

Improving early detection surveillance and emergency disease response to Ostreid herpesvirus using a hydrodynamic dispersion model:

Updating disease management areas for the South Australian oyster industry

Shane Roberts, Charles James, Matthew Bansemer, Frank Colberg, Saima Aijaz, Kaine Jakaitis, Eric Schulz and John Middleton

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Research	er Contact Details	FRDC Co	FRDC Contact Details			
Name:	Dr Shane Roberts	Address:	25 Geils Court			
Address:	Level 14, 25 Grenfell Street, Adelaide		Deakin ACT 2600			
		Phone:	02 6285 0400			
Phone:	(08) 8429 0505	Fax:	02 6285 0499			
Email:	shane.roberts@sa.gov.au	Email: Web:	frdc@frdc.com.au www.frdc.com.au			

In submitting this report, the researcher has agreed to FRDC publishing this material in its edited form.

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Abbreviations

CSIRO DMA DNA	Commonwealth Scientific and Industrial Research Organisation Disease Management Area Deoxyribonucleic acid
ISA	Infectious salmon anaemia
FRDC	Fisheries Research and Development Corporation
NSW	New South Wales
OsHV-1	Ostreid Herpesvirus 1 microvariant
PIRSA	Primary Industries and Regions South Australia
POMS	Pacific Oyster Mortality Syndrome
ROMS	Regional Ocean Modelling System
SA	South Australia
SARDI	South Australian Research and Development Institute
SAIMOS	Southern Australian Integrated Marine Observing System
SAOGA	South Australian Oyster Growers Association
SAROM	South Australian Regional Ocean Model
SASQAP	South Australian Shellfish Quality Assurance Program
TGM	Two Gulf Model

Executive Summary

The commercial oyster growing industry in South Australia is worth up to \$40 million / year and includes over 336 licenced aquaculture sites covering approximately 959 hectares across the State. Pacific Oyster Mortality Syndrome (POMS) is a considerable threat to the industry, and is currently present in the feral oyster population in Port Adelaide, which is located ~60 km from the closest commercial oyster growing region. Regular State-wide disease surveillance has not detected the virus in farming regions to date.

Rapid predictive capability of viral spread through water during an aquatic disease outbreak is an epidemiologist's dream, and up until now has not been achievable. A biophysical particle tracking model for Ostreid herpesvirus 1 microvariant (OsHV-1) that causes POMS was developed to determine virus spread during disease outbreaks in South Australian coastal waters. Model outputs from 23 hypothetical outbreaks across the State have provided valuable information for PIRSA to review and update current Disease Management Areas (DMAs) for POMS. Outputs from this project will greatly enhance future disease surveillance programs and emergency responses.

Prior to this project, disease management (prevention, preparedness and response) for POMS included 11 distinct DMAs (i.e. biosecurity zones) across the State, which is based on an assumed viral dispersal distance of 5 NM (<10 km) between Pacific oyster populations. This project aimed to provide more accurate estimates of viral dispersal distances in each growing region and to validate or update current DMAs.

The biophysical model developed for POMS during this project is underpinned by a hydrodynamic particle tracking model, which was developed in a previous FRDC research project (FRDC 2016/005: Middleton et al., 2017). The biophysical model couples oceanographic parameters (e.g. currents, tides, wind and water temperature) with known biological information (including host population location, viral production, temperature dependent virus survival time in water, viral DNA detection time in water, particle infectivity, trajectory limits) to predict viral dispersal. Assumptions and limitations of the model are discussed in the report.

Particle tracking time in the model represents survival of the virus (up to 2 days) and the DNA (up to 22 days), noting that average maximum distances travelled are reported for 20 days (adequate data replication). Modelled data were analysed to determine convex hulls (polygons) at 2, 4, 7, 14 and 22 days to provide information on particle connectivity between sites and regions. Data are presented as maps, regression analyses and summary tables. Particle dispersal distance varied between sites, regions and seasons. The maximum distance live virus (2 day lifespan) travelled from a point source varied from 5.2 km (90th percentile = 1.9 km) in Proper Bay during Summer to 44.1 km (90th percentile = 36.1 km) in Stansbury (Yorke Peninsula) during Summer. The maximum distance viral DNA can travel by 20 days (Season dependent) varied from 7.8 km (90th percentile = 7.5 km) from Thevenard wharf (Denial Bay) during spring to 310 km (90th percentile = 297.7 km) from Coffin Bay during summer. Across all sites and seasons, the average maximum dispersal distance after 2 days was 20.2 km, while the 90th percentile of particles reached 11.7 km.

These dispersal distances for each site were used to determine and update the DMAs for oyster growing regions in South Australia. Boundaries of all DMAs were updated to reflect a more accurate estimate of viral dispersal distance based on unique oceanography at each site. Two oyster growing regions which were previously considered separate DMAs (Denial Bay and Smoky Bay biosecurity zones) were merged due to overlapping particle dispersal polygons demonstrating connectivity between sites (moderate to high risks of disease spread through water). Future disease responses in this area will consider both bays as one DMA. This could also reduce sampling effort in this region for future surveillance, as they are now considered epidemiologically linked. Furthermore, understanding the prevailing hydrodynamics influencing the trajectory and dispersal distance of virus plumes will inform sampling locations within regions since early detection surveillance aims to bias sampling towards high-risk areas.

The biophysical model is now developed for each oyster growing region in South Australia and can be used in real time to provide predictive capability up to 3 days for future emergency responses to POMS. The model is flexible to track other passive particles (other pathogens, harmful algae blooms, chemicals, toxins or oil spills) or motile particles (e.g. parasites or larvae) given appropriate biological inputs and assumptions. The modelled data are presented in this report in such a way to allow dispersal distances to be determined for any passive particle up to 22 days.

This project demonstrated an effective collaboration between different fields of science (oceanography, epidemiology and virology) to achieve the outcomes, which have substantially improved future early detection surveillance, and emergency disease preparedness. The model is now being used by PIRSA and the oyster growing industry for these activities. In addition, this model has already been used to provide real-time monitoring of a harmful algae bloom (*Karenia mikimotoi*) that was threating fisheries and aquaculture sectors (southern bluefin tuna, yellowtail kingfish and abalone) around Port Lincoln during 2019.

Key words

Pacific Oyster (*Magallana gigas*, syn. *Crassostrea gigas*), Pacific Oyster Mortality Syndrome (POMS), Ostreid herpesvirus, disease management area, surveillance, biophysical model, hydrodynamics, particle tracking, hydrodynamic model, marine connectivity.

Introduction

Pacific Oyster Mortality Syndrome (POMS), caused by the Ostreid Herpesvirus type 1 (OsHV-1) microvariant, is associated with mass mortalities in Pacific oysters. POMS has impacted oyster growing regions in Europe, New Zealand and Australia. In Australia, POMS caused sudden high oyster mortalities and economic impacts in New South Wales (first detected in 2010) and Tasmania (first detected in 2016). In February 2018, POMS was also detected in feral Pacific oysters in Port Adelaide, South Australia. In all three Australian jurisdictions, OsHV-1 has been effectively contained to initially infected areas for years.

The detection of POMS in the Port River puts the States \$40 million / year oyster farming industry at risk. The closest oyster farming region to the Port River is ~60 km away, while the closest oyster hatchery is ~25 km away. Substantial amounts of resources have been committed to contain OsHV-1 to Port Adelaide. For example, there is a ban on the removal of bivalves from the infected area, associated compliance activities, feral oyster destruction at strategic locations, surveillance, technical advice to vessel owners particularly in relation to biofouling management, and a communications and awareness campaign. There are a number of pathways by which OsHV-1 can spread to new areas, including through movement of infected oysters (e.g. livestock, bait/berley, vessel biofouling), contaminated equipment and through water from an infected area (Rodgers et al., 2019).

Understanding dispersal and transmission of OsHV-1 through water is important for epidemiology and disease management purposes, in particular for determining Disease Management Areas (DMAs) and risk based early detection surveillance. Dispersal and transmission is governed by biological characteristics of the virus (e.g. viral survival outside the host, infective concentration, and temperature dependent activity) and hydrodynamics. While there is some information on OsHV-1 survival and decay in water (Vigneron et al., 2004; Martenot et al., 2015; Hick et al., 2016), there is a lack of information on dispersal of OsHV-1 based on hydrodynamic variables (Paul-Pont et al., 2014; Pernot et al., 2016; Rodgers et al., 2019).

In the absence of specific information on pathogen dispersal distance in water, for the purpose of policy, zoning and establishing DMAs an assumed distance of 5 km (Aldrin et al., 2011; WA Policy, 2017; Landos et al., 2019) or 5 NM (<10 km) (Department of Agriculture, 2009; Stevens, 2012; Australia's National Abalone Health Accreditation Program; PIRSA Policy; Australian import permit conditions for Salmonid products) has been used for viral and bacterial infections based on previous literature (e.g. Needham, 1995; Jarp and Karlsen, 1997; McClure et al., 2005; Aldrin et al., 2011). However, some pathogens can spread further than 5 NM in open marine systems due to their biology (e.g. parasites, protozoan spores), site-specific oceanographic conditions, vectors (e.g. scavengers) or fomites (e.g. vessels).

For OsHV-1, the nationally agreed emergency response plan (Department of Agriculture, 2015) suggests that the establishment of DMA boundaries must take into account dispersal of virus through water, including local oceanography and wild oyster populations. It is advised to overestimate the size of DMAs and change their area as required during the response or if more knowledge becomes available. In South Australia, biosecurity zones (or DMAs) for oyster growing regions have previously been determined based on an assumed viral dispersal distance of 5 NM (<10 km) between Pacific oyster populations including farmed and known wild populations (Figure 1). These DMAs represent epidemiologically distinct oyster populations with the same OsHV-1 dispersal risk through water, and are used for surveillance, disease management, livestock movement restrictions during mortality investigations and emergency disease responses.



Figure 1. Map of South Australia identifying the 11 Pacific Oyster Biosecurity Zones used prior to this report, based on 5 nautical mile buffers surrounding commercial oyster growing regions to define epidemiological units. These zones are now referred to as Disease Management Areas (DMAs).

Recently, a validated hydrodynamic particle tracking model of South Australian waters (eSA-Marine: <u>www.pir.sa.gov.au/research/esa_marine</u>) was developed to provide predictive capability for particle dispersal such as larvae, pathogens, harmful algae blooms and toxins (FRDC 2016/005: Middleton et al., 2017). McLeay et al. (2016) previously used this model to predict the dispersal of prawn larvae in South Australia for example.

The development of coupled biological-physical models (herein referred to as biophysical models) can provide more accurate information about particle dispersal at given geographic locations to improve epidemiology and disease management (Salama and Rabe, 2013). The framework for developing such a model includes inputting biological parameters of the particles (e.g. virus) into a hydrodynamic model with underlying assumptions and limitations governing the modelled output data (see Figure 2). Biological validation is an important final step particularly for particles that are active (e.g. sea lice). For passive particles (e.g. virus), the use of an already validated hydrodynamic model provides good predictive power for particle trajectories. This has been attempted previously for OsHV-1 to determine DMAs in New Zealand (Pande et al., 2015). Dispersal polygons were created around the maximum trajectory for each particle. A convex-hull algorithm is used to create a polygon that encompasses all the points where particles released from the farm were recorded. If polygons overlap, then they are considered at risk of infection and form one DMA (Morrisey et al., 2011: Figure 3). While Pande et al (2015) did not document OsHV-1 dispersal distance and used an assumed lifespan of 1 day, from their map of dispersal polygons it can be inferred that maximum dispersal distance was at least 15 km in 1 day. More recent research suggests OsHV-1 can survive in water for approximately 2 days (Martenot et al., 2015; Hick et al., 2016).

In this project, our aim was to develop a biophysical particle tracking model for OsHV-1 in South Australia. The hydrodynamic model incorporates real observed data for ocean currents, tides, wind forcing and water temperature. Hypothetical outbreaks of OsHV-1 were modelled for up to 22 days (maximum survival time for OsHV-1 DNA) across 23 sites in South Australia for Spring, Summer and Autumn in 2018-19. The results provide more accurate information to determine DMAs for South Australian oyster farming regions, and will improve future emergency disease responses, early detection surveillance and the epidemiological understanding of OsHV-1. This project will also provide valuable information on predicted trajectories for other passive particles such as other pathogens, harmful algae blooms, chemicals, toxins or oil spills.



Figure 2. Framework for developing methods for investigating the environmental transmission of disease causing agents, from Salama and Rabe (2013).

Objectives

There were three Objectives of the project:

- 1. To model viral particle dispersal at key locations around South Australia, including commercial oyster growing areas, known feral oyster populations, key ports (potential feral oysters), and incorporating seasonal oceanographic parameters
- 2. Using hydrodynamic model outputs, identify epidemiological units (DMAs) to inform surveillance, disease management and emergency disease response activities
- 3. Demonstrate how hydrodynamic model outputs of predicted viral particle dispersal can be used to develop a risk-based surveillance design for the detection of OsHV-1

Method

Development of a Biophysical Model

Hydrodynamic Model

Model data (e.g. currents, tide, wind, temperature) were used where there was at least 22 consecutive days available, which was sourced from the e-SA marine system: <u>https://pir.sa.gov.au/research/esa_marine</u>. Compiled data-assimilating model output in South Australia for Autumn 2018 (9 April – 1 May), Spring 2018 (29 October – 20 November) and Summer 2019 (5 – 27 January) scenarios were used.

