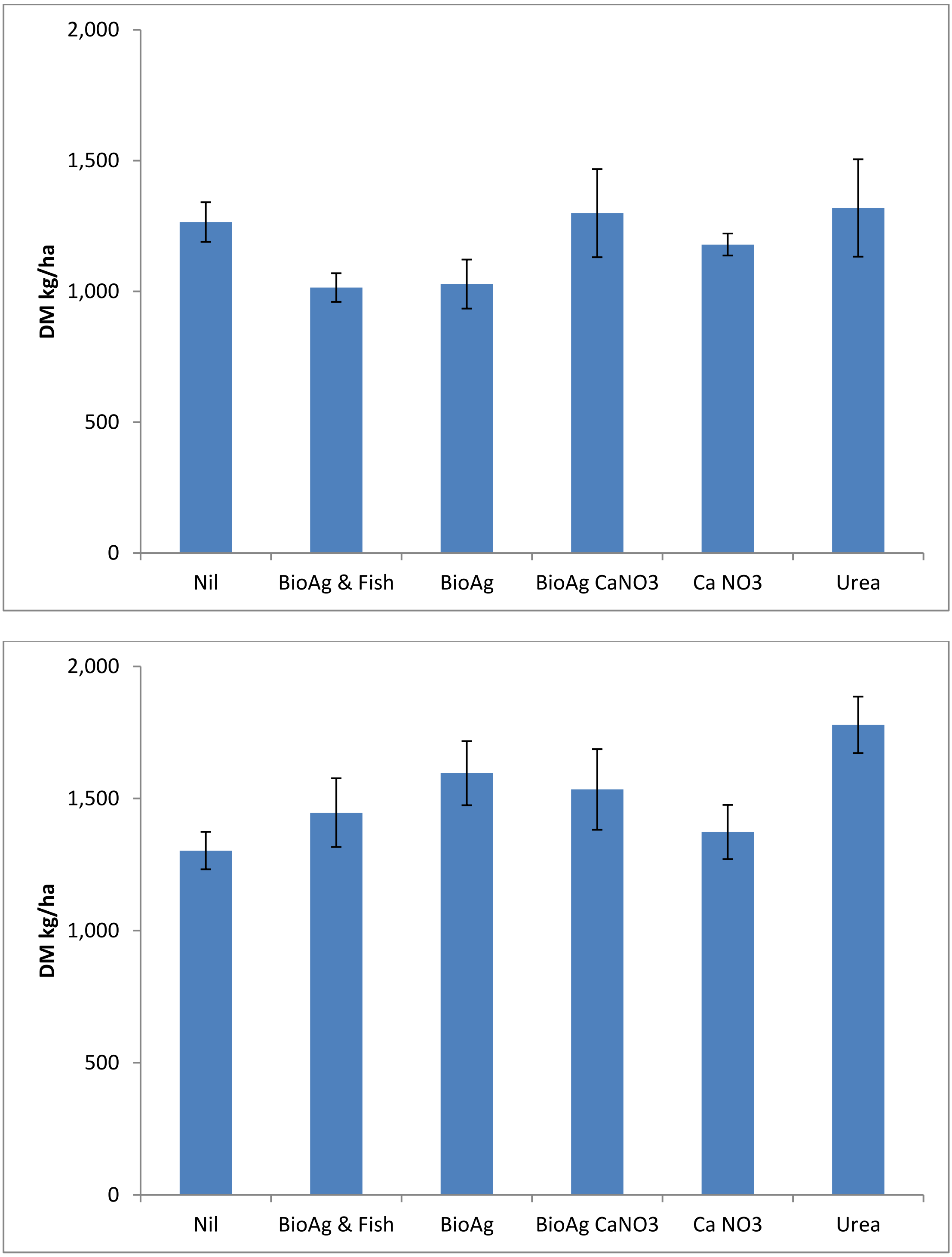


Figure 46. Effect of fertiliser on estimated pasture production on shallow (upper) and deep soil phases (lower) ± SEM.

Pasture growth was consistently greater on the deep soil phase than on the shallow soil phase in the 2016 -2 018 seasons (Figure 47). Fertiliser seasonal averages in 2017 -18, either from directly harvested dry matter yields (Table 5, Figure 48), or from the more extensive, but less precise, combined harvested and estimated yields (Table 6), differed with soil phase. Urea and BioAg stimulants were the two highest yielding applications on both soils, followed by calcium nitrate. BioAg stimulants with calcium nitrate ranked next, then BioAg stimulants with fish.

