



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

Research Stimulus Fund

Final Report

'Maximising the Genetic Potential of Young Elite Bulls - A Multidisciplinary Approach; BullMax'

DAFM Project Reference No: RSF 11/S/116

Start date: 01/1/13

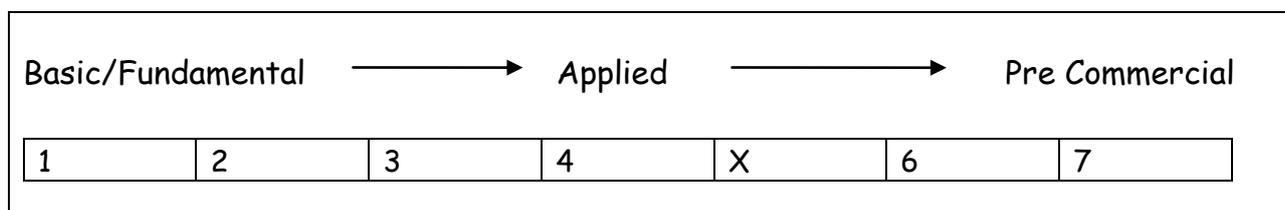
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Please place one "x" below in the appropriate area on the research continuum where you feel this project fits



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

Priority Area (s)	Priority Area I - Sustainable Food Production and Processing
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1. Rationale for Undertaking the Research

Since the abolition of milk quotas the Irish dairy industry has been in a phase of rapid expansion. The underlying basis for this is that Ireland's seasonal grass-based milk production system possesses a significant cost advantage in the form of low-cost milk production as well as environmental sustainability. This system is highly dependent on excellent reproductive performance with compact calving to coincide with the start of the grass growing season. While milk production per cow has increased since the 1970's, reproductive performance has dramatically declined and in order to combat this decline in fertility, Ireland, through the Irish Cattle Breeding Federation (ICBF), has implemented an Economic Breeding Index (EBI). This is a profit index that enables farmers to select the most elite and profitable bulls to breed replacement heifers which will have an increased ability to produce more milk solids per cow per year from grass.

In 2009, Ireland began to select dairy bulls for use in artificial insemination (AI) programmes using genomic selection and now potential AI bulls, with the highest EBI values, are being identified within weeks of birth. Once these young bulls have undergone puberty they enter into an AI centre but yield a low volume of semen and demand for their semen far exceeds supply. Therefore, it is imperative that the industry devises a clear and repeatable protocol for the rearing of young dairy bulls from birth, so as to facilitate collection of high volumes of quality semen in their first breeding season. In parallel to this, and given that over 60% of dairy inseminations occur in a 6-week period in April/May in Ireland, the extensive use of liquid semen is a viable option, whereby, the number of insemination doses per ejaculate could be increased by up to 10-fold compared to frozen semen. Also, the potential for the use of sexed semen is immense, especially during rapid herd expansion; however, its effect on fertility has never been evaluated under an Irish grass-based system.

2. Research Approach

Key industry stakeholders, who worked alongside the research team for the duration of the project informed the hypothesis of this project. The team used a combination of *in vitro* and *in vivo* research approaches. To characterise the early calthood nutritional regulation of puberty we employed a calf model using nutritional and health treatments indicative of that used in industry as the control, while focusing on time periods around puberty and sexual maturation which were relevant to the industry. Our work relating to liquid semen storage focused on improving the status quo, firstly, by completing a wide range of experiments on the effect of sperm concentration, diluents, temperature and antioxidants on sperm physiology using state-of-the art *in vitro* techniques such as flow cytometry, computer assisted sperm analysis and *in vitro* fertilisation. Once treatments were narrowed down, we completed large scale field trials using tens of thousands of inseminations. Similarly, we completed a large scale field trial to ascertain the fertility of both liquid and frozen-thawed sexed semen when inseminated into both cows and heifers under the Irish seasonal grass based dairy and beef production systems. This was backed

up with *in vitro* sperm functional data which characterised the physiology of sperm during storage. Finally, we used a systems modelling approach to assess the role of sexed semen under differing herd expansion scenarios.