Ocean circulation within Spencer Gulf and Gulf St Vincent were simulated using the Regional Ocean Modelling System (ROMS). ROMS is a high resolution, three-dimensional ocean model that incorporates a time step of 50 s to allow the model to solve the dominant tidal currents occurring in the Gulfs. The resultant models for the two gulfs is called the Two Gulf Model (TGM) (500 m grid, 1 hourly outputs). This model has previously been validated against 'now-cast' model output from satellite information and the Southern Australian Integrated marine Observing System (SAIMOS) which includes moorings and field surveys (McLeay et al., 2016; Middleton et al., 2017 FRDC 2016/005). That data includes sea level, ocean currents, tides and temperature.

For the West Coast, conditions for velocity, temperature and salinity in two-dimensions at the open boundaries were obtained from the output of the South Australian Regional Ocean Model (SAROM) (2.5 km grid, 1 hourly outputs, 180 s time step). The SAROM model is a large-scale model previously developed by Middleton et al. (2013).

Biological Characteristics

Latest research on OsHV-1, as well as discussions with virologists, were used to incorporate the current known biological characteristics of OsHV-1 into the biophysical model.

Survival

Particle tracking time in the model represents survival of the virus and the DNA. Under laboratory conditions, the virus can remain infective for two days in water at 20°C (Hick et al., 2016) and 2.25 days (54h) at 16°C (Martenot et al., 2015). Further, viral DNA can be detected in seawater up to 22 days at 4°C and 12 days at 20°C from macerated infected larvae under laboratory conditions (Vigneron et al., 2004).

It has been suggested that these timeframes may be extended in field conditions, where the virus may be bound up and protected in biological material (S. Corbeil [CSIRO] pers. comms; P. Hick and R. Whittington [Uni. Sydney] pers. comms). However, particle survival time may also be influenced by thinner or degraded mucus (Roberts and Powell, 2005), virus dilution, viral decay (e.g. from exposure to sunlight and microbiota), and reduced viral infectivity (Garver et al., 2013).

Therefore, for this model viral survival was assumed to be 2 days. For comparison, previous dispersal models in the literature have assumed a lifespan of 24 hours for OsHV-1 (Pande et al., 2015) and ISA virus in salmon (Murray et al., 2005). Viral DNA was assumed to last up to 22 days, with the latter being conservative (worst case scenario) given the water temperatures in South Australia (generally over 12°C; Roberts et al., 2012; Roberts et al., 2019). In the model, virus particles will be released and tracked continuously for 22 days. Key time points for analyses will be 2 days (virus survival time), 4 days, 7 days, 14 days and 20 days (to ensure enough data points are available for analyses). If particle survival or infective time in water is found to be different in

the future, this study provides data and regression analyses to determine dispersal distance for up to 22 days.

Temporal

PIRSA's Disease Response Plan is triggered when oyster mortalities occur in water temperatures over 17°C (Roberts et al., 2013). The virus is active (risk of infection) during warmer seasons when water temperature is over 16°C (Rodgers et al., 2019), while outbreaks typically occur at over 18°C in Tasmania (Ugalde et al., 2018), or over 19°C in NSW (Rodgers et al., 2019). The effect of seasonality (e.g. Spring, Summer and Autumn) will be investigated during the times of year when average water temperatures are above 17°C. Outputs of the eSA-Marine models where 22 days of continuous data were available for each season were Autumn 2018 (9 April – 1 May), Spring 2018 (29 October – 20 November) and Summer 2019 (5 – 27 January). These hydrodynamic outputs were used to determine dispersal for each season and at each of the 23 sites of interest.

Particle characteristics

Due to the small size of the viral particles (less than approximately 0.1µm), and their non-motile nature, each particle is considered passive in the model. Viral dispersal through water likely occurs within aggregate particulate matter (e.g. oyster faeces, sloughed oyster tissue cells, eggs, mucus from an infected oyster) (Paul-Pont et al., 2013; Evans et al., 2014; Martenot et al., 2015; Whittington et al., 2018). A single infected oyster can have between 100,000 and 30 million virions per mg tissue (or ml water) (Paul-Pont et al., 2015). Therefore it can shed a high number of viral particles (>100,000 virions per mg or ml). Given that an infective dose is >5,000 virions under laboratory conditions (Paul-Pont et al., 2015), each particle in this model represents 10,000's of virions and is therefore assumed to potentially infect a new oyster population.

During an outbreak, virus shedding occurs constantly from the infected population until the population dies off. We can assume a population would be "tens of thousands to hundreds of thousands" of oysters. However only a proportion of them would be infected, shedding virus and dying / decaying at any one time. During an outbreak, while the virus incubation period is only 4-5 days (Paul-Pont et al., 2014), and the kill rate can be >90%, infection and mortality sporadically moves through the population. So the outbreak can last for weeks / months. Therefore, the model will continuously release 200 particles every hour for the whole model run (22 days) for marine sites, which provides 1000's of particles being tracked during any day.

Sites

Prior to this project, PIRSA had separated its commercial oyster growing regions into 11 biosecurity zones (DMAs) based on an assumed disease spread distance of 5 NM (~10 km) for the purposes of disease management and emergency response (PIRSA documents: "POMS Surveillance Strategy 2017" file reference A3073039; "Proposed Permit System for Oyster Movements in SA" file reference A3442328).

For each of the 11 oyster growing regions, 23 sites have been chosen to run the biophysical model. Sites were chosen based on:

- Closest lease or hatchery to an adjacent biosecurity zone (growing region) and/or
- Closest known feral oyster population, or port, wharf or marina where, within or adjacent to a biosecurity zone (growing region).

Sites were chosen with input from key stakeholders (South Australian Oyster Growers Association [SAOGA], Primary Industries and Regions SA [PIRSA] South Australian Shellfish Quality Assurance Program [SASQAP], PIRSA Biosecurity SA and Aquatic Biosecurity Pty Ltd). Hypothetical virus particles were released at the identified sites.

Biophysical Model

The biological model is coupled with the three-dimensional hydrodynamic models to an offline Lagrangian particle-tracking model (LTRANS, North et al., 2006, 2008). For similar detailed methods see McLeay et al. (2016) and Fowler (2019). Initial particle positions were centred on 23 sites determined by PIRSA. Two hundred (200) particles were released every 1 hour at each site and tracked from start to end of day 22. Particle positions were stored every 30 minutes. This represented a large amount of modelled data, for example 96,200 2-day old particles were tracked over 481 release times for a single site during one season. Each particles age (in days) was stored along with particle ID.

Dispersal of particles were measured by enclosing particles with polygons formed by calculating the convex hull (in Matlab "ind=convhull(x,y); plot(x(ind),y(ind));") which represents the smallest convex set that encloses all particles. Note that age at which the POMS virus ceases to be infectious is 2 days. Particles are classified as infectious (age = 0 to 2 days) and non-infectious but still detectible by DNA analysis (age = 2 to 22 days). Convex hulls are calculated for 2 days, 4 days, 7 days, 14 days and 22 days.

For data analyses (regression and ANOVA), a time duration of 20 days was analysed (instead of 22 days) to ensure adequate replication. During the 22 day model run, for each season, the number of release times (or simulations) and therefore total number of particles tracked for each time duration were as follows:

- Day 2 = 481 releases with 96,200 particles tracked
- Day 4 = 433 releases with 86,600 particles tracked
- Day 7 = 361 releases with 72,200 particles tracked
- Day 14 = 193 releases with 38,600 particles tracked
- Day 20 = 49 releases with 9,800 particles tracked

Particle tracking outputs were reported as "Maximum" dispersal distance and "90th percentile" of particles. Maximum dispersal distance represents the furthest any of the 1000's of particles can travel under the given hydrodynamic conditions. 90th percentile distance is the distance at which the 180th particle travels at each release time (out of 200 particles), then the distance at which the 90th percentile of those particles travel from all hourly releases.

Analyses

Connectivity Between Sites and Regions

Convex hulls, or polygons, provide information on connectivity between sites and regions.

In Pande et al. (2015) and Morrisey et al. (2011), dispersal polygons were drawn around each particle tracked after the lifespan of the particle (i.e. 24 hours in both papers). A convex-hull algorithm is used to create a polygon that encompasses all the points where particles released from the farm were recorded. If polygons overlap, then they consider it a risk of infection and they form one disease management unit (one zone). Separate dispersal polygons are generated for different DMAs. If a farm lies within a dispersal plume from another area, it is considered at risk (Figure 3).



Figure 3. Representation of process by which biosecurity zones (or DMAs) are defined, through particle dispersion areas of overlap, adapted from Morrisey et al. (2011).

Based on Figure 3, polygons in this project were classed as follows:

- 2 day plume = high risk (90% of particles as per Samsing et al., 2017),
- 2 day maximum hull = moderate risk,
- 4 days = low risk,
- 7 days = unlikely risk
- 14 days = negligible risk
- 22 days = negligible risk

Statistical Analyses

IBM SPSS, Version 26 for Windows (IBM SPSS Inc., Chicago, IL, USA) was used for all statistical analyses. Homogeneity of variances and normality among mean values were assessed using Levene's test and the Shapiro–Wilk test, respectively. The average maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior were used in a two-factor ANOVA to determine the interaction between site (within a disease management zone) and season (summer, spring and autumn). When significant interactions were observed, post-hoc tests were used to detect significant differences between all treatments combinations (Tukey's post hoc). If there were no significant interactions, the interaction was not included in the model and main effects were analysed. Regression analyses (linear, quadratic or logarithmic) were also used to determine the relationship between maximum dispersal distance and days and also the 90th percentile of particles and days. A significance level of P < 0.05 was used for all statistical tests. All values are presented as means \pm standard deviation (SD), unless otherwise stated.

Results

Updated OsHV-1 DMAs for South Australia's oyster industry are shown in Figure 4. This was based on two day particle dispersal polygons, across all seasons and sites, with overlapping polygons (which indicate medium and high risk connectivity) defining individual DMAs. Dispersal polygons were based on the maximum distance any single particle travelled during 481 simulations (using compiled data-assimilating model output from 2018 and 2019), which provides a conservative and scientifically robust level of management.



Figure 4. Disease Management Areas (DMA) for the South Australian oyster industry. Proper Bay and Boston / Louth Bay are currently considered two separate DMAs. If a significant feral oyster population is detected in the Boston Bay area in the future, those two DMAs should be merged into one (Proper, Boston and Louth Bay).

Denial Bay and Smoky Bay Disease Management Area

The Smoky and Denial Bay DMA is displayed in Figure 5. This DMA is contained and bounded by a line commencing at mean high water springs closest to 32° 11' 5.924" South, 133° 21' 34.052" East, then south-westerly to 32° 11' 27.784" South, 133° 21' 25.564" East, then south-easterly to 32° 30' 57.985" South, 133° 51' 21.197" East, then beginning north-westerly following the line of mean high water springs to the point of commencement.

The Smoky and Denial Bay DMA is based on the combined maximum hull from the biophysical modelling of OsHV-1 particles from Site 1, 2, 3 and 4 during Spring, Summer and Autumn (Figure 6 - 10).

The relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 11 and Table 1. Combined across all sites, the maximum dispersal distance for 2 days was 26.2 km (90th percentile = 19.0 km) during Spring and increased to 34.6 km (90th percentile = 25.4 km) after 20 days during summer (Table 1). The maximum dispersal distance of OsHV-1 particles after 2 days was significantly influenced by season (Spring > Summer > Autumn; P < 0.001) and site (Site 3 > Site 1 > Site 4 > Site 2; P < 0.001), although there was a significant interaction between these factors (P < 0.001) indicating that particle dispersal patterns for each site depend on the season (Table 2).



Figure 5. Disease Management Area for Denial Bay and Smoky Bay. Note that 'aquaculture licences' represent all active aquaculture licence holders as at the date of this report, and may not necessarily represent oyster farms only (see <u>www.aginsight.sa.gov.au/</u>).



Figure 6. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in Denial Bay (Site 1; - 32.2560°, 133.6689°).



Figure 7. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from Thevenard wharf in Denial Bay (Site 2; - 32.1492°, 133.6403°).



Figure 8. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in Smoky Bay (Site 3; - 32.3642°, 133.8578°).



Figure 9. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from the township of Smoky Bay (Site 4; -32.3784°, 133.9309°).



Figure 10. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Denial Bay and Smoky Bay at Site 1 (-32.2560°, 133.6689°), 2 (-32.1492°, 133.6403°), 3 (32.3642°, 133.8578°) and 4 (-32.3784°, 133.9309°).



Figure 11. Maximum dispersal distance and 90th percentile of particles at 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Denial Bay and Smoky Bay at Site 1, 2, 3, and 4.

