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Prediction of nitrogen (N) leaching from urine events using modelling tools such as OVERSEER is important for regulation of dairy farm outputs and practices. Risk of N leaching is greatest during winter, particularly in systems which adopt high stocking densities such as those grazing fodder beet and kale crops. However large prediction errors for leaching values from OVERSEER are recognised, highlighting the need for more quantitative information on urination behaviour of dairy cows grazing winter forages. The study was conducted at Ashley Dene research farm (-43.65 º North, 172.33 º East) using 24 high producing Friesian x Jersey dairy cows fed two diets representative of industry practice. Diet treatments were 10 kg DM of fodder beet with 5.6 kg DM of ryegrass baleage (FB) or 16.4 kg DM of kale with 6.4kg of oat straw (KA) . All cows were fitted with a urine harness for 24 to 48 hours which collected data on number, volume, and timing of urine events. Urine patch areas were estimated using the urine harness data and a calibration curve of urine volume and wetted area in the paddocks being grazed.

Urine volumes were similar (27.6 L/cow/day, p=0.988) though behaviour was affected by diet whereby FB cows urinated less frequently (8.42 vs 10.1 events/cow/day for FB and KA respectively, p=0.128) but with more volume per event than KA (3.58 vs 2.71 L/cow/day for FB and KA respectively, p=0.04). On their own, similarity in total volumes (FB vs. KA) could not be explained by intake of water (55 vs. 69 L/cow/day, p<0.001), N (237 vs. 472 g N/cow/day, p<0.001), DM (13.4 vs. 17.1 kg cow/day, p<0.001), potassium (2.7 vs. 2.9 g/cow/day, p=0.055) or sodium (0.53 vs. 0.40 g/cow/day, p<0.001). The urine patch areas on the FB grazing area were smaller at 0.16m² than the kale patches at 0.23m² which is attributed to differences in paddock surface microtopography. The smaller, more dense deposits of N in urine patches and the stocking rate being three times as great resulted in a higher predicted volume of N leached from the fodder beet paddock at 123 kg/ha.
compared with 82 kg/ha for the kale paddock. The results of this study provide new information regarding the urination behaviour of the livestock and the N losses from winter grazing in New Zealand systems. There is a need for more confidence in the measurement techniques used for future studies.

**Keywords:** New Zealand, Canterbury, *Brassica oleracea*, *Beta vulgaris*, urine depositions, urine paddock coverage, dry dairy cows, urine behaviour.
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New Zealand dairy farmers, especially those in cooler regions such as the South Island, face a major challenge over winter in growing sufficient pasture to meet the energy requirement of the livestock in late gestation (Dalley, 2011). Farmers aim to put body condition on livestock during the period before calving to maximise reproductive performance and therefore overall farm productivity (Edwards et al., 2014). Pasture growth rates in Canterbury over this period range from 0 to 15 kg of dry matter (DM) per day (DairyNZ, 2010) which does not provide adequate nutrition for the typical stocking rates and energy requirements of cows (DairyNZ, 2014; Mandok et al., 2013). It is common practice for dairy cows to graze winter forages such as kale (*Brassica oleracea*) and fodder beet (*Beta vulgaris*) *in situ* off the milking platform as these crops provide high quality and quantity of feed over the entire winter period (Brown et al., 2007; Rugoho et al., 2014).

The low nitrogen (N) requirement of dairy cows in winter relative to the dietary N intake results in excess N being excreted largely in the cow urine which is deposited onto the soil surface at N rates equivalent to 800 - 1300 kg/ha (Eckard et al., 2010; Laubach et al., 2014). The urine from grazing livestock is the main driver of N leaching on dairy farms (de klein et al., 2010) and grazing of winter forages *in situ* results in a high density of urine patches across the paddock due to the high stocking rates able to be fed on this area (Judson & Edwards, 2008; Pleasants et al., 2007). Consequently the leaching from winter forage grazing by dairy cows accounts for up to 24% of the whole farms annual leaching losses despite representing only up to 9% of the farm area (Chrystal et al., 2012).

N from agriculture leached in the form of nitrate into ground and surface water has negative implications for the environment and human and livestock health (Cameron et al., 2002). Current and forthcoming policies seek to regulate and limit the nutrient losses from farms particularly leached N (Williams et al., 2014). This will be achieved with use of modelling software such as OVERSEER which use knowledge of farm inputs and systems processes to predict N losses on an area basis. However incomplete knowledge regarding the processes in grazing systems results in assumptions being used in the models which limits their ability to accurately predict N leaching losses (Cichota & Snow, 2009). Therefore further data is needed regarding N leaching from pastoral farming including from wintering platforms.

For a given soil type and climate it is the N content of urine, the volume of urine deposited, and the timing of urine deposit that determines the level of N leached from pastoral dairy farms (Romera et
al., 2012). There is a need for further knowledge regarding the urination behaviour of dairy cattle as models currently use average values (Cichota & Snow, 2009) of which there are few from dairy cows in New Zealand pastoral systems. The daily urination volume is highly variable ranging from 8.7 to 54.7L (Ravera et al., 2015; Betteridge et al., 1986). Average volumes for urine events have averaged 2-3L (Ravera et al., 2015; Betteridge et al., 2013) and frequency of urination has a large range of 2 to 73 daily events (Betteridge et al., 1986; Betteridge et al., 2013; Ravera et al., 2015; Jenkinson et al., 2014). The wetted area of the paddock surface resulting from a given volume of urine has been found to differ when falling on the surface of the grazing area of dairy cows grazing kale and fodder beet winter forages influencing the concentration of N deposited (Ravera et al., 2015). The volume of urination from dairy cows is influenced by diet DM and crude protein (CP) levels as well as intake of water, sodium (Na), and potassium (K) (Bannick et al., 1999; Khelil-Arfa et al., 2012; Holter & Urban, 1992). The objective of the current research was to provide quantitative information on urination volumes and investigate the influence of factors which drive urine and N outputs.
Chapter 2
Literature Review

2.1 Wintering Objectives

New Zealand dairy farmers, especially those in cooler regions such as the South Island, face a major challenge over winter in growing sufficient pasture to meet the energy requirement of the livestock in late gestation (Dalley, 2011). Farmers aim to put condition on their livestock during the period before calving (Edwards et al., 2014). A lower body condition score (BCS) at calving by 1 unit has been found to extend the post calving anoestrous interval by 7 to 10 days (McDougall et al., 1995) which would have an effect on the animals’ reproductive performance and therefore overall farm productivity. A BCS of 5 at calving is desirable (Judson & Edwards, 2008) thus the energy requirement of dry, pregnant cows putting on condition is 75 MJ ME/day (ARC, 1980) which results in a minimum daily DM requirement of 7 kg DM. Mandok et al. (2013) argued that in New Zealand’s outdoor grazing systems, ME requirement were greater than those reported by ARC and that winter requirement are likely to be closer to 100 MJ ME/d. Given that the average stocking rate in the South Island is 3.04 cows per hectare (DairyNZ, 2014) this would require a pasture growth rate of at least 21 kg DM/ha/day. Pasture growth rates range between 0 and 15 kg DM/ha (DairyNZ, 2010) during this period and grazing cows on heavy soils results in pugging and reduced long-term pasture growth rates. Subsequently in situ grazing of brassica crops over winter off of the milking platform is a common strategy to mitigate this problem though housing cows over winter is has increased in frequency of use in the South Island to mitigate the environmental effects of wintering cows (Chrystal et al., 2012).

2.2 Crop Systems

Kale has been widely used in Canterbury as a winter forage crop for dairy systems due to its high DM yield and quality relative to that of pasture (Brown et al., 2007; Rugoho et al., 2014). Brassica crops have the advantage of growing and accumulating DM with a base temperature of 0⁰C (de Ruiter et al., 2009). Kale crops have been identified to supply 12 MJ ME/kg DM where the quality does not experience much decline over the winter period as the plant matures (Judson & Edwards, 2008). Alternative forages have been proposed, such as fodder beet, with the aim of better aiding the cows to regain their body condition than kale (Greenwood et al. 2011; Rugoho et al. 2014). A suggested major factor in the varying ability of kale to put sufficient condition on dairy cows is the utilisation of the crop which has been found to range. Judson and Edwards (2008) identified the utilisation of kale
to vary from 40 to 90% on commercial farms in the South Island which affected the cows’ ability to consume the target mass of feed and was likely due to inaccuracies in crop allocation. The winter forage crops are often break fed and supplemented with a conserved feed such as straw and baleage to maintain the fibre level in the feed and to modify feed intake rate and behaviour (Judson & Edwards, 2008).

2.2.1 Yield of Crops

Fodder beet is a high yielding crop which produces between 11 and 32.8 t DM/ha (Goh & Magat 1989; Chakwizira et al., 2012). The fodder beet cultivar ‘Rivage’ was grown at the Ashley Dene Research Farm in 2012 and 2013 yielding 18.5 and 21.8 t DM/ha respectively (Edwards et al., 2014). At the same time, kale was also grown on Ashley Dene Research Farm and yielded an average of 14.6 t DM/ha which is in the expected range for the majority of kale grown in Canterbury of 10 to 16 t DM/ha (Judson & Edwards, 2008). Major factors determining the yield of the crop are the sowing date, cultivar, and location (soil and climate). Edwards et al. (2014) showed fodder beet to be more efficient in its use of N fertiliser with more than double the yield of feed per kg of N fertiliser applied compared with kale at 99 and 44.9 kg DM/kg N respectively. Fodder beet was effectively eaten by cows achieving 99.5% utilisation, 10% higher than was achieved for kale in the same study. Both kale and fodder beet are relatively low cost feeds to grow at 9-11 c/kg DM for fodder beet and 12.6 c/kg DM for kale (de Ruiter et al. 2007; DLF Seeds, n.d.; Gibbs, 2011).

