Growing confidence in forestry's future Research Programme

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Technical Note

Track-sprayer screening trials for testing new mid-rotation treatment options – 2015/16

Summary:

This study investigated the effect of a range of potential mid-rotation spray treatments on enhancing growth in young Pinus radiata plants in pots. In addition, it also discusses the scope of foliar spray treatments for improving management practices and potentially reducing/minimising environmental impacts. A total of 13 products were tested across a range of rates and these combinations resulted in 29 individual treatments which were monitored over seven months. At the end of the growing season, a product called Phos-Pot™ (Grochem) applied at rates between 4.2 and 10 L/ha provided the greatest height and diameter gains. Foliar applications of urea at between 7.82 and 15.65 kg/ha was the best based on stem dry weight and product costs. Positive growth effects to biomass allocation were also shown using microbial products such as, Nutri-Life 4/20™ (Nutri-Tech), Amino-Max™ (Nutri-Tech). There is scope to improve the economics of some of these treatments further.

Plant processes such as photosynthetic capacity and allocation to stem growth was increased in several treatments. Specifically, the photosynthetic capacity was improved in 54% of treatments and a maximum gain in photosynthetic capacity of 11.8% was shown for Perk Supa™ (Key Industries). Products increased the efficiency of stem growth as a proportion of total biomass by 6.6% across Urea rates and the greatest gain to stem allocation was observed from the Amino-Max (14.8%).

Results from these screening trials suggest several pathways for improving forest productivity and it is proposed to test some of these foliar treatments in operational field trials. Options for forest growers interested in getting involved are to:

i) At relatively fertile sites, field test applications of Phos-Pot (4.2 L/ha) and Amino-Max (< 10 L/ha) for boosting growth.
ii) Monitor disease resistance in response to application of Phos-Pot, Perk Supa, Nutri-Life 4/20 and BioPlex ™ (Nutri-Tech) products which are thought to stimulate the P.radiata immune system and hence reduce disease. Further work is needed to confirm this hypothesis.
iii) If growers are considering treatment options alternative to those tested here, the protocols set up for this study provides a low risk and cost effective method for accurately testing different options, and
iv) Assist with field trials where possible and discuss treatments if interested, given that several options look promising at this preliminary stage.

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Introduction

This project was initiated to screen a large range of new treatment options. We investigated several foliar fertiliser treatment options to determine their potential for enhancing growth in mid-rotation stands while minimising environmental effects and improving cost-effectiveness compared with conventional practices.

The initial treatment strategies were identified in the "Review of mid-rotation management options for increasing growth, quality or value from New Zealand plantation forestry" [1]. Products chosen in this study primarily consisted of nitrogen (N) in different forms or in combination with other products e.g. humic acids, phosphorus (P), copper (Cu), sulphur (S) or plant hormones. Nitrogen was chosen as the key ingredient because it has the potential to boost growth across most Forest Growers sites, particularly at the beginning and end of each growing season, whereas, other nutrients are typically only growth limiting at specific mid-rotation stands.

The screening of these products was undertaken in pot trials as this is a faster and more economic method to screen a large range of new treatments compared to operational trials in mid-rotation stands. Therefore, an efficient screening protocol based on seedlings was established in this study with the aim to identify promising products which would then be trialled in field trials

Methods

Thirteen different products, each with up to three different rates (see Appendix 1) were tested on recently lifted GF19 P. radiata seedlings growing in four litre liver pails. In total, 29 treatments were applied. Each treatment was represented by 25 plants in total. Each application was repeated 5 separate times on 5 sets of different plants.

Products were mixed in a solution of water and applied using the Scion track-sprayer in mid Oct 2015 and seedling growth was monitored until May 2016.

Following operational practices, the target application rate was 100 L/ha. During treatment the rate applied was highly consistent across the 140 individual spray applications. The application rate varied from 93 – 108 Litres/ha, with an average of 102.83 +/- 2.79 (StdDev). This equates to a 2.71% Coefficient of Variation (CV), and a ten-fold improvement in precision compared with operational spray targets (30% CV).

Response measures

Soil nutrition, foliage chemistry, photosynthetic efficiency, plant size, biomass allocation were measured and changes analysed over the course of a growing season (Oct 2015 through to May 2016).

Available nutrients in soils were determined by water extraction, using a 1 part soil to 1.5 times volume of water for each product. The filtrate was analysed by ICP-OES (Hill's Laboratories, Hamilton).

Foliage samples were collected in late winter after a period of approximately two months post establishment.

Chlorophyll fluorescence of plants was assessed as a proxy for growth efficiency and nutrient response. Chlorophyll fluorescence was used as it is a non-destructive method for assessing a range of plant responses such as stress and optimum nutrition[2]. The photosynthetic characteristics of new season's foliage was assessed on five replicates per treatment over several different occasions.

Initial measures of nutrition and plant size were used to account for variations un-related to treatment effects and the volume index of

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seedlings was log transformed to improve compliance with analysis of covariance.