Table 1.	Maximum dispersal distance (km) and 90 th percentile of particles (in parentheses) at
2, 4, 7, 14 a	and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer
and Autum	n in Denial Bay and Smoky Bay at Site 1, 2, 3, and 4.1

Site	Season	2	4	7	14	20	Equation (max)	r ² (max)	P value (max)
Site 1	Summer	28.0	29.8	32.6	32.6	36.6	$y = 3.337 \ln(x) + 25.48$	0 90	P = 0.015
	Spring	(24.7) 30.2 (24.4)	(27.5) 34.8 (28.0)	(28.8) 34.8 (28.0)	(28.8) 34.8 (28.0)	(28.8) 34.8 (28.0)	y = 1.616ln(x) + 30.733	0.55	P = 0.149
	Autumn	(24.4) 30.1 (25.3)	(20.0) 32.9 (29.8)	(20.0) 32.9 (30.0)	(20.0) 32.9 (30.0)	(20.0) 32.9 (30.0)	y = 1.028 ln(x) + 30.368	0.55	P = 0.149
	All seasons	29.4 (24.8)	32.5 (28.4)	33.4 (28.9)	33.4 (28.9)	34.8 (28.9)	$y = 1.994 \ln(x) + 28.86$	0.85	P = 0.025
Site 2	Summer	7.7 (6.7)	7.8 (7.5)	7.8 (7.6)	7.8 (7.7)	7.8 (7.7)	y = 0.046ln(x) + 7.719	0.57	P = 0.140
	Spring	7.7 (6.4)	7.8 (7.3)	7.8 (7.3)	7.8 (7.5)	7.8 (7.5)	$y = 0.036 \ln(x) + 7.736$	0.64	P = 0.103
	Autumn	5.9 (4.6)	7.6 (6.3)	7.8 (7.1)	7.8 (7.5)	7.8 (7.6)	$y = 0.706 \ln(x) + 6.040$	0.64	P = 0.103
	All seasons	7.1 (5.9)	7.8 (7.1)	7.8 (7.3)	7.8 (7.6)	7.8 (7.6)	$y = 0.263 \ln(x) + 7.1649$	0.64	P = 0.104
Site 3	Summer	34.0 (29.2)	47.2 (38.8)	50.6 (47.0)	50.6 (47.2)	50.6 (47.4)	y = 6.470ln(x) + 34.122	0.70	P = 0.077
	Spring	38.6 (29.3)	49.8 (38.5)	49.8 (38.5)	50.1 (38.5)	50.1 (38.5)	y = 4.178ln(x) + 39.611	0.58	P = 0.135
	Autumn	21.8 (18.0)	37.5 (31.0)	40.9 (31.8)	40.9 (31.8)	50.5 (31.8)	y = 10.289 ln(x) + 18.454	0.85	P = 0.027
	All seasons	31.5 (25.5)	44.9 (36.1)	47.1 (39.1)	47.2 (39.2)	50.4 (39.3)	$y = 6.979 \ln(x) + 30.729$	0.77	P = 0.050
Site 4	Summer	30.9 (16.4)	38.3 (17.5)	42.4 (17.6)	43.2 (17.6)	43.3 (17.6)	y = 5.199ln(x) + 29.593	0.84	P = 0.029
	Spring	28.2 (16.1)	31.1 (17.2)	31.1 (17.2)	31.1 (17.2)	31.1 (17.4)	$y = 1.013 \ln(x) + 28.553$	0.55	P = 0.149
	Autumn	17.4 (11.2)	30.8 (16.7)	30.8 (16.7)	34.5 (17.3)	34.5 (17.3)	$y = 6.693 \ln(x) + 16.699$	0.78	P = 0.048
	All seasons	25.5 (14.6)	33.4 (17.1)	34.8 (17.2)	36.3 (17.4)	36.3 (17.4)	$y = 4.302 \ln(x) + 24.948$	0.79	P = 0.041
All sites	Summer	25.2 (19.2)	30.8 (22.8)	33.4 (25.2)	33.6 (25.3)	34.6 (25.4)	y = 3.7631ln(x) + 24.228	0.84	P = 0.028
	Spring	26.2 (19.0)	30.9 (22.8)	30.9 (22.8)	30.9 (22.8)	30.9 (22.8)	y = 1.7115ln(x) + 26.658	0.57	P = 0.140
	Autumn	18.8 (14.8)	27.2 (20.9)	28.1 (21.4)	29.1 (21.7)	31.4 (21.7)	$y = 4.6791 \ln(x) + 17.89$	0.82	P = 0.033

¹ Data is for 2 days (96,200 particles from 481 releases), 4 days (86,600 particles from 433 releases), 7 days (72,200 particles from 361 releases), 14 days (38,600 particles from 193 releases) and 20 days (9,800 particles from 49 releases). The model was run for 22 days with hourly releases of 200 particles, while data analyses was based on the furthest any of the 200 particles can travel.

Table 2.	Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles
after 2 days	Juring Spring, Summer and Autumn in Denial Bay and Smoky Bay at Site 1, 2, 3, and
1 1	

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
Site 1	Summer Spring Autumn	19.2 ± 4.8^{b} 11.9 ± 7.1 ^d 13.5 ± 7.7 ^c
Site 2	Summer Spring Autumn	6.2 ± 0.9^{g} 4.8 ± 1.4^{h} 4.5 ± 0.6^{h}
Site 3	Summer Spring Autumn	21.7 ± 6.9 ^a 11.9 ± 9.3 ^d 10.5 ± 5.5 ^e
Site 4	Summer Spring Autumn	$14.0 \pm 4.6^{\circ}$ 8.0 ± 5.9 ^f 6.0 ± 3.9 ^g
ANOVA ² Site Season Site × Season		P < 0.001 P < 0.001 P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior. ² Where significant main effects were detected, post-hoc tests were used to determine differences between means (Tukeys test; P < 0.05). For the significant interaction (Site × Season; P < 0.05), difference in site are compared

across all season (one-factor ANOVA, Tukeys test), values without a common superscript are significantly different (a indicates the highest value; P < 0.05).

Streak Bay and Haslam Disease Management Area

The Streaky Bay and Haslam DMA is displayed in Figure 12. This DMA is contained within and bounded by a the line commencing at mean high water springs closest to 32° 28' 15.467" South, 134° 5' 10.954" East, then south-westerly to 32° 28' 31.598" South, 134° 4' 50.902" East, then south-westerly to 32° 28' 51.546" South, 134° 4' 30.662" East, then south-easterly to 32° 30' 26.615" South, 134° 4' 45.311" East, then south-easterly to 32° 42' 42.300" South, 134°7'38.899" East, then south-easterly to 32° 42' 53.420" South, 134° 7' 50.848" East, then south-easterly to the location on mean high water springs closest to 32° 43' 12.648" South, 134° 7' 57.191" East, then beginning easterly following the line of mean high water springs to the point of commencement.

The Streaky Bay and Haslam DMA is based on the combined maximum hull from the biophysical modelling of OsHV-1 particles from Site 5, 6 and 7 during Spring, Summer and Autumn (Figure 13 - 16).

The relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 17 and Table 3. Combined across all sites, the maximum dispersal distance for 2 days was 18.1 km (90th percentile = 11.8 km) during Spring and increased to 25.9 km (90th percentile = 21.3 km) after 20 days during summer (Table 3). The maximum dispersal distance of OsHV-1 particles after 2 days was significantly influenced by season (Spring > Summer > Autumn; P < 0.001) and site (Site 6 > Site 7 > Site 5; P < 0.001), although there was a significant interaction between these factors (P < 0.001) indicating that particle dispersal patterns for each site depend on the season (Table 4).



Figure 12. Disease Management Area for Streaky Bay and Haslam. Note that 'aquaculture licences' represent all active aquaculture licence holders as at the date of this report, and may not necessarily represent oyster farms only (see <u>www.aginsight.sa.gov.au/</u>).



Figure 13. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in Streaky Bay (Haslam) (Site 5; -32.5436°, 134.2007°).



Figure 14. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in Streaky Bay (Site 6; - 32.6362°, 134.2255°).



Figure 15. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from the township in Streaky Bay (Site 7; - 32.7941°, 134.2109°).



Figure 16. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Streaky Bay at Site 5 (-32.5436°, 134.2007°), 6 (-32.6362°, 134.2255°) and 7 (-32.7941°, 134.2109°).



Figure 17. Maximum dispersal distance and 90th percentile of particles at 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Streaky Bay and Haslam at Site 5, 6 and 7.

		Days							
Site	Season	2	4	7	14	20	Equation (max.)	r² (max)	P values (max)
Site 5	Summer	11.7 (9.2)	14.1 (11.0)	16.9 (11.3)	17.2 (11.6)	18.7 (12.2)	$y = 2.905 \ln(x) + 10.114$	0.94	P = 0.007
	Spring	14.1 (9.3)	14.1 (10.5)	14.1 (10.5)	14.1 (10.5)	14.1 (11.0)	NA	NA	NA
	Autumn	9.8 (7.0)	15.4 (9.9)	15.4 (9.9)	15.4 (10.3)	15.4 (10.3)	$y = 2.025 \ln(x) + 10.393$	0.55	P = 0.149
	All seasons	11.8 (8.5)	14.5 (10.5)	15.5 (10.6)	15.6 (10.8)	16.1 (11.2)	$y = 1.6431 \ln(x) + 11.522$	0.82	P = 0.033
		04.5	045	007	007	007			
Site 6	Summer	21.5 (15.8)	24.5 (21.1)	26.7 (21.7)	26.7 (21.7)	26.7 (22.9)	y = 2.238ln(x) + 20.921	0.81	P = 0.039
	Spring	20.5 (14.7)	24.7 (19.9)	24.7 (19.9)	24.7 (19.9)	24.7 (20.3) 22.7 (15.1)	y = 1.489ln(x) + 20.955	0.55	P = 0.149
	Autumn	11.0 (7.9)	20.5 (15.1)	22.7 (15.1)	22.7 (15.1)		y = 4.552ln(x) + 11.154	0.69	P = 0.080
	All seasons	17.1 (12.8)	23.2 (18.7)	24.7 (18.9)	24.7 (18.9)	24.7 (19.4)	$y = 2.7598 \ln(x) + 17.677$	0.71	P = 0.075
		40 5	07.0	00 4					
Site 7	Summer	19.5 (13.8)	27.6 (23.7)	32.1 (28.6)	32.3 (28.6)	32.3 (28.7)	$y = 5.347 \ln(x) + 18.443$	0.80	P = 0.042
	Spring	19.8 (11.6)	29.1 (22.5)	31.4 (27.1)	31.4 (27.1)	31.4 (27.1)	y = 4.501ln(x) + 19.938	0.70	P = 0.079
	Autumn	8.0 (5.5)	15.7 (10.9)	25.3 (17.1)	31.2 (17.1)	31.2 (17.1)	y = 10.733 ln(x) + 1.552	0.96	P = 0.004
	All seasons	15.8 (10.3)	24.1 (19.0)	29.6 (24.3)	31.7 (24.3)	31.7 (24.3)	y = 6.8603 ln(x) + 13.311	0.89	P = 0.016
					 (
All sites	Summer	17.6 (12.9)	22.1 (18.6)	25.3 (20.5)	25.4 (20.6)	25.9 (21.3)	$y = 3.4966 \ln(x) + 16.492$	0.85	P = 0.026
	Spring	18.1 (11.8)	22.6 (17.6)	23.4 (19.2)	23.4 (19.2)	23.4 (19.5)	$y = 1.9967 \ln(x) + 18.317$	0.66	P = 0.094
	Autumn	9.6 (6.8)	17.2 (12.0)	21.2 (14.0)	23.1 (14.2)	23.1 (14.2)	$y = 5.77 \ln(x) + 7.6998$	0.88	P = 0.017

Table 3. Maximum dispersal distance (km) and 90th percentile of particles (in parentheses) for 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Streaky Bay and Haslam at Site 5, 6 and 7.¹

¹ Data is for 2 days (96,200 particles from 481 releases), 4 days (86,600 particles from 433 releases), 7 days (72,200 particles from 361 releases), 14 days (38,600 particles from 193 releases) and 20 days (9,800 particles from 49 releases). The model was run for 22 days with hourly releases of 200 particles, while data analyses was based on the furthest any of the 200 particles can travel.

Table 4. Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles after 2 days using biophysical modelling during Spring, Summer and Autumn in Streaky Bay and Haslam at Site 5, 6 and 7.¹

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
Site 5	Summer Spring Autumn	8.1 ± 1.7^{c} 5.5 ± 2.8^{ef} 5.2 ± 2.0^{fg}
Site 6	Summer Spring Autumn	12.5 ± 3.4^{a} 7.8 ± 4.6 ^c 6.0 ± 2.0 ^e
Site 7	Summer Spring Autumn	10.5 ± 3.4^{b} 7.1 ± 3.9 ^d 4.9 ± 1.4 ^g
ANOVA ² Site Season Site × Season		P < 0.001 P < 0.001 P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior.