2.2.2 Composition of Kale and Fodder Beet

The two crops have similar energy content at 12.2-13.5 MJ ME/kg DM (table 2.1), though the CP and fibre content of kale is greater than that of fodder beet. The forages have similar digestibility at 80-85 g/100g DM. Fodder beet mineral content was analysed across sites in the South Island and seasons by Gibbs (2011) where calcium (Ca) content was determined to be 0.35% DM. K content has been identified at 9.7 g/kg DM and Na at 1.1 g/kg DM (Chakwizira et al., 2013). Kale Ca levels are in the range of 20 to 40 g/kg DM, magnesium (Mg) at 1-7 g/kg DM, Na at 1 g/kg DM, and K at 17-50 g/kg DM (Nutrimix, n.d.; de Ruiter et al., 2009). Water content is greater in kale though the variation in DM content, CP and fibre are often compensated for by the type and amount of supplement fed, making the diet winter diet similar for the two systems.
Table 2.1 Nutritional composition of fodder beet and kale obtained from various trials at Ashley Dene Research Farm from 2010 to 2013.

<table>
<thead>
<tr>
<th></th>
<th>Fodder Beet</th>
<th>Kale</th>
</tr>
</thead>
<tbody>
<tr>
<td>DM%</td>
<td>16.5</td>
<td>14.0</td>
</tr>
<tr>
<td>ME (MJ/kg DM)</td>
<td>12.2</td>
<td>12.8</td>
</tr>
<tr>
<td>N (%DM)</td>
<td>1.7</td>
<td>1.7</td>
</tr>
<tr>
<td>CP %DM</td>
<td>10.6</td>
<td>10.9</td>
</tr>
<tr>
<td>NDF (%DM)</td>
<td>20.6</td>
<td>17.1</td>
</tr>
<tr>
<td>ADF (%DM)</td>
<td>-</td>
<td>10.4</td>
</tr>
<tr>
<td>DOMD (g/100g DM)</td>
<td>-</td>
<td>80.3</td>
</tr>
</tbody>
</table>

DM=dry matter, ME=metabolisable energy, N=nitrogen, CP=crude protein, NDF=neutral detergent fibre, ADF=acid detergent fibre, DOMD=digestible organic matter.

2.3 Requirements of Non-Lactating Dairy Cows in Late Gestation in Winter

2.3.1 Energy Requirement

Non-lactating dairy cows in late gestation grazing winter forages require energy from their diet to meet their needs for maintenance with a low activity level, for the gain in liveweight associated with their increasing BCS, and to support gestation. Roche et al. (2005) determined the energy requirement of cows in late gestation to be 1.05 MJ ME/kg LWT\(^{0.75}\)/day which for a 510 kg cow gaining condition during the winter period resulted in a daily energy requirement of 127 and 129 MJ ME for cows grazing kale and fodder beet respectively. Studies have highlighted the disparity between the energy requirement of non-lactating dairy cows in late gestation, their energy intakes, and their performance (Edwards et al., 2014; Greenwood et al., 2011). Where the estimated daily energy requirement to enable cows to reach performance targets have been up to 44 MJME lower than their apparent intakes. This could be due to underestimations in energy requirement for maintenance and liveweight gain (Roche et al., 2005; Mandok et al., 2013), higher intake to meet metabolisable protein demands on forages with low CP content (Westwood & Mulcock, 2012) at 12.6 and 10.6% DM for kale and fodder beet respectively (Edwards et al., 2014), and/or the consumption of anti-nutritional compounds such as S-methylcysteine sulfoxide in the kale which can limit performance (Barry & Manley, 1985). It is also possible that the energy supply of the feed is overestimated due to the negative effect that high intake and the cold environment can have on the digestibility of the feed not having been taken into account (Mairon & Christopherson, 1992).

Edwards et al. (2014) determined apparent intake of non-lactating dairy cows in late gestation to be 13.1 kg DM for cows fed fodder beet and ryegrass baleage and 14.2 kg DM for cows fed kale and oat straw. The daily intake of cows fed kale and oat straw was similar to that determined by Miller et al. (2012) which was 14.5 kg DM. The intakes were considered to be high partially due to the low NDF...
content of the forages at 26.3% DM for the kale crop and 20.6% DM for the fodder beet crop. The relatively low fibre content of the forages increased the intake as there was less physical limitation due to gut fill (Mertens, 1994). This intake led to energy intakes of 168 and 155 MJ ME/kg DM for cows in the kale and fodder beet treatments respectively.

2.3.2 Protein Requirement

A major inefficiency regarding N in pastoral dairy systems is the greater N requirement of plants for optimum growth relative to the lower dietary N requirement of grazing livestock (Ledgard et al., 2000). Dairy cattle have a minimum requirement for N in their diet which, if not met, can limit production which is 1.8% of DM and is greater for young/lactating or growing cattle at 3 and 2.2% of DM respectively (Pacheco & Waghorn, 2008). A large (675kg) non-lactating dairy cow in late gestation requires CP in her diet as 12% of DM (Merck Manuals, 2014) equating to 1.92% DM as N. Kale forage supplies 2 to 2.2% DM as N (Miller et al., 2012) and fodder beet supplies 1.7% DM as N (Edwards et al., 2014; Jenkinson et al., 2014). Any imbalance between the animal requirement and forage content of N causes large amounts of N to be excreted (Eckard et al., 2010). Laubach et al. (2013) identified that 88.5% of the ingested N was excreted of which 68.8% of that was in urine. N intake of 400 g/day is the critical point above which the urinary N output increases exponentially with increasing N intake (Castillo et al., 2000). Kebreab et al. (2001) identified that increasing N content in the feed results in greater N concentrations in urine. Faecal N output increases with a constant gradient of around 7.5 g N per 1kg DM of feed ingested (Castillo et al., 2000).

2.3.3 Mineral Requirement

The demands of livestock for minerals are greater for higher producing animals however they are fairly consistent on a per kg DM of feed basis where changes in feed intake provide the correct level of minerals in the diet (Sykes, n.d.). Lactating 500 kg dairy cows producing 10 to 30L of milk daily require Ca and Mg in the order of 3.2 and 0.8-1.4 g/kg DM respectively (Grace, Knowles, & Sykes, 2010). The mineral requirement of a large (675 kg) dairy cow in late gestation are 0.48, 0.40, 0.14, and 0.62% DM for Ca, Mg, Na, and K respectively (Merck Manuals, 2014). Dairy cattle in late gestation have a requirement for Ca in the diet of less than 60 g/kg DM where a greater supply can restrict the cows’ ability to mobilise Ca reserves post calving (de Ruiter et al., 2009).

2.3.4 Water Requirement

Although water provides no energy to an animal, it is a vital part of their diet and makes up 60% of their mass (Keenan, 1988). Water is essential to dairy cows for all of their life processes including elimination of waste materials, dissipation of excess body heat, and maintenance of osmotic
pressure in cells and tissues (Khelil-Arfa et al., 2012). Water is constantly lost from the body and must be replenished in order to maintain hydration. Water enters the body through direct consumption during drinking from a water source such as troughs, as well as indirectly through water in feed and water produced during metabolic processes and the oxidation of nutrients. Daily water intake is highly variable in dry cows and was found by Paquay et al., (1970a) to range from 8-64 L. Water intake has been found to be affected by ambient temperature as a behavioural regulator of water consumption when it exceeds 27°C (Senn et al., 1996). It is expected that at ambient temperatures outside cows thermal neutral zone of -5 to 21°C (Meyer et al., 2004) thermoregulatory processes such as shivering and sweating would be activated which would alter the body’s water usage. Feed moisture content is well correlated with daily water intake ($r^2$=0.87) and intake of a diet with 13.5-20% DM will result in a water intake of 5.5 kg for each additional kg of DM consumed (Parquay et al., 1970b).

2.4 Factors Affecting N Leaching

2.4.1 Urine Patch Dynamics

It is the excess dietary N supplied to livestock which results in urine patches depositing N in concentrations of 800-1300 kg/ha (Eckard et al., 2010) of which 8 to 20% is leached (Cameron et al., 2002). Typically the major driver of N leaching on dairy forms is urine from grazing animals rather than applications of fertiliser or effluent (de Klein et al., 2010). The effects of N fertiliser use on N losses are indirect in that increased use of N fertiliser results in greater pasture production and therefore stocking rates incurring greater deposits of N in concentrated form (figure 2.1). It has been proposed that grazing forages with elevated levels of condensed tannins and water soluble carbohydrates to lower urinary N concentration (Waghorn et al., 2007; Woodward et al., 2004). However species which would achieve this such as Lotus corniculatus and Lotus pedunculatus are not suitable as winter forages as they do not yield the same quantity of feed as currently used crops such as kale (Miller et al., 2012). There is potential to supplement a winter forage diet based on grazing kale with conserved feed with elevated levels of condensed tannins (Miller et al., 2012; Powell et al., 2009).
For a given soil type and climate it is the N content of urine, the volume of urine deposited, and the timing of urine deposit that determines the level of nitrate leached from pastoral dairy farms (Romera et al., 2012). Urine is deposited unevenly across a paddock in patches (Di & Cameron, 2002) with frequencies of deposition which are greater in some areas such as gateways in pastoral dairy farming (Matthew et al., 1988; McDowell, 2006). The patches have been found to overlap (figure 2.2) which has been suggested by modelling to have the effect such that a double urine patch would leach three times the N as a single patch (Shorten & Pleasants, 2007).

Figure 2.1 Annual losses per hectare of pasture with increasing stocking rate of dairy cows. Modelling by McGechan & Topp (2004).

Figure 2.2 The incidence of single and double urine patch areas with increasing stocking rate of dairy cows on pasture (Pleasants et al., 2007).
The paddock coverage of urine patches depends on the type of animal grazing and the stocking rate (Pleasants et al., 2007). Cattle urine patches of a 2 L volume covering an area of 0.38 - 0.42 m² have been found to penetrate to a depth of 400 mm where 11% of the N applied in the urine patch was leached (Williams & Haynes, 1994). Romera et al. (2012) determined that 8% of a pasture paddock area grazed by dairy cattle was covered in multiple urine patches where 39% of the total urine volume was deposited on overlapping patch areas. On a typical pastoral dairy farm 85% of the urine patches are deposited in the paddock with the remainder on raceways and cattle yards (Draganova et al., 2012). Moir et al. (2011) identified that an increase of 10,000 grazing hours/ha/year results in a 33% increase in the surface area of the paddock covered in urine patches annually (figure 2.3). Where 5 days of 300 to 400 cows grazing a paddock equates to more than 6000 grazing hours.

