An Roinn Talmhaíochta,
Bia agus Mara
Department of Agriculture,
Food and the Marine

Research Stimulus Fund

Final Report

'Classical and Genotypic methods of increasing grassland productivity through Breeding Next Generation Grass'

DAFM Project Reference No: 11 S 109

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Collaborating Research Institutions and Researchers:
Teagasc Oakpark, Teagasc Grange, Teagasc Oakpark – Celeup, AFBI

Please place *one* "x" below in the appropriate area on the research continuum where you feel this project fits

Basic/Fundamental ⟶			Applied ⟶			Pre Commercial
1	2	3	4	5	6	7
			*			

Please specify priority area(s) of research this project relates to from the National Prioritisation Research Exercise* (NRPE) report;

Priority Area (s)	Smart and sustainable Food production and Processing

Research in this priority area seeks to improve the competitiveness of agriculture, food and the wider bioeconomy, to support sustainable farming and the environment, and to encourage diversification of the rural economy and enhance the quality of life in rural areas. Technologies related to Sustainable Food Production and Processing are being developed in response to minimising agriculture's impact on the environment, and supporting the efficient and sustainable use of resources (e.g. water, soil and fertilisers) in the food production process over the long-term.
The impact of this project in the overall development of increased sustainability for food production systems cannot be over emphasized. Grazed grass is the fundamental building block of out food production system and this project only reinforces Ireland commitment to improving the environmental, economic and social resilience of the system.

Key words: *Grass breeding, Perennial Ryegrass, Genomic Selection, Sustainable Livestock Systems*

1. Rationale for Undertaking the Research

Grazed grass should provide the basis of sustainable livestock systems in all regions of Ireland, as grazed grass is the cheapest source of feed available (Finneran et al., 2011). This project set out to establish new selection tools to achieve optimal breeding and evaluation goals for perennial ryegrass. The quantification of breeding progress (through cultivar longevity) was undertaken on commercial farms. This was a novel project approach to grassland research which has not previously been undertaken in the world.

Speeding up the rate improvement in breeding elite ryegrasses that also increase the age to which swards can be maintained at full productivity before renewal. This will contribute to enhancing the competitiveness of pasture-based systems of production, to preserving the rural landscape and to promoting an environmental and animal welfare friendly image for livestock production.

2. Research Approach

This project adopted a multidisciplinary approach, whereby grassland researchers, modellers, nutritionists, geneticists, grass breeders, grass biotechnologists, molecular biotechnologists and recommended list evaluators came together to understand and increase the knowledge base in grassland production and grass breeding/biotechnology. The experimental approach in Tasks 1 and 2, followed that of long-term experimental studies, which was completed under grazing and frequent harvesting. A new approach was used in Task 1, whereby a large commercial farm study was used to identify the long-term performance and persistence of grass cultivars. In Task 2, data from the EU Common Catalogue examined the impact of sward structure on animal performance. This work can have major implications in using sward structural parameters in breeding goals. In Task 3, a modelling approach was used. Breeding goals need to be in line with the requirements of

profitable commercial grassland farms. The Moorepark Dairy Systems Model (MDSM; Shalloo et al., 2004) has been successfully used to develop economic indices for the EBI and the grass selection index; it was used again in this task to establish the economic impact of changing the main breeding goals in grass breeding programs. One of the main objectives of this research task is to further establish the economic effects of sward longevity and persistency. In Task 4 to 6, the use and application of genomic selection will be scoped out in perennial ryegrass breeding. While grass breeding is a slow process the application of GS could decrease the generations required to produce new cultivars by 5-8 years. The starting point was to establish a phenotyping population for use in genomic selection studies, this will be 2000 plants generated from within the Oakpark grass breeding program. The design of the training population (e.g., number of genotypes, number of populations and the population mating structure vis-à-vis full-sib families, half sib families and synthetic varieties) will be of vital importance for determining marker effects. The follow-on process was to establish a robust, high throughput GBS protocol that effectively interrogates the SNP content of the gene space of perennial ryegrass at a density likely to be useful for GS. The selection of the appropriate statistical model to identify QTL (quantitative trait loci) and their effects. The final aspect of this work was to develop a protocol for the incorporation of GS in commercial perennial ryegrass breeding programmes. This was successfully achieved.

