Technical Series

White clover The forgotten component of high producing pastures?



RHIZOBIA RESEARCH IN NZ

HOW DO RYEGRASS PLANTS CHANGE AS A DAIRY PASTURE AGES? FORAGES TO REDUCE URINE PATCH N LEACHING

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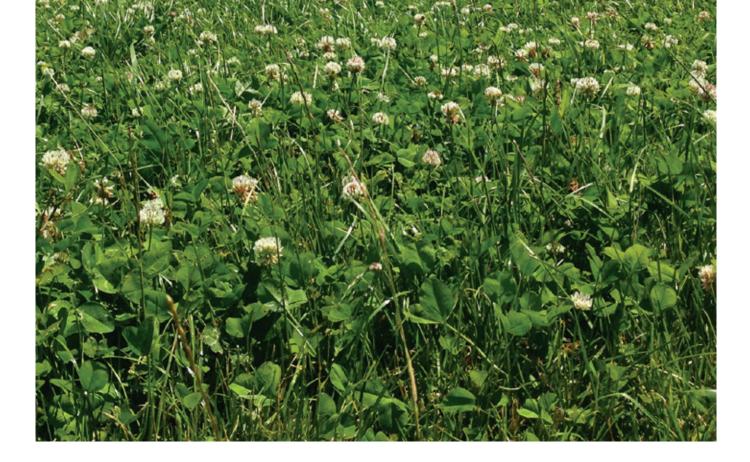
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White clover: the forgotten component of high producing pastures?

In this article we will cover some of the fundamental aspects of the growth and development of white clover plants. These provide the basis for understanding why white clover requires careful management and how more value might be obtained from it.

Key findings

- Understanding how white clover grows and interacts with ryegrass allows farmers to manage pastures for greater white clover content.
- Timing of the first grazing of newly sown pasture is important; it should occur when white clover seedlings are small rosettes and before lateral expansion of the plant begins, to avoid damaging new stolons.
- Between 1 and 2.5 years after sowing the clover taproot dies and the plant fragments into smaller plants. At this time, farmers often observe a decline in clover content.
- Two factors that influence the ability of white clover to thrive are nitrogen and light. Farmers can improve white clover content by controlling pre-grazing pasture mass to limit shading from ryegrass, and post-grazing pasture mass to avoid over grazing.



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Introduction

The beneficial features of white clover have long been extolled - these are primarily its ability to fix atmospheric nitrogen (N) and contribute to the N economy of pasture¹, its high nutritive value which supports higher milksolids production per unit of feed consumed compared with ryegrass², and its ability to increase annual pasture production when sown in a mixture with ryegrass compared with pure ryegrass³. However, white clover seldom reaches its potential in modern grazed pastures.

Establishment

Traditionally, new ryegrass-white clover pasture was sown into a cultivated seedbed following a cash or forage crop. Cultivation prepared a seedbed, depleted populations of pest and diseases and reduced soil mineral N to lower levels. Good seedbed preparation and consolidation allowed control over sowing depth such that seed could be lightly covered with soil to the optimal 5 mm depth, and broadcasting provided an even spatial distribution of seed⁴.

In the past 20-30 years there have been changes in sowing techniques, primarily grass-to-grass renewal and greater use of direct drills. These changes have affected virtually all the factors listed above, to the detriment of white clover establishment:

- Soil nitrogen is not depleted by the intervening crop and high soil N means more competition for the white clover seedling from the companion ryegrass.
- There is also reduced ability to sow clover seed consistently at shallow depths
- There is greater presence of pests (e.g. slugs) and diseases, and a less favourable (for white clover) spatial distribution of seed.
- There is greater competition for light and nutrients within the drill row from ryegrass or weed species, and from other white clover seedlings.
- There is also no opportunity to create a fine, firm, moist and weed-free seedbed through cultivation⁴.

To improve white clover establishment if new pasture is direct drilled, it is essential to:

- Double spray existing pasture with herbicide before direct drilling. This ensures weed species are eliminated
- Stick to the 'traditional' clover sowing rate of 3-4 kg seed/ ha, but use a moderate grass seeding rate (e.g. 18 kg/ha for diploids and 22 kg/ha for tetraploids)
- Use a drill with a small seeds box. Drop white clover seed onto the soil surface from the small seeds box in front of coulters sowing the ryegrass. Cover seed with a brush or bar harrow to ensure the correct sowing depth (ideally 5 mm).
- Apply slug bait at sowing.

Life cycle stages of the white clover plant

From germination, the growth and development of white clover can be separated into three distinct morphological phases⁴ (see figures 1-3 opposite).

