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# Opportunities and challenges for breeding perennial ryegrass cultivars with improved livestock production potential

#### T.J. Gilliland<sup>1†</sup>, T. Ball<sup>2</sup>, D. Hennessy<sup>3</sup>

<sup>1</sup>Institute of Global Food Security, School of Biological Sciences, Queen's University Belfast, United Kingdom <sup>2</sup>Deutsche Saatveredelung AG (DSV) United Kingdom Ltd, Top Dawkins Barn, Wardington, Banbury, OX17 1FE, England, UK <sup>3</sup>Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland

#### Abstract

This review addresses key factors and impediments that govern the efficient transfer of nutrient energy from primary producing grassland to ruminant milk and meat. The review focuses on permanent improved grasslands, defined as "swards maintained at a high production potential by grass-to-grass renewal", frequently of a 5- to 10-yr longevity. Breeding progress to date is examined as are the primary objectives for the next generation of cultivars. This involves aligning grass productivity to ruminant demand in three primary aspects, namely intake potential, nutritional value and productivity profile. The opportunity to selectively improve plant traits affecting sward structure, chemical composition, seasonality and ability to persist and perform under farm conditions is evaluated. The EU context involves appraising the impact of variables such as grass species and cultivar, regional abiotic stresses (water, temperature, nutrients, soil type, etc.), biotic stresses from disease and pests, regional diversity in sward management strategies, and the opportunity to minimise the environmental footprint of ruminant farming.

#### **Keywords**

Cultivar • livestock •perennial ryegrass • performance

## Introduction

This paper is not intended to provide a comprehensive review of contemporary ryegrass genetics and agronomy knowledge, or to critique grass breeding strategies for improving productivity, climate tolerance or disease and pest resistance. Such bodies of work already exist (e.g. Humphreys, 1997; Conaghan & Casler, 2011; Kole, 2013a, 2013b). Rather, this paper focuses on current and future opportunities and challenges for improving the efficiency of nutritional energy transfer from primary producing perennial ryegrass (Lolium perenne L.) cultivars to ruminant milk and meat production. To identify the opportunities and understand the challenges require a holistic appreciation of the multifactorial nature of the grass seed industry, as argued by van Wijk & Reheul (1991). In addition to the process of breeding (genetics, genomics, epigenetics, logistics, response timelines and economics), there are official and regional evaluation thresholds designed to promote those cultivars that best furnish a diversity of on-farm end-user requirements (regional practices in grazing, "cut-n-carry" and conservation, tolerance of localised climatic, edaphic, disease and pest stresses). In addition to this is the substantial

complicating factor that grass herbage is not an end product in itself but must be processed through ruminants into meat and milk, which adds key performance requirements associated with intake, ingestion and metabolic utilisation. This multifactorial requirement for cultivar improvement can be more simply defined in terms of three key factors of critical farm-level importance, as follows:

#### "A proficient and sustained delivery of highly utilisable, highyielding herbage".

"Proficiency" requires cultivars that make optimum use of soil nutrients and light capture with the capability to maintain high productivity under varying (or specific) sward management regimes (seasonal grazing patterns, ensilability, set-stocked/ rotational/zero grazed, intensive, extensive, etc.) and regional growing stresses (disease, pest, temperature and moisture). "Sustained" reflects the need for cultivars with greater predictability and reliability across a growing season, over years and throughout as wide a climatic and edaphic range as possible.

<sup>&</sup>lt;sup>†</sup>Corresponding author: T.J. Gilliland E-mail: tj.gill@hotmail.co.uk



"Utilisable" encompasses possibly the greatest challenge for cultivar improvement, namely addressing the poor conversion rates of herbage mass grown, and particularly its protein components, into ruminant product, while profiling productivity closer to livestock demand across a growing season.

Breeding progress in these key factors will, by implication, also address important EU and global political and regulatory policies on environmental protection (Osterburg *et al.*, 2010). Proficient use of input resources reduces nutrient leakage, sustaining a tolerance to stresses also contributes more resilience to climatic change, and better animal utilisation reduces the release of phosphate and nitrogenous compounds (ammonia, nitrous oxide) and methane by ruminants into air and ground waters (Jarvis *et al.*, 1996). There are also substantial economic gains for farm business through a greater contribution from home-grown herbage (Dillon *et al.*, 2005), which in turn benefits rural businesses and communities. These are all factors that are attracting increased political and societal scrutiny (Osterburg *et al.*, 2010).

It has long been known and validated that grazing management to control the sward structure and nutritive value impacts substantively on grazing performance (Pérez-Prieto & Delagarde, 2012) to the extent that it can be modelled (Delagarde *et al.*, 2011); however, this does not preclude the need for breeding progress in these characteristics as only breeding can raise the potential value of grassland above current ceilings. An evaluation of the opportunities and challenges to achieve this can be categorised into a number of key drivers, as described in the following section.