3. Research Achievements/Results

The economic ranking of grass varieties from DAFM recommended lists has been made and incorporated into the DAFM RL. Within task 3 the successful adoption of the Pasture profit index has been successful in Ireland and is well adopted by the grassland industry. Task 1 and 2 were feeder Tasks to Task 3. The introduction of on farm evaluation studies is very unique in the world of grassland research; however this project has shown its impact to the adoption and integration of the PPI. Subsequent improvements to the PPI and additional traits to the index will be added in the next years based on the findings of this project.

The area of genomic selection was established into a grass breeding program in Task 4 to 6. The successful establishment of a training population for heading date and rust resistance, measurements were completed and appropriate statistical protocol enforced. The subsequent successful adaptation of a GS protocol was establishment and deployed to the grass breeding program during the course of the project. Both of these results will have significant impacts in future years for the Irish grassland industry.

4. Impact of the Research

This work had multiple impacts, the research undertaken has many positive impacts on grassland and the information disseminated on grass varieties. The evaluation of grass varieties showed clearly how varieties differed in milk production and canopy morphology. The introduction of the Pasture profit index was widely adopted by the Irish grassland

industry and gave clear guidance to the industry on the route forward for grass breeding and evaluation. The creation of a stakeholder group to discuss, generate new ideas and roll out he index was extremely positive and had major impacts. The introduction of genomic selection to the grass breeding program has had many positive effects on developing a clear thought process on how integrate an important technology into classical plant breeding.

4(a) Summary of Research Outcomes

(i) Collaborative links developed during this research

The on farm evaluation of grass varieties has developed a clear network of participating farmers and industry stakeholders, this has been vital for the increase of awareness of the development of the Pasture Profit index and the role out of on farm grass variety evaluations. A number of industry collaborations have been built from this work.

(ii) Outcomes where new products, technologies and processes were developed and/or adopted

In Work package three, new ideas and traits have been generated from the on-farm variety work, this will have a positive impact on the future proofing of the Pasture Profit Index. It will add enormously to developing new traits for the index which will have future impacts for the Irish Grassland Community.

In Work Package four the establishment of a sufficiently large training populations in forage breeding required significant investment and is beyond the capabilities of many small-medium sized breeding programmes. The work completed in this task has enabled Teagasc to establish and accumulate a database of phenotypes on a large training population over numerous years; thereby laying the groundwork for a selection scheme that can exploit genomics-assisted breeding. Teagasc is focussed on breeding forage grasses for the Irish agri-food sector and adoption of new technologies, such as genomic selection, will ensure we can continue to bring high-performing cultivars to market.

(iii) Outcomes with economic potential

On farm grass variety evaluation has clearly shown the importance of evaluating grass varieties on commercial farms. End users are directly impacted by the performance of individual varieties, this work clearly showed this impact.

It will happen after the project end date; However we expect to develop new varieties from genomic selection form the developments within this project.

(iv) Outcomes with national/ policy/social/environmental potential

The outcomes of this project have leveraged further Teagasc internal strategic funding to continue genomic selection on new populations of interest to the Teagasc breeding programme, e.g. tetraploid breeding populations. Furthermore, plant material from the training population has been intercrossed and full-sib families developed for yield evaluation (established in 2018 for evaluation in 2019-2020). This will enable the development of predictive models for forage yield using parental genotypes (task five); followed by implementation of genomics-assisted breeding for yield, quality and crown rust resistance.
In the future, we would expect DAFM to bring on a new RSF call, on grassland and would we will anticipate putting another project through this call.

4 (b) Summary of Research Outputs

(i) Peer-reviewed publications, International Journal/Book chapters.

O'Donovan M, McHugh N, McEvoy M, Grogan D, Shalloo L (2017). Combining seasonal yield, silage dry matter yield, quality and persistency in an economic index to assist perennial ryegrass variety selection. Journal of gricultural Science 2017;155(4):556-568; doi 10.1017/s0021859616000587.

Byrne N, Gilliland TJ, McHugh N, Delaby L, Geoghegan A, O'Donovan M (2017).Establishing phenotypic performance of grass varieties on Irish grassland farms. Journal of Agricultural Science 2017;155(10):1633-1645; doi Doi: 10.1017/s0021859617000740.

Arojju, S.K., Barth, S., Milbourne, D., Conaghan, P., Velmurugan, J., Hodkinson, T.R. and Byrne, S.L., 2016. Markers associated with heading and aftermath heading in perennial ryegrass full-sib families. *BMC Plant Biology*, *16*(1), p.160.

Byrne SL, Conaghan P, Barth S, Arojju SK, Casler M, Michel T, Velmurugan J, Milbourne D (2017) Using variable importance measures to identify a small set of SNPs capable of predicting heading date in perennial ryegrass. Scientific Reports 7: 3566, doi:10.1038/s41598-017-03232-8

Arojju SK, Conaghan P, Barth S, Milbourne D, Casler MD, Hodkinson TR, Michel T & Byrne SL (2018) Genomicprediction of crown rust resistance in *Lolium perenne*. BMC Genetics, doi.org/10.1186/s12863-018-0613-z

Using variable importance measures to identify a small set of SNPs capable of predicting heading date in perennial ryegrass (2016) Byrne SL, Conaghan P, Barth S, Arojju SK, Casler M, Milbourne D. *Scientific Reports* (under review).

Genomic prediction of crown rust resistance in perennial ryegrass and factors influencing predictive ability (2016) Arojju SK, Barth S, Milbourne D, Conaghan P, Hodkinson T, Casler M, Byrne SL. *G3* (under review).

(ii) Popular non-scientific publications and abstracts including those presented at conferences

Cashman, P. O'Donovan, M Gilliland, T. McEvoy, M. Quantifying ground score on perennial ryegrass swards exposed to different grazing regimes. Agricultural Research Forum, 2014 pp89

Cashman, P. O'Donovan, M Gilliland, T. McEvoy, M. Population selection within perennial ryegrass cultivars under simulated grazing. Grassland Science in Europe, Vol 19 - EGF - The future of European Grasslands, 833.

McEvoy, M. McHugh, M. O'Donovan, M Grogan, D. and Shalloo, L. Pasture Profit index; updated economic values and inclusion of persistency. Grassland Science in Europe, Vol 19 - EGF - The future of European Grasslands, 843.

McEvoy, M. O'Donovan, M. Grogan, D. and Shalloo, L. Using the grass economic index to quantify the economic performance of perennial ryegrass cultivars. Agricultural Research Forum, 2014 pp55

Cashman, P. O'Donovan, M Gilliland, T. McEvoy, M. Grazing utilisation of perennial ryegrass cultivarsAgricultural Research Forum, 2014 pp56

Cashman, P. O'Donovan, M Gilliland, T. McEvoy, M. Milk production performance of lactating daoiry cows grazing four different perennial ryegrass cultivars Agricultural Research Forum, 2014 pp60

M O'Donovan Pasturebase Ireland - a national grassland database. Plant biology europ FESPB/EPSO 2014 Congress

M O'Donovan Moorepark Open day 01/07/2015

N Byrne Moorepark Open day 01/07/2015

N Byrne, D Berry, A Geoghegan, L Shalloo, T Gilliland, M O'Donovan. Evaluation of fry matter yield of ryegrass varieities on irish grassland farms. Grassland and Forages in high output dairy farming systems, Vol 20 - EGF - Grassland Science in Europe, 383.

J McDonagh, M McEvoy, T Gililland, M O'Donovan. Perennial ryegrass variety ranking responeses to inclusion of white clover and altered nitrogen fertility. Grassland and

Forages in high output dairy farming systems, Vol 20 - EGF - Grassland Science in Europe, 457.

J McDonagh, M McEvoy, T gilliland, M O'Donovan. Milk production responses of lactating dairy cows to sward structure differences between perennial ryegrass varieties. Agricultural research forum 2015 pp 38.