Seedling phase - This slow growing phase lasts approximately 1-3 months during which the seedling reaches a compact rosette with 2-3 branches and 10-20 mm lateral spread. There is no stem elongation. During this phase the seedling is vulnerable to light competition from the faster growing and more erect ryegrass seedling. Therefore, the timing of first grazing is important. Ideally this should occur just before new stolons start to elongate and while the seedling is still small and compact.

Delaying first grazing could be counterproductive to the small

white clover seedlings because the new, elongating stolons could be physically damaged or removed completely before they have been able to establish their own anchoring root system. This timing is more crucial for direct-drilled pastures where the competition from ryegrass is more intense.

Expansion phase - Also known as the tap-rooted phase, which lasts for 1-2 years and during which the plant rapidly expands and branches. Secondary stolons radiate from the central taproot/primary stolon, with lower orders of stolons progressively formed as branching increases. A single plant may have 15-20 growing points from the various hierarchical levels of stolons. Roots begin to form at the nodes of these lower-order stolons. Large nodal roots anchor the stolon well and keep it at or slightly below the soil surface.

During this phase pastures can have good proportions of white clover, and even white clover dominance if soil N levels are low.

Similar to ryegrass, white clover requires a year to fully establish⁵. Thus, even beyond the first few grazings it is important to avoid treading damage to seedlings, especially as soils are wetting up going into late autumn and winter. Grazing management also needs to balance the requirement for light by white clover (i.e. minimal shading) with the need to avoid grazing too intensely as this can increase stolon loss if the nodal roots are not well formed.

From about 1 year after sowing, death of the taproot begins to occur in some plants and most taproots have died by 2.5 years from sowing. During this period the white clover plant population consists of a mix of tap-rooted plants, and plants that have reached the ultimate, clonal phase in the life-cycle.

Clonal phase - Once the taproot has died the plant fragments and from this point for the life of the pasture the population of white clover consists of smaller plants of varying size that grow from the stolon tip and decay from the stolon base.

This gives plants the ability to creep and exploit new favourable niches for growth. Often about the time of this transition to a clonal population, a 'clover decline' will be observed. Without a taproot and relying on smaller nodal roots, plants are more vulnerable to moisture stress, and damage by pests and diseases such as leaf mosaic virus, slugs and rootfeeding nematodes and insects including grass grub, white fringed weevil and clover root weevil.

This contributes to seasonal differences in the growth and abundance of white clover in our pastures.

White clover proportion in established pastures

White clover typically contributes <20% of total annual pasture production in NZ dairy pastures⁶. Greater proportions of white clover are not easily achieved or sustained without other intervention (e.g. herbicide suppression of grass). Compared with ryegrass, white clover growth is more limited by low temperatures from mid-autumn to mid-spring in most regions. For this reason, the proportion of clover in pasture during this period is low. White clover grows best with warm temperatures

(the optimum for growth is 24°C compared with about 20°C for ryegrass) provided soil moisture is adequate. However, without irrigation its potential for growth with those warmer temperatures is often limited by dry conditions during summer.

Natural cycles impact clover's density.

In addition to the effects of temperature and soil moisture described above, there are natural cycles in the proportion of white clover in mixed pastures^{7,8,9}. These cycles are driven largely by the way ryegrass and white clover respond to N (whether supplied from soil mineralisation or fertiliser) and the effects of this on their relative abilities to capture light. Under high soil N, ryegrass grows more quickly than clover because mineral N uptake requires less plant energy than the combination of N uptake and N fixation that clover uses for growth. This competition from ryegrass suppresses clover, but the ryegrass also uses soil N and reduces it to lower levels. At low soil N the competitive advantage switches to clover by virtue of its "stoloniferous" growth habit, and its ability to fix its own N. Then, eventually in this cyclic process the build-up of N from clover fixation reaches the point where ryegrass again becomes more competitive. In addition, urine patches have very high levels of N and strong competition from fast-growing ryegrass effectively excludes white clover for quite long periods.

Management is a balancing act for density and quality.

Unlike factors such as temperature and moisture (and insects and diseases) that are difficult to control (availability of irrigation being an exception), farmers have some control over N (through N fertiliser application rates) and light availability to clover plants. The frequent reports of difficulty in maintaining adequate proportions of white clover in dairy pastures can often (there are exceptions, for example, the recent experience of the devastating effects of clover root weevil through most of the country) be attributed to combinations of these factors and changes in management practices that have affected them. Grazing management of pastures to promote the proportion of white clover is a delicate balance between what the clover plants themselves require and what is required to manage the competitive effects from the ryegrass i.e. allowing clover to grow well while preventing ryegrass from dominating.

Steps to better clover management.