3. Research Achievements/Results

Rearing protocols for young bulls:

We have clearly demonstrated that bulls which grow faster in the early calthood period have a more developed hypothalamic-pituitary-testicular axis as assessed through semen production as well as through gene expression, protein expression and hormonal profiles in comparison to dairy bulls of similar age but which grow slower. Specifically, bulls on a high plane of nutrition in the first 6 months of life reach puberty sooner than bulls on a low-moderate plane of nutrition. Increasing the plane of nutrition, of previously diet restricted bulls, during the second 6 months of life did not advance puberty, despite dramatically improving bodyweight gain. In addition, bulls on a high plane of nutrition for the first 18 weeks of life had altered gene expression in their testes which lead to more advanced stages of spermatogenesis within their testes. Dietary supplementation with either n-3 or n-6 PUFA to young post-pubertal dairy bulls altered the fatty acid composition of the sperm but had no effect on semen quality. In addition, we have shown that gradual weaning at 10-12 weeks of age does not induce a systemic immune response in dairy calves, however, health management practices should be tailored to specific breeds.

Development of Protocols on the use of Liquid Semen

We have completed a number of studies on the *in vitro* assessment of bull sperm stored at varying concentrations and at varying temperatures. The overall conclusion from this work was that sperm have better *in vitro* characteristics (in terms of motility, and oxidative stress) when stored at lower sperm concentrations and temperature changes between 5 and 22 °C do not have a detrimental effect on sperm. A large scale field trial was conducted and based on a combination of non-return rates (NRR's) and calving rates a lower sperm number resulted in a reduction in calving rates following 2 days of storage. The addition of antioxidants to semen diluents had no effect on sperm quality in liquid semen stored for up to 5 days, as assessed *in vitro*. While seminal plasma composition varied between high and low fertility bulls the addition of seminal plasma from high fertility bulls to caudal epididymal sperm had no beneficial effect on sperm physiology.

Sexed Semen

A field trial conducted in 2013 demonstrated that sexed semen resulted in significantly reduced pregnancy rates compared to a conventional control, irrespective of whether the sexed semen was insemination as fresh or frozen-thawed. A similar trend was observed in both dairy heifers and cows. The poor performance of liquid sexed semen was attributed to abnormal levels of head-to-head agglutination as assessed *in vitro*. Systems modelling of the use of sexed semen in expanding dairy herds has demonstrated that despite lower conception rates, the targeted use of sexed semen will result in faster herd expansion.

4. Impact of the Research

4(a) Summary of Research Outcomes

(i) Collaborative links developed during this research

This project has raised the international profile of the researchers, their groups, their institutions and of Ireland as leaders in the area of bull fertility. Indeed this cumulated in the researchers hosting an International Bull Fertility Conference, under the auspices of the British Society for Animal Science, in May 2018 in Westport (~250 delegates). It has aided in the development of new relationships with the leading cattle breeding companies in Ireland and Internationally (UK, France, Denmark, USA) and led to the development of collaborations with world renowned scientists from agri-food, basic biochemistry and mathematical backgrounds. Collectively these collaborations have facilitated the researchers to secure additional funding so as to progress this innovative work in conjunction with industry.

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

We have developed novel nutritional and health based strategies to facilitate the advancement of puberty in elite young dairy bulls so that when they reach sexual maturation they have the capacity to produce high volumes of excellent quality semen as early in life, as possible. We have also developed a clear protocol on the number of sperm per insemination dose for liquid semen as well as the optimum storage conditions in terms of diluents and temperature for semen to be stored in. Where relevant, these protocols have been implemented by both Irish and European cattle breeding companies. We completed the first ever field trial of sexed semen under Irish conditions and despite the lower conception rates than that of conventional non-sex sorted semen, the outcomes from our systems modelling work has illustrated that the targeted use of sexed semen will result in faster herd expansion.