# Opportunities and challenges of improving key breeding drivers

For the purposes of this overview, the key target traits for breeding improved livestock performance characteristics into perennial ryegrass cultivars are subdivided into "gross production", "animal nutrition", "animal intake", "environmental impactors" and "performance consistency".

#### Gross production drivers

There is documented evidence from Northwestern Europe that breeders have successfully and consistently achieved a productivity gain rate of around +0.4–0.6% per annum depending on the yield component and region (van Wijk & Reheul, 1991; Humphreys, 1999; Wilkins & Humphreys, 2003; Sampoux *et al.*, 2011; McDonagh *et al.*, 2016). Similar gains have also been reported in New Zealand (Easton *et al.*, 2002). However, along with the traditionally other important breeding traits of persistence and disease resistance, the primary targets for improvement have differed little over 45 yr (Cooper & Breeze, 1971; Parsons *et al.*, 2011). There are

isolated examples of breeding for novel traits such as watersoluble carbohydrates (WSC) (Humphreys, 1989) or more recently digestible fibres (Parijs et al., 2017), but these are either isolated single-breed examples or of disputed value. Other novel approaches such as introgression of Festuca drought resistance genes into Lolium (Humphreys & Thomas, 1993) or exploratory studies into creating F1 hybrid ryegrasses using cytoplasmic male sterility (Deutsche Saatveredelung AG and Norddeutsche Pflanzenzucht Hans-Georg Lembke KG; personal communication) have not been financially or functionally suited to large-scale/routine breeding. So genetic gain has required progressive increases in total shoot production, making grass yield gains arguably at least as good a breeding achievement as in arable crops. Yield gain alone does not, however, achieve the requirements for greater proficiency, sustainability and utilisation of the herbage produced.

#### Animal nutrition drivers

A review of grassland productivity in Northern Ireland (AFBI, 2017) showed that the average utilised yield on farm was estimated at 5.0 t DM/ha per year (dairy 7.5 t DM/ha per year; beef and sheep 4.1 t DM/ha per year). In Ireland, the average grass utilised on dairy farms in 2015 was 7.8 t DM/ha (Hanrahan et al., 2018). These values fall within a wide variation of what is achievable across Europe, largely due to the broad range in grassland productivity potential and grass-fed livestock densities, impacted by differences in farming practices, for example, nitrogen (N) fertiliser use, mowing and grazing intensity and in supplementary diet levels (Chang et al., 2015). Hence, even within France, grass utilisation varies between 3 and 7 t DM/ha with a small number of farms greater than 7 t DM/ha in Brittany (L. Delaby, INRAE, personal communication). In contrast, the Northern Irish grass growth monitoring service (GrassCheck, www. agrisearch.org/grasscheck) has recorded on-farm yields of 14-16 t DM/ha per year with utilisation approaching 80% on the top 1% of best managed farms. However, as detailed elsewhere, the efficiency of use of ingested herbage energy and protein is disconcertingly low at around only 30% of the total intake. Underutilisation of grass grown has negative implications for nutrient use efficiency in grassland systems (Anon, 2016). It is also the key biological limit to livestock performance from grass, and so not a grass production ceiling but an animal intake and metabolisation shortfall. While this dynamic undoubtedly has evolutionary roots as ruminants evolved to only achieve maintenance plus one calving annually from natural grassland, commercial targets, as exemplified in advisory and press publications, typically set challenging targets of 5,000 L of milk from forage (Price, 2015; Mayne, 2018) and beef live weight gains in the region of 500 kg/yr (Irwin, 2019; Lively, 2019). To derive as much

as possible of this from grass requires breeding advances in the intake and nutritional traits to achieve 80% utilisation on farm. In this context, relatively modest genetic gains have been made in digestibility, at around 0.5–1.0 g/kg DM per annum (Wilkins & Humphreys, 2003; McDonagh *et al.*, 2016). Furthermore, Wilkins & Humphreys (2003) concluded that the traits which impact on nutritive value include crude protein (CP) concentration, WSC, neutral detergent fibre (NDF) and organic matter digestibility (OMD). Selection for high WSC has been shown to improve CP metabolism of grazed grass (Miller *et al.*, 2001) and silage (Merry *et al.*, 2006), and to reduce N excreted in the urine. Therefore, this is an area where greater breeding emphasis is now required to satisfy leading grassland farmers' requirements.