While there are natural limits to white clover content in pastures, the following management principles will help get the best performance that is possible.

During growth following grazing, white clover elevates its leaves, supported on elongating petioles, into the upper levels of the growing canopy to intercept light. Because they are elevated a high proportion are removed at the next grazing, and growth is dependent on initiation of new leaves. In winter when white



Figure 1: A white clover seedling at the rosette stage of growth. When sown in autumn, white clover can germinate more rapidly than ryegrass. But leaf production is slow and branches form but do not elongate, as shown in this picture of the small vertical primary stem surrounded by a ring of short secondary branches. The result is a compact rosette plant form with few branches and a small spread, and dependent on the central taproot. This form may last 1-3 months dependent on temperature.



Figure 2: A white clover plant in the early expansion phase, also dependent on a taproot. The primary stem of the rosette stage plant rarely elongates. Expansion of the plant comes from the secondary branches, of which there are 6 on average but up to 15, and can be very rapid. This picture from the underside of a plant shows that, in early expansion, root formation at the nodes close to the taproot is relatively weak, perhaps an expression of dominance by the strong central taproot system.

This illustrates the need for careful timing of the first grazing, to avoid developing stolons being stripped from the soil surface. This is more important in the large-leafed, slower branching cultivars (i.e. dairy types) than the smaller leafed, rapidly branching cultivars.

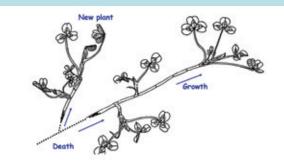


Figure 3: The third, clonal phase is the normal status of white clover populations in established mixed, permanent pastures. Through the process of continual growth at the stolon tip and decay at the stolon base, a relatively stable population of plants of varying size and complexity is maintained.

clover is growing slowly or possibly not at all, and ryegrass is growing, infrequent grazing that allows high herbage mass to accumulate and causes prolonged shading is also detrimental to white clover and these effects extend in the subsequent growing season.

During spring white clover plants are at their smallest size in their annual growth cycle (due to stolon death exceeding new stolon growth) and are, therefore, fragile and particularly susceptible to mismanagement (e.g. treading damage on wet soils; high herbage mass and shading) and environmental stresses.

During the growing season, grazing too frequently (i.e. not allowing pastures to reach at least 15 cm height) can curtail white clover leaf formation and elongation resulting in progressively fewer, smaller leaves with each successive grazing. Grazing too hard (i.e. residuals of less than 4 cm), especially during summer, may also be detrimental to white clover growth, and under dry conditions, its persistence⁶. Dairy pastures with low density can leave white clover exposed to over-grazing during periods of feed shortage (e.g. summer). This, combined with low soil moisture and high soil surface temperatures, often results in plant death. Large-leaved, dairy-type cultivars are more vulnerable to this than are small-leaved cultivars.

In dairying the natural cycle between ryegrass and clover has been disrupted by use of high rates of N fertiliser. This means that the competitive advantage is with ryegrass most of the time. Maintaining white clover while also using N fertiliser are not necessarily incompatible, provided grazing management controls the competition from ryegrass for light¹⁰, particularly from late winter and through early spring.

In summary, use the following guide to improve clover establishment and its contribution to established pastures.

	New pasture	Established pasture
Autumn	Clover seedlings are very vulnerable to shading. The first grazing requires a delicate balance between preventing shading by ryegrass and treading damage to the small and compact seedling. Do not graze lower than 3-4 cm.	
Winter	Clover seedlings are sensitive to treading damage from heavy animals on wet soils. Use young stock if available.	Avoid accumulating long pasture because ryegrass can shade the slow growing clover at this time of year.
Spring	Avoid shutting for silage as this will intensify shading by ryegrass.	Graze as per normal management for this time of year.
Summer		Maintain residuals of at least 4-4.5 cm, and 28-30 day grazing round especially under dry conditions.

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A renaissance of rhizobia research in NZ

The use of white clover to improve soil N fertility is often accepted as 'New Zealand's economic advantage'.

Key findings

- White clover and biological N-fixation has long been NZ's economic advantage. To maintain this, we require clovers and rhizobia bacteria that are capable of maintaining their relationship in competitive pasture environments.
- After 60 years there have been no changes to the rhizobia strain TA1 used in clover, despite changes to clover cultivars, pasture swards and stocking methods.
- Matching more suitable and enduring rhizobia to modern clover cultivars is underway, but getting new suitable strains takes time.
- The actual effectiveness of soil rhizobia populations ranged from 125% of TA1's ability (i.e. the natural strains were far more effective than TA1) to just 32%
- Establishing "elite" rhizobia requires them to be persistent and competitive with rhizobia that occur naturally in the soil where clover grows.
- In future, establishing proportions of elite rhizobia may occur using new delivery methods, other than seed coating.