Biomass measurements

Six seedlings were selected from each of the 29 treatments. Seedlings were selected to represent the range of volume across each treatment. This subset of seedlings was based on two small seedlings from the 10% volume index quantile, two averaged sized from the 50% quantile and two large seedlings from the 90% quantile range of each treatment. This allowed the analysis to take into account the effect of seedling size on allocation, while avoiding the influence of extremes.

A total of 174 seedlings were removed from their containers, their roots were washed of soil, the above and below ground components separated and then dried in an oven till constant weight. Stems and branches were stripped of their needles and weighed separately.

Plants were maintained in a relatively wind sheltered area with drip irrigation, at the Scion nursery in Rotorua. The spring period had above average rainfall of 311 mm rain evenly spread over the three months from Oct 2015 till end of Jan 2016 (see Appendix 2).

Results

Initial Soil Chemistry

Soil samples taken prior to treatment showed differences for some of the treatment groups. Five of the products had significantly greater soil nutrition than the control, and one product had lower nutrients (Table 1). The soil media of Phos-Pot, Perk Supa, GA$_3$, and Sulphur plus nitrogen treatments had greater levels of either nitrate-N, P, Ca, Mg and S.

Analysis of variance indicated that four products started with significantly greater ($p < 0.05$) soil nutrition pre-treatment compared with the control (Fig. 1). Greater soil nutrition on average, prior to treatment, of Perk Supa, Phos-Pot, Nutri-Carb-N and GA$_3$ allowed more nutrient uptake from the soil media during the two months until treatment, which in turn potentially gave these plants a head start.

Table 1. Soil nutrient concentrations prior to treatment, sorted alphabetically by product. Note: Nutrient concentrations shown in milligrams per litre. Number in italics indicate higher concentrations than the control and underlined values indicate concentrations lower than the control.

Product	NH_4	NO_3	Ca	K	Mg	Na	P	S
Amino-max	2	23	27	24	7	2	3	15
Control	3	22	29	23	8	3	3	16
Cu + N	0.5	13	20	18	6	2	2	13
DAP	0.5	21	26	22	7	2	3	12
GA3	*12*	*41*	*54*	*47*	*17*	*3*	*9*	*40*
GA4/7	2	22	32	23	9	2	3	18
GA4/7 + N	1	18	31	20	9	2	2	20
Nutri-Carb-N	0.5	17	30	20	8	2	2	19
Nutri-Life	*8*	*45*	*63*	*39*	*17*	*3*	*6*	*38*
Perk Supa	*15*	*41*	*55*	*47*	*16*	*3*	*11*	*44*
Phos-Pot	*4*	*36*	*43*	*33*	*13*	*3*	*5*	*26*
S + N	*4*	*37*	*49*	*29*	*14*	*3*	*4*	*23*
Urea	0.5	10	19	16	5	2	2	13

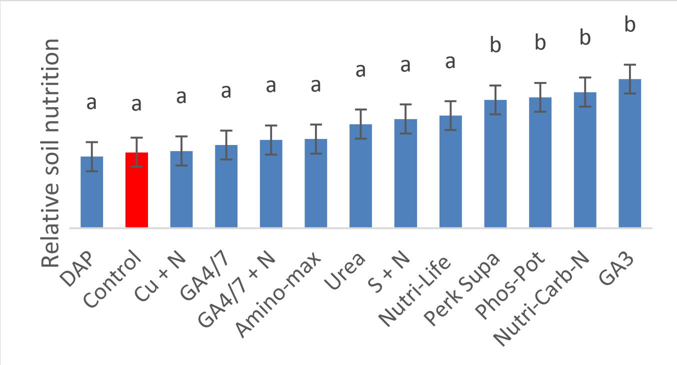

Figure 1. Relative soil nutrition* prior to treatment for each product. Treatments with the same letter are not significantly different from the control ($p < 0.5$). Error bars indicate one standard error of the mean. *Because of the variation across different elements (such as high Ca 3352 and low Zn 4.7 ppm) would mask the likelihood of significance of low values, soil nutrients were standardised to a mean of one and then differences were tested.

Initial Foliage Nutrition

The average foliage nutrition prior to treatments was high in copper and iron and low in nitrogen (Table 2). These reflect pre lifting nursery treatments and low nitrogen availability, typical of early spring.

Table 2. Foliage nutrient concentrations prior to treatments. Adjusted for needle weight and averaged across treatment series. SE = one standard error of the mean.

Nutrient	Average	SE
AL (ppm)	241.64	14.13
B (ppm)	9.23	0.38
C (%)	51.09	0.11
Ca (%)	0.28	0.01
Cu (ppm)	11.81	0.88
Fe (ppm)	70.94	4.36
K (%)	0.95	0.03
Mg (%)	0.12	0.01
Mn (ppm)	294.77	19.21
N (%)	0.91	0.02
Na (%)	0.02	0.001
P (%)	0.11	0.01
S (%)	0.24	0.01
Zn (ppm)	44.38	8.58

Treatment effects on plant photosynthesis

On average photosynthetic capacity increased rapidly from October till it reached a maximum in late December, in conjunction with peak solar intensity.