² Where significant main effects were detected, post-hoc tests were used to determine differences between means (Tukeys test; P < 0.05). For the significant interaction (Site × Season; P < 0.05), difference in site are compared across all season (one-factor ANOVA, Tukeys test), values without a common superscript are significantly different (^a indicates the highest value; P < 0.05).

Coffin Bay Disease Management Area

The Coffin Bay DMA is displayed in Figure 18. This DMA is contained within and bounded by a line commencing at mean high water springs closest to 34° 20' 49.110" South, 135° 21' 26.449" East, then north-westerly to 34° 20' 38.951" South, 135° 21' 17.039" East, then north-westerly to 34° 19' 57.749" South, 135° 21' 4.828" East, then north-westerly to 34° 19' 51.719" South, 135° 20' 52.368" East, then north-westerly to 34° 19' 44.692" South, 135° 20' 28.630" East, then north-westerly to 34° 19' 3.324" South, 135° 17' 37.896" East, then north-westerly to 34° 18' 24.566" South, 135° 13' 12.173" East, then westerly to 34° 18' 24.426" South, 135° 12' 53.532" East (Point 8), then south-westerly to 34° 18' 40.061" South, 135° 12' 35.708" East (Point 9), then south-westerly to 34° 25' 37.938" South, 135° 11' 26.286" East (Point 10), then south-easterly to the location on mean high water springs closest to 34° 26' 5.410" South, 135° 11' 43.091" East (Point 11), then beginning north-easterly following the line of mean high water springs to the point of commencement.

The Coffin Bay DMA is based on the combined maximum hull from the biophysical modelling of OsHV-1 particles from Site 8 during Spring, Summer and Autumn (Figure 19).

The relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 20 and Table 5. Combined across all seasons, the maximum dispersal distance for 2 days was 21.5 km (90th percentile = 12.9 km) and increased to 162 km (90th percentile = 141.5 km) after 20 days (Table 5). Of all 23 sites across the state, Coffin Bay showed the greatest maximum particle dispersal distance at 20 days with potential viral DNA travelling 310 km (90th percentile =297.7 km) during summer. The maximum dispersal distance of OsHV-1 particles after 2 days was significantly influenced by season (summer > spring > autumn; P < 0.001) (Table 6).



Figure 18. Disease Management Area for Coffin Bay. Note that 'aquaculture licences' represent all active aquaculture licence holders as at the date of this report, and may not necessarily represent oyster farms only (see <u>www.aginsight.sa.gov.au/</u>).



Figure 19. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in Coffin Bay (Site 8; -34.5386°, 135.3554°).



Figure 20. Maximum dispersal distance and 90th percentile of particles at 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Coffin Bay at Site 8.

Table 5. Maximum dispersal distance (km) and 90th percentile of particles (in parentheses) at 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Coffin Bay at Site 8.¹

	Season	days							
Site		2	4	7	14	20	Equation (max.)	r² (max)	P value (max)
Site 8	Summer	28.9 (16.0)	63.4 (40.6)	138.9 (98.1)	257.4 (216.0)	310.0 (297.7)	y = -0.542x ² + 27.959x - 31.023	0.99	P = 0.002
	Spring	19.3 (11.2)	39.2 (21.5)	68.7 (21.7)	103.4 (64.9)	132.1 (94.2)	y = -0.2019x ² + 10.555x	0.99	P = 0.006
	Autumn	16.4 (11.6)	33.9 (17.6)	33.9 (18.8)	35.4 (22.6)	43.8 (32.7)	y = 9.5878ln(x) + 14.13	0.80	P = 0.040
	All season	21.5 (12.9)	45.5 (26.6)	80.5 (46.2)	132 (101.1)	162 (141.5)	y = -0.2301x ² + 12.697x	0.99	P = 0.001

¹ Data is for 2 days (96,200 particles from 481 releases), 4 days (86,600 particles from 433 releases), 7 days (72,200 particles from 361 releases), 14 days (38,600 particles from 193 releases) and 20 days (9,800 particles from 49 releases). The model was run for 22 days with hourly releases of 200 particles, while data analyses was based on the furthest any of the 200 particles can travel.
Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles Table 6. after 2 days using biophysical modelling during Spring, Summer and Autumn in Coffin Bay at Site 8.¹

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
Site 8	Summer Spring Autumn	11.8 ± 4.8^{a} 7.3 ± 3.6 ^b 6.9 ± 3.3 ^b
ANOVA ² Season		P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior. ² Where significant main effects were detected, post-hoc tests were used to determine differences between means

⁽Tukeys test; ^a indicates the highest value; P < 0.05).

Proper Bay and Boston / Louth Bay Disease Management Areas

The Proper Bay and Boston / Louth Bay DMA is displayed in Figure 21a.

The Proper Bay DMA (Figure 21a) comprises the State waters contained within and bounded by a line commencing at mean high water springs closest to 34° 45' 9.594" South, 135° 53' 28.082" East, then south-easterly to 34° 45' 38.336" South, 135° 54' 29.340" East, then south-easterly to the location on mean high water springs closest to 34° 46' 24.632" South, 135° 55' 35.029" East, then beginning south-westerly following the line of mean high water springs to the point of commencement.

The Boston / Louth Bay DMA (Figure 21a) comprises the State waters contained within and bounded by a line commencing at mean high water springs closest to 34° 47' 39.595" South, 136° 0' 56.268" East, then south-easterly to 34° 47' 42.101" South, 136° 1' 7.298" East, then north-easterly to 34° 47' 31.657" South, 136° 2' 51.295" East, then north-easterly to 34° 46' 25.374" South, 136° 5' 1.036" East, then north-easterly to 34° 45' 42.322" South, 136° 6' 14.760" East, then north-easterly to 34° 44' 52.202" South, 136° 7' 10.355" East, then north-easterly to 34° 44' 34.908" South, 136° 7' 26.378" East, then north-easterly to 34° 43' 50.743" South, 136° 7' 57.540" East, then north-easterly to 34° 40' 43.727" South, 136° 9' 44.608" East, then north-easterly to 34° 38' 56.292" South, 136° 10' 39.324" East, then north-easterly to 34° 38' 39.577" South, 136° 10' 45.455" East, then north-westerly to 34° 37' 3.439" South, 136° 10' 28.582" East, then north-westerly to 34° 30' 12.395" South, 136° 7' 49.184" East, then north-westerly to 34° 29' 55.486" South, 136° 7' 42.445" East, then north-westerly to 34° 29' 48.098" South, 136° 7' 22.400" East, then north-westerly to the location on mean high water springs closest to 34° 29' 44.898" South, 136° 7' 0.120" East, then beginning south-westerly following the line of mean high water springs to the point of commencement

If a feral Pacific oyster population is discovered in the Boston Bay area in the future (scenario 2, Figure 21b) the Boston / Louth Bay DMA would include Proper Bay in one larger DMA.

The Proper Bay and Boston / Louth Bay DMA (Figure 21a) are based on the combined maximum hulls after 2 days from the biophysical modelling of OsHV-1 particles at Site 9, 11 and 12 during Spring, Summer and Autumn (Figure 22, 25, 26 and 28). Biophysical modelling of OsHV-1 particles at Site 10 during Spring, Summer and Autumn (Figure 23 and 27) from hypothetical feral oyster populations on the Port Lincoln town wharf were not included when defining the Boston and Louth Bay DMA because there are currently no known feral oyster populations in this area. However, if feral oyster populations are detected around the Port Lincoln township area, these 2 DMAs would be linked and considered as one DMA for disease surveillance and emergency response purposes (see scenario 2 map; Figure 21b).

For Sites 9 and 10, the relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 24 and Table 7. Site 9 (Proper Bay) had the shortest maximum dispersal distance after 2 days out of all 23 sites investigated in the current project (Max = 5.2 km, 90th percentile = 1.9 km during Summer). Autumn conditions caused the greatest maximum dispersal distance of OsHV-1 particles after 2 days at Site 9 (Max = 6.6 km, 90th percentile = 2.7 km). Particle dispersal from Site 10 was further, with a maximum dispersal distance after 2 days occurring during autumn (Max = 28.1 km, 90th percentile = 16.8 km), and maximum particle dispersal peaking during Spring after 20 days (Max = 137.4 km, 90th percentile = 96.4 km). The maximum dispersal distance of OsHV-1 particles after 2 days after 2 days was significantly influenced by season (Autumn > Spring > Summer; P < 0.001) and site (Site10 > Site 9; P < 0.001), although there was a significant interaction between these factors (P < 0.001) indicating that particle dispersal patterns for each site depend on the season (Table 8).

For Sites 11 and 12, the relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 29 and Table 9. Combined across all sites, the maximum dispersal distance for 2 days was 19.3 km (90th percentile = 6.1 km) during Autumn and increased to 134.7 km (90th percentile = 64.3 km) after 20 days during summer (Table 9). The maximum dispersal distance of OsHV-1 particles after 2 days was significantly influenced by season (Autumn > Spring > Summer; P < 0.001) and site (Site 11 > Site 12; P < 0.001), although there was a significant interaction between these factors (P < 0.001) indicating that particle dispersal patterns for each site depend on the season (Table 10).



Figure 21. Disease Management Areas (DMA) for (a) the Proper Bay DMA and the Boston Bay and Louth Bay DMA which does not consider any feral oyster populations in the Boston Bay area, and (b) a single Boston Bay and Louth Bay DMA which incorporates the Proper Bay area as scenario 2 (hypothetical feral oyster population in Boston Bay). Note that 'aquaculture licences' represent all active aquaculture licence holders as at the date of this report, and may not necessarily represent oyster farms only (see <u>www.aginsight.sa.gov.au/</u>).



Figure 22. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in Proper Bay (Site 9; - 34.7610°, 135.8616°).



Figure 23. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from the Port Lincoln wharf in Boston Bay (Site 10; - 34.7163°, 135.8703°).



Figure 24. Maximum dispersal distance and 90th percentile of particles at 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Port Lincoln at Site 9 and 10.

				Days					
		2	4	7	14	20	Equation (max.)	r² (max)	P value (max)
Site 9	Summer	5.2 (1.9)	6.4 (3.2)	7.6 (4.7)	19.1 (5.3)	19.1 (5.6)	y = 0.897x + 3.055	0.90	P = 0.013
	Spring	6.0 (2.6)	13.1 (3.7)	23.1 (5.6)	44.8 (7.3)	68.6 (7.6)	y = 3.418x - 1.019	0.99	P < 0.001
	Autumn	6.6 (2.7)	7.8 (5.0)	21.3 (7.0)	38.7 (7.0)	55.6 (7.0)	y = 2.800x - 0.317	0.99	P < 0.001
	All seasons	5.9 (2.4)	9.1 (4.0)	17.3 (5.8)	34.2 (6.5)	47.8 (6.7)	y = 2.3713x + 0.5728	0.99	P < 0.001
		117	26.2	44.0	06 F	121.0			
Site 10	Summer	(7.7)	26.3 (11.1)	44.9 (25.1)	96.5 (47.7)	(76.7)	y = 6.789x - 0.389	1.00	P < 0.001
	Spring	21.5 (13.8)	26.6 (17.7)	48.1 (25.1)	99.6 (55.7)	9.6 137.4 55.7) (96.4)	y = 6.704x + 3.648	1.00	P < 0.001
	Autumn	28.1 (16.8)	52.0 (24.9)	62.1 (37.7)	89.6 (66.9)	98.9 (85.4)	y = 3.686x + 31.496	0.92	P = 0.009
	All seasons	21.4 (12.8)	35.0 (17.9)	51.7 (29.3)	95.2 (56.8)	123.7 (86.2)	y = 5.726x + 11.585	1.00	P < 0.001
All sites	Summer	9.9	16.4	26.2	57.8	77.0	v = 3.843x + 1.333	0 99	P < 0.001
	Cuminor	(4.8)	(7.1)	(14.9)	(26.5)	(41.1)		0100	
	Spring	(8.2)	(10.7)	(15.4)	(31.5)	(52.0)	y = 5.061x + 1.315	0.99	P < 0.001
	Autumn	17.4 (9.8)	29.9 (15.0)	41.7 (22.3)	64.1 (36.9)	77.2 (46.2)	y = 26.05ln(x) - 4.2597	0.98	P = 0.001

Table 7. Maximum dispersal distance (km) and 90th percentile of particles at 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Port Lincoln at Site 9 and 10.¹

Table 8. Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles after 2 days using biophysical modelling during Spring, Summer and Autumn in Port Lincoln at Site 9 and 10.¹

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
Site 9	Summer	2.5 ± 0.5^{c}
	Spring	$2.7 \pm 0.6^{\circ}$
	Autumn	$3.0 \pm 0.9^{\circ}$
Sita 10	Summor	72, 21b
Sile IU	Summer	$7.3 \pm 2.1^{\circ}$
	Spring	8.3 ± 4.0^{a}
	Autumn	8.8 ± 6.4^{a}
ANOVA ²		
Site		P < 0.001
Season		P < 0.001
Site × Season		P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior. ² Where significant main effects were detected, post-hoc tests were used to determine differences between means (Tukeys test; P < 0.05). For the significant interaction (Site × Season; P < 0.05), difference in site are compared across all season (one-factor ANOVA, Tukeys test), values without a common superscript are significantly different (^a indicates the highest value; P < 0.05).



