

# Didymo National Delimiting Survey

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# 1 Executive Summary

- 1.1 In November 2007, MAF and the didymo regional partner groups conducted a national delimiting survey to monitor the spread of didymo. In total, samples were analysed from 145 sites, including 74 North Island sites and 71 South Island sites. Ten positive or suspect positive new sites were found, all in the South Island.
- 1.2 The majority of the ten South Island new sites are very close to other waterways that already are infected with didymo. Typically they are geographically close to infected sites, downstream, as is the case with the Clarence River in North Canterbury, or directly connected to a waterway that is infected with didymo, as is the case with the Rakaia River in Canterbury.
- 1.3 The survey followed a different model compared to earlier delimiting surveys. Previous surveys had been contracted to an external organisation to coordinate sample collection and analysis. This November survey instead had significant input from the regional partner groups, and used a web based database to coordinate sample collection and analysis. The model used for this survey supports a transition from MAF quarterly national surveys to partner driven, regionally based surveying.

## 2 Purpose

- 2.1 The purpose of the national delimiting surveys is to monitor the spread of didymo to waterways where it has not previously been found. This survey is the final didymo national delimiting survey commissioned by MAF.
- 2.2 The primary aim of the delimiting surveys is to determine how widely didymo has spread into new waterways in the South Island, and to detect if didymo is present in the North Island.
- 2.3 The aim of this November survey was to involve the didymo partner groups, especially North Island partners, so that they can become more familiar with the tools and processes for monitoring didymo.

## 3 Approach

- 3.1 Previously, didymo national delimiting surveys have been contracted to a single provider to coordinate, collect and analyse samples and report results. This survey was instead coordinated by MAF, working with the partner groups and laboratories. The survey also used a new database system to track and record sample results, and a new DNA detection process to analyse North Island samples.
- 3.2 The Didymo Samples Database (released in September 2007) was central to the sample collection and sample analysis processing because it enabled information to be accurately tracked from collection through to final analysis. The database had only been available for a few months prior to the survey, meaning that regional partners and the two laboratories did not have much time to become familiar with using it.
- 3.3 This survey was the first time that the DNA process had been used on such a wide scale. The DNA process was used for all North Island sampling, with sampling kits provided by the University of Waikato.

### 3.4 PARTNER INPUT

- 3.4.1 The approach taken for this survey relied heavily on the regionally based partner groups to coordinate sample collection in each region. This put considerable responsibility on the regional partner coordinators who, in many instances, were working with teams of sample collectors from several different organisations. The commitment of the regional coordinators and cooperation evident between the partner groups meant the sample collection worked well.
- 3.4.2 Sampling sites were initially identified by the North and South Island Didymo Long Term Management Operations Managers (MAF). The sites were then discussed and confirmed with regional partner groups. Sites were selected on a combination of risk, accessibility, value and suitability for didymo to establish.
- 3.4.3 Once the sites were confirmed, the partner groups were asked to submit a cost breakdown to MAF for cost recovery. Some regions and groups did not seek to recover some or all of their costs. In some regions this was because the delimiting survey sites

have been incorporated into ongoing regular monitoring programmes. Just one site needed helicopter access.

- 3.4.4 During October, North Island sample collectors were invited to attend a sample collection training day at the University of Waikato, Hamilton. The training was to show sample collectors the processes required to collect samples for DNA analysis. The training day was well attended, with generally positive feedback, including attendees finding the day a good opportunity for meeting people from other regions. At the training, the first of the DNA sample collection kits were distributed.
- 3.4.5 Matt Dale, Water Resource Scientist from Otago Regional Council, provided field training for West Coast Regional Council staff.

