High UMF® honey production from manuka plantations

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Abstract
High UMF® (unique manuka factor) manuka honey is in demand because of its strong anti-bacterial properties. However, production is limited and variable; establishing manuka plantations using seedlings grown from seed sourced from areas producing high quality honey is a potential solution. This study compared establishment, growth and nectar dihydroxyacetone content of four manuka provenances (geographically localised varieties) and indigenous manuka at Maxwell and Tutira. Survival was >90% in most provenances. Normalised (80 ºBx) nectar dihydroxyacetone content in plantation manuka was generally higher than the indigenous. At Maxwell provenances A (7757 mg/kg) and C (10 561 mg/kg) had significantly higher normalised dihydroxyacetone than indigenous manuka (4173 mg/kg) and at Tutira provenance D (12 157 mg/kg) was significantly higher than indigenous (5111 mg/kg). The results confirm the potential to produce high UMF® honey from manuka plantations.

Keywords: manuka, nectar, dihydroxyacetone, sugar, provenances

Key messages
- Plantation manuka sourced from selected provenances (varieties) produce normalised nectar DHA double that of indigenous manuka
- Basal stem diameter measurements provide an accurate estimate of biomass (carbon sequestration)
- High UMF honey from manuka plantations could increase returns on marginal land.

Introduction
Manuka (*Leptospermum scoparium*) is a successional species found throughout New Zealand particularly where fire and land disturbance has occurred. It has traditionally been regarded as a weed, often establishing on steep erosion-prone hill country. The economics of pastoral production on this land class are marginal; economic farm surplus on East Coast hard hill country has ranged from -$17.55 (2007-2008) to $255.76/ha (2011-2012) in recent years, for example (Beef + Lamb New Zealand 2015). Many hill country farmers wanting to improve and diversify income while minimising erosion risk adopt plantation forestry but this can be economically marginal depending on log prices, scale and location (Future Forests Research 2015).

New Zealand honey exports increased from 102 to $233 million from 2011 to 2015 (Statistics NZ 2015), primarily due to increased production and value of manuka honey. High manuka honey values (18 to $100/kg) (Bagrie *et al*. 2015), particularly honey with high 'unique manuka factor' (UMF®) rating (Allen *et al*. 1991) has resulted in a surge of interest in harvesting honey from natural stands and establishing manuka plantations using seed of provenances (geographically localised varieties) with high UMF® potential (Hamilton *et al.* 2013). An investment of about $2000/ha may produce an internal rate of return of >10 % and up to 16.4 % depending on production, quality, value and income share (Bagrie et al. 2015). Carbon credits may also generate income; 40.2 tonne/ha CO_2 (stands< 100 ha) can be claimed by year 10 (MPI 2015a), currently worth about $400/ha (Carbon News 2016).

The UMF® rating of manuka honey is determined by the methylglyoxal content, an antibacterial/fungal compound. Methylglyoxal forms in manuka honey over time through natural chemical transformation of dihydroxyacetone (DHA), present in manuka nectar. Nectar DHA varies between stands, seasons and individual plants in the same stand (Williams *et al*. 2014) but the reasons for this variability are not clear (Adams *et al*. 2009). Production of high UMF® honey suitable for medical dressings, in particular, is insufficient to meet demand (MPI 2015b).

A Primary Growth Partnership (PGP) programme aims to increase production of high value honey from manuka plantations (MPI 2015b). Assessing the genetic influence on nectar characteristics of plantation manuka at different sites is a major component of the programme. This paper presents results of the assessment of manuka plantations including different provenances and indigenous manuka at two sites.

Methods
Sites and plant material
The manuka PGP includes assessment of manuka provenances at a number of sites, mostly in the North Island. This paper reports on results from two contrasting sites; Maxwell (Whanganui) and Tutira (Hawkes Bay). Both sites were established in August 2011 on steep (25º) northwest facing slopes from pasture. Weeds: scattered manuka and gorse at Maxwell

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was controlled with metsulfuron herbicide (aerial) pre-plant, and pre-plant spot spraying (75 cm diameter) with glyphosate at Tutira. Soils are primarily sandstone with some tephra at Tutira.