![Figure 2.3 Annual dairy cow grazing hours on paddock vs. proportion of paddock covered in patches r²=0.77 (Moir et al., 2011).](image)

### 2.4.2 Factors Affecting Urine Volume

#### 2.4.2.1 Water Loss

In dry dairy cows the majority of water loss from the body is in the form of urine as well as being excreted in faeces and evaporated from body surfaces including the skin and respiratory passages (Frandson et al., 2006). Losses in faecal matter have been found to be between 4 and 30 kg/day which can be affected by dietary crude fibre (r=0.50) (Parquay et al., 1970b). It was also found that the water loss in the form of faeces is also influenced by the water content of feed (r=0.52), congruous with the findings of Holter and Urban (1992). Moisture losses through saliva and sweat were estimated to account for 18% of total daily water losses in lactating dairy cows in a study by...
Holter and Urban (1992). Church et al. (1988) observed that losses of water from respiratory passages as a result of inspired oxygen is influenced by ambient temperature.

2.4.2.2 Urine in Cows

The kidneys regulate the body’s water, PH, and electrolyte balance through adjusting the composition and volume of urine which is formed there. This occurs in response to changes in metabolism or intake of feed and water (Frandson et al., 2006). Therefore the volume of urine excreted depends on the level of fluid and minerals needing to be expelled from the body. Animals fed a diet with a greater CP content consume more water and therefore will excrete a greater volume of water in their urine (Bannik et al., 1999). Khelil-Arfa et al. (2012) found that the feed CP content was well correlated to urine output ($r^2$=0.77) were is was the major determining factor in the volume of urine excreted. Holter and Urban (1992) found that dietary CP was very strongly correlated to urinary output volume ($r^2$=0.92) with DM intake and feed DM content also being significant factors. Urine production is also affected by the levels of Na and K that are ingested (Bannick et al., 1999). The volume of urinary output has been found to vary from 1.1 to 32.8 kg per day in non-lactating dairy cows and is the greatest contributor to urea excretion (Parquay et al., 1970b) and to daily water loss (Parquay et al., 1970a).

2.4.3 Urination N Content

The urine N concentrations have been identified to be lower for cows grazing kale and fodder beet as a winter forage crop at 2.3 to 4.0 g N/L (Miller et al., 2012; Ravera et al., 2015) than concentrations measured for cows grazing traditional pastures of ryegrass and white clover at 5.7 g N/L (Totty et al., 2013) and 6.1 g N/L (Edwards et al., 2015). The lower N content in the urine is suggested to be due to the greater water content of the kale diet resulting in a diluting effect (Ledgard et al., 2007) and the lower N content of the diet. Urinary N levels of grazing dairy cows were found to average 9.5 g N/L ranging between 1.2 and 24.7 g N/L (Betteridege et al., 2013). Jenkinson et al. (2014) identified that although the total N intakes of non-lactating dairy cows in late gestation were significantly different at 289 g/kg DM for those fed kale and 228 g/kg DM for those fed fodder beet winter forages where total DM intake was similar, the urinary (at 2.2 g N/L for both treatments) and faecal (at 2.1% of DM for both treatments) N contents were not significantly different. This agrees with the findings of Edwards et al. (2014) that urinary N content did not significantly differ between cows grazing either kale (2.3 g N/L) or fodder beet (2.1 g N/L).
2.4.4 The Volume of Nitrate Leached

The conditions most favouring heavy N leaching are a high level of N in the form of nitrate in the soil followed by or coinciding with a period of heavy drainage through the soil. Nitrate is readily leached when there is drainage as it is negatively charged so is repelled by the clay content of soils in temperate regions (Di and Cameron, 2002). Therefore the major determinants of the degree of N leaching are the level of N accumulated in the soil which is a result of numerous factors (figure 2.4) and the degree of drainage through the soil occurring (Cameron, Di, & Moir, 2013). Major factors contributing to the level of N leaching include land use season, climate, and the soil properties.

![Conceptual model of N inputs and outputs for the potentially leachable N pool (Di & Cameron, 2000).](image)

2.4.5 Seasonal Changes and Climatic Conditions

The majority of nitrate leaching occurs during late autumn, winter, and early spring for many areas of New Zealand largely due to the rate of evapotranspiration being exceeded by the rate of moisture deposition due to rainfall. This results in nitrate being leached when coinciding with the soil being at or near field capacity (McLaren and Cameron, 1996). These times of year are generally when the temperatures and therefore plant growth are low leading to reduced nitrate uptake by plants. The nitrate levels then build up in the soil which, in conjunction with high rainfall and drainage, causes winter to typically be the period of greatest nitrate leaching.
2.4.6 Soil Profile and drainage

The rate at which water drains through the soil directly is determined by the soil’s texture and structure and affects the level of N leached which is greater in soils with more drainage (McLaren and Cameron, 1996). At greater rates of drainage less N is removed from the soil solution through denitrification, immobilisation, and plant uptake (Cameron et al., 2002). N leaching occurs more quickly in lighter sandy soils as they have a lower field capacity than clay and silt loam soils (Cameron et al., 2002). Drainage is also influenced by the soils’ porosity which is increased by earthworms, plant roots, or wetting and drying cycles (Cameron et al., 2013). Water movement through the soil profile is also increased by artificial agricultural drainage systems which have been found to increase N leaching. For example a paddock with molepipe drainage in a continuously grazed beef system had consistently 70 kg N/ha/yr greater leaching than a similar paddock without artificial drainage (Schofield et al., 1993).

2.4.7 Land Use

Land use intensification typically increases N leaching (McLaren and Cameron, 1996) as the total level of N inputs increases the risk of N leaching increases exponentially regardless of the type of N input (de Klein et al., 2010). Intensively grazed systems such as New Zealand pastoral dairy farms have relatively high N leaching levels due to high stocking rates and large N fertiliser inputs. Dairying systems with fertiliser inputs of 360 kg N/ha/yr are highly intensive uses of land which can leach up to 110 kg N/ha/yr. Grazing winter forages in situ contributes a disproportionate amount to the farm’s total N leaching profile. The high level of N leaching over winter is due to high stocking rates causing a dense deposition of urine patches, low temperatures leading to low plant growth rates, and the use of intense soil drainage systems (Dalley, 2011). The high yielding forage crops such as kale and fodder beet are typically break fed with high stock numbers to harvest the forage resulting in dense stock numbers on small area (Judson & Edwards, 2008). Although winter grazing represents only 4 to 9% of the total farm system area, it is responsible for 11 to 24% of the farm’s total N losses through leaching as displayed in figure 2.5 (Chrystal et al., 2012). A trial (Chrystal et al., 2012) modelled six farm systems determining that the nitrate leaching losses for a typical farm system in which stock graze winter forages over the entire dry period are the greatest. It was identified that the winter grazing leached around 58 kg N/ha/year which was more than twice the level of the 23 kg N/ha/year leached from main farm block. All of the systems in which stock grazed winter forages followed the same pattern in that the leaching during the winter period was twice that of the home farm block.
2.5 Regulation of N Losses from Dairy Farms

2.5.1 Consequences of Nitrate Leaching

N leaching in the form of nitrate (NO$_3^-$) from agricultural systems increases the nitrate levels in ground and surface waters which poses a threat to its quality for wildlife, recreation, and human consumption (Cameron et al., 2002). Elevated nitrate levels in drinking water are considered to be especially harmful to the health of infants as it can lead to the disorder methaemoglobinaemia which interferes with the oxygen carrying capacity of the blood (Cameron et al., 2002). Levels of greater than 40-100 mg NO$_3^-$/L are considered to be potentially dangerous to livestock for which it can cause abortions and methaemoglobinaemia in cattle (Di and Cameron, 2002). Nitrate leached into waterways such as rivers, estuaries, and lakes can cause environmental damage due largely to its contribution to eutrophication where algae blooms excessively at the expense of aquatic life (Di and Cameron, 2002).

2.5.2 Regulations and Use of OVERSEER

The National Policy Statement for Freshwater Management came into effect July 1$^{st}$ 2011 mandating that regional authorities set and manage land uses within water quality limits (Williams et al., 2014).
This was in response to agricultural N leaching losses hence these regional authorities are developing plans to manage water quality and reduce nutrient levels in surface and groundwater with particular focus on N and leaching from agricultural sources (Williams et al., 2013). The plans aim to regulate nutrient losses rather than limiting inputs which presents a challenge as losses are more difficult to monitor and quantify (Williams et al., 2013). Environment Canterbury, Otago Regional Council, Environment Southland, Waikato Regional Council, and Environment Bay of Plenty currently specify using OVERSEER to estimate nutrient losses from agricultural properties (Williams et al., 2013). The initial purpose of OVERSEER was to aid fertiliser and nutrient management on pastoral farms (Cichota & Snow, 2009; Williams et al., 2013) however it has undergone further development to evaluate farming systems, their nutrient losses, and environmental effects (Cichota & Snow, 2009). Empirical relationships, readily available data from “existing” farms, and internal databases are used in the OVERSEER model to estimate paddock nutrient inputs and outputs and present them as a nutrient budget (Cichota & Snow, 2009). It is therefore well suited to New Zealand environmental conditions and management practices and requires minimal inputs of data that are easily obtained by the farmer and are significant aspects of a farming system (Cichota & Snow, 2009; Williams et al., 2013). Incomplete knowledge of how natural processes and systems occur produces the need for assumptions to be used in models which results in them having limitations hence models should be considered as only simplified descriptions of natural processes. Although models have a large degree of uncertainty, they are continually updated with improving knowledge of the processes and systems involved which improves reliability (Cichota & Snow, 2009). There is a need for further knowledge regarding the urination behaviour of dairy cattle as models currently use average values (Cichota & Snow, 2009) of which there are few from dairy cows in New Zealand pastoral systems and grazing winter forages in situ.

2.6 Current Data on Urine Volumes

There is a large degree of variability of the urination behaviour of dairy cows hence most of the models used to describe N cycling in a farming system use average values (Cichota & Snow, 2009). The urination events of dairy cows varies greatly in terms of their frequency, volume, and concentration of N and there is currently limited data available to quantify the frequency and volume variables. The majority of the published information on urination behaviour has been conducted with cows in metabolism crates rather than grazing in the field.
2.6.1 Urination Volume and Frequency

A study conducted on grazing steers (Betteridge et al., 1986) found that urination behaviour over a 24 hr period varied greatly. Frequency varied from 13 to 73 times and total daily urination volume ranged from 5.8 to 54.7 L. Betteridge et al., (2013) found that the average volume of a single urination of a grazing dairy cow was 2.1 L ranging from 0.3 to 7.83 L per urination. Villetta and Robichaud et al., (2011) and Aland et al. (2002) found the number of urination events over 24 hrs for housed dairy cows to have a large range of 7 and 3 to 19 averaging 9.3 events per day. Similarly Castle et al. (1950) reported urination frequency to be 9.8 for grazing dairy cows. Other studies have found the number of urination events per cow per day to average around 14 (McLeod et al., 2009; Draganova et al., 2010). Jenkinson et al. (2014) determined grazing dry cows in late gestation grazing either kale or fodder beet to not have significantly different urination behaviour with similar urination frequency per cow in a six hour period (an equivalent of 10.2 and 11.6 events/day respectively) and duration of each event (9.3 and 8.9 seconds respectively).