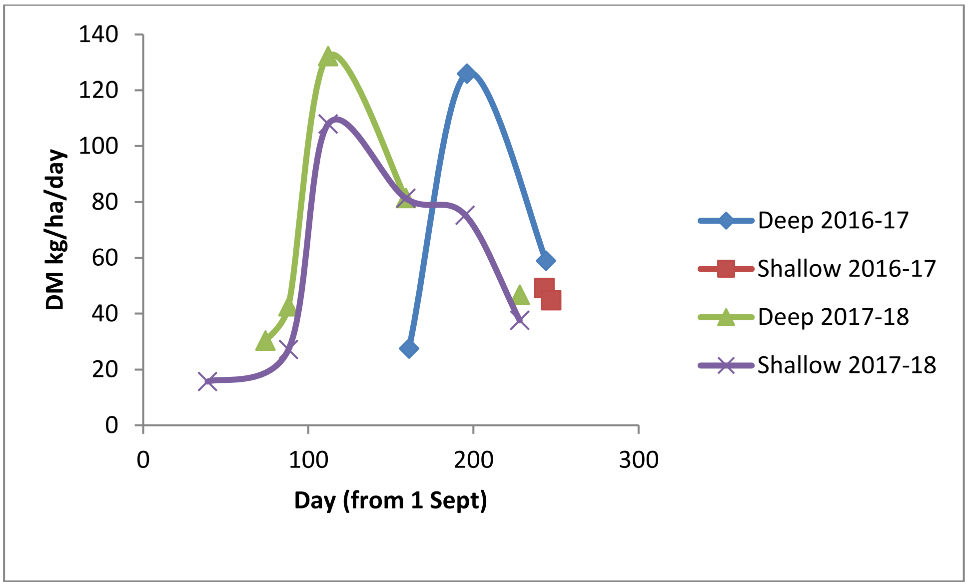


Figure 47. Effect of soil phases on pasture production 2016 - 2017 and 2017 – 2018.

Table 5. Change in harvested DM relative to unfertilised pasture 2017-2018 (%).

Fertiliser	% Change	
	Shallow	Deep
Nil	0	0
BioAg stimulants	2	28
BioAg & Fish Hydrolysate	-11	3
BioAg & Calcium Nitrate	-4	11
Calcium Nitrate	-2	14
Urea	23	54

Table 6. Change in combined estimated and DM yields production relative to unfertilised pasture 2017-2018 (%).

Fertiliser	% Change	
	Shallow	Deep
Nil	0	0
BioAg stimulants	-1	20
BioAg & Fish Hydrolysate	-7	16
BioAg & Calcium Nitrate	-7	17
Calcium Nitrate	-4	19
Urea	2	33

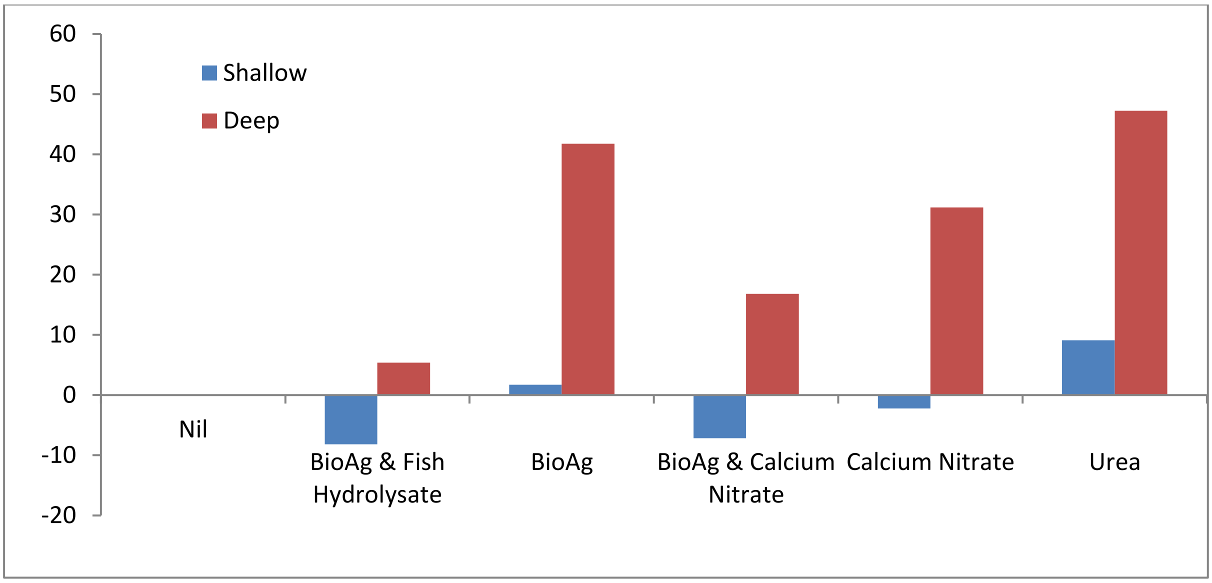


Figure 48. Change in relative DM production from unfertilised pasture in the2017-2018 season.

