Section A

Institution: University of St Andrews

Unit of Assessment: UoA 05: Biological Sciences

Title of case study: Successful aquaculture genetics business underpinned by research on the physiology and genetics of fish muscle

Period when the underpinning research was undertaken: 2000 - 2016

Details of staff conducting the underpinning research from the submitting unit:

Name(s):	Role(s) (e.g. job title):	Period(s) employed by submitting HEI:
Ian A. Johnston	Professor	01 January 1976 - 31 December 2019
Daniel Macqueen	Research Fellow	01 January 2008 - 31 December 2012

Period when the claimed impact occurred: 01 August 2013 – 31 December 2020

Is this case study continued from a case study submitted in 2014? No

Section B

1. Summary of the impact

Future growth in seafood consumption is predicted to come from aquaculture, and the application of genetics is essential for meeting this demand. Research from St Andrews on the physiology and genetics of fish muscle (2000 – 2012) included the discovery of selection markers for increasing fillet yield and superior texture and underpinned the success of a spin-out company which has grown since 2013 into a mature multi-species aquaculture genetics business trading globally (www.xelect-genetics.com). Xelect methods enable aquaculture companies to select for and test for fillets of higher value. From August 2016 to December 2020, Xelect has provided breeding programme management and analytic services with broad reach to 14 countries involving 16 species on behalf of more than 25 companies, including a top six global salmon producer. By improving hatchery management and protecting endemic wild fish stocks from introduced fish, Xelect contributes to improved sustainability and growth of the global aquaculture sector, whilst creating highly skilled employment for a team of 15 people (headcount: 15; FTEs: 15; 11 with PhDs) in the UK.

2. Underpinning research

The Johnston research group has explored the structure, function and evolution of fish muscle. Research on Atlantic salmon revealed that the recruitment of muscle fibres continues for more than a year after they transfer from freshwater to seawater, with the final fibre number varying by more than three-fold between individuals (R1). Furthermore, final fibre number varied by up to 20% depending upon the water temperature during a critical window of development between the end of somite formation and the eyed stage of the embryo (R1). The number and density of muscle fibres at the time of harvest in salmon had a significant effect on growth rate, texture, colour and the propensity for post-mortem gaping of the fillet (R2) (an important source of economic loss during processing) and to have moderate to high levels of heritability. The influence of growth rate, smolt duration and environment on the growth and texture of Atlantic salmon muscle was also determined and a novel method for measuring the tensile strength of fillets was developed (R3) which provides a better predictor of gaping than previously described instrumental texture measurements. Other studies demonstrated that muscle cellularity was subject to rapid evolution, resulting in large reductions in fibre number and increases in fibre size in dwarf landlocked arctic charr and sticklebacks relative to ancestral populations, indicating the adaptability of these economically important traits.

A series of papers investigated gene expression and transcriptomics in fish muscle in relation to circadian rhythms, fasting and refeeding, diet, acclimation temperature and selection for body size at age (e.g. R4). This work particularly focused on pathways involved in growth regulation and protein breakdown. Specific genes and pathways that were upregulated or downregulated following the cessation of muscle fibre recruitment were identified in a model fish species (R5). The salmonid lineage underwent a whole genome duplication (WGD), with 50% of duplicated genes being retained in extant species. Phylogenetic analysis of an extensive set of gene paralogues coupled with molecular clock analysis provided an accurate estimate for the timing of the whole genome duplication event in relation to subsequent speciation of the lineage (R6). Other studies identified examples of sub-functionalisation of the expression of paralogous genes involved myogenesis and growth that had been retained from the teleost- and lineage-specific WGDs, including the MyoD family of master transcription factors. Building on this body of work, genetic markers for increased fillet yield and firmness were discovered using a candidate gene approach and Genome Wide Association Analysis. This research led to the following patents:
1: European Patent (EP2766494B1) which can select up to 3.6% higher fillet yield in Atlantic Salmon than unselected fish.
2: World patent WO2013/054107 A1 1117448.9 2011.10.10 GB Accepted Journal date 2017.08.03.
3. Australian patent 521300

3. References to the research

The underpinning research has involved fundamental scientific studies producing 87 publications in peer-reviewed scientific journals, a relevant selection of which are presented here. R4 had been submitted to REF2014.

