

Final pest risk analysis for *Pepino mosaic virus* and pospiviroids associated with tomato seed

February 2021



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Acronyms and abbreviations

Term or abbreviation	Definition
ALOP	Appropriate level of protection
BICON	Australia's Biosecurity Import Condition System
BIRA	Biosecurity Import Risk Analysis
ССЕРР	Consultative Committee on Emergency Plant Pests
IPPC	International Plant Protection Convention
ISPM	International Standard for Phytosanitary Measures
NPPO	National Plant Protection Organisation
NSW	New South Wales
NT	Northern Territory
PCR	Polymerase Chain Reaction
PFA	Pest Free Area
PRA	Pest Risk Analysis
Qld	Queensland
RT-PCR	Reverse Transcription - Polymerase Chain Reaction
SA	South Australia
SPS Agreement	WTO agreement on the Application of Sanitary and Phytosanitary Measures
The department	The Department of Agriculture, Water and the Environment
Tas.	Tasmania
Vic.	Victoria
WA	Western Australia
WTO	World Trade Organization

Summary

Australia imports large quantities of vegetable seeds annually and depends heavily on these imports to produce a range of crops, including tomato crops.

The geographic ranges of seed-borne pathogens are expanding globally, and new risks frequently emerge. The agricultural seeds trade has become globalised and is evolving—seed lines are typically developed and commercially multiplied in different countries and rapidly shipped and sold internationally. As seed is moved from country to country for seed production, seed lines may be exposed to a broad range of pathogens and the likelihood that these pathogens may enter Australia via imported seeds may increase.

In 2008, Australia implemented emergency measures to manage the risk posed by *Potato spindle tuber viroid* (PSTVd) associated with imported tomato seed (*Solanum lycopersicum*). These measures were then revised in 2012 and 2013, including the addition of measures to manage the risks posed by five more pospiviroids—*Columnea latent viroid* (CLVd), *Pepper chat fruit viroid* (PCFVd), *Tomato apical stunt viroid* (TASVd), *Tomato chlorotic dwarf viroid* (TCDVd) and *Tomato planta macho viroid* (TPMVd) and *Pepino mosaic virus* (PepMV).

The World Trade Organisation Agreement on the Application of Sanitary and Phytosanitary Measures and the International Plant Protection Convention require that any phytosanitary measure applied against a pest must be technically justified. Countries may take emergency actions, including emergency measures, when a new or unexpected phytosanitary risk is identified, as specified in the International Standard for Phytosanitary Measures (ISPM) No. 1. Such measures should be evaluated by pest risk analysis or other comparable examination as soon as possible, to ensure that the continuance of the measure is technically justified.

The Australian Government Department of Agriculture, Water and the Environment initiated this pest risk analysis (PRA) to assess the risks posed by five pospiviroids and PepMV associated with imported tomato seed. The regulatory status of PSTVd is under review as a process separate to this PRA, therefore a risk assessment for this viroid is not presented in this final report.

This PRA concluded that the unrestricted risks of four pospiviroids (CLVd, PCFVd, TASVd and TCDVd) and PepMV on the tomato seeds for sowing pathway do not achieve the appropriate level of protection (ALOP) for Australia. Pest risk management measures are therefore required to mitigate the risks posed by these five quarantine pests to achieve the ALOP for Australia.

In addition to the department's standard seeds for sowing import conditions, three risk management options are recommended for seeds of *Solanum lycopersicum* (tomato) and hybrids of this species:

- Option 1. Polymerase chain reaction (PCR) test—an option that is applicable to all five quarantine pests associated with tomato seed.
 - PCR using sample size of 20,000 seeds or 20% of small seed lots to verify freedom from the detectable presence of CLVd, PCFVd, TASVd, TCDVd and PepMV.
- Option 2. Enzyme-linked immunosorbent assay (ELISA) test—an option that is applicable only to PepMV.

- ELISA test using sample size of 20,000 seeds or 20% of small seed lots to verify freedom from the detectable presence of PepMV.
- Option 3. Heat treatment—an option that is applicable only to PepMV.
 - Dry heat treatment at 80 °C for 72 hours.

If the required treatment or testing is undertaken off-shore, phytosanitary certification is required with the additional declaration that this has been conducted in accordance with Australia's requirements.

Alternatives to testing or treatment, such as sourcing seed from pest-free areas or pest-free places of production, or sourcing seed produced under a systems approach, may be considered. Supporting documentation demonstrating pest free area status, pest free place of production status, or details of a proposed systems approach will be required for the department to consider these options on a case-by-case basis.

The department, in a separate process, is evaluating whether PSTVd should be a regulated pest. Therefore, the recommended pest risk management measures specified in this final report will apply to this viroid on imported tomato seeds, until this process has concluded. Stakeholders will then be notified of any decision and next steps.

Comments raised by stakeholders on the *Draft pest risk analysis for Pepino mosaic virus and pospiviroids associated with tomato seed* were taken into consideration in the preparation of this final report. Key responses are presented in Appendix D.

The key changes made in the final report are:

- The removal of TPMVd due to insufficient evidence for this viroid to be considered on the tomato seed for sowing pathway
- Retaining the ELISA test as an option to manage the risk of introducing PepMV.

Editorial revision has occurred between the draft and this final report to address issues raised by stakeholders and for improved consistency and clarity, including moving the former Chapters 3, 4 and 5 to Appendices, where they provide supplementary material to the risk assessments.

In conclusion, this PRA demonstrates that the continuation of measures for CLVd, PCFVd, TASVd, TCDVd and PepMV is technically justified. The recommended pest risk management measures are also broadly consistent with the emergency measures they replace.

1 Introduction

1.1 Australia's biosecurity policy framework

Australia's biosecurity policies aim to protect Australia against the risks that may arise from exotic pests entering, establishing and spreading in Australia, thereby threatening Australia's unique flora and fauna, as well as those agricultural industries that are relatively free from serious pests.

The risk analysis process is an important part of Australia's biosecurity policy development. It enables the Australian Government to formally consider the level of biosecurity risk that may be associated with proposals to import goods into Australia. If the biosecurity risks do not achieve the appropriate level of protection (ALOP) for Australia, risk management measures are recommended to reduce the risks to an acceptable level. If the risks cannot be reduced to an acceptable level, the goods will not be imported into Australia until suitable measures are identified or developed.

Successive Australian Governments have maintained a stringent, but not a zero risk, approach to the management of biosecurity risks. This approach is expressed in terms of the ALOP for Australia, which is defined in the *Biosecurity Act 2015* as providing a high level of protection aimed at reducing risk to a very low level, but not to zero.

Australia's risk analyses are undertaken by the Department of Agriculture, Water and the Environment using technical and scientific experts in relevant fields and involves consultation with stakeholders at various stages during the process.

Risk analyses may take the form of a biosecurity import risk analysis (BIRA) or a review of biosecurity import requirements (such as scientific reviews of existing policy and import conditions, pest-specific assessments, weed risk assessments, biological control agent assessments or scientific advice).

Further information about Australia's biosecurity framework is provided in the *Biosecurity Import Risk Analysis Guidelines 2016* located on the <u>Department of Agriculture, Water and the</u> <u>Environment</u> website.

1.2 This risk analysis

1.2.1 Background

The Australian Government Department of Agriculture, Water and the Environment initiated this pest risk analysis (PRA), consistent with IPPC standards, to consider the risks posed by pospiviroids and *Pepino mosaic virus* associated with imported tomato seed, and the technical justification for phytosanitary measures to mitigate the risks.

This PRA also fulfils Australia's international obligations under the IPPC and ISPM 1 to consider the technical justification for the emergency measures introduced and revised between 2008 and 2013. The emergency measures were for *Pepino mosaic virus* (PepMV), *Columnea latent viroid* (CLVd), *Pepper chat fruit viroid* (PCFVd), *Potato spindle tuber viroid* (PSTVd), *Tomato apical stunt viroid* (TASVd), *Tomato chlorotic dwarf viroid* (TCDVd) and *Tomato planta macho viroid* (TPMVd) associated with tomato seeds imported into Australia.

1.2.2 Scope

This final report provides pest risk assessments for *Columnea latent viroid*, *Pepper chat fruit viroid*, *Tomato apical stunt viroid*, *Tomato chlorotic dwarf viroid* and *Pepino mosaic virus* associated with tomato (*Solanum lycopersicum*) seeds for sowing, from all sources, imported into Australia. It also presents a review of the emergency measures implemented for these pathogens and, as appropriate, will recommend the pest risk management measures required to mitigate the risks posed by the identified quarantine pests to achieve the ALOP for Australia.

Information about *Potato spindle tuber viroid* and *Tomato planta macho viroid* was evaluated in this pest risk analysis, but pest risk assessments for these viroids are not presented. Discussion of the status of these viroids is presented in Chapter 3.

Risk assessments are not given for the identified quarantine pests in wild tomato species (*Solanum chilense, S. chmielewskii, S. parviflorum, S. peruvianum* and *S. pimpinellifolium*) because there is insufficient evidence of the pests being associated with the seeds of these species. Consequently, the scope of this PRA is limited to the identified quarantine pests associated with seeds of *Solanum lycopersicum* (tomato) and hybrids of this species (including crosses with wild tomato species).

1.2.3 Emergency measures

Prior to 1992, tomato (*Solanum lycopersicum*) seed was a restricted seed for which importation into Australia required an import permit, hot water treatment and immersion in trisodium phosphate solution under quarantine supervision. In 1992, the former Australian Quarantine and Inspection Service (AQIS) reviewed the quarantine status of tomato seed. New import conditions that removed the treatment requirements were announced in Quarantine Circular Memorandum (Plants) 1992/95 on 2 December 1992. The requirement for seed treatment was removed on the basis that *Phoma lycopersici, Tomato mosaic virus* and *Clavibacter michiganensis* subsp. *michiganensis* were present in Australia and were not under official control. Tomato seed was permitted entry after visual inspection by AQIS on arrival in Australia, following the same conditions as other permitted seeds.

Initiation of emergency measures (2008)

Following incursions of PSTVd in Australia in 2001, 2004, 2006 and 2007, the Consultative Committee on Emergency Plant Pests (CCEPP) requested that the former Biosecurity Australia undertake a PRA for PSTVd associated with tomato seed, which commenced in September 2007. As PSTVd was then recognised as seed-transmitted in tomato, Australia introduced emergency measures for PSTVd in tomato seed on 25 June 2008 (G/SPS/N/AUS/225).

The emergency measures for tomato seed required that consignments were accompanied by a Phytosanitary Certificate endorsed with one of the following additional declarations:

• The tomato seed in lot(s) [numbers] in the consignment was grown in [Country] in an area that is free of *Potato spindle tuber viroid*, based on an official survey covering the complete range of potato spindle tuber viroid hosts, OR

• The tomato seed in lot(s) [numbers] in the consignment was derived from seed and pollen parent plants grown by [producer] in [country] that were tested during the growing period and found free of *Potato spindle tuber viroid*.

Following concerns raised by stakeholders about the ability of exporting countries to meet the requirements for seeds already produced, an extra additional declaration option was provided (7 August 2008):

• No symptoms of diseases caused by *Potato spindle tuber viroid* have been observed on the plants at the place of production during their complete cycle of vegetation.

For tomato seed lines that were not certified free from PSTVd by one of the above declarations, Australia required a test be undertaken on arrival for PSTVd on a sample of 20,000 seeds. Seed lines that tested positive for PSTVd were either re-exported or destroyed.

A sample size of 20,000 seeds was considered necessary to detect low levels of contamination of seed lines by PSTVd-infected tomato seed. This sample size was supported by an investigation in the United Kingdom of an outbreak in a greenhouse crop from which the contamination of a seed lot could be estimated (Mumford, Jarvis & Skelton 2004).

Revision I (February 2012)

Following an incursion of PSTVd in Queensland (Qld) in 2011, a CCEPP meeting in April of that year recommended that the emergency measures implemented in 2008 be strengthened.

The emergency measures were revised (3 February 2012; G/SPS/N/AUS/225/Add.1), removing the additional declaration based on freedom from symptoms and replacing it with one based on a requirement for a test for PSTVd of a 20,000 seed sample from each seed lot. Samples were to be tested using a reverse-transcription polymerase chain reaction (RT-PCR). Acknowledging that there were several possible protocols to test for this viroid, the emergency measures did not stipulate a specific protocol for overseas laboratories. However, the option for an additional declaration based on area freedom was retained.

Emergency measures were also introduced for PepMV associated with tomato seeds. An additional declaration for PepMV was required, with options for testing a 3,000 seed sample, area freedom or the testing of parent plants.

Revision II (May 2012)

As a result of testing on arrival, *Tomato chlorotic dwarf viroid* was detected in tomato seed that had been produced in a country in Africa and a country in Europe. The emergency measures were revised (23 May 2012; G/SPS/N/AUS/225/Add.2) to include this viroid with options for testing a seed sample, area freedom or testing of parent plants.

Revision III (November 2012)

As a result of testing on arrival, PCFVd was detected in seed that had been produced in the Middle East and Asia. The emergency measures were again revised (16 November 2012; G/SPS/N/AUS/225/Add.3) to require testing a seed sample or testing parent plants. Emergency measures requiring testing of seed or parent plants were also introduced for CLVd, TASVd and TPMVd.

Under the revised emergency measures, the option of additional declaration based on area freedom was removed. This option required an official survey to establish area freedom. No requests for this option were received by the department. Area freedom based on absence of records was not accepted by the department, as Australian testing results showed that such claims were incorrect on several occasions. Furthermore, tomato seed is freely traded between countries, and this appeared to be often occurring without testing for pospiviroids.

Revision IV (2013)

The additional declaration option based on parent plant testing was removed (21 November 2013). This option required the testing of all parent plants, and no requests for this option were received by the department. An option based on the partial testing of parent plants was permitted for a time until testing detected pospiviroids in tomato seed imported under this option.

A summary of these emergency measures and subsequent revisions is presented in Table 1.1.

Table 1.1. Summary of emergency measures for pospiviroids and *Pepino mosaic virus* associated with tomato seed

Date	SPS notification	Pest	Action/measure
25 Jun 2008	G/SPS/N/AUS/225	PSTVd	Seed derived from a pest free area OR from tested parent plants.
3 Feb 2012	G/SPS/N/AUS/225 /Add.1	PSTVd, PepMV	Seed tested for PSTVd and PepMV OR derived from tested parent plants OR from pest free areas or properties as shown by official surveys.
25 May 2012	G/SPS/N/AUS/225 /Add.2	PSTVd, PepMV and TCDVd	Seed tested for PSTVd, PepMV and TCDVd OR derived from tested parent plants OR from pest free areas or properties as shown by official surveys.
16 Nov 2012	G/SPS/N/AUS/225 /Add.3	CLVd, PCFVd, PSTVd, TASVd, TCDVd, TPMVd and PepMV	Seed tested for CLVd, PCFVd, PSTVd, TASVd, TCDVd, TPMVd and PepMV OR derived from tested parent plants.
21 Nov 2013 (current)	-	CLVd, PCFVd, PSTVd, TASVd, TCDVd, TPMVd and PepMV	The option based on parent plants testing was removed. All tomato seed to be tested for the six pospiviroids and PepMV.

Current emergency measures

The current emergency measures on tomato seed for the management of identified pospiviroids and *Pepino mosaic virus* are summarised below:

- For PepMV—Polymerase Chain Reaction (PCR) test OR Enzyme-linked immunosorbent assay (ELISA) test using sample size of 3,000 seeds or 20% of small seed lots.
- For pospiviroids (CLVd, PCFVd, PSTVd, TASVd, TCDVd, TPMVd)—PCR test using sample size of 20,000 seeds or 20% of small seed lots.

Seed lots tested off-shore must be accompanied by the laboratory test report and an official government Phytosanitary Certificate endorsed with the additional declaration that the consignment has undergone mandatory testing in accordance with Australian import conditions.

Domestic arrangements

The Australian Government is responsible for regulating the movement of goods such as plants and plant products into and out of Australia. However, the state and territory governments are responsible for plant health controls within their individual jurisdiction. Legislation relating to resource management or plant health may be used by state and territory government agencies to control interstate movement of plants and their products. After imported plants and plant products have been cleared by Australian Government biosecurity officers, they may be subject to interstate movement regulations/arrangements. It is the importer's responsibility to identify and ensure compliance with all requirements.

1.2.4 Consultation

The draft report was released on 8 August 2018 (Biosecurity Advice 2018-19 and SPS notification G/SPS/N/AUS/455) for comment by stakeholders, for a period of 60 days that concluded on 8 October 2018. The department received eleven written technical submissions on the draft report.

All submissions were carefully considered and, where relevant, changes were made to the final report. A summary of key stakeholder comments and the department's responses are provided in Appendix D.

Prior to the implementation and revision of emergency measures, the requirements were published on the department's website and the department's former Import Conditions system (ICON). The department also received comments from stakeholders when the emergency measures were implemented and revised.

Between 2008 and 2013, the department communicated with tomato growers, importers, seed trading businesses, and the National Plant Protection Organisations (NPPOs) of seed exporting countries about the introduction and revision of the emergency measures. Interested parties were provided with information by letter or email, and discussions were held by telephone, by teleconference or in face-to-face meetings.

Australia notified the World Trade Organisation (WTO) of the emergency measures in May 2008 (WTO notification G/SPS/N/AUS/225) and notified the WTO and the International Plant Protection Convention (IPPC) of the amendments to the emergency measures in February, May and November 2012 (G/SPS/N/AUS/225/Add.1; G/SPS/N/AUS/225/Add.2; G/SPS/N/AUS/225/Add.3).

The Australian Seed Federation (ASF) was informed of the emergency measures prior to their introduction in 2008. Approximately seven months before the emergency measures were amended in 2012, the department wrote to tomato seed importers and tomato fruit producers to explain the proposed emergency measures and the reasons for the changes. In 2011 teleconferences were held at which seed importers, including Australian representatives of major seed companies, and other stakeholders commented and asked questions about the department's planned revisions. The ASF and seed businesses communicated directly with the department in response to the emergency measures on several occasions, as did representatives of seed businesses.

In 2011 and 2012, the International Seed Federation (ISF) commented on the emergency measures. The department responded formally, providing further information on the emergency measures, and updating some references to ISF material in the documentation on the measures.

CCEPP meetings concerning PSTVd were held in 2008, 2011 and 2012, and CCEPP meetings concerning PCFVd were held in 2013. The Australian state and territory Chief Plant Health Managers were represented at these meetings, and the emergency measures on tomato seed were discussed and supported. Teleconferences were also held with the Chief Plant Health Managers while the department was developing the proposal for revising the emergency measures in 2011 and 2012, and the managers supported the amendments to the measures.

1.2.5 Next Steps

This final report will be published on the department's website with a notice advising stakeholders of its release. The department will also notify the WTO Secretariat about the release of the final report. The biosecurity requirements recommended in this final report will form the basis of the revised import conditions published in the Biosecurity Import Conditions (BICON) system.

2 Method for pest risk analysis

This chapter sets out the method used for the pest risk analysis (PRA) in this report. The Department of Agriculture, Water and the Environment has conducted this PRA in accordance with the International Standards for Phytosanitary Measures (ISPMs), including ISPM 2: *Framework for pest risk analysis* (FAO 2019a) and ISPM 11: *Pest risk analysis for quarantine* pests (FAO 2019c) that have been developed under the 'World Trade Organization Agreement on the Application of Sanitary and Phytosanitary Measures' (WTO 1995).

A PRA is 'the process of evaluating biological or other scientific and economic evidence to determine whether an organism is a pest, whether it should be regulated, and the strength of any phytosanitary measures to be taken against it' (FAO 2019b). A pest is 'any species, strain or biotype of plant, animal, or pathogenic agent injurious to plants or plant products' (FAO 2019b). This definition is also used in the *Biosecurity Act 2015*.

Biosecurity risk consists of two major components: the likelihood of a pest entering, establishing and spreading in Australia from imports, and the consequences should this happen. These two components are combined to give an overall estimate of the risk.

Unrestricted risk is estimated taking into account the existing commercial production practices of the exporting country and that, on arrival in Australia, the department will verify that the consignment received is as described on the commercial documents and its integrity has been maintained.

Restricted risk is estimated with phytosanitary measure(s) applied. A phytosanitary measure is 'any legislation, regulation or official procedure having the purpose to prevent the introduction and/or spread of quarantine pests, or to limit the economic impact of regulated non-quarantine pests' (FAO 2019b).

A glossary of the terms used in the risk analysis is provided at the end of this report.

The PRAs are conducted in the following three consecutive stages: initiation, pest risk assessment and pest risk management.

2.1 Stage 1 Initiation

The initiation of a pest risk analysis involves identifying the pest(s) and pathway(s) that should be considered for risk analysis in relation to the identified PRA area. According to ISPM No. 2 (FAO 2019a), a PRA process may be initiated because of:

- identification of a pathway that presents a potential pest risk (a means of pest introduction or spread)
- identification of a pest that may require phytosanitary measures (a pest may have been detected or intercepted, a request made to import it, or it may have been reported elsewhere)
- review or revision of existing phytosanitary policies and priorities; or
- identification of an organism not previously known to be a pest.

This PRA was initiated by the department to review and, if appropriate, revise the current emergency measures introduced in 2012 and 2013 (see section 1.2.3).

PepMV and TCDVd have been regulated as quarantine pests for Australia since February 2012 and May 2012, respectively, and CLVd, PCFVd, TASVd and TPMVd have been regulated as quarantine pests for Australia since November 2012.

For this pest risk analysis, the 'PRA area' is defined as Australia for CLVd, PCFVd, TASVd, TCDVd, TPMVd and PepMV.

2.2 Stage 2 Pest risk assessment

A pest risk assessment is an 'evaluation of the probability of the introduction and spread of a pest, and the magnitude of the associated potential economic consequences' (FAO 2019b). The pest risk assessment provides technical justification for identifying quarantine pests and for establishing phytosanitary import requirements.

2.2.1 Pest categorisation

Pest categorisation identifies which of the pests with the potential to be on the commodity are quarantine pests for Australia and require pest risk assessment. A 'quarantine pest' is a pest of potential economic importance to the area endangered thereby and not yet present there, or present but not widely distributed and being officially controlled (FAO 2019b).

The process of a pest categorisation is summarized by ISPM No. 11 (FAO 2019c) as a screening procedure based on the following criteria:

- identity of the pest
- presence or absence in the PRA area
- regulatory status
- potential for establishment and spread in the PRA area
- potential for economic consequences (including environmental consequences) in the PRA area.

The quarantine pests were carried forward for pest risk assessment.

2.2.2 Assessment of the probability of entry, establishment and spread

Details of how to assess the 'probability of entry', 'probability of establishment' and 'probability of spread' of a pest are given in ISPM 11 (FAO 2019c). The SPS Agreement (WTO 1995) uses the term 'likelihood' rather than 'probability' for these estimates. In qualitative PRAs, the department uses the term 'likelihood' for the descriptors it uses for its estimates of likelihood of entry, establishment and spread. The use of the term 'probability' is limited to the direct quotation of ISPM definitions.

A summary of this process is given, followed by a description of the qualitative methodology used in this risk analysis.

Likelihood of entry

The likelihood of entry describes the likelihood that a quarantine pest will enter Australia as a result of trade in a given commodity, be distributed in a viable state in the PRA area and subsequently be transferred to a host. ISPM 11 (FAO 2019c) states that the likelihood of entry of

a pest depends on the pathways from the exporting country to the destination, and the frequency and quantity of pests associated with them. ISPM 11 (FAO 2019c) lists various factors which should be taken into account when assessing the likelihood of entry.

