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# Overview

# Remote microscopy: a success story in Australian and New Zealand plant biosecurity

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Abstract Rapid and accurate identification of organisms is crucial to many research and applied outcomes. Diagnostics is a critical first step in determining the significance of suspected biosecurity threats posed by emergency plant pests and other invasive pests and pathogens. Traditionally, the biological specimens needing identification are physically mailed to a dispersed community of taxonomic experts for determination. While effective, this is an expensive, labour-intensive and slow process, often taking days to receive a confirmed identification. Remote microscopy creates virtual, real-time networks of experts using web-based cameras mounted on microscopes that allow interactive access to real-time images of scientific specimens from anywhere in the world via the Internet. Trials conducted by the Australian Quarantine and Inspection Service and the Ministry of Agriculture and Forestry Biosecurity New Zealand to test the efficacy of remote microscopy in plant quarantine settings showed that in Australia a diagnosis to a level at which realistic biosecurity decisions could be made occurred on 77% of occasions, while in New Zealand high impact exotic pest status was determined during 92% of the diagnostic events, and regulatory status was determined during 96% of events. These positive results are leading towards the expansion of remote microscopy throughout Australia, New Zealand and into South-East Asia, as well as widening its role as part of online diagnostic frameworks.

Key words AQIS, biosecurity, identification, MAFBNZ, quarantine, remote diagnostics.

# INTRODUCTION

As the world moves increasingly in the direction of digital and virtual environments, the biosecurity community needs to utilise technological advances for fast and accurate diagnostics. The global environment of increased international travel and trade, and the associated incursion risk from invasive and dangerous species also demands diagnostic tools of a high calibre. These factors, combined with a need to utilise existing resources more efficiently and effectively, emphasise the need for increased diagnostic capacity in biosecurity. While most taxonomic expertise resides in museums and research organisations, high-quality diagnostic services are essential to the success of government biosecurity programs through identification-based risk-mitigation processes.

Until recently, biological specimens requiring identification have been mailed to taxonomic experts. Posting of specimens can be costly and unreliable, causing delays in the identification of potentially serious incursions. A recent advance in rapid diagnostics has been the emailing of still images to experts (Disbury *et al.* 2008). While still an immediate approach, this method provides only limited insight into the nature of the pest because of limited interactivity between the two identifying parties (Disbury *et al.* 2008). These limitations can have serious repercussions where exotic pests and international trade are concerned. In these high-stake situations, prompt and accurate diagnosis is paramount.

Remote microscopy (RM) facilitates real-time identification of animal or plant specimens (Kumarasinghe 2008). This is achieved by using web-enabled video cameras mounted on microscopes, allowing live 'streaming' of images to a web address. This web link can then be accessed by anybody (e.g. a specialist entomologist/taxonomist) with access to the Internet. Direct communication between an expert and a specimen holder using RM equipment facilitates a very high level of interactivity. The specialist can instruct the microscope user via telephone or a web-based collaboration tool on how to manipulate the specimen to highlight diagnostic characters. This real-time, highly interactive communication improves

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upon current remote diagnostic systems (such as emailing images) as the person operating the microscope does not require a detailed knowledge of insect morphology in order to take photos of diagnostic characters. Used in this way, RM utilises expert knowledge to its full interactive potential, greatly increasing diagnostic capacity while providing more accurate and timely results.

Early experimentation with scanning electron microscopes (SEM) showed that information such as images, X-ray spectra and parameter data could be shared among geographically dispersed investigators in real time through the Internet (Caldwell *et al.* 1997; Chand *et al.* 1997).

The potential for this technology to be applied in biosecurity settings is great. This is because fast, accurate diagnostics of intercepted plant pests are crucial for the effective treatment, management and containment of biosecurity risks at the border (Stanaway *et al.* 2001; Maynard *et al.* 2004). This type of service also can produce favourable economic outcomes in the way of cost savings related to reducing fumigations/treatments at the border and not having to courier specimens to remote diagnosticians.