R1. Johnston, I.A., Manthri, S., Alderson, R., Smart, A., Campbell, P., Nickell, D., Robertson, B., Paxton, C.G.M. and Burt, M.L. (2003). **Freshwater environment affects growth rate and muscle fibre recruitment in seawater stages of Atlantic salmon** (*Salmo salar*). J. Exp. Biol. 206, 1337-1351. DOI:10.1242/jeb.00262.
R2. Johnston, I.A., Alderson, D., Sandham, C., Dingwall, A., Mitchell, D., Selkirk, C., Nickell, B., Baker, R., Robertson, B., Whyte, E, A. and Springate, J (2000). **Muscle fibre density in relation to the colour and texture of smoked Atlantic salmon** (*Salmo salar* L.). Aquaculture 189, 335-349. DOI: 10.1016/S0044-8486(00)00373-2.
R3. Ashton, T.J., Michie, I., and **Johnston, I.A**. (2010) **A Novel Tensile Test Method to Assess Texture and Gaping in Salmon Fillets**. Journal of Food Science. 75(4), 182-190. DOI: 10.1111/j.1750-3841.2010.01586.x.
R4. Macqueen, D. J., M. L. Delbridge, Manthri, S. and **Johnston, I.A**. (2010). **A newly classified vertebrate calpain protease, directly ancestral to CAPN1 and 2, episodically evolved a restricted physiological function in placental mammals.** Molecular Biology & Evolution 27(1), 1886-1902. DOI: 10.1093/molbev/msq071.
R5. Johnston, I.A., Lee, H-T., **Macqueen, D.J.**, Paranthaman, K., Kawashima, C., Anwar, A., Kinghorn, J.R. and Dalmay, T. (2009). **Embryonic temperature affects muscle fibre recruitment in adult zebrafish: genome-wide changes in gene and microRNA expression associated with the transition from hyperplastic to hypertrophic growth phenotypes**. J. Exp. Biol. 212, 1781-1793. DOI: 10.1242/jeb.029918.

R6. Macqueen, D.J. and **Johnston, I. A.** (2014). **A well-constrained estimate for the timing of the salmonid whole genome duplication reveals major decoupling from species diversification**. Proc. Roy. Soc. B. 281: 20132881. DOI: 10.1098/rspb.2013.2881.

4. Details of the impact

An increase in the demand for fish coupled with over-exploitation of wild fish stocks has led to the strong growth of aquaculture globally (>50% of harvested fish already comes from aquaculture - FAO 2016; http://www.fao.org/3/a-i5555e.pdf). The application of genetic selection is essential for a modern profitable industry because of the need to avoid inbreeding (which results in a loss of fecundity and high incidence of deformities) and to drive generational gains in growth, disease resistance, feed efficiency, survival and flesh quality. Our basic research on the biology of fish muscle (described in section 2) found application through a series of collaborations with the salmon farming industry supported by UK Research Councils Link and Industrial Partnership grants (2000-2012). The research conducted and commercial contacts made led to the formation of an aquaculture genetics company called Xelect (2012-13). Xelect's early activities centred around licensing genetic marker IP [patents 1 and 2 listed at the end of section 2 for increased fillet field and trade secrets for muscle number (2015)] that have been used to select Atlantic salmon with superior fillet yield and flesh quality for Chile (MultiExport Foods) and Canada (Cermaq).

Xelect management of breeding programs reduces costs and increases income: In 2016, the business expanded into providing complete management support for breeding programmes, and this now accounts for 80% of revenues. The company makes genetics affordable to even small and medium sized producers without them having to invest in the large interdisciplinary team needed to apply advanced breeding technologies. Xelect currently manages and supports breeding programmes in 14 countries involving 16 species on behalf of more than 25 companies, including a top six global salmon producer. Xelect's services include the development of genetic tools, licensing IP, genotyping services, quantitative genetics calculations and practical hatchery support. The company supports pedigree-based combined family selection, marker assisted selection and genomic selection. The return on investment in genetics to producers is recognised to be very significant. "*Using Xelect's markers, companies can add GBP600 to the value of a tonne of farmed salmon*" (S1 page 1 column 3), and the net benefit for a single small breeding programme for sea bream was estimated at EUR2,900,000 over 10 years (S2 page 1 Abstract). Xelect has become an established brand within the aquaculture industry as evidenced by extensive coverage in the trade press (S3-S6).