For the purpose of considering the likelihood of entry, the department divides this step into two components:

- **Likelihood of importation**—the likelihood that a pest will arrive in Australia when a given commodity is imported.
- **Likelihood of distribution**—the likelihood that the pest will be distributed, as a result of the processing, sale or disposal of the commodity, in the PRA area and subsequently transfer to a susceptible part of a host.

Likelihood of establishment

Establishment is defined as the 'perpetuation for the foreseeable future, of a pest within an area after entry' (FAO 2019b). In order to estimate the likelihood of establishment of a pest, reliable biological information (for example, lifecycle, host range, epidemiology and survival) is obtained from the areas where the pest currently occurs. The situation in the PRA area can then be compared with that in the areas where it currently occurs, and expert judgement used to assess the likelihood of establishment.

Likelihood of spread

Spread is defined as 'the expansion of the geographical distribution of a pest within an area' (FAO 2019b). The likelihood of spread considers the factors relevant to the movement of the pest, after establishment on a host plant or plants, to other susceptible host plants of the same or different species in other areas. In order to estimate the likelihood of spread of the pest, reliable biological information is obtained from areas where the pest currently occurs. The situation in the PRA area is then carefully compared with that in the areas where the pest currently occurs and expert judgement used to assess the likelihood of spread.

Assigning likelihoods for entry, establishment and spread

Likelihoods are assigned to each step of entry, establishment and spread. Six descriptors are used: high; moderate; low; very low; extremely low; and negligible (Table 2.1). Definitions for these descriptors and their indicative probability ranges are given in Table 2.1. The indicative probability ranges are only provided to illustrate the boundaries of the descriptors and are not used beyond this purpose in qualitative PRAs. These indicative probability ranges provide guidance to the risk analyst and promote consistency between different pest risk assessments.

Likelihood	Descriptive definition	Indicative range
High	The event would be very likely to occur	0.7 < to ≤ 1
Moderate	The event would occur with an even likelihood	$0.3 < to \le 0.7$
Low	The event would be unlikely to occur	$0.05 < to \le 0.3$
Very low	The event would be very unlikely to occur	0.001 < to ≤ 0.05
Extremely low	The event would be extremely unlikely to occur	$0.000001 < to \le 0.001$
Negligible	The event would almost certainly not occur	$0 < to \le 0.000001$

Combining likelihoods

The likelihood of entry is determined by combining the likelihood that the pest will be imported into the PRA area and the likelihood that the pest will be distributed within the PRA area, using a matrix of rules (Table 2.2). This matrix is then used to combine the likelihood of entry and the likelihood of establishment, and the likelihood of entry and establishment is then combined with the likelihood of spread to determine the overall likelihood of entry, establishment and spread.

For example, if the likelihood of importation is assigned a descriptor of 'low' and the likelihood of distribution is assigned a descriptor of 'moderate', then they are combined to give a likelihood of 'low' for entry. The likelihood for entry is then combined with the likelihood assigned for establishment of 'high' to give a likelihood for entry and establishment of 'low'. The likelihood for entry and establishment of 'low'. The likelihood for entry and establishment of 'low'. The likelihood for entry and establishment is then combined with the likelihood assigned for spread of 'very low' to give the overall likelihood for entry, establishment and spread of 'very low'. This can be summarised as:

importation x distribution = entry [E]	low x moderate = low
entry x establishment = [EE]	low x high = low
[EE] x spread = [EES]	low x very low = very low

Table 2.2 Matrix of rules for combining likelihoods

	High	Moderate	Low	Very low	Extremely low	Negligible
High	High	Moderate	Low	Very low	Extremely low	Negligible
Moderate Low		Low	Very low	Extremely low	Negligible	
Low			Very low	Very low	Extremely low	Negligible
Very low				Extremely low	Extremely low	Negligible
Extremely low					Negligible	Negligible
Negligible						Negligible

Time and volume of trade

One factor affecting the likelihood of entry is the volume and duration of trade. If all other conditions remain the same, the overall likelihood of entry will increase as time passes and the overall volume of trade increases.

The department normally considers the likelihood of entry on the basis of the estimated volume of one year's trade. This is a convenient value for the analysis that is relatively easy to estimate

and allows for expert consideration of seasonal variations in pest presence, incidence and behaviour to be taken into account. The consideration of the likelihood of entry, establishment and spread and subsequent consequences takes into account events that might happen over a number of years even though only one year's volume of trade is being considered. This difference reflects biological and ecological facts, for example where a pest or disease may establish in the year of import but spread may take many years.

The use of a one-year volume of trade has been taken into account when setting up the matrix that is used to estimate the risk and therefore any policy based on this analysis does not simply apply to one year of trade. Policy decisions that are based on the department's method that uses the estimated volume of one year's trade are consistent with Australia's policy on appropriate level of protection and meet the Australian Government's requirement for ongoing quarantine protection. If there are substantial changes in the volume and nature of the trade in specific commodities then the department will review the risk analysis and, if necessary, provide updated policy advice.

2.2.3 Assessment of potential consequences

The objective of the consequence assessment is to provide a structured and transparent analysis of the potential consequences if the pests or disease agents were to enter, establish and spread in Australia. The assessment considers direct and indirect pest effects and their economic and environmental consequences. The requirements for assessing potential consequences are given in Article 5.3 of the SPS Agreement (WTO 1995), ISPM 5 (FAO 2019b) and ISPM 11 (FAO 2019c).

Direct pest effects are considered in the context of the effects on:

- plant life or health
- other aspects of the environment.

Indirect pest effects are considered in the context of the effects on:

- eradication, control
- domestic trade
- international trade
- non-commercial and environmental.

The direct and indirect consequences were estimated over four geographic levels, defined as:

Local—an aggregate of households or enterprises (a rural community, a town or a local government area).

District—a geographically or geopolitically associated collection of aggregates (generally a recognised section of a state or territory, such as 'Far North Queensland').

Regional—a geographically or geopolitically associated collection of districts in a geographic area (generally a state or territory, although there may be exceptions with larger states such as Western Australia).

National—Australia wide (Australian mainland states and territories and Tasmania).

The magnitude of the potential consequence at each of these levels was described using four categories, defined as:

Indiscernible—pest impact unlikely to be noticeable.

Minor significance—expected to lead to a minor increase in mortality/morbidity of hosts or a minor decrease in production but not expected to threaten the economic viability of production. Expected to decrease the value of non-commercial criteria but not threaten the criterion's intrinsic value. Effects would generally be reversible.

Significant—expected to threaten the economic viability of production through a moderate increase in mortality/morbidity of hosts, or a moderate decrease in production. Expected to significantly diminish or threaten the intrinsic value of non-commercial criteria. Effects may not be reversible.

Major significance—expected to threaten the economic viability through a large increase in mortality/morbidity of hosts, or a large decrease in production. Expected to severely or irreversibly damage the intrinsic 'value' of non-commercial criteria.

The estimates of the magnitude of the potential consequences over the four geographic levels were translated into a qualitative impact score (A–G) using Table 2.3. For example, a consequence with a magnitude of 'significant' at the 'district' level will have a consequence impact score of D.

	Geographic scale				
Magnitude	Local	District	Region	Nation	
Indiscernible	А	А	А	А	
Minor significance	В	С	D	Е	
Significant	С	D	Е	F	
Major significance	D	Е	F	G	

Table 2.3 Decision rules for determining the consequence impact score based on the magnitude of consequences at four geographical scales

Note: In earlier qualitative PRAs, the scale for the impact scores went from A to F and did not explicitly allow for the rating 'indiscernible' at all four levels. This combination might be applicable for some criteria. In this report, the impact scale of A to F has been changed to become B–G and a new lowest category A ('indiscernible' at all four levels) was added. The rules for combining impacts in Table 2.4 Decision rules for determining the overall consequence rating for each pest were adjusted accordingly.

The overall consequence for each pest is achieved by combining the qualitative impact scores (A–G) for each direct and indirect consequence using a series of decision rules (Table 2.4). These rules are mutually exclusive, and are assessed in numerical order until one applies.

Rule	The impact scores for consequences of direct and indirect criteria	Overall consequence rating
1	Any criterion has an impact of 'G'; or more than one criterion has an impact of 'F'; or a single criterion has an impact of 'F' and each remaining criterion an 'E'.	Extreme
2	A single criterion has an impact of 'F'; or all criteria have an impact of 'E'.	High
3	One or more criteria have an impact of 'E'; or all criteria have an impact of 'D'.	Moderate
4	One or more criteria have an impact of 'D'; or all criteria have an impact of 'C'.	Low
5	One or more criteria have an impact of 'C'; or all criteria have an impact of 'B'.	Very Low
6	One or more but not all criteria have an impact of 'B', and all remaining criteria have an impact of 'A'.	Negligible

Table 2.4 Decision rules for determining the overall consequence rating for each pest

2.2.4 Estimation of the unrestricted risk

Once the assessment of the likelihood of entry, establishment and spread, and potential consequences are completed, the unrestricted risk can be determined for each pest or groups of pests. This is determined by using a risk estimation matrix (Table 2.5) to combine the estimates of the likelihood of entry, establishment and spread and the overall consequences of pest establishment and spread. Therefore, risk is the product of likelihood and consequence.

When interpreting the risk estimation matrix, note the descriptors for each axis are similar (for example, low, moderate, high) but the vertical axis refers to likelihood and the horizontal axis refers to consequences. Accordingly, a 'low' likelihood combined with 'high' consequences, is not the same as a 'high' likelihood combined with 'low' consequences—the matrix is not symmetrical. For example, the former combination would give an unrestricted risk rating of 'moderate', whereas, the latter would be rated as a 'low' unrestricted risk.

Likelihood of pest entry,	Consequences of pest entry, establishment and spread					
establishment and spread	Negligible	Very low	Low	Moderate	High	Extreme
High	Negligible risk	Very low risk	Low risk	Moderate risk	High risk	Extreme risk
Moderate	Negligible risk	Very low risk	Low risk	Moderate risk	High risk	Extreme risk
Low	Negligible risk	Negligible risk	Very low risk	Low risk	Moderate risk	High risk
Very low	Negligible risk	Negligible risk	Negligible risk	Very low risk	Low risk	Moderate risk
Extremely low	Negligible risk	Negligible risk	Negligible risk	Negligible risk	Very low risk	Low risk
Negligible	Negligible risk	Negligible risk	Negligible risk	Negligible risk	Negligible risk	Very low risk

Table 2.5 Risk estimation matrix

2.2.5 The appropriate level of protection (ALOP) for Australia

The SPS Agreement defines the concept of an 'appropriate level of sanitary or phytosanitary protection (ALOP)' as the level of protection deemed appropriate by the WTO Member establishing a sanitary or phytosanitary measure to protect human, animal or plant life or health within its territory.

Like many other countries, Australia expresses its ALOP in qualitative terms. The ALOP for Australia, which is defined in the *Biosecurity Act 2015*, is a high level of sanitary or phytosanitary protection aimed at reducing risk to a very low level, but not to zero. The band of cells in Table 2.5 marked 'very low risk' represents the ALOP for Australia.

2.3 Stage 3 Pest risk management

Pest risk management describes the process of identifying and implementing phytosanitary measures to manage risks to achieve the ALOP for Australia, while ensuring that any negative effects on trade are minimised.

The conclusions from a pest risk assessment are used to decide whether risk management is required and if so, the appropriate measures to be used. Where the unrestricted risk estimate does not achieve the ALOP for Australia, risk management measures are required to reduce this risk to a very low level. The guiding principle for risk management is to manage risk to achieve the ALOP for Australia. The effectiveness of any recommended phytosanitary measures (or combination of measures) is evaluated, using the same approach as used to evaluate the unrestricted risk, to ensure it reduces the restricted risk for the relevant pest or pests to achieve the ALOP for Australia.

ISPM 11 (FAO 2019c) provides details on the identification and selection of appropriate risk management options and notes that the choice of measures should be based on their effectiveness in reducing the likelihood of entry of the pest.

Examples given of measures commonly applied to traded commodities include:

- options for consignments—for example, inspection or testing for freedom from pests, prohibition of parts of the host, a pre-entry or post-entry quarantine system, specified conditions on preparation of the consignment, specified treatment of the consignment, restrictions on end-use, distribution and periods of entry of the commodity
- options preventing or reducing infestation in the crop—for example, treatment of the crop, restriction on the composition of a consignment so it is composed of plants belonging to resistant or less susceptible species, harvesting of plants at a certain age or specified time of the year, production in a certification scheme
- options ensuring that the area, place or site of production or crop is free from the pest—for example, pest-free area, pest-free place of production or pest-free production site
- options for other types of pathways—for example, consider natural spread, measures for human travellers and their baggage, cleaning or disinfestations of contaminated machinery
- options within the importing country—for example, surveillance and eradication programs
- prohibition of commodities—if no satisfactory measure can be found.

Risk management measures are identified for each quarantine pest where the level of biosecurity risk does not achieve the ALOP for Australia. Relevant measures are presented in Chapter 4 of this report.

3 Pest risk assessment for quarantine pests

3.1 Introduction

This risk assessment was initiated to fulfil Australia's obligations under the IPPC and ISPM 1 (FAO 2016a) to review the current emergency phytosanitary measures implemented on tomato seed to manage the risk of *Pepino mosaic virus* (PepMV), *Columnea latent viroid* (CLVd), *Pepper chat fruit viroid* (PCFVd), *Potato spindle tuber viroid* (PSTVd), *Tomato apical stunt viroid* (TASVd), *Tomato chlorotic dwarf viroid* (TCDVd) and *Tomato planta macho viroid* (TPMVd).

3.1.1 Potato spindle tuber viroid (PSTVd)

Emergency measures for PSTVd were initiated in 2008 after several incursions of PSTVd in tomato crops (see section 1.2.3). At that time Australia was believed to be free of PSTVd, and several incursions of PSTVd were eradicated, but despite these actions the viroid became established in some areas of Australia. This status change was reported to the IPPC in 2015 (AUS-66/1).

Under the IPPC, a country may regulate a plant pest if it meets the definition of a quarantine pest or a regulated non-quarantine pest (RNQP). A quarantine pest is one of 'potential economic importance to the area endangered thereby and not yet present there, or present but not widely distributed and being officially controlled' (FAO 2019b), whereas an RNQP is one 'whose presence in plants for planting affects the intended use of those plants with an economically unacceptable impact and which is therefore regulated within the territory of the importing contracting party' (FAO 2019b). If phytosanitary measures are implemented at a national boundary based on a claim that a pest is an RNQP, the measures must be supported by a pest risk assessment that specifically addresses the requirements of the relevant ISPMs (ISPM 16 and 21), including the requirement for regulation of the pest in plants for planting within the territory.

Assessments of potential quarantine pests differ from assessments of potential RNQPs; whereas the former evaluate 'the probability of the introduction and spread of a pest and the magnitude of the associated potential economic consequences,' the latter evaluate 'the probability that a pest in plants for planting affects the intended use of those plants with an economically unacceptable impact' (FAO 2019b).

A preliminary evaluation indicates that PSTVd is seed transmitted in tomato and several other solanaceous crop species, and that trade in seed is a pathway for the introduction of the viroid. The preliminary evaluation also suggests that the viroid can become established in production systems from infected seeds and can cause economic damage, although it is probably excluded from certain Australian production systems by government and industry biosecurity activities.

In collaboration with Australian state and territory authorities, the department is evaluating the current status of PSTVd in Australia. This evaluation is being undertaken as a separate process to the finalisation of this pest risk analysis (PRA), in part because policy for PSTVd covers a broader range of hosts than tomato seed. The department intends to publish a pest risk assessment if it finds that the viroid continues to meet the criteria for regulatory control.

3.1.2 Tomato planta macho viroid (TPMVd)

TPMVd has infected tomato crops in Mexico since the disease was first observed in 1969, and has caused significant disease and yield loss (Diener 1987; Orozco Vargas & Galindo-Alonso 1986). Infected plants only produce small fruit that have no commercial value (Galindo, Smith & Diener 1982).

It is not clear that TPMVd is associated with traded tomato seed. TPMVd has been shown to be transmitted through tomato seed to seedlings in experiments at rates of up to 4.4% (Yanagisawa & Matsushima 2017), but no report of traded tomato seed contaminated with the viroid was found. One report indicated that tomato plants infected with TPMVd produced fruit with few to no seeds (Orozco Vargas & Galindo-Alonso 1986), so it is possible that seed infected with the viroid is rarely produced. This viroid has not been detected in tomato seed sent to Australia during eight years of mandatory testing of imports (Constable et al. 2019).

TPMVd was assessed in the *Draft pest risk analysis for Pepino mosaic virus and pospiviroids associated with tomato seed* and phytosanitary measures were proposed to manage risks that were thought to be presented by the viroid. After considering comments from stakeholders, the department re-evaluated the assessment of this viroid species and concluded that at this time there is insufficient evidence of association with traded tomato seed (the pathway). Accordingly, although it remains a quarantine pest for Australia, specific measures will not be recommended for this viroid on this pathway.

3.1.3 Pest risk analysis of pospiviroids and Pepino mosaic virus (PepMV)

This pest risk analysis (PRA) assesses imports of tomato seed as a potential pathway by which regulated pathogens may enter Australia. PepMV, CLVd, PCFVd, TASVd and TCDVd have been detected in tomato seed lots sent to Australia during the period of application of emergency measures. Confirming that this pathway could lead to outbreaks, there is evidence that all of the pathogens are transmitted from infected tomato seed to seedlings when the seed is germinated (Table 3.1).

Pest risk assessments are presented in sections 3.2 to 3.6. The risks are estimated for imports as if they occurred without any testing for PepMV or pospiviroids (the unrestricted risks). The department considers the risks posed by tomato seeds exported from all countries to be equivalent, taking into account the rapid international transport of tomato seed and the sources of infected tomato seed lots intercepted by Australia and other countries (Appendices A, B and C). The pest risk assessments have considered these factors as well as the epidemiology of the pathogens, data from Australian testing for the pathogens, uncertainties in the distributions of the pathogens and the complex features of tomato seed production (Appendices A, B and C).

In summary, the risk assessments and the additional information indicate the following:

- It is likely that PepMV and four pospiviroids (CLVd, PCFVd, TASVd and TCDVd) will be present in tomato seed lots sent to Australia for commercial tomato production, as international controls to prevent the production and trade of pathogen-infected tomato seeds do not adequately reduce this risk.
- Australia has intercepted tomato seed lots contaminated with PepMV-infected seeds and pospiviroid-infected seeds, including seeds infected by CLVd, PCFVd, TASVd or TCDVd. PepMV and the pospiviroids have also been detected by other countries in traded tomato

seed lots. The rates of detection show that internationally traded tomato seed lots are sometimes infected by these pathogens. Analysis of international outbreaks indicates these pathogens are transported with seed and introduced to crops through planting of infected seed.

- It is likely that these pathogens will establish and spread in Australia if no measures are taken to mitigate the risks of introduction.
- PepMV and pospiviroids are transmitted through tomato seed to seedlings, as indicated by molecular testing of seed, seed transmission experiments and information from outbreaks. Outbreaks of the pathogens in tomato crops often occur in other countries and some of these outbreaks have been linked to infected seed.
- If the regulated pathogens were to spread in Australia, substantial damage to crops and considerable control and eradication costs are likely. Infections of PepMV and pospiviroids can cause significant disease in tomato crops and reduce tomato crop yields and the quality of fruit. PCFVd also is a risk to capsicum crops and PepMV can affect pepino and basil crops.

Pathogens subject to emergency measures	Acronym	Detected in commercial tomato seed lots	Transmitted through tomato seed
Pepino mosaic virus	PepMV	Yes ^a	Yes (Córdoba-Sellés et al. 2007; Hanssen et al. 2010)
Columnea latent viroid	CLVd	Yes ^b	Yes (Matsushita & Tsuda 2016)
Pepper chat fruit viroid	PCFVd	Yes ^b	Yes (Yanagisawa & Matsushima 2017)
Tomato apical stunt viroid	TASVd	Yesb	Yes (Antignus, Lachman & Pearlsman 2007; Antignus et al. 2006)
Tomato chlorotic dwarf viroid	TCDVd	Yesb	Yes (Singh & Dilworth 2009)

Table 3.1 Summary of seed transmission and detection of listed plant pathogens

a Detected in traded tomato seed sent to Australia in 2020 and by countries in Europe (see Table A.1)

b Detected in tomato seed lots sent to Australia between 2013 and 2017 by Australian testing (see Appendix B). Seed lots that were found to be carrying the regulated pathogens were re-exported or destroyed.

3.2 Pepino mosaic virus

Pepino mosaic virus (PepMV), a member of the *Potexvirus* genus, was first identified in 1974 when it was isolated from diseased *Solanum muricatum* (pepino) in Peru (Jones, Koenig & Lesemann 1980). It was first reported in tomato in 1999 when it appeared in crops in Germany, the Netherlands, and the United Kingdom (van der Vlugt et al. 2002). An intercontinental outbreak subsequently ensued with the virus infecting tomato crops in Asia, Africa, the Americas and many countries in Europe.

The virus spreads readily in tomato crops, being mechanically transmitted by horticultural workers who become contaminated by handling infected plants (Hanssen & Thomma 2010; Jones, Koenig & Lesemann 1980; Spence et al. 2006; Wright & Mumford 1999). The virus is also transmitted by plant-to-plant contact, by the greenhouse whitefly (*Trialeurodes vaporariorum*) and bumble-bees (*Bombus impatiens*), and through water in hydroponic crops (Lacasa et al. 2003; Noël, Hance & Bragard 2014; Shipp et al. 2008).

PepMV is seed-borne and seed transmitted and has been detected many times in consignments of traded tomato seed since 2001 (Clark & Crook 2012). Although transmission from contaminated seeds to seedlings occurs infrequently, at an incidence reported to range between

2% and 0.005% (Córdoba-Sellés et al. 2007; Hanssen et al. 2010), it is believed that transmission via contaminated seeds is responsible for initiating outbreaks and for transport of the virus to regions where it was previously not known (Moreno-Pérez et al. 2014; Werkman & Sansford 2010).

Since the virus is readily transmitted, entire crops can become infected (Wright & Mumford 1999). The virus can cause wilting or stunting of plants. However, it commonly causes a range of less severe symptoms and in many instances does not cause any visible symptoms on the vegetative parts of plants (EPPO 2014d; Hanssen & Thomma 2010; Spence et al. 2006). Fruit from infected plants may be unmarketable or may be downgraded, and fruit can be affected on plants that are otherwise asymptomatic (Hanssen & Thomma 2010).

The virus also infects basil and a wide range of wild and weedy plants, including species from the Amaranthaceae, Asteraceae, Boraginaceae, Brassicaceae, Chenopodiaceae, Convolvulaceae, Malvaceae, Plantaginaceae, Polygonaceae and Solanaceae (Córdoba, Martinez-Priego & Jordá 2004; Papayiannis, Kokkinos & Alfaro-Fernández 2012; Soler et al. 2002). Infected wild plants and weeds may act as reservoirs of the virus, increasing the likelihood of establishment and spread (Córdoba, Martinez-Priego & Jordá 2004; Papayiannis, Kokkinos & Alfaro-Fernández 2012; Soler et al. 2002). The virus also infects close relatives of tomato (*Solanum lycopersicum*), some of which are cultivated and are commonly called wild tomato, namely: *Solanum chilense*, *S. chmielewskii, S. parviflorum, S. peruvianum* and *S. pimpinellifolium*.

In this pest risk assessment, a risk scenario is considered whereby PepMV enters Australia in tomato seed, the seed is planted, and the virus is transmitted within a tomato crop and to other hosts.