Steve Wakelin, AgResearch

Given environmental challenges associated with N and nitrates confronting the agricultural sector, the maintenance of grassclover pastures that can self-regulate the N cycle and reduce the amount of leachable N in the bulk soil, is highly desirable. This can only work, however, if clovers can compete well with the grass. For this, effective rhizobia are needed.

Rhizobia are a group of soil bacteria that form specialised symbiosis (partnership) with clover roots (see Figure 1). When this symbiosis is effective, the rhizobia converts atmospheric N into forms the clover can use, known as Biological N Fixation (BNF), while the clover provides energy and suitable reduction – oxidation (REDOX) conditions for the bacteria.

The rhizobia strain currently used in NZ to inoculate white clover seed is called TA1. TA1 was first discovered in Australia more than 60 years ago; yet there has been no attempt since to match modern clover cultivars with new rhizobia. It is surprising that we rely on 60-year-old rhizobia to drive nitrogen fixation in the most important pastoral legume in New Zealand. Furthermore, the genetic potential of new clover varieties, each representing decades of investment in breeding and selections, are being under-realised if they are not matched with elite rhizobia. The gap between what is 'current' and 'potentially achievable' in this research area is enormous.

But can we actually find rhizobia that fix more N, for the same or lower energy cost to the clover, than TA1?

Figure 1: Our ability to grow white clover in pastures is often referred to as New Zealand's economic advantage. Clover, and other legumes, form a symbiosis with root-nodule forming rhizobia bacteria. Within root nodules, the rhizobia convert atmospheric nitrogen to plant available forms. Over time, this naturally increases the N-fertility of pastures, and improves forage quality. Improving on this symbiosis may provide multiple benefits for New Zealand's pastoral sector.



Investment in clover-rhizobia research

Key opportunities to increase the legume: rhizobia performance are being sought. These may be realised through:

- Discovery of high N-fixing strains of rhizobia that persist in New Zealand's soils, in current and future climatic conditions, and, that support clover growth in our farming systems.
- New methods of commercial inoculation of clover seed that support higher numbers and survival of rhizobia.
- Targeted inoculation of rhizobia into soils with depleted and/or ineffective rhizobia populations.
- Matching of rhizobia strains to the host clover, or other legume, genotype.

We are discovering elite rhizobia

Over 3000 rhizobia strains have been isolated from sites throughout New Zealand. Screening is underway to test these strains for ability to improve white clover growth through nitrogen fixation. Already, we have found many strains that significantly outperform TA1 in terms of N-fixation on white clover. These results are exciting; they demonstrate that gains for New Zealand's pastoral farmers may be achieved simply by replacing the old TA1 strain with newly discovered isolates of rhizobia from NZ pastures. The current screening process is very time consuming and expensive; it could take years to process just the strains collected. (Figure 2) As such, the development of genotypic markers as tools to support selection of rhizobia in vitro, potentially based on a number of different traits, is important.

Figure 2: Laboratory screening of rhizobia for high nitrogen fixation on clover is laborious and time consuming. To improve the rate of screening, modern genetic tools are being developed for rhizobia. This will enable rapid, cheaper, 'marker-assisted selection' of rhizobia strains from very large collections.



Opportunities in NZ's pastoral soils

The discovery of new rhizobia must be supported by knowing where they can best be used in pastoral systems. Indeed, many pastoral soils in New Zealand already contain high numbers of rhizobia. Is there room to improve on these natural populations? If so, where?

A random survey of 26 pastures found that despite the high population size, the actual effectiveness of soil rhizobia populations ranged from 125% of TA1's ability (i.e. the natural strains were far more effective than TA1) to just 32%. Most pastoral soils in NZ have a relatively ineffective populations. The reason behind this is unclear; however, it is not associated with low numbers of rhizobia in the soil. Rather, we think these populations of rhizobia have inherently low performance compared with TA1. Although these findings indicate the improvement of rhizobia populations will provide much greater benefit than originally anticipated, it might also be more difficult to achieve.

Delivery is essential for success

Realising the potential of elite rhizobia, and better matching between legume × rhizobia × environment, means that new strains must be successfully established in pastoral soils. This is currently done by coating legume seed with rhizobia (Figure 3). However, as legume plants grow, they will also form symbioses with rhizobia already present in the soil. This is a more pertinent issue for long-term pastures, and when annual legumes are a component of pastures. A significant challenge will be establishing elite rhizobia in the resident soil population - i.e. how do we build genetic gain in wider rhizobia populations?