(iii) Outcomes with economic potential

Collectively, this project has resulted in improvement early life management strategies of bulls as well as the better use of semen. This enables the industry to maximise the number of replacement heifers entering the dairy herd which are sired from young elite AI bulls. Advancing puberty and sexual maturation of elite young dairy sires will make more semen available from each bull earlier. This is especially important in seasonal grass based production systems whereby young genomically selected bulls could be heavily used in their first season at 12-15 months of age. Maximising the numbers of inseminations from each ejaculate can be achieved through liquid semen by putting a lower sperm number in each semen straw (3 million sperm if used within 1 day and 5 million if used within 2.5 days). The increased usage of sexed semen can facilitate more rapid herd expansion.

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

Through the increased production and better use of semen from young elite sires the results of this project will increase on-farm productivity and efficiencies. It will aid in maximising the kgs of milk solids per ha, thereby, reducing the environmental footprint of the dairy sector by having a lower number of more productive cows.

4 (b) Summary of Research Outputs

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

- Byrne, C.J., Fair, S., English, A.M., Cirot, M., Staub, C., Lonergan, P., Kenny D.A. (2018). Plane of nutrition before and after 6 months of age in Holstein-Friesian bulls: I. Effects on performance, body composition, age at puberty, and postpubertal semen production. *Journal of Dairy Science* 101, 3447-3459.
- Byrne, C.J., Fair, S., English, A.M., Urh, C., Sauerwein, H., Crowe, M.A., Lonergan, P., Kenny D.A. (2018). Plane of nutrition before and after 6 months of age in Holstein-Friesian bulls: II. Effects on metabolic and reproductive endocrinology and identification of physiological markers of puberty and sexual maturation. *Journal of Dairy Science* 101, 3460-3475.
- English, A.M., Byrne, C. J., Cormican, P., Waters, S.M., Fair, S. Kenny, D.A. (2018). Effect of Early Calf-Hood Nutrition on the Transcriptional Regulation of the Hypothalamic-Pituitary-Testicular axis in Holstein-Friesian Bull Calves. *Scientific Reports* (Accepted - In press).
- English, A.M., Kenny, D.A., Byrne, C.J., Sauerwein, H., Urh, C., Crowe, M.A., Staub, C., Waters, S.M., Fair, S. (2018). Role of early life nutrition on regulating the hypothalamic-anterior pituitary-testicular axis of the bull. *Reproduction* 156, 283-297.
- Byrne, C.J., Fair, S., English, A.M., Holden, S.A., Dick, J.R., Lonergan. P., Kenny, D.A. (2017). Dietary polyunsaturated fatty acid supplementation of young post-pubertal dairy bulls alters the fatty acid composition of seminal plasma and spermatozoa but has no effect on semen volume or sperm quality. *Theriogenology* 90, 289-300.
- Kenny, D.A., Byrne, C.J. (2018). The effect of nutrition on timing of pubertal onset and subsequent fertility in the bull. *Theory to Practice - International Bull Fertility Conference, Westport, Ireland, 27-30 May 2018. Animal.* 12(S1), S36-S44. doi:10.1017/S1751731118000514
- Fair S, Lonergan P (2018) Review: Understanding the causes of variation in reproductive wastage among bulls. *Animal.* doi: 10.1017/S1751731118000964.
- Byrne, C.J., Fair, S., English, A.M., Johnston, D., Lonergan, P., Kenny, D.A. (2017). Effect of milk replacer and concentrate intake on growth rate, feeding behaviour

and systemic metabolite concentrations of pre-weaned bull calves of two dairy breeds. *Animal* 11, 1531-1538.