3.2.1 Likelihood of entry

The likelihood of entry is considered in two parts, the likelihood of importation (the likelihood that PepMV will arrive when host tomato seeds for sowing are imported) and the likelihood of distribution (the likelihood that PepMV will be viable and be transferred to a suitable host in Australia).

Likelihood of importation

The likelihood that PepMV will be imported on host tomato seeds for sowing is assessed as **High**.

PepMV has been detected in many traded tomato seed lots, and the virus has been detected in crops in many countries, including seed-producing countries. Australian laboratories have detected the virus in imported tomato seed lots during the period of application of emergency measures. Additionally, large volumes of tomato seed are imported into Australia each year and planted.

The supporting evidence for this assessment is provided.

• PepMV is present in countries in Asia, Africa, the Americas and Europe, but there is uncertainty about the details of its geographic distribution (CABI 2020a; EPPO 2000b, 2001b; Roggero 2001; Werkman & Sansford 2010), partly because outbreaks can go unreported and the pathogen is moved with seed trade (Appendix A).

- Seed producers may be unaware that fruit selected for seed extraction is infected with PepMV. PepMV infected plants may be asymptomatic or exhibit symptoms similar to those caused by other pathogens (Clark & Crook 2012).
- PepMV is easily spread by standard crop handling procedures. It is spread when tools, hands and clothing become contaminated and by direct plant-to-plant contact (Hanssen & Thomma 2010; Spence et al. 2006; Wright & Mumford 1999). The virus may be spread from weeds to seed crops and from fruit production crops to seed crops.
- PepMV was detected in the seed coat fraction in both immature and mature tomato seeds, but not in the embryo (Ling 2008).
- Tomato seeds are cleaned using standard processes during or after extraction. This cleaning process is thought to substantially reduce the quantity of virus inoculum (Córdoba-Sellés et al. 2007; EPPO 2005c; Hanssen et al. 2010). Current evidence suggests cleaning does not eliminate the virus from large batches of PepMV-contaminated seeds (Córdoba-Sellés et al. 2007; Hanssen et al. 2010), and no study was found during the preparation of this report showing the effect on transmission when commercial quantities of seed were cleaned.
- PepMV has been reported in tomato crops grown for seed production in Chile (Carreno 2005; Munoz et al. 2002).
- PepMV-infected seeds were detected in two commercially traded tomato seed lots sent to Australia. One of the lots was produced in Central America and the other lot was produced in Europe.
- The European Plant Protection Organisation has reported PepMV contaminated tomato seed lots in consignments from Chile, China, India, Israel, Italy, the Netherlands, Senegal, Thailand, USA and Vietnam (Clark & Crook 2012).
- Large quantities of tomato seed are imported into Australia annually from suppliers in many countries, including from countries that are known to have PepMV in tomatoes. The department's records indicate that on average 760kg of tomato seed are imported into Australia annually.
- PepMV-infected seeds contaminating a seed consignment cannot be detected by visual inspection.

Likelihood of distribution

To have an impact a pest must be transported in or on a pathway and must then be capable of transferring to a suitable host. The likelihood of this transfer occurring depends on the dispersal mechanisms of the pest and the intended use of the commodity.

The likelihood that PepMV will be distributed across Australia on imported tomato seeds for sowing and be transferred from the resulting plants to a suitable host is assessed as **High**.

This assessment is made primarily because imported tomato seed is distributed for planting throughout Australia, and if PepMV-infected seeds are present in an imported seed lot, it is likely the seeds and virus will remain viable.

- Tomato seed is imported for planting for greenhouse and field production of tomato fruit. Seed will be distributed for planting in domestic gardens and in nurseries, greenhouses, and farms in production areas throughout Australia.
- If present, PepMV is very likely to be present in an infectious state in the seed when it is planted.

• PepMV is likely to survive in seed for long periods of time as the virus particles are very stable. PepMV can survive more than 90 days in dried plant material (Blancard 2012).

Overall likelihood of entry (importation × distribution)

The likelihoods of importation and distribution of PepMV are combined to give an overall likelihood of entry using the matrix of rules for combining likelihoods (Table 2.2).

The overall likelihood that PepMV will enter Australia and be transferred to a suitable host via tomato seed for sowing is assessed as **High**.

3.2.2 Likelihood of establishment

The likelihood that PepMV will establish within Australia will depend upon the availability of host plants and the reproductive and survival strategies of the virus. Based on an evaluation of these factors, the likelihood of establishment is assessed as **High**.

This assessment is made primarily because very large numbers of imported tomato seeds are sown in Australia, because PepMV is transmitted from infected seeds to seedlings, and because there is evidence of the virus establishing in tomato crops and weeds in other countries.

- Since 1999 the virus has become established in many countries in Africa, Asia, Europe and the Americas (CABI 2020b).
- PepMV is transmitted from tomato seeds to seedlings (Carreno 2005).
- The rate of PepMV transmission via seeds depends on the time of seed harvest, the tomato variety, and seed cleaning and disinfection methods (Córdoba-Sellés et al. 2007; Hanssen & Thomma 2010; Ling 2008). The rate may be up to 1.8% but can be as low as 0.005% after seed cleaning (Córdoba-Sellés et al. 2007; Hanssen et al. 2010).
- Millions of tomato plants are grown each year in Australia from imported seed. Although the rate of transmission of PepMV from cleaned contaminated seeds is probably very low (Córdoba-Sellés et al. 2007; Hanssen & Thomma 2010; Ling 2008), given the numbers of tomato seeds that are planted, it is probable that infected seedlings will emerge.
- An expert opinion indicated that 'one seed giving rise to an infected seedling is very likely to spread PepMV to other plants and finally infect the whole crop' (Werkman & Sansford 2010).
- PepMV infections may go unnoticed as sometimes infected plants are asymptomatic.
- PepMV infections in tomato may not be recognised because they are difficult to distinguish visually from infections of other disease agents (Blancard 2012; EPPO 2011e).
- Tomatoes are grown commercially in greenhouses and in the field in all states of Australia, including regions with temperate and tropical climates. They are grown during spring and summer in the field in the cooler regions of southern Australia, and all year round in other regions of Australia and in greenhouses.
- The climates of regions in Australia where tomatoes are grown are generally similar to the climates of the areas where PepMV has established in other countries. The environmental conditions in commercial greenhouses in Australia are very similar to those in greenhouses in countries where outbreaks of PepMV have occurred.

3.2.3 Likelihood of spread

The likelihood of spread describes the likelihood that PepMV, once having entered Australia on imported tomato seeds and become established, will spread from a point of introduction to new areas.

Based on a comparison of factors relevant to the expansion of geographic distributions of PepMV in the source and destination areas, the assessed likelihood of spread is **High**.

This assessment is made primarily because there is evidence the virus has spread widely and quite rapidly in other countries. Additionally, the virus infects weed species and it is spread by normal horticultural activities. Moreover, it may be transported inadvertently by contaminated agricultural equipment and with infected crop residues, and it is also likely to be spread by infected pollen, seed, seedlings, and by insects.

- PepMV has spread in many countries in Africa, Asia, Europe and the Americas (CABI 2020b). Surveys indicated that the virus became very widespread in Europe in the 2000s (Werkman & Sansford 2010).
- Tomato is widely grown in home gardens, greenhouses and the field in all states and territories of Australia.
- PepMV is easily spread by standard crop handling procedures. It is spread when tools, hands and clothing become contaminated or by direct plant-to-plant contact (Hanssen & Thomma 2010; Spence et al. 2006; Wright & Mumford 1999).
- PepMV could be spread to tomato crops in new areas if contaminated machinery or tools are moved between areas, and could be moved on workers' hands if people work in more than one area within a short period of time (Hanssen & Thomma 2010; Spence et al. 2006; Wright & Mumford 1999).
- PepMV also infects other cultivated plant species. PepMV infects basil, pepino and potato (CSL 2005; Davino et al. 2009; Papayiannis, Kokkinos & Alfaro-Fernández 2012).
- The virus may spread from tomato to other host plant species including weed species. PepMV was recorded in field and greenhouse tomatoes in Cyprus as well as 20 weed species in the field of that country in 2009 (Papayiannis, Kokkinos & Alfaro-Fernández 2012). The weeds infected in Cyprus included the species *Malva parviflora, Sonchus oleraceus, Solanum nigrum, Convolvulus arvensis, Chrysanthemum segetum* and *Calendula arvensis*. These weed species occur in Australia and some are widespread and abundant.
- PepMV infects plant species in the families Amaranthaceae, Asteraceae, Boraginaceae, Brassicaceae, Chenopodiaceae, Convolvulaceae, Malvaceae, Plantaginaceae, Polygonaceae and Solanaceae (Córdoba, Martinez-Priego & Jordá 2004; Papayiannis, Kokkinos & Alfaro-Fernández 2012; Soler et al. 2002). Since many wild and weedy species in these plant families exist in Australia, PepMV in tomato plants may be transmitted to weeds in a crop or weeds or wild plants growing near a crop. The virus may be sustained in the alternative hosts.
- Weeds could act as a reservoir for the virus (Córdoba, Martinez-Priego & Jordá 2004; Werkman & Sansford 2010). Infection of weeds may accelerate or consolidate the spread of the virus, as more inoculum may be present at a location when weeds are infected. This may provide more opportunities for the virus to spread.
- PepMV can be transmitted by bumble-bees which contribute to the spread of the virus between tomato plants (Lacasa et al. 2003; Shipp et al. 2008). Bumble-bees can also

transmit PepMV between different plant species, for example, from tomato plants to the weed species *Solanum ptycanthum*, *S. sarrachoides* or *Datura stramonium*, and from these species back to tomato (Stobbs et al. 2009). The bumble-bee species *Bombus terrestris* is present in Tasmania but it is not present on the mainland of Australia.

- PepMV could be transmitted through soil by the chytrid fungus *Olpidium virulentus* (Alfaro-Fernández et al. 2010). *Olpidium virulentus* is present in Australia (Maccarone et al. 2010). PepMV may be transmitted by the fungus between field-grown tomatoes and may be transmitted by the fungus in hydroponic systems via the irrigation water.
- PepMV infections may go unrecognised. The symptoms are similar to those caused by other viruses and viroids. An unrecognised infection may not be controlled or eradicated, and thus, may spread.

3.2.4 Overall likelihood of entry, establishment and spread

The overall likelihood of entry, establishment and spread is determined by combining the individual likelihoods of entry, of establishment and spread using the matrix rules for combining descriptive likelihoods (Table 2.2).

The overall likelihood that PepMV will enter Australia, be distributed in a viable state to susceptible hosts, establish in that area and subsequently spread within Australia is assessed as **High**.

3.2.5 Consequences

The consequences of entry, establishment and spread of PepMV in Australia have been estimated according to the methods described in Table 2.3.

Based on the decision rules described in Table 2.4, that is, where the potential consequences of a pest with respect to one or more (but not all) criteria have an impact of 'E', the overall consequences are estimated to be **Moderate**.

This assessment is made because the virus may cause substantial losses in tomato crops, and these losses would be amplified by spread of the virus. If there is an incursion, it is likely that eradication and control would be attempted by state and territory governments, but given the characteristics of the virus, these activities would prove costly and difficult.

Criterion	Estimate and rationale
Direct	
Plant life or health	E – significant at the regional level
	PepMV is an important pathogen of tomato crops and can cause significant yield losses. In the 2018–19 financial year, Australian tomato production was estimated to have a gross value of \$674.2 million (Horticulture Innovation Australia 2020).
	PepMV infection directly affects tomato fruit production by reducing yields and affecting the quality of fruit. In the United Kingdom, business losses from PepMV infections in tomato crops were estimated to range from £3.8 million per season to £37.5 million per season over a period of three years (Alleweldt 2011). The Australian tomato industry is several times larger than that in the United Kingdom and hence, there is the potential for greater losses in Australia.

	The incidence of infected plants varies in tomato crops, with one estimate indicating a range of incidence from $10-90\%$ (Soler-Aleixandre et al. 2005b)
	Trials have shown the effect on tomato yields and quality vary depending on virus variant (Alleweldt 2011; Peters et al. 2011; Peters et al. 2010). One variant of the virus was found to have little effect on tomato yields and quality. However other variants were found to cause a reduction in yield of about 5–15% (Peters et al. 2011). Trials have also shown yields of class 1 fruits to be reduced by certain variants by about 14% to more than 38% (Spence et al. 2006). Additional grading of tomato fruit due to PepMV infection would add to production costs.
	Fruit from infected tomato plants may be discoloured and have a marbled or mosaic appearance, or may split and become open so that the seed and flesh is exposed (Hanssen & Thomma 2010). The degree of symptoms in the fruit depends on the virus variant and probably on the cultivar of tomato (Fakhro et al. 2011; Peters et al. 2011; Peters et al. 2010).
	In some cases, PepMV-infected tomato plants only express mild symptoms on the vegetative parts or may be asymptomatic (Peters et al. 2011; Peters et al. 2010). However, in other cases, tomato plants have developed more serious symptoms including necrosis, deformed growth, wilting, plant collapse and plant death (Hanssen & Thomma 2010; Polston 2008; Soler-Aleixandre et al. 2005b).
	Symptoms and adverse effects may be worse when the virus infects tomato plants together with other pathogens. One report of plants infected with a mixed infection of PepMV and the chytrid fungus <i>Olpidium brassicae</i> indicated that wilting and collapse were common symptoms (Soler-Aleixandre et al. 2005b). A mixed infection with tomato chlorosis virus (ToCV) also produced heightened symptoms and greater losses than were expected to be induced by either pathogen alone (Davino et al. 2008). Increased effects were also noted in PepMV and <i>Verticillium</i> sp. co-infected plants (Mumford & Jones 2005).
	PepMV has been reported to infect basil in Sicily, causing chlorosis on young leaves (Davino et al. 2009). However, no significant economic yield loss has been recorded in this crop.
Other aspects of the	D – minor significance at the regional level
environment	PepMV also infects species from the Amaranthaceae, Asteraceae, Brassicaceae, Boraginaceae, Chenopodiaceae, Convolvulaceae, Malvaceae, Plantaginaceae, Polygonaceae and Solanaceae (Córdoba, Martinez-Priego & Jordá 2004; Papayiannis, Kokkinos & Alfaro-Fernández 2012). There are Australian native species of plants in all these families, and many other species in these families are naturalised weeds in Australia. Some of these wild and naturalised plant species may be infected by PepMV and their abundance or health might be affected.
environment Indirect	PepMV also infects species from the Amaranthaceae, Asteraceae, Brassicaceae, Boraginaceae, Chenopodiaceae, Convolvulaceae, Malvaceae, Plantaginaceae, Polygonaceae and Solanaceae (Córdoba, Martinez-Priego & Jordá 2004; Papayiannis, Kokkinos & Alfaro-Fernández 2012). There are Australian native species of plants in all these families, and many other species in these families are naturalised weeds in Australia. Some of these wild and naturalised plant species may be infected by PepMV and their abundance or health might be affected.
environment Indirect Eradication, control	PepMV also infects species from the Amaranthaceae, Asteraceae, Brassicaceae, Boraginaceae, Chenopodiaceae, Convolvulaceae, Malvaceae, Plantaginaceae, Polygonaceae and Solanaceae (Córdoba, Martinez-Priego & Jordá 2004; Papayiannis, Kokkinos & Alfaro-Fernández 2012). There are Australian native species of plants in all these families, and many other species in these families are naturalised weeds in Australia. Some of these wild and naturalised plant species may be infected by PepMV and their abundance or health might be affected. E – major significance at the district level If an incursion of PepMV was to occur in an Australian tomato crop it is likely that eradication would be attempted. In the past decade several incursions of pospiviroids in tomato crops have been eradicated.
environment Indirect Eradication, control	 PepMV also infects species from the Amaranthaceae, Asteraceae, Brassicaceae, Boraginaceae, Chenopodiaceae, Convolvulaceae, Malvaceae, Plantaginaceae, Polygonaceae and Solanaceae (Córdoba, Martinez-Priego & Jordá 2004; Papayiannis, Kokkinos & Alfaro-Fernández 2012). There are Australian native species of plants in all these families, and many other species in these families are naturalised weeds in Australia. Some of these wild and naturalised plant species may be infected by PepMV and their abundance or health might be affected. E – major significance at the district level If an incursion of PepMV was to occur in an Australian tomato crop it is likely that eradication would be attempted. In the past decade several incursions of pospiviroids in tomato crops have been eradicated. Spread in field crops is likely to be difficult to control, and lead to greater costs because the virus may also infect wild and weedy plants. This appears to have happened in Europe where endemic weed species have been reported as natural alternative hosts, (Córdoba, Martinez-Priego & Jordá 2004; OEPP-EPPO 2009), and these plants may have become reservoirs that perpetuate PepMV infection. If weeds are infected, eradication and control of PepMV may be more costly and less likely to succeed.
environment Indirect Eradication, control	 PepMV also infects species from the Amaranthaceae, Asteraceae, Brassicaceae, Boraginaceae, Chenopodiaceae, Convolvulaceae, Malvaceae, Plantaginaceae, Polygonaceae and Solanaceae (Córdoba, Martinez-Priego & Jordá 2004; Papayiannis, Kokkinos & Alfaro-Fernández 2012). There are Australian native species of plants in all these families, and many other species in these families are naturalised weeds in Australia. Some of these wild and naturalised plant species may be infected by PepMV and their abundance or health might be affected. E – major significance at the district level If an incursion of PepMV was to occur in an Australian tomato crop it is likely that eradication would be attempted. In the past decade several incursions of pospiviroids in tomato crops have been eradicated. Spread in field crops is likely to be difficult to control, and lead to greater costs because the virus may also infect wild and weedy plants. This appears to have happened in Europe where endemic weed species have been reported as natural alternative hosts, (Córdoba, Martinez-Priego & Jordá 2004; OEPP-EPPO 2009), and these plants may have become reservoirs that perpetuate PepMV infection. If weeds are infected, eradication and control of PepMV may be more costly and less likely to succeed. PepMV is not eliminated when infected plants are killed with herbicide, so to control the virus and prevent spread, infected plants are killed with herbicide, so to control the virus and prevent spread, infected plants in a crop by surveying and testing, but if the aim is eradication and the virus spreads quickly, the entire crop may need to be cleaned before it could be used again. Equipment would need to be sterilized using bleach. Typically, when eradication is attempted, plants, propagating material, machinery and implements may not be moved from properties where an outbreak has been detected.

	If PepMV became established in an Australian state, restrictions might be introduced on the interstate trade of affected propagative material, including seed. These restrictions could lead to loss of domestic markets.
	Tomato seed, seedlings and transplants are traded across Australia. This trade might be interrupted if an outbreak of PepMV occurred. It is suspected that the international outbreak of PepMV and its spread across Europe was due in part to trade in infected seed (Córdoba-Sellés et al. 2007).
International trade	D – significant at the district level
	Australia exports a a very small proportion of its fresh tomato fruit crop, which might be affected if PepMV became established in Australia. Australia has markets for fresh tomatoes to New Zealand, Singapore, Hong Kong, Brunei-Darussalam, Malaysia, New Caledonia, Indonesia, French Polynesia, Fiji, and USA (DAWE 2020; HAL 2012).
Non-commercial and environmental	A –indiscernible at the local level
	No evidence was found indicating environmental and non-commercial indirect effects.

3.2.6 Unrestricted risk estimate

The unrestricted risk estimate for PepMV is **Moderate**. Likelihoods and consequences are combined using the risk estimation matrix shown in Table 2.5. A summary of the risk estimates leading to this unrestricted risk estimate is given in Table 3.2 in Section 3.7.

3.3 Columnea latent viroid

Columnea latent viroid (CLVd) was found in several cultivars of tomato in the Netherlands in 1989, 1993 and 1995, and in tomato samples from Belgium in 1996 (EPPO 2005a; Verhoeven et al. 2004). It was identified in tomato plants from Portugal in 2006, and was confirmed in nurseries producing tomato plants in England in 2007 (Monger & Mumford 2006; Nixon et al. 2009). CLVd was also reported in France in 2007 (CSL 2007). Tomato plants infected with CLVd develop disease symptoms including stunting, chlorosis, bronzing and leaf distortion (NAPPO 2007).

CLVd is transmitted through tomato seeds to seedlings (FERA 2009b; Marach 2008; Matsushita & Tsuda 2016). Previous outbreaks in greenhouses are suspected to have been introduced with infected seed (FERA 2009b; Verhoeven, Hammond & Stancanelli 2017). The viroid is mechanically transmitted, and it has been reported to spread rapidly through greenhouse tomato crops, likely by plant-to-plant contact and through normal horticultural activities when agricultural equipment becomes contaminated (FERA 2009b).

In this pest risk assessment, a risk scenario is considered whereby CLVd enters Australia in tomato seed, the infected seed is planted, and the viroid is transmitted within a tomato crop and to other hosts.

3.3.1 Likelihood of entry

The likelihood of entry is considered in two parts, the likelihood of importation (the likelihood that CLVd will arrive when host tomato seeds for sowing are imported) and the likelihood of distribution (the likelihood that CLVd will be viable and be transferred to a suitable host in Australia).

Likelihood of importation

The likelihood that CLVd will be imported on host tomato seeds for sowing is assessed as **High**.

This assessment is made because CLVd has been detected in tomato seed lots sent to Australia during the period of the application of emergency measures, and the viroid has been reported in crops in many countries, including tomato seed producing countries. Additionally, large volumes of tomato seed are imported into Australia each year.

The supporting evidence for this assessment is provided.

- CLVd has been found in tomato crops in Asia (Thailand), Europe (United Kingdom, Belgium, France, the Netherlands, Germany and Portugal) and North America (Canada and USA) (CSL 2008; Hadidi et al. 2003; Mumford et al. 2006; NCBI 2007; Steyer et al. 2009; Verhoeven et al. 2004; Werkman, Verhoeven & Roenhorst 2007).
- Pospiviroids infect tomato plants systemically. Pospiviroid RNA has been found in the sap, leaves, roots, sepals, petals, ovaries, stamens and seed of tomato (Antignus, Lachman & Pearlsman 2007; Koenraadt et al. 2009; Lykke et al. 2010; Singh & Dilworth 2009; Singh et al. 2006; Singh, Nie & Singh 1999; Zhu et al. 2001). Pospiviroid RNA has been detected in the embryonic tissues of the seed in some hosts (Antignus, Lachman & Pearlsman 2007; EPPO 2016b; Matsushita & Tsuda 2014b).
- Seed extracted from tomato fruit naturally infected with CLVd has been found to carry CLVd RNA (FERA 2009b).
- CLVd was detected in 13 lots of tomato seed sent to Australia from the Middle East and Asia between February 2012 and October 2013.
- Large quantities of tomato seed are imported into Australia annually from suppliers in many countries for the production of tomato crops. The department's records indicate that on average 760kg of tomato seed are imported into Australia annually.

Likelihood of distribution

To have an impact a pest must be transported in or on a pathway and must then be capable of transferring to a suitable host. The likelihood of this transfer occurring depends on the dispersal mechanisms of the pest and the intended use of the commodity.

The likelihood that CLVd will be distributed across Australia on imported tomato seeds for sowing and be transferred from the resulting plants to a suitable host is assessed as **High**.

This assessment is made primarily because imported tomato seed is distributed throughout Australia for sowing and if CLVd-infected seeds are present in an imported seed lot, it is likely the seeds and virus will remain viable.