### **3.5 DIDYMO SAMPLES DATABASE**

- 3.5.1 In September 2007, the Didymo Samples Database went into operation. The purpose of the database is to provide a central repository for results of didymo sampling sites (and the samples taken at each site) that can be accessed using the Internet.
- 3.5.2 Prior to the database, regional partner groups (including MAF and laboratories) individually kept records of sampling sites and results in each region, often on Excel spreadsheets. The problem with this was that sharing sampling information was difficult and prone to information being missed. The centralised database has significantly improved information management.
- 3.5.3 For the survey, the database provided the central information channel between sample collectors and the laboratories. Sample collectors were asked to enter sample collection information into the database, print off a sample collection form and send the form with the sample to the laboratory. The laboratory was then able to easily and accurately complete the sample record in the database by using the unique site identification number from the printed sample collection form.
- 3.5.4 Printing the sample collection form and sending it with each sample effectively minimises sample results from being mixed up with the wrong waterway during the analysis and result recording process. It was a very successful process and it is expected that all partner groups and laboratories will continue to use it for future sample collection and analysis processing.
- 3.5.5 The database is password protected. Regional partner groups are provided with access that allows them to add, delete and edit all records entered by their individual organisation. The laboratories have access that allows them to change and edit records that have been entered by other organisations. In this way the laboratories are able to complete the result section of each sample collection record. Read only access is available to anyone else.

### **3.6 NIWA, MICROSCOPIC ANALYSIS**

- 3.6.1 NIWA's Christchurch laboratory completed all the sample analysis for the South Island. In total, samples from 71 sites were analysed with results entered directly into the Didymo Samples Database.

- 3.6.2 All samples were analysed microscopically using the microscopic analysis protocol (Protocol for microscopic analysis samples for the detection of *Didymosphenia geminata*). The protocol can be found on the Biosecurity New Zealand website at the following address <http://www.biosecurity.govt.nz/pest-and-disease-response/pests-and-diseases-watchlist/didymosphenia-geminata/protocols>
- 3.6.3 NIWA provided two sampling kits to each regional partner group.
- 3.6.4 The DNA process was not considered to be the most efficient approach for the South Island because of its high sensitivity and the wide spread of didymo throughout many South Island catchments.

### 3.7 UNIVERSITY OF WAIKATO, DNA DETECTION

- 3.7.1 Samples collected in the North Island were analysed by the University of Waikato using the DNA detection process. Samples from 74 sites were analysed.
- 3.7.2 The DNA process was chosen to be used for the North Island sampling for several reasons. These include:
- Highly sensitive. The DNA detection process increases the chance of very low levels of didymo being detected in a waterway.
  - Reduced sample collection costs. Samples can be taken from the lower reaches of waterways that tend to be more accessible.
  - Greater coverage of waterways. A single sample is often all that is required to be collected from each catchment, meaning that sample collectors were able to collect samples from a greater range of waterways.
- 3.7.3 The survey was the first time that the DNA detection process had been used on a significant scale (it had previously been used for some sampling in Fiordland). The DNA approach did present some risks for the survey. To minimise these risks, The University of Waikato provided a training day for sample collectors in October. During the training and in all subsequent communications, considerable emphasis was placed on the importance of cleaning sampling equipment thoroughly to avoid cross-contamination of samples. The sensitivity of the DNA detection process means that a very thorough cleaning process is required to not only kill any didymo in the sampling equipment, but to actually breakdown any DNA so that it cannot be detected.
- 3.7.4 To assist sample collectors to avoid contaminating samples a detailed, step-by-step sample collection guide was developed by the University. A copy of the guide is included in every DNA sample collection kit. Each sampling kit has sufficient equipment to take up to 20 samples, and can be restocked.
- 3.7.5 The DNA detection process (A sensitive genetic-based detection capability for *Didymosphenia geminata* (Lyngbye) M. Schmidt: Phases Two and Three) can be found on the Biosecurity New Zealand website at the website address in paragraph 3.6.2.

## 4 Other didymo related events

- 4.1 During the period October to December 2007 two related events occurred that impacted on the survey. The first was the discovery of dead didymo cells in water samples taken from rivers in the Central North Island, and the second was confirmation that didymo was in the Rangitata Diversion Race (RDR), in central Canterbury. The main impact of these two events on the survey was that it took longer for some regions to complete their sample collections.
- 4.2 The North Island find of dead cells raised considerable national media coverage, while the RDR find was covered extensively by regional media.