#### Animal intake drivers

The other pillar of utilisation is physical intake by grazing animals. Smit et al. (2005a) showed significant differences between six perennial ryegrass (Lolium perenne L.) cultivars for sward surface height (SSH), bulk density (BD), proportion of green leaf (PGF), tiller density (TD), tiller weight (TW) and length of sheath (LS), but not extended tiller height (ETH) or length of leaf blade (LLB). They further reported that herbage intake was significantly associated with SSH and PGF (Smit et al., 2005b). As this was only observed in one of two experimental years, inconsistency might limit the on-farm benefits from breeding advances in these intake characters. In contrast, a number of experiments have shown that the greater the free leaf lamina (FLL) (Wims et al., 2013; Cashman, 2014) and/or sward leaf content (Gowen et al., 2003; Flores-Lesama et al., 2006; Beecher et al., 2015), the greater the animal performance. For example, Cashman (2014) and Wims et al. (2013) found an average difference of 1.6 kg milk/cow per day between grazed perennial ryegrass cultivars with the highest and lowest FLL content. Furthermore, McDonagh (2017) and Byrne et al. (2018) showed that FLL length is a good indicator of grass utilisation because as it increases, the pseudostem and true stem contents decline, sward digestibility rises, and post-grazing sward height reduces. McDonagh (2017) also found a strong relationship between pre-grazing FLL length measured through the growing season in grazed swards and flag-leaf length in spaced plants. In addition, Sampoux et al. (2011), when comparing seven natural perennial ryegrass populations and 21 cultivars from the last 40 yr, found a clear association of leaf and lamina lengths with spring and summer DM yields. Their data showed a possible negative impact of long leaves on sward persistency, but concluded that breeding for longer leaves and a high leaf elongation rate would improve the interception efficiency of incident radiation during re-growth, which they expect would most significantly increase spring yields.

Tubritt et al. (2018) found significant differences in postgrazing sward heights (3.7-4.8 cm) between 30 perennial ryegrass cultivars when cattle grazed. Disappointingly, there was also a significant negative relationship between grazed yield and post-grazing sward height as the lowest yielding cultivars had the lowest post-grazing sward heights and vice versa. There was, however, clear evidence that this relationship was not obligated ( $R^2 = 0.41$ ) as, for example, some cultivars with similarly good post-grazing sward height values differed significantly by around 3 t DM/ha in grazed grass yield. While consistency over and within years still needs to be established, this evidence of cultivar diversity indicates a potential breeding trait to improve animal utilisation. Furthermore, if ongoing investigation confirms that secondary head development is reduced following lower postgrazing sward heights, this trait would also indicate enhanced herbage grazing guality on farm, which is known to further enhance utilisation (O'Donovan & Delaby, 2005).

Furthermore, McDonagh (2017) showed that differences in FLL, measured in swards, were a good indicator of intake by grazing cattle and that these differences correlated closely to spaced plant-measured leaf length differences (spLL), both when swards were in the reproductive ( $R^2 = 0.88$ ) and vegetative ( $R^2 = 0.99$ ) phases. Given that the sward and spaced plant measurements came from distantly separated locations and different years, FLL/spLL appears to be a trait that both impacts on animal intake/utilisation and is amenable to selection in breeders' plant nurseries.

The magnitude of the benefit to grassland farming of improved grass quality and intake has recently been estimated from AFBI studies in Northern Ireland. Improving grass utilisation by 1 t DM/ha, combined with improved grass quality, can potentially increase the margin over feed costs by £204–334/ha per year on dairy farms or £160–218/ha on beef farms (Anon, 2016). This dairy benefit was largely driven by reduced concentrate feed costs while the improved beef performance was due to a 21% per ha reduction in concentrates, an increase of 19% per ha in stocking rate and an improved live weight gain of 35% per ha from grass. Similarly, Teagasc figures show that a 1 t DM/ha increase in grass utilisation on dairy farms in Ireland is worth an additional €173 net profit/ha (Hanrahan *et al.*, 2018).

#### Environmental impact drivers

There is increasing regulatory pressure on grassland farming to mitigate emissions of greenhouse gases (GHGs), to reduce nutrient losses to ground waters and to sequester carbon into soil sinks (ACRE, 2007). In the UK, 59% of total agricultural ammonia emissions come from ruminant farming (24% beef, 31% dairy, 4% sheep; Misselbrook *et al.*, 2016) with agricultural livestock producing upwards of 9% of total anthropogenic GHG emissions as methane and nitrous oxide (Gill *et al.*, 2010). In a less urbanised/industrialised region such as Northern Ireland, ruminant emissions rise to over 70% of total emissions. In dairy cattle offered 35% concentrates and 65% fresh grass, livestock metabolism studies have shown the following:

- Of the gross energy intake, 30% was lost in excretions, 6% as methane, 36% lost as heat, 23% retained in milk with 5% retained in the body (Hynes *et al.*, 2016a).
- Of the total protein fraction, tracked as total N, only 27% and 2% was retained in milk and body, respectively, with 34% lost in faeces and 37% in urine (Hynes *et al.*, 2016b).
- Of the phosphate ingested, only 33% was transferred to milk with 3.5% retained and the remaining 63.5% excreted, almost entirely within faeces (Ferris *et al.*, 2010).

A critical grass-breeding challenge is, therefore, to decrease these losses by better transfer of the ingested herbage into animal product. The nutritive composition of the grass has a major role to play here, with increased metabolisable energy content of herbage shown to improve conversion into animal product which, for example, can lower nitrous oxide (N<sub>2</sub>O) emissions by reducing N excretion in the urine (Miller et al., 2001). Increasing herbage WSC content has also been implicated in reducing enteric methane eructation from ruminants (Martin et al., 2010; Shibata & Terada, 2010). Here again there is opportunity for breeding intervention as there are many studies reporting cultivar differences in, for example, WSC, CP, fatty acids, fibre digestibility and DM digestibility (DMD) (Wilkins et al., 2000; Miller et al., 2001; Gilliland et al., 2002; Tas et al., 2005; Merry et al., 2006; Downing & French, 2009) with varying evidence of improved intake and animal outputs.