- Tomato seed is imported for planting for greenhouse and field production of tomato fruit. Seed will be distributed to greenhouses and farms in production areas throughout Australia, and for planting in domestic gardens.
- It is very likely CLVd will survive in tomato seed for long periods. The related viroid PSTVd has been found to endure for many years in seed when stored at room temperature (Singh, Boucher & Wang 1991).
- CLVd will be distributed with the seed to production areas throughout Australia and, if present, is very likely to be present in an infectious state in the seed when it is planted.

Overall likelihood of entry (importation × distribution)

The likelihoods of importation and distribution of CLVd are combined to give an overall likelihood of entry using the matrix of rules for combining likelihoods (Table 2.2).

The overall likelihood that CLVd will enter Australia and be transferred to a suitable host via tomato seed for sowing is assessed as **High**.

3.3.2 Likelihood of establishment

The likelihood that CLVd will establish within Australia will depend upon the availability of host plants and the reproductive and survival strategies of the viroid. Based on an evaluation of these factors, the likelihood of establishment is assessed as **High**.

This assessment is made primarily because very large numbers of imported tomato seeds are sown in Australia, CLVd is transmitted from infected seed to seedlings and there is evidence of the viroid establishing in tomato crops in other countries.

- CLVd has been found in Thailand and in several countries in Europe and North America (CSL 2008; Hadidi et al. 2003; Mumford et al. 2006; NCBI 2007; Steyer et al. 2009; Verhoeven et al. 2004; Werkman, Verhoeven & Roenhorst 2007). CLVd has been found in tomato plants for seed production in an open field in Thailand (Tangkanchanapas et al. 2005).
- CLVd is transmitted through tomato seed to seedlings (Marach 2008). Some seedlings grown from CLVd-infected tomato seed are thus likely to be infected by the viroid. The germination rates of tomato seed infected with CLVd may be reduced, as has been reported for tomato seed infected with other pospiviroids (Benson & Singh 1964).
- Millions of tomato plants are grown each year in Australia from imported seed.
- Outbreaks of CLVd in tomato crops in the United Kingdom in 2007 are believed to have been caused by infected seed, since seed was the common factor linking the outbreaks (FERA 2009b).
- CLVd is mechanically transmitted from plant-to-plant by contaminated cutting tools and machinery, on worker's hands and by plant-to-plant contact (FERA 2009b).
- CLVd may spread in greenhouse crops before disease symptoms become apparent or are recognised. Infected tomato plants in a field crop are less likely to be detected because of the lower intensity of horticultural activity in these crops.
- CLVd may survive between tomato crops in contaminated greenhouses and in infected crop residues and infect plants subsequently grown in the vicinity in later seasons. The related viroid PSTVd is stable under most environmental conditions and can remain infective in hydrated plant material for several months and in dry material for over a year, and is expected to survive composting processes (Mikkelsen, Elphinstone & Jensen 2005).
- Tomatoes are grown commercially in greenhouses and fields in all states of Australia, including regions with tropical and temperate climates. They are grown during the spring and summer in the field in the cooler regions of southern Australia, and all year round in other regions of Australia and in greenhouses.
- The climates of these regions in Australia where tomatoes are grown are generally similar to the climates of the areas where CLVd has been found. The environmental conditions in commercial greenhouses in Australia are very similar to those in greenhouses in countries where outbreaks of CLVd have occurred.

- It is possible that CLVd may infect solanaceous weeds in the vicinity of infected greenhouses and field crops and become established in an area.
- CLVd outbreaks in tomato crops have been reported from the Netherlands and Belgium (EPPO 2005a; Verhoeven et al. 2004), Portugal (Monger & Mumford 2006), England (Nixon et al. 2009) and France (CSL 2007).

3.3.3 Likelihood of spread

The likelihood of spread describes the likelihood that CLVd, once having entered Australia on imported tomato seeds and become established, will spread from a point of introduction to new areas.

Based on a comparison of factors relevant to the potential expansion of the geographic range of the pathogen in the source and destination areas, the likelihood that CLVd will spread is assessed as **Moderate**.

This assessment is made because the viroid is spread by normal horticultural activities, and it may also be dispersed with trade in tomato seed and seedlings, and with the disposal of crop residues. The viroid might also be transmitted by bumble-bees and certain aphids.

The supporting evidence for this assessment is provided.

- Tomato is widely grown in home gardens, greenhouses and the field in all states and territories of Australia.
- Pospiviroids can spread rapidly through greenhouse crops because of the density of planting and the intensity of horticultural activity (Diener 1971; Hammond & Owens 2006).
- CLVd could be spread to tomato crops in new areas if contaminated machinery or tools are moved between areas, and could be moved on worker's hands if people work in more than one area within a short period of time (FERA 2009a).
- Transport of crop residues infected with CLVd to new areas and their disposal near tomato, weed or ornamental hosts could result in the mechanical transfer of the viroid to these hosts and result in its spread.
- CLVd could be introduced into new areas in tomato seed, as it can be present in seed and seed to seedling transmission has been demonstrated (FERA 2009b; Matsushita & Tsuda 2016).
- Seedlings are moved over long distances within Australia for planting and infected seedlings could introduce CLVd into new areas.
- CLVd could also be transmitted by plant-feeding insects or in pollen, as other pospiviroids are sometimes transmitted by these mechanisms (Antignus, Lachman & Pearlsman 2007; EPPO 1997; Fernow, Peterson & Plaisted 1970; Kryczynski, Paduch-Cichal & Skreczkowski 1988; Querci et al. 1997; Singh, Boucher & Somerville 1992; Syller, Marczewski & Pawlowicz 1997).

3.3.4 Overall likelihood of entry, establishment and spread

The overall likelihood of entry, establishment and spread is determined by combining the likelihoods of entry, of establishment and of spread using the matrix rules for combining descriptive likelihoods (Table 2.2).

The overall likelihood that CLVd will enter Australia, be distributed in a viable state to susceptible hosts, establish in that area and subsequently spread within Australia is assessed as **Moderate**.

3.3.5 Consequences

The consequences of entry, establishment and spread of CLVd in Australia have been estimated according to the methods described in Table 2.3.

Based on the decision rules described in Table 2.4, that is, where the potential consequences of a pest with respect to one or more (but not all) criteria have an impact of 'D', the overall consequences are estimated to be **Low**.

This assessment is made because the viroid may cause substantial losses in tomato crops, and these losses would be amplified by spread of the viroid. If there is an incursion, it is likely that eradication and control would be attempted by state and territory governments, but given the characteristics of the viroid, these activities could prove costly and difficult.

Criterion	Estimate and rationale			
Direct				
Plant life or health	D – significant at the district level			
	CLVd is an important pathogen of tomato crops and potentially can cause significant yield losses. In the 2018–19 financial year, Australian tomato production was estimated to have a gross value of \$674.2 million (Horticulture Innovation Australia 2020).			
	During an outbreak of CLVd in the United Kingdom, 50–60% of a tomato crop became infected and symptoms were severe, which include severe leaf distortion, bronzing and 'crunchy' leaf (FERA 2009b; Nixon et al. 2009; Sansford & Morris 2009). Yield losses of tomato fruit of 26–100% have been measured in experiments (Marach 2008).			
Other aspects of the environment	C – minor significance at the district level			
	CLVd may infect other plant species of the family Solanaceae in addition to tomato. Native and naturalised Solanaceae are components of Australian ecosystems that might be infected by CLVd and their abundance or health might be affected.			
Indirect				
Eradication, control	D – significant at the district level			
	If an incursion of CLVd were to occur in a tomato crop, and it was detected, it would probably be controlled by eradication. The extent of an outbreak will depend on many factors including the transmission characteristics of the viroid variant, environmental conditions, the transport of plants and machinery between properties, the movement of workers and the activity of insect vectors, if present.			
	CLVd infections may go unrecognised because a number of other viroid species produce symptoms identical or very similar to those induced by CLVd (Hammond, Smith & Diener 1989; Verhoeven et al. 2004). An outbreak of CLVd may not be recognised or reported until it has spread to several crops, properties and species.			
	During an outbreak, the infected property and adjoining and nearby properties will be surveyed. Samples from surveys will be tested in state government laboratories. RT-PCR will be used to test the samples and detect the viroid. Surveillance and laboratory tests are costly.			
	Viroids are not eliminated when infected plants are killed so infected plant material is buried. Equipment is sterilized using bleach. Typically, plants, propagating material, machinery and implements may not be moved from properties where outbreaks have been detected.			
Domestic trade	C – minor significance at the district level			
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	If CLVd became established in an Australian state, restrictions might be introduced on the interstate trade of affected seed and seedlings. If this occurred it could lead to the loss of markets.			
	No movement of machinery from the affected properties is permitted during an eradication campaign.			
International trade	D – significant at the district level			
	Australia exports a proportion of its fresh tomato fruit crop, which might be affected if CLVd became established in Australia. Australia has markets for fresh tomatoes to New Zealand, Singapore, Hong Kong, Brunei-Darussalam, Malaysia, New Caledonia, Indonesia, French Polynesia, Fiji, and USA (DAWE 2020; HAL 2012).			
Non-commercial and	A –indiscernible at the local level			
environmental	No evidence was found indicating environmental and non-commercial indirect effects.			

3.3.6 Unrestricted risk estimate

The unrestricted risk estimate for CLVd is **Low**. Likelihoods and consequences are combined using the risk estimation matrix shown in Table 2.5. A summary of the risk estimates leading to this unrestricted risk estimate is given in Table 3.2 in Section 3.7.

3.4 Pepper chat fruit viroid

Pepper chat fruit viroid (PCFVd) is a relatively newly identified pospiviroid that causes disease in tomato and capsicum (*Capsicum annuum* L.). It was first reported in greenhouse capsicum crops in the Netherlands in 2006 and reoccurred in 2007 in the same species and location (Verhoeven et al. 2009). In 2009, it was isolated from tomato plants in Thailand, and in 2010 in capsicum plants in Canada (Punyapitak 2004; Reanwarakorn, Klinkong & Porsoongnurn 2011; Verhoeven et al. 2011). It was suspected that the viroid had been present in tomato in Thailand for several years. Infected tomato plants are stunted, the leaves becoming necrotic, distorted and discoloured, and the fruit are small.

PCFVd is transmitted through tomato seed to seedlings at rates of up to 1.4% (Yanagisawa & Matsushima 2017). Sequenced isolates published in GenBank from Thailand are 95–99% similar to sequences from outbreaks in Canada and the Netherlands. The geographic distribution of the viroid suggests it was probably transported to Canada and the Netherlands in infected seed.

In this pest risk assessment, a risk scenario is considered whereby PCFVd enters Australia in tomato seed, the seed is planted, and the viroid is transmitted within a tomato crop and to other hosts.

3.4.1 Likelihood of entry

The likelihood of entry is considered in two parts, the likelihood of importation (the likelihood that PCFVd will arrive when host tomato seeds for sowing are imported) and the likelihood of distribution (the likelihood that PCFVd will be viable and be transferred to a suitable host in Australia).

Likelihood of importation

The likelihood that PCFVd will be imported on host tomato seeds for sowing is assessed as **High**.

This assessment is made because PCFVd has been detected in tomato seed lots sent to Australia during the period of application of the emergency measures, and the viroid has been reported in several countries, including tomato seed producing countries. Additionally, large volumes of tomato seed are imported into Australia each year.

The supporting evidence for this assessment is provided.

- PCFVd has been found in tomato in Thailand and in capsicum from the Netherlands and Canada (Punyapitak 2004; Reanwarakorn, Klinkong & Porsoongnurn 2011; Verhoeven et al. 2011; Verhoeven et al. 2009). It has been found both in greenhouse and field-grown plants.
- Pospiviroids infect tomato plants systemically. Pospiviroid RNA has been found in the sap, leaves, roots, sepals, petals, ovaries, stamens and seed of tomato (Antignus, Lachman & Pearlsman 2007; Koenraadt et al. 2009; Lykke et al. 2010; Singh & Dilworth 2009; Singh et al. 2006; Singh, Nie & Singh 1999; Zhu et al. 2001). Pospiviroid RNA has been detected in the embryonic tissues of the seed of some hosts (Antignus, Lachman & Pearlsman 2007; EPPO 2016b; Matsushita & Tsuda 2014b).
- PCFVd was detected in eleven lots of tomato seed sent to Australia and tested on arrival between February 2012 and October 2013, with infected seed produced in two countries in Europe, one in the Middle East and two countries in Asia.
- Large quantities of tomato seed are imported into Australia annually from suppliers in many countries for the production of tomato crops. The department's records indicate that on average 760kg of tomato seed are imported into Australia annually.

Likelihood of distribution

To have an impact a pest must be transported in or on a pathway and must then be capable of transferring to a suitable host. The likelihood of this transfer occurring depends on the dispersal mechanisms of the pest and the intended use of the commodity.

The likelihood that PCFVd will be distributed across Australia on imported tomato seeds for sowing and be transferred from the resulting plants to a suitable host is assessed as **High**.

This assessment is made primarily because imported tomato seed is distributed for planting throughout Australia and, if PCFVd-infected seeds are present in an imported seed lot, it is likely the seeds and viroid will remain viable.

The supporting evidence for this assessment is provided.

- Tomato seed is imported for planting for greenhouse and field production of tomato fruit. Seed will be distributed for planting to greenhouses and farms in production areas throughout Australia, and for planting in domestic gardens.
- It is very likely PCFVd will survive in tomato seed for long periods. The related viroid PSTVd has been found to endure for many years in seed when stored at room temperature (Singh, Boucher & Wang 1991).
- PCFVd will be distributed with the seed to production areas throughout Australia, and if present, is very likely to be present in an infectious state in the seed when it is planted.

Overall likelihood of entry (importation × distribution)

The likelihoods of importation and distribution of PCFVd are combined to give an overall likelihood of entry using the matrix of rules for combining likelihoods (Table 2.2).

The overall likelihood that PCFVd will enter Australia and be transferred to a suitable host via tomato seed for sowing is assessed as **High**.

3.4.2 Likelihood of establishment

The likelihood that PCFVd will establish within Australia will depend upon the availability of host plants and the reproductive and survival strategies of the viroid. Based on an evaluation of these factors, the likelihood of establishment is assessed as **High**.

This assessment is made primarily because very large numbers of imported tomato seeds are sown in Australia, PCFVd is transmitted from infected seed to seedlings, and there is evidence of the viroid establishing in tomato crops in other countries.

- PCFVd has been found in Thailand, the Netherlands and Canada (Punyapitak 2004; Reanwarakorn, Klinkong & Porsoongnurn 2011; Verhoeven et al. 2011; Verhoeven et al. 2009).
- Some seedlings grown from PCFVd-infected tomato seed are very likely to be infected by the viroid, as seed to seedling transmission has been demonstrated for PCFVd (Yanagisawa & Matsushima 2017). The viroid may be seed transmitted in capsicum (Verhoeven et al. 2009).
- The germination rates of tomato seed infected with PCFVd may be reduced, as has been reported for tomato seed infected with other pospiviroids (Benson & Singh 1964).
- Millions of tomato plants are grown each year in Australia from imported seed.
- PCFVd is mechanically transmitted in tomato (Reanwarakorn, Klinkong & Porsoongnurn 2011; Verhoeven et al. 2009). Pospiviroids are spread mechanically from infected plants by contaminated agricultural equipment and on worker's hands during horticultural activities and by plant-to-plant contact (Conde, Connelly & Pitkethley 1996; Diener 1971; Hammond & Owens 2006; Horticulture New Zealand 2008; Manzer & Merriam 1961; Singh & Dhar 1998).
- PCFVd may spread in greenhouse crops before disease symptoms become apparent or are recognised. Infected tomato plants in a field crop are less likely to be detected because of the lower intensity of horticultural activity in these crops.
- Tomatoes are grown commercially in greenhouses and in the field in all states of Australia, including regions with tropical and temperate climates. They are grown during the spring and summer in the field in the cooler regions of southern Australia, and all year round in other regions of Australia and in greenhouses.
- The climates of these regions in Australia where tomatoes are grown are generally similar to the climates of the areas where PCFVd has been found. The environmental conditions in commercial greenhouses in Australia are very similar to those in greenhouses in countries where outbreaks of PCFVd have occurred.
- It is possible that PCFVd may infect solanaceous weeds in the vicinity of infected greenhouses and field crops and become established in an area.
- An incursion of PCFVd in a greenhouse tomato crop in South Australia was eradicated. The incursion was the first record of PCFVd in Australia, suggesting it was introduced in tomato seed, and that seed to seedling transmission occurred.

3.4.3 Likelihood of spread

The likelihood of spread describes the likelihood that PCFVd, once having entered Australia on imported tomato seeds and become established, will spread from a point of introduction to new areas.

Based on a comparison of factors relevant to the expansion of geographic distributions of PCFVd in the source and destination areas, the assessed likelihood of spread is **Moderate**.

This assessment is made because the viroid is spread by normal horticultural activities, and it may also be dispersed with trade in tomato seed and seedlings, and with the disposal of crop residues. The viroid might also be transmitted by bumble-bees and certain aphids. Tomato is widely grown in Australia in home gardens, greenhouses and the field in all states and territories of Australia.

The supporting evidence for this assessment is provided.

- Pospiviroids can spread rapidly through greenhouse crops because of the density of planting and the intensity of horticultural activity (Diener 1971; Hammond & Owens 2006).
- PCFVd could be spread to tomato crops in new areas if contaminated machinery or tools are moved between areas, and could be moved on worker's hands if people work in more than one area within a short period of time (Reanwarakorn, Klinkong & Porsoongnurn 2011; Verhoeven et al. 2009).
- Transport of crop residues infected with PCFVd to new areas and their disposal near tomato, weed or ornamental hosts could result in the mechanical transfer of the viroid to these hosts and result in its spread.
- PCFVd could be introduced into new areas in tomato seed, as it can be present in seed and seed to seedling transmission has been demonstrated (Singh & Dilworth 2009).
- Seedlings are moved over long distances within Australia for planting and infected seedlings could introduce PCFVd into new areas.
- PCFVd could also be transmitted by plant feeding insects or in pollen, as other pospiviroids are sometimes transmitted by these mechanisms (Antignus, Lachman & Pearlsman 2007; EPPO 1997; Fernow, Peterson & Plaisted 1970; Kryczynski, Paduch-Cichal & Skreczkowski 1988; Querci et al. 1997; Singh, Boucher & Somerville 1992; Syller, Marczewski & Pawlowicz 1997).

3.4.4 Overall likelihood of entry, establishment and spread

The overall likelihood of entry, establishment and spread is determined by combining the individual likelihoods of entry, of establishment and spread using the matrix rules for combining descriptive likelihoods (Table 2.2).

The overall likelihood that PCFVd will enter Australia, be distributed in a viable state to susceptible hosts, establish in that area and subsequently spread within Australia is assessed as **Moderate**.

3.4.5 Consequences

The consequences of entry, establishment and spread of PCFVd in Australia have been estimated according to the methods described in Table 2.3.

Based on the decision rules described in Table 2.4, that is, where the potential consequences of a pest with respect to one or more (but not all) criteria have an impact of 'D', the overall consequences are estimated to be **Low**.

This assessment is made because the viroid may cause substantial losses in tomato and capsicum crops, and these losses would be amplified by spread of the viroid. If there is an incursion, it is likely that eradication and control would be attempted by state and territory governments, but given the characteristics of the viroid, these activities could prove costly and difficult.

Criterion	Estimate and rationale				
Direct					
Plant life or health	D – significant at the district level				
	PCFVd is an important pathogen of capsicum crops and can potentially cause significant yield losses. This viroid can also cause disease in tomato crops. In the 2018–19 financial year, Australian capsicum and tomato production were estimated to have gross values of \$180 million and \$674.2 million respectively (Horticulture Innovation Australia 2020).				
	Fruit from infected capsicum plants are small, being reduced in size by up to 50% and probably unmarketable (Punyapitak 2004; Verhoeven et al. 2011; Verhoeven et al. 2009). Capsicum plants may be mildly stunted.				
	PCFVd was reported to naturally infect tomato plants in Thailand. Affected tomato plants were stunted and showed leaf necrosis, distortion, and discoloration (Reanwarakorn, Klinkong & Porsoongnurn 2011).				
Other aspects of the	C – minor significance at the district level				
environment	PCFVd may infect other plant species of the Solanaceae, in addition to capsicum and tomato. Native and naturalised Solanaceae are components of Australian ecosystems that might be infected by PCFVd and their number or health might be affected. These plants provide food for native animals. <i>Solanum centrale</i> (bush tomato) is widespread in arid regions of central Australia.				
Indirect					
Eradication, control	D – significant at the district level				
	If an incursion of PCFVd were to occur in a capsicum or tomato crop, and it was detected, it would probably be controlled by eradication. The extent of an outbreak will depend on many factors including environmental conditions, the transport of plants and machinery between properties, the movement of workers and the activity of insect vectors, if present.				
	PCFVd infections may go unrecognised because a number of other viroid species produce symptoms identical or very similar to those induced by PCFVd (Hammond, Smith & Diener 1989; Verhoeven et al. 2004). An outbreak of PCFVd may not be detected until it has spread to several crops, properties and species.				
	During an outbreak, the infected property and adjoining and nearby properties will be surveyed. Samples from surveys will be tested in state government laboratories. RT-PCR will be used to test the samples and detect the viroid. Surveillance and laboratory tests are costly.				
	Viroids are not eliminated when infected plants are killed so infected plant material is buried. Equipment is sterilized using bleach. Typically, plants, propagating material, machinery and implements may not be moved from properties where outbreaks have been detected.				
Domestic trade	C – minor significance at the district level				
	If PCFVd became established in an Australian state, restrictions might be introduced on the interstate trade of affected seed and seedlings, and if this occurred it could lead to the loss of markets.				

	No movement of machinery from the affected properties is permitted during an eradication campaign.
International trade	D – significant at the district level
	Australia exports a very small proportion of its fresh tomato and capsicum fruit crop. These export markets might be affected if PCFVd became established in Australia. For example, Australia has markets for fresh tomatoes to New Zealand, Singapore, Hong Kong, Brunei-Darussalam, Malaysia, New Caledonia, Indonesia, French Polynesia, Fiji, and USA (DAWE 2020; HAL 2012).
Non-commercial and environmental	A –indiscernible at the local level
	No evidence was found indicating environmental and non-commercial indirect effects.

3.4.6 Unrestricted risk estimate

The unrestricted risk estimate for PCFVd is **Low**. Likelihoods and consequences are combined using the risk estimation matrix shown in Table 2.5. A summary of the risk estimates leading to this unrestricted risk estimate is given in Table 3.2 in Section 3.7.

3.5 Tomato apical stunt viroid

Tomato apical stunt viroid (TASVd) has infected tomato crops in several countries since the first known outbreak in Côte d'Ivoire in 1981, and has caused significant disease and yield loss in outbreaks in Israel and Tunisia (Antignus et al. 2002; Verhoeven, Jansen & Roenhorst 2006; Walter, Thouvenel & Fauquet 1980). In Côte d'Ivoire, TASVd was found in tomato plants grown in the open field, whereas it was found in greenhouse crops in Israel and Tunisia (Antignus, Lachman & Pearlsman 2007; Verhoeven, Jansen & Roenhorst 2006; Walter, Thouvenel & Fauquet 1980). TASVd has also been recorded from tomato crops in Indonesia, Niger and Senegal (Candresse et al. 2007; Candresse, Smith & Diener 1987; Walter 1987). The viroid has been reported in several ornamental plants in the Netherlands, including *Cestrum* sp. and *Solanum jasminoides* (Verhoeven et al. 2012).