This paper presents the results of a trial to test the efficacy of RM in quarantine/biosecurity settings in Australia (Australian Quarantine and Inspection Service (AQIS)) and New Zealand (Ministry of Agriculture and Forestry Biosecurity New Zealand (MAFBNZ)). Specifically, we evaluated RM for the identification of plant pest invertebrate specimens at the border.

# METHODS

The AQIS and MAFBNZ use real-time RM, the essence of which is the connection of non-expert to expert, through the Internet, for the purposes of pest identification, training or knowledge sharing, and is possible in a variety of forms (Table 1).

We used an RM setup of a microscope, a broadband (ADSL) Internet connection, a microscope-mounted camera and a console that acts as a web server to 'stream' realtime images to a widely accessible IP address. The systems used at AQIS and MAFBNZ use Nikon DS-Fi1 cameras mounted to microscopes and Nikon DS-L2 web servers that are connected to the Internet via an independent broadband connection. The image of the specimen in question is viewed through the microscope and captured by the camera that transfers the image to the console. The console then acts as an independent web server and broadcasts the image, via the Internet, to an IP address. The RM set-up does not require a computer at the site that is broadcasting the feed.

When AQIS or MAFBNZ personnel intercept a potential exotic pest at the border they make contact with a general taxonomist (operational scientist/entomologist) who, via RM, performs 'taxonomic triage', either identifying the pest themselves or accessing a network of specialist taxonomists to aid

| Remote microscopy | Microscope camera                     | $\wedge$ | Internet | + | Communication tool        | $\wedge$ | > Expert/taxonomic triage             | ٨        | > Identification/diagnosis/ |
|-------------------|---------------------------------------|----------|----------|---|---------------------------|----------|---------------------------------------|----------|-----------------------------|
|                   |                                       |          |          |   |                           |          |                                       |          | training                    |
| This trial        | Nikon DS-L2 console and DS-Fi1 camera | ٨        | Internet | + | Telephone                 | $\wedge$ | AQIS and MAFBNZ operational personnel | $\wedge$ | Identification              |
| Alternatives†     | PC + alternative microscope camera    | ٨        | Internet | + | Telephone                 | $\wedge$ | Virtual taxonomic environment         | $\wedge$ | Identification              |
|                   | – Oľ –                                |          |          |   | – 0ľ –                    |          | – 0r –                                |          | Diagnosis                   |
|                   | portable USB microscope + web-based   |          |          |   | web-based collaboration   |          | taxonomic triage                      |          | Training                    |
|                   | collaboration software                |          |          |   | tools (e.g. Skype, WebEx) |          |                                       |          |                             |
|                   |                                       |          |          |   | – 0ľ –                    |          |                                       |          |                             |
|                   |                                       |          |          |   | SMS                       |          |                                       |          |                             |
|                   |                                       |          |          |   | – 0ľ –                    |          |                                       |          |                             |
|                   |                                       |          |          |   | email                     |          |                                       |          |                             |
| Example           | Nikon DS-L2 and DS-Fi1                | $\wedge$ | Internet | + | Telephone                 | $\wedge$ | PaDIL: remote microscope diagnostics  | $\wedge$ | Identification              |
|                   |                                       |          |          |   |                           |          |                                       |          | Training                    |

Table 1 Model for remote microscopy showing alternative configurations

in making the identification. The expert makes the identification by viewing the link and communicating instructions on the positioning and focus of the specimen to the person possessing the specimen. This achieves a fully live interactive RM identification session.

The AQIS trial was conducted over 8 months. At the start of this trial, Nikon DS-Fi1 cameras mounted to microscopes were installed at four AQIS locations: Canberra, Perth, Adelaide and Townsville. Subsequently, the AQIS RM network expanded to include Cairns, Gladstone, Mackay, Brisbane, Darwin, Melbourne and Sydney. In the case of AQIS border interceptions, once a determination was made, a decision could be made on the related consignment. The specimens were then sent by post to the entomologist for verification and confirmation of the identification. The time saved through using RM vs. the standard method (post) was recorded by the remote identifier. Any cost saving (e.g. not having to precautionary fumigate or post specimens) through the use of RM was also recorded.