Statistical analysis of the harvested dry matter during both growing seasons showed a significant difference in pasture production between soil phases (ANOVA $P < 0.05$) but not between fertiliser applications ($P = 0.63$). There was also no statistically significant difference using the combined DM.

3. Discussion

Nitrogen deficiency limited pasture production at both Southland and Canterbury. The N supplying fertilisers calcium nitrate and urea increased yield in Southland by 11% - 43% [6] and by 14% to 53% in Canterbury. The positive 49% response to fish hydrolysate in Southland in 2016 - 2017 showed that it also effectively supplied N though its effect in Canterbury was less, giving a 6% increase on the most responsive deep soil phase

The application of Soil and Seed bio-stimulant and subsequent Root & Shoots bio-stimulant, increased base fertiliser yield in soils with good or reasonable fertility levels by 15% -19% in Southland and by 3% to 15% in Canterbury in 2016 – 2017. This demonstrates applications further increased N availability, or other limiting nutrients or micronutrients at both sites. Consistent with this, application of Soil and Seed plus Root and Shoots without any fertiliser increased yield at Southland by 12% and by 28% in Canterbury on the deep soil phase in the 2017- 2018 season.

The low application rates of hydrolysate, Soil and Seed and Roots & Shoots gave increases in pasture yield similar to those from calcium nitrate or urea directly supplying far greater amounts of N. Fish hydrolysate contained 2.2% N, 1.6% P and 0.3% potassium compared with 46% N in urea and 17% N in calcium nitrate. Soil and Seed contains 0.3% N and 2% P.

This response, despite the far greater quantities of N supplied in the mineral fertilisers supports BioAg's claim that their products and programme stimulate soil biology which may then be responsible for additional nutrient or micronutrient supply resulting in increased pasture production. The decrease in P and S levels during the trial may indicate that pasture uptake of these nutrients may also limit production and that soil biological mobilisation of P and S may also contribute to the observed yield increases compared with unfertilised pasture or when added to the N supplying fertilisers.

Statistical analysis showed BioAg combinations significantly or highly significantly increased pasture production in the 2016-17 Southland assessment (Table 2) but not in the following season or in the Canterbury assessments (Tables 5, 6). As the pasture response in the 2017-18 Southland season and soil biological activity were both affected by rainfall, soil moisture may have affected results.

Similar beneficial trends with BioAg applications occurred in Canterbury, notably a 28% increase with Roots and Shoots without fertiliser, but these did not reach the threshold for statistical significance. This could be due to greater experimental variability within soil phases, or to differing microbiological communities resulting from the high fertility inputs, particularly of urea at 50—70 kg/ha of urea following dairy grazing rounds compared with far lower inputs for the Southland sheep and beef pastures.

[6] Urea application increased yield at the Southland Site in a simultaneous trial run for Southern Humates.

In the synchronous companion trial with urea, humate and gauno at Southland, DNA assessment showed fertiliser and humate application significantly altered microbial population composition and affected microbial activity, directly corresponding to pasture production. It is probable the same microbiological effect is operating with BioAg applications. BioAg applications appear to beneficially increase pasture production in both South Island sheep and beef and dairy pastures. This is consistent with other reported research results[7].

The significant difference between soil phases at Rakaia is consistent with results from Lincoln College in similar soils in the same Canterbury environment where soil structure and moisture holding capacity greatly affect pasture daily growth rates (Figure 49). It is possible that this may equally extend to microbiological populations and activity.