The effect of treatments significantly influenced plant photosynthetic capacity and light use efficiency. Treatments improved the photosynthetic capacity of 54% of the tested products compared with the control. Perk Supa showed the maximum gain of 11.8%. Reduced photosynthetic capacity was observed in three treatments (not shown here). Regression analysis also indicated ($p<.0001$) that gains in both the photosynthetic capacity and light use efficiency were associated with greater quantities (kg/ha) of P, in relation to N and K levels. The quantity of N, P and K in different products explained 32% of the variation in photosynthetic capacity and 39% of the light use efficiency.

Plant size and growth responses

Prior to treatment the mean height and diameter (+/- standard deviation) of the plants were 35.1 (+/- 2.16) cm and 7.6 (+/- 1.4) mm. Of the 725 seedlings, 667 survived until final assessments in May. A mortality rate of 8% occurred randomly amongst all treatments of the trial.

Initially, the volume index ($0.3*ht*\pi r^2$) of three of the 28 treatments was larger than the controls ($p<0.1$). Assessment of height and diameter growth increment helped to account for these differences. Treatments grew up to a maximum of 54 cm in height and 8 mm in diameter prior to the final assessment in May. Over time the variation between treatment responses generally increased.

Height growth

In May, 78% of treatments had seedling heights greater than the control (Fig. 2). Gibberellic acid ($GA_{4/7}$) at 6.6 grams per hectare produced the greatest height gains of 14.8% (7cm). The other $GA_{4/7}$ rates (10 and 12.5 grams/ha) also consistently produced significantly greater seedling heights. Treatments supplying only N, at different rates, were consistently ($p<.0001$) reduced in height compared with the controls.

Diameter growth

In May, 54% of treatments had greater diameter growth ($p>0.05$) than the controls (Fig. 3). All three rates of Phos-Pot were associated with the most diameter growth (10.4-10.7% gain), followed by the $GA_{4/7}$ treatments. Treatments supplying only N provided 3.2-3.4% gains in diameter over the controls. DAP fertiliser at rates of 20, 40 and 50 kg/ha had reduced diameter growth compared with controls. Similar to Urea this contrasted with superior early season responses to height.

Volume increment

Combining height and diameter into a volume index for each treatment helps to attribute the relative gains and losses of these criteria collectively. Volume increment was increased by a maximum of 5.6% over the untreated controls by the Phos-Pot product. Amino-Max™ ranked next best and then the plant hormone $GA_{4/7}$, featuring consistently across different rates (Fig. 4). Products that resulted in a reduced volume index at the end of season were primarily conventional forms of N (Urea) and P (DAP). It is possible that this indicates mineral forms of nutrition had long term detrimental effects on secondary sources of nutrition such as provided by microbial activity.

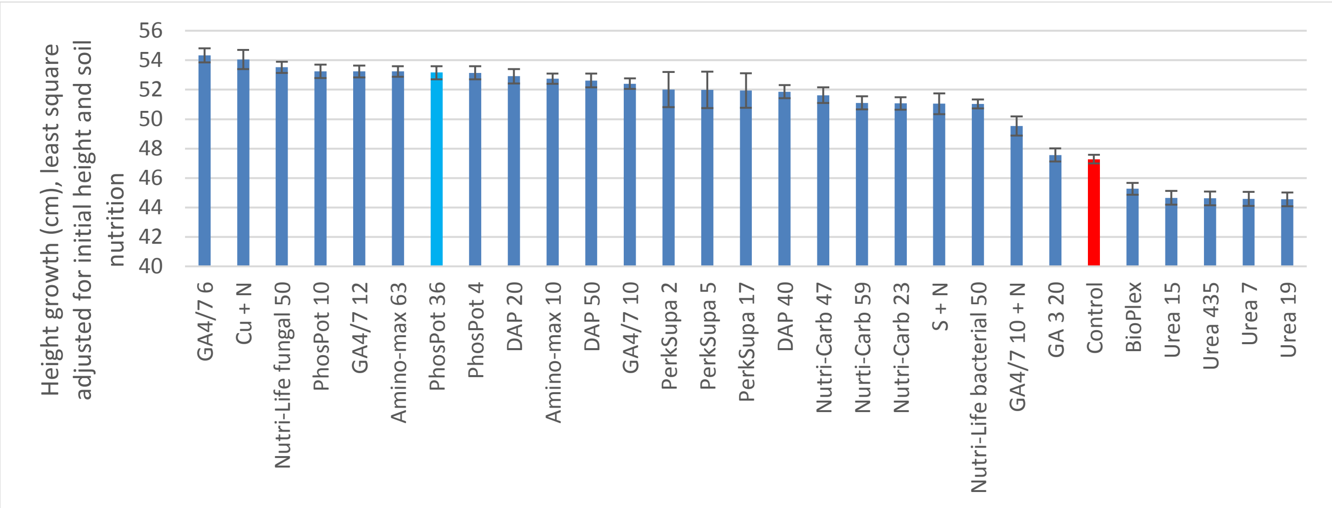

Figure 2. Mean seedling height growth as at the end of season measure. Bars = 1 StdErr of the mean.