#### Performance consistency drivers

If farmers are to place greater reliance on grass for their livestock nutrient supply, then greater predictability and reliability across varying growing conditions are required. Talbot (1984), in assessing the sources of variation in grass cultivar trials located across the UK, concluded that years and locations imposed the biggest variances and were interchangeable. Therefore, cultivar-testing protocols that involve multiple years, locations and retesting cycles identify those cultivars that not only achieve higher overall performances but also have a greater resilience than those not approved.

On a macro "EU-wide" scale, Table 1 shows the number of perennial ryegrass cultivars registered on the EU Common Catalogue of Varieties (https://ec.europa.eu/food/plant/plant\_propagation\_material/plant\_variety\_catalogues\_databases\_en, 2017) compared to several major forage grass species and arable crops.

Widely adapted cultivars are preferable from an agribusiness aspect as production costs are lower if fewer cultivars are produced in larger quantities. There are, however, complicating agri-economic factors underlying decisions on which country or how many countries a breeder will submit a new cultivar for registration and subsequent commercialisation, including testing costs, market volume, market structure of competing companies as well as the inability to penetrate that market without a local independent recommendation. Furthermore, after making multiple national submissions to gain regionally approved performance data, breeders can reduce ongoing maintenance costs by only retaining one member state registration for EU market rights. Region-specific requirements for high disease resistance (e.g. rust resistance in France, Puccinia spp.) or high winter hardiness (e.g. in Germany and Nordic regions) impose large differentials in the regional adaptation of grass cultivars, but no greater than among arable crops. So, while the data need to be interpreted with considerable caution and given that arable crops mostly have higher value and higher volume markets than perennial ryegrass and must be resown annually, there is no evidence from the numbers of registered cultivars that perennial ryegrass is more widely adapted across Europe than arable cultivars, despite its allogamy.

At a national level, Long *et al.* (2010) reported that of the 120 perennial ryegrass cultivars recommended in the UK and Ireland, only 10% were on all four recommended lists (England and Wales, Ireland, Northern Ireland, and Scotland) and 43% of the cultivars were only approved in one region (Table 2).

This confirms observations from studies on cultivar ranking consistency. Wilkins (1989) and Wims *et al.* (2009) reported re-ranking under different growing conditions and concluded that this justified the need for regional recommended lists and explained regional variations in breeding gains reported

Forage grasses	Perennial ryegrass 6701	Italian ryegrass 248	Hybrid ryegrass 107	
	Meadow Fescue 103	Tall Fescue 324	Cocksfoot 160	Festulolium 44
Arable Crops	Barley 2-row 985	Barley 6-row 393	Wheat 2417	Durum 512
	Oats 358 (+39 naked)	Potato 1632	Sugar beet 1645	Linseed 182

Table 1: Number of cultivars registered on the EU Common Catalogue 2017

<sup>1</sup>Estimated from a total listing of 1093 amenity and forage cultivars

Total number of recommended perennial ryegrass cultivars		120
Proportion recommended by:	Four testing authorities	10%
	Three testing authorities	25%
	Two testing authorities	22%
	One testing authority	43%

 
 Table 2: Comparison of recommendation consistency in the UK and Ireland 2010

by Wilkins & Humphreys (2003). This is understandable as a cultivar that is dormant enough to survive a northern Britain winter would be expected to have low spring yields in the south-west of Ireland, where the winter period is much shorter and prolonged frost is uncommon. Conversely, cultivars that perform well in the south-west of Ireland by not being winter dormant tend to suffer winter damage at northern sites. However, even in a relatively confined and benign maritime region such as Northern Ireland, substantial production variations can occur over relatively short distances. Table 3a shows the variation in yields and weather variances recorded across 30 dairy and beef farms in the six counties of Northern Ireland during May 2017. All these farms were operating to an optimum management level in what were not extreme weather conditions. In this year, growth in Armagh and Down was 0.95 t DM/ha lower than the average of the other counties and 1.25 t DM/ha less than Antrim. By contrast in 2018, when