Container grown seedlings of four provenances (Table 1) from stands known to produce high UMF® honey (J. Stephens pers. comm.) were planted at Maxwell, (1600 plants/ha) and three provenances (B, C and D) at Tutira (1100 plants/ha). Planting rates were estimated by considering site occupancy at maturity and local factors affecting survival including brush weed competition.

Sample plots

Four 0.02 ha circular plots were established in each provenance at both sites. Assessment of survival, height and basal stem diameter in each plot was carried out in September and November 2014 at Maxwell and Tutira, respectively. Height was measured with a graduated height pole and basal stem diameter with a digital calliper at ground level; two calliper readings were taken, the first across the widest point of the stem and the second at 90º. Stem basal diameter was calculated using the formula below:

Basal stem diameter = √(diameter 1 x diameter 2)

Mean stem diameter = quadratic mean.

Nectar collection

Nectar from the plantation manuka was sampled from 3 plants in each plot during peak flowering, October/November 2014. At both sites, provenance A was the earliest, B and C intermediate and D last to flower. Before sampling flowers were enclosed in fine mesh bags for at least 24 h without rain to allow nectar to accumulate. Five µl of de-ionised water was pipetted onto the hypanthium of flowers; the water and nectar mix were retrieved (pipette) into vials, immediately placed on ice and stored at -80°C until analysis. Fifteen flowers were sampled from each plant and nectar pooled for analysis of DHA and sugar. The indigenous manuka at both sites was sampled by randomly selecting 10 plants during peak flowering (December 2014). At Maxwell, no assessment of provenance B was possible because of uncontrollable gorse regrowth, and nectar quality for provenance D is not included due to limited nectar sampling.

Table 1 Provenance descriptions.

Provenance	Description	Origin
A	var. incanum	Northern North Island
B	var. scoparium	Central North Island
C	var. scoparium	Southern North Island
D	var. linifolium	Central North Island

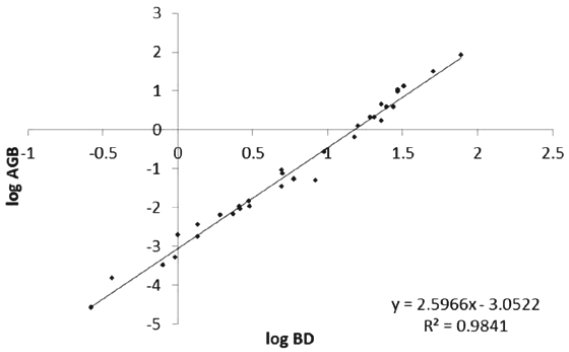

Figure 1 Relationship between the ln stem basal diameter (BD) and the ln above ground biomass (ABG), provenance A.

Nectar DHA content was determined using reverse phase HPLC with a diode array detection method (Windsor *et al*. 2012). Nectar sugar (sucrose, glucose and fructose) content was determined using HPLC with a refractive index detector (Hamilton *et al*. 2013). All nectar samples were normalised to 80 ^{0}Bx to standardise the DHA values.

Biomass allometry

The relation between basal stem diameter and above ground biomass was investigated by destructively harvesting 2 year old (Maxwell) and 4 year old (Massey University) provenance A plants. Sampled plants (33) were weighed, subsamples taken and weighed, oven-dried (85ºC) and reweighed to calculate dry matter % and total biomass. The relationship between basal stem diameter and above ground biomass was estimated using linear regression between the natural log of basal stem diameter and the natural log of above ground biomass (Scott *et al*. 2000).

Results

Growth

At Maxwell the mean height of individual provenances was 1.7, 1.9 and 1.8 m for provenance A, C and D, respectively, while mean basal stem diameter for individual provenances was 28.1, 34.8 and 30.7 cm, respectively (Table 2). There was greater variation in both height and basal stem diameter at Tutira; height of provenances B, C and D was 1.6, 1.5 and 2.3 m, respectively, while basal stem diameter was 30.2, 22.9 and 31.0 cm respectively. At Tutira, provenance D was significantly taller than other provenances ($P< 0.001$) while mean basal stem diameter of provenance C was significantly lower than other provenances ($P<0.002$). Survival was good (>90%) in provenances C and D but poor in provenance A (<40%) at Maxwell whereas survival was good in all provenances at Tutira (Table 2).