- Byrne, C.J., Fair, S., English, A.M., Urh, C., Sauerwein, H., Crowe, M.A., Lonergan, P., Kenny, D.A., (2017). Effect of breed, plane of nutrition and age on growth, scrotal development, metabolite concentrations and on systemic gonadotropin and testosterone concentrations following a GnRH challenge in young dairy bulls. *Theriogenology* 96, 58-68.
- English, A.M., Waters, S.M., Cormican, P., Byrne, C.J., Fair, S., Kenny D.A. (2017). Effect of Early Calf-Hood Nutrition on the Transcriptomic Profile of Subcutaneous Adipose Tissue in Holstein-Friesian Bulls. *BMC Genomics* 19, 281.
- Holden, S.A., Fernandez-Fuertes, B., Murphy, C., Whelan, H., O'Gorman, A., Brennan, L., Butler, S.T., Lonergan, P., Fair, S. (2017). Relationship between in vitro sperm functional assessments, seminal plasma composition and field fertility following AI with either non-sorted or sex-sorted bull semen. *Theriogenology* 87, 221-228.
- Holden, S.A., Fernandez-Fuertes, B., Murphy, E.M., Lonergan, P., Fair, S. (2017). Effect of seminal plasma from high- and low-fertility bulls on cauda epididymal sperm function. *Reproduction Fertility and Development* 29, 2457-2465.
- Holden, S.A., Murphy, C., Moreno, J.F., Butler, S.T., Cromie, A.R., Lonergan, P., Fair, S. (2017). In vitro characterisation of fresh and frozen sex-sorted bull spermatozoa. *Reproduction Fertility and Development* 29, 1415-1425.
- Johnston D, Earley B, Cormican P, Kenny D.A., McCabe M., Kelly A.K., McGee, M., Waters, S.M. (2016). Characterisation of the whole blood mRNA transcriptome in Holstein-Friesian and Jersey calves in response to gradual weaning. *PLOS ONE* 11, p.e0159707.
- Johnston, D., Kenny, D.A., McGee, M., Waters, S.M., Kelly, A.K., Earley, B. (2016). Electronic feeding behavioural data as indicators of health status in dairy calves. *Irish Journal of Agriculture and Food Science* 55, 159-168.
- Murphy, C., Shalloo, L., Hutchinson, I.A., Butler, S.T. (2016). Expanding the dairy herd in pasture-based systems: The role of sexed semen within alternative breeding strategies. *Journal of Dairy Science* 99, 6680-6692.
- Johnston, D., Kenny, D.A., Kelly, A.K., McCabe, M.S., McGee, M., Waters, S.M., Earley, B. (2015). Characterisation of haematological profiles and whole blood relative gene expression levels in Holstein-Friesian and Jersey bull calves undergoing gradual weaning. *Animal* 10, 1547-56.
- Butler, S.T., Hutchinson, I.A., Cromie, A.R., Shalloo, L. (2014). Applications and cost benefits of sexed semen in pasture-based dairy production systems. *Animal* 8, 165-172.