Xelect techniques improve quality selection for farmed fish: Xelect has developed techniques to measure soft flesh and gaping (R3), leading to quality control processes that have been adopted by Mowi (world's largest salmon producer), Dawnfresh (UK's largest sea trout producer), Young's Seafood (UK) and Pacific Seafoods (USA) (major processor and integrated producer respectively) for their routine quality assessments during processing. This has enabled companies to assess the impact of production practices, e.g. growth, harvest time, and feeding regimes on flesh quality losses, supplementing gains made through genetic selection (S7).

Contributing to the Economy of the UK and global aquaculture: By July 2020 Xelect employed a team of 15 people (headcount: 15; FTEs: 15; 11 with PhDs) including aquaculture specialists, quantitative geneticists, molecular biologists, bioinformaticians and computer scientists. The company had sales revenues approximately GBP1,200,000 per year by year-end 2020 (approximately 90% exports) and was profitable with no borrowings. Xelect has successfully adapted its operations in response to Covid-19 resulting in new ways of working. Five new strategic partnership contracts have been signed in 2020 with companies operating in Greece, Kenya, Portugal, Uganda, Vietnam, and Zambia. Since genetic gain is only achieved in each generation, which is 4 years for salmon, the first quantitative validation of simulated genetic gains will only be available at the end of 2021.

Improving hatchery management: Xelect developed software between 2017 and 2020 that contributes to better hatchery management across the aquaculture industry. The software considers Estimated Breeding Values (EBVs), relatedness and non-genetic factors such as biomass, weight, sex, and photoperiod regime to optimally configure each spawning tank in the hatchery, resulting in increases in growth of 8-15% in offspring whilst maintaining strict inbreeding control. The software is in routine use across hatcheries in Croatia, Greece, Mexico, and Australia. The director of the Group Hatcheries sector for Selonda Aquaculture noted "*Xelect has helped us make significant progress in improving the genetic diversity of our broodstock and solving the complex organisational challenges of breeding group-spawning species ... to produce faster-growing and robust production fish with improved feed efficiency*" (S8). The freshwater manager for Loch Duart Ltd noted "*It is through the use of their expert services that Loch Duart have been able to transform our broodstock program to help benefit the stock*" (S9).

Protecting endemic wild fish stocks: Some salmonid species are farmed as all-female triploids which are sterile such that escapees cannot interbreed with wild stocks and reduce their fitness. Efforts to produce sterile triploid eggs by commercial breeders are rarely 100% effective. The Xelect next day triploidy testing service based on flow cytometry enables producers of sterile trout, artic charr, and salmon eggs to ship their product to customers with an independent certificate of the average ploidy. Xelect is the only company in Europe offering triploidy testing and has 100% of the market, carrying out more than 7,000 determinations each year for breeding companies in Denmark, Finland, Norway, Poland, Slovenia and the UK. In addition, we routinely test batches of eggs supplied to Dawnfresh the UK's largest trout producer to ensure they are 100% triploid and all-female (using a molecular sex determination method).

5. Sources to corroborate the impact

S1. Xelect was used as an impact case study by BBSRC - http://www.bbsrc.ac.uk/documents/1511-xelect-pdf/

S2. Janssen, K., Saatkamp, H., and Komen, H. (2018). Cost-benefit analysis of aquaculture breeding programs. Genetics Selection Evolution 50(2) https://dx.doi.org/10.1186%2Fs12711-018-0372-3

S3. Fish Farmer Magazine article (15 Feb 2020) https://www.fishfarmermagazine.com/news/rapid-response-to-get-the-right-genes/

S4. Fish Focus Aquaculture article (2019) https://fishfocus.co.uk/xelect-express/

S5. Seafood Source article (9 April 2020) https://www.seafoodsource.com/news/aquaculture/how-genetics-is-leveling-the-aquaculture-playing-field

S6. The Fish Site article (3 August 2020) https://thefishsite.com/articles/a-novel-way-of-assessing-broodstock-value

S7. Proprietary software – OptiMate facilitates the selective breeding of group-spawning species by solving the complex problems of batch re-organisation. https://xelect.co.uk/breeding-programme-management/ (Particularly Item 3)

S8. Letter from the Director of Group Hatcheries sector, Selonda Aquaculture, Greece

S9. Letter from Freshwater Manager, Loch Duart Ltd, Scotland