there was severe moisture stress during much of the main growing season of late June, July and August in some of the regions, daily growth rates were down by 6.4 kg DM/ha overall, with the Antrim and Armagh regions down by 20% in daily growth and by similar reduction in total yield (Table 3b). Such weather extreme responses are not uncommon at a micro level. For example, the long-term perennial ryegrass cultivar performance trials at AFBI Crossnacreevy, Northern Ireland, show 10-yr average yields for perennial ryegrass of 12.3 t DM/ha yr under a simulated grazing management and 16.4 t DM/ha yr under a conservation management, with an annual variation of +/-5.5 t DM and +/-7.2 t DM/ha per year, respectively (Meehan, 2016). Despite this baseline production variation due to seasonal conditions, commercially successful cultivars must not significantly re-rank in their key characteristics. Otherwise, it would be impossible to provide a reliable measure of their relative potential on different farms and in different years. There is both longstanding and recent evidence to show this is not a weakness within the scope of normal management practices. McDonagh (2017) found that eight perennial ryegrass cultivars did not re-rank to any great extent in production, quality or persistence, despite different N application rates and defoliation frequencies. Similarly, Aldrich & Elliott (1974) found no re-ranking between cutting and grazing systems. So, breeding programmes have delivered an acceptable level of resilience in terms of relative cultivar performance, albeit not an immunity to more

 Table 3: Variation in weather and grass growth parameters between NI countries for GrassCheck dairy farms (McConnell, 2018, personal communication)

Region (county)	Total rainfall (mm)	Average daily temperature (°C)	Daily growth rate (kg DM/ha)	Total yield (t DM/ha)
		3a) May 2017		
Antrim	49.9	12.0	101.9	3.16
Armagh	42.4	12.4	63.4	1.97
Down	35.3	11.9	60.3	1.87
Fermanagh	46.8	12.3	87.8	2.72
Londonderry	35.7	12.2	89.0	2.76
Tyrone	53.2	12.3	92.2	2.86
Average	43.9	12.2	82.4	2.56
	3b	) Growing season to end of August 2	018	
Antrim	297	13.1	53.4	9.0
Armagh	295	13.7	46.8	7.9
Down	233	13.2	49.5	8.4
Fermanagh	330	12.4	64.0	10.8
Londonderry	302	12.4	59.8	10.1
Tyrone	351	13.0	68.1	11.5
Average	301	13.0	56.9	9.5

acute weather events such as severe droughts, excessive rainfall or temperature extremes that cause sward production collapses. Challenges of this magnitude, although predicted to be more frequent given climate change modelling (EASAC, 2018), are currently beyond the capability of grass breeding to address either locally or EU-wide. The only partial remedial measures come not from breeding but through local advisory monitoring services such as GrassCheck (www.agrisearch. org/grasscheck). This uses a growth prediction model to forecast expected grass yields 2 wk in advance. Therefore, although breeders desire cultivars that are successful over wide climatic ranges to maximise their sales volume, this is not where increased breeding effort is most required to improve livestock performance from grass.

## **Breeding opportunities and challenges**

Given the considerable challenges posed by the breeding drivers discussed previously, questions arise as to what opportunities breeders have to effectively address them. The British Society of Plant Breeders was asked to survey its perennial ryegrass breeders for this paper. This survey was sent to all members operating in the UK market, which included both UK-based breeders and breeders from Denmark, France, Ireland and the Netherlands. The survey asked them to score their selection priorities for a total of 33 traits, across five groupings, as either A (very important to essential), B (somewhat important/important) or C (useful/irrelevant):

- 8 Productivity Traits: Total herbage production; spring herbage production; summer herbage production; autumn herbage production; first-cut silage yield; second-cut silage yield; third-cut silage yield; overwinter/low temperature growth
- 9 Herbage Quality: Spring grass quality; summer grass quality; autumn grass quality; digestibility; CP content; WSC content; fibre content; fatty acid profile; tannin content
- *4 Structural Parameters*: Lamina length; leaf area index; erect/prostrate habit; sward density
- 7 Resistance Factors: Rust resistance; mildew resistance; Drechslera resistance; other diseases, persistence/ longevity; drought tolerance; cold tolerance/winter kill
- 5 Specialist Characters: Nitrogen use efficiency; phosphorus use efficiency; utilisation under grazing; livestock output measure; lower methane emissions

Only four traits were A-classed by all breeders (total and spring production, spring quality and digestibility), with a further six A-classed by a majority of breeders (first-cut silage yield, sward density, rust and *Drechslera* resistance, utilisation under grazing and persistence/longevity). Fatty acid

profile and tannin content were C-classed as irrelevant by all breeders, but otherwise there was no clear consensus on the priorities of the remaining traits. Interestingly, one breeder had only a single "essential trait" (total yield), while another breeder listed 23 of the 33 traits as A-class priority. Overall, productivity and resistance traits retained high priority with herbage quality and structure less so, despite their importance to animal productivity as already described (Table 4). Specialist characters were also given a high priority, though there was little consensus on the priorities between breeders. A number of "additional" characters were reported by the breeders, each largely specific to an individual breeder. These included fibre or cell wall digestibility, re-heading, livestock wear, tillering, poaching/wear resistance and seed yield.

These results reflect the challenge of breeding for genetic gain in a crop that does not have a singularity of end-use and with the ultimate product depending on the efficiency of the ruminant "end user". When viewed as a totality, they give some indication of the challenge of achieving a multifactorial improvement by targeting progress across a wide diversity of traits.