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

- English A.M., Fair S., Byrne C.J., Waters S.M., Kenny D.A. (2017). Effect of early calf-hood nutrition on the hypothalamic-pituitary axis in Holstein-Friesian bulls. American Association of Animal Science, Baltimore, MD, 8th-12th July 2017
- English A.M., Fair S., Cormican P., Byrne C.J., Waters S.M., Kenny D.A. (2017). Effect of plane of nutrition on the transcriptomic profile of subcutaneous adipose tissue in Holstein-Friesian bull calves. 36th International Society of Animal Genetics Conference, Dublin, Ireland, July 16th-21st 2017.
- Fair, S. (2017). Maximising the use of elite genomically selected bulls through fresh semen. EU AI Vets Meeting, Aranjuez, Spain, 19th October 2017.
- Fair, S. (2017). Metabolome profiling and its relationship to sperm quality. European Society for Domestic Animal Reproduction. Bern, Switzerland, August 24th 2017
- Kenny, D. A., Heslin, J., Byrne, C. J. (2017) Early onset of puberty in cattle: implications for gamete quality and embryo survival. 44th Annual Conference of the IETS January 13-16, 2018 Shangri-La Hotel Bangkok, Bangkok, Thailand. *Reproduction, Fertility and Development*. 30(1):101-117.
- Kenny, D.A., Lonergan, P., Byrne, C.J., English, A.M., Fair, S. (2016). Nutrition and young bull semen production. EU AI Vets Annual Meeting, Salzburg, Austria, 27th - 28th October, 2016.
- English, A.M., Byrne, C., Cassidy, J., Fair, S., Kenny, D.A., (2016). The effect of early life plane of nutrition on testicular development of Holstein Friesian bull calves. British Society of Animal Science, Chester, UK, United Kingdom 6th April 2016.
- Fair, S., Lonergan, P., Butler, S.T., Murphy, C. (2016). Non-return rate as a predictor of calving rate in seasonal-calving pasture-based dairy herds. 10th Biennial Meeting of the Association for Applied Animal Andrology (AAAA), June 24-26, 2016; Tours, France.
- Holden, S.A., Lonergan, P. and Fair S. (2016). Effect of Seminal Plasma on the Rheotaxis Response of Caudal Epididymal Sperm. 18th International Congress on Animal Reproduction. June 26-30, 2016; Tours, France
- Holden, S.A., Lonergan, P. and Fair S. (2016). The effect of antioxidants on the in vitro quality of fresh bull sperm stored in an egg yolk based diluent. 10th Biennial Meeting of the Association for Applied Animal Andrology, June 24-26, 2016; Tours, France. Published in *Animal Reproduction Science* 169: 122.
- Murphy, C., Shalloo, L., and Butler, S.T. (2016). The influence of sexed semen use and genotyping on genetic gain in seasonal-calving pasture-based dairy herds. *Anim Reprod Sci* 169, 129. Association of Applied Animal Andrology, Jun 24 - 26 2016.

- Murphy, C., Shalloo, L., Hutchinson, I.A. and Butler, S.T. (2016). The potential role of sexed semen within expanding pasture-based dairy herds. The 18th International Congress of Animal Reproduction, Jun 26 - 30 2016.
- Byrne, C.J., English, A.M., Fair, S., Lonergan, P., Kenny, D.A. (2016). Effect of altering plane of nutrition during the first and second six months of life on age at puberty onset in Holstein Friesian bulls. British Society of Animal Science. Chester, UK, April 2016.
- Byrne, C.J., English, A.M., Fair, S., Lonergan, P., Kenny, D.A. (2016). Effect of plane of nutrition during the first and second six months of life on age at puberty and subsequent semen production in Holstein Friesian bulls. 18th International Congress on Animal Reproduction, Tours, France. 27th June 2016
- Byrne, C.J., English, A.M., Fair, S., Lonergan, P., Kenny, D.A. (2016). Effect of plane of nutrition during the first and second six months of life on age at puberty and blood metabolites in Holstein Friesian bulls. 67th Annual Meeting of European Association of Animal Production. 29th August 2016.
- Byrne, C.J., English, A.M., Fair, S., Lonergan, P., Kenny, D.A. (2016). Plane of nutrition affects scrotal skin thickness and scrotal temperature in pre-pubertal Holstein-Friesian bulls. Association for Applied Animal Andrology (AAAA), 10th Biennial Conference, Tours, France. 26th June 2016
- Byrne, C.J., English, A.M., Fair, S., Lonergan, P., Kenny, D.A. (2016). The effect of dietary omega-6 and omega-3 fatty acid supplementation on semen volume and quality in young post-pubertal dairy bulls. British Society of Animal Science. Chester, UK, April 2016.
- English A.M., Byrne C., Waters, S.M., Fair, S., Kenny, D.A., (2016). The effect of early life plane of nutrition on blood metabolites and testicular development of Holstein Friesian bull calves. Association of Applied Animal Andrology, Tours, France, 27th June 2016
- English A.M., Byrne C., Waters, S.M., Fair, S., Kenny, D.A., (2016). The effect of early calf-hood nutrition and age on the expression of genes associated with testicular development of Holstein-Friesian bulls. 18th International Congress of Animal Reproduction, Tours, France, 27th June 2016
- Holden, S. Fernandez-Fuertes, B., Lonergan, P., Fair, S. (2015). Effect of seminal plasma from high and low fertility bulls on epididymal sperm function. In Proceedings of European Domestic Animal Reproduction. Albena September 17th and 18th 2015
- Holden, S.A., Fernandez, B., Murphy, C., O'Gorman, A., Brennan, L., Butler, S.T., Lonergan, P., Fair, S. (2015). The use of in vitro assessments to predict bull fertility. Agricultural Research Forum. Tullamore. March 2015 9th and 10th p.94
- Johnston, D., Kenny, D.A., Cormican, P., McGee, M., Waters, S.M., Kelly, A.K., McCabe M., Earley, B. (2015). A comparison of the whole blood mRNA

transcriptome, between Holstein-Friesian and Jersey calves, preceding and succeeding gradual weaning. In: Proceedings of the Irish Agricultural Research Forum. Tullamore. March 9th and 10th 2015. p.149.

- Johnston, D., Kenny, D.A., Cormican, P., McGee, M., Waters, S.M., Kelly, A.K., McCabe M., Earley, B. (2015). Characterisation of the whole blood mRNA transcriptome in Holstein-Friesian and Jersey calves during gradual weaning. In: Proceedings of the annual meeting of Association for Veterinary Teaching and Research Work (Irish Branch). Teagasc Grange, 2015
- Byrne, C.J., English, A.M., Fair, S., Crowe, M.A., Lonergan, P., Kenny, D.A. (2015). Effect of breed, plane of nutrition and age on systemic gonadotrophin and testosterone concentrations, following a GnRH challenge in young dairy bulls. Agricultural Research Forum. Tullamore. March 9th and 10th 2015.
- Kenny, D.A., Lonergan, P., Byrne, C.J., English, A.M., Fair, S. (2015). Effect of nutrition on the onset of puberty and post pubertal fertility of bulls. EU AI Vets Meeting, Varna, Bulgaria, September 15th 2015
- Byrne, C.J., English, A.M., Fair, S., Lonergan, P., Kenny, D.A. (2015). The effect of dietary polyunsaturated fatty acid supplementation on semen volume and quality in young post-pubertal dairy bulls. Agricultural Research Forum. Tullamore. March 9th and 10th 2015.
- Byrne, C.J., English, A.M., Fair, S., Lonergan, P., Kenny, D.A. (2015). Effect of plane of nutrition on growth rate, feeding behaviour and systemic metabolite concentrations in pre-weaned bull calves of two contrasting dairy breeds. British Society of Animal Science. Chester, UK, April 2015.
- Holden, S.A., Murphy, C., Moreno, J.F., Cromie, A.R., Butler, S.T., Lonergan, P., Fair, S. (2014). The in vitro assessment of sex-sorted fresh and frozen bull sperm. British Society for Animal Science; Westport, Co Mayo; 19-21st May 2014
- Holden, S.A., Murphy, C., Moreno, J.F., Cromie, A.R., Butler, S.T., Lonergan, P., Fair, S. (2014). In vitro analysis of sex sorted fresh and frozen bovine semen. Agricultural Research Forum; Tullamore, Co Offaly; 10-11th March 2014
- Johnston, D., Kenny, D.A., Rubio Contreras, R., Waters, S.M., Kelly, A.K., McGee, M., Earley, B. (2014). The effect of gradual weaning on haematological profiles in artificially reared Holstein-Friesian and Jersey calves fed different planes of nutrition Agricultural Research Forum; Tullamore, Co Offaly; 10-11th March 2014
- Johnston, D., Kenny, D.A., Rubio Contreras, R., Waters, S.M., Kelly, A.K., McGee, M., Earley, B. (2014). The effect of gradual weaning on the relative expression levels of selected genes in the leukocyte environment of artificially reared Holstein - Friesian and Jersey calves. Agricultural Research Forum; Tullamore, Co Offaly; 10-11th March 2014
- Johnston, D., Kenny, D.A., Rubio Contreras, R., Waters, S.M., Kelly, A.K., McGee, M., Earley, B. (2014). The effect of gradual weaning on leukocyte relative gene