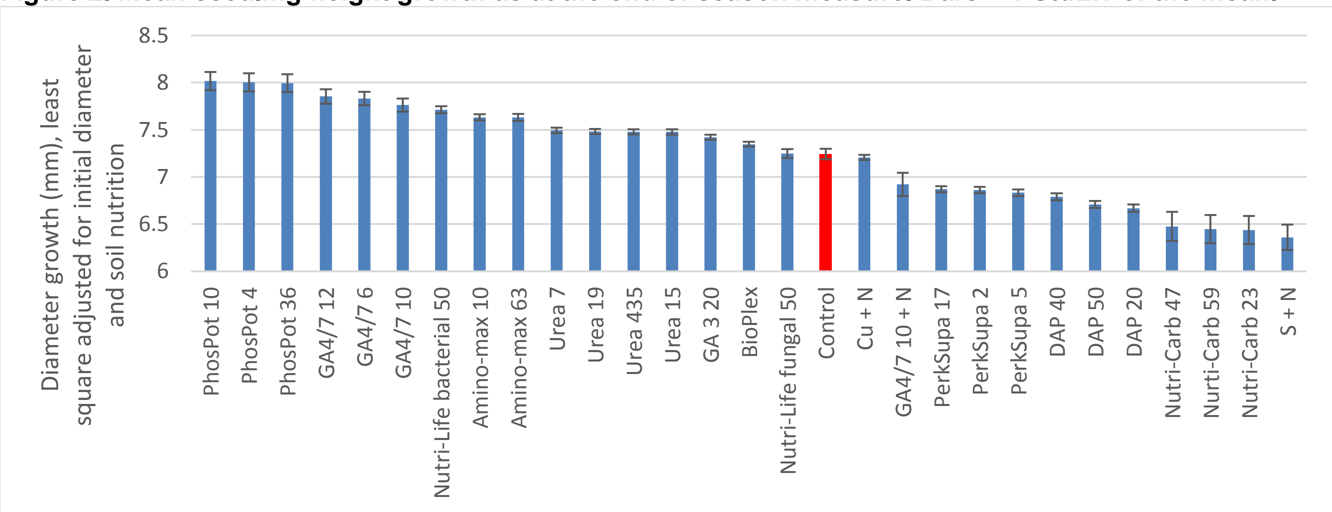

Figure 3. Mean seedling diameter growth as at the end of season measure.

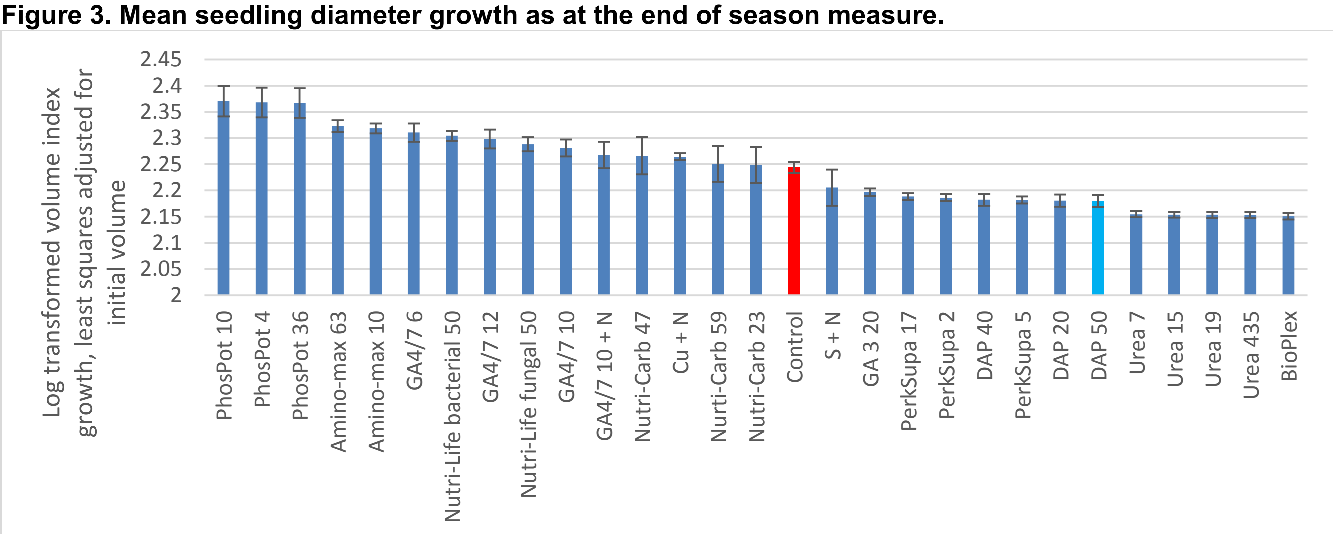

Figure 4. Mean seedling volume index growth as at the end of season measure.