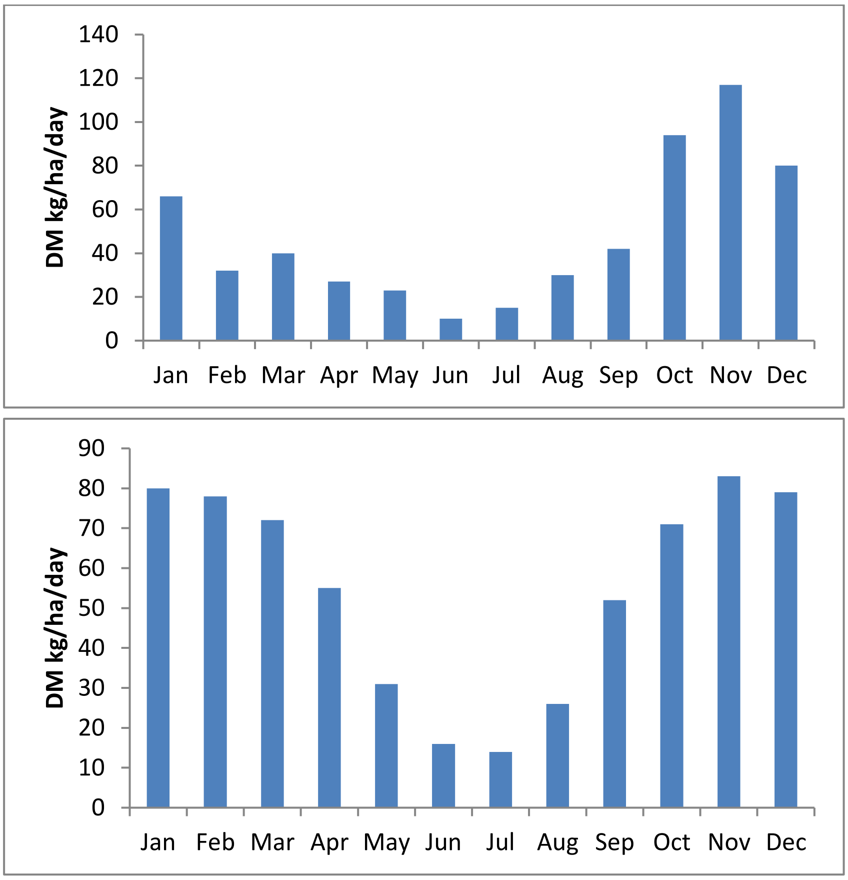


Figure 49. Lincoln daily growth rates (upper) light sandy loam (lower) heavy clay.

The results from this trial provide evidence that biological stimulants or microbial additions increase pasture yield, plausibly by influencing the soil microbiome. This may provide a feasible approach for supplying pasture nutrient requirements in New Zealand pastoral systems.

[7] Barton, A. 2016. BioAg Company Overview. (https://morellofert.com.au/wp-content/uploads/2015/08/Trial-data.pdf); BioAg. Better soils. Better crops. Better stock. 2019. (https://www.bioag.co.nz).

4. Acknowledgements

This was a huge trial and would not have been possible without help from many people: I thank Steven Haswell and Kim Strang, BioAg Ltd.; Bruce Hore, Agriganics Ltd.; Malcolm Sinclair, Chris Nel and staff, Southern Humates Ltd.; and Michael Richards, HydroBoost Ltd. for their for their assistance with trial planning, establishment and assessment; Steve Booker and Dave Jackway, Rotokaia Farm Ltd, for site provision, fencing and assistance with grazing management.

5. Appendices

5.1: Southland topsoil chemistry

Soil Test	Value	Medium lower range value	% of Medium lower range
Volume weight g/mL	0.88	0.60	
Soil Acidity pH	6.1	5.8	105
Resin P mg/kg	23	25	92
Olsen Phosphorus mg/ L	7	15	47
Phosphorus (Mehlich 3)	16	30	53
Total' Phosphorus mg/kg	656		
Potassium me/100g	0.12	0.30	40
Potassium % BS	0.7		
Potassium MAF quick test units	2		
Potassium (Mehlich 3)	16	44	36
Calcium me/100g	10.9	3.0	363
Calcium % BS	59.0		
Calcium MAF quick test units	12.0		
Calcium (Mehlich 3)	1824	650	281
Total' Calcium mg/kg	5,020		
Magnesium me/100g	0.5	0.8	63
Magnesium % BS	2.7		
Magnesium MAF quick test units	10.0		
Magnesium (Mehlich 3)	56.7	90.0	63
Sodium me/100g	0.10	0.20	50
Sodium % BS	0.5		
Sodium MAF quick test units	4.0		
Sodium (Mehlich 3)	21.0	40.0	
Total Base Saturation %	63	55	115
Anion storage capacity %	48	30	160
Cation Exchange Capacity me/100g	18	12	150

Appendix 1 continued.