J McDonagh, M McEvoy, T gilliland, M O'Donovan. Effect of perennial ryegrass ploidy and nitrogen input on white clover content and stolon mass under intensive grazing. Agricultural research forum 2015 pp 43.

N Byrne, D Berry, A Geoghegan, L Shalloo, T Gilliland, M O'Donovan. Evaluation of fry matter yield of ryegrass varieties on irish grassland farms. Agricultural research forum 2015 pp 45.

5. Scientists trained by Project

Total Number of PhD theses: 4

Justin McDonagh, PHD, Queens University Belfast, Teagasc Moorepark - Improving the productivity of perennial ryegrass (Lolium perenne L.) pastures in dairy systems in Ireland; Genetic gain, genotype by management interactions and grazing efficiency.

Pat Cashman, PHD, Queens University Belfast, Tegasc, Moorepark – Differential productivity and persistency responses to simulated and animal grazing of perennial ryegrass genotypes.
Janaki Velmarugan, PHD, TCD, Teagasc Oakpark, Celup – Markers associated with heading and aftermath heading in perennial ryegrass full sib families.

Nicky Byrne, PHD, Queens University Belfast, Teagasc Moorepark - Improving evaluation and selection criteria of grass varieties by linking phenotypic performance from research and commercial farm environments

Total Number of Masters theses: 0

6. Permanent Researchers

Institution Name	Number of Permanent staff contributing to project	Total Time contribution (person years)
Teagasc Moorepark	2	1.16
Oakpark Grass	1	0.95
Oak Park Celup	2	1.39
AFBI	1	0.02
Total	**6**	**3.52**

7. Researchers Funded by DAFM

Type of Researcher	Number	Total Time contribution (person years)
Post Doctorates/Contract Researchers	None None	
PhD students	2	2.59
Masters students	None	
Temporary researchers	None	
Other		
Total	**2**	**2.59**

8. Involvement in Agri Food Graduate Development Programme

Name of Postgraduate / contract researcher	Names and Dates of modules attended
N/A	

9. Project Expenditure

Total expenditure of the project:	€622,690.57
Total Award by DAFM:	€622,690.57
Other sources of funding including benefit in kind and/or cash contribution(specify):	€0

Breakdown of Total Expenditure

Category	Teagasc Grassland Moorepark and Grange	Teagasc,Grassland Oakpark	Teagasc Celup	AFBI	Total
Contract staff	125,459.82	54,203.67	114,014.41	0	293,677.90
Temporary staff					
Post doctorates					
Post graduates	25,856.27	20,300.66		0	46,156.93
Consumables	4,711.44	15,240.21	19,108.23		39,059.88
Travel and subsistence	7,753.43	3,167.61	3,173.20		14,094.34
Sub total	163,780.96	92,912.15	136,295.84	0	392,989.05
Durable equipment	18,226.71		4,500		4,500
Other		27,958.81	61,119.28		107,304.80
Overheads	49,134.29	27,873.65	40,888.78		117,896.72
Total	**231,141.96**	**148,744.61**	**242,804.0**	**0**	**622,690.57**

10. Leveraging

Work Package 4 (Genomic selection) has been crucial in laying the groundwork for future research activity focussed on expanding Teagasc activities in genomics-assisted forage breeding. Using the established population as a foundation, research activities are on-going and will continue for the foreseeable future, and we are actively competing for H2020 funds directly on the back of work completed in this task.

In WP five, knowledge and expertise gained on high throughput genome-wide genotyping in plant genomes and associated analysis of big genomic data sets has resulted in establishment of in-house expertise in forage genotyping and analysis. These capabilities are being leveraged for deployment of genomics-assisted breeding strategies in the Teagasc programme.

11. Future Strategies

The on farm grass variety evaluation of grass varieties is continuing post this project and has been extended to 120 farms. This work will be to clover on farm evaluation in the next months. The genomic selection incorporation into the grass breeding program is ongoing and has been stimulated from this work. The first breeding crosses from this work were made in 2017 and are continuing to be progressed in the current year.