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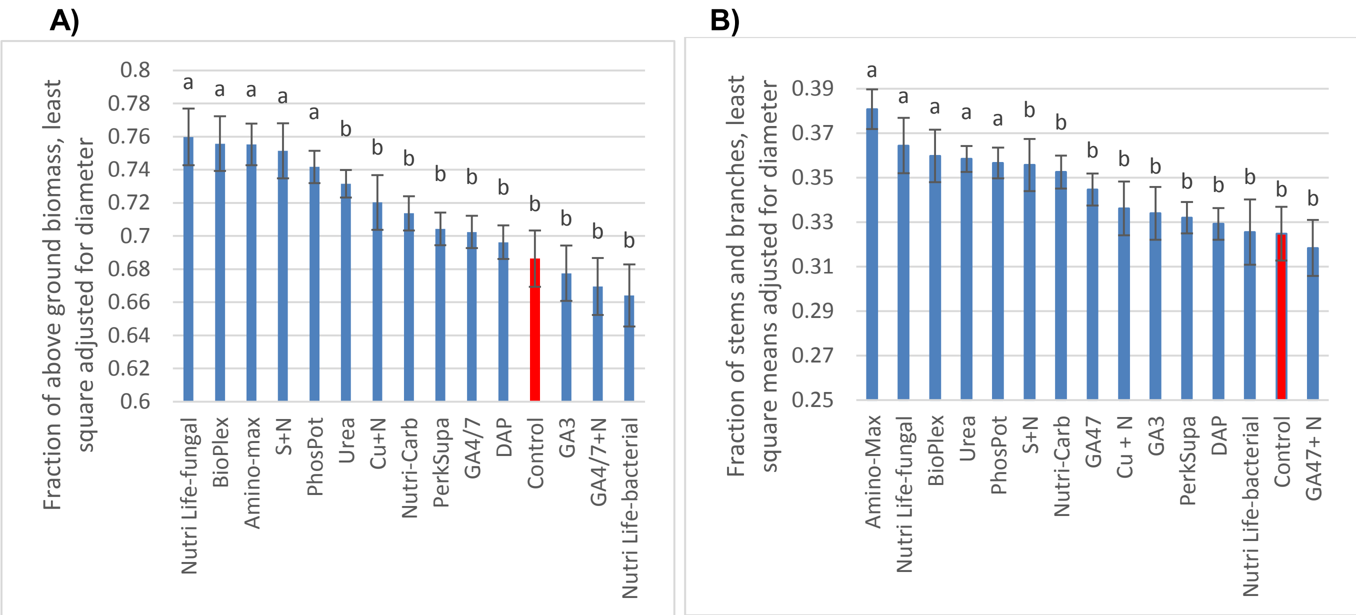

Figure 6. a) Fraction of seedling biomass in the above-ground components and b) the stem + branches components for identified products. Least square adjusted mean values are plotted as at the end of season harvest. Bars indicate one standard error of the mean. Treatments with the same letter are not significantly different from the Control ($p < 0.01$).

Biomass and differences in allocation

Analysis indicated that the size of the plants chosen for biomass assessment was not significantly different across treatments ($p=0.92$). (Fig 5.)

Above ground biomass

The fraction of above ground biomass (stems & branches + foliage) in relation to total biomass *increased* with increasing seedling diameter ($p<.0001$) and height ($p=0.0002$), but not volume index ($p=0.83$). This suggests that treatments may influence both height and diameter, but the above ground biomass fraction is maintained for a given sized plant.

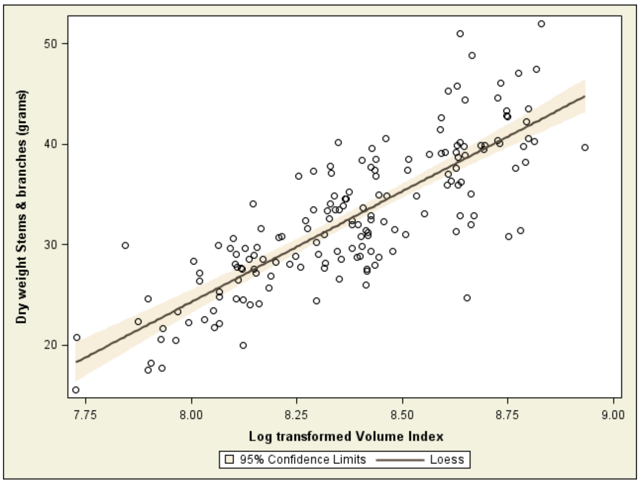

Figure 5. Stem and branch dry weights of seedlings treated with a range of spray products in Oct 2015 and harvested in May 2016.

Similarly, the proportion of foliage decreased consistently with increasing volume index ($p<.0001$), but the allocation to above ground biomass was maintained by increased stem and branch mass with increased volume index ($p<.0001$).