TASVd is transmitted through tomato seed and may be introduced into greenhouses via infected seed (Antignus, Lachman & Pearlsman 2007; Antignus et al. 2002). TASVd has a moderately wide host range, and some infected hosts are asymptomatic (Singh, Ready & Nie 2003b). It is mechanically transmitted and is reported to spread rapidly through greenhouse tomato crops, probably on contaminated agricultural equipment and by plant-to-plant contact (Antignus et al. 2002). It is also transmitted by bumble-bees (Antignus, Lachman & Pearlsman 2007).

In this pest risk assessment, a risk scenario is considered whereby TASVd enters Australia in tomato seed, the seed is planted, and the viroid is transmitted within a tomato crop and to other hosts.

3.5.1 Likelihood of entry

The likelihood of entry is considered in two parts, the likelihood of importation (the likelihood that TASVd will arrive when host tomato seeds for sowing are imported) and the likelihood of distribution (the likelihood that TASVd will be viable and be transferred to a suitable host in Australia).

Likelihood of importation

The likelihood that TASVd will be imported on host tomato seeds for sowing is assessed as **High**.

This assessment is made because TASVd has been detected in tomato seed lots sent to Australia during the period of application of emergency measures, and the viroid has been reported in several countries, including tomato seed producing countries. Additionally, large volumes of tomato seed are imported into Australia each year.

The supporting evidence for this assessment is provided.

- TASVd has been found in tomato crops in Côte d'Ivoire (Walter, Thouvenel & Fauquet 1980), Indonesia (Candresse, Smith & Diener 1987), Israel (Antignus, Lachman & Pearlsman 2007), Niger (Walter 1987), Senegal (Candresse et al. 2007) and Tunisia (Verhoeven, Jansen & Roenhorst 2006).
- TASVd was found consistently in greenhouse tomato crops in Israel over several years from 1999 (Antignus, Lachman & Pearlsman 2007; Antignus et al. 2002; Candresse et al. 2007).
- TASVd infects tomato plants systemically. TASVd RNA has been found in the sap, leaves, roots, sepals, petals, ovaries, stamens, and seed of tomatoes (Antignus, Lachman & Pearlsman 2007). Disinfestation of seed harvested from TASVd-infected plants could not prevent viroid transmission to progeny seedlings, indicating that the viroid may be able to invade the embryonic tissue of the seed (Antignus, Lachman & Pearlsman 2007).
- TASVd was detected in three tomato seed lots sent to Australia from a country in Asia between February 2012 and October 2013.
- Large quantities of tomato seed are imported into Australia annually from suppliers in many countries to produce tomato crops. The department's records indicate that on average 760kg of tomato seed are imported into Australia annually.

Likelihood of distribution

To have an impact a pest must be transported in or on a pathway and must then be capable of transferring to a suitable host. The likelihood of this transfer occurring depends on the dispersal mechanisms of the pest and the intended use of the commodity.

The likelihood that TASVd will be distributed across Australia on imported tomato seeds for sowing and be transferred from the resulting plants to a suitable host is assessed as **High**.

This assessment is made primarily because imported tomato seed is distributed for planting throughout Australia, and if TASVd-infected seeds are present in an imported seed lot, it is likely the seeds and viroid will remain viable.

- Tomato seed is imported for planting for greenhouse and field production of tomato fruit. Seeds will be distributed for planting to greenhouses and farms in production areas throughout Australia, and for planting in domestic gardens.
- It is very likely TASVd will survive in tomato seed for long periods. The related viroid PSTVd has been found to endure for many years in seed when stored at room temperature (Singh, Boucher & Wang 1991).
- TASVd in seed will be distributed with the seed to production areas throughout Australia, and if present, is very likely to be present in an infectious state in the seed when it is planted.

Overall likelihood of entry (importation × distribution)

The likelihoods of importation and distribution of TASVd are combined to give an overall likelihood of entry using the matrix of rules for combining likelihoods (Table 2.2).

The overall likelihood that TASVd will enter Australia and be transferred to a suitable host via tomato seed for sowing is assessed as **High**.

3.5.2 Likelihood of establishment

The likelihood that TASVd will establish within Australia will depend upon the availability of host plants and the reproductive and survival strategies of the viroid. Based on an evaluation of these factors, the likelihood of establishment is assessed as **High**.

This assessment is made primarily because very large numbers of imported tomato seeds are sown in Australia, TASVd is transmitted from infected seed to seedlings, and there is evidence of the viroid establishing in tomato crops in other countries.

- TASVd has been found in Israel and Indonesia, and in several countries in Africa (Antignus, Lachman & Pearlsman 2007; Candresse et al. 2007; Candresse, Smith & Diener 1987; Verhoeven, Jansen & Roenhorst 2006; Walter, Thouvenel & Fauquet 1980).
- Some seedlings grown from TASVd-infected tomato seeds are very likely to be infected by the viroid. The viroid is transmitted through tomato seed to seedlings and a transmission rate of up to 80% has been reported (Antignus, Lachman & Pearlsman 2007).
- The germination rates of TASVd infected tomato seeds may be reduced, as has been reported for tomato seeds infected with other pospiviroids (Benson & Singh 1964).
- Millions of tomato plants are grown each year in Australia from imported seeds.
- Outbreaks of TASVd in greenhouses in Israel probably arose from infections of single isolated plants (Antignus et al. 2002).
- TASVd is mechanically transmitted (Antignus, Lachman & Pearlsman 2007; Singh & Dhar 1998; Singh, Ready & Nie 2003b) and is spread on contaminated machinery, tools and worker's hands during horticultural activities, including grafting, pruning and cutting.
- TASVd may spread in greenhouse crops before disease symptoms become apparent or are recognised. Infected tomato plants are less likely to be detected in field crops because of the lower intensity of horticultural activity in these crops.
- TASVd may survive between tomato crops in contaminated greenhouses and in infected crop residues, and infect plants subsequently grown in the vicinity in later seasons. The related viroid PSTVd is stable under most environmental conditions, and can remain infective in hydrated plant material for several months and in dry material for over a year, and is expected to survive composting processes (Mikkelsen, Elphinstone & Jensen 2005).
- Tomatoes are grown commercially in greenhouses and in the field in all states of Australia, including in regions with tropical and temperate climates. They are grown during the spring and summer in the field in the cooler regions of southern Australia, and all year round in other regions of Australia and in greenhouses.
- The climates of these regions in Australia where tomatoes are grown are generally similar to the climates of the areas where TASVd has been found. The environmental conditions in

commercial greenhouses in Australia are very similar to those in greenhouses in countries where outbreaks of TASVd have occurred.

• It is possible that TASVd may infect solanaceous weeds in the vicinity of infected greenhouses and field crops, and become established in a new area; *Solanum nigrum, Datura inoxia, D. metel* and other solanaceous species can be infected when inoculated with the viroid (Singh, Ready & Nie 2003b).

3.5.3 Likelihood of spread

The likelihood of spread describes the likelihood that TASVd, once having entered Australia on imported tomato seeds and become established, will spread from a point of introduction to new areas.

Based on a comparison of factors relevant to the expansion of geographic distributions of TASVd in the source and destination areas, the assessed likelihood of spread is **Moderate**.

This assessment is made because the viroid is spread by normal horticultural activities and it may also be dispersed with trade in tomato seed and seedlings, and with the disposal of crop residues. The viroid might also be transmitted by bumble-bees and certain aphids.

- Hosts of TASVd are widespread in Australia, as tomatoes are grown commercially in all states and territories of Australia. The solanaceous ornamental hosts *Cestrum* sp., *Solanum laxum* and *S. pseudocapsicum* are widely grown.
- TASVd is mechanically transmitted (Singh, Ready & Nie 2003b) and could be spread to new areas on contaminated machinery, tools and worker's hands during horticultural activities, including grafting, pruning and cutting.
- In greenhouse crops in Israel, TASVd spread quickly, almost always along plant rows, with entire crops becoming infected (Antignus et al. 2002).
- An entire greenhouse crop in Tunisia was quickly infected by TASVd (Verhoeven, Jansen & Roenhorst 2006).
- Bumble-bees (*Bombus terrestris*) can transmit TASVd during pollination (Antignus, Lachman & Pearlsman 2007). Bumble-bees are present in Tasmania but they are not present elsewhere on the mainland of Australia.
- Transport of crop residues infected with TASVd to new areas and their disposal near tomato, weed or ornamental hosts could result in the spread of TASVd to these hosts via mechanical transfer.
- TASVd could be introduced into new areas via tomato seed, as it can be present in seed and seed to seedling transmission has been demonstrated for this viroid in tomato (Antignus, Lachman & Pearlsman 2007).
- Seedlings are transported long distances within Australia for planting and infected seedlings could introduce TASVd into new areas. An incursion of the related viroid PSTVd was initiated at Mansfield, Victoria in 2012 after seedlings were transported from Perth, Western Australia, to the property in Victoria.
- TASVd could also be transmitted by plant feeding insects or in pollen, as other pospiviroids are sometimes transmitted by these mechanisms (EPPO 1997; Fernow, Peterson & Plaisted 1970; Kryczynski, Paduch-Cichal & Skreczkowski 1988; Querci et al. 1997; Singh et al. 2006; Syller, Marczewski & Pawlowicz 1997).

3.5.4 Overall likelihood of entry, establishment and spread

The overall likelihood of entry, establishment and spread is determined by combining the individual likelihoods of entry, of establishment and spread using the matrix rules for combining descriptive likelihoods (Table 2.2).

The overall likelihood that TASVd will enter Australia, be distributed in a viable state to susceptible hosts, establish in that area and subsequently spread within Australia is assessed as **Moderate**.

3.5.5 Consequences

The consequences of entry, establishment and spread of PCFVd in Australia have been estimated according to the methods described in Table 2.3.

Based on the decision rules described in Table 2.4, that is, where the potential consequences of a pest with respect to one or more (but not all) criteria have an impact of 'D', the overall consequences are estimated to be **Low**.

This assessment is made because the viroid may cause substantial losses in tomato crops and these losses would be amplified by spread of the viroid. If there is an incursion, it is likely that eradication and control would be attempted by state and territory governments, but given the characteristics of the viroid, these activities could prove costly and difficult.

Criterion	Estimate and rationale		
Direct			
Plant life or health	D – significant at the district level		
	TASVd is an important pathogen of tomato crops and can cause significant yield losses. In the 2018–19 financial year, Australian tomato production was estimated to have a gross value of \$674.2 million (Horticulture Innovation Australia 2020).		
	TASVd-infected tomato plants are stunted and their leaves become deformed, yellow and brittle (Antignus et al. 2002; Candresse et al. 2007; Spieker, Marinkovic & Sänger 1996; Verhoeven, Jansen & Roenhorst 2006). Fruit from infected plants is small and discoloured, ripening is delayed, and storage life is greatly reduced (Antignus et al. 2002; Candresse et al. 2007). In greenhouse tomato crops TASVd can spread quickly and infect entire crops (Antignus et al. 2002; Verhoeven, Jansen & Roenhorst 2006). Up to 100% disease incidence could be observed with heavy yield losses (EPPO 2017b).		
Other aspects of the	C – minor significance at the district level		
environment	Species from the Asteraceae, Chenopodiaceae, Scrophulariaceae and Solanaceae were infected when inoculated with TASVd (Singh, Ready & Nie 2003b). Native and naturalised species of these plant families may be infected by this viroid and their number or health may be affected. Native and naturalised species of Chenopodiaceae, Asteraceae, Scrophulariaceae and Solanaceae are components of Australian ecosystems and provide food for native animals. <i>Solanum centrale</i> (bush tomato) is widespread in arid regions of central Australia.		
Indirect			
Eradication, control	D – significant at the district level		
	If an incursion of TASVd were to occur in a tomato crop, and it was detected, it would probably be controlled by eradication. The extent of an outbreak will depend many factors including environmental conditions, the transport of plants and machinery		

	between properties, the movement of workers and the movement of insect vectors, if present.
	TASVd infections may go unrecognised because a number of other viroid species produce symptoms identical or very similar to those induced by TASVd (Singh, Ready & Nie 2003b). An outbreak of TASVd may not be detected until it has spread to several crops, properties and species.
	During an outbreak, the infected property and adjoining and nearby properties will be surveyed. Samples from surveys will be tested in state government laboratories. RT-PCR will be used to test the samples and detect the viroid. Surveillance and laboratory tests are costly.
	Viroids are not eliminated when infected plants are killed so infected plant material is buried. Equipment is sterilized using bleach. Typically, plants, propagating material, machinery and implements may not be moved from properties where outbreaks have been detected.
Domestic trade	C – minor significance at the district level
	If TASVd became established in an Australian state, restrictions might be introduced on the interstate trade of affected seed and seedlings, and if this occurred it could lead to the loss of markets.
	No movement of machinery from the affected properties is permitted during an eradication campaign.
International trade	D – significant at the district level
	Australia exports a very small proportion of its fresh tomato fruit crop, which might be affected if TASVd became established in Australia. Australia has markets for fresh tomatoes to New Zealand, Singapore, Hong Kong, Brunei-Darussalam, Malaysia, New Caledonia, Indonesia, French Polynesia, Fiji, and USA (DAWE 2020; HAL 2012).
Non-commercial and environmental	A –indiscernible at the local level No evidence was found indicating environmental and non-commercial indirect effects.

3.5.6 Unrestricted risk estimate

The unrestricted risk estimate for TASVd is **Low**. Likelihoods and consequences are combined using the risk estimation matrix shown in Table 2.5. A summary of the risk estimates leading to this unrestricted risk estimate is given in Table 3.2 in Section 3.7.

3.6 Tomato chlorotic dwarf viroid

Tomato chlorotic dwarf viroid (TCDVd) was first described from a greenhouse tomato crop in Canada in 1996 (Sabaratnam 2012; Singh, Nie & Singh 1999). The viroid causes significant disease and economic losses in greenhouse tomato crops (Matsushita et al. 2008; Singh, Nie & Singh 1999) and has now been reported from countries in Asia, Europe, the Middle East and North America (Table B.1).

TCDVd is transmitted through tomato seed to seedlings at rates of up to 80% (Singh & Dilworth 2009). Its spread through greenhouse tomato crops in Japan was consistent with mechanical contact (Matsushita, Usugi & Tsuda 2009). Bumble-bees have also been shown to transmit TCDVd during pollination (Matsushita, Usugi & Tsuda 2009).

In this pest risk assessment, a risk scenario is considered whereby TCDVd enters Australia in tomato seed, the seed is planted, and the viroid is transmitted within a tomato crop and to other hosts.

3.6.1 Likelihood of entry

The likelihood of entry is considered in two parts, the likelihood of importation (the likelihood that TCDVd will arrive when host tomato seeds for sowing are imported) and the likelihood of distribution (the likelihood that TCDVd will be viable and be transferred to a suitable host in Australia).

Likelihood of importation

The likelihood that TCDVd will be imported on host tomato seeds for sowing is assessed as **High**.

This assessment is made because TCDVd has been detected in tomato seed lots sent to Australia during the period of application of emergency measures and the viroid has been reported in several countries, including tomato seed producing countries. Additionally, large volumes of tomato seed are imported into Australia each year.

The supporting evidence for this assessment is provided.

- TCDVd has been found in greenhouse tomato crops in Canada (Singh, Nie & Singh 1999), USA (Ling et al. 2009), Japan (Matsushita et al. 2008), France (Candresse et al. 2010) and Mexico (Ling & Zhang 2009).
- Pospiviroids infect tomato plants systemically. Pospiviroid RNA has been found in the sap, leaves, roots, sepals, petals, ovaries, stamens and seed of tomato (Antignus, Lachman & Pearlsman 2007; EPPO 2016b; Koenraadt et al. 2009; Lykke et al. 2010; Matsushita & Tsuda 2014b; Singh & Dilworth 2009; Singh et al. 2006; Singh, Nie & Singh 1999; Zhu et al. 2001).
- A high percentage of tomato seed from TCDVd-infected plants can contain the viroid (Singh & Dilworth 2009).
- TCDVd was detected in seven lots of tomato seed sent to Australia and tested on arrival between February 2012 and October 2013. The seed was produced in two countries in Europe, one in the Americas, one in Africa and one in the Middle East.
- Large quantities of tomato seed are imported into Australia annually from suppliers in many countries to produce tomato crops. The department's records indicate that on average 760kg of tomato seed are imported into Australia annually.

Likelihood of distribution

To have an impact a pest must be transported in or on a pathway and must then be capable of transferring to a suitable host. The likelihood of this transfer occurring depends on the dispersal mechanisms of the pest and the intended use of the commodity.

The likelihood that TCDVd will be distributed across Australia on imported tomato seeds for sowing and be transferred from the resulting plants to a suitable host is assessed as **High**.

This assessment is made because imported tomato seed is distributed for planting throughout Australia, and if TCDVd-infected seeds are present in an imported seed lot, it is likely the seeds and viroid will remain viable.

- Tomato seeds are imported for planting for greenhouse and field production of tomato fruit. Seeds will be distributed to greenhouses and farms in production areas throughout Australia, and for planting in domestic gardens.
- It is very likely TCDVd will survive in tomato seed for long periods. The related pospiviroid PSTVd has been found to endure for many years in seed when stored at room temperature (Singh, Boucher & Wang 1991).
- TCDVd present in seed will be distributed with the seed to production areas throughout Australia, and if present, is very likely to be present in an infectious state in the seed when it is planted.

Overall likelihood of entry (importation × distribution)

The likelihoods of importation and distribution of TCDVd are combined to give an overall likelihood of entry using the matrix of rules for combining likelihoods (Table 2.2).

The overall likelihood that TCDVd will enter Australia and be transferred to a suitable host via tomato seed for sowing is assessed as **High**.

3.6.2 Likelihood of establishment

The likelihood that TCDVd will establish within Australia will depend upon the availability of host plants and the reproductive and survival strategies of the viroid. Based on an evaluation of these factors, the likelihood of establishment is assessed as **High**.

This assessment is made primarily because very large numbers of imported tomato seeds are sown in Australia, TCDVd is transmitted from infected seed to seedlings, and there is evidence of the viroid establishing in tomato crops in other countries.

- Some seedlings grown from TCDVd infected tomato seed are very likely to be infected by the viroid. TCDVd is transmitted through tomato seed to seedlings and a transmission rate of up to 80% has been reported (Singh & Dilworth 2009).
- Seed to seedling transmission has been demonstrated for commercial seed, as two out of 250 pools of 10 plants tested positive for TCDVd infection in a grow-out test of seed from which an outbreak developed in France (Candresse et al. 2010).
- The germination rates of TCDVd infected tomato seeds may be reduced, as has been reported for tomato seeds infected with other pospiviroids (Benson & Singh 1964).
- Millions of tomato plants are grown each year in Australia from imported seeds.
- TCDVd is mechanically transmitted (Matsushita, Usugi & Tsuda 2009; Singh & Dilworth 2009) and its spread along rows of tomato plants in Japan suggests that transmission resulted from mechanical contact (Matsushita et al. 2008). Pospiviroids are spread mechanically from infected plants by contaminated agricultural equipment, on worker's hands during horticultural activities and by plant-to-plant contact (Conde, Connelly & Pitkethley 1996; Diener 1971; Hammond & Owens 2006; Horticulture New Zealand 2008; Manzer & Merriam 1961; Singh & Dhar 1998).
- TCDVd may spread in greenhouse crops before disease symptoms become apparent or are recognised. Infected tomato plants in a field crop are less likely to be detected because of the lower intensity of horticultural activity in these crops.

- TCDVd may survive between tomato crops in contaminated greenhouses and in infected crop residues, and infect plants subsequently grown in the vicinity in later seasons. The related viroid PSTVd is stable under most environmental conditions and can remain infective in hydrated plant material for several months and in dry material for over a year, and is expected to survive composting processes (Mikkelsen, Elphinstone & Jensen 2005).
- Tomato crops are grown commercially in greenhouses and in the field in all states of Australia. TCDVd has been found overseas infecting greenhouses crops in temperate areas.
- The environmental conditions in commercial greenhouses in Australia are very similar to those in greenhouses in countries where outbreaks of TCDVd have occurred.
- Beside tomato, TCDVd has been found infecting the ornamentals *Brugmansia sanguine* (Verhoeven et al. 2010), *Petunia* × *hybrida* (James et al. 2008; Verhoeven et al. 2007b) and *Vinca minor* (Singh & Dilworth 2009). These ornamentals could act as reservoirs of TCDVd if they were infected.
- TCDVd has recently been detected in asymptomatic eggplant (*Solanum melongena* L.), in which seed transmission rates between 7.7% and 100% have been recorded (Gramazio et al. 2019). Infected eggplant crops may act as a source of inoculum for transmission of TCDVd to tomato.
- Outbreaks of TCDVd have occurred in Canada, USA, Japan, and Mexico (Candresse et al. 2010; Ling & Zhang 2009; Matsushita et al. 2008; Singh, Nie & Singh 1999).

3.6.3 Likelihood of spread

The likelihood of spread describes the likelihood that TCDVd, once having entered Australia on imported tomato seeds and become established, will spread from a point of introduction to new areas.

Based on a comparison of factors relevant to the expansion of geographic distributions of TCDVd in the source and destination areas, the assessed likelihood of spread is **Moderate**.

This assessment is made because the viroid is spread by normal horticultural activities and it may also be dispersed with trade in tomato seed and seedlings, and with the disposal of crop residues. The viroid is transmitted by bumble-bees and might be transmitted by aphids.

- Hosts of TCDVd are widespread in Australia. Tomatoes and eggplants are grown commercially across Australia and the solanaceous ornamental hosts *Brugmansia sanguinea*, *Petunia × hybrida*, *Verbena × hybrida* and *Vinca minor* are also widely grown.
- Pospiviroids can spread rapidly through greenhouse crops because of the density of planting and the intensity of horticultural activity (Diener 1971; Hammond & Owens 2006).
- TCDVd could be spread to tomato crops in new areas if contaminated machinery or tools were moved between areas and could be moved on worker's hands if people work in more than one area within a short period of time (Sabaratnam 2012).
- Movement of TCDVd-infected crop residues to new areas, and their disposal near tomato, weed or other hosts could result in the mechanical transfer of the viroid to those hosts and result in its spread.
- TCDVd could be introduced into new areas by tomato seed, as it can be present in seed, and seed to seedling transmission has been demonstrated for this viroid in tomato (Singh & Dilworth 2009).

- Seedlings are moved over long distances within Australia for planting and infected seedlings could introduce TASVd into new areas. An incursion of the related viroid PSTVd was initiated at Mansfield, Victoria in 2012 after seedlings were transported from Perth, Western Australia, to the property in Victoria.
- The movement of nursery stock or of the ornamental hosts of TCDVd, *Brugmansia sanguinea*, *Petunia* × *hybrida*, *Verbena* × *hybrida* and *Vinca minor*, could spread the viroid to new areas.
- TCDVd can be spread by bumble-bees (*Bombus terrestris*) (Matsushita, Usugi & Tsuda 2009). Bumble-bees are not present on mainland Australia, but they are present in Tasmania.
- TCDVd could also be transmitted by plant feeding insects or in pollen, as other pospiviroids are sometimes transmitted by these mechanisms (EPPO 1997; Fernow, Peterson & Plaisted 1970; Kryczynski, Paduch-Cichal & Skreczkowski 1988; Querci et al. 1997; Singh et al. 2006; Syller, Marczewski & Pawlowicz 1997).

3.6.4 Overall likelihood of entry, establishment and spread

The overall likelihood of entry, establishment and spread is determined by combining the individual likelihoods of entry, of establishment and spread using the matrix rules for combining descriptive likelihoods (Table 2.2).