Ravera et al. (2015) studied the urination behaviour of pregnant dry dairy cows grazing kale and fodder beet over winter using a urine harness. The frequency of urination events ranged from 8 to 21 events per day for cows grazing kale where cows grazing fodder beet was lower ranging from 3 to 11 events. The total daily urine output for cows grazing kale was greater ranging from 19.2 to 47.3 L and ranging from 8.7 to 25.2 L for cows grazing fodder beet. The average volume for a single urine event was similar for both treatments at 2.37 L and this varied from events less than 0.5 L and those up to 8.6L. The events of larger volume (above 5 L) were found to occur between 0600 and 0730 hrs or between 2200 and 0000 hrs. There was great variability between individual cows in the groups grazing each treatment with low replicate numbers (5 for kale and 8 for fodder beet) hence identifying significant differences between the urinary behaviour of cows in each group was difficult. The urine patch area in the paddock was investigated for each forage crop where patches were smaller for fodder beet (area=0.109*volume) for a given urine volume compared with patches in the kale paddock (area=0.190*volume). The trial found the average volume for a single urine event to be 2.37 L for both treatments which would result in a urine patch area of 0.47 m² on a kale paddock and 0.25 m² on the fodder beet paddock. The paddock leaching loss estimations were 77.82 kg/ha/yr for the fodder beet paddock and 53.79 kg/ha/yr for the kale paddock. However, the study by Ravera et al. (2015) represents only a single year and a small sample size. More data is required to gain confidence in previous findings and to improve prediction accuracy in future models.
Chapter 3
Materials and Methods

3.1 Experimental site and design

The experiment was conducted at the Lincoln University Ashley Dene research farm (-43.65 º North, 172.33 º East) with the approval of the Lincoln University Animal Ethics committee (#620) from the 23rd of June to the 18th of July 2015. The soil type was Lismore/ Balmoral shallow stony loam soil structure. The site was converted from dryland pasture to irrigated dairy support land in 2011.

The experiment was a cross-sectional study in a randomised complete block design to determine the effect of diet on urine volume and frequency of dairy cows in late gestation. The trial was repeated over 4 weeks with each week acting as a temporal block and 8 animals used each time totalling 32 cows. Animals for this trial were selected from an existing winter feed trial where cows had already adjusted to the treatment diet for over a month. The winter feed trial had 50 pregnant, non-lactating, spring calving, Friesian x Jersey cows from the Lincoln University Research Dairy Farm in each treatment which were blocked according to calving date, BCS, liveweight, age, and breeding value. The two treatments in this study were either a kale or fodder beet winter system. These treatments were chosen as representative of current industry practices. Cows for the trial in this study were selected (from cows in paddocks 1, 3, 7, and 10) using restricted randomisation as the sample size was relatively small (n = 12) and urine volumes are known to have large between-cow variability (Ravera et al. 2015). Consequently animals of similar liveweight and days since conception were used.

For each block the pen layout was the same but a new area in the paddock was set up at the end of each week. The dates during which each repetition was completed were: the 23rd to the 26th of June, 31st of June to 3rd of July, the 6th to the 10th of July, and the 15th to the 18th of July. These dates were chosen due to being largely during the week when various technicians and resources were available. They also best afforded the opportunity to obtain 48 hrs of data from each subject. The first 3 weeks used 4 animals of each treatment and the 4th used 5 cows of the fodder beet treatment and 3 of the kale treatment. Hence a total of 17 cows were used in the fodder beet treatment and 15 in the kale treatment.

There were 2 diets based on kale (cultivar, Regal) and fodder beet (cultivar, Rivage) winter forage crops. The kale was cut from crop paddock 1a-c where is was direct drilled into oat (Avena sativa L.) stubble on
the 17th October at a rate of 5 kg/ha. Applied to the crop was 200 and 15 kg/ha of diammonium phosphate and boron (10% B) at sowing, and 102 kg N/ha urea was applied to the crop as it grew. The fodder beet was taken from paddock 3a-c which was sprayed on the 15th of September with Norton + Bentanal Forte + Goltix then ploughed on the 25th. On the 7th of October the paddock was sprayed with the same mix of herbicides before being cut and rolled. The seed was then sown on the 20th of October at a rate of 100,000 seeds per hectare and 250, 350, 200 and 15 kg/ha respectively of CropMaster 20, Na chloride, K chloride and boron (10% B) were applied at sowing. At two points during the growing season 85 kg/ha. During the growing season the crops were maintained under a lateral irrigator. Plate 3.1 indicates the area at Ashley Dene Research Farm where the trial was conducted which is in paddock 4 in the purple area. Forage was taken from the northern ends of paddocks 1 and 3a.

Plate 3.1 Site Map

3.2 Animals and Management

All cows were selected from cows from the Lincoln University Research Dairy Farm which were already transitioned onto the forages. 32 crossbreed Friesian x Jersey cows in total were included in the study. Cows used in the trial were 6 ± 0.46 years of age with a liveweight of 513 ± 8.6kg, a BCS of 5 ± 0.09, and a breeding worth of 120 ± 8.3. The milk production during the 2014/15 season averaged 353 ± 14.2 kg milk solids and their calving due date ranged from the 30th of July to the 5th of September.

Cows were kept in pens formed of temporary electrical fencing which measured 48m² with water available ad lib (plates 3.2 and 3.3). Cows were offered 16.43 kg DM of kale supplemented with 6.38
kg DM of oat straw or 10.04 kg DM of fodder beet supplemented with 5.59 kg DM of ryegrass baleage. The supplement was fed at 0800 h and the forage at 1000 h which was cut and carried to the cows. All feed offered and refused was weighed using a trailer with inbuilt scales.

Plate 3.2 Cows in kale treatment wearing urine harnesses consuming supplemented oat straw. Plate 3.3 Cows in fodder beet treatment consuming forage ration.

Plate 3.4 is a representative diagram displaying the penned area of the trial which is located at the southern end of paddock 4 at Ashley Dene Research Farm. Each pink or purple square represents to 48m² pen in which a cow lived during her time in the trial. The pink pens housed cows in the kale treatment and the purple housed cows in the fodder beet treatment. Each pen had a water trough and a gate which opened into the alley between the rows of pens for each treatment. Supplement and forage were fed along opposite fence lines for each pen i.e. along the east or west side. Water intake from troughs was measured through the use of a ruler where the height of each trough was recorded at 2pm each day before and after the re-filling of the trough.
3.3 Sample Analysis

3.3.1 Dietary Composition Analysis

Representative samples were taken from the diet offered each week and refusals each day for both treatments. Forage samples were washed and divided into leaf (including petiole) and stem/bulb. The components were then weighed to determine the leaf:stem/bulb ratio of the offered and refused diet. One set of sub samples of all forage components and supplement was oven dried at 60°C for 48 hours and reweighed to determine DM content. Another set of sub samples were frozen and freeze dried. The freeze-dried samples were then ground through a 1-mm sieve, and scanned by near infra-red spectrophotometer (NIRS, NIRSystems 5000, Foss, Maryland, USA) to determine forage digestibility, fibre, and CP content. Remaining freeze-dried samples were analysed by an Elementar (Variomax CN Analyser. Analysensysteme GmbH, Hanau, Germany) to determine the content of minerals: Ca, Mg, K, and Na.

3.3.2 Urine Composition Analysis

Urine samples were collected from 7 cows in each crop treatment during the trial period. Urine samples were taken mid-stream after manual stimulation of the vulva, acidified below a pH of 4.0 using concentrated sulphuric acid to prevent volatilization, and then stored at -20°C. Thawed samples were assessed for urine N% using an N-analyser (Vario MAX CN, Elementar Analysensysteme, Hanau, Germany). Urea, ammonia, and creatinine contents were also determined (RX daytona Benchtop Clinical Chemistry Analyzer, Randox Laboratories US Limited).
3.3.3 Faecal Measurements and Composition Analysis

To determine moisture and N losses from faecal events the number of faecal events were recorded and faecal samples were taken from defecations in the cows’ pens after their first 24 hours there. Representative samples were taken into a 400 ml container from beneath the surface crust of each faecal patch. Each sample was stirred before subsampling into a smaller 60 ml container. Sub samples were either dried at 100⁰C for 48 hours to determine DM content or frozen then freeze dried and ground through a 1mm sieve for composition analysis. The N% was determined through the use of an Elementar (Vario MAX CN, Elementar Analysensysteme, Hanau, Germany) and the OM% through furnace combustion at 550⁰C for 4 hours.

3.4 Urine Volume and Frequency

Urine volume and frequency was determined using the urine harness developed by Ravera et al. (2015). In this system a flow meter connected to a glove through which the cow urinated sent information to the data logger which was carried by the cow in the pocket of a cow cover (plate 3.5). The data logger automatically recorded the time, duration, and volume of urine flow through the meter.

Plate 3.5 Urine harness attached to cows (Ravera et al., 2015).

Urine event recordings with flow rates of less than 80 mL per second were removed to produce a data set of “true urination events”. The small events with low values were deemed likely to have
been due to movement of urine through the meter due to shaking of the meter as the cows moved. Incomplete data sets without a minimum of 24 hours of recorded urinations were removed as were sets with numerous unusually high values i.e. above 40 L. On occasion the urine harnesses failed either due to equipment failure or cows breaking the equipment. This resulted in 12 cows from the fodder beet and 14 from the kale treatment with at least 24 hours of usable data.

3.4.1 Harness Validation Measurements

Urination was stimulated for cows wearing the harness and the urine event was collected in a measuring jug with the time of event recorded. Observed urine events of cows wearing the harness also had their time recorded. This data was then compared to the data logger output. Figure 3.1 displays the relationship between measured and flow meter values to be close to 1.0 the goodness of fit was lower than expected resulting in a standard error of the estimate of 500 ml.

![Figure 3.1 Quadratic regression of measured versus flow meter volumes from urine events. Regression $y = 1.0075x$, r-sq = 0.61, standard error of the estimate = 0.512.](image)

3.5 Urine Patch Area

To determine ground coverage of urine events on kale and fodder beet areas, a calibration curve was produced by pouring measured quantities of water from the average height of cow vulva onto the grazing areas of each treatment. Volumes poured were 1000, 1500, 2000, 2500, 3000, 3500, 4000, 4500, and 5000 ml with 4 measurements taken for each volume. Fluorescent paint was sprayed around the perimeter of the wetted area and a photo was taken including a fencing standard for scaling (plate 3.6). Care was taken to ensure the entire patch was included in the photos which were analysed using the software SketchandCalc to calculate the patch area.
3.6 Calculations of Leaching Loss

Paddock leaching losses were calculated using the following equation:

\[ N_L = (N_{L1} \times P_1) + (N_{L2} \times P_2) \]

- \[ N_L \] = annual average NO\(_3\)-N leaching losses from a grazed field
- \[ N_{L1} \] = N leaching losses at the urine patch
- \[ N_{L2} \] = N leaching losses at non-urine patch areas
- \[ P_1 \] = proportion of area covered by urine patch areas
\( P_2 = \) proportion of area covered by non-urine patch areas

Lysimeter data from the site was collected in 2014 which was used for the leaching loss estimations (Cameron, unpublished). From that data the N leaching losses used for non-urine patch areas were estimated to be 16 kg N/ha for the fodder beet treatment, and 17 kg N/ha for the kale treatment. In order to estimate leaching losses for urine patches the N load per average urination was multiplied by the percentage of N load leached.

The urine patch N load = \( \frac{\text{urine } N \text{ conc } \times \text{average urination event volume}}{\text{average urine patch area}} \). The percentage of N load being leached was taken from the 2014 lysimeter data (Cameron, unpublished) and was estimated to be 34% of the fodder beet treatment and 55% for the kale treatment. The proportion of paddock area covered by urine patches was calculated as:

\[
\text{Covered in Urine Patch (%) = } \left( \frac{\text{average number urinations per 24hrs}}{\text{total area grazed}} \right) \times (\text{number cows}) \times (\text{days on paddock})
\]

The area not covered in urine patches could then be estimated through subtracting the obtained value from 1. The values for these areas could then be inserted into the equation to estimate paddock leaching losses.

### 3.7 Statistical Analysis

All data was processed using Microsoft Excel 2013. All diagrams and graphs were produced using Microsoft Excel 2013. The intake of minerals, water, CP, fibre, etc and the composition of animal samples were assessed between treatments using one-way ANOVA and Minitab 16. A regression model was used to calculate the slope coefficient and its standard error of equations for the urine patch calibration for each treatment.

The urination data was analysed using Minitab 17 with a General Linear Model. Ten cows of each treatment had 48 hours of urination data from the harness logger and another 6 cows in total had only 24 hours of data. There were unequal numbers of data sets from each temporal block e.g. 8 for kale and 4 for fodder beet. One-way ANOVA was performed to assess any differences between the urination behaviour for cows with 24 hours or 48 hours of data which yielded no significant results. In order to use all of the data obtained each set of 24 hours of data was treated as independent regardless of some sets coming from the same cow. The non-independence of samples and the small, unequal sample sizes resulted in urination behaviour data which breached ANOVA assumptions. A general linear model was used to incorporate a number of statistical tests including ANOVA and t-tests. One-way ANOVA was performed to compare the urination behaviour of cows
included in each week and several statistically significant differences were found hence the temporal blocks were included as fixed factors in the general linear model analysis of urination behaviour where the diet treatment was the random factor.
4.1 Climatic Conditions

Climatic conditions during the experiment are presented in figures 4.1 and 4.2. The mean maximum temperature was 11.5°C and the mean minimum temperature 0.8°C. Minimum temperatures fell below zero on 11 of the days and total accumulated rainfall was 36 mm over the experiment.

Figure 4.6 Maximum and minimum temperatures during the experimental period

Figure 4.7 Rainfall and wind speed during the experimental period
4.2 Diet Composition

Table 4.1 displays the composition of the diet components for cows in each treatment. At time of study the crop yields from paddocks being sampled were 24.8 ± 2.54 t DM/ha for fodder beet and 14.3 ± 1.44 t DM/ha for kale (de Ruiter, unpublished). Compared with fodder beet, the leaf constituted a greater proportion of the forage DM for the kale plants and the DM% was similar for each of the diet components i.e. supplement, leaf, and stem/bulb. The fibre content was greatest in the supplement which had lower DM digestibility. The CP content of fodder beet bulb and kale stem were similar at 15% of the DM. However CP in leaf of kale was nearly three times that of fodder beet leaf. Apart from N, mineral concentrations tended to be greater in the stem or bulb of both crops. K and Na were higher in fodder beet while Mg was relatively low in both crops.

Table 4.1 Nutrient and mineral composition (g/kg dry matter unless otherwise stated) of kale and fodder beet and supplements

<table>
<thead>
<tr>
<th></th>
<th>Fodder Beet</th>
<th>Kale</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Leaf</td>
<td>Bulb</td>
</tr>
<tr>
<td>% of Plant</td>
<td>0.177</td>
<td>0.823</td>
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<tr>
<td>Dry Matter%</td>
<td>23.4</td>
<td>18.7</td>
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<tr>
<td>Neutral Detergent Fibre</td>
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<td>355</td>
</tr>
<tr>
<td>Acid Detergent Fibre</td>
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<td>256</td>
</tr>
<tr>
<td>Dry Matter Digestibility %</td>
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<tr>
<td>Sodium</td>
<td>3.91</td>
<td>7.78</td>
</tr>
</tbody>
</table>

4.3 Apparent Animal Dietary Intake

Apparent intake of DM and nutritive components are presented in table 4.2. The cows in the kale treatment consumed 3.7 kg more DM than those in the fodder beet treatment (p<0.001). Due to increased apparent DM intake and high N and Ca the apparent intake of these minerals was also greater for cows on kale (p<0.001). The fibre intake was more than 1 kg greater for cows in the kale treatment for both neutral detergent fibre and acid detergent fibre. The CP and N intakes were significantly higher at almost double the intake for cows consuming the kale diet relative to those in the fodder beet treatment. The mineral intake of K and Mg were also greater for those in the kale treatment however the Ca and Na intakes were greater for cows in the fodder beet treatment.
Table 4.2 Apparent dietary intake of non-lactating dairy cows in late gestation (per cow per day)

<table>
<thead>
<tr>
<th>Intake</th>
<th>Fodder Beet</th>
<th>Kale</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dry Matter of crop (kg)</td>
<td>7.3 ± 2.1</td>
<td>11.2 ± 1.5</td>
<td>-</td>
</tr>
<tr>
<td>Dry Matter of Supplement (kg)</td>
<td>5.6</td>
<td>6.4</td>
<td>-</td>
</tr>
<tr>
<td>Total Dry Matter (kg)</td>
<td>13.4 ± 0.3</td>
<td>17.1 ± 0.5</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Acid Detergent Fibre (kg)</td>
<td>2.93 ± 0.6</td>
<td>3.87 ± 0.1</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Neutral Detergent Fibre (kg)</td>
<td>4.27 ± 0.07</td>
<td>5.71 ± 0.2</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Crude Protein (kg)</td>
<td>1.48 ± 0.03</td>
<td>2.95 ± 0.09</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Calcium (g)</td>
<td>0.309 ± 0.002</td>
<td>1.80 ± 0.983</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Potassium (g)</td>
<td>2.662 ± 0.06</td>
<td>2.884 ± 0.08</td>
<td>0.055</td>
</tr>
<tr>
<td>Magnesium (g)</td>
<td>0.222 ± 0.006</td>
<td>0.262 ± 0.01</td>
<td>0.004</td>
</tr>
<tr>
<td>Sodium (g)</td>
<td>0.534 ± 0.01</td>
<td>0.403 ± 0.02</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

4.4 Urinary Behaviour

Cows showed similar urine behaviour for both treatments resulting in a total daily urine volume of 27 L/cow/day for kale and fodder beet (Table 4.3, Appendix A). Urine frequency, the minimum and maximum urine event volumes, and the total daily volume adjusted for liveweight did not differ between treatments. The average and median urine event volumes were significantly different where the volumes for cows in the fodder beet treatment were greater than those in the kale treatment by 87 mL for the average volume and 77 mL for the median volume. The average volume per kg of liveweight trended towards being significant with cows in the fodder beet treatment again having greater values. Numerous urination behaviour variables were significantly affected by their temporal block including total daily volume, average urine event volume, and total daily volume per kg of liveweight.

Table 4.3 Urination behaviour of non-lactating dairy cows in late gestation fed fodder beet and kale based diets obtained using a urine harness

<table>
<thead>
<tr>
<th></th>
<th>Fodder beet</th>
<th>Kale</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Volume (L/day)</td>
<td>27.8 ± 3.06</td>
<td>27.4 ± 2.80</td>
<td>0.988</td>
</tr>
<tr>
<td>Frequency (#/day)</td>
<td>8.42 ± 0.98</td>
<td>10.1 ± 0.71</td>
<td>0.128</td>
</tr>
<tr>
<td>Average Volume (L)</td>
<td>3.58 ± 0.36</td>
<td>2.71 ± 0.24</td>
<td>0.04</td>
</tr>
<tr>
<td>Total Volume/LWT (mL/kg)</td>
<td>68.0 ± 0.62</td>
<td>54.0 ± 0.48</td>
<td>0.815</td>
</tr>
<tr>
<td>Average Volume/LWT (mL/kg)</td>
<td>5.37 ± 5.76</td>
<td>5.12 ± 6.52</td>
<td>0.063</td>
</tr>
<tr>
<td>Minimum Volume (L)</td>
<td>1.67 ± 0.24</td>
<td>1.30 ± 0.15</td>
<td>0.221</td>
</tr>
<tr>
<td>Maximum Volume (L)</td>
<td>6.25 ± 0.73</td>
<td>5.71 ± 0.55</td>
<td>0.696</td>
</tr>
<tr>
<td>Median Volume (L)</td>
<td>3.11 ± 0.31</td>
<td>2.34 ± 0.20</td>
<td>0.047</td>
</tr>
</tbody>
</table>
4.5 Animal Samples

The number of faecal events per day did not significantly differ between diets nor did their DM contents (table 4.4). The ash and N contents did differ, significantly and very significantly respectively. Where the N level in the faeces was 1.94% DM for cows consuming the kale diet and 2.52% DM for those consuming the fodder beet diet. The urine N content was greater for cows in the kale treatment at 4.6 g N/L compared with 3.7 g N/L for cows in the fodder beet treatment though the difference was not significant. The urea and ammonia content of the urine was similar for cows in both treatment groups. The creatinine levels were significantly higher for cows in the kale treatment at 4.36 mmol/L where the fodder beet level was 3.92 mmol/L.