expression levels in dairy calves. Proceedings of the EAAP Annual Meeting 2014 Copenhagen, Denmark, 25-29th August 2014

- Murphy, C., Holden, S.A, Lonergan, P., Fair, S. (2014). Optimising the storage temperature and sperm concentration of liquid stored bull sperm. British Society for Animal Science; Westport, Co Mayo; 19-21st May 2014
- Murphy, C., Holden, S.A, Lonergan, P., Fair, S. (2014). The effect of fluctuating storage temperature on liquid stored bull sperm. Association for Applied Animal Andrology; Newcastle Australia; 8-10th September 2014
- Murphy, C., Holden, S.A., Fair, S. (2014). The effect of storage temperature and sperm concentration on liquid stored bull sperm. Agricultural Research Forum; Tullamore, Co Offaly; 10-11th March 2014

(iii) National Report

- Murphy, C., Maicas, C., Holden, S.A., Shalloo, L., Butler, S.T. (2017). Sexed semen: Grow your dairy herd and increase beef output at the same time. Moorepark '17 Open Day. Moorepark '15 Open Day, attended by 10,000 farmers
- Kenny, D.A., Byrne, C.J., English, A.M., Fair, S. (2016). Bull fertility: Important issues and current research. Teagasc Grange Open Day Booklet. 188-191. ISBN: 978-1-84170-627-6.
- Murphy, C., Shalloo, L., Butler, S.T. (2015). Potential benefit of sexed semen in Ireland. Moorepark '15 Open Day, attended by 10,000 farmers
- Butler, S.T., Hutchinson, I.A. (2014). Is there a role for sexed semen in seasonal-calving systems? Proceedings of the 2014 Positive Farmers Conference, Clonmel, Ireland. 15th January 2014
- Butler, S.T., Hutchinson, I.A., Cromie, A.R. (2013). The future role of sexed semen in Irish dairy farming. Teagasc National Dairy Conference. 12th November 2013

(iv) Workshops/seminars at which results were presented

- Kenny, D.A., Keogh, K., Byrne, C.J. (2018). Impact of early life nutrition on the molecular and physiological regulation of puberty onset in the bull. American Dairy Science Association, Annual meeting, Knoxville, Tennessee. 24-27th June 2018. The Professional Animal Scientist. (Accepted - In press).
- Kenny DA. (2016). The early life nutritional physiology of the male calf and effects on age at onset of puberty and beef carcass production. The 1st International Symposium on Young Ruminants. Beijing, China, 28th to 31st October, 2016.
- Kenny, D.A., Fair, S. (2015). Invited Seminar on a review of Bull Nutrition and Fertility. Cogent AI Centre, Chester, UK. November 5th, 2015

(v) Intellectual Property applications/licences/patents

(vi) Other

5. Scientists trained by Project

Total Number of PhD theses: 3

1. Shauna Holden. Optimising the use of fresh semen: Insights for use with young genomically-selected bulls. University of Limerick. Graduated August 2017
2. Colin Byrne. An Examination of the Effects of Nutrition on Age at Puberty and Subsequent Fertility in Dairy-Bred Bulls. PhD thesis submitted to the National University of Ireland, 2017. Graduated June 2017.
3. Anne-Marie English. The Molecular Control of the Hypothalamic-Pituitary-Testicular Axis in the Bull. PhD thesis submitted to the University of Limerick, 2017. Graduated August 2018.