In New Zealand, the system was set up at three MAFBNZ locations: MAFBNZ Cargo site in Auckland, MAFBNZ Plant Health and Environment Laboratory (PHEL), Auckland and MAFBNZ PHEL Christchurch. Inspectors (non-specialists) at the cargo site were encouraged to use the technology to liaise with entomologists at the laboratories in order to identify intercepted invertebrate specimens.

At the PHEL, MAFBNZ, the remote microscopy trial was conducted in parallel to the conventional laboratory identification on samples from routine inspection and clearance of consignments. During the trial, whenever multiple specimens of the same taxa were encountered while inspecting a product line, these specimens were stored and each week the cargo inspectors contacted the laboratory entomologists to conduct the remote diagnosis on those samples. These specimens were then sent to the laboratory for confirmation of the identifications. The time spent per remote identification was limited to 10 min.

#### RESULTS

#### Australia

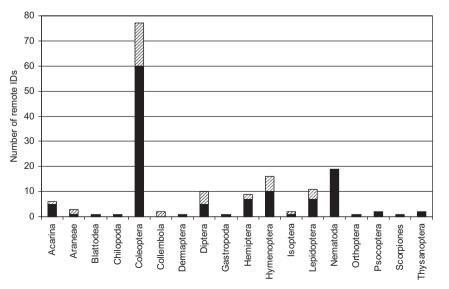
A total of 165 specimens were identified using RM at seven locations. In most cases (128 or 77%), the level of identification allowed AQIS scientists to make preliminary decisions to treat or not the goods involved. Of these, 100 (60%) were diagnosed to genus level, and 62 of those (37%) to species level. The technology therefore allows Operational Science Program (OSP) personnel to 'filter' requests for diagnoses of some pest groups, thus saving AQIS and its clients (importers, exporters, etc.) time and money.

During the trial, 19 fumigation treatments were avoided due to identifications made using real-time cameras in Perth, Brisbane, Townsville and Adelaide. This is because in some cases, fumigations result from clients opting for treatment instead of diagnosis – this culture of treat rather than diagnose is due to the delays experienced in the transport of specimens from outports to OSP scientists. Real-time camera technology permits preliminary diagnosis within hours of interception, thereby allowing AQIS operational staff to make decisions and provide technical advice within similar time frames.

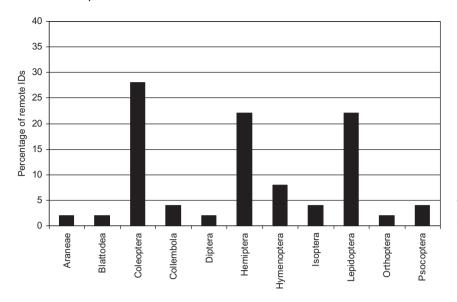
An average of 2.64 days was saved each time a specimen was examined by RM. This was a direct result of not having to courier or post specimens to OSP scientists. Consequently, decisions in relation to treatment were able to be made on the same day, often within the 10 min. This also led to cost savings through fewer precautionary fumigations, which equated to \$2850 or \$356.25/month (calculated on fumigating a 20 foot shipping container at \$150 each).

A broad range of invertebrate types, especially Coleoptera, Nematoda and Hymenoptera, were examined (Fig. 1). In the vast majority of cases where RM was implemented, the level of identification was sufficient to make an informed decision on what action to undertake. Identifications of beetles and ants

Fig. 1. Types of intercepted invertebrates examined remotely by Australian Quarantine and Inspection Service (AQIS) Operational Science Program (OSP) scientists using remote microscopy (RM) technology. Solid sections of the bars represent instances in which a decision was able to be made based on identifications conducted remotely, and the hatched section of the bars represent instances where a decision could not be made based on RM information alone.



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*Fig. 2.* Types and percentages of border intercepted arthropods examined remotely by Ministry of Agriculture and Forestry Biosecurity New Zealand (MAFBNZ) Plant Health and Environment Laboratory (PHEL) scientists using remote microscopy.

 Table 2
 Types of intercepted invertebrates examined remotely

 by AQIS OSP scientists using remote microscopy

|                | Life stage/             | Numbers | Percentage |
|----------------|-------------------------|---------|------------|
|                | regulatory status/      |         |            |
|                | level of identification |         |            |
| Life stage     | Immature                | 7       | 4          |
|                | Adult                   | 158     | 96         |
| Regulatory     | Regulated <sup>†</sup>  | 53      | 32         |
| status         | Non-regulated‡          | 72      | 44         |
|                | Unable to make          | 40      | 24         |
|                | decision via RDRM       |         |            |
| Level of       | Order                   | 18      | 11         |
| identification | Family                  | 47      | 28         |
|                | Genus                   | 38      | 23         |
|                | Species                 | 62      | 38         |

†Pests for which actions such as fumigation, re-export or destruction would be undertaken if they were intercepted. ‡Pests for which no actions would be undertaken if they were intercepted, i.e. they would be released.

AQIS, Australian Quarantine and Inspection Service; OSP, Operational Science Program.

were very successful, with 77% and 62% of identifications, respectively, leading to a decision on what action to take. The use of RM of nematodes, however, proved most successful, with 100% of identifications using the technology leading directly to a decision to release, fumigate, re-export or destroy the consignment.

## **New Zealand**

A total of 51 intercepted specimens were examined remotely by PHEL staff using RM. A range of arthropod types was examined (Fig. 2), including immatures and adults.

The results of the New Zealand study are comparable with the Australian results (Table 2,3). In most cases, specimens were diagnosed to a level at which a preliminary categorisation could be made, i.e. high-impact exotic pest status was determined for 47 of 51 specimens (92%) and regulatory status was *Table 3* Details of border intercepted arthropods examined remotely by MAFBNZ PHEL scientists using remote microscopy

|                | Life stage/<br>regulatory status/<br>level of identification | Numbers | Percentage |
|----------------|--|---------|------------|
| Life stage     | Immature   | 13      | 26         |
| e              | Adult  | 38      | 74         |
| Regulatory     | HIEP†  | 0       | 0          |
| status         | Not HIEP   | 47      | 92         |
|                | HIEP status unsure<br>(unable to make<br>this decision)      | 4       | 8          |
|                | Regulated‡   | 44      | 86         |
|                | Non-regulated§   | 5       | 10         |
|                | Regulated status<br>unsure (unable to<br>make this decision) | 2       | 4          |
| Level of       | Order  | 11      | 21         |
| identification | Family   | 21      | 41         |
|                | Genus  | 10      | 20         |
|                | Species  | 9       | 18         |

†High impact exotic pest. ‡Pests for which measures and actions would be undertaken if they were intercepted or detected. §Pests for which action would not be undertaken if they were intercepted or detected.

MAFBNZ, Ministry of Agriculture and Forestry Biosecurity New Zealand; PHEL, Plant Health and Environment Laboratory.

determined for 49 specimens (96%) using RM. The accuracy of the diagnosis was very high, with only 1 incorrect identification out of 51. However, the taxonomic level of identification using RM was low due to the time limit of 10 min per specimen. This restricted time limit was set to enable categorisation of intercepted pests rather than trying to achieve identification to the species level.

## DISCUSSION

Remote microscopy innovations are changing the process of identification of biological specimens by bringing taxonomic expertise to the specimen rather than the specimen to the expert, providing nearly immediate identifications. With taxonomic expertise dwindling worldwide, RM offers a powerful means of improving communication and increasing diagnostic capacity. Connecting experts with problematic specimens in real time has the potential to save organisations time, money and resources, as well as provide more accurate identifications.