The overall likelihood that TCDVd will enter Australia, be distributed in a viable state to susceptible hosts, establish in that area and subsequently spread within Australia is assessed as **Moderate**.

3.6.5 Consequences

The consequences of entry, establishment and spread of TCDVd in Australia have been estimated according to the methods described in Table 2.3.

Based on the decision rules described in Table 2.4, that is, where the potential consequences of a pest with respect to one or more (but not all) criteria have an impact of 'D', the overall consequences are estimated to be **Low**.

This assessment is made because the viroid may cause substantial losses in tomato crops and these losses would be amplified by spread of the viroid. If there is an incursion, it is likely that eradication and control would be attempted by state and territory governments, but given the characteristics of the viroid, these activities could prove costly and difficult.

Criterion	Estimate and rationale		
Direct			
Plant life or health	D – significant at the district level		
	TCDVd is an important pathogen of tomato crops and can cause significant yield losses. In the 2018–19 financial year, Australian tomato production was estimated to have a gross value of \$674.2 million (Horticulture Innovation Australia 2020).		
	Tomato plants infected by TCDVd show top bunching, leaf curling symptoms (Candresse et al. 2010). Commonly observed symptoms are stunting, bunchiness, reduced leaves and fruit, leaf chlorosis, leaf and petiole necrosis, downward bending of leaves and fruit distortion (Sabaratnam 2012).		

	TCDVd has been found to spread in greenhouse tomato crops in Canada (Singh, Nie & Singh 1999), Japan (Matsushita et al. 2008) and France (Candresse et al. 2010).				
Other aspects of the	C – minor significance at the district level				
environment	Native and naturalised species of Solanaceae may be infected by TCDVd and their number or health may be affected. Native and naturalised Solanaceae are components of Australian ecosystems and provide food for native animals. <i>Solanum centrale</i> (bush tomato) is widespread in arid regions of central Australia.				
Indirect					
Eradication, control	D – significant at the district level				
	If an incursion of TCDVd were to occur in a tomato crop, and it was detected, it would probably be controlled by eradication. The extent of an outbreak will depend on many factors including the environmental conditions, the transport of plants and machinery between properties, the movement of workers and the activity of insect vectors, if present.				
	TCDVd infections may go unrecognised because a number of other viroid species produce symptoms identical or very similar to those induced by TCDVd (Singh, Ready & Nie 2003b). An outbreak of TCDVd may not be detected until it has spread to several crops, properties and species.				
	During an outbreak, the infected property and adjoining and nearby properties will be surveyed. Samples from surveys will be tested in state government laboratories. RT-PCR will be used to test the samples and detect the viroid. Surveillance and laboratory tests are costly.				
	Viroids are not eliminated when infected plants are killed so infected plant material is buried. Equipment is sterilized using bleach. Typically, plants, propagating material, machinery and implements may not be moved from properties where outbreaks have been detected.				
Domestic trade	C – minor significance at the district level				
	If TCDVd became established in an Australian state, restrictions might be introduced on the interstate trade of affected seed and seedlings, and if this occurred it could lead to the loss of markets.				
	No movement of machinery from the affected properties is permitted during an eradication campaign.				
International trade	D – significant at the district level				
	Australia exports a very small proportion of its fresh tomato fruit crop, which might be affected if TCDVd became established in Australia. Australia has markets for fresh tomatoes to New Zealand, Singapore, Hong Kong, Brunei-Darussalam, Malaysia, New Caledonia, Indonesia, French Polynesia, Fiji, and USA (DAWE 2020; HAL 2012).				
Non-commercial and	A –indiscernible at the local level				
environmental	No evidence was found indicating environmental and non-commercial indirect effects.				

3.6.6 Unrestricted risk estimate

The unrestricted risk estimate for TCDVd is **Low**. Likelihoods and consequences are combined using the risk estimation matrix shown in Table 2.5. A summary of the risk estimates leading to this unrestricted risk estimate is given in Table 3.2 in Section 3.7.

3.7 Unrestricted risk estimate

Unrestricted risk is the result of combining the likelihood of entry, establishment and spread with the assessed outcome of consequences. Likelihoods and consequences are combined using the risk estimation matrix shown in Table 2.5.

Table 3.2 summarises the unrestricted risk estimates for PepMV, CLVd, PCFVd, TASVd and TCDVd associated with tomato seed for sowing.

Pest name	Entry	Establishment	Spread	Consequences	URE
Pepino mosaic virus	High	High	High	Moderate	Moderate
Columnea latent viroid	High	High	Moderate	Low	Low
Pepper chat fruit viroid	High	High	Moderate	Low	Low
Tomato apical stunt viroid	High	High	Moderate	Low	Low
Tomato chlorotic dwarf viroid	High	High	Moderate	Low	Low

Table 3.2 Unrestricted risk estimates for PepMV and pospiviroids in tomato seeds for sowing

3.8 Pest risk assessment conclusions

The unrestricted risk estimates for PepMV, CLVd, PCFVd, TASVd and TCDVd do not achieve the ALOP for Australia. Accordingly, risk management measures are required for these pests.

4 Pest risk management

Pepino mosaic virus (PepMV), Columnea latent viroid (CLVd), Pepper chat fruit viroid (PCFVd), Tomato apical stunt viroid (TASVd) and Tomato chlorotic dwarf viroid (TCDVd) present unrestricted risks that do not achieve the appropriate level of protection (ALOP) for Australia. Consequently, the department recommends risk management measures to reduce the risk posed by these pests to levels that achieve the ALOP for Australia. The recommended risk management measures are described in this chapter.

The status of *Potato spindle tuber viroid* (PSTVd) in Australia is under continuing evaluation. This evaluation is being undertaken as a process separate to the finalisation of this pest risk analysis (PRA) because policy for PSTVd covers a broader range of hosts than tomato seed. Until this evaluation is completed, PSTVd will continue to be regulated at the Australian border.

4.1 Recommended risk management measures

The recommended risk management measures are largely consistent with the emergency measures that they will replace, but some amendments are recommended. The differences between the recommended risk management measures and the emergency measures include an increase of the sample size taken for testing for PepMV, an option of heat treatment to manage PepMV, and the removal of measures for wild tomato seeds. The rationale for these changes is explained in Appendix D.

A combination of phytosanitary measures is recommended for seeds of *Solanum lycopersicum* (tomato) and hybrids of this species (including crosses with wild tomato species) that includes a test or a treatment for each identified pest:

- Option 1. Polymerase chain reaction (PCR) test—an option that is applicable to all quarantine pests associated with tomato seed.
 - PCR using sample size of 20,000 seeds or 20% of small seed lots to verify freedom from detectable presence of PepMV, CLVd, PCFVd, TASVd and TCDVd.
- Option 2. Enzyme-linked immunosorbent assay (ELISA) test—an option that is applicable to PepMV only.
 - ELISA test using sample size of 20,000 seeds or 20% of small seed lots to verify freedom from detectable presence of PepMV.
- Option 3. Heat treatment—an option that is applicable to PepMV only.
 - Dry heat treatment at 80°C for 72 hours.

Test conditions, including subsample sizes, will be specified on BICON.

It is recommended that testing of seeds of wild tomato species (*Solanum chilense, S. chmielewskii, S. parviflorum, S. peruvianum* and *S. pimpinellifolium*) for the regulated pathogens should cease.

4.1.1 Phytosanitary certification

Seed lots of *Solanum lycopersicum* and hybrids of this species that are tested off-shore by PCR must be accompanied by a laboratory test report and an official government Phytosanitary Certificate endorsed with the following additional declaration:

• 'The consignment of tomato seed comprises [*insert number of seed lots*] seed lot(s); for each seed lot, seeds were tested by PCR [*insert laboratory name(s) and report number(s)*] on a sample size of [*insert sample size i.e. 20,000 seeds or 20% of small seed lots*] and found free from [*name of the pests*]';

Seed lots that are tested by ELISA or treated by dry heat for PepMV must also be accompanied by the laboratory test report and an official government Phytosanitary Certificate endorsed with the following additional declaration:

• 'The consignment of tomato seed comprises [*insert number of seed lots*] seed lot(s); for each seed lot, seeds were tested by ELISA [*insert laboratory name(s) and report number(s)*] on a sample size of [*insert sample size i.e. 20,000 seeds or 20% of small seed lots*] and found free from PepMV'.

OR

• 'The consignment of tomato seed comprises [*insert number of seed lots*] seed lot(s); for each seed lot, seeds were treated by dry heat at 80°C for 72 hours for control of PepMV'.

4.2 Evaluation of recommended risk management measures

The recommended pest risk management measures (Table 4.1) are designed to reduce the pest risk for each identified quarantine pest to a very low level, which will achieve the ALOP for Australia.

Recommended measure	Effect of the measure	Risk estimates after measures (restricted risk)
Option 1. PCR test	Testing to verify freedom from PepMV, CLVd, PCFVd, TASVd and TCDVd will reduce the risk of introducing these pests into Australia.	Very low
Option 2. ELISA test	Testing to verify freedom from PepMV will reduce the risk of introducing this pest into Australia.	Very low
Option 3. Heat treatment	Treatment of seeds with dry heat will reduce the risk of introducing PepMV into Australia.	Very low

Table 4.1 Evaluation of the recommended pest risk management measures impact on risk estimates

4.3 Standard import conditions

Under Australia's existing policies, seeds of tomato and wild tomato species are subject to the department's standard import conditions for seeds. These import conditions will remain in place. The standard import conditions are:

- Each shipment must be packed in clean, new packaging and be clearly labelled with the full botanical name of the species.
- Where the seed lot weight is greater than 10kg, mandatory International Seed Testing Association (ISTA) sampling of each consignment must be used to establish freedom from weed seed contamination. This testing may be performed at department-approved ISTA laboratories overseas, or on arrival at Australian-accredited facilities.
- Where seed lots are less than or equal to 10kg, a biosecurity officer must conduct a visual inspection of each consignment on arrival in Australia for freedom from live insects, soil,

disease symptoms, contaminant seed, other plant material (for example, leaf and stem material, fruit pulp, and pod material), animal material (for example, animal faeces and feathers) and any other extraneous contamination of biosecurity concern.

It is the importer's responsibility to ensure compliance with the requirements of all other regulatory and advisory bodies associated with importing commodities into Australia. These include the Australian Government Department of Home Affairs, the Therapeutic Goods Administration, the Australian Pesticides and Veterinary Medicines Authority, the Office of the Gene Technology Regulator, and state and territory departments of agriculture.

4.4 Consideration of alternative measures

Australia recognises the principle of equivalence, namely, 'the situation where, for a specified pest risk, different phytosanitary measures achieve a contracting party's appropriate level of protection' (FAO 2019b). ISPM 24 (FAO 2017c) provides guidelines for the determination and recognition of equivalence of phytosanitary measures.

Where formal recognition of equivalence is required, the NPPO of the exporting country must provide a technical submission detailing relevant evidence for the proposed measures for consideration by the department.

Several ISPMs provide further guidance on alternative pest risk management options that may be appropriate to achieve the objective of freedom from the quarantine pests identified in this review. These include:

- ISPM 4: Requirements for the establishment of pest free areas (FAO 2017b)
- ISPM 10: *Requirements for the establishment of pest free places of production and pest free production sites* (FAO 2016b)
- ISPM 14: The use of integrated measures in a systems approach for pest risk management (FAO 2019d)

These alternative pest risk management options are discussed in the following sections.

4.4.1 Sourcing seeds from pest-free areas

The establishment and use of a pest free area (PFA) by an NPPO provides assurance that specific pests are not present in a delimited geographic area. The delimitation of a PFA should be relevant to the biology of the pest concerned.

The requirements for establishing PFAs are set out in ISPM 4 (FAO 2017b). This ISPM defines a PFA as 'an area in which a specific pest does not occur as demonstrated by scientific evidence and in which, where appropriate, this condition is being officially maintained'. A PFA may concern all or part of several countries and is managed by the NPPO of the exporting country. The establishment and use of a PFA by an NPPO allows an exporting country to export plants and other regulated articles to an importing country without having to apply additional phytosanitary measures providing certain requirements are met.

Requirements for an NPPO to establish and maintain a PFA include:

- systems to establish freedom (general surveillance and specific surveys)
- phytosanitary measures to maintain freedom (regulatory actions, routine monitoring, and extension advice to producers)
- checks to verify freedom has been maintained.

NPPOs that propose to use area freedom as a measure for managing risks posed by the quarantine pests identified in this review must provide the Department of Agriculture, Water and the Environment with an appropriate submission demonstrating area freedom for its consideration.

4.4.2 Sourcing seeds from pest-free places of production

Requirements for establishing pest free places of production are set out in ISPM 10 (FAO 2016b). The concept of 'pest freedom' allows exporting countries to provide assurance to importing countries that plants, plant products and other regulated articles are free from a specific pest or pests and meet the phytosanitary requirements of the importing country. Where a defined portion of a place of production is managed as a separate unit and can be maintained pest free, it may be regarded as a pest free production site.

Requirements for an NPPO to establish and maintain a pest free place of production or a pest free production site as a phytosanitary measure include:

- systems to establish pest freedom
- systems to maintain pest freedom
- verification that pest freedom has been attained or maintained
- product identity, consignment integrity and phytosanitary security.

Where necessary, a pest free place of production or a pest free production site must also establish and maintain an appropriate buffer zone.

Administrative activities required to support a pest free place of production or a pest free production site include documentation of the system and maintenance of adequate records about the measures taken. Review and audit procedures undertaken by an NPPO are essential to support assurance of pest freedom and for system appraisal. Bilateral agreements or arrangements may also be needed.

NPPOs that propose to use pest free places of production as a measure for managing risks posed by the quarantine pests identified in this review must provide the Department of Agriculture, Water and the Environment with an appropriate submission demonstrating pest free place of production status, for its consideration.

4.4.3 Sourcing seeds produced under a systems approach

ISPM 14 (FAO 2019d) provides guidelines on the use of systems approaches to manage pest risk. According to ISPM 14 (FAO 2019d), 'a systems approach requires the integration of different measures, at least two of which act independently, with a cumulative effect' to achieve the appropriate level of protection.

A systems approach could provide an alternative to relying on a single measure to achieve the ALOP of an importing country or could be used where no single measure is available. A systems approach is often tailored to specific commodity–pest–origin combinations and may be developed and implemented collaboratively by exporting and importing countries. The importing country specifies the appropriate approach after considering technical requirements, minimal impact, transparency, non-discrimination, equivalence, and operational feasibility.

NPPOs that propose to use a systems approach as a measure for managing risks posed by the quarantine pests identified in this review must provide the Department of Agriculture, Water and the Environment with an appropriate submission describing their preferred systems approach and rationale, for its consideration.

Seed disinfestation by chemical and heat treatments

The department has considered evidence that *Pepino mosaic virus* and pospiviroids may be eliminated from seed by using chemical or heat treatments (Appendices A & B). Whereas evidence was found that supported the use of a dry heat treatment to eliminate *Pepino mosaic virus*, no equivalent evidence was found in the preparation of this report that showed pospiviroids could be eliminated from large numbers of seeds by a chemical or heat treatment. If an application is received to use seed disinfestation as an alternative to testing for the pospiviroids, the department will require efficacy data that includes sufficient replicates and testing of sufficient numbers of seeds to show that the method will reliably eliminate the proposed heat treatment for *Pepino mosaic virus*, the department will require efficacy data that include sufficient replicates and testing of sufficient numbers of seeds to show that the method will require efficacy data that include sufficient replicates and testing of sufficient numbers of seeds to show that the method will require efficacy data that include sufficient replicates and testing of sufficient numbers of seeds to show that the method will reliably eliminate the virus from large seed lots.

Visual inspection of seed production crops

Phytosanitary certification of tomato seed lots based on visual inspection of the seed production crop was accepted for a period under the emergency measures. However, incursions of one of the pathogens continued to occur in Australia (Section 1.2.3). Testing of seed sent to Australia indicated that many seed lots that were certified in this way were contaminated with infected seeds (Section 1.2.3). The department concluded that the measure was insufficient for the regulated pathogens.

4.5 Review of import conditions

The department reserves the right to review these import conditions if there is reason to believe that the pest or phytosanitary status of these organisms has changed or is likely to change. Similarly, a review may be required, for example, where scientific evidence or other information subsequently becomes available which improves knowledge of, or decreases uncertainty in treatment efficacy and/or the equivalence of measures.

5 Conclusion

The findings of this pest risk analysis for *Pepino mosaic virus* and pospiviroids (*Columnea latent viroid, Pepper chat fruit viroid, Tomato apical stunt viroid* and *Tomato chlorotic dwarf viroid*) associated with tomato seed are based on a comprehensive scientific analysis of relevant literature.

The Department of Agriculture, Water and the Environment considers that the risk management measures recommended in this report will provide an appropriate level of protection against the identified quarantine pests associated with tomato seed.

Appendix A: Pepino mosaic virus

Pepino mosaic virus biology

Pepino mosaic virus (PepMV) was originally detected on pepino plants (*Solanum muricatum*) in Peru in 1974 (Jones, Koenig & Lesemann 1980). It was first reported affecting major crops when it was found infecting tomato in Germany, the Netherlands and United Kingdom in 1999 (van der Vlugt et al. 2002; van der Vlugt et al. 2000; Wright & Mumford 1999). PepMV has since been detected in many countries in Europe and in countries in Africa, Asia and Americas, with this transcontinental distribution probably produced by recent rapid spread (Gómez, Sempere & Aranda 2012) (Table A.2).

PepMV has been found infecting tomato and pepino crops and basil (*Ocimum basilicum*) (Davino et al. 2009; Hanssen & Thomma 2010). Experiments have shown the virus can infect capsicum and potato, but as yet no natural infections have been reported in these plants (Blystad et al. 2015; EPPO 2014d; Hanssen & Thomma 2010).

In addition to cultivated plants, PepMV infects a range of weed species and species of wild plants (CSL 2005; Soler et al. 2002). Several of these alternative hosts are from the family Solanaceae, whereas others are from the Amaranthaceae, Asteraceae, Boraginaceae, Brassicaceae, Chenopodiaceae, Convolvulaceae, Malvaceae, Plantaginaceae and Polygonaceae (Córdoba, Martinez-Priego & Jordá 2004; Papayiannis, Kokkinos & Alfaro-Fernández 2012; Soler et al. 2002).

Presence in seeds and floral parts

PepMV spreads through plants systemically, and has been detected in leaves, fruits, roots, flower parts and on seeds (Alfaro-Fernández et al. 2009; Ling 2008; Mehle et al. 2014; Özdemir 2010; Schwarz et al. 2010; Shipp et al. 2008).

PepMV is detected in tomato flowers (Özdemir 2010; Shipp et al. 2008) and within the flowers, PepMV particles have been detected in the stigma, petals, anthers and anther filaments (Ling 2008). No report was found indicating PepMV particles are present in pollen, but the virus has been detected on bumble-bees (*Bombus impatiens*) that had been foraging on infected plants (Shipp et al. 2008) suggesting that pollen may carry the virus and may be contaminated with virus particles.

PepMV has been detected many times internationally in commercially traded tomato seed lots (Table A.1). One significant experiment indicated the virus is present in the seed coat (testa) and seed membranes, as well as on seed surfaces (Ling 2008), but the virus was not detected in embryonic tissue (Ling 2008).

Transmission of PepMV

PepMV is mechanically transmitted and highly transmissible to tomato plants. The virus spreads within crops when plants are pruned or suffer minor abrasions, as occurs when plants are handled, and also when plants touch each other (Jones, Koenig & Lesemann 1980; Spence et al. 2006; Wright & Mumford 1999). PepMV can also be transmitted when tools and worker's hands and clothes become contaminated (Hanssen & Thomma 2010; van der Vlugt 2009). PepMV particles are relatively stable at room temperature and can remain infectious for several weeks in plant debris, and on contaminated surfaces and in water (van der Vlugt 2009).

The virus can be transmitted by insects. PepMV is transmitted by the greenhouse whitefly (*Trialeurodes vaporariorum*) and bumble-bees (*Bombus impatiens*), the former achieving transmission when feeding on the plants, the latter when pollinating (Lacasa et al. 2003; Noël, Hance & Bragard 2014; Shipp et al. 2008). It has been suggested that infection often occurs when flowers are pollinated by bumble-bees and then spreads to other parts of tomato plants.

PepMV is transmitted through water after being released from the roots of infected plants. The motile zoospores of the fungus *Olpidium virulentus* may be required for transmission through water or may just assist transmission (Alfaro-Fernández et al. 2010; Ling & Scott 2007; Mehle et al. 2014; Schwarz et al. 2010).

Transport of live plant material, including seeds and seedlings, is likely to be responsible for the introduction of the virus into crops (Córdoba-Sellés et al. 2007) and into geographic regions (Werkman & Sansford 2010). Bumble-bees can also transmit the virus between adjacent crops (Shipp et al. 2008).

Symptoms in tomato

PepMV-infected plants typically develop distorted leaves that have a blistered appearance. The leaves may also develop chlorosis, yellow angular spots, severe leaf mosaics and necrosis symptoms. Brown streaks may appear on the stems, which may also become necrotic (Hanssen & Thomma 2010). Plants may develop the 'nettle-head' form, with the upper young leaves and shoots becoming stunted (Hanssen & Thomma 2010). PepMV-infected plants may be stunted or distorted or may wilt (EPPO 2014d; Soler-Aleixandre et al. 2005a).

Fruit from infected plants may be discoloured and have a marbled or mosaic appearance with patches of yellow and red or green and red (Hanssen & Thomma 2010). Fruit may split and become open so that the seed and flesh is exposed (Hanssen & Thomma 2010).

The degree of visible disease on the vegetative parts of PepMV-infected tomato plants varies widely, with some plants exhibiting severe symptoms, others expressing mild symptoms and some being asymptomatic. Importantly, asymptomatic infected plants and plants with mild symptoms are difficult to recognise and may be missed when crops are inspected. The range of symptom expression may be due to environmental factors (Chitambar 2015). Low temperatures and low light conditions are reported to favour the appearance of more pronounced symptoms (van der Vlugt 2009). Infected tomato plants with no vegetative symptoms may still develop symptoms on their fruits.

Seed transmission

It is generally accepted that PepMV is transmitted from infected seeds to seedlings (Moreno-Pérez et al. 2014; Werkman & Sansford 2010). One of the first reported experiments on seed transmission of PepMV was based on germinating a small number of seeds (n=50) in a 'grow-out test' or 'grow-out experiment' (Salomone & Roggero 2002). No infected plants were detected. However, an experiment using a larger number of seeds (n=168), indicated a transmission rate of about 1.8% (Córdoba-Sellés et al. 2007). A much greater number of seeds was used (n=87,000) in a third published grow-out experiment that confirmed transmission via seed (Hanssen et al. 2010). In this third experiment, the seed was cleaned to 'industry standards' using an acid and enzymatic treatment (Hanssen et al. 2010). The overall seed transmission rate was estimated to be 0.026%, after cleaning, and seed transmission rates for different tomato lines were estimated to vary from 0.057% to 0.005% (Hanssen et al. 2010).

PepMV has not been found in seed embryonic tissue and for this reason it is usually considered to be 'seed-borne' by virologists rather than 'seed-transmitted' (Ling 2008). PepMV virus particles are largely present on the outside of seeds, but they have also been detected in the seed coat (testa) (Ling 2008). Virus on the exterior of seeds may infect seedlings as they break through the seed coat.

Interceptions of PepMV-infected tomato seed

In 2020, Australian laboratories detected seeds infected with PepMV in two commercially traded tomato seed lots that had been sent to Australia for planting. One of the lots was produced in Central America and the other in Europe.