This will likely be achieved by selecting more competitive and persistent rhizobia, by knowing when to apply new rhizobia to soils to increase the chances of successful establishment, or use of legume cultivars that will preferentially partner with the elite rhizobia (supporting persistence of elite strains in soil).

Figure 3: Using coated seed is recommended as it is currently the best method for introducing rhizobia into pastoral soils. The effectiveness of coating as a carrier of rhizobia depends on survival of the bacteria, and whether the clover plant forms symbioses with the 'background' rhizobia naturally present in many soils. Delivery technologies to further improve the establishment of elite rhizobia into pastoral systems are being investigated.



When can we expect our next generation rhizobia and pasture productivity?

In the near future, it is likely that the rhizobium strain TA1 will be replaced with newly identified elite strains. The release of these new strains will be supported by delivery of ancillary knowledge to help farmers make informed decisions about when and where new rhizobial strains will return a benefit.

New methods for delivery of rhizobia to farming systems are also likely to be developed. Collectively, these will ensure that farmers will have a much stronger opportunity to maintenance of grass-clover pastures that can, to an extent, self-regulate the N cycle. This will result in less undesirable loss of N from the soil, and reduce need for fertiliser N inputs. This can only be a winwin situation.

A cknowledgments

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Photographs courtesy of Mrs Dalin Brown, Lincoln University.



How do ryegrass plants change as a dairy pasture ages?

Poor pasture persistence continues to be a hot topic among dairy farmers across New Zealand. This is especially so in the upper North Island where, under some circumstances, new perennial ryegrass pastures last only 3-4 years before they need to be replaced.

Key points:

- The genetic make-up and growth habit of perennial ryegrass plant populations can change over time as a result of the 'survival of the fittest' principle.
- This could explain why the initial yield advantage of a new pasture tends to decline over time.
- A study comparing different cultivars and sowing rates was set up in three regions to track changes.
- After four years, plants were not genetically different from the original cultivar sown, though leaf length of the survivors was slightly shorter.
- Therefore, there was no dramatic shift in the composition of the ryegrass populations, such as might be expected if a particular genetic group of better adapted plants had preferentially survived in the plots while others had died.
- The results were consistent across regions, ryegrass cultivars and sowing rates.



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Ryegrasses slip, slipping away.

When these rapid collapses occur, it is likely that several factors such as droughts, insect pests and soil type combine to push ryegrass over the edge, resulting in plant death. But even when these major stress events are absent, a gradual 'leakage' of yield in the years after sowing is commonly reported.

There are three possible things happening when new pastures are deemed to have failed to persist¹. Understanding these processes and finding ways of preventing them or managing around them are key targets for AgResearch and DairyNZ researchers.

The first is the rapid collapse, or 'implosion', phenomenon noted above. When this happens, the results are obvious: the pasture opens up, weeds (e.g. paspalum) invade, and pasture growth in autumn and winter drops through the floor. This can happen even with the best perennial ryegrass genetics and management practices in place, and signals that the environment is dominating everything else. Perennial ryegrass then becomes, effectively, a short-term option so alternatives such as annual or Italian ryegrasses, or more reliance on annual crops, come into play. More work is required to understand the relative economics and sustainability of these options for affected areas.

Secondly, sown ryegrass plants could die out in the pasture but be replaced by 'volunteer' old ryegrass from the seed bank buried in the soil. In this case, the change would be subtle and hard to see. The seasonal yield profile of the pasture might change because the volunteers are likely to be most productive in spring and less productive in summer and autumn. To confirm this, we would need to generate a DNA fingerprint for plants in the pasture to distinguish the volunteers from the sown plants. Recent research indicates that this is a minor process in our dairy pastures and can be largely ruled out as a cause of poor persistence².

The third possibility is that the sown plants survive in good numbers, but the survivors lose their yield advantage as the environment 'selects' plants with the genes that enable them to survive while the more-productive plants that established originally die out. In this case, the changes would also be subtle and, again, we would need to use DNA fingerprints to confirm what has happened³. We can also use the phenotype (the shape, size and growth habit) of survivor plants to check if there has been some 'shift' in the ryegrass plant population.

Unravelling the processes

In the third example above, a reasonable passage of time is required to allow the processes to play out. The best way to check is to collect survivor plants from pastures sequentially (say, once a year, for at least 5 years), grow them in a common nursery, and compare their DNA and phenotype with plants grown from the original seed line that have not been through the same environmental 'sieve' as the survivors.

