

## **Cullen Scholarship: Epidemiological examination of Salmonid Rickettsial Septicaemia (SRS) in Irish Atlantic salmon farms (PhD Award).**

### **Background**

Salmonid rickettsial septicaemia (SRS) is currently described as one of the major health challenges in Irish salmonid aquaculture. SRS was first described in Chile in the late 1980's. Disease outbreaks were later reported in the UK, Ireland, Norway and Canada. *Piscirickettsia salmonis*, a gram-negative, coccoid-like, intracellular bacteria, is the aetiological agent of SRS or piscirickettsiosis. SRS is a severe disease that has been responsible for major economic losses in the aquaculture industry since its appearance in 1989. Historically, the incidences and impact of *P. salmonis* in Ireland were very low and required minimal intervention (antibiotics). The frequency of occurrence in Ireland over the last number of years has increased significantly and *P. salmonis* is now emerging as one of the main infectious disease challenges in marine salmon farming, with the yearly number of cases requiring antibiotic intervention on the rise. The disease is now one of the most significant drivers of antibiotic use in salmon farming in Ireland, accounting for 80% of antibiotics used in 2022.

The development of effective control strategies for the disease has been limited due to a lack of knowledge about the biology, intracellular growth, transmission, virulence and cultivation of the organism. SRS has become increasingly insidious and difficult to control. Failure of antibiotic treatment is common, and currently used vaccines show variable long-term efficacy. Currently, there is no vaccine licenced for use in Ireland. A detailed investigation into the distribution, seasonality, and virulence of this bacteria in an Irish context is needed to better understand the disease. Knowledge acquisition in this area is essential for the development of potential mitigation measures for the disease, including the development of autogenous vaccines.

The proposed programme of research will generate detailed epidemiological data for *P. salmonis* in the Irish context, in addition to information on the efficacy of current biosecurity and farm management practices. Development of fast, reliable and sensitive molecular diagnostic tools are necessary for early detection of the pathogen and to enable early intervention and mitigation of disease outbreaks. Relative and absolute quantification PCR methods will be also be developed, optimized and validated as part of this research. As there is currently no vaccine available for use in Ireland, there is an urgent need for increased investigation and development in this area.

### **Proposal**

We propose a **structured four-year PhD** on a full-time basis to undertake studies into the investigation of *Piscirickettsia salmonis* prevalence, epidemiology and control in farmed salmonids in Ireland through all stages of production. The project will investigate a range of parameters in order to increase our understanding on the prevalence and transmission of *P. salmonis*, virulence and origin. The project

should support the development of a mitigation strategy for this disease through improved diagnostics and vaccine development. The project will aim to:

- Conduct longitudinal studies to determine prevalence and seasonality of the bacteria in farmed Atlantic salmon.
- Investigate potential routes of transmission through examination of suspect vector species.
- Establish enrichment solid agar and cell culture cultivation methods for the bacteria based on published literature.
- Develop rapid, sensitive, quantifiable and reliable molecular diagnostic tests for *P. salmonis* using real-time PCR and digital droplet PCR.
- Perform phylogenetic and genotypic assessment of all isolates to determine origin and strain variance.
- Establish an autogenous vaccine to be used in Irish aquaculture.

### **Location of Scholar**

The scholar will be based for circa 50% of their four years at the Marine Institute HQ Rinville, Galway.

### **Outcome**

The outcome of this project will be to increase our knowledge of SRS and to develop technologies to facilitate mitigation for Irish aquaculture. The development of improved diagnostics (agar culture, cell culture and quantitative PCR) is important for the Fish Health Unit in their role as the National Reference Laboratory for fish diseases. The development of an autogenous vaccine is an essential output from this project as a key component to form part of on-farm management strategies.

### **Links with the Marine Institute Strategic Plan 2023-2027**

This proposal falls under Strategic Priority “Delivering impact through research and innovation” with strong links to Strategic Priority “Transforming our knowledge, advice and services”.

### **Specific Requirements**

The Fellow should have a primary degree/post-graduate qualification in biological sciences with a strong molecular biology component. The host institute should have a proven capability in vaccine development and disease epidemiology.

### **Financial Details**

Scholarships will be up to €27,500 per annum (maximum funding of €110,000 over four years). This amount comprises a maintenance award of €18,500 (Irish Research Council rate effective 1-Jan-21\*) to the student as well as payment of fees to the host higher education institution (HEI). The maximum fees payable to the HEI will be €6,000 per annum. The scholarship award also includes a budget of up to €3,000 per annum for eligible research costs (travel & subsistence, publication costs, consumables and other costs e.g. laptop) for the sole use of the student, and are payable on a reimbursement basis direct to the host institution where the postgraduate student (scholar) is registered. There are no overheads payable on the scholarship. Publication costs are intended to cover publications on which the scholar is listed as first author and are published under Open Access. Scholars can also supplement this funding by applying to the Marine Institute's Networking & Marine Research Communication Awards annual call.