The publicly available EPPO and EUROPHYT records include reports of 37 interceptions of infected seed (Table A.1). However, the European PRA for PepMV indicates there were 64 notifications of non-compliance on seed between 2000 and May 2010, and that 106 infected tomato seed lots were detected through surveys in 2009 (Werkman & Sansford 2010). These detections showed the virus was present in several countries where it had not been previously reported. The virus was found in seed lots from countries in Africa, Europe, the Middle East and the Americas and in many seed lots from Asia. The detections showed that tomato seed production crops were often infected. This suggests that PepMV often goes undetected or unreported in seed production systems, for although PepMV-infected seed lots have been detected more than 150 times, infection of a seed production crop by the virus has only been reported twice (EPPO 2005c).

Year	Type of commodity	Country of origin	Country of destination	Number	Reference
2000	Seed	Netherlands	UK	1	(EPPO 2000a)
2004	Seed	Netherlands	Bulgaria	1	(EPPO 2004b)
2004	Seed	Netherlands	UK	1	(EPPO 2004e)
2005	Seed	Chile	France	1	(EPPO 2005b)
2005	Seed	Madagascar	France	1	(EPPO 2005b)
2007	Seed	Chile	France	1	(EPPO 2007a)
2008	Seed	Netherlands	Czech Republic	1	(EPPO 2008b)
2010	Seed	China	France	2	(EPPO 2010a)
2010	Seed	China	Poland	1	(EPPO 2010a)
2010	Seed	India	France	2	(EPPO 2010a)
2010	Seed	Israel	France	1	(EPPO 2010a)
2010	Seed	Senegal	France	1	(EPPO 2010a)
2010	Seed	Thailand	France	9	(EPPO 2010a)

Table A.1 Tomato seed carrying PepMV intercepted by other countries

Year	Type of commodity	Country of origin	Country of destination	Number	Reference
2010	Seed	USA	France	1	(EPPO 2010a)
2010	Seed	China	France	2	(EPPO 2010b)
2011	Seed	Israel	Bulgaria	1	(EPPO 2011c)
2011	Seed	USA	France	1	(EPPO 2011c)
2011	Seed	China	Italy	1	(EPPO 2011b)
2011	Seed	China	France	1	(EPPO 2011d)
2012	Seed	China	Germany	1	(EPPO 2012a)
2012	Seed	Italy	Malta	1	(EPPO 2012c)
2012	Seed	Vietnam	France	1	(EPPO 2012c)
2013	Seed	Italy	Malta	1	(EPPO 2013a)
2014	Seed	Chile	France	1	(EPPO 2014a)
2019	Seed	Dominican Republic	Undisclosed EU country	1	(EUROPHYT 2019)
2020	Seed	China	Undisclosed EU country	1	(EUROPHYT 2020)

Outbreaks of PepMV in other countries

Outbreaks of PepMV in tomato crops have been reported internationally, as have interceptions of PepMV-infected tomato seeds. The reports come from three sources: the scientific literature, a European PRA for PepMV (Werkman & Sansford 2010) and the European and Mediterranean Plant Protection Organization (EPPO). Tables A.1 and A.2 collate the reports from the EPPO and the scientific literature, but not the European PRA.

PepMV probably originated in South America, but the reports show there has been a widespread international outbreak of the virus across Africa, Asia, Europe and the Americas (Table A2) (CABI 2011; Clark & Crook 2012; Gómez, Sempere & Aranda 2012; Werkman & Sansford 2010). This kind of long-distance, transcontinental spread is best explained by transport of the virus with seed shipments. Seedlings and transplants are unlikely to have been traded from continent to continent and to remote locations, or if such trade in plant materials has occurred it is likely to have been very limited. The reports also suggest that control of the virus by current commercial processes has not been fully effective.

The international outbreak probably began in 1999 and has continued since then with more than 100 local outbreaks (Table A.2) (CABI 2011; Clark & Crook 2012; Gómez, Sempere & Aranda 2012). Before 1999 there were no reports of the virus in tomato crops, but in 1999 the virus was found in greenhouse tomato crops in Germany, the Netherlands, and the United Kingdom (EPPO 2002c; van der Vlugt et al. 2000). In 2000, PepMV was detected in tomato fruit production at 15 locations in four countries in Europe (Table A.2). In 2001, the virus was detected in tomato crops at 18 locations in ten countries in Europe and the Americas, and in 2002, it was detected at nine locations in seven countries in Europe and the Americas (Table A.2). There were relatively few reports of local outbreaks after 2011, possibly because they had become unremarkable.

The European PRA for PepMV provided a summary of surveys for PepMV by member states (MS) between 2000 and 2010 (Werkman & Sansford 2010). Fruit production sites and three categories of plant material were surveyed: tomato seed, plants for planting and fruit being marketed. Significantly, the European surveys provided evidence of many more outbreaks and detections than had been reported in other public sources (Werkman & Sansford 2010). The PRA indicated that 80 out of 461 fruit lots on the European market were found to be infected when surveyed in 2009 (Werkman & Sansford 2010).

It is suspected that the international outbreak was due in part to trade in infected seed (CABI 2011; Clark & Crook 2012). Many infected seed lots have been intercepted in Europe (Table A.1). The nearly simultaneous outbreaks in three European countries in 1999 (EPPO 2002c), suggest that infected tomato seed was probably the source. Local spread by other transmission mechanisms between the three locations is unlikely to account for the correlated timing, as local spread takes longer to cover such distances. Tellingly, PepMV was detected in seed from the Netherlands in 2000 (EPPO 2000a) and in a seed crop in Chile in 2001 (EPPO 2005c).

Considering the detections of the virus in crops and in seed, the European PRA for PepMV concluded that 'there is uncertainty regarding the exact distribution of PepMV' (Werkman & Sansford 2010). New Zealand also published a risk analysis of PepMV in tomato seeds and considered that this virus was likely more widespread than currently officially recognised (CABI 2011; Clark & Crook 2012).

Outside Europe, PepMV has been found affecting tomato crops in Canada, China, Chile, Ecuador, Guatemala, Israel, Peru, Mexico, Morocco, South Africa, Thailand and in the USA (Carmichael et al. 2011; Klap et al. 2020; Ling & Zhang 2011; Werkman & Sansford 2010).

Year	Facility	Country	Number of reports	References
1999	Tomato crops	UK	Unknown	(EPPO 2000a; Wright & Mumford 1999)
1999	Tomato crops	Netherlands	Unknown	(van der Vlugt et al. 2000)
1999	Fruit production	Germany	1	(EPPO 2002c)
2000	Fruit production	UK	3	(EPPO 2001a)
2000	Fruit production	Germany	1	(EPPO 2001a, 2002c; Lesemann et al. 2000)
2000	Fruit production	Netherlands	5	(EPPO 2001a)
2000	Fruit production	Spain	6	(EPPO 2003e)
2001	Glasshouse tomato plants	Italy	1	(EPPO 2001a; Roggero 2001)
2001	Fruit production (glasshouses)	Finland	6	(EPPO 2001a; KTTK 2002; Lemmetty et al. 2011)
2001	Glasshouse tomato plants	Germany	1	(EPPO 2001b, 2002c)
2001	Glasshouse tomato plants	Canada	1	(French et al. 2001)
2001	Fruit production	USA	4	(French et al. 2001)
2001	Glasshouse tomato plants	Sweden	1	(EPPO 2002c)
2001	Fruit production	Norway	1	(EPPO 2002c)
2001	Fruit production	Belgium	1	(EPPO 2003e)

Table A.2 Reports of PepMV outbreaks in tomato

Year	Facility	Country	Number of reports	References
2001	Fruit production (Nursery)	Denmark	1	(EPPO 2003e)
2001	Tomato crops for seed	Chile	1	(EPPO 2005c)
2002	Glasshouse tomato plants	Poland	1	(EPPO 2002c, 2003b)
2002	Fruit production (Nursery)	Denmark	1	(EPPO 2003e)
2002	Fruit production	Belgium	1	(EPPO 2003e)
2002	Fruit production (glasshouses)	France	1	(EPPO 2003e)
2002	Tomato crops	Peru	1	(EPPO 2002b)
2002	Fruit production	Ireland	1	(EPPO 2003e)
2002	Fruit production	UK	3	(EPPO 2003e)
2003	Fruit production (glasshouses)	Finland	1	(EPPO 2003a)
2003	Fruit production (glasshouses)	Germany	1	(EPPO 2003c)
2003	Tomato plants (non-commercial plastic house)	Slovakia	1	(EPPO 2004a)
2004	Fruit production (glasshouse)	Bulgaria	1	(EPPO 2004b)
2004	Fruit production	UK	5	(EPPO 2004e)
2004	Fruit production (glasshouses)	Hungary	1	(EPPO 2004c)
2004	Tomato crops	Switzerland	1	(EPPO 2006; Staubli 2005)
2002	Tomato crops for seed	Chile	1	(EPPO 2005c)
2005	Tomato crops	Chile	1	(EPPO 2005c)
2005	Glasshouse tomato plants	Italy	1	(Davino et al. 2006)
2005	Fruit production	Poland	Unknown	(Pospieszny & Borodynko 2006)
2006	Fruit production	Austria	3	(EPPO 2007b)
2007	Glasshouse tomato plants	USA	Unknown	(EPPO 2008c)
2008	Fruit production (glasshouse)	Czech Republic	1	(EPPO 2008b)
2008	Tomato crops	Spain	1	(Alfaro-Fernández et al. 2008)
2008	Tomato crops (glasshouse)	Turkey	1	(Özdemir 2010)
2008	Fruit production	South Africa	1	(Carmichael et al. 2011)
2009	Tomato crops (glasshouse)	Turkey	1	(Özdemir 2010)
2009	Tomato crops (glasshouse)	Cyprus	1	(EPPO 2012d)
2010	Fruit production (glasshouse)	Italy	6	(EPPO 2011g)
2010	Fruit production (field)	Cyprus	3	(Papayiannis, Kokkinos & Alfaro- Fernández 2012)
2010	Fruit production (glasshouse)	Greece	2	(Efthimiou et al. 2011; EPPO 2012d)
2010	Fruit production (glasshouse)	Mexico	1	(EPPO 2012d; Ling & Zhang 2011)
2011	Fruit production (glasshouse)	Syria	1	(Fakhro et al. 2010)
2011	Glasshouse tomato plants	Italy	2	(EPPO 2011h)
2011	Fruit production	Finland	1	(EPPO 2011f)
2011	Fruit production	UK	3	(EPPO 2012e)
2011	Fruit production (glasshouse)	Croatia	3	(Novak, Milanovic & Kajic 2012)
2011	Tomato crops	Greece	Unknown	(Efthimiou et al. 2011)

Year	Facility	Country	Number of reports	References
2011	Tomato crops	Mexico	Unknown	(Ling & Zhang 2011)
2011	Tomato crops	South Africa	Unknown	(Carmichael et al. 2011)
2012	Fruit production	Switzerland	3	(EPPO 2012f)
2012	Fruit production (glasshouse)	Lithuania	2	(Šneideris et al. 2013)
2015	Fruit production	Spain	2	(EPPO 2016a)
2016	Fruit production (glasshouse)	Morocco	7	(Imane 2016; Souiri et al. 2017)
2019	Fruit production (glasshouse)	Israel	1	(Klap et al. 2020)

Appendix B: Pospiviroids

Pospiviroid biology

Viroids consist of circular duplexed ribonucleic acid (RNA) molecules, typically 239 to 401 nucleotides long (Gross et al. 1978; Steger & Riesner 2003).

Pospiviroids are generally easily transmitted 'mechanically' when plants are intentionally cut or accidentally abraded during normal horticultural activities. Transmission typically occurs through contact with contaminated pruning tools, farm equipment, clothing and people's hands (Owens & Verhoeven 2009; Sabaratnam 2012; Singh, Ready & Nie 2003b). They can also be transmitted through grafting and vegetative propagation, and they are naturally transmitted by contact between neighbouring plants, through pollen and seed and, in some specific circumstances, by aphids and bumble-bees (Galindo 1988; Galindo, Lopez & Aguilar 1986; Owens & Verhoeven 2009; Salazar et al. 1983; Singh 1970; Singh, Boucher & Somerville 1992; Singh, Ready & Nie 2003b).

Pospiviroid disease in tomatoes

Infection levels of viroids in tomato crops can vary, typically being below 10% and sometimes below 1%, but in some cases reaching a large proportion of the crop and in a few cases nearly 100% (Antignus et al. 2002; EFSA 2011). Most spread within tomato crops is by mechanical transmission (Antignus et al. 2002; Owens & Verhoeven 2009; van Brunschot et al. 2014; Verhoeven et al. 2004). Whereas there are many reports of pospiviroid infections in greenhouse tomato crops, few reports of infected tomato field crops were found (Barbetti et al. 2012; Ling et al. 2012; Mackie et al. 2016; Pur Rahim et al. 2009).

Infected tomato plants show a range of symptoms. In most reported infections, tomato plants are stunted and have chlorotic or bronzed leaves that are distorted or small. Some plants may lose leaves or have brittle leaves or develop patches of necrosis on the leaves, and in some cases on the stems (Behjatnia 1996; Galindo, Smith & Diener 1982; Martínez-Soriano et al. 1996; McClean 1948; Mishra et al. 1991; Owens 1990; Owens, Candresse & Diener 1990; Singh 1973; Singh, Nie & Singh 1999; Verhoeven et al. 2004; Walter 1987). Infected plants usually produce fewer and smaller fruit, and in some severe cases no fruit at all (Owens & Verhoeven 2009; Singh, Ready & Nie 2003b).

Diagnosis is difficult because the symptoms are neither distinctive nor diagnostic. Different pospiviroids cause similar symptoms, which may be easily confused with symptoms caused by other pathogens, and possibly by herbicide damage (Blancard 2012; EFSA 2011).

The severity of symptoms varies considerably, with the most severely affected plants being stunted and expressing most of the typical symptoms, and the most mildly affected plants having few or no symptoms. Symptom severity is believed to be dependent upon the variant of the viroid, the cultivar of the tomato, and temperature and light levels (EFSA 2011; Singh, Ready & Nie 2003a). Symptom severity also varies as much within some pospiviroid species as between species (EFSA 2011; Runia & Peters 1980).

Some tomato cultivars do not produce symptoms (being asymptomatic) when infected by certain pospiviroid variants, or may only produce very mild symptoms (Barbetti et al. 2012;

O'Brien 1972; Owens & Verhoeven 2009; Singh 1973; Stark-Lorenzen et al. 1997). Asymptomatic infections cannot be detected visually, but only by sampling and laboratory testing.

Tomato seedlings infected with pospiviroid species may not produce symptoms for more than six weeks after germination, and some may not produce symptoms at all when grown from infected seeds (Kryczynski, Paduch-Cichal & Skreczkowski 1988) (Singh & Dilworth 2009). Viroid replication and symptom development is enhanced by high temperatures and light levels (Singh 1983; Singh, Ready & Nie 2003a).

Viroid presence in seeds and floral parts

Pospiviroids that naturally infect tomato have been detected in samples of tomato seed (Antignus, Lachman & Pearlsman 2007; Chambers et al. 2013; FERA 2009b; Marach 2008; Singh & Dilworth 2009). However, whether a pospiviroid infection is present within the seed or only on the seed surface has been questioned. This localisation may influence transmission through seed, testing protocols, and whether treating tomato seed with chemical agents will eliminate the viroid.

The localisation of pospiviroid RNA in plant tissues in general has been investigated using *Potato spindle tuber viroid* (PSTVd), *Tomato apical stunt viroid* (TASVd) and *Tomato chlorotic dwarf viroid* (TCDVd). PSTVd RNA has been detected within tomato seed and in all the floral parts of infected tomatoes that have been tested, including the ovaries, ovules and pollen (Lykke et al. 2010; Singh 2006; Singh & Dilworth 2009; Zhu et al. 2001). Similarly, experiments have shown the RNA of TASVd and TCDVd is present within the seed of infected tomatoes, and TASVd RNA is present in the petals, sepals, ovaries and stamens of infected tomatoes (Antignus, Lachman & Pearlsman 2007; Singh & Dilworth 2009). These findings are consistent with earlier work that showed that PSTVd RNA is present in the pollen, sepals, fruit and true botanical seed of infected potato plants (Fernow, Peterson & Plaisted 1970; Salazar et al. 1983; Singh, Boucher & Somerville 1992; Singh, Boucher & Wang 1991). These findings are also consistent with later work demonstrating that PSTVd RNA is present within petunia seeds, ovaries and embryos (EPPO 2016b; Matsushita & Tsuda 2014b).

Pospiviroid RNA is also found in tomato fruit flesh (van Brunschot et al. 2014) and hence will be present on seed surfaces. However, PSTVd, TASVd and TCDVd were not eliminated from seeds when the seeds were soaked in bleach (sodium hypochlorite) or alkali solutions to destroy RNA, confirming that the viroids are protected within seeds as well as being present on seed surfaces (Antignus, Lachman & Pearlsman 2007; Singh & Dilworth 2009).

The seed cleaning and localisation data suggest that attempts to eliminate pospiviroids by treating seeds will not be effective, given that the pospiviroid RNA may be within the seeds. However, such processes are likely to reduce the amount of pospiviroid RNA present in seed samples, making it more difficult to detect if seed samples are tested.

Viroid transmission through tomato seeds to seedlings

The pospiviroids subject to emergency measures and considered in this PRA have been shown to be transmitted through tomato seeds in experiments where seedlings are grown from seeds collected from infected parent plants (Table B.1, and references therein).

Seed-transmission of pospiviroids may be due to the presence of pospiviroid RNA in embryos, ovules and pollen. However, some seedlings may be infected by pospiviroid RNA present on seed surfaces or in seed coats, given that pospiviroids are easily mechanically transmitted and because abrasion occurs when the seeds germinate and break through the seed coat.

Experiments indicate that the rate of transmission varies widely (Table B.1). Experiments to assess PSTVd transmission through tomato seeds show the extent of the variation, with one experiment failing to observe transmission, several observing transmission rates of 5–10%, and one finding a transmission rate of 50.9% (Table B.1). These inconsistencies suggest that unrecognised environmental, physiological, epidemiological, or genetic factors affecting seed transmission have not been replicated or controlled in the reported grow-out experiments. The inconsistencies indicate that there is a level of uncertainty, but the balance of evidence indicates seed transmission.

The rate of transmission varies with the host plant and probably also varies between and within pospiviroid species. The rate of transmission of isolates of both TASVd and TCDVd through tomato seed has been estimated to be 80% (Antignus, Lachman & Pearlsman 2007; Singh & Dilworth 2009).

There may be several explanations for experiments where no transmission was detected through tomato seeds to seedlings, and for variations in the rate of transmission (Fox & Monger 2011). It was suggested that the temperature at which parent plants are grown and when seedlings are germinating may affect transmission (Chung & Pak 2008). Pospiviroid seed transmission may also be reduced in some tomato cultivars (Matsushita & Tsuda 2016; Singh, Boucher & Somerville 1992).

Sequence variation is known to influence viroid biology (Singh, Ready & Nie 2003a; Yanagisawa et al. 2019) and might affect seed transmission. In one example, a variant of PSTVd with mild effects was transmitted through small numbers of seeds, whereas transmission of a variant with severe effects was not detected (Khoury et al. 1988). In another example, transmission through tomato seed was detected using a variant of TCDVd that differed at a few nucleotide positions from a variant of TCDVd that apparently was not transmitted (Singh & Dilworth 2009; Singh, Nie & Singh 1999).

It is possible that variations in pospiviroid RNA levels in the seed also affect seed transmission. Tests of PSTVd in infected tomato seeds found that the level of viroid RNA was very variable, with some individual seeds being 'highly contaminated' whereas other seeds had 'low PSTVd concentrations' (Lykke et al. 2010).

Publication	Viroid species	Host plant	Transmission rate ^a
Benson and Singh (1964)	PSTVd	Solanum lycopersicum	7.9–11.1%
Hunter, Darling and Beale (1969)	PSTVd	Solanum tuberosum	87–100%
Fernow, Peterson and Plaisted (1970)	PSTVd	Solanum tuberosum	0-100%
Singh (1970)	PSTVd	Solanum lycopersicum	6–11%
		Solanum tuberosum	6–12%

Table B.1 Experiments on pospiviroid transmission through seed of Solanaceae to seedlings

Publication	Viroid species	Host plant	Transmission rate ^a
Singh and Finnie (1973)	PSTVd	Scopolia sinensis	71%
Grasmick and Slack (1987)	PSTVd	Solanum tuberosum	100%
Khoury et al. (1988)	PSTVd	Solanum lycopersicum	5%
Kryczynski, Paduch-Cichal and Skreczkowski (1988)	PSTVd	Solanum lycopersicum	>10%
Singh, Nie and Singh (1999)	TCDVd	Nicotiana physaloides Nicotiana debneyi Physalis angulata Physalis oridana Solanum lycopersicum Solanum tuberosum	No transmission observed
Lebas et al. (2005)	PSTVd	Capsicum annuum	No transmission
		Solanum lycopersicum	observed
Antignus, Lachman and Pearlsman (2007)	TASVd	Solanum lycopersicum	80%
Marach (2008)	Columnea latent viroid (CLVd)	Solanum lycopersicum	Transmitted but transmission rate not reported
Koenraadt et al. (2009)	TCDVd	Solanum lycopersicum	No transmission observed
Singh and Dilworth (2009)	TCDVd	Solanum lycopersicum	80%
Verhoeven et al. (2009)	Pepper chat fruit viroid (PCFVd)	Capsicum annuum	19%
Candresse et al. (2010)	TCDVd	Solanum lycopersicum	<1% b
Fox and Monger (2011)	CLVd	Solanum lycopersicum	No transmission observed
van Brunschot et al. (2014)	PSTVd	Solanum lycopersicum	<1% b
Matsushita and Tsuda (2014a)	PSTVd	Petunia × hybrida	51.7-78%
Faggioli et al. (2015)	PSTVd	Solanum lycopersicum	No transmission observed
	CLVd	Solanum lycopersicum	No transmission observed
	TASVd	Solanum lycopersicum	No transmission observed
Simmons, Ruchti and Munkvold (2015)	PSTVd	Solanum lycopersicum	50.9%
Matsushita and Tsuda (2016)	PSTVd	Solanum lycopersicum	0-90.2%
		Solanum melongena	No transmission observed
		Capsicum annuum	0-0.5%
		Petunia × hybrida	81%
	TCDVd	Solanum lycopersicum	No transmission observed
		Solanum melongena	No transmission observed
		Capsicum annuum	No transmission observed

Publication	Viroid species	Host plant	Transmission rate ^a
		Petunia × hybrida	25%
	TASVd	Solanum lycopersicum	No transmission observed
	CLVd	Solanum lycopersicum	0-100%
		Solanum melongena	No transmission observed
Yanagisawa and Matsushima	PCFVd	Capsicum annuum	0%
(2017)		Petunia × hybrida	0-91.9%
		Solanum lycopersicum	0-1.4%
	PSTVd	Petunia × hybrid	20.8-97.7%
Batuman et al. (2019)	PSTVd	Solanum lycopersicum	0-2%
	TASVd	Solanum lycopersicum	0-2%
Bhuvitarkorn and Reanwarakorn (2019)	CLVd	Solanum melongena	2.3-82%
Gramazio et al. (2019)	TCDVd	Solanum melongena	7.7–100%
Verhoeven et al. (2020)	CLVd	Capsicum annuum	No transmission observed
	PCFVd	Capsicum annuum	No transmission observed
	PSTVd	Capsicum annuum	No transmission observed
	TASVd	Capsicum annuum	No transmission observed

a. Transmission rate estimates have usually been made by determining the number of infected seedlings grown from a known number of seeds from infected parent plants and has not involved any statistical method or tests of the proportion of seeds carrying the viroid. **b.** Candresse et al. (2010) and van Brunschot et al. (2014) reported results of grow-out tests from commercially produced seed lots and hence the rates of transmission are probably affected by dilution of seeds from infected parent plants.