Soil Test	Value	Medium lower range value	% of Medium lower range
Sulphate Sulphur mg/kg	4	10	40
Extractable Organic Sulphur	7	15	47
Total' Sulphur mg/kg	463	600	77
Available N (15 cm depth) kg/ha	256	150	
Total Nitrogen %	0.3	0.3	103
Anerobically mineralisable N µg/g	195		
Anerobic N / Total N Ratio	6.3		
Organic Matter %	7.8		
Total Carbon %	4.5		
Carbon / Nitrogen Ratio	14.6		
Iron (Mehlich 3)	275.0		
Managanese (Mehlich 3)	15.4	8.0	193
Zinc (Mehlich 3)	0.51	0.40	128
Copper (Mehlich 3)	0.5	0.4	50
Boron (Mehlich 3)	0.54	0.60	90
Cobalt (Mehlich 3)	0.1	0.3	33
Aluminium (Mehlich 3)	1089	900	121
Total' Copper mg/kg	5		125
Total' Molybdenum mg/kg	0.4		
Total' Cobalt mg/kg	3.00		
Total' Selenium mg/kg	0.5	0.5	100
Total' Cadmium mg/kg	0.23		

5.2: Canterbury topsoil chemistry

Element	Range	Shallow Soil Phase Sample1	Sample2	Ave	Deep Soil Phase Sample1	Sample2	Ave
Total Exchange capacity (M.E.)		13.0	14.2	**13.6**	11.4	12.9	**12.2**
pH		6.8	6.7	**6.8**	6.2	6.5	**6.4**
Organic Matter %		3.6	3.7	**3.6**	3.5	3.4	**3.4**
Nitrogen lbs/acre	Desired	95.3	97.5	**96.4**	95.3	93.0	**94.2**
Sulphate S ppm	Actual	7.8	13.5	**10.6**	16.8	6.7	**11.8**
Olsen P	Desired	271	272	**272**	269	270	**270**
P_2O_5 lbs/acre	Actual	278	375	**327**	249	275	**262**
Calcium	Desired	3,966	4,320	**4,143**	3,373	3,717	**3,545**
lbs/acre	Actual	4,082	4,147	**4,115**	3,087	3,562	**3,324**
Magnesium	Desired	419	457	**438**	368	393	**381**
lbs/acre	Actual	640	741	**690**	482	586	**534**
Potassium	Desired	448	479	**463**	405	430	**418**
lbs/acre	Actual	238	483	**360**	319	198	**259**
Sodium lbs/acre	Actual	133	158	**146**	198	169	**184**
Base Saturation %	Desired						
Calcium (60 - 70)	60 - 70%	70.0	65.3	**67.6**	60.4	65.2	**62.8**
Magnesiun (10 - 20)	10 - 20%	18.3	19.4	**18.9**	15.8	17.9	**16.8**
Potassium (2 - 5)	2- 5 %	2.1	3.9	**3.0**	3.2	1.9	**2.5**
Sodium (0.5 - 3)	0.5 - 3%	2.0	2.2	**2.1**	3.4	2.7	**3.0**
Other bases		4.6	4.7	**4.7**	5.2	4.9	**5.1**
Hydrogen (10 - 15)	10 - 15%	3.0	4.5	**3.8**	12.0	7.5	**9.8**
Micronutrients ppm							
Iron		514	428	**471**	563	500	**531**
Manganese		63	97	**80**	68	71	**70**
Zinc		9.3	10.0	**9.6**	9.1	8.2	**8.7**
Molybdenum		1.5	1.4	**1.4**	1.4	1.5	**1.4**
Copper		1.3	1.3	**1.3**	1.0	1.0	**1.0**
Boron		1.0	0.7	**0.8**	0.9	0.8	**0.8**
Cobalt		0.6	0.5	**0.5**	0.6	0.6	**0.6**

5.3: Statistical Analysis of significant fertiliser responses.

Table 7. Analysis of variance for fertiliser effect on pasture production, and Tukey multiple comparison of means, Southland, February 2017.

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Fertiliser	4	28017491	7004373	4.118	0.010
Residuals	26	44223754	1700914		

Fertiliser	Statistical difference	Significance P
Nil	a	
Calcium Nitrate	ab	0.034
Calcium Nitrate & Soil & Seed	b	0.034 *
Fish Hydrolysate	b	0.016 *
Fish Hydrolysate & Soil & Seed	b	0.022 *

Significance code: * = P < 0.05

Table 8. Analysis of variance for fertiliser effect on pasture production, Tukey multiple comparison of means, Southland, April 2017.

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Fertiliser	4	11895727	2973932	3.393	0.0239
Residuals	26	21909685	876387		

Fertiliser	Statistical difference	Significance P
Nil	a	
Calcium Nitrate	ab	0.149
Calcium Nitrate & Soil & Seed	ab	0.066 •
Fish Hydrolysate	ab	0.060 •
Fish Hydrolysate & Soil & Seed	b	0.038 *

Significance codes: • P < 0.1; * = P < 0.05, treatments with different letters differ significantly.