\*There is a national review underway of the PhD annual student stipend payment, which is expected to result in a rate increase, and the Marine Institute will adjust the total funding accordingly following the completion of this review.

### **Marine Institute Co-Supervisor**

Dr. Samantha White, Fish Health Unit, Marine Environment & Food Safety Services (MEFS)

[Samantha.White@marine.ie](mailto:Samantha.White@marine.ie)

### **References**

Aravena, P. *et al.* (2020). PCR-RFLP Detection and Genogroup Identification of *Piscirickettsia salmonis* in Field Samples. *Pathogens*. **9**: 358. doi:10.3390/pathogens9050358

Jones, S. R. M. (2019). Characterization of *Piscirickettsia salmonis* and salmonid rickettsial septicaemia to inform pathogen transfer risk assessments in British Columbia. *DFO Can. Sci. Advis. Sec. Res. Doc.* 2019/020. v + 21 p.

Karatas, S. *et al.* (2008). Real time PCR detection of *Piscirickettsia salmonis* from formalin-fixed paraffin-embedded tissues. *Journal of Fish Diseases*. **31**: 747-753. doi:10.1111/j.1365-2761.2008.00948.x

Levipan, H. A. *et al.* (2022). Collective behaviour and virulence arsenal of the fish pathogen *Piscirickettsia salmonis* in the biofilm realm. *Frontiers in Cellular and Infection Microbiology*. **12**: 1067514. doi: 10.3389/fcimb.2022.1067514

Makrinos, D. L. and Bowden, T. J. (2017). Growth characteristics of the intracellular pathogen *Piscirickettsia salmonis*, in tissue culture and cell-free media. *Journal of Fish Diseases*. **40**: 1115-1127. doi:10.1111/jfd.12578

- Marshall, S. *et al.* (1998). Minimally Invasive Detection of *Piscirickettsia salmonis* in Cultivated Salmonids via the PCR. *Applied and Environmental Microbiology*. **64**(8): 3066-3069.
- Marcos-Lopez, M. *et al.* (2017). *Piscirickettsia salmonis* infection in cultured lumpfish (*Cyclopterus lumpus* L.). *Journal of Fish Diseases*. **40**:1625-1634. Doi:10.1111/jfd.12630
- Olivares, J. and Marshall, S. H. (2010). Determination of minimal concentration of *Piscirickettsia salmonis* in water columns to establish a fallowing period in salmon farms. *Journal of Fish Diseases*. **33**: 261-266. doi:10.1111/j.1365-2761.2009.01119.x
- Otterlei, A. *et al.* (2016). Phenotypic and genetic characterization of *Piscirickettsia salmonis* from Chilean and Canadian salmonids. *BMC Veterinary Research*. **12**: 55. doi: 10.1186/s12917-016-0681-0
- Palmer, R. *et al.* (1996). A *Piscirickettsiosis*-like disease in farmed Atlantic salmon in Ireland – isolation of the agent. *Bull. Eur. Ass. Fish Pathol.* **17**(2): 68.
- Reid, H. I. *et al.* (2004). Isolates of *Piscirickettsia salmonis* from Scotland and Ireland Show Evidence of Clonal Diversity. *Applied and Environmental Microbiology*. **70**(7): 4393-4397. doi: 10.1128/AEM.70.7.4393–4397.2004
- Rozas, M. and Enriquez, R. (2014). *Piscirickettsiosis* and *Piscirickettsia salmonis* in fish: a Review. *Journal of Fish Diseases*. **37**:163-188. doi:10.1111/jfd.12211
- Saavedra, J. *et al.* (2017). Prevalence, geographic distribution and phenotypic differences of *Piscirickettsia salmonis* EM-90-like isolates. *Journal of Fish Diseases*. **40**: 1055-1063. doi:10.1111/j.1365-2761.2009.01119.x
- Vargas, D. *et al.* (2021). The Analysis of Live-Attenuated *Piscirickettsia salmonis* Vaccine Reveals the Short-Term Upregulation of Innate and Adaptive Immune Genes in Atlantic Salmon (*Salmo salar*): An In Situ Open-Sea Cages Study. *Microorganisms*. **9**: 703. doi:10.3390/microorganisms9040703
- Yanez, A. J. *et al.* (2012). Broth medium for the successful culture of the fish pathogen *Piscirickettsia salmonis*. *Diseases of Aquatic Organisms*. **97**:197-205. doi: 10.3354/dao02403
- Yanez, A. J. *et al.* (2013). Two novel blood-free solid media for the culture of the salmonid pathogen *Piscirickettsia salmonis*. *Journal of Fish Diseases*. **36**:587-591 doi:10.1111/jfd.12034