Table 4.4 Composition of dung and urine of non-lactating dairy cows in late gestation with winter diets based on fodder beet and kale forages

<table>
<thead>
<tr>
<th></th>
<th>Fodder Beet</th>
<th>kale</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Dung</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of events</td>
<td>6.94 ± 2.48</td>
<td>7.38 ± 0.40</td>
<td>0.552</td>
</tr>
<tr>
<td>DM%</td>
<td>23.5 ± 0.08</td>
<td>25.0 ± 0.80</td>
<td>0.182</td>
</tr>
<tr>
<td>OM%</td>
<td>54.7 ± 0.03</td>
<td>67.9 ± 2.48</td>
<td>0.001</td>
</tr>
<tr>
<td>N% DM</td>
<td>2.52 ± 0.20</td>
<td>1.94 ± 0.05</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>Urine</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N%</td>
<td>0.37 ± 0.05</td>
<td>0.46 ± 0.10</td>
<td>0.227</td>
</tr>
<tr>
<td>NH3 (mmol/L)</td>
<td>2.17 ± 0.31</td>
<td>2.57 ± 0.79</td>
<td>0.052</td>
</tr>
<tr>
<td>Creatinine (mmol/L)</td>
<td>3.92 ± 0.55</td>
<td>4.36 ± 0.82</td>
<td>0.019</td>
</tr>
<tr>
<td>Urea (mmol/L)</td>
<td>70.65 ± 8.65</td>
<td>86.8 ± 24.0</td>
<td>0.29</td>
</tr>
</tbody>
</table>

4.6 Water and N Balance

The N intake was significantly greater for cows in the kale treatment consuming more than double the level than those in the fodder beet treatment (Table 4.5, p<0.001). Because digestibility of the diets was similar and urine volumes were similar, over 50% of ingested N in the kale treatment is unaccounted for. The daily N intake was plotted against the daily total volume of urine which displayed a weak, positive relationship for cows in the fodder beet treatment and a moderate, positive relationship for cows in the kale treatment.
Table 4.5 N balance of non-lactating dairy cows in late gestation fed winter diets based on fodder beet and kale forages. All values in grams

<table>
<thead>
<tr>
<th></th>
<th>Fodder Beet</th>
<th>Kale</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nitrogen Intake</td>
<td>237 ± 4.5</td>
<td>471.85 ± 15.11</td>
</tr>
<tr>
<td>Nitrogen in Faeces</td>
<td>63.2 ± 2.32</td>
<td>82.6 ± 1.64</td>
</tr>
<tr>
<td>Nitrogen in Urine</td>
<td>106.07 ± 11.94</td>
<td>125.53 ± 12.84</td>
</tr>
<tr>
<td>Nitrogen Retained in Foetus &amp; Liveweight Gain</td>
<td>20 g (from ARC 1980)</td>
<td>20 g</td>
</tr>
<tr>
<td>Unaccounted for Nitrogen</td>
<td>47.6 ± 14.3</td>
<td>264 ± 15.1</td>
</tr>
<tr>
<td>Nitrogen Intake vs. Daily Urine Volume</td>
<td>Coefficient: 0.656 $R^2$: 0.125</td>
<td>Coefficient: -0.450 $R^2$: 0.403</td>
</tr>
</tbody>
</table>

The direct water intake of the animals from the trough was not found to differ between treatments, though due to high DM intake on kale diets the indirect water consumption from the kale diet was significantly higher than fodder beet resulting in a significantly higher total water intake (table 4.6). The water in the faeces was greater for cows in the kale treatment which had 10L greater estimated respiration and insensible losses. The total water intake and daily urine volumes were plotted against each other displaying a moderate, negative relationship for cows in the fodder beet treatment and a weak, positive relationship for those in the kale treatment.

Table 4.6 Water balance of non-lactating dairy cows in late gestation fed winter diets based on fodder beet and kale forages. All values in litres

<table>
<thead>
<tr>
<th></th>
<th>Fodder Beet</th>
<th>Kale</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water from Trough</td>
<td>11.47 ± 4.48</td>
<td>9.68 ± 2.69</td>
<td>0.339</td>
</tr>
<tr>
<td>Water from Feed</td>
<td>43.1 ± 1.2</td>
<td>59.6 ± 2.1</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Total Water Intake</td>
<td>54.5 ± 1.25</td>
<td>69.3 ± 2.06</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Water in Faeces</td>
<td>8.33 ± 0.42</td>
<td>13.1 ± 0.59</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Water in Urine</td>
<td>28.7 ± 3.23</td>
<td>27.4 ± 2.80</td>
<td>0.988</td>
</tr>
<tr>
<td>Estimated Respiration/Insensible Losses</td>
<td>18.5 ± 3.6</td>
<td>28.9 ± 2.3</td>
<td>0.02</td>
</tr>
<tr>
<td>Water Intake vs. Daily Urine Volume</td>
<td>Coefficient: -1.71 $R^2$: 0.439</td>
<td>Coefficient: 0.589 $R^2$: 0.190</td>
<td>-</td>
</tr>
</tbody>
</table>

4.7 Urine Patch Coverage

The equations obtained from the calibration in figure 4.3 of urine patch areas were polynomial with an $r^2$ for fodder beet of 0.67 and for kale of 0.94 (figure 4.3). The urine patch area is greater for volumes landing on the kale grazing area compared with those on the fodder beet area and this results in smaller average urine patch sizes on the fodder beet paddock (table 4.7). However the coverage per hectare of the paddock is greater for the fodder beet paddock owing to the higher stocking density.
Table 4.7 Winter forage fodder beet and kale paddock urine patch coverage grazed in situ by non-lactating dairy cows in late gestation

<table>
<thead>
<tr>
<th></th>
<th>Fodder beet</th>
<th>Kale</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calibration Equation</td>
<td>0.0122x^2 – 0.0416x + 0.1547</td>
<td>0.0064x^2 + 0.0452x + 0.0609</td>
</tr>
<tr>
<td>Mean Volume from Harness (L)</td>
<td>3.58 ± 0.36</td>
<td>2.71 ± 0.24</td>
</tr>
<tr>
<td>Frequency from Harness (#/day)</td>
<td>8.42 ± 0.98</td>
<td>10.11 ± 0.71</td>
</tr>
<tr>
<td>Mean Predicted Patch (m²)</td>
<td>0.16</td>
<td>0.23</td>
</tr>
<tr>
<td>Minimum Patch Size (m²)</td>
<td>0.12</td>
<td>0.12</td>
</tr>
<tr>
<td>Maximum Patch Size (m²)</td>
<td>0.37</td>
<td>0.32</td>
</tr>
<tr>
<td>Stocking Density (cows/ha)</td>
<td>50</td>
<td>16.67</td>
</tr>
<tr>
<td>Daily Paddock Coverage (m²/ha)</td>
<td>67.36</td>
<td>38.76</td>
</tr>
</tbody>
</table>

4.8 Estimation of Paddock N Losses

The leaching N loss from non-urine patch areas was similar for both paddocks however the proportion of the paddock covered by urine patches and the loss of N form urine patches was almost double for the fodder beet paddock (table 4.8). This is a product of smaller urine patch areas leading to greater N loading and greater stocking rates. The proportion of the paddock determined to be covered by urine patches was 45.4% for the fodder beet and 24.3% for the kale paddock. The total N losses over the winter period were calculated to be 122.5 kg from the fodder beet paddock and 82.2 kg from the kale paddock.
Table 4.8 Estimated paddock N (nitrogen) leaching losses from fodder beet and kale winter forage paddocks grazed *in situ* by non-lactating dairy cows in late gestation

<table>
<thead>
<tr>
<th></th>
<th>Fodder Beet</th>
<th>Kale</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time Grazed (days)</td>
<td>60</td>
<td>60</td>
</tr>
<tr>
<td>Area Grazed (ha)</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>Cows #</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>Urine Events/24h #</td>
<td>8.42</td>
<td>10.1</td>
</tr>
<tr>
<td>N% in Urine</td>
<td>3.7</td>
<td>4.6</td>
</tr>
<tr>
<td>Average Urine Volume (L)</td>
<td>3.58</td>
<td>2.71</td>
</tr>
<tr>
<td>Average Urine Patch Area (m²)</td>
<td>0.16</td>
<td>0.23</td>
</tr>
<tr>
<td>Non-Patch N Loss (kg)*</td>
<td>16</td>
<td>17</td>
</tr>
<tr>
<td>% N Loading Lost from Patch*</td>
<td>34</td>
<td>55</td>
</tr>
<tr>
<td>Urine N Load Average Urination (kg)</td>
<td>13</td>
<td>12</td>
</tr>
<tr>
<td>Urine Patch N Load (kg)</td>
<td>817</td>
<td>541</td>
</tr>
<tr>
<td>Leaching Loss from Urine Patch (kg)</td>
<td>278</td>
<td>298</td>
</tr>
<tr>
<td>% Urine Patch Coverage</td>
<td>41</td>
<td>23</td>
</tr>
<tr>
<td>% Not Urine Coverage</td>
<td>59</td>
<td>77</td>
</tr>
<tr>
<td>Loss from Patch Area (kg)</td>
<td>114</td>
<td>69.3</td>
</tr>
<tr>
<td>Loss from Non-Patch Area (kg)</td>
<td>9.4</td>
<td>13</td>
</tr>
<tr>
<td>Total N Loss from Paddock (kg/ha)</td>
<td>123</td>
<td>82</td>
</tr>
</tbody>
</table>

*Values sourced from Cameron (unpublished data).
Chapter 5
Discussion

The aim of this research was to quantify the urination behaviour of dairy cows wintered on either kale or fodder beet and investigate factors responsible for similarities and differences in urination behaviour. The results of this research found that total urine excretion of cows consuming a fodder beet or kale based diet were similar, despite differences in water, N, DM, CP, and mineral intake.