This was the aim of the 'Linear trial' which was established by DairyNZ in autumn 2011 at three experimental sites: Northland (Jordan Valley Farm), Waikato (Scott Farm), and Canterbury (Lincoln University Dairy Farm). Four perennial ryegrass cultivars were sown in each region; Commando and Alto (modern diploids), Halo (modern tetraploid), and Nui (older diploid). Nui was infected with the standard endophyte (SE) while the three modern cultivars were all infected with AR37 endophyte. Nui was included because farmers often raise concerns that new cultivars are less persistent than old ones.

Ryegrass seed was sown at five seeding rates (equivalent to 6, 12, 18, 24, and 30 kg seed/ha), because there is also a view that high ryegrass seeding rates (>18 kg seed /ha) reduce plant size and physical survival during the first year after sowing, with subsequent negative effects on sward persistence⁴. Pastures were rotationally grazed by dairy cows when herbage mass reached 2500-3500 kg DM/ha and with a target post-grazing residual of 1600 kg DM/ha.

Vegetative cuttings from all three regions have been collected each autumn for 5 years from pastures sown to Alto and Nui at either 12 kg seed/ha or 30 kg seed/ha. These cuttings have been grown in a spaced plant nursery at AgResearch Ruakura (Figure 1) with 96 replicate plants of each site/cultivar/treatment combination.

The plants are being used to study changes over time in DNA fingerprints and growth habit; by using a nursery, a fair comparison is enabled by ensuring plants are grown under the same environmental conditions.

Figure 1. Four year old Alto and Nui survivor plants growing in the Ruakura spaced plant nursery.



Measuring genetic change over time

Perennial ryegrass cultivars are genetically diverse because ryegrass is an out-crossing species. Hence, each seed that is sown carries a slightly different genetic makeup, the extent of which is governed by the breeders' choice of the parent plants (usually between 4 and 12 in number) that are selected to constitute the cultivar. This means that the 'survival of the fittest' principle can operate on the population because individual plants will differ in their ability to survive in their environment.

DNA markers are a useful tool for investigating genetic changes in forage plant populations, over time or in response to different environmental factors^{3,5}. DNA marker technology was employed to monitor the genetic make-up of the Alto AR37 and Nui SE plants. A panel of DNA markers, called simple sequence repeats (SSRs), was used to 'fingerprint' the Alto and Nui survivor plants growing in the Ruakura nursery. SSR fingerprints of the survivor populations were then compared with those from a sample of the seed originally used to sow the pasture treatments.

Results to-date have revealed that the plants still present in the plots after four years are not genetically different from the cultivar seed that was originally sown. This finding suggests that no dramatic shift has occurred in the composition of the populations, such as might be expected if a particular genetic group of better-adapted plants had preferentially survived in the plots while others had died out.

These trends are true across all three sites for both Alto AR37 (representing 'modern' genetics) and Nui SE ('older' genetics).

Growth habit of the surviving ryegrass plants from the Linear trial swards

The growth habit of the year four Nui SE and Alto AR37 survivor plants was very similar to those raised from the original seed. This result is not surprising, given that the genotyping tests demonstrated very little genetic change. There was no difference in individual leaf weight between the four-year-old survivor plants, and plants raised from the original seed. However, length of the longest leaf blade on each plant was 7% shorter for Alto survivor plants compared with Alto grown from seed. For Nui, the leaf-length of the survivors was 13% shorter than those grown from seed.

Within a cultivar, individual plants will vary in leaf length. We assume that, under the 'best practice' grazing management applied to the pastures, plants with longer leaves have lost a higher proportion of leaf tissue to grazing at each grazing event, gradually weakening the plants and making them more susceptible to other stresses. Over time, more of the longer leaved plants may have died out. The changes in leaf length are relatively small, after four years of grazing, and this probably explains why no differences showed up in the genotyping results.

Discussion and conclusions

We have found that the genetic composition of four-year-old survivor ryegrasses is not significantly different when compared with the original seed sown, and with only very minor changes in leaf length. To answer to the question 'How do ryegrass plants change as a dairy pasture ages?' – it seems very little. These results suggest that ryegrass persistence issues are not caused by progressive loss of less well-adapted plants from the sward, as the sown cultivars have survived and are remaining true to type. This pattern is consistent for new and old cultivars, so there is no evidence that new cultivars are genetically poorer in their ability to survive over time. Other factors, such as grazing management, climatic conditions and insect damage, are the most likely contributors to poor pasture persistence, particularly when two or more stresses are operating at the same time.

For good pasture renewal outcomes, continue following best practise pasture renewal methods, use certified seed, and select appropriate cultivars and endophytes using the DairyNZ Forage Value Index. Careful grazing during establishment and in times of adverse climatic conditions will also aid the persistence of pastures.

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Focus on forages to reduce urine patch N leaching

Practical options for reducing the environmental impacts of intensive, pasture-based livestock systems are required to help farmers meet stringent regulations on nitrate leaching.