All except one breeder reported that their germplasm resources were entirely either "elite/improved" (existing commercial cultivars) or "adapted" (of known favourable traits but not from existing cultivars). This one breeder was using between 1 and 5% of "unadapted" germplasm (of high genetic diversity without trait assessment, e.g. wild populations). So virtually all of the breeding effort is seeking to achieve improvements from within the gene pool of current cultivars and associated material.

There have been concerns expressed in some quarters regarding diversity bottlenecks due to this type of recurrent selection strategies (Yong-Bi, 2015), and there are knowledge gaps requiring more research into genetic diversity changes under plant breeding. However, as perennial ryegrass is allogamous, it requires several maternal plants of sufficient diversity to overcome the self-incompatibility genes (Klaas *et al.*, 2011) and ensure commercially viable seed production capability. This implants a greater phenotypic variance within grass cultivars compared to autogamous or clonal cereals.

 Table 4: Percentage of traits in each group assigned to one of three classes of importance: A = very important/essential; B = somewhat important/important; C = useful/irrelevant

Perennial ryegrass trait group	A	В	C
Productivity	55	30	14
Herbage quality	35	35	30
Structural parameters	32	46	21
Resistance factors	52	38	10
Specialist characters	54	11	34

This should make recurrent selection from existing cultivars successful and likely places perennial ryegrasses at a lower risk of entering a diversity bottleneck.

Evidence of this comes from the statutory Plant Breeders' Rights registration schemes in the UK. Table 5 shows the magnitude of differences between plants within registered cultivars for several characteristics measured on spaced plants in Distinctness, Uniformity and Stability tests of perennial ryegrass (data from AFBI Plant Testing Station, Crossnacreevy, Northern Ireland). These data are from registered cultivars, which are therefore "uniform" and so show that a difference in heading date between plants of 2.7-5.4 d can exist. Similarly, flag leaves can range between 3.6 and 6.1 cm in length and 1.2 and 2.2 cm in width, giving a leaf area range of 1.7-3.3 cm<sup>2</sup>. Therefore, there is variance within registered cultivars that can be exploited to improve some traits that are important for animal performance and justifies breeders' focus on using elite maternal germplasm. However, to make progress on a multifactorial basis may require more innovative approaches. It was notable that only one breeder reported using molecular selection methods in their current breeding programme, though several stated that they had future plans to do so. Genomic selection (GS) can facilitate multivariate selection to address the multifactorial resilience requirement, though grasses have lagged behind other crops in the use of these tools. This is partly due to allogamy, as it is harder to capture gene variants within cultivars that are effectively populations compared to autogamous or clonal crops. However, innovations such as the use of mixed models for genomic-wide selection (Bernardo & Yu, 2007) have been shown to enhance the efficiency of selective breeding (Lorenzana & Bernardo, 2009), and Yabe et al. (2018) have demonstrated the potential of GS for breeding by mass selection in a model allogamous species. Costs have, however, been prohibitive in the past and although now

becoming cheaper still represent a significant investment. So, the following three recent examples of genomics in grass breeding have all involved investment from public or academic funders.

- The forage grass breeding programme of the Institute of Biological, Environmental and Rural Sciences in Wales (www.aber.ac.uk/en/ibers) exploits its academic links within the University of Aberystwyth to conduct "public good" breeding and attract funding from the Government (Defra, www.gov.uk/government/organisations/department-forenvironment-food-rural-affairs) and research funders such as the Biotechnology and Biological Sciences Research Council (BBSRC) (www.bbsrc.ac.uk), the Technology Strategy Board (www.gov.uk/government/organisations/ technology-strategy-board) and a seed industry company.
- Teagasc, Oakpark, Ireland, for a medium-sized genomics screening capability, required Government (Department of Agriculture, Food and the Marine [DAFM]) funding (www. teagasc.ie).
- . DLF A/S, in Denmark, linked with Aarhus University (www. au.dk/en) for a genomics programme in 2011 and after 5 yr had examined around 1,800 ryegrass families, all with phenotypic data and had identified 1.8 million DNA markers (approximately 1% of the 2.7 Gb ryegrass diploid genome). This was an "Industrial PhD studentship" at AU, jointly run by DLF A/S and supported by public funding from the Danish Ministry of Education, through the Council for Industrial PhD Education (11-109967), (http://ufm.dk/ en/research-and-innovation/funding-programmes-forresearch-and-innovation/find-danishfunding-programmes/ programmes-managed-by-innovation-fund-denmark/ industrial-phd). The work demonstrated the potential for GS in perennial ryegrass (Fè et al., 2015) and has been further supported through a "public good" methane emissions study, partnered with Tystoftefonden (www.tystofte.dk) and

Character name	Flowering date	Spring angle	Spring height	Spring width	Spring shape
Units	days	degrees <sup>1</sup>	cm	cm	cm
Maximum	5.4	12.4	11.7	11.3	0.28
Minimum	2.7	7.1	3.9	6.8	0.10
Average	3.8	9.8	8.6	9.1	0.18
Character name	Tiller height <sup>2</sup>	Plant width <sup>3</sup>	Flag leaf length	Flag leaf width	Flag leaf area
Units	cm	cm	cm	mm	cm²
Maximum	13.6	12.7	6.1	2.2	3.3
Minimum	9.0	7.7	3.6	1.2	1.7
Average	11.6	10.3	5.0	1.7	2.6

<sup>1</sup>Angle from ground level

<sup>2</sup>At ear emergence

funded under the Green Development and Demonstration Program, Ministry of Environment and Food, Denmark (www.mst.dk/service/om-miljoestyrelsen).

Other grass breeders, such as DSV, target specific traits, seeking causal genes for characters in quality traits (e.g. lignin synthesis) and plant structure (e.g. lamina length). There is a growing body of scientific evidence and experience in cytogenetics, genotyping, next-generation sequencing and bioinformatics to support further progress in this area. Evidence of synteny across genera in the Gramineae (or Poaceae), such as Brachypodium, rice (Oryza sativa), sorghum (Sorghum bicolor) and more recently the barley (Hordeum vulgare) genome (Mayer et al., 2011), novel approaches such as Targeting Induced Local Lesions in Genomes (TILLING) (Manzanares et al., 2016), or categorisation of specific quantitative trait loci, for example, leaf length in ryegrass (Barre et al., 2009) and improved cell wall degradability (Bolwell, 2000; Barrière et al., 2003) are opening new opportunities. Therefore, these examples indicate that new opportunities to breed for greater resilience and to expect breeding progress on a multifactor are no longer such an unattainable goal. However, the challenge for breeding business models is to find funding streams where they cannot entirely self-fund genomic breeding.

#### Cultivar evaluation opportunities and challenges

For many years, official evaluation schemes across Europe sought evidence of improvement in individual traits, the most important of which were DM yield and persistency, with resistance to any acute regional disease/climatic factor being a baseline requirement. Latterly, digestibility was introduced, but improvement continued to be required on a trait-by-trait basis, with "overall performance" only used for marginal pass/ fail decisions. It was accepted that these traits were detached from the animal product as animal-based trialling demands resources beyond what most testing authorities can provide or breeders afford to fund. Hence, small plot field trials are expected to continue for large-scale cultivar evaluations to reduce candidate numbers to a smaller set of new elite performing cultivars (Conaghan et al., 2008). However, given the body of evidence presented previously showing the need for progress in characteristics that are now known to impact on animal intake, nutrition and environmental footprint, a more multifactorial approach to cultivar evaluation is clearly required. There are already a number of evaluation schemes that use indices to provide an overall performance indicator for perennial ryegrasses.

The French small plot evaluation scheme (Reglement technique d'examen des varietes de plantes fourrageres et

a gazon; www.gnis.fr/reglementation-semences) applies an index rating to cultivars after they have passed the baseline evaluation criteria. Coefficients are applied to the yields (1–100% on measured values and 1–9 for notes) for a range of characteristics (total and seasonal yields, reheading, resistance to diseases, operational flexibility, flexibility of the foliage, persistence/sustainability and nutritional value – total N composition/acid detergent fibre [ADF] content/ soluble sugar content), to give an overall rating. This allows for inferiority in some characters to be offset by favourable expression in others, but with an elimination threshold set for characters of major agronomic importance, such as reheading and rust resistance.

 Similarly, the Dutch Plantum index for grasses (Protocol Beslissingen Opname en Afvoer Engels Raai Voeder 2019, Commissie Samenstelling Aanbevelende Rassenlijst; https://rassenlijst.info/ajax\_frontoffice/filemanager/files/ wp-uploads/2018/11/Engels-raaigras-beslissingen-2019def.pdf) assigns weighting and scale factors to total yield, first-cut silage yield, crown rust and ground cover, with minimum standards for winter hardiness and all but the first-cut yield.

These indices attempt to represent the overall value of a cultivar for animal production, based on the small plot characters, as listed. An alternative approach is to form indices based on the calculated financial value to the farm business of the herbage produced. So, DairyNZ uses the Forage Value Index (www.dairynz.co.nz/feed/pasture-renewal/selectpasture-species/about-fvi) in New Zealand. This is calculated from the economic value of the seasonal yields on a regional basis and each cultivar's DM production level for each seasonal period. Cultivars are given a star rating to indicate an estimated annual value to the farm of -\$78 to +\$29 (1 star) up to +\$351 to +\$458 (5 star), using an online selection tool. In Ireland, Teagasc have developed a Pasture Profit Index (PPI; www.teagasc.ie/crops/grassland/pasture-profitindex) that provides predicted economic values for cultivars that have been recommended by the Government (DAFM). This research-based index (McEvoy et al., 2011; O'Donovan et al., 2016) assigns a financial value to each cultivar based on the seasonal (spring, summer, autumn) and total yield, persistency and digestibility of the herbage produced in small plot trials. In both of these financial-based indices, there has been a very strong engagement of farmers and an invigorated interest in grass cultivars, more production from grass and reseeding with better cultivars.