Data indicated that the above ground biomass differed by the main effect of treatment ($p=0.094$) and this was confirmed by an interaction between volume index and treatment ($p=0.035$). This provides greater certainty that some treatments were associated with a change in the relationship (slope) between above ground biomass and volume index. Six products increased the above ground biomass as a proportion of total dry weight compared with the controls (Fig. 6a). The Nutri-Life 4/20™ fungal treatment increased the proportion of above ground biomass the most, by up to 10.7%.

Stem growth

The biomass allocation to stem is the most valuable component to a forest manager. Allocation was significantly increased by Amino-Max 17.3%, Nutri-Life 4/20 fungal and Nutri-Life BioPlex™, Urea 10.4% and Phos-Pot 9.8% (Fig. 6b). No products significantly reduced the fraction of biomass allocation to stem when size was taken into account compared with the control.

Discussion

The mechanisms creating benefits of each product are not yet fully understood. However, a single application of a range of products influenced seedlings throughout the growing season. Preliminary analysis in February (not shown) indicated several differences in the ranking of both height and diameter growth responses compared with those assessed in May. For example Urea influenced diameter growth more than all but the Phos-Pot products prior to February. The Urea treatments continued to increase seedling diameter post February but the relative rate was decreased compared with other products, such as $GA_{4/7}$. This suggests that some products may either: i) signal or assist plants to transfer through growth stages at different times, ii) delay the effectiveness of some products or, iii) provide short term mineral nutrient gains, which results in detrimental feedback influences, such as discouraging microbial activity.

A unique aspect of this study was the investigation of mineral fertilisers, plant hormones, immune system stimulants and fungicides on biomass allocation. We observed that a number of products increased the above ground biomass and specifically the proportion of stem mass. As biomass lags stem growth by about a month[3], early gains of height and diameter are then followed by changes in foliage biomass. Our study supports early height and diameter growth followed by increased proportions of above ground biomass.

Height growth

Treatments supplying only N were consistently reduced in height compared with the controls. This supports a view that plant height is likely to be less with high fertility[4]. This may also indicate that N influences diameter growth at the end of the growing season, in preference of height growth.

Immune system stimulants

Phosphite is an emerging biostimulant [5] and this trial tested Phos-Pot which contains 400g/L Phosphorous Acid (Phosphite) with urea to aid absorption. Typically conversion from phosphite ($H_2PO_3^-$) to the plant available form phosphorus (H_2PO_4), takes about 3-4 months of bacterial and fungal oxidation. We found that Phos-Pot significantly increased volume index and stem biomass of radiata seedlings. Although, it is thought that phosphorus limited plants may be sensitive to phosphite applications and display phytotoxic symptoms. This suggests that Phos-Pot might not be suitable for forest stands deficient in soil P.

Microbial products

The microbial based Nutri-Life 4/20 fungal treatment provided benefits to the above ground biomass and these contrasted with the Nutri-Life 4/20 bacterial treatment. It remains un-confirmed if this benefit was provided directly though the foliage or if there was a soil based response. Our evidence supports the general view that trees benefit from fungal dominated soils.

Plant hormones

This study provides evidence for the first time that plant hormones can be applied successfully at operational rates in New Zealand. The plant hormone $GA_{4/7}$ provided maximum height growth (Fig. 2), significant diameter growth (second to Phos-Pot) and volume index gains at the end of the season. The proportion of above ground biomass and stem and branch allocation was similar to the controls (Fig. 6 a&b). This suggests that $GA_{4/7}$ can be successfully applied without fear of creating an abnormal tree because biomass is allocated within natural proportions. Also that the stimulating benefits to growth potentially act for less than a season long. Suggesting that it may be impractical to apply $GA_{4/7}$ to a mature stand unless there are synergistic effects when combined with another product. The combination with low rates of Urea, did not appear to provide potential synergistic gains.

GA_3, a much cheaper option than $GA_{4/7}$, improves grass growth[6], but did not improve radiata seedling growth significantly. The potential for GA_3 to provide significant gains to radiata appears low.

Fungicides

The fungicidal treatments of copper at double the Dothistroma rates (Cu 2.8kg/ha + N 3.2kg/ha) and Lime of Sulphur (0.3 L/ha + N 3.2 kg/ha) did not seem to provide beneficial growth as measured by stem volume index. Although, the copper had increased height growth and the lime of sulphur treatment had reduced diameter growth compared with the control. The allocation to above ground biomass was improved compared with the control. Suggesting that the foliage biomass was increased. But any synergistic benefits from the addition of nitrogen to these treatments was not shown in comparison with Urea alone.

Fraction of foliage biomass

Considering that next year's growth, and the potential for on-going growth benefits from treatments may be related to this year's foliage mass, this characteristic might be important. Treatments were therefore ranked according to the fraction of foliage (Appendix 3). For the least squares adjusted means, this suggested that the treatments with the highest probability of producing on-going treatment responses ($p<0.01$) were:

1. Bio-Plex 0.7 kg/ha
2. Phos-Pot 4.2 L/ha
3. Lime of Sulphur 0.3 L/ha + N 3.2 kg/ha
4. Nutri-Life 4/20

Urea at 435 kg/ha also ranked high ($p=0.066$).

To test if foliage contributes to the on-going benefit to seedling growth rates, ideally these treatments would be grown on into 2017 and compared with the remaining controls.

Economic considerations

Cost is a critically important consideration with regard to the development of beneficial treatments. The basis for our comparison for practical reasons was the cost of products, based on the trial requirements. Until such time as operational trials are complete, this is the only method available. At present we believe this is a valid comparison because all treatments were applied using the same operational parameter (100 L/ha application rate), and so the comparison is consistent across treatments (Fig. 7).

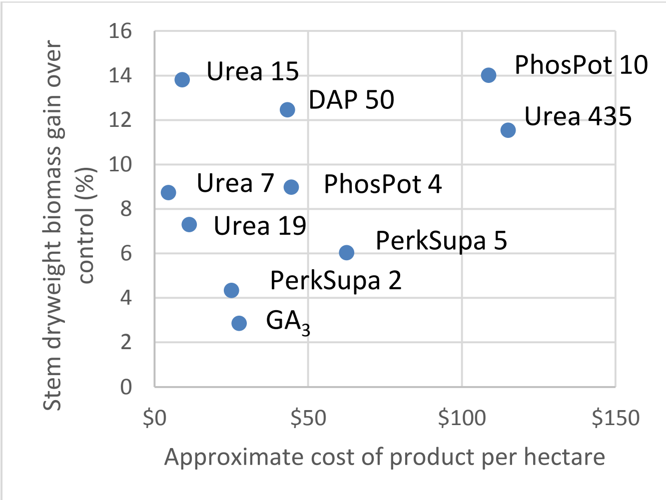

Figure 7. Cost of optimal treatments as determined by relative gains in dry weight biomass at end of the growing season compared with the untreated control.

On a cost and gain in dry weight basis, foliar applications of Urea at 15kg/ha appear 12 times more cost effective than conventional Urea treatments over a single growing season (Fig. 7). In terms of environmental cost, that equates to 29 times less added nitrogen into the environment. Phos-Pot at 10.25 L/ha provides a marginal productivity gain over conventional treatments.

Forest sites compared with screening trial soil media

We set up the trial to identify a worst case scenario by planting seedlings into a relatively fertile mix, those gains identified by treatments might be expected to be greater in the field and at specific sites. Our measured responses partially reflect high nitrogen availability of the soil, with some additional gains provided by the treatments supplying phosphorus.

By applying treatments to ‘plant foliage’ – the driver of growth – and attempting to directly influence foliage nutrition, the unnecessary manipulation of a tree’s substantial nutrient pool (below ground, and within the stem) is potentially avoided during a single season.

It appears that radiata seedlings may proportionally increase height firstly, then diameter and finally build foliage biomass. So to build more efficient foliage (see earlier on the chlorophyll fluorescence) perhaps nutritional applications could be more beneficial if applied at specific times such as from December after height growth? Or applications could be optimised to stimulate specific growth patterns such as height early in the season, with products like $GA_{4/7}$, which then may potentially have a multiplying effect on the following growth processes.

Conclusions

There are several ways to assess product performance. This study shows that the potential advantages are not always measured accurately by changes in height and diameter. We suggest that the above ground biomass is the preferred option for the screening trials undertaken here, because it includes the stem, a direct measure of current season’s growth, and also the foliage, which will contribute to enhanced growth in the following season.

Several of these treatments show good potential as new spray options. Advantages such as a 12 fold decrease in cost of product was enabled by more efficient use when applied as a liquid foliar application. Foliar application is one key way to increase treatment efficiencies and also fertilisers increased the relative efficiency of stem growth by up to 14.7%, another key mechanism enabling advantages.

At this time we have not determined the effects to wood quality because of the preliminary nature of these treatments and the small material being tested.

We recommend that beneficial treatments require further development and testing in operational settings. A series of field trials is currently being planned for 2016 and 2017 across up to 10 sites throughout New Zealand.

Recommendations

This study provided data which supported consistent trends across rates for the following products:

- Phos-Pot, optimal rates were determined to be less than the 4.2 L/ha tested in this study. Thus further minor operational gains in cost may be obtained.
- Perk Supa, optimal rates were determined to be less than the 2 L/ha tested in this study. Again further minor operational gains in costs may be obtained.
- $GA_{4/7}$, significant gains and optimal rates were shown at less than 6.6 grams/ha tested in this study. The high cost of a commercial product limits $GA_{4/7}$ use in forestry at present.
- DAP, optimal rates were shown to be greater than 50kg/ha. Unfortunately the limit to solubility is 56kg/100L, so an alternative product or formulation for liquid applications is recommended.
- Urea, optimal rates require further testing, but appear to be around the 10.8 kg/ha.
- Given the identified above ground benefits (Fig. 6a&b) Nutri-Life 4/20 has the potential to be a very cost effective option at operational scale because of the brewing process and its ability to cheaply seed a batch process with plant growth promoting microbes.
- Amino-Max at less than 10 L/ha is a viable alternative for boosting allocation to stem.