Figure 25. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from an oyster hatchery in Louth Bay (Site 11; - 34.6490°, 135.9376°).



Figure 26. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine site in Louth Bay (Site 12; -34.6024°, 135.9122°).



Figure 27. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Boston and Louth Bay at Site 9 (-34.7610°, 135.8616°), Site 10 (-34.7163°, 135.8703°), Site 11 (-34.6490°, 135.9376°) and Site 12 (-34.6024°, 135.9122°).



Figure 28. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Boston and Louth Bay at Site 9 (-34.7610°, 135.8616°), Site 11 (-34.6490°, 135.9376°) and Site 12; -34.6024°, 135.9122°).



Figure 29. Maximum dispersal distance and 90th percentile of particles at 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Boston and Louth Bay at Site 11 and 12.

Table 9.	Maximum dispersal distance (km) and 90 th percentile of particles for 2, 4, 7, 14 and
20 days using	g biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in
Boston and L	outh Bay at Site 11 and 12.1

				days			_		
Site	Season	2	4	7	14	20	Equation (max.)	r ² (max)	P value (max)
Site 11	Summer	19.9 (13.7)	36.1 (18.3)	62.0 (29.4)	108.8 (63.4)	151.0 (92.9)	y = 7.213x + 7.794	1.00	P < 0.001
	Spring	21.9 (12.4)	33.2 (18.6)	60.1 (29.8)	98.6 (69.4)	123.8 (105.1)	y = 5.737x + 13.611	0.99	P = 0.001
	Autumn	23.7 (10.6)	38.8 (21.7)	72.9 (36.7)	81.6 (65.7)	100.6 (82.5)	y = 33.249ln(x) – 0.714	0.96	P = 0.003
	All seasons	21.1 (12.2)	36.1 (19.5)	65.0 (32.0)	96.4 (66.2)	125.1 (93.5)	y = 5.633x + 15.933	0.98	P = 0.001
Site 12	Summer	10.9 (1.8)	19.4 (5.8)	29.3 (10.0)	77.2 (16.7)	118.4 (35.7)	y = 6.067x – 5.999	0.99	P < 0.001
	Spring	13.5 (1.7)	23.0 (5.8)	38.7 (14.9)	83.7 (28.5)	103.3 (48.2)	y = 5.212x + 3.422	0.99	P = 0.001
	Autumn	15.0 (1.6)	35.0 (8.2)	58.5 (17.3)	78.0 (32.0)	94.9 (62.1)	y = 34.419ln(x) – 10.229	0.99	P < 0.001
	All seasons	13.1 (1.6)	25.8 (6.6)	42.2 (14.1)	79.6 (25.7)	105.5 (48.7)	y = -5.14x + 4.92	1.00	P < 0.001
All sites	Summer	15.4 (7.8)	27.8 (12.1)	45.7 (19.7)	93.0 (40.0)	134.7 (64.3)	y = 6.664x + 0.897	0.99	P <0.001
	Spring	17.7 (7.0)	28.1 (12.2)	49.4 (22.4)	91.2 (49.0)	113.5 (76.6)	y = -0.111x ² + 7.918x + 0.327	0.99	P = 0.002
	Autumn	19.3 (6.1)	36.9 (15.0)	65.7 (27.0)	79.8 (48.8)	97.7 (72.3)	y = 33.834ln(x) - 5.4713	0.98	P = 0.001

Table 10. Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles after 2 days using biophysical modelling during Spring, Summer and Autumn in Boston and Louth Bay at Site 11 and 12.¹

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
Site 11	Summer Spring Autumn	7.3 ± 2.1^{a} 8.3 ± 4.0^{b} 8.1 ± 4.2^{c}
Site 12	Summer Spring Autumn	4.0 ± 2.1^{d} 3.9 ± 2.1^{d} 3.3 ± 2.2^{e}
ANOVA ² Site Season Site × Season		P < 0.001 P < 0.001 P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior. ² Where significant main effects were detected, post-hoc tests were used to determine differences between means (Tukeys test; P < 0.05). For the significant interaction (Site × Season; P < 0.05), difference in site are compared across all season (one-factor ANOVA, Tukeys test), values without a common superscript are significantly different (^a indicates the highest value; P < 0.05).

Franklin Harbor Disease Management Area

The Franklin Harbor DMA is displayed in Figure 30. This DMA is contained within and bounded by a line commencing at mean high water springs closest to 33° 45' 50.346" South, 136° 55' 54.048" East, then south-easterly to 33° 46' 7.500" South, 136° 56' 3.001" East, then south-easterly to 33° 46' 48.198" South, 136° 56' 38.659" East, then easterly to 33° 46' 49.825" South, 136° 56' 54.625" East, then north-easterly to 33° 46' 23.938" South, 136° 58' 52.396" East, then north-easterly to 33° 46' 13.004" South, 136° 59' 17.293" East, then north-easterly to 33° 44' 38.285" South, 137° 0' 29.178" East, then north-westerly to 33° 44' 31.178" South, 137° 0' 26.147" East, then north-westerly to 33° 43' 43.799" South, 137° 0' 0.000" East, then north-westerly to the location on mean high water springs closest to 33° 43' 29.435" South, 136° 59' 53.178" East, then beginning westerly following the line of mean high water springs to the point of commencement.

The Franklin Harbor DMA is based on the combined maximum hulls from the biophysical modelling of OsHV-1 particles from Site 13 during Spring, Summer and Autumn (Figure 31).

The relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 32 and Table 11. Combined across all seasons, the maximum dispersal distance for 2 days was 7.2 km (90th percentile = 3.1 km) and increased to 70.0 km (90th percentile = 22.4 km) after 20 days (Table 11). The maximum dispersal distance of OsHV-1 particles after 2 days was significantly influenced by season (Autumn > Summer > Spring; P < 0.001) (Table 12).



Figure 30. Disease Management Area for Franklin Harbor. Note that 'aquaculture licences' represent all active aquaculture licence holders as at the date of this report, and may not necessarily represent oyster farms only (see <u>www.aginsight.sa.gov.au/</u>).



Figure 31. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in Franklin Harbor (Site 13; - 33.7018°, 136.9276°).



Figure 32. Maximum dispersal distance and 90th percentile of particles for 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Franklin Harbor at Site 13.

Table 11.	Maximum dispersal distance (km) and 90 th percentile of particles for 2, 4, 7, 14 and
20 days using	g biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in
Franklin Hark	por at Site 13. ¹

				Days					
Site	Season	2	4	7	14	20	Equation (max)	r ² (max)	P value (max)
Site 13	Summer	5.8 (2.8)	11.2 (4.1)	31.6 (6.3)	60.7 (12.4)	81.9 (27.5)	y = 0.0035x ² + 4.1693x - 1.805	1.00	P = 0.004
	Spring	7.2 (2.8)	15.0 (4.5)	32.2 (6.3)	54.9 (9.0)	78.6 (9.3)	y = -0.066x ² + 5.792x - 7.429	1.00	P = 0.004
	Autumn	8.6 (3.7)	12.2 (6.2)	27.3 (8.9)	43.2 (18.4)	49.4 (30.4)	y = -0.109x ² + 4.757x - 2.102	0.99	P = 0.010
	All seasons	7.2 (3.1)	12.8 (4.9)	30.4 (7.2)	53.0 (13.3)	70.0 (22.4)	$y = -0.070x^2 + 5.08x - 3.779$	1.00	P = 0.004

Table 12. Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles after 2 days using biophysical modelling during Spring, Summer and Autumn in Franklin Harbor at Site 13.¹

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
Site 13	Summer	3.4 ± 0.7^{b}
	Sprina	$3.2 \pm 0.8^{\circ}$
	Autumn	3.8 ± 1.3^{a}
ANOVA ²		
Season		P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior.

² Where significant main effects were detected, post-hoc tests were used to determine differences between means (Tukeys test; P < 0.05).

Fitzgerald Bay Disease Management Area

The Fitzgerald Bay DMA is displayed in Figure 33. This DMA is contained within and bounded by a line commencing at mean high water springs closest to 32° 59' 43.440" South, 137° 45' 9.925" East, then southerly to 33° 4' 54.062" South, 137° 45' 15.520" East, then south-easterly to 33° 5' 26.732" South, 137° 46' 52.835" East, then easterly to 33° 5' 28.194" South, 137° 47' 16.624" East, then north-easterly to 33° 5' 8.966" South, 137° 48' 25.628" East, then north-easterly to 33° 3' 52.110" South, 137° 52' 18.563" East, then north-easterly to 33° 2' 23.226" South, 137° 55' 39.058" East, then north-easterly to 33° 2' 12.746" South, 137° 55' 53.706" East, then northeasterly to 33° 2' 0.416" South, 137° 56' 8.732" East, then north-easterly to 33° 1' 40.069" South, 137° 56' 13.632" East, then northerly to 33° 1' 20.071" South, 137° 56' 14.122" East, then northwesterly to 32° 56' 27.132" South, 137° 54' 42.260" East, then north-westerly to 32° 54' 37.678" South, 137° 53' 57.509" East, then north-westerly to 32° 53' 16.699" South, 137° 53' 21.905" East, then north-westerly to 32° 49' 1.488" South, 137° 51' 13.370" East, then north-westerly to 32° 44' 41.111" South, 137° 49' 0.113" East, then north-westerly to 32° 44' 5.222" South, 137° 48' 32.134" East, then south-westerly to the location on mean high water springs closest to 32° 44' 9.096" South, 137° 48' 6.451" East, then beginning south-easterly following the line of mean high water springs to the point of commencement.

The Fitzgerald Bay DMA is based on the combined maximum hulls from the biophysical modelling of OsHV-1 particles from Site 14 during Spring, Summer and Autumn (Figure 34).

The relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 35 and Table 13. Combined across all seasons, the maximum dispersal distance for 2 days was 20.7 km (90th percentile = 12.8 km) and 54.8 km (90th percentile = 36.5 km) after 20 days (Table 13). The maximum dispersal distance of OsHV-1 particles after 2 days was significantly influenced by season (Autumn > Spring > Summer; P < 0.001) (Table 14).



Figure 33. Disease Management Area for Franklin Harbor. Note that 'aquaculture licences' represent all active aquaculture licence holders as at the date of this report, and may not necessarily represent oyster farms only (see <u>www.aginsight.sa.gov.au/</u>).



Figure 34. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in Fitzgerald Bay (Site 14; - 32.8759°, 137.7972°).



Figure 35. Maximum dispersal distance and 90th percentile of particles for 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Fitzgerald Bay at Site 14.

Table 13.	Maximum dispersal distance (km) and 90 th percentile of particles (in parentheses) for
2, 4, 7, 14 an	d 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer
and Autumn	in Fitzgerald Bay at Site 14. ¹

				Days					
Site	Seasons	2	4	7	14	20	Equation (max.)	r² (max)	P value (max)
					40 -		0.000/		
Site 14	Summer	21.4 (9.1)	24.2 (17.4)	31.5 (21.9)	48.5 (27.1)	62.5 (27.1)	y = 2.3394x + 15.639	0.99	P < 0.001
	Spring	20.8́ (16.6)	30.9 (19.5)	54.0 (29.0)	67.1 (54.1)	67.2 [′] (54.1)	y = 22.253ln(x) + 4.998	0.95	P = 0.005
	Autumn	19.9 (12.6)	26.4 (19.8)	32.8 (23.7)	34.3 (28.4)	34.6 (28.4)	y = 6.4653ln(x) + 17.083	0.90	P = 0.014
	All seasons	20.7 (12.8)	27.1 (18.9)	39.4 (24.9)	50.0 (36.5)	54.8 (36.5)	y = 15.503ln(x) + 8.4443	0.99	P = 0.001

Table 14. Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles after 2 days using biophysical modelling during Spring, Summer and Autumn in Fitzgerald Bay at Site 14.¹

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
Site 14	Summer Spring Autumn	8.8 ± 3.7^{b} 9.9 ± 5.0^{a} 10.2 ± 4.0^{a}
ANOVA ² Season		P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior.

² Where significant main effects were detected, post-hoc tests were used to determine differences between means (Tukeys test; P < 0.05).

Eastern Yorke Peninsula Disease Management Area

The Eastern Yorke Peninsula DMA is displayed in Figure 36. This DMA is contained within and bounded by a line commencing at mean high water springs closest to 35° 9' 21.150" South, 137° 41' 6.018" East, then south-easterly to 35° 9' 33.098" South, 137° 41' 19.601" East, then southeasterly to 35° 10' 19.942" South, 137° 42' 27.392" East, then south-easterly to 35° 10' 48.011" South, 137° 43' 17.299" East, then south-easterly to 35° 11' 27.110" South, 137° 45' 14.962" East, then south-easterly to 35° 11' 45.550" South, 137° 46' 25.997" East, then south-easterly to 35° 12' 21.290" South, 137° 49' 22.141" East, then easterly to 35° 12' 21.172" South, 137° 52' 8.180" East, then north-easterly to 35° 12' 15.124" South, 137° 52' 39.835" East, then north-easterly to 35° 11' 49.153" South, 137° 53' 37.738" East, then north-easterly to 35° 11' 5.572" South, 137° 54' 38.498" East, then north-easterly to 35° 7' 42.056" South, 137° 55' 51.701" East, then northeasterly to 34° 57' 45.828" South, 137° 59' 12.872" East, then north-easterly to 34° 57' 7.438" South, 137° 59' 21.516" East, then north-easterly to 34° 56' 26.614" South, 137° 59' 30.131" East, then north-easterly to 34° 40' 44.396" South, 138° 2' 33.752" East, then north-westerly to 34° 37' 44.929" South, 138° 2' 19.482" East, then north-westerly to 34° 34' 54.401" South, 138° 1' 35.969" East, then north-westerly to 34° 24' 3.125" South, 137° 57' 38.153" East, then north-westerly to the location on mean high water springs closest to 34° 23' 25.962" South, 137° 57' 23.861" East, then beginning south-westerly following the line of mean high water springs to the point of commencement.

The Eastern Yorke Peninsula DMA is based on the combined maximum hull from the biophysical modelling of OsHV-1 particles from Site 15, 16, 17 and 18 during Spring, Summer and Autumn (Figure 37-41).

The relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 42 and Table 15. Combined across all sites, the maximum dispersal distance for 2 days was 35.8 km (90th percentile = 22.5 km) and increased to 86.2 km (90th percentile = 73.8 km) after 20 days during summer (Table 15). The maximum dispersal distance of OsHV-1 particles after 2 days was significantly influenced by season (Summer > Spring > Autumn; P < 0.001) and site (Site16 > Site 15 > Site 18 > Site 17; P < 0.001), although there was a significant interaction between these factors (P < 0.001) indicating that particle dispersal patterns for each site depend on the season (Table 16).



Figure 36. Disease Management Area for Eastern Yorke Peninsula. Note that 'aquaculture licences' represent all active aquaculture licence holders as at the date of this report, and may not necessarily represent oyster farms only (see www.aginsight.sa.gov.au/).



Figure 37. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease at Port Vincent on Yorke Peninsula (Site 15; -34.7709°, 137.8915°).



Figure 38. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease at Stansbury on Yorke Peninsula (Site 16; -34.8871°, 137.8460°).



Figure 39. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease at Coobowie on Yorke Peninsula (Site 17; -35.0595°, 137.7444°).



Figure 40. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from Klein Point Jetty on Yorke Peninsula (Site 18; - 34.9611°, 137.7750°).



Figure 41. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Yorke Peninsula at Site 15; (-34.7709°, 137.8915°), 16 (-34.8871°, 137.8460°), 17 (-35.0595°, 137.7444°) and 18 (-34.9611°, 137.7750°).



Figure 42. Maximum dispersal distance and 90th percentile of particles for 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Yorke Peninsula (Site 15, 16, 17 and 18).

		Days					_	_	
Site	Season	2	4	7	14	20	Equation (max.)	r ² (max)	P value (max)
									/
Site 15	Summer	41.7	57.0	62.3	70.3	73.3	$y = 13.229 \ln(x) +$	0.96	P = 0.003
	a .	(33.1) 33.7	(45.2) 49.9	(52.9) 59.3	(59.1) 61.6	(64.0) 65.3	35.365 v = 12.953ln(x) +		5
	Spring	(25.5)	(43.9)	(53.0)	(53.4)	(56.5)	28.929	0.91	P = 0.013
	Autumn	24.7 (14.5)	37.9 (24.2)	46.0 (33.5)	48.4 (42.2)	51.8 (43.0)	y = 11.174ln(x) + 20.185	0.92	P = 0.010
	All	33.4	48.2	55.9	60.1	63.5	y = 12.452ln(x) +	0.94	P = 0.007
	seasons	(24.4)	(37.8)	(46.4)	(51.6)	(54.5)	28.160	0.01	1 - 0.007
0:4+ 40	0	44.1	68.0	73.1	83.9	86.7	$y = 17.53 \ln(x) +$	0.00	D 0.000
Site 16	Summer	(36.1)	(53.1)	(65.0)	(72.1)	(76.9)	37.295	0.93	P = 0.008
	Spring	33.9 (28.1)	60.1 (51.5)	69.6 (61.3)	74.9 (67.8)	75.8 (67.8)	y = 17.250in(x) + 29.537	0.86	P = 0.024
	Autumn	23.9	39.5 (55.6	58.9	64.6	$y = 17.300 \ln(x) +$	0.94	P = 0.006
	All	(15.5) 34.0	(27.9) 55.9	(42.8) 66.1	(50.4) 72.5	(54.4) 75.7	15.078 v = 17.36ln(x) +		D
	seasons	(26.6)	(44.2)	(56.4)	(63.5)	(66.4)	27.303	0.92	P = 0.009
		18.1	30.3	62.6	03.6	96.9	$y = 36.465 \ln(y)$		
Site 17	Summer	(5.4)	(14.0)	(27.7)	(66.8)	(78.2)	8.337	0.99	P = 0.001
	Spring	20.6	32.8	70.1 (20.0)	86.0 (68.0)	94.6 (70.1)	y = 34.359ln(x) -	0.96	P = 0.004
	Autumn	(3.3) 14.5	30.2	(29.9) 40.9	(00.0) 60.3	73.3	$y = 25.015 \ln(x) -$	0.00	P < 0.001
		(6.7) 17.7	(10.9)	(19.4) 57.0	(43.9)	(59.3)	4.501	0.99	F < 0.001
	seasons	(5.8)	(13.6)	(25.7)	(59.6)	(72.2)	y = 31.94011(x) - 6.131	0.99	P < 0.001
Site 18	Summer	39.2 (15.2)	50.0 (29.1)	70.8 (44.3)	83.6 (69.7)	87.8 (76.3)	y = 22.436ln(x) + 22.924	0.98	P = 0.002
	Spring	25.8	52.0	70.6	82.9	85.3	$y = 25.979 \ln(x) +$	0.95	P = 0.005
	opinig	(12.4) 23 1	(25.3) 34 9	(49.2) 42 6	(69.7) 61 4	(77.5) 72 1	13.138 v = 21.086ln(x) +	0100	
	Autumn	(8.9)	(17.5)	(29.2)	(44.0)	(59.5)	6.079	0.98	P = 0.001
	All seasons	29.3 (12.2)	45.6 (24.0)	61.3 (40.9)	76.0 (61.2)	81.7 (71-1)	y = 23.167ln(x) + 14 047	0.99	P < 0.001
	0000010	(12.2)	(←1.0)	(+0.0)	(01.2)	(* 1. 1)	1 1 0 1 1		
All sites	Summer	35.8	53.6 (35 4)	67.2 (47.5)	82.8 (67 0)	86.2 (73.8)	y = 22.415ln(x) + 21.812	0.99	P < 0.001
	Spring	(22.5) 28.5	48.7	67.4	76.3	80.3	$y = 22.635 \ln(x) +$	0.96	P - 0 004
	Spring	(17.8) 21 5	(34.1) 35.6	(48.3) 46.3	(64.7) 57.2	(70.2)	16.512	0.90	i° – 0.004
	Autumn	(11.4)	(20.1)	(31.2)	(45.2)	(54.0)	9.210	1.00	P < 0.001

Table 15. Maximum dispersal distance (km) and 90th percentile of particles (in parentheses) for 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Yorke Peninsula at Site 15, 16, 17 and 18.¹

Table 16.	Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles
after 2 days	using biophysical modelling during Spring, Summer and Autumn in Yorke Peninsula at
Site 15, 16,	17 and 18.1

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
Site 15	Summer Spring Autumn	23.9 ± 7.0^{b} 14.6 ± 8.1 ^d 10.1 ± 4.4 ^f
Site 16	Summer Spring Autumn	26.7 ± 7.0ª 15.6 ± 9.1 ^d 10.7 ± 5.0 ^{ef}
Site 17	Summer Spring Autumn	8.1 ± 2.7^{h} 7.4 ± 3.4 ^h 7.2 ± 2.8 ^h
Site 18	Summer Spring Autumn	17.1 ± 5.5 ^c 11.3 + 5.8 ^e 8.3 ± 3.3g ^h
ANOVA ² Site Season Site × Season		P < 0.001 P < 0.001 P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior.

² Where significant main effects were detected, post-hoc tests were used to determine differences between means (Tukeys test; P < 0.05). For the significant interaction (Site × Season; P < 0.05), difference in site are compared across all season (one-factor ANOVA, Tukeys test), values without a common superscript are significantly different (^a indicates the highest value; P < 0.05).

Outer Harbor and West Beach Disease Management Area

The Outer Harbor and West Beach DMA is displayed in Figure 43. This DMA is contained within and bounded by a line commencing at mean high water springs closest to 34° 37' 15.600" South, 138° 25' 19.369" East, then south-westerly to 34° 38' 24.968" South, 138° 23' 28.068" East, then south-westerly to 34° 38' 30.102" South, 138° 23' 24.119" East, then south-westerly to 34° 38' 56.155" South, 138° 23' 7.242" East, then south-easterly to 35° 7' 15.395" South, 138° 27' 33.880" East, then easterly to the location on mean high water springs closest to 35° 7' 14.387" South, 138° 28' 2.510" East, then beginning northerly following the line of mean high water springs to the point of commencement.

The Outer Harbor and West Beach DMA is based on the combined maximum hull from the biophysical modelling of OsHV-1 particles from Site 19 and 20 during Spring, Summer and Autumn (Figure 44 and 45).

The relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 46 and Table 17. Combined across all sites, the maximum dispersal distance for 2 days was 20.3 km (90th percentile = 14.1 km) and increased to 71.7 km (90th percentile = 66.4 km) after 20 days during summer (Table 17). The maximum dispersal distance of OsHV-1 particles after 2 days was significantly influenced by season and site (Site 19: Summer > Spring > Autumn, P < 0.001; Site 20: Summer > Autumn > Spring, P < 0.001) (Table 18). A statistically significant interaction (P < 0.001) indicated that particle dispersal patterns are differ at each site across seasons (Table 18).



Figure 43. Disease Management Area for Outer Harbor and West Beach.



Figure 44. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from an oyster hatchery at West Beach (Site 19; - 34.9536°, 138.5042°).



Figure 45. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from the Port River mouth that contains an feral oyster population (Site 20; -34.7786°, 138.4808°).



Figure 46. Maximum dispersal distance and 90th percentile of particles for 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in West Beach and Port River mouth at Site 19 and 20.

Table 17.	Maximum dispersal distance (km) and 90 th percentile of particles (in parentheses) for
2, 4, 7, 14 an	d 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer
and Autumn	in West Beach and Port River mouth at Site 19 and 20. ¹

				Days					
Site	Season	2	4	7	14	20	Equation (max)	r² (max)	P value (max)
Site 19	Summer	23.2 (17.5)	37.1 (26.3)	52.4 (41.9)	71.6 (60.2)	79.9 (75.2)	y = 25.220ln(x) + 4.107	1.00	P < 0.001
	Spring	17.7 (14.0)	31.3 (23.9)	38.2 (31.1)	48.8 (40.3)	50.8 (43.1)	$y = 14.444 \ln(x) + 9.454$	0.98	P = 0.001
	Autumn	19.1 (8.3)	24.9 (13.5)	25.9 (14.2)	34.0 (24.1)	46.7 (24.1)	y = 1.404x + 16.898	0.97	P = 0.003
	All seasons	20.0 (13.3)	31.1 (21.3)	38.8 (29.1)	51.5 (41.6)	59.1 (47.5)	y = 16.764ln(x) + 7.7084	1.00	P < 0.001
Site 20	Summer	17.4 (10.7)	30.1 (19.7)	42.5 (30.4)	60.1 (45.1)	63.5 (57.6)	y = 20.973ln(x) + 2.21	0.99	P < 0.001
	Spring	14.5 (8.2)	28.1 (16.2)	33.4 (25.1)	42.9 (35.5)	43.6 (36.0)	y = 12.665ln(x) + 8.025	0.97	P = 0.003
	Autumn	19.3 (12.6)	29.5 (21.3)	34.0 (25.6)	47.6 (35.3)	47.6 (35.3)	y = 12.935ln(x) + 10.611	0.97	P = 0.002
	All seasons	17.1 (10.5)	29.2 (19.1)	36.6 (27.0)	50.2 (38.6)	51.6 (43.0)	y = 15.524ln(x) + 6.9484	0.99	P = 0.001
All sites	Summer	20.3 (14.1)	33.6 (23.0)	47.5 (36.2)	65.9 (52.7)	71.7 (66.4)	y = 23.097ln(x) + 3.158	1.00	P < 0.001
	Spring	16.1 (11.1)	29.7 (20.0)	35.8 (28.1)	45.8 (37.9)	47.2 (39.6)	y = 13.555ln(x) + 8.7393	0.98	P = 0.002
	Autumn	19.2 (10.5)	27.2 (17.4)	29.9 (19.9)	40.8 (29.7)	47.1 (29.7)	y = 11.782ln(x) + 10.088	0.97	P = 0.002

Table 18. Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles after 2 days using biophysical modelling during Spring, Summer and Autumn in West Beach and Port River mouth at Site 19 and 20.¹

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
	_	
Site 19	Summer	11.7 ± 5.0 ^a
	Spring	7.7 ± 4.0^{b}
	Autumn	$6.9 \pm 3.0^{\circ}$
Site 20	Summer	7.8 ± 3.8^{b}
	Spring	5.5 ± 2.7^{d}
	Autumn	6.4 ± 4.4^{c}
ANOVA		
Site		P < 0.001
Season		P < 0.001
Site × Season ¹		P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior. ² Where significant main effects were detected, post-hoc tests were used to determine differences between means (Tukeys test; P < 0.05). For the significant interaction (Site × Season; P < 0.05), difference in site are compared across all season (one-factor ANOVA, Tukeys test), values without a common superscript are significantly different (^a indicates the highest value; P < 0.05).

Kangaroo Island Disease Management Area

The Kangaroo Island DMA is displayed in Figure 47. This DMA is contained within and bounded by a line commencing at mean high water springs closest to 35° 35' 34.055" South, 137° 25' 48.364" East, then north-easterly to 35° 33' 57.733" South, 137° 26' 47.890" East, then northerly to 35° 32' 36.481" South, 137° 26' 52.991" East, then north-easterly to 35° 28' 35.803" South, 137° 27' 49.871" East, then north-easterly to 35° 26' 28.230" South, 137° 29' 27.215" East, then north-easterly to 35° 25' 46.420" South, 137° 32' 39.322" East, then south-easterly to 35° 27' 11.786" South, 137° 35' 37.651" East, then south-easterly to 35° 47' 47.278" South, 138° 13' 43.043" East, then south-easterly to 35° 49' 33.434" South, 138° 14' 7.411" East, then south-westerly to 35° 49' 40.994" South, 138° 13' 4.030" East, then south-westerly to 35° 50' 33.000" South, 138° 8' 16.501" East, then south-westerly to the location on mean high water springs closest to 35° 50' 35.556" South, 138° 8' 2.432" East, then beginning northerly following the line of mean high water springs to the point of commencement.

The Kangaroo Island DMA is based on the combined maximum hull from the biophysical modelling of OsHV-1 particles from Site 21, 22 and 23 during Spring, Summer and Autumn (Figure 48-51).

The relationships between maximum dispersal distance and 90th percentile of OsHV-1 particles and time (2, 4, 7, 14 and 20 days) during Spring, Summer and Autumn is displayed in Figure 52 and Table 19. Combined across all sites, the maximum dispersal distance for 2 days was 30.4 km (90th percentile = 5.4 km) during Autumn, while the maximum dispersal distance for 20 days was 152.8 km (90th percentile = 100.5 km) after 20 days during summer (Table 19). The maximum dispersal distance of OsHV-1 particles after 2 days was significantly influenced by season (Autumn > Summer > Spring; P < 0.001) and site (Site 22 > Site 21 > Site 23; P < 0.001), although there was a significant interaction between these factors (P < 0.001) indicating that particle dispersal patterns for each site depend on the season (Table 20).



Figure 47. Disease Management Area for Kangaroo Island. Note that 'aquaculture licences' represent all active aquaculture licence holders as at the date of this report, and may not necessarily represent oyster farms only (see www.aginsight.sa.gov.au/).



Figure 48. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in Kingscote on Kangaroo Island (Site 21; -35.7372°, 137.6862°).



Figure 49. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from Kingscote jetty that contains feral oyster populations on Kangaroo Island (Site 22; -35.6551°, 137.6450°).



Figure 50. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn from a marine lease in American River on Kangaroo Island (Site 23; -35.7670°, 137.7969°).



Figure 51. Biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn on Kangaroo Island at Site 21 (35.7372°, 137.6862°), 22 (-35.6551°, 137.6450°) and 23 (-35.7670°, 137.7969°).


Figure 52. Maximum dispersal distance and 90th percentile of particles for 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Kangaroo Island at Site 21, 22 and 23.

				Days					
		2	4	7	14	20	Equation (max)	r² (max)	P value (max)
		10.0	20.0	547	00.0	115 0	y = 7.007y		
Site 21	Summer	(4.9)	(9.0)	(24.8)	(67.9)	(77.4)	y = 7.097X - 0.474	0.99	P = 0.001
	Spring	12.2 (5.3)	27.5 (12.6)	53.6 (21.5)	101.7 (48.6)	124.9 (64.2)	y = 6.383x + 4.000	0.98	P = 0.001
	Autumn	23.2 (3.6)	39.4 ´ (11.9)	39.7 (18.0)	54.9 (28.5)	65.0 (45-4)	y = 16.830ln(x) + 11 942	0.951	P = 0.005
	All seasons	(0.0) 15.5 (4.6)	32.3 (11.2)	(10.0) 49.3 (21.4)	82.2 (48.4)	(40.4) 111.9 (62.3)	y = 5.183x + 9.507	0.99	P < 0.001
Site 22	Summer	27.8	44.6	66.0	114.1	174.8	y = 7.944x +	0.99	P < 0.001
	Spring	(10.3) 20.9 (7.6)	43.6 (16.1)	(40.7) 60.0 (41.4)	(64.4) 96.7 (69.7)	(133.4) 112.2 (74.1)	$y = 40.099 \ln(x) - 10.797$	0.99	P = 0.001
	Autumn	56.2 (10.1)	96.0 (39.9)	104.5 (59.0)	118.7 (65.5)	118.7 (71.8)	y = 25.847ln(x) + 48.856	0.88	P = 0.019
	All seasons	35.0 (9.5)	61.4 (27.5)	76.8 (47.0)	109.8 (73.2)	135.2 (93.1)	y = 42.07ln(x) + 2.3657	0.98	P = 0.001
Site 23	Summer	12.9	34.2	56.5	97.9	137.8	y = 6.700x +	0.99	P < 0.001
	Spring	(2.6) 7.8 (2.4)	(5.9) 27.8 (4.2)	(19.0) 60.0 (10.7)	(62.3) 92.8 (48.5)	(90.9) 110.3 (52.9)	4.905 y = 45.885ln(x) - 28.894	0.99	P < 0.001
	Autumn	(2.1) 11.8 (2.4)	23.0	50.3 (9.4)	(1010) 55.2 (22.9)	(38 7)	$y = 26.717 \ln(x) - 8.519$	0.95	P = 0.005
	All seasons	10.8 (2.4)	28.3 (5.5)	55.6 (13.0)	(44.5)	107.8 (60.8)	y = 41.707ln(x) – 23.666	0.98	P = 0.001
All sites	Summer	17.2 (6.1)	36.3 (13.8)	59.1 (28.1)	100.6 (71.6)	152.8 (100.5)	y = 7.2472x + 5.075	0.99	P < 0.001
	Spring	13.6 (5.1)	33.0 (11.0)	57.9 (24.5)	97.1 (55.6)	115.8 (63.8)	y = 45.540ln(x) - 24.507	0.98	P = 0.001
	Autumn	30.4 (5.4)	52.8 [´] (19.4)	64.8 (28.8)	76.3 (38.9)	86.3 (51.9)	y = 23.131ln(x) + 17.427	0.98	P = 0.001

Table 19. Maximum dispersal distance (km) and 90th percentile of particles (in parentheses) for 2, 4, 7, 14 and 20 days using biophysical modelling of OsHV-1 particles during Spring, Summer and Autumn in Kangaroo Island at Site 21, 22 and 23.¹

¹ Data is for 2 days (96,200 particles from 481 releases), 4 days (86,600 particles from 433 releases), 7 days (72,200 particles from 361 releases), 14 days (38,600 particles from 193 releases) and 20 days (9,800 particles from 49 releases). The model was run for 22 days with hourly releases of 200 particles, while data analyses was based on the furthest any of the 200 particles can travel.

Table 20. Statistical analyses of mean (N=481) dispersal distance (km) of OsHV-1 particles after 2 days using biophysical modelling during Spring, Summer and Autumn in Kangaroo Island at Site 21, 22 and 23.¹

Site	Season	Mean maximum dispersal distance of OsHV-1 particles after 2 days
Site 21	Summer	4.9 ± 1.7 ^{de}
	Spring	5.3 ± 2.3 ^d
	Autumn	4.2 ± 3.3^{e}
Site 22	Summer	12.6 ± 5.5^{a}
	Spring	$8.0 \pm 3.8^{\circ}$
	Autumn	9.7 ± 7.1^{b}
Site 23	Summer	4.2 ± 1.6^{e}
	Spring	3.3 ± 0.8^{f}
	Autumn	3.3 ± 1.5^{f}
ANOVA ²		
Site		P < 0.001
Season		P < 0.001
Site × Season		P < 0.001

¹ The model was run for 22 days with hourly releases of 200 particles. Data analyses was based maximum dispersal of 200 particles at the top of the hour (e.g. 12:00:00, 1:00:00) that were released two days prior.

² Where significant main effects were detected, post-hoc tests were used to determine differences between means (Tukeys test; P < 0.05). For the significant interaction (Site × Season; P < 0.05), difference in site are compared across all season (one-factor ANOVA, Tukeys test), values without a common superscript are significantly different (^a indicates the highest value; P < 0.05).

Discussion

Modelled viral dispersal distances (Objective 1)

The predicted trajectory and dispersal distance of OsHV-1 particles from 23 sites in oyster growing regions throughout South Australia were modelled and analysed during Autumn 2018, Spring 2018 and Summer 2019. Importantly, a biophysical model (administered by SARDI and Bureau of Meteorology) based on the e-SA Marine system (www.pir.sa.gov.au/research/esa_marine) is now developed and operational for each oyster growing region in South Australia and can be used in real time to provide predictive capability up to 3 days for future emergency responses. Furthermore, the model is flexible, and can be used to track other passive particles (other pathogens, harmful algae blooms, chemicals, toxins or oil spills) or motile particles (e.g. parasites or larvae) given appropriate biological inputs and assumptions.

For all 23 sites and seasons, the average maximum dispersal distance for live OsHV-1 virus (2 day lifespan) based on modelled data from this project was 20.2 km (moderate risk), while the 90th percentile of particles was 11.7 km (high risk). The average for the 90th percentile is close to the assumed disease spread distance of 10 km used by international and national policy makers in the absence of geographically specific hydrodynamic information.

An epidemiological unit is defined as a group of animals that share approximately the same risk of exposure to a disease agent, within a location (OIE, 2013). Prior to this project, an epidemiological unit for disease management and response for OsHV-1 assumed a distance of ~10 km. For OsHV-1 (assumed 2 day survival time), this project has demonstrated that in the absence of site specific hydrodynamic information, DMAs could be based on 12 km for high risk management and 20 km for moderate risk management. However, due to the statistically significant interactions between site and season data in the current project, we recommended that site specific hydrodynamic conditions are considered and investigated.

Particle dispersal trajectory and distance significantly varied between regions, sites and seasons. The maximum dispersal distance live OsHV-1 virus (2 day lifespan) varied from 5.2 km (90th percentile = 1.9 km) in Proper Bay (Port Lincoln) during Summer up to 44.1 km (90th percentile = 36.1 km) in Stansbury (Yorke Peninsula) during Summer. The maximum distance viral DNA by 20 days varied from 7.8 km (90th percentile = 7.5 km) at Thevenard wharf (Denial Bay) during spring to 310 km (90th percentile =297.7 km) from Coffin Bay during summer. In comparison, Pande et al. (2015) reported dispersal distances of at least 15 km over 1 day using a biophysical particle tracking model.

OsHV-1 is suggested to disperse in aggregated particles (e.g. oyster faeces, sloughed oyster tissue cells, eggs, mucus from an infected oyster), which could be protective increasing viral survival time in situ (Paul-Pont et al., 2013; Evans et al., 2014; Whittington et al., 2015; Whittington et al., 2018). However, virus dilution, viral decay and reduced viral infectivity may reduce the survival and infectivity time in water (Garver et al., 2013). For example, once shed into the marine environment, the abundance of Infectious Hematopoietic Necrosis Virus (IHNV) virions is modulated by sunlight and the growth of natural biota present in the seawater. IHNV virions decayed very slowly in sterilized seawater while rates as high as k = 4.37/day were observed in natural seawater. Decay rates were further accelerated when exposed to sunlight with virus infectivity reduced by six orders of magnitude within 3 hours of full sunlight exposure (Garver et al., 2013). Given

the above, and based on current research (Martenot et al., 2015; Hick et al., 2016), the assumed 2-day survival of OsHV-1 may be a sufficient assumption. Nevertheless, if survival or infective time in water are found to be different in the future, this study provides data and regression analyses to determine dispersal distance for up to 22 days.

Infective distances for other pathogens have been reported previously, and are largely based on patterns of infection during real outbreaks. Laferty and Ben-Horin (2013) detected dilute DNA of the infectious Withering Syndrome Rickettsia-Like Organism throughout the water column at almost 20 km from a Californian abalone aquaculture facility. For Viral Haemorrhagic Septicaemia (VHS) on a turbot farm in Scotland, a conservative distance at which virus concentration was determined to fall below 1 infectious virion / m³ was 20 km, which was used to determine at risk farms (Munro 1996). Jarp and Karlsen (1997), and McClure et al. (2005) determined that a distance of up to 5 km between farms posed a high risk of infection with Infectious Salmon Anaemia (ISA). A 5 km restricted zone and a 10 km observation zone was enforced by the Norwegian Food Safety Authority around ISA-infected farms (Aldrin et al. 2011). Needham (1995) reported that furunculosis spread between salmon farms up to 10 km away during outbreaks in Canada.

Dispersal distances can differ for invertebrate parasites. For example, Gargan et al. (2007) found that the highest numbers of salmon lice were recorded at sites less than 20 km from salmon farms, and beyond 30 km low levels were recorded. Modelled sea lice dispersal recently shown to be on average 10.8 km in Spring and 18.9 km in winter (Samsing et al 2017), although the 90th percentile of sea lice reached 118 km. In Fitzgerald Bay, Chambers and Ernst (2005) investigated the effect of tidal currents on the dispersal of skin fluke (*Benedenia seriolae*) eggs and on the infection rates of *B. seriolae* on sentinel yellowtail kingfish (*Seriola lalandi*) near a yellowtail kingfish farm. The authors reported dispersal distance of 4 km after 40.7 hours and 8 km after 6.3 days, with farms down-current at greatest risk. In contrast, for passive viral particles, the current project found the maximum dispersal distance across all seasons for Fitzgerald Bay for 2 days was 20.7 km (90th percentile = 12.8 km) and after 7 days was 39.4 km (90th percentile = 24.9 km).

Non-viable OsHV-1 DNA (dead virus) can be detected in seawater up to 22 days at 4°C and 12 days at 20°C from macerated infected larvae under laboratory conditions (Vigneron et al., 2004). In our study, the long distances that viral DNA may be able to travel (up to 310 km from Coffin Bay after 20 days during summer) may hold implications for both early detection surveillance and emergency disease response. For example, PCR techniques cannot currently distinguish between live and dead virus, so positive PCR test results in the absence of oyster mortalities may not necessarily indicate subclinical or latent infection, but should be further investigated to rule out DNA contamination from a distant outbreak. In that scenario further tracing, surveillance and use of other diagnostic tools to detect viable vs non-viable virus would need to be considered. Furthermore, dispersal of non-viable viral DNA may even provide some insight into the survival and immunity of oysters (Green and Speck, 2018) within and near POMS outbreaks.

Key assumptions and limitations of this project to note include:

- Model outputs are for hydrodynamic conditions during Autumn 2018, Spring 2018 and Summer 2019
- Modelled data are for 23 specific sites located in South Australia
- OsHV-1 survival time for infection was assumed to be 2 days
- Every modelled particle was assumed to represent 10,000's of aggregated OsHV-1 virions (infective dose) and were considered infectious to a new adjacent oyster population for up to 2 days

- Maximum dispersal distance of any one particle was used to define DMAs as per Pande et al (2015), which is conservative as suggested by the national emergency disease response plan for OsHV-1 (Department of Agriculture, 2015)
- Populations were defined as known farmed and feral Pacific oyster populations (primary host species). Other feral Pacific oyster populations, other susceptible hosts or environmental reservoirs identified in the future may influence DMA boundaries
- OsHV-1 DNA can be detected for 22 days (although temperature dependent).

Robust justification of these assumptions are outlined in the Methods section. Furthermore, the modelled data are presented in this report in such a way to allow dispersal distances to be determined for any passive particle up to 22 days. If survival or infective time changes with new knowledge in the future, it can be inferred from the maps, graphs and tables provided in this report.

Disease Management Areas (Objective 2)

For the purpose of future surveillance and emergency disease response, OsHV-1 DMAs for South Australia's oyster industry are now updated from Figure 1 to Figure 4. This was based on two day particle dispersal polygons, across all seasons and sites, with overlapping polygons (which indicate medium and high risk connectivity) defining individual DMAs. Dispersal polygons were based on the maximum distance any single particle travelled during 481 simulations (using data-assimilating model output for 2018 and 2019), which provides a conservative and scientifically robust level of management.

Feral Pacific oyster populations are a disease risk that potentially link DMAs. Unlike other jurisdictions, South Australia has few feral oyster populations. Based on previous feral oyster pest surveys (conducted in 2009 and 2010), and the surveys conducted during the 2018 POMS disease response, there are five known significant wild pacific oyster populations identified in South Australia; Port Adelaide, Ceduna, Coffin Bay, Kangaroo Island and Yorke Peninsula (Port Vincent). This project considered farmed oyster sites (marine and land-based hatcheries), known feral pacific oyster populations and hypothetical feral oyster populations. It is worth noting that if in the future a feral oyster population is observed in Boston bay then the Proper Bay and Boston / Louth bay DMAs should be merged (Figure 21).

Risk-based surveillance (Objective 3)

Disease surveillance programs that aim for early detection generally bias sampling towards high risk areas to increase the chance of detection, if a pathogen is present. For example, PIRSA's current OsHV-1 early detection surveillance program biases sampling of oysters to warmer months of the year (>16°C), sites close to ports / harbours, small farmed oysters (<15mm size) and also includes feral oysters.

Prevailing currents and particle dispersal can also be used to bias sampling towards high risk areas. For example, the Yorke Peninsula DMA consists of three oyster farming areas: Port Vincent (north), Stansbury (middle) and Coobowie (south) as well as a known small feral oyster population at Klein Point Jetty in the south. The prevailing hydrodynamics is in a northerly direction such that if Klein Point jetty became infected then a plume of viral particles would be expected throughout the Yorke Peninsula DMA, and it would be likely that Stansbury and Port Vincent would become infected soon after (see Figure 40). However, if Port Vincent became infected the viral plume would unlikely be detected in the more southern areas based on 2018/19 data (see Figure 37). The risk of spread to more

southern sites from Port Vincent is moderate in accordance with Figure 3 (page 18), and the rate of spread would be slower. So in either scenario, if OsHV-1 is detected using PCR in the Yorke Peninsula DMA, the greatest chance of detection would be in the more northern farming regions. Therefore early detection surveillance could bias sampling to oyster populations in the north (Figure 53).

During this project in early 2019, the biophysical model for Boston Bay was used to provide real-time monitoring of a harmful algae bloom (*Karenia mikimotoi*) that was threating fisheries and aquaculture sectors (southern bluefin tuna, yellowtail kingfish and abalone) around Port Lincoln. SARDI oceanographers provided regular (daily) updates of the algae blooms predicted movement for up to 3 days in advance. That information was used to determine where to sample the bloom (surveillance) and for emergency response planning (e.g. movement of sea-cages). Fortunately, the harmful algae bloom did not reach critical levels to initiate an emergency response, but both government and industry were well prepared. No fish mortalities in the Boston Bay area occurred.



Figure 53. The Yorke Peninsula Disease Management Area (DMA) (left), and an example of sampling bias based on dispersal trajectories for early detection surveillance where 150 oyster samples are to be collected from the DMA (assumed surveillance design of 95% confidence of detecting the virus if it were at least 2% prevalent) (right).

Conclusion

Biophysical modelling is a useful tool for disease surveillance, disease management and for use during emergency disease response. Pathogen (particle) dispersal distance, and trajectory, through water during a disease outbreak is an important factor for epidemiology and emergency disease response. Particle dispersal is significantly influenced by the unique hydrodynamics at each geographical location and season.

New maps and descriptions of OsHV-1 DMAs for South Australia's oyster industry have been created based on two day particle dispersal across all seasons and sites (see Figure 4 and Appendix 1). From the methods used, this provides a conservative and scientifically robust level of disease management for future emergency responses. The outputs of this project can also be used to improve early detection surveillance in the future where sampling strategies can target specific hydrodynamics in each region and season.

The biophysical model is now developed for each oyster growing region in South Australia and can be used in real time to provide predictive capability up to 3 days for future emergency responses to POMS. The model is flexible enough to track other passive particles (other pathogens, harmful algae blooms, chemicals, toxins or oil spills) or even motile particles (e.g. parasites or larvae) given appropriate biological inputs and assumptions.

Implications

Outputs and outcomes from this project are already being used by both government and industry for POMS prevention, preparedness and disease management strategies.

The detection of POMS in the Port River put at risk the States \$40 million / year oyster farming industry, which contributes approximately \$74.1 Million to the State's economy (approximately 67% generated in regional South Australia) and directly employs an estimated 418 FTE (Econsearch, 2017). The impact of POMS in South Australia is predicted to be significant for regional communities (which includes impacts to economy, people, public administration and social settings), particularly for small regional towns which heavily rely on the oyster industry.

These outputs and outcomes will significantly enhance future rapid emergency disease responses if POMS were to be detected in an oyster growing region, where the aim would be to contain the virus and reduce the risks of spread to other geographical areas

This project has also demonstrated how pathogens can spread through marine waters, and how a biophysical particle tracking model can enhance disease management and response. These methods may be of importance to other jurisdictions and elsewhere in the world.

Further to the above, during 2019, the biophysical particle tracking model was used by PIRSA and industry to assist in the monitoring and emergency preparedness for a harmful algae bloom (*Karenia mikimotoi*) detected in the Boston Bay area. Information was being used to monitor the bloom in the right locations, and if trigger levels were reached livestock could have been moved or isolated from the bloom area. Fortunately algae concentrations did not reach trigger levels. Located in this area is the Australian southern bluefin tuna

industry (worth \$130 million), the finfish industry (worth \$18 million), abalone aquaculture (worth \$11.4 million) and the mussel industry (worth \$3 million) (www.pir.sa.gov.au/___data/assets/pdf_file/0003/297372/ZONING_IN_Aquaculture_Report_2015-16_optimised.pdf)

Recommendations and Further Development

While this biophysical model provides high confidence in passive particle dispersal trajectories and distances, biological validation (see Figure 2) under natural in situ conditions would be a useful next step (e.g. eDNA survey of adjacent waters during a POMS outbreak). Furthermore, while validated hydrodynamic data are available for most sites, collection of oceanographic data (e.g. currents, temperature, sea level) in remaining locations, as well as over the long term, would improve future forecasts and help further validate particle tracking.

Paul-Pont et al. (2013), Evans et al. (2014) and Whittington et al. (2015, 2018) have suggested that OsHV-1 appears to disperse in aggregated particles (e.g. oyster faeces, sloughed oyster tissue cells, eggs, mucus from an infected oyster), which could be protective increasing viral survival time in situ. However, virus dilution, viral decay, reduced viral infectivity etc may reduce survival time in water (Garver et al., 2013). In-situ research would provide more accurate estimates of virus survival in the marine environment, and this biophysical model would greatly benefit that area of work.

We recommend that a biophysical particle tracking model is developed for other important aquaculture regions, including interstate and overseas, to improve early detection surveillance and emergency disease response.

Extension and Adoption

Project updates, outputs and outcomes have been presented at both industry and scientific conferences, including:

- the Eighth International Symposium on Aquatic Animal Health (ISAAH) at Prince Edward Island, Canada on the 2-6 September 2018 <u>https://isaah2018.files.wordpress.com/2018/08/8-30-18-oral-presentationssearchable-pdf-final1.pdf</u>
- South Australian Oyster Growers Association annual seminar, Streaky Bay, 22-23 August 2019.
- FRDC Aquatic Animal Health and Biosecurity conference, July 2019: <u>https://events.csiro.au/Events/2019/February/27/5th-FRDC-Conference-on-Aquatic-Animal-Health-Biosecurity</u>

This project has also been communicated through the PIRSA / SAOGA POMS working group which meets regularly.

Outputs and outcomes are being adopted by government (PIRSA) and industry for the purpose of early detection surveillance and POMS emergency management, and have already been used for monitoring and response preparedness during a harmful algae bloom in Boston Bay during 2019. In addition, PIRSA's emergency management plan is being reviewed to incorporate this new knowledge and information, including the new DMAs.

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Project Materials Developed

This project developed a biophysical particle tracking model for regions around the South Australian coastline. The model is based on the e-SA marine system (<u>https://pir.sa.gov.au/research/esa_marine</u>), is administered by SARDI Aquatic Sciences and the Bureau of Meteorology (BOM) and can be accessed by contacting SARDI using the contact details on the above website.

Output files from the project also included movie files for all 23 sites and each season (Spring, Summer, Autumn) which are available upon request. Movie files may be accessible on the FRDC website.

Appendices

Appendix 1 (PIRSA file reference A4194664) of this report provides the description of each Disease Management Area for POMS in South Australia. This information is provided in a way that it can be used for legislative requirements during an emergency response (e.g. livestock movement restrictions during a suspected or confirmed POMS outbreak).