Our results indicate that RM is indeed an effective tool for identifying problematic specimens in a variety of quarantine settings. The results from both Australia and New Zealand show that RM facilitates accurate, fast and meaningful identifications to a level where biosecurity decisions can be made. Immediate diagnoses were achieved the vast majority of the time and importantly, rapid operational responses to these identifications could be made.

The direct 'downstream' outcomes to these faster responses include cost savings for importers by not incurring demurrage fees at ports while specimens are being diagnosed, negating the chance of specimens being lost or damaged in transit and a reduction in precautionary fumigations that result directly from importers being provided with almost immediate decisions regarding treatment. These findings are the first of their kind and should pave the way for wider use of RM.

Since the start of this trial RM has expanded throughout Australia, New Zealand and into South-East (SE) Asia. Currently, AQIS nodes exist at Cairns, Townsville, Mackay, Gladstone, Brisbane, Sydney, Melbourne, Adelaide, Perth, Karratha and Darwin and MAFBNZ nodes at Auckland and Christchurch. General plant health nodes exist in Australian state-based Departments of Primary Industries at Cairns, Toowoomba, Brisbane, Orange, Canberra, Perth, Darwin, Katherine, Kununurra and in Thailand (Bangkok and Chang Rai), Lao PDR (Vientiane), Vietnam (Hanoi) and Singapore. These nodes draw on expertise from all over Australia and New Zealand to effect diagnosis, training and knowledge sharing as part of a wider 'Remote Microscope Network'. The expansion of an RM network into the SE Asian and Oceanic regions will be beneficial to our pre-border surveillance and biosecurity protection. Much of the current RM proliferation is focussed on fruit fly relevant areas and commodities within Australia in line with other national initiatives.

Our results indicate that RM lends itself to forming part of virtual taxonomic environments, opening up dynamic links among institutions and specimen collections around the world. They demonstrate that RM is a potentially powerful tool that will complement virtual taxonomy in the future. It is through virtual taxonomic environments that RM has the potential to influence the biosecurity community, by up-skilling its non-specialist users. The transfer of knowledge that occurs during RM will be captured and stored in these virtual environments in the form of image and information libraries.

Connecting specimens to experts in real time has been shown to be effective with the use of RM. This is the most common use of RM, but the technology also has a variety of other applications. RM can be used in collections, museums and universities as a tool to facilitate the sharing of knowledge and the virtual sharing of specimens. RM also has applications in training where an expert may train people in several locations at once via RM connections. RM has the potential to greatly augment the development of virtual taxonomy. Virtual tools such as Pests and Diseases Image Library (PaDIL) (http://www.PaDIL.gov.au), the Plant Biosecurity Toolbox (http://www.PaDIL.gov.au/PBT), the Biosecurity Bank (http:// www.biosecuritybank.com) and future virtual collaborations are likely to benefit from developments in RM. Virtual taxonomic environments can utilise RM to build on existing diagnostic capacity and allow people from all over the world to share information and collaborate to solve taxonomic problems. Some future RM Network interactions will be based from a portal housed within PaDIL. PaDIL, along with the Plant Biosecurity Toolbox and the BioSecurity Bank, are powerful virtual tools to be used in conjunction with RM that will allow RM users to enhance their identification, training and knowledge sharing sessions. However, PaDIL-based RM use is not, and will not be, exclusive. Future RM use from PaDIL will be structured with the portal providing protocols for use and expert contact, an expert register and useful links to other PaDIL resources.

Remote microscopy could also form part of the repertoire of tools and collaborative endeavours that contribute to any virtual taxonomic environment. A workspace such as this could serve to promote real-time remote diagnostics as a tool to support biosecurity and sustainable agriculture, remote teaching and training delivery, and virtual curatorial systems.

Another potential application of RM is for researchers in the field of systematic studies. They can remotely access primary type specimens (holotypes) of species as part of the process of checking if a new species being described is indeed new or has already been described. Taxonomists can access type specimens in museums and national collection centres through RM and compare with their specimens without actually visiting or obtaining loan specimens from the institution. This saves researchers' time, costs and helps preserve the quality and safety of specimens.

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