Teagasc have implemented a further level of farmer involvement by establishing a network of 66 farms to evaluate 11 currently recommended cultivars, sown in monoculture under intensive grazing systems (Byrne *et al.*, 2017). The farmer monitors herbage production through weekly paddock cover estimations (O'Donovan et al., 2002) and uploads data into PastureBase Ireland (PBI), a farmer decision support tool and database (Hanrahan et al., 2017). Sward guality and ground score are measured by trained technicians at fixed periods during the season. In 2017, there was a range of 1.9 t DM/ha in total annual DM production, 44 g/kg DM for DMD and 0.7 for ground score. There are a number of implications of this scheme. It requires a large number of farmers who will commit and adhere to the evaluation scheme and incurs greater variances as not all cultivars are on all farms, sown in the same year and must account for regional conditions and farmer practice differences. It also cannot be conducted until after the initial small plot scheme has identified the few elite cultivars from among the many candidates. It does, however, produce performance data under actual farm grazing, across the actual range of farming conditions and greatly enthuses the "co-research" farmers (and their neighbours). The final phase in this PPI/PBI initiative is to adopt characters as described in earlier sections that enhance animal intake and utilisation and reduce nutrient losses to the environment, so that the indices and evaluations comprise a multifactorial calculation of key animal performance drivers.

An unexpected implication has been that some of these coresearch farmers are moving away from using mixtures to sowing single cultivars. As they have gained experience in using single cultivar swards and identified specific ones on which their livestock has optimised, they have sought to expand the area available to graze. Without this precision management experience, the loss in flexibility gained from mixtures may be detrimental to less grass-skilled farmers. If this became common practice, there would also be considerable problems for breeders and seed producers meeting the needs of a cultivar-demanding market that could change rapidly. However, the "milchindex" trademark has been successfully used by DSV to market cultivars of high digestibility and quality in Germany as a premium brand in an otherwise price-sensitive market. This typifies the potential benefits that breeders might gain if grass cultivars were more specifically defined for their animal performance potential to farmers.

#### **Concluding remarks**

It is widely accepted that improved efficiency of animal production from grass is the ultimate goal of forage grass breeding for European temperate regions (Wilkins & Humphreys, 2003), and identifying cultivar characteristics that can be highly utilised and influence animal performance is important from all aspects of grassland (Wims *et al.*, 2013). Grass breeders must, as always, seek to be at the forefront of such grassland improvement. Today, this is increasingly driven by the twin need to fulfil leading grassland farmers' requirements for quantifiable animal performance at grass

and the regulatory imperatives to reduce GHG loses to the atmosphere and nutrients to ground waters. As overviewed in this paper, there is ample research evidence of grass characteristics that will enhance animal intake, metabolic utilisation and so livestock production from home-grown grazed and conserved perennial ryegrass. These traits will equally help lower the environmental impact of ruminant farming. It is also clear, however, that in assessing the evidence for such "animal performance" characters, there are often qualifications needed regarding repeatability, accuracy, magnitude of the value, interaction with and independence from other characters, as alluded to. Notably, the evidence from Smit et al. (2005b) of inconsistent animal performance responses to the expressed differences in grass structural traits indicates that it will be incumbent on farmers to manage their swards with sufficient precision to fully gain from any such breeding advances. Leading grassland farmers are, however, already operating at this level of expertise. Therefore, none of this should be an impediment to grass evaluators now actively adopting novel characteristics, particularly as the use of indices provides scope to account for any uncertainties, while still delivering a multifactorial assessment of the animal value of new cultivars. With these in place, breeders will be able to target improvement in multiple traits, which the straw poll indicated is within their current capability either by conventional methods or through their ambitions around GS. There is, however, some evidence that developing cultivars with high animal-value characters could lead to more use of single cultivar swards with implications for the business models of breeding companies and seed producers.

Taking the available evidence overall, it is clear that published grassland research has identified a number of opportunities to improve animal performance from grass by adopting intakeand digestion-supporting traits into candidate cultivar listing procedures. For these traits, further research in not necessary nor are sources of new diversity required, beyond that currently available among commercial breeding stocks. The challenge is therefore for cultivar evaluators to adopt these traits into their listing decision processes. Even where the benefits are difficult to quantify, indices and expert panel weightings can be used to promote candidate cultivars with high expression of these characteristics. This will then encourage and challenge breeders to include these traits in the selection of their elite synthetics and drive genetic gain towards enhanced livestock productivity from perennial ryegrass. Evidence of cultivars with improved animal performance and reduced environmental footprint potential will be highly valued by leading farmers and government policy regulators. The benefit to breeders is that once grass cultivars are individually recognised for their financial contribution to farm efficiency and profitability, the opportunity to broker a seed price that better reflects their true value and which can better offset the cost of development becomes an attainable goal. This should also act as a catalyst for continued research into new means of enhancing the livestock production potential of perennial ryegrass to the benefit of all the stakeholders.

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