Based on these findings, combined with some additional development of ideas, the best treatments for use across the field trials are likely to be at least the following:

1. Granular Urea at 450 kg/ha – conventional practice.
2. Foliar N as 11 kg/ha Urea (5.1 kg/ha of N)
3. Foliar P as 4.45 L/ha Phos-Pot (0.49 kg/ha of P)
4. Foliar N+P at a ratio of 10.4:1 (5.1 kg/ha N as Urea @ 11kg/ha, 0.49 kg/ha of P as PerkSupa @ 2.17 L/ha)
5. Un-treated control

Acknowledgements

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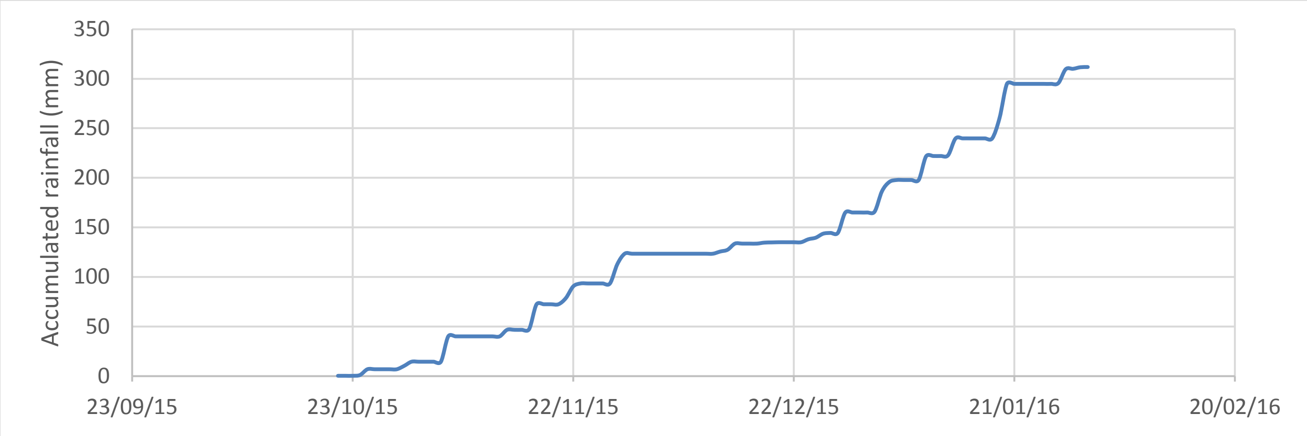
Appendix 1. **Products tested.**

Product	Rate applied	N	P	K	Approximate Cost/ha
			(kg/ha)		
Amino-Max	10 L/ha	0.57			$132.15
Amino-Max	63.2 L/ha	3.602			$835.17
Bio-Plex	0.7 kg/ha				$71.11
Control					$0.00
Cu+ N	2.8 kg/ha	3.6			$252.41
DAP	20 kg/ha	3.6	4		$17.30
DAP	40 L/ha	7.2	8		$34.60
DAP	50 kg/ha	9	10		$43.25
GA3 + N	20 g/ha	3.6			$27.50
GA4/7	10 g/ha				$1,363.19
GA4/7	12.5 g/ha				$2,034.50
GA4/7	6.6 g/ha				$1,074.22
GA4/7 + N	10 g/ha	3.6			$1,627.60
Lime of S+N	0.03 L/ha	3.6			$288.90
Nutri-Carb-N	23.74 L/ha	3.604			$54.60
Nutri-Carb-N	47.45 L/ha	7.203			$109.00
Nutri-Carb-N	59.26 L/ha	8.996			$136.20
Nutri-Life 4/20 Bacterial	0.5 kg/ha				$318.37
Nutri-Life 4/20 Fungal	0.5 kg/ha				$330.64
PerkSUPA	17.4 L/ha		4.002	6.612	$217.50
PerkSUPA	2 L/ha		0.46	0.76	$25.00
PerkSUPA	5 L/ha		1.15	1.9	$62.50
Phos-Pot	10.25 L/ha	0.205	1.128	1.948	$108.65
Phos-Pot	36.4 L/ha	7.28	4.004	6.916	$385.84
Phos-Pot	4.2 L/ha	0.084	0.462	0.798	$44.52
Urea	15.65 kg/ha	7.2			$9.00
Urea	19.56 kg/ha	9			$11.25
Urea	435 kg/ha	200			$115.00
Urea	7.82 kg/ha	3.6			$4.50

Appendix 2. Accumulated rainfall over the critical treatment period from October 2015 until Jan 31, 2016.



Appendix 3. The proportion of biomass ranked by the quantity of foliage. Note, not Least Squares adjusted for plant size, hence different ranking according to the discussion text.