Biomass

A strong linear relationship between log stem basal diameter and log above ground biomass ($R^2 = 0.98$) was identified (Figure 1). This suggests that measurement of stem basal diameter will be sufficient to allow accurate estimation of above ground biomass and ultimately, above ground carbon.

Nectar quality

Maxwell

Normalised DHA in plantation manuka ranged from 7757 mg/kg (provenance A) to 10 561 mg/kg (provenance C) significantly higher (P=0.04) than the indigenous manuka (4173 mg/kg). Low normalised DHA content in the indigenous manuka is a result of significantly lower (P=0.04) nectar DHA (4.1 mg/kg) compared to the plantation manuka (Table 3). There were no differences in nectar sugar content.

Tutira

The normalised nectar DHA content of indigenous manuka was significantly lower (P=0.002) than provenance D (Table 3); normalised nectar DHA of provenance D was also significantly higher than provenance C (P=0.03). Similarly, the low normalised nectar DHA in the indigenous manuka at Tutira is primarily a result of low nectar DHA content (3.9 mg/g). The difference in nectar DHA between the indigenous manuka and provenance C was significant (P=0.05). No differences in nectar sugar were found at Tutira.

Relationship between nectar DHA content and normalised DHA

Scatterplots of nectar DHA content versus normalised DHA content for the plantation and indigenous manuka at both sites reveal a weak relationship (Figure 2). The correlation coefficients were r=0.63 (P=0.01) and r=0.40 (P=0.02) for Maxwell and Tutira, respectively. The nectar DHA content range in the indigenous manuka was lower than the plantation manuka at both sites. Normalised DHA of some plants was high despite low nectar DHA, a result of low nectar sugar content. For both sites and all provenances the coefficient of

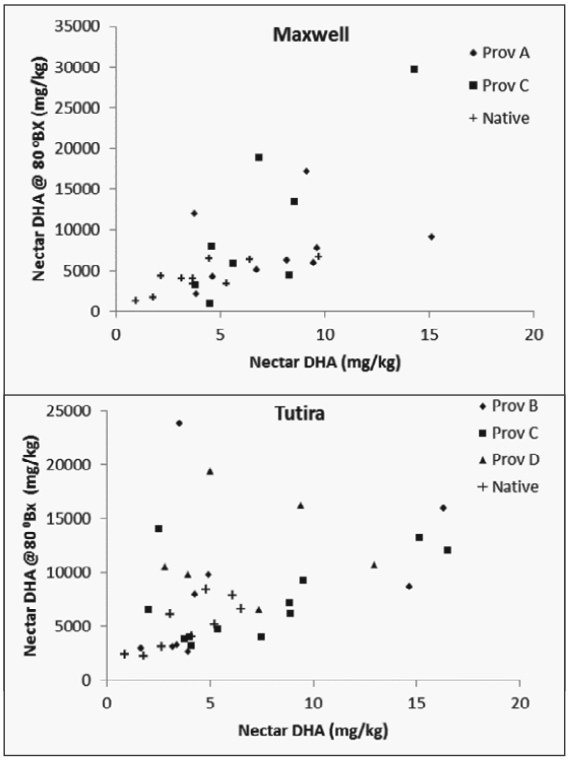

Figure 2 Scatterplots of nectar dihydroxyacetone (DHA) content and normalised nectar DHA content at Maxwell (top) and Tutira (bottom) for indigenous and plantation Manuka in 2014.

Table 2 Growth (±SE) and survival of manuka provenances at Maxwell (September 2014) and Tutira (November 2014).

Provenance	Height (m)	Basal diameter (cm)	Survival (%)
	Maxwell		
A	1.7±0.1	28.1±6.1	<40
C	1.9±0.1	34.8±3.7	> 90
D	1.8±0.2	30.7±4.0	> 90
	Tutira		
B	1.6±0.1	30.2±1.6	>90
C	1.5±0.1	22.9±1.4	>90
D	2.3±0.1	31.0±1.5	>90

Table 3 Nectar dihydroxyacetone (DHA) (mg/kg), sugar (mg/kg) and normalised dihydroxyacetone content (mg/kg 80 °Bx) (± SEM) of plantation and indigenous Manuka at Maxwell and Tutira in 2014.