### 4.3 NORTH ISLAND LABORATORY CONTAMINATION

- 4.3.1 In late October 2007, dead didymo cells were found in samples collected from rivers in the Central North Island, during Genesis Energy's routine monitoring programme. An investigation was initiated involving teams of people from the regional partners, laboratories, and MAF. The investigation, which involved extensive re-sampling to establish the extent of the didymo find, gained a very high profile in the media given the national significance of the rivers and because didymo had been found in the North Island.
- 4.3.2 A subsequent audit and trace back of the sampling process found that the dead cells had been a result of cross-contamination in the laboratory that supplied the sample collection containers. The significance of the cross-contamination showed just how easily sample collection equipment can be contaminated. This emphasises the need for sampling equipment to be very carefully cleaned, especially when DNA analysis is being used.

### 4.4 RANGITATA DIVERSION RACE, MID CANTERBURY

- 4.4.1 The RDR takes water from the Rangitata River for irrigation, stock water, and power generation, and discharges water into the Hinds, Ashburton and Rakaia Rivers in Mid Canterbury. The RDR was not part of the survey although a sampling site at SH1 on the Rakaia River was. A sample taken from the Rakaia River in mid November found no trace of didymo.
- 4.4.2 In early December, visible didymo was found at the beginning of the RDR. Given the likelihood that didymo infected water was flowing into the Rakaia, another sample was taken from the SH1 site on the Rakaia River. Live didymo was subsequently confirmed at this site in December 2007.
- 4.4.3 It is not clear why didymo was not found in the earlier sample taken from the Rakaia River. Further sampling of the Ashburton River was also negative.