Total Number of Masters theses: 0

6. Permanent Researchers

Institution Name	Number of Permanent staff contributing to project	Total Time contribution (person years)
UL	1	0.8
Teagasc Grange	4	1.08
Teagasc Moorepark	2	2.68
UCD	1	0.2
Total	8	4.76

7. Researchers Funded by DAFM

Type of Researcher	Number	Total Time contribution (person years)
Post Doctorates/Contract Researchers	1	2
PhD students	3*	8.05
Masters students	0	0
Temporary researchers	0	0
Other		
Total	4	10.05

*7 PhD students inputted into the project. Of these, 3 students had their stipends funded on this project.

8. Involvement in Agri Food Graduate Development Programme

NA

9. Project Expenditure

Total expenditure of the project: €666,418

Total Award by DAFM: €672,598

Other sources of funding including benefit in kind and/or cash contribution(specify): €0

Breakdown of Total Expenditure

Category	UL Institution 1	Teagasc Grange Institution 2	Teagasc Moorepark Institution 3	UCD Institution 4	Total
Contract staff					
Temporary staff					
Post doctorates			89936		89936
Post graduates	85962			86647	172609
Consumables	50263	51269		22036	123568
Travel and subsistence	15199	3343	3213	4403	26158
Sub total	151424	54612	93148	113085	412270
Durable equipment	52517	2142	767		55427
Other	68073	7410			75483
Overheads	44985	16384	27945	33925	123239
Total	317000	80548	121860	147011	666418

10. Leveraging

The following are research grants secured from various funding agencies on bull fertility.

Funding secured from Science Foundation Ireland (SFI)

- SFI Investigators Programme (2017; €1.94 million). Title: An integrated multidisciplinary approach to revolutionise dairy cattle breeding, through the application of state-of-the-art technology to advance the identification, sexual maturation, fertility and availability of semen from genetically elite sires

Funding secured from the Department of Agriculture Food and the Marine (DAFM)

- Research Stimulus Fund (2015; €1.1 million). Title: New strategies to predict and monitor semen fertility

Funding secured from Enterprise Ireland (EI)

- Commercialisation Fund (2014; €233,000). Title: Development of a Sperm Filtration Device

Funding secured from the Irish Research Council (IRC)

- Enterprise Partnership PhD Scheme. Student started in January 2018
- Postgraduate Scholarship Scheme PhD. Student started in October 2017
- Employment based PhD Scheme. Student started in February 2014.
- New Foundations Scheme. Three small projects funded over 3 years.
- Postgraduate Scholarship Scheme PhD. Student started in September 2013

11. Future Strategies

The results from this DAFM funded project have laid the foundation for the researchers to secure additional competitive funding and building critical mass in the area of bull fertility research in Ireland. The researchers have secured follow on grants from SFI, EI, DAFM and the IRC to work on both basic and applied themes. All of these grants have significant input from the Irish cattle breeding companies. The researchers have broadened their collaborations and are now working with a multi-disciplinary team of world renowned scientists from agri-food, basic biochemistry and mathematical backgrounds together with key industry stakeholders in order to address bull fertility problems limiting the potential growth of Ireland's agri-food industry. Indeed, these collaborations have led to a recent H2020 funding application on the epigenetic regulation of bull fertility.

Taken together, the newly funded projects will characterise DNA and other biochemical markers of early puberty as well as unravelling the metabolic control of this complex trait so that it can be consistently advanced, through strategic early life dietary management. In addition, through the application of state-of-the-art genetic, physiological, immunological and molecular approaches, combined with *in vitro* sperm functional and bioinformatics analyses we aim to develop biomarkers to predict and monitor semen fertility. In parallel to this, the use of both liquid and sex sorted semen in the dairy sector is being further developed and trialled under Irish conditions. This novel, multidisciplinary and industry focused work will revolutionise cattle breeding in Ireland through augmenting the current genomic selection based cattle breeding programme and will further enhance the country's reputation as a world leader in the application of genomic technology to livestock breeding.