Provenance	DHA	Sugar	DHA 80 °Bx
		Maxwell	
A	7.8±1.14	961.4±125.6	7757.0±1430.8
C	7.07±1.08	1071.3±361.8	10561±3059.9
Indigenous	4.1±0.81	779.4±85.3	4173.0±594.1
		Tutira	
B	6.2±1.78	711.9±132.8	8677.8±2391.7
C	7.4±1.36	870.7±106.5	7309.5±1116.9
D	6.9±1.55	515.1±140.2	12157.0±1922.8
Indigenous	3.9±0.65	608.6±62.5	5111.4±765.2

variation of nectar sugar content (range 41 to 106%) was greater than that of the indigenous manuka (35 and 31% at Maxwell and Tutira, respectively).

Discussion

Survival of manuka plants 3 years after establishment was variable. Provenance A had relatively poor survival at Maxwell. Initial survival of this provenance has been good at other sites (Hamilton *et al*. 2013) but subsequent browsing by goats has resulted in considerable losses. Goat browsing is an on-going problem at Maxwell and Tutira despite control. Northern provenances have shown greater late winter/early spring growth than indigenous plants suggesting they may be more palatable to goats at a time when browse is limited (Crouchley 1980).

Weed control in establishing manuka is problematic because of the limited choice of effective selective herbicides, particularly those able to kill brush weeds; controlling brush weeds before planting manuka is crucial. Herbaceous species are also capable of supressing manuka seedlings. Pre-plant spot spraying with a mixture of glyphosate and simazine can give good control of grass and broadleaf weeds at low cost (Harrington *et al*. 2015). Post-plant herbicides capable of killing most grass and broadleaf weeds include haloxyfop and clopyralid. However, our experience has been that post-plant spot spraying can be hindered by difficulty finding seedlings once the spring flush of growth has begun. Aerial spraying overcomes this and is cost effective.

The growth of plantation manuka was satisfactory compared with typical height and stem basal diameter growth rates for planted manuka (Tane's Tree Trust 2011). However, comparatively low plant densities in plantation manuka mean that biomass/ha (and carbon) at the time of assessment was low.

The plantation manuka produced normalised nectar DHA levels about double the indigenous manuka indicating potential to produce high UMF® honey (Hamilton *et al*. 2013). Flowering in the plantation manuka occurred earlier than the indigenous at both sites. Provenance A was first to flower, provenance D the last, but there was overlap in the flowering period. Early flowering in provenance A has been reported previously (Hamilton *et al*. 2013).

The nectar DHA levels measured here are high, particularly the plantation manuka. For example, Williams *et al*. (2014) recorded nectar DHA/total sugar ratios in different regions ranging between <0.001 (low) to 0.002 (high) (one region). In this study indigenous manuka had a mean DHA/total sugar of 0.0053 and 0.0064 at Maxwell and Tutira, respectively (Table 3). DHA/total sugar in the plantation manuka ranged from 0.009 to 0.013. The nectar sampling method used by Williams *et al*. (2014) involved washing flowers without bagging compromising the ability to recover sugar; nectar is much more likely to be removed by insects before sampling, if flowers are not bagged.

There was considerable variation in the nectar characteristics at both sites. Williams *et al*. (2014) suggested that rain and insect activity were potential causes of variability, however, the nectar sampling method used (bagging; 24 h rain-free) will have negated these factors. Normalised nectar DHA ranged from <5000 mg/kg to over 20 000 mg/kg (Figure 2). Many of the high normalised DHA results are a consequence of low nectar sugar content. The virtue of these high values is questionable since bees vary their foraging behaviour in response to factors including temperature and nectar sugar content (Roubik & Buchmann 1984).

The nectar DHA contents measured in the plantation manuka at both sites confirms the potential to produce high UMF® honey from plantations. Future work will include measuring honey yield and quality from plantations.

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