5.1 Total Urine Output

The mean urine volume (27 L/cow/day) and range in urine volumes (12 – 50 L/cow/day) are similar to values of 9 – 47 L/cow/day reported in a previous study under similar conditions (Ravera et al. 2015). The values obtained for total daily volume of urine, average urine event volume, and urination frequency for both treatments are in the expected range. This study is the second time that this model of urine harness has been used and the validation of the flow meter on the harness showed a standard error of the estimate of 500 ml based on 11 measurements (figure 3.6). The deviation of the harness recorded volume from the measured volume was not consistently lower or higher. A study (Cao et al., 2009) using a urine collection device on cattle found average daily urine output volume to be lower than was identified in this study at 15.7 L however these cows were likely consuming a diet with lower water content. Ravera et al. (2015) noted significantly higher urine volumes with kale diets at 30 L compared with cows on fodder beet 18 L which was attributed to the greater intake of CP and DM for cows fed kale. The daily urine output volume is influenced by the level of fluid and minerals needing to be expelled from the body as well as the CP and DM intake (Bannik et al., 1999; Khelil-Arfa et al., 2012). As the intakes of water, DM, CP, and N were significantly greater for cows fed kale it was expected that the volume of urine excreted would have been larger.

Water intake through drinking and eating is recognised to influence urine output (Frandson et al., 2006). In this study there were significant differences in water intake with cows on kale consuming 15L litres more than cows on FB. The percentage of water intake which was unaccounted for in urine and faeces equated to 33.3% for fodder beet and 41.5% for cows on kale which could be regarded as insensible water loss. There is little published data regarding the respiratory and insensible water losses of dairy cattle. Holter & Urban (1991) determined 18% of water loss to be accounted for by sweat and respiration in lactating dairy cattle in their thermoneutral zone which is much lower than unaccounted water in the present study. The partitioning of insensible water losses are highly dependent on ambient temperature (Church et al., 1988) however the temperatures during the trial...
were within the thermoneutral zone of dairy cattle (Meyer et al., 2004). The values for water intake from the trough were obtained from few animals (n=16) yielding a high standard error, e.g. 11.47 L ± 4.48 L, where the values for cows in the same treatment on the same day ranged from 7 to 37 L.

Another source of error in accounting for water losses may be in that excreted in faeces. Faecal DM% and digestibility of the diet was used to determine faecal water loss. The diet digestibility was predicted through NIRS and may not be accurate as the DM intake levels were high particularly for the kale diet which is 150% of their maintenance requirements (Roche et al., 2005). At high intake levels and cold temperatures the digestibility of feeds is reduced (Mairon & Christopherson, 1992). When the digestibility values for the feed are 10% lower the water loss in faeces is increased which reduces respirations and insensible water losses to 12.81 and 18.50L for cows in the fodder beet and kale treatments respectively. There is evidence to suggest possible error in the measurements due to the large volume of ingested water that is not accounted for in excretion of urine and faeces. The regression of water intake and volume of water excreted in urine displayed a moderate, negative relationship for cows in the fodder beet treatment ($r^2=0.44$) and a weak, positive relationship for those in the kale treatment ($r^2=0.19$). Indicating that the water intake may have influenced the urine volume of cows fed fodder beet but possibly not for cows fed kale.

The level of N excreted in urine has been found to be highly correlated with the N levels in the diet (Castillo et al., 2000) and the N intake with the total daily urine excretion (Bannik et al., 1999). The N intake was high for all cows though the intake for those fed kale N was double that of cows fed fodder beet. This is a result of high DM intake and high N content in the forage. The kale plant contained N in the concentration of 2.74% DM which is greater than published values of 2.0-2.2% DM (Edwards et al., 2014; Miller et al., 2012; Jenkinson et al., 2014) though it is similar to values obtained from kale grown at this site of 3.0% DM (Rugoho et al., 2010). Eckard et al. (2010) determined that 75-95% of the ingested N by ruminants in excreted. Given this the excreted N level as 75% of ingested would be expected to be 178 g for the fodder beet treatment and 354 g for the kale treatment. However the excreted N for cows in the kale treatment was 228 g which is less than 50% of the ingested N and resulting a large amount of unaccounted for N. The large quantity of unaccounted N could possibly be due to an underestimation of urinary N content (see section 5.2 and figure 5.2). The regression of N intake and total daily urine output showed a weak, positive relationship for cows in the fodder beet treatment ($r^2=0.13$) and a moderate, positive relationship for cows in the kale treatment ($r^2=0.40$). This suggests that the N intake was a possible factor in the urine volume of cows in the fodder beet but is not likely to have been a major factor for cows in the kale treatment. The intake of K was 0.2 g greater for cows fed kale (p=0.055) and Na intake was 0.13 g greater for cows fed fodder beet (p<0.001). Increases in the intake of these minerals has been found
to increase urine output (Frandsen et al., 2006) however it is not clear if the differing intake levels in the diet were having an effect in the current study.

Major factors that have been found to increase urine volume are CP and DM intake (Bannik et al., 1999; Khelil-Arfa et al., 2012; Holter and Urban, 1992). The DM intake was 3 kg greater for cows fed the kale diet (p<0.001) and the CP intake was almost double that of the cows fed fodder beet (p<0.001). The DM intakes for each diet are similar to those fed to cows by Ravera et al. (2015) however for the previous trial the DM was concluded to have caused the greater urine volume for cows fed kale. The DM content of the diets was averaged over the course of the trial which may not be representative of the actual DM% variation and would have an effect on the intake estimations. The similar urine volumes of cows in either treatment in this trial may be due to other factors influencing the urine output volume such as the Na intake which was significantly higher for cows fed fodder beet. The temporal block the cows were in had a significant interaction with total daily urine output (P value=0.003) suggesting that factors caused by differences in conditions each week could have influenced the daily urine volume. Some researchers use creatinine as a proxy for urine output. David et al., (2015) determined the creatinine content in the urine to be inversely related to the daily volume of urinary output in sheep. The creatinine level was significantly greater for cows in the kale treatment at 4.36 mmol/L compared with 3.92 mmol/L for those in the fodder beet treatment. This would indicate cows in the fodder beet treatment to have a greater volume of urine excreted though this was not the case.

Though the winter diets supplied significantly greater intakes of DM, water, CP, and N which have been identified to increase urine volume in previous research, the total daily urine volumes of cows fed each forage were similar. There are some possible sources of error in the measurement such as further validation of the urine harness needed, DM% of the feed, faecal water underestimated due to incorrect feed digestibility values, and the water intake from the trough. In general the similarity in urine volumes was unexpected and could not be attributed to a single factor measured in this study.

### 5.2 Soil N loading

Nitrate leaching from urine events are driven by volume per urination, N concentration per urination, area of deposition of urination, soil properties and drainage (Li et al., 2012, figure 5.1). In this study the volume of urine in a single event ranged between 0.7 and 11.4 L and averaged 3.6 and 2.7 L for fodder beet and kale respectively. Published values for non-lactating dairy cows in late gestation grazing winter forages in situ in New Zealand show urine volumes per event ranged between 0.5 and 8.6 L and averaged 2.37 L (Ravera et al., 2015). This trial found the fodder beet event volumes to be of greater volume but less frequent (8.42 events in 24 hours) than the events for cows in the kale
treatment (10.11 events in 24 hours). Therefore the difference in urine event volumes was compensated for by the change in frequency resulting in similar total daily urine volume output for cows in both treatments. The frequency of urine events measured by Ravera *et al.* (2015) was similar to those in this trial at 8.2 and 12.3 events per day for cows in the fodder beet and kale treatments respectively.

Soil N loading is also influenced by urinary N content as mentioned above which is linked to N intake (Eckard *et al.*, 2010). The N intake was significantly greater (p<0.001) for cows in the kale treatment and the urine N content was also greater at 4.6 g/L compared with 3.7 g/L for cows in the fodder beet treatment. These results are consistent with those of Ravera *et al.* (2015) where the N intake was also greater for cows consuming kale and urinary N levels were 4.9 g/L and 4.0 g/L for cows grazing kale and fodder beet respectively. These urine N levels are lower than those of lactating dairy cows grazing traditional pastures of ryegrass and white clover at 5.7 g N/L (Totty *et al.*, 2013) and 6.1 g N/L (Edwards *et al.*, 2015). It is possible that the spot samples of urine were collected at times of day when the N content was relatively low. Betteridge *et al.* (2013) identified urinary N content to be lowest during the 5 hours following feeding (figure 5.2). The spot samples in this study were taken within 4 hours of the feed being given to the animals hence it is possible that the N excreted in urine was greater than the values in this study. Consequently the N loading for each urination was 12.5 and 13.3 g N per urination for kale and fodder beet respectively.
The dispersion of urine as it hits the ground affects N leaching because it is a factor in the concentration of N deposited on the paddock surface and soaked into the soil (Li et al., 2012, figure 5.3). The ground topography differed visually between treatments where the fodder beet grazing area had deeper hoof imprints while the kale grazing area was relatively more even as is displayed in plate 3.6. The fodder beet paddock had three times the stocking density of the kale paddock where trampling by cows and greater numbers of urine events on the area would lead to a wetter, softer area with deeper mud. Urine events deposited on already wet areas soak into the soil more quickly so the urine fluid does not spread as far across the surface. The paddock in which the fodder beet was grown was conventionally ploughed during the previous September and the consumption of the plants themselves results in the loosening of topsoil as plants are removed as 40-50% of the bulb is buried in the soil (DLFseeds, 2013). Therefore there is more soft soil available at the surface to form mud compared with the kale paddock into which the seed was direct drilled resulting in far less soil disturbance. The topography of the fodder beet paddock surface due to deeper hoof prints in the fodder beet paddock muddy surface as well as the deep craters left from the bulb removal resulted in the urine pooling into smaller areas resulting in smaller areas of urine patches as shown in plate 3.6. Therefore although the average urine volume was greater for cows in the fodder beet treatment the average urine patch was of a smaller area.
The urine patch areas were estimated using the urine patch area calibration curves and urine event volume as determined by the harness. From average urination volumes, this resulted in predicted urine patch areas of 0.16 m$^2$ in the fodder beet paddock and 0.23 m$^2$ in the kale paddock. In a similar study with dry cows Ravera et al. (2015) reported patch areas for cows grazing fodder beet of 0.19 m$^2$ and 0.24 m$^2$ for the kale patches, which are similar to those reported here. However, the results of Ravera et al. (2015) were based on areas from smaller average urine volumes, reflecting differences in calibration coefficients. The authors of that study recognised the role of climate and soil topography. As stated above the topography of the paddock surface is a major factor in the wetted area of each urination event. Ravera et al. (2015) conducted their study at the same trial site in 2014 using the same paddocks as were used in this trial. The fodder beet crop yield during this trial was 24.8 t DM/ha which is similar to the yield at that time last year of 24.7 t DM/ha (de Ruiter, unpublished) hence a similar amount of soil disturbance would have occurred due to bulb removal each year. The rainfall was similar during the period leading up to and during the time that the urine patch calibration was produced in 2014 when Ravera et al. (2015) and in 2015 for this trial. Hence the wet ground area could be assumed to be the same having no great effect on the differences in patch areas.