Key findings

- The urine patch is the major source of nitrogen loss to the environment on dairy farms.
- Different forages can be used to reduce nitrate leaching, either by lowering the nitrogen loading in urine patches or increasing nitrogen uptake from the urine patch.
- Nitrogen concentration of urine from cows grazing plantain was significantly lower than for cows grazing perennial ryegrass-white clover based pasture.
- Italian ryegrass significantly reduced nitrate leaching compared to perennial ryegrass-white clover pastures. This was due to greater cool season plant growth increasing nitrogen uptake during late-autumn, winter and early-spring.
- Plantain and Italian ryegrass based pastures may be useful for reducing nitrate leaching while maintaining or increasing milksolids production.



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Introduction

Nitrogen (N) from urine patches is a major contributor to N leaching¹, due to the high loading rate of N in urine patches compared with the capacity of many plant species to take up the N. There are two ways in which plants can be used to reduce nitrate leaching losses.

First, by growing forages that lead to livestock excreting urine with a lower concentration of N. Second, by using forages to increase N uptake from the urine patch once the urine is deposited on the soil surface. The Forages for Reduced Nitrate Leaching (FRNL) programme is investigating both pathways.

Lower N loading in the urine patch

Previous studies have demonstrated that cows grazing 'diverse' pastures containing the herbs plantain (*Plantago lanceolata*)

and chicory (*Cichorium intybus*) excrete urine which has lower N concentrations compared with cows grazing standard perennial ryegrass-white clover pastures^{2,3}.

More recent studies have focussed specifically on feeding plantain as a strategy to reduce nitrogen excretion. At Lincoln University, milk production and urinary N concentration were measured in late lactation dairy cows grazing a perennial ryegrass-white clover pasture, pure plantain, or a pasture comprised of 50% perennial ryegrass-white clover and 50% pure plantain by ground area. All cows were offered a similar herbage allowance (Table 1)⁴.

Table 1. Mean milk yield and composition for dairy cows grazing perennial ryegrass-white clover pasture, plantain or 50-50 pasture-plantain⁴.

	Standard Pasture	Pure Plantain	50% Pasture Plantain
Milk			
Milksolids (kg/cow/day)	1.50	1.67	1.60
Milk protein (%)	4.28	4.34	4.29
Milk fat (%)	6.16	5.80	5.52
Lactose (%)	4.95	5.05	5.07
Milk urea (mmol/L)	11.2	9.9	10.9
Urine			
N concentration (g N/L)	5.4	2.4	3.6
Faeces			
%DM	10.9	15.7	12.6
N (%)	3.4	3.5	3.3

Daily milksolids production per cow was 0.17 kg MS greater for cows grazing plantain than cows grazing pasture, with cows grazing 50-50 pasture-plantain intermediate. A striking result was that the urine-N concentration was 56% lower for plantain and 33% lower for 50-50 pasture-plantain than pasture. Previous studies have shown that the excretion of N in urine is linearly related to N intake⁵. However, in this experiment the difference in apparent N intake between pasture and plantain was small at 11 g N/cow/ day, and is unlikely to be sufficient to explain the large difference in urine N concentration. Subsequent studies have indicated that cows grazing the plantain excrete a greater volume of urine which may have contributed to lower N concentration through dilution. Indeed, there is direct evidence to show that plantain causes a diuresis effect (increased urination) when it is ingested by sheep, possibly by reducing reabsorption of water in the kidneys6.

Thus, plantain could offer a pathway toward reducing nitrogen

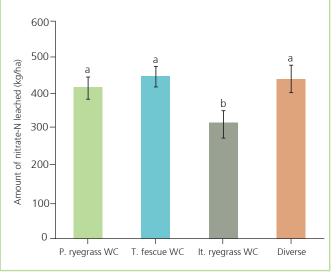
losses to the environment without negative impacts on milk production. The focus of research is now turning to identification of the best ways to capture the potential benefits of the plants in a whole farm system, and ensuring that farmers get credit for adopting the mitigation in the Overseer model.

Capturing soil nitrate

Initial glasshouse studies with a range of grass, legume and herb species have shown a strong positive relationship between N uptake in the cool season and lower nitrate leaching loss⁷. Building on this, lysimeter studies in FRNL have compared nitrate leaching losses from a range of pasture mixes. Large undisturbed soil monolith lysimeters (50 cm diameter x 700 cm deep) were collected from the different pasture mixtures on the Lincoln University Research Dairy Farm and treated with urine to measure the leaching loss. In the first of three studies⁸, nitrate leaching from simulated urine patches was measured under four different pasture types: perennial ryegrass/white clover (P. ryegrass WC), tall fescue/white clover (T. fescue WC), Italian ryegrass/white clover (It. ryegrass WC) and perennial ryegrass/Italian ryegrass/ white clover/red clover/chicory/plantain (Diverse).