Outbreaks in tomato crops in other countries

Outbreaks of the pospiviroids, which are subject to emergency measures and considered in this PRA, have occurred in tomato crops in disparate locations across the world are frequently reported (Table B.2 and references therein). These outbreaks and the paucity of prior records indicate the pospiviroids are emerging pathogens (Anderson et al. 2004).

The reports of outbreaks present a picture of global distribution that is best explained by international transport of the pathogens with seed, and their introduction to crops through seed, along with instances of local transmission. The reports of the pospiviroids spreading from one continent to another, across great distances and to isolated locations emphasize the importance of long-distance spread, which is best explained by transport with seed shipments. Seedlings and transplants are unlikely to have been traded in significant volumes from continent to continent, over great distances or to remote locations, as it is considerably easier to transport seed. Examples of this kind of evidence of spread include the appearance of CLVd in Africa, Asia and Europe, PCFVd in Asia, Europe and North America, PSTVd in Australia and New Zealand, TASVd in Africa, Asia, and Europe and TCDVd in Asia, Europe and North America (Table B.2).

Viroid species	Countries	Report	
Columnea latent viroid	Belgium	(Verhoeven et al. 2004)	
	The Netherlands	(Verhoeven et al. 2004)	
	Thailand	(Tangkanchanapas et al. 2005)	
	Portugal	(CSL 2007; Monger & Mumford 2006)	
	France	(CSL 2007; Steyer et al. 2009)	
	United Kingdom	(CSL 2007; Nixon et al. 2009; Sansford & Morris 2009)	
	Italy	(Parrella, Crescenzi & Pacella 2011)	
	Mali	(Batuman & Gilbertson 2013)	
Pepper chat fruit viroid	The Netherlands	(Verhoeven et al. 2009)	
	Canada	(Verhoeven et al. 2011)	
	Thailand	(Reanwarakorn, Klinkong & Porsoongnurn 2011)	
Potato spindle tuber viroid	New Zealand	(Elliot et al. 2001; Lebas et al. 2005)	
	The Netherlands	(EPPO 2003d; NPPO the Netherlands 2013; Verhoeven et al. 2004; Werkman, Verhoeven & Roenhorst 2007)	
	Germany	(EPPO 2004d; Werkman, Verhoeven & Roenhorst 2007)	
	United Kingdom	(CSL 2008; FERA 2011; Mumford, Jarvis & Skelton 2004)	
	Belgium	(Verhoeven et al. 2007a; Werkman, Verhoeven & Roenhorst 2007)	
	Canada	(Werkman, Verhoeven & Roenhorst 2007)	
	Austria	(EPPO 2008a)	
	Italy	(Navarro et al. 2009)	
	Japan	(Matsuura et al. 2010)	
	United States	(Li et al. 2012; Ling et al. 2012; Ling & Sfetcu 2010)	
	Ghana	(Batuman et al. 2013)	
	Dominican Republic	(Ling & Li 2014)	
Tomato apical stunt viroid	Cote d'Ivoire	(Walter 1987; Walter, Thouvenel & Fauquet 1980)	
	Indonesia	(Candresse, Smith & Diener 1987)	
	Israel	(Antignus et al. 2000)	
	Tunisia	(Verhoeven, Jansen & Roenhorst 2006)	
	Senegal	(Candresse et al. 2007)	
	The Netherlands	(Verhoeven et al. 2012)	
	France	(EPPO 2013b)	
	Ghana	(Batuman et al. 2013)	
	Italy	(Parrella & Numitone 2014)	
Tomato chlorotic dwarf viroid	Canada	(Singh, Nie & Singh 1999)	
	United States	(Ling et al. 2009; Olmedo-Velarde et al. 2019; Verhoeven et al. 2004)	
	Japan	(Matsushita et al. 2008)	
	Mexico	(Ling & Zhang 2009)	
	France	(Candresse et al. 2010)	

Table B.2 Reports of pospiviroid outbreaks in tomato outside Australia
Viroid species	Countries	Report
	Norway	(Fox et al. 2013)

International interceptions and initiation of outbreaks from seed

Since 2002, countries in Europe and the Middle East have intercepted tomato seed carrying PSTVd (Table B.3). These interceptions provide a likely explanation for the international outbreaks of PSTVd (Table B.2), as they suggest the outbreaks were probably caused by the inadvertent distribution of PSTVd-infected tomato seeds through the international seed trade.

It is likely that outbreaks of other viroid species in tomato crops (Table B.2) have the same cause, namely trade in infected tomato seed. Australia has intercepted tomato seed infected with several pospiviroid species, supporting this broader inference. It seems likely that other countries have not detected the pospiviroid species because seed imports were not tested for the wider range of pospiviroid species.

Year	Type of commodity	Country of origin	Country of destination	Number	Reference
2002	Seed	India	Austria	1	(CABI-EPPO 2002)
2002	Seed	Thailand	Austria	2	(EPPO 2002a)
2007	Seed	Israel	Not named	1	(European Commission 2008)
2008	Seed	Netherlands	Austria	1	(EPPO 2008a)
2008	Seed	Israel	Not named	1	(European Commission 2008)
2011	Seed	China	Not named	1	(EUROPHYT 2012b)
2011	Seed	China	Israel	1	(EPPO 2011a)
2011	Seed	Kenya	Israel	1	(EPPO 2011a)
2011	Seed	Netherlands	Israel	1	(EPPO 2011a)
2011	Seed	USA	Israel	1	(EPPO 2011a)
2011	Seed	China	Italy	1	(EPPO 2011b)
2012	Seed	China	Not named	1	(EUROPHYT 2012c)
2012	Seed	China	Austria	1	(EPPO 2012c)
2012	Seed	China	Not named	1	(EUROPHYT 2012a)
2012	Seed	China	Austria	1	(EPPO 2012b)
2014	Seed	China	Italy	1	(EPPO 2014c)
2014	Seed	China	Slovenia	1	(EPPO 2014b)
2015	Seed	China	Denmark	1	(EPPO 2015)
2017	Seed	China	Italy	1	(EPPO 2017a)

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There is also evidence that links outbreaks directly to infected tomato seed. In one instance, a trial line of tomato seed that was planted in the same greenhouse as a large crop was found to be the source of an outbreak of PSTVd in that crop (van Brunschot et al. 2014). A sample of 370 seeds from the trial line was grown out and one seedling was found to be infected with PSTVd. The PSTVd isolate from the seedling had the same genetic sequence as the PSTVd identified in the outbreak, and it was concluded that the infected trial line was the source.

In another example, outbreaks of CLVd in four tomato crops of the same cultivar in the United Kingdom were linked to seed, although follow-up experiments were unable to demonstrate seed transmission (Fox & Monger 2011; Nixon et al. 2009). An investigation of the outbreaks found that a seed lot used for the crops was carrying the same viroid at a low level (Fox & Monger 2011).

In a third example, an outbreak of TCDVd in tomato crops in France was linked to a seed lot that was found to be carrying the viroid (Candresse et al. 2010).

By contrast, Koenraadt et al. (2009) did not find a single seedling infected among 4,000 grown using seed from parent plants that were infected with TCDVd. Similarly, Faggioli et al. (2015) failed to observe transmission of four other pospiviroids via seed to approximately 4700 tomato seedlings. It is possible that some environmental factor reduced or eliminated seed transmission in these experiments, or that the transmission rate may have been so small as to be undetectable when these numbers of seeds were germinated.

Taken together, the evidence indicates some outbreaks are initiated by infected seeds in traded seed lots. The evidence also indicates outbreaks may be initiated when very few infected seeds are present in otherwise uninfected seed lots (Candresse et al. 2010; Fox & Monger 2011; van Brunschot et al. 2014). When few seeds are infected and transmission rates are small, the significant numbers of seeds that are planted to establish tomato crops is an important factor. It is noted that outbreaks of other seed-transmitted pathogens can arise from very few infected seeds and can arise even when the pathogen has a low seed transmission rate (Agarwal & Sinclair 1996).

Pospiviroid sources: reservoirs and alternative hosts

Scientists have proposed two other possible causes of outbreaks of pospiviroids in tomato crops, both being relevant to Australia. Firstly, it was suggested that the viroids are transmitted from ornamental plants, as the viroids have been detected many times in ornamental plants from the family Solanaceae (Shiraishi et al. 2013; Verhoeven et al. 2012). Secondly, pospiviroids have been detected in wild plants and weeds in two countries, and it has been proposed that these plants may be sources of the viroids.

Although it is possible some weeds act as reservoirs at some locations, the outbreaks of pospiviroids in different countries (Table B.2) cannot be explained simply in terms of natural spread from these weeds. No natural transmission pathway involving weeds has been identified or proposed that could explain the long-distance movement between crops in different continents.

Interceptions of pospiviroid-infected seed by Australia

Since 2009, Australian laboratories have detected tomato seed lots contaminated with pospiviroid-infected seeds through on-arrival testing under the emergency measures.

Constable et al. (2019) and Dall et al. (2019) presented an analysis of the numbers of contaminated tomato seed lots and the pospiviroid species detected through the testing done by the Australian laboratories between 2008 and 2016. More than 2,000 seed lots were tested over the period. Pospiviroids were detected in more than 10% of the seed lots in the first years of

mandatory testing, but the proportion of lots that were contaminated with infected seeds declined after 2011 to less than 5%.

Of the pospiviroids subject to emergency measures and considered in this PRA, CLVd, PCFVd, PSTVd, TCDVd and TASVd were detected. These pospiviroids were detected in seed lots exported from countries from every production region. However, none of the tested seed lots were found to be carrying PepMV.

Overall, 21 countries produced pospiviroid-infected seed that was sent to Australia between 2012 and 2017, including three countries in Africa, five in the Americas, eight in Asia, four in Europe and one in the Middle East. When viewed collectively, this data from seed testing showed these pathogens have a very wide geographic distribution, and that seed crops are infected in every major production region.

Appendix C: Commercial tomato seed production and trade

Tomato seed production and trade

Tomato fruit is a significant vegetable crop globally. The 'scale of production, trade and distribution has increased tremendously' in recent years (GSPP 2013), and therefore also tomato seed production and its trade. Australian tomato fruit production crops are predominantly grown from imported seed.

The department's records indicate that on average 760kg of tomato seed are imported into Australia annually. Most imported seed are thought to be first generation (F1) hybrid seed produced by cross pollination (hybridisation) of parental lines. Tomato hybrids are reported to have better vigour, uniformity, disease resistance and stress tolerance, and to have desirable horticultural traits including early fruiting, longer shelf life and consistent yield (IIGB 2016).

The production process begins with plant breeding and involves the production of parent lines which are usually hybridised to produce the seed. After harvesting the fruit, the seed is extracted and separated from the pulp using processes that clean the seed. Typically, the pulp is fermented for several hours, washed with an acidic solution, and then washed with water several times. This extraction process may be undertaken on site by the farm workers. The seed may be further treated with chemicals including fungicides, and it may be primed for germination or pelleted. These treatments often occur in another country to that where the seed was produced. After extraction and treatment, seed lots may be stored for several years. Portions of lots are sold to fruit production growers and nurseries in many countries.

International tomato seed production

The hybridisation step is important as it is labour intensive, and for this reason hybrid tomato seed is commonly produced in countries where labour costs are low (IIGB 2016; ISF 2017a).

Hybrid tomato seed sent to Australia is produced from crops grown in countries in Asia, Europe, Africa, the Middle East and the Americas.

The IIGB noted that:

• Seeds present significant biosecurity risks due to the numerous complex, variable international production pathways, including contracted farms in countries where biosecurity might not always be consistent with Australian standards (IIGB 2016).

The production of hybrid tomato seed lots often involves activities in several countries (IIGB 2016; ISF 2017a). Plant lines used to produce hybrid seed may be grown, selected and multiplied in two or three countries successively (IIGB 2016; ISF 2017a; Werkman & Sansford 2008). As an example, two parental lines may be bred in the Netherlands, then larger quantities of seed of these lines (basic seed) may be produced in France or Spain, and this basic seed may then be grown in Thailand or China where the tomato flowers are cross-pollinated to produce the hybrid seed (IIGB 2016; ISF 2017a). In another example observed by the department, parental lines bred and selected in a country in the Northern Hemisphere were sent to a country in the Southern Hemisphere where breeding and selection continued so that two seasons of breeding were achieved in a single year.

Following its production, tomato seed may be trans-shipped via airfreight through other countries. Some tomato seed sent to Australia is trans-shipped through other countries, for example France, Israel, Japan, the Netherlands or the USA.

Lot numbers are usually used to identify seed production lots produced on one farm or field in one season. The IIGB was informed that in Thailand seed lot numbers were retained unaltered from the field production site through to the sale of the seed (IIGB 2016).

During its processing and shipment, tomato seed from one lot is commonly divided up into a series of batches. Batches may be treated differently and are commonly re-packaged. Each time a batch is divided, treated, or repackaged, the batch and its derivatives are usually assigned new batch numbers.

Production supervision

When considering biosecurity, it is notable that seed trading businesses selling tomato seed to Australian growers do not usually produce the seed, nor do they fully supervise the production. Instead the supervision is sub-contracted to businesses that work in the countries where the seed is produced (Tay 2002; Venkateswarlu 2007). Subcontractors that organise production, commonly contract out production of seed to many different growers (Tay 2002; Venkateswarlu 2007).

The IIGB observed these business relationships in Thailand and noted that:

- Major international seed companies contract out vegetable seed production (including tomato and carrot seeds) in one or more of around 25 countries (IIGB 2016).
- Larger seed companies typically contract out the production and multiplication processes to farmers, farmers' associations, or private firms, often in countries with low production costs (IIGB 2016).

The IIGB (2016) reviewed the production of hybrid tomato seed by one company in Thailand and noted a range of phytosanitary and traceability measures. It is not clear whether the same phytosanitary measures, or similar ones, are practiced in other countries or practised by other seed production businesses.

The measures in Thailand noted by the IIGB included:

- inspections by field supervisors, who are employees of the subcontracting business, to monitor crop production and sanitation practices and ensure records of pest and disease incidents and the use of chemicals are maintained.
- inspection by a visiting plant pathologist, who is an employee of the parent seed trading company, to ensure that pest and disease incidents are managed as early as possible, and ensure that production practices meet agreed protocols and requirements.
- collection of leaf samples from diseased plants by the plant pathologist which are sent to the parent company laboratory for testing.
- inspections by quarantine inspectors of the Thailand Department of Agriculture.

Biosecurity concerns related to tomato seed production systems

The level of biosecurity practised in tomato seed production systems varies considerably. The phytosanitary measures noted by the IIGB may reduce certain phytosanitary risks. However, other aspects of tomato seed production may introduce or increase phytosanitary risks.

Parental plants for tomato seed production are commonly grown in open fields (IIGB 2016) and so the plants are exposed to invertebrates that may transmit seed-transmitted plant pathogens to the plants. Equally importantly, because hybridisation involves emasculation of flowers and pollination by hand (Cheema & Dhaliwal 2005) it has the potential to mechanically transmit and spread certain plant pathogens.

By growing plant lines in several different countries successively, the plant lines may be exposed to a greater range of pathogens than present in a single country. Furthermore, the places where seed production crops are grown change relatively often, as tomato crops are typically rotated every year in response to pest and pathogen pressures (Gould 2013). Moreover, the location of crops will change as the organisers and farm businesses, which are independent of the seed trading businesses, make decisions about subcontracted seed production work every year (Venkateswarlu et al. 2015).

In describing the tomato seed production process, the IIGB (2016) provided evidence that:

- workers in the tomato seed crops become contaminated by plant sap;
- while the identities of suspected bacterial infections of tomato plants were investigated, tomato plants suspected to be infected by viruses were disposed of without the infection necessarily being investigated, and
- seed infected with PepMV might be packaged before it is disinfected.

This evidence is significant because the virus species and viroids reviewed in this report are transmitted when worker's hands and equipment are contaminated. Under these circumstances, the pathogens are transmitted through plant sap and by minor accidental abrasions of plants.

The evidence provided by the IIGB (2016) is also significant because when the causes of plant disease symptoms are not investigated in a seed production crop, as indicated for suspected viral infections, the business and the country concerned may not be aware of the health status of the exported seed. Furthermore, the packaging of seed from plants infected with PepMV that has not been decontaminated may contaminate packaging materials, equipment and other seed lots.

Seed lots infected with the pospiviroids have been detected many times by Australian laboratory testing, and seed lots infected with PepMV have been detected many times by European testing (Appendices A and B). Based on this evidence, it appears that the standard commercial practices used by the tomato seed production industry do not ensure that exported seed is free of these pathogens. In many instances infecting pathogens are not detected or identified until the seed reaches the Australian border.

Exporting countries usually manage the phytosanitary risks of exported seed by certifying the phytosanitary condition of seed lots. Exporting countries commonly visually inspect seed crops to assure themselves that certain pests are not present in the crop. However, many infections cannot be detected by visual inspection. It is difficult to identify PepMV-infected and

pospiviroid-infected plants (Appendices A and B), and sometimes these plants are asymptomatic. It is also difficult to comprehensively inspect large crops and many small farms. Therefore, visual inspection of crops is not a suitable method for detecting these pathogens.

The International Seed Federation (ISF) has also recognised that phytosanitary certification of seed can be challenging because the destination of the seed may not be known when the seed is produced (ISF 2017a). Failing to retain the seed lot number, which distinguishes the place and season of production, and re-packaging and re-labelling of seed lots may also make phytosanitary certification difficult or unreliable.

Another element of the phytosanitary risk relates to the distribution and cultivation of trial lines or trial lots of tomato seed. Fruit production businesses and seed businesses collaborate to grow trial lines to determine their suitability for Australian conditions. Tomato trial lines are usually grown at the same time and in the same place as larger fruit production crops. Significantly, trial lines are sometimes a source of pospiviroid outbreaks (van Brunschot et al. 2014) and some trial lines of seeds have been found to be contaminated with infected seeds by Australian seed testing (Appendix B).

Appendix D: Issues raised on the draft report and responses

The Department of Agriculture, Water and the Environment released the '*Draft pest risk analysis for Pepino mosaic virus and pospiviroids associated with tomato seed*' in August 2018 for stakeholder consultation. A WTO SPS notification G/SPS/N/AUS/455 was issued at that time.

Comments were received on the draft report of the pest risk analysis from stakeholders, including industry representatives, trading partners and state and territory governments. A summary of the key issues raised, and the department's responses, is provided.

Issue 1: Host and pest status

Absence of the identified pests from Australia

Stakeholders suggested that the identified pospiviroids and *Pepino mosaic virus* (PepMV) may be present in Australia, but no specific evidence was provided in support of this claim.

Australia's national biosecurity system is extensive, complex and multi-layered, with complementary measures applied off-shore, at the border and on-shore. It is a shared responsibility undertaken by a broad range of participants, covering all Australian governments, industry bodies and other stakeholders. Measures are in place to prevent the entry of quarantine pests and to mitigate the risk of their establishment and spread should they enter Australia. Post-border elements include general surveillance, routine monitoring, extension advice to producers and diagnostic services, as well as specific surveys. If a pest incursion occurs, Australian governments respond with coordinated regulatory actions with the affected parties.

Australia implemented emergency measures in response to the emerging risks posed by the identified pospiviroids and PepMV on the tomato seed pathway. Prior to their implementation, incursions of some of these pests occurred, but the pests were contained and eradicated from Australia. Since the implementation in 2013 of mandatory testing for these pests, no further incursions have been recorded.

Collectively, this biosecurity system and the specific regulatory actions taken provide an appropriate level of assurance that the identified pospiviroids and PepMV are not present in Australia.

Status of Potato spindle tuber viroid

A stakeholder asked for more information about the department's work to regulate *Potato spindle tuber viroid* (PSTVd).

The department is still evaluating whether PSTVd should be a regulated pest. This evaluation is being undertaken as a process separate to the finalisation of this pest risk analysis (PRA), in part because policy for PSTVd covers a broader range of hosts than tomato seed. Until this process is completed, PSTVd will continue to be regulated at the Australian border.

Wild tomato species

A stakeholder was concerned that the content of the PRA provided in the draft report did not clearly convey how the PRA assessed the potential presence of the pests in seeds of wild tomato species.

Risk assessments are not given in this report for wild tomato species (*Solanum chilense*, *S. chmielewskii*, *S. parviflorum*, *S. peruvianum* and *S. pimpinellifolium*) because there is insufficient evidence for the identified pospiviroids or *Pepino mosaic virus* being associated with the seeds of these species. The scope of this PRA is limited to the identified quarantine pests associated with cultivated tomato (*Solanum lycopersicum*).

Issue 2: Pest risk assessment

Pathway association

Stakeholders suggested that the draft PRA did not provide sufficient evidence that tomato seed imports are a pathway for the identified pests.

The department re-examined the evidence of association of the identified pospiviroids and PepMV with the tomato seed pathway (Chapter 3, Appendices A and B). It was concluded that the evidence of association with this pathway was sufficient for PepMV, *Columnea latent viroid* (CLVd), *Pepper chat fruit viroid* (PCFVd), *Tomato apical stunt viroid* (TASVd) and *Tomato chlorotic dwarf viroid* (TCDVd). However, it was considered that there was insufficient evidence for *Tomato planta macho viroid* (TPMVd) being associated with this pathway. This viroid has not been intercepted during extensive testing of imported tomato seed by Australian laboratories, and no reports were found of it being intercepted internationally, nor was other clear evidence found of the viroid being associated with traded tomato seeds. This final report has been amended accordingly. The decision to exclude TPMVd from the PRA may be reviewed if new evidence for seed association emerges.

Information derived from the International Seed Federation (ISF) Regulated Pest List Database was cited by a stakeholder in support of a claim that the draft report had not proven that tomato seed was a pathway for the identified pospiviroids and PepMV. This database does not comprehensively consider the available evidence for association of a pest with the seed pathway or provide reasoned conclusions. Additionally, the database does not provide pest risk assessments consistent with international standards. Furthermore, the department does not necessarily accept some claims made in the database about association of pathogens with seeds or their capacity for seed transmission.

Risk ratings

Stakeholders queried the justification for the risk ratings for entry and establishment of the identified pests in the PRA.

The department conducted the pest risk analysis in accordance with the International Plant Protection Convention (IPPC) and the International Standards for Phytosanitary Measures (FAO 2011, 2016a), and after considering the available evidence, decided to maintain the risk ratings for entry and establishment. These ratings were supported by evidence of association with traded tomato seed and evidence of seed transmission (Table 3.1; Chapter 3 and Appendices A and B).

The department also re-evaluated the evidence of damage to crops and the risk ratings for some potential consequences of pospiviroid infections were moderated from E to D. Although the risk estimates for CLVd, PCFVd, TASVd and TCDVd changed to Low with this moderation, the resultant unrestricted risk estimates did not achieve the ALOP for Australia.

Prevalence of infected lots

Stakeholders were concerned that estimates of the prevalence of infected tomato seed lots were inaccurate and suggested that data from seed lots tested in other countries should be included.

The department estimated the prevalence of pospiviroid-infected seