

DEVELOPING NUTRIENT EFFICIENT CLOVERS FOR NEW ZEALAND FARMERS

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Abstract

Our current strategy is focussed on improving phosphate acquisition efficiency from the soil in white clover, rather than internal phosphate use efficiency. Introgression of traits from some of white clover's close wild relatives into adapted cultivars provides access to more variation than is found in the white clover gene pool alone. For example, *Trifolium uniflorum*, a Mediterranean wild clover has several characteristics indicating adaptation to low fertility soils. If it is crossed with white clover, and the hybrid progeny is then crossed again with white clover, the backcross (BC1) progeny are genetically 75% white clover and above ground they closely resemble typical white clovers. Under controlled conditions, some of these BC1 hybrids grow better than white clover in soils with Olsen P values in the 10 – 20 range. Of more importance may be their characteristic of investing heavily in root growth in lower P soils, i.e. scavenging for P when it is in short supply. As soil P increases they switch over to shoot growth, but unlike white clover they accumulate P in their roots where it is protected from grazing, and made progressively available for shoot growth over a longer period. This trait would be particularly valuable in clovers for hill farming where P fertiliser inputs are less frequent than for lowland systems.

Introduction

The benefits of having more clover in hill pastures are well known, and although clover is still routinely included in lowland seed mixtures, its significance in contemporary high nitrogen (N) fertiliser use dairy systems is debatable. In hill farming, growing more clover promotes animal production directly by improving forage quality, and also by stimulating grass yields and quality through increased N cycling. The main limitations to clover growth in New Zealand soils are soil phosphorus (P) availability levels, seasonal moisture stresses of varying severity and duration, phytotoxic levels of subsoil aluminium in acid soils, and competition from grasses for nutrients, water and light. We briefly review recent research underpinning development of clover with improved nutrient use efficiency for application in New Zealand farming.

Conventional breeding approaches

P responsiveness is genetically controlled in white clover and the heritability of the trait is at levels where selection for the trait is practicable (Caradus et al. 1992). However differences among glasshouse selections for P responsiveness in controlled conditions could not be repeated at a hill farm site (Caradus & Dunn 2000), presumably because stresses other than P were more important. We have recently shown that repeated selection for higher rates of white clover shoot growth in New Zealand breeding programmes over the last 85 years, has

been accompanied by a 10% increase in internal P use efficiency (PUE) (Crush et al. 2015). In this context, PUE is defined as shoot dry weight per unit total plant P uptake. The agronomic significance of this change in PUE is unknown because no P response field trials involving contemporary clover cultivars have been reported, that we are aware of. However there are obviously limits to how far internal PUE can be increased because of the importance of P in most plant metabolic processes. However having some inorganic P held in reserve in the cell vacuoles is probably good insurance against short term fluctuations in P supply from the soil. Consequently the focus of the search to reduce the P fertiliser requirement of clover is now on P acquisition efficiency, i.e. the ability of clover to acquire P from the soil, by manipulating root system morphology and architecture.

Interspecific hybridisation

The discovery that white clover (*Trifolium repens*) is a hybrid between *T. occidentale* and *T. pallescens* (Ellison et al. 2006) has opened the possibility of creating new hybrids of white clover carrying traits of value contributed from its close wild relatives.

T. uniflorum is a Mediterranean species that usually grows in low scrubland that is communally grazed by goats, and burnt off periodically. There are no nutrient inputs apart from what may be recycled in dung and ashes. Nothing was known about the physiology of *T. uniflorum* but it was assumed to be adapted to low soil moisture. A preliminary experiment in sand culture showed that *T. repens* × *T. uniflorum* hybrids backcrossed once to white clover (BC1 hybrids = 75% white clover genes) had higher growth rates than their parental white clover cultivars (Nichols et al. 2014a), at shoot tissue P concentrations, below the minimum requirement for white clover (McNaught 1970). The superior growth of some *T. repens* × *T. uniflorum* interspecific hybrids under low P supply has been confirmed in soil at Olsen P levels in the 10 – 20 mg kg⁻¹ range (Nichols et al. 2014b). One such hybrid family demonstrated a much more plastic allocation of biomass to roots and shoots as soil P levels changed than the white clover parental cultivar. In the lower P soils the hybrid invested much more in root growth than white clover did, presumably because it was scavenging for P. However, as soil P levels increased the hybrid switched rapidly to shoot growth, at the expense of root growth. This characteristic would be useful in hill farm systems where P fertiliser use is often less frequent and at lower rates. Another trait demonstrated by *T. uniflorum* and some hybrids was sequestration of P in the roots as soil P levels increased (Nichols & Crush 2015). We assume that this is an adaptation to intermittent increases in P supply (e.g. from animal manure), where the pulse of P is stored below grazing height and progressively used for shoot growth. Some similar results were reported for Tahora white clover by Hart (1986). This cultivar was bred from hill country ecotypes, presumably with adaptation to low soil P. As P supplies increased, Tahora retained more P in its stolons than Huia white clover – a cultivar bred from lowland ecotypes.

Increasing the moisture stress resistance of forage legumes is important because predictions for future climate forecast increasing frequency and severity of drought in many regions of the country. Competition for irrigation water will intensify in those lowland areas where water is available. Where irrigation is not an option, such as on many hill farms, clover with improved moisture stress resistance will be essential. The interaction between soil moisture and nutrient uptake rates in forages may become much more important under future, drier climates. Interspecific hybrids between *T. repens* and *T. uniflorum* have been shown to have increased drought resistance compared with white clover (Nichols et al. 2014c, 2015). The improved drought resistance was associated with a range of root traits and shoot physiological traits.

Subsoil Al toxicity in acid soils inhibits rooting depth and therefore the volume of exploitable soil water and nutrients. There is variation between and within forage species for tolerance of Al toxic soils (Caradus & Crush 1996; Wheeler *et al.* 1992) but few Al-tolerant cultivars have been released. Nothing is known about the Al tolerances of white clover's wild relatives and this will be examined in future research.

Reducing the ryegrass inefficiency factor

Clover has always been targeted in attempts to improve pasture PUE because it competes poorly with companion grasses for P, so the fertiliser requirement of mixed swards is up to three times greater than that needed to grow the sward components separately (Dunlop & Hart 1987). Ryegrass is extremely competitive for soil P because, compared with white clover, it has finely divided roots with long root hairs (Jackman & Mouat 1972; Evans 1977) and lower rates of P efflux from roots back into the soil (Dunlop & Phung 1999). The turnover rate for fine roots of ryegrass is high - up to $8x\ yr^{-1}$ (Reid & Crush 2013) - so that fine roots are constantly growing into fresh, unexploited soil. The research question is - can we reduce P uptake by the ryegrass component of the sward so more P is available to the clover, allowing a reduction in P fertiliser input rates? This is a new approach and two lines of research will be followed. Shoot P concentration in ryegrass is genetically controlled at heritability levels that suggest it should be possible to breed for lowered shoot P (Cooper 1973, Easton *et al.* 1997). If less P is sequestered in the grass component of the sward, there will be more available to the clover. The second approach will attempt to modify the root system of ryegrass, which is extraordinarily well adapted to low P soils, so that it is less competitive against clover for soil P. Shortening its root hairs will be the first strategy to be examined.

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