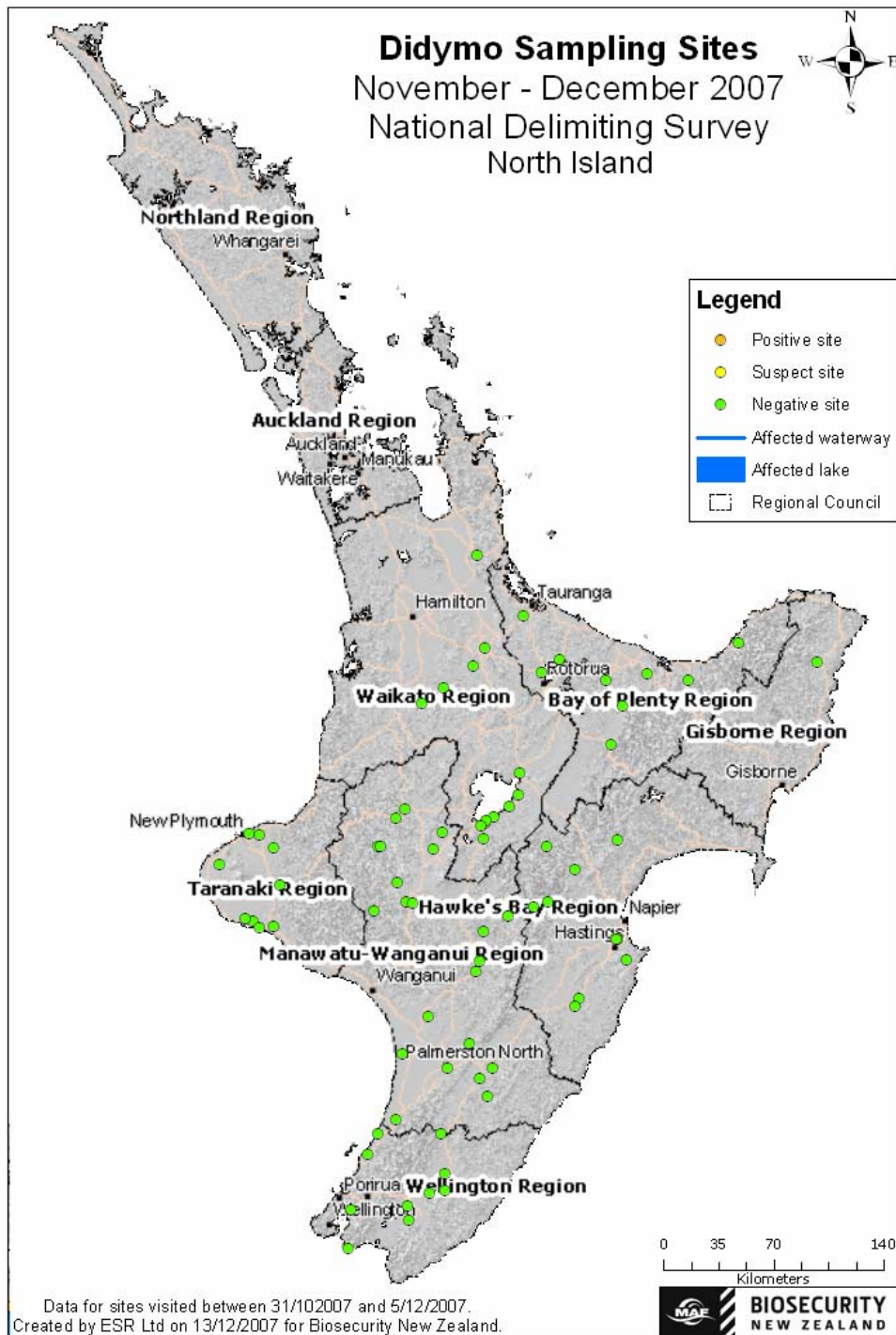
## 5 Costs

- 5.1 The cost of this survey has been approximately \$60,000. This includes the cost of collecting samples, analysing samples, training, travel, and sample collection kits. Some costs are not included in this figure, such as MAF or partner staff time when recovery costs were not sought.

## 6 Results

### 6.1 NORTH ISLAND, SITES AND MAP

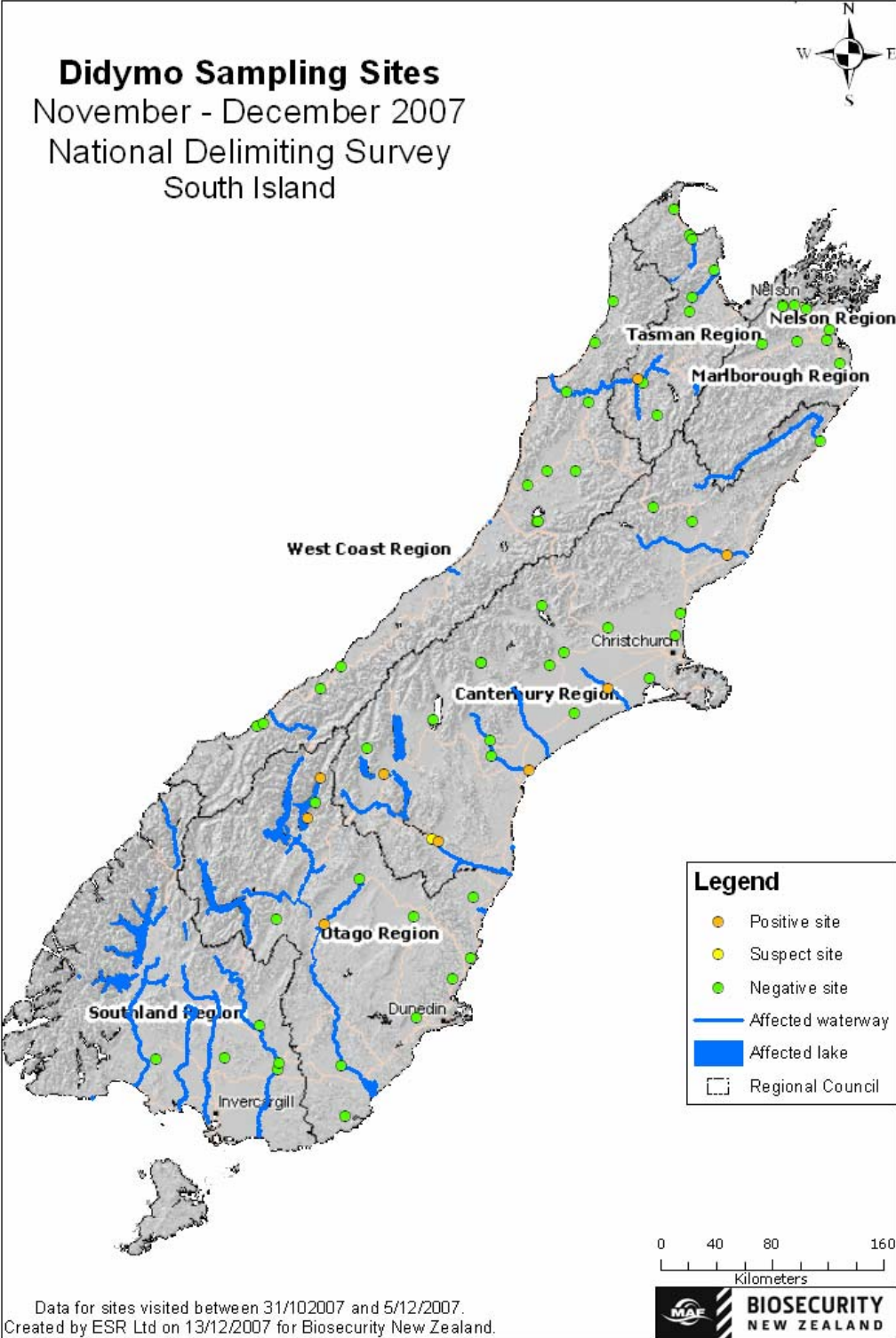
6.1.1 The following map and table show the North Island sites that were included in the survey. All North Island sites tested negative. The map shows only the sites from the survey. All sites can be viewed in the Didymo Samples Database.



| Region             | River              | SiteLocation                       | Result   |
|--------------------|--------------------|------------------------------------|----------|
| Bay of Plenty      | Kaituna            | Trout Pool Rd                      | Negative |
| Bay of Plenty      | Motu               | Hauptu                             | Negative |
| Bay of Plenty      | Ngongotaha Strm    | Railway Bridge                     | Negative |
| Bay of Plenty      | Rangitaiki         | Downstream of Waiohou              | Negative |
| Bay of Plenty      | Tarawera           | U/s golf course, true right        | Negative |
| Bay of Plenty      | Waioeka            | Entrance to gorge                  | Negative |
| Bay of Plenty      | Whakatane          | Grace Road                         | Negative |
| Bay of Plenty      | Whirinaki River    | Whirinaki Rd, Murupara             | Negative |
| Gisborne           | Mata River         | Matarika Road                      | Negative |
| Hawkes Bay         | Wairoa River       | Power Station                      | Negative |
| Hawkes Bay         | Mohaka             | Pakātutu Rd Bridge                 | Negative |
| Hawkes Bay         | Mohaka             | Taharua confl.                     | Negative |
| Hawkes Bay         | Mohaka             | Te Hoe Confluence                  | Negative |
| Hawkes Bay         | Ngaruroro          | Kuripapango                        | Negative |
| Hawkes Bay         | Ngaruroro          | u/s Chesterhope Br.                | Negative |
| Hawkes Bay         | Tararua            | Napier Taihape RD Bridge           | Negative |
| Hawkes Bay         | Tukituki           | SH2                                | Negative |
| Hawkes Bay         | Tukituki           | Waimarama                          | Negative |
| Hawkes Bay         | Tutaekuri          | SH50 Bridge                        | Negative |
| Hawkes Bay         | Waipawa            | SH2 Br, Waipawa                    | Negative |
| Manawatu/Whanganui | Whanganui          | Pipiriki                           | Negative |
| Manawatu/Whanganui | Whanganui          | d/s Retaruke (Wades Landing)       | Negative |
| Manawatu/Whanganui | Whanganui          | Cherry Grove                       | Negative |
| Manawatu/Whanganui | Whanganui          | at Hohotaka Bridge                 | Negative |
| Manawatu/Whanganui | Mangatepopo        | at intake                          | Negative |
| Manawatu/Whanganui | Whakapapa          | at Kakahi                          | Negative |
| Manawatu/Whanganui | Makotuku           | u/s Raetihi                        | Negative |
| Manawatu/Whanganui | Manganui o te Ao   | Ruatiti Rd Bridge                  | Negative |
| Manawatu/Whanganui | Mangawhero         | Pakih Road                         | Negative |
| Manawatu/Whanganui | Retaruke           | Retaruke Township                  | Negative |
| Manawatu/Whanganui | Hautapu            | u/s Rangitikei River               | Negative |
| Manawatu/Whanganui | Makuri             | Tuscan Hills                       | Negative |
| Manawatu/Whanganui | Manawatu           | at Teacher's College               | Negative |
| Manawatu/Whanganui | Manawatu           | Hopelands Reserve                  | Negative |
| Manawatu/Whanganui | Mangatainoka       | SH2                                | Negative |
| Manawatu/Whanganui | Maowhango          | Maowhango                          | Negative |
| Manawatu/Whanganui | Ohau               | SH1                                | Negative |
| Manawatu/Whanganui | Pohangina          | Mais Reach                         | Negative |
| Manawatu/Whanganui | Rangitikei         | Onepuhi                            | Negative |
| Manawatu/Whanganui | Rangitikei         | Mangaweka                          | Negative |
| Manawatu/Whanganui | Rangitikei         | Springvale Bridge                  | Negative |
| Manawatu/Whanganui | Rangitikei         | Pukeokahu                          | Negative |
| Taranaki           | Kapuni             | SH45                               | Negative |
| Taranaki           | Kaupokonui         | Glenn Rd u/s bridge                | Negative |
| Taranaki           | Manganui           | Bristol Rd d/s from bridge at trib | Negative |
| Taranaki           | Mangaoraka         | Corbett Rd                         | Negative |
| Taranaki           | Patea              | at Skinner Rd                      | Negative |
| Taranaki           | Stony River        | SH45                               | Negative |
| Taranaki           | Tahwhiti Stream    | TFG hatchery Hawera                | Negative |
| Taranaki           | Waingongoro        | Ohawe Beach                        | Negative |
| Taranaki           | Waiwhakaiho        | Constance Rd d/s of bridge.        | Negative |
| Waikato            | Ohinemuri          | Karangahake                        | Negative |
| Waikato            | Pokaiwhenua        | Arapuni Road                       | Negative |
| Waikato            | Puniu              | Bayley Road                        | Negative |
| Waikato            | Waihou             | Okorere d/s of Br                  | Negative |
| Waikato            | Waipa              | Toa Bridge                         | Negative |
| Waikato            | Hinemaiaia         | Hinemaiaia                         | Negative |
| Waikato            | Tongariro          | Bain Pool                          | Negative |
| Waikato            | Tongariro          | Blue Pool                          | Negative |
| Waikato            | Waikato River      | Reids Farm                         | Negative |
| Waikato            | Tauranga Taupo     | First exit point                   | Negative |
| Waikato            | Waimarino          | Highway Bridge                     | Negative |
| Waikato            | Waiotaka R         | Highway Bridge                     | Negative |
| Waikato            | Waitahanui         | Waitahanui Bridge                  | Negative |
| Wellington         | Hutt River         | Bulcott                            | Negative |
| Wellington         | Orongorongo River  | Mouth                              | Negative |
| Wellington         | Otaki River        | Mouth                              | Negative |
| Wellington         | Ruamahanga River   | Pukio                              | Negative |
| Wellington         | Ruamahanga River   | McLays                             | Negative |
| Wellington         | Ruamahanga River   | Gladstone                          | Negative |
| Wellington         | Tauherenikau River | Websters                           | Negative |
| Wellington         | Waingawa River     | South Road                         | Negative |
| Wellington         | Waiohine River     | Bicknells                          | Negative |
| Wellington         | Waikanae River     | Greenaway Road                     | Negative |

## 6.2 SOUTH ISLAND, SITES AND MAP

6.2.1 The following map and table show the South Island sites that were included in the survey. Dead didymo cells were found at the sites marked “suspect positive”. In some instances “suspect positive” sites were resampled as a precaution. Live didymo was found at the sites marked “positive”.



| Region      | River             | Site/Location   | Result           |
|-------------|-------------------|---|------------------|
| Canterbury  | Ashley River      | at SH 1 below bridge  | Negative         |
| Canterbury  | Deep Creek        | Left at Mt Potts airstrip, 1km towards Rangitata through paddocks | Negative         |
| Canterbury  | Harper River      | above Coleridge diversion   | Negative         |
| Canterbury  | Hope River        | at SH7 bridge   | Negative         |
| Canterbury  | North Ashburton   | 500 m u/s SH72 Br   | Negative         |
| Canterbury  | Opuha             | below skiptons bridge   | Negative         |
| Canterbury  | Rakaia            | 300 m d/s Gorge Br  | Negative         |
| Canterbury  | Selwyn            | Coes Ford   | Negative         |
| Canterbury  | Upper Rangitata   | Below Coal Creek  | Negative         |
| Canterbury  | Waiau             | Leslie Hills Rd Br  | Negative         |
| Canterbury  | Waimakariri       | 50m below jetboat ramp  | Negative         |
| Canterbury  | Waimakariri       | d/s Gorge bridge  | Negative         |
| Canterbury  | Opihi             | Raincliff u/s of bridge   | Negative         |
| Canterbury  | Hakataramea       | 1.3 km from SH82  | Positive         |
| Canterbury  | Hurunui           | 500m below SH 1 bridge  | Positive         |
| Canterbury  | Ohau River        | 300 m u/s L Ruataniwha  | Positive         |
| Canterbury  | Rakaia            | sh1   | Positive         |
| Canterbury  | Awakino           | at SH 83 Bridge   | Suspect Positive |
| Canterbury  | Opihi             | Waipopo   | Suspect Positive |
| Canterbury  | Dobson            | Glen Lyon Rd @ head of lake                                       | Negative         |
| Canterbury  | Lake McGregor     | Bridge  | Negative         |
| Canterbury  | Clarence River    | SH 1 under road bridge  | Negative         |
| Marlborough | Awatere River     | Seddon rail/road bridge   | Negative         |
| Marlborough | Kaituna           | Reading Road  | Negative         |
| Marlborough | Opawa             | car park by Hagley building                                       | Negative         |
| Marlborough | Pelorus Bridge    | Pelorus Bridge  | Negative         |
| Marlborough | Rai River         | Rai Falls   | Negative         |
| Marlborough | Staircase Stream  |   | Negative         |
| Marlborough | Wairau            | Below SH1 bridge  | Negative         |
| Marlborough | Wairau            | The Narrows   | Negative         |
| Marlborough | Wakamarina River  | State Highway Bridge  | Negative         |
| Otago       | Catlins River     | Tawanui Rd, SOE Site  | Negative         |
| Otago       | Dingle Burn       | near homestead  | Negative         |
| Otago       | Dunstan Creek     | Beatie Rd Br  | Negative         |
| Otago       | Kauru             | Kakanui Valley Rd   | Negative         |
| Otago       | Nevis River       | d/s of crossing   | Negative         |
| Otago       | Pomahaka          | 300 m d/s Blach Bridge  | Negative         |
| Otago       | Shag River        | Palmerston  | Negative         |
| Otago       | Taieri            | Kokonga Bridge  | Negative         |
| Otago       | Waikouaiti River  | Bucklands Crossing  | Negative         |
| Otago       | Hunter River      | Cascade creek   | Positive         |
| Otago       | Manuherikia River | Galloway Road   | Positive         |
| Otago       | Timaru River      | d/s bridge  | Positive         |
| Otago       | Taieri            | at Outram   | Negative         |
| Southland   | Mataura River     | Gore, Surrey Street   | Negative         |
| Southland   | Orauea            | Feldwick Road   | Negative         |
| Southland   | Otapiri           | Otapiri Gorge Rd Br   | Negative         |
| Southland   | Waikaia River     | below Waipounamu Bridge   | Negative         |
| Southland   | Waikaka Stream    | River Road, Gore  | Negative         |
| Tasman      | Anatoki River     | u/s of One Spec road bridge                                       | Negative         |
| Tasman      | Aorere River      | Rockville bridge  | Negative         |
| Tasman      | Mangles           | SH6 Bridge  | Negative         |
| Tasman      | Matakitaki        | Mt Ella 36  | Negative         |
| Tasman      | Pearce            | By old toilet, 2km from Mot River                                 | Negative         |
| Tasman      | Riwaka river      | Rugby Ground, SH60  | Negative         |
| Tasman      | Waikoropupu River | downstream of Pupu Springs  | Negative         |
| Tasman      | Wangapeka         | Walters Peak  | Negative         |
| Tasman      | Matiri            | 1.5km d/s bridge  | Positive         |
| West Coast  | Ahaura            | Ahaura at SH 7 Bridge   | Negative         |
| West Coast  | Arnold            | SH 7 Bridge   | Negative         |
| West Coast  | Bruce Stream      | North Base  | Negative         |
| West Coast  | Grey              | SH7- Waipuna corner   | Negative         |
| West Coast  | Inangahua         | Landing   | Negative         |
| West Coast  | Jacob's River     | SH 6 bridge   | Negative         |
| West Coast  | Karamea River     | SH 67 bridge  | Negative         |
| West Coast  | Mokihinui River   | SH 67 bridge Seddonville  | Negative         |
| West Coast  | Ohikaniu River    | SH 6 bridge   | Negative         |
| West Coast  | Okuru River       | North Bank Okuru Rd   | Negative         |
| West Coast  | Orangipuku        | Kumara Inchbonnie Rd Bridge                                       | Negative         |
| West Coast  | Paringa River     | SH 6 bridge   | Negative         |
| West Coast  | Turnbull River    | Turnbull Rd   | Negative         |

## 7 Lessons learnt and follow up actions

- 7.1 The devolved approach of the survey highlighted the need to have just one person in each region coordinating sample collection – so that people can be kept informed about what was happening, and where people can seek assistance. In most instances this worked very well, especially considering that many regional partner groups comprise people from several different organisations. In just a few instances there were minor break-downs in communications which resulted in samples not being collected on time or data not being entered into the database, which in turn affected the overall completion time for the survey.
- 7.2 Related to the above communications issues, it had been the intention that the Didymo Samples Database would be the only way that regional partners would contact the laboratories. This approach was taken to maximise the use of the database, and more importantly, to reduce the time for laboratory staff to chase up or match samples back to sampling sites. In most cases the database worked extremely well as the information channel between samplers and the two laboratories. However, both the University of Waikato and NIWA did have to contact or were contacted directly by sample collectors, usually to follow up on missing samples or samples that had been sent to the laboratory without the printed sample collection form from the database.
- 7.3 Similarly, one of the bigger challenges of tracking results throughout the survey and for this report has been matching up the sample site details from the original list of sites agreed with regional partners, and the site descriptions entered into the Didymo Samples Database. In future, greater emphasis should be placed on communicating to sample collectors the need to enter sites into the database that correspond very clearly to the agreed sampling sites, or cross-referencing them clearly with the original agreed list of sites.
- 7.4 Overall feedback from partners and laboratories was that the database worked very well. A few minor enhancements were identified to make it better to use.
- 7.5 The final area where there seemed to be some confusion was around the interpretation of sampling results. A relatively simple criterion has been established to categorise sample results. This is generally as follows: “Negative” if there is no sign of didymo present in the sample, “Suspect Positive” if only dead cells or one or two unhealthy live cells is present in the sample, and “Positive” if the sample contains healthy live didymo. This was worked through satisfactorily as the survey progressed.