The total N leached at the farm level is expressed as kilograms of N per hectare. Each urine event as described above has to be multiplied by the number of events per cow per day, the number of cows and the duration those animals are held in a given area to produce predictions of N leaching. The paddock coverage of urine patches per hectare was estimated to be 41% for the fodder beet paddock and 23% for the kale paddock. Though the average urine patches are smaller for the fodder beet paddock there was 3 times the stocking density of animals on the paddock area resulting in almost twice the paddock covered with urine patches. The urine deposits of grazing livestock is the major driver of N leaching on farms (de Kelin et al., 2010) hence the smaller more concentrated
deposits of N in urine across a greater area of the paddock results in a greater volume of N leached from the fodder beet paddock. The estimated N leached from each paddock in this study was 123 and 82 kg/ha for the fodder beet and kale paddock respectively. Ravera et al. (2015) predicted N leaching losses of 78 kg/ha for the fodder beet paddock and 54 kg/ha for the kale paddock when grazed by non-lactating dairy cows in late gestation at the same stocking rates as in this study. Chrystal et al., (2012) determined the N leached from winter grazing on kale to be 53 kg/ha which is close to the value of Ravera et al. (2015) for the kale paddock. The estimated N leached from the winter grazing paddocks was 45 and 28 kg/ha greater for this study than for Ravera et al. (2015).

A major source of the difference between the N loss estimations between this and the previous study (Raveral et al 2015) is the lysimeter data used to determine the proportion of N in urine and non-urine patches that is leached. The proportion N leached for this study was higher compared with values used by Ravera et al. (2015, table 5.1). The lysimeter data used and the potentially larger pool of leachable N in the soil are possible factors causing the greater level of N to be leached in this study. The level of N that is readily leachable in the soil determines the volume of N that will be lost during periods of drainage through the soil (Cameron, Di, & Moir, 2013). Denitrification is the process through which leachable nitrate is converted to N gases and this activity is lowered at temperatures of less than 5°C (Maag & Vinther, 1996). During the period of February to July 2014 the temperature fell below 5°C on 52 days. Comparatively during the same period in 2015 the temperature was below 5°C on 73 days which may have caused less removal of N from the leachable pool through denitrification. Kale has a base temperature of 0°C (de Ruiter et al., 2009) and fodder beet grows above 8°C (Feedipedia, 2015). The temperature fell below 0°C on 20 and 26 days in 2014 and 2015 respectively and the temperatures were generally lower in 2015 hence the plant uptake of N from the leachable pool may have been lower at this time. These lower temperatures in 2015 could account for some of the greater N leached as less was removed from the soil mineral pool through denitrification and plant uptake (Figure 2.4).

<table>
<thead>
<tr>
<th></th>
<th>Fodder Beet</th>
<th></th>
<th>Kale</th>
</tr>
</thead>
<tbody>
<tr>
<td>% Paddock Coverage – Urine Patches</td>
<td>41</td>
<td>61.43</td>
<td>23</td>
</tr>
<tr>
<td>% N lost from Urine Patches</td>
<td>34</td>
<td>32.16</td>
<td>55</td>
</tr>
<tr>
<td>N leached from Non-Urine Patch Area (kg/ha)</td>
<td>16</td>
<td>11.5</td>
<td>17</td>
</tr>
<tr>
<td>Paddock N Loss (kg/ha)</td>
<td>123</td>
<td>77.82</td>
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5.3 Conclusions

Despite numerous factors that have previously been found to affect urinary output being significantly different, the total daily urine excretion of cows from each treatment was similar. Identification of the factors causing the daily urine output to the similar was not possible therefore further research into the mechanisms behind the effects of the kale and fodder beet diets on urination would be valuable. Also further improvement and validation of the urine harness is recommended for future studies. However this study has provided estimations of N leached from kale and fodder beet winter forage paddocks for which there is a still a need. This information can be compared with the N leaching results of the suction cups and lysimeters from the trial site in the 2015 winter season to test the accuracy of the estimations. The data regarding urine patch area and coverage is a significant contribution to the little current knowledge of the urine patch dynamics on winter grazing areas. More information in this area is needed to enable models such as OVERSEER to make more accurate predictions of N losses from farm systems. This would have effects for farmers in terms of their nutrient output limits which will require adjustments in their nutrient budgeting.
# Appendix A Urination Behaviour of non-lactating dairy cows grazing winter forages fodder beet and kale in situ

<table>
<thead>
<tr>
<th>Cow</th>
<th>Days</th>
<th>TRT</th>
<th>Cow (L)</th>
<th>8 ± 0</th>
<th>2.32 ± 0.20</th>
<th>4.47 ± 0.38</th>
<th>35.77 ± 3.07</th>
<th>1.44 ± 0.19</th>
<th>4.00 ± 0.19</th>
<th>2.03 ± 0.06</th>
<th>520</th>
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<tbody>
<tr>
<td>21</td>
<td>2</td>
<td>f</td>
<td>18.60 ± 1.60</td>
<td>25.98</td>
<td>9</td>
<td>2.89</td>
<td>5.80</td>
<td>52.16</td>
<td>0.79</td>
<td>4.60</td>
<td>2.96</td>
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<tr>
<td>28</td>
<td>2</td>
<td>f</td>
<td>24.43 ± 2.12</td>
<td>35.14</td>
<td>12 ± 1</td>
<td>2.86 ± 0.87</td>
<td>6.21 ± 1.90</td>
<td>76.38 ± 29.02</td>
<td>0.83 ± 0.51</td>
<td>4.06 ± 1.62</td>
<td>2.41 ± 1.11</td>
</tr>
<tr>
<td>78</td>
<td>2</td>
<td>f</td>
<td>35.14 ± 13.35</td>
<td>39.09</td>
<td>7</td>
<td>5.58</td>
<td>9.90</td>
<td>69.31</td>
<td>2.59</td>
<td>10.29</td>
<td>4.50</td>
</tr>
<tr>
<td>150</td>
<td>1</td>
<td>f</td>
<td>17.94 ± 4.98</td>
<td>175</td>
<td>2</td>
<td>2</td>
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<tr>
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<td>f</td>
<td>37.92 ± 5.37</td>
<td>184</td>
<td>2</td>
<td>4</td>
<td>1</td>
<td>3</td>
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<tr>
<td>213</td>
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<td>f</td>
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<td>0.5</td>
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<td>2</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>327</td>
<td>2</td>
<td>f</td>
<td>21.31 ± 8.36</td>
<td>338</td>
<td>2</td>
<td>15.5 ± 0.15</td>
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<td>5</td>
<td>2</td>
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<td>2</td>
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<tr>
<td>30</td>
<td>2</td>
<td>k</td>
<td>24.44 ± 11.63</td>
<td>52</td>
<td>1</td>
<td>27.29</td>
<td>10</td>
<td>2</td>
<td>7</td>
<td>2.73</td>
<td>4.74</td>
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<tr>
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<td>2</td>
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<td>195</td>
<td>2</td>
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<td>2.64 ± 0.78</td>
<td>5.06 ± 1.81</td>
<td>52.95 ± 18.69</td>
<td>1.50 ± 0.05</td>
<td>4.75 ± 2.26</td>
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<tr>
<td>198</td>
<td>2</td>
<td>k</td>
<td>29.44 ± 1.17</td>
<td>221</td>
<td>2</td>
<td>11 ± 1</td>
<td>3.54 ± 1.05</td>
<td>7.75 ± 2.30</td>
<td>87.55 ± 33.08</td>
<td>1.19 ± 0.70</td>
<td>7.85 ± 3.12</td>
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<td>k</td>
<td>46.86 ± 1.31</td>
<td>280</td>
<td>2</td>
<td>7 ± 4</td>
<td>1.55 ± 0.36</td>
<td>3.35 ± 0.78</td>
<td>26.55 ± 18.87</td>
<td>0.90 ± 0.38</td>
<td>3.54 ± 2.05</td>
</tr>
<tr>
<td>288</td>
<td>1</td>
<td>k</td>
<td>16.41</td>
<td>303</td>
<td>2</td>
<td>12.5 ± 2.5</td>
<td>2.02 ± 0.08</td>
<td>4.18 ± 0.16</td>
<td>52.64 ± 12.48</td>
<td>0.90 ± 0.07</td>
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<tr>
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<td>1</td>
<td>k</td>
<td>13.57</td>
<td>335</td>
<td>1</td>
<td>15.41</td>
<td>1.71</td>
<td>3.37</td>
<td>30.34</td>
<td>0.88</td>
<td>4.60</td>
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<tr>
<td>340</td>
<td>2</td>
<td>k</td>
<td>30.83 ± 0.21</td>
<td>352</td>
<td>2</td>
<td>10 ± 1</td>
<td>3.42 ± 0.55</td>
<td>7.24 ± 1.16</td>
<td>73.56 ± 18.82</td>
<td>1.61 ± 0.54</td>
<td>5.70 ± 2.07</td>
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</table>

TRT=treatment; Vol=volume; freq=frequency; LWT=liveweight; min=minimum; max=maximum.
References