Nitrate leaching losses were at around 25% lower under Italian ryegrass WC than under the other pasture species examined (Figure 1).

Figure 1: Total amount of nitrate-N leached from urine patches in the 2011/12 lysimeter study (NB the N loss from the paddock will be a fraction of this loss depending on the urine patch coverage). Means with a letter in common are not significantly different at the 5% level⁸.



The reduction in nitrate leaching under Italian ryegrass WC was attributed to the greater growth rate, and therefore uptake of N from the soil, of the Italian ryegrass during the winter months. Nitrogen uptake of It. ryegrass WC was 1.63 kg N/ha/d, compared with 1.35 kg N/ha/d for P. ryegrass WC and 1.425 kg N/ha/d for T. fescue WC. A second study, conducted in the

Waikato, found no significant difference in nitrogen uptake between P. ryegrass and T. fescue which, when compared with the Canterbury study, was possibly due to the warmer winter conditions in the Waikato allowing the T. fescue to continue growing. Thus, forage options for reducing the nitrate leaching problem may not conform to a 'one size fits all' solution: different forages may work better in some regions than others, depending on the growth characteristics of the species, the regional climate and the timing of the main drainage periods during the year.

In the third study⁹, nitrate-N leaching losses were again lower (35%) from Italian ryegrass than from perennial ryegrass/ WC. This study also included lucerne as a comparison. N losses from the lucerne crop were nearly double those from perennial ryegrass/WC, probably because of the low cool-season growth and, therefore, low N uptake of the lucerne. Poor N uptake in cooler seasons resulted in an excess of nitrate remaining in the soil which was ultimately leached over the main drainage period during the winter.

Future work

These findings were achieved under controlled experimental conditions, and so the N leaching reduction in a commercial setting must be determined. In addition, future experiments will determine the amount of plantain needed in the diet to reduce urinary N, and if mixtures of plantain and Italian ryegrass will offer even greater reductions in N leaching than those associated with one species alone.

Acknowledgement

Forages for Reduced Nitrate Leaching is a DairyNZled collaborative research programme across the primary sector delivering science for better farming and environmental outcomes. The aim is to reduce nitrate leaching through research into diverse pasture species and crops for dairy, arable and sheep and beef farms. The main funder is the Ministry of Business, Innovation and Employment, with co-funding from research partners DairyNZ, AgResearch, Plant & Food Research, Lincoln University, Foundation for Arable Research and Landcare Research.

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Science snapshot

DairyNZ levy funded or supported science

Once-a-day milking during late lactation¹

Once-a-day (OAD) milking during late lactation is often used as a strategy to manage situations with low feed availability and to increase body condition score (BCS). However, the effect of OAD on cow intake, BCS and milk production had not been measured.

To examine the effect of late lactation OAD, fifty-two lactating, pregnant Holstein-Friesian dairy cows were either milked OAD or twice-a-day (TAD) from mid-January until dry-off (12 weeks).

Cows milked OAD:

- Produced 20% less milk during the 12-week period. However, milk fat and protein concentration were greater than cows milked TAD; this resulted in 10% less milksolids. Over the full season, this equated to approximately 4% less total milksolids per cow.
- Had greater blood glucose concentrations, and a reduced expression of genes involved in glucose production in the liver, due to reduced demand for glucose for milk production.
 - Had marginally lower intakes (17.8 vs. 18.2 kg DM).
- Gained an additional 0.25 BCS units, and were 13 kg heavier than cows milked TAD, by dry-off. However, the positive effect on BCS gain did not occur until the cows had been milked OAD for over 6 weeks.
- Blood concentrations of fatty acids and the expression of genes involved in the breakdown of fatty acids indicated reduced use of body tissue stores consistent with the live weight gain.

Next season effects

As cows milked OAD had a higher BCS at dry-off, these cows required less feed than the TAD cows to reach pre-calving target of BCS 5.0. Post-calving, all cows were milked TAD, with no difference in milksolids production, supporting the theory that the mammary gland "resets" itself during the dry period. So, there was no carry-over effect of late lactation OAD into milk production in the next lactation.

Summary

Milking cows OAD for 84 days in late lactation:

- Had little effect on cow intakes
- Increased BCS by 0.25 units
- Decreased milksolids production by 10% during that time and full lactation milk production by 4%
- Reduced feed required during the dry period to achieve target pre-calving BCS

These effects should be factored in when considering OAD milking as a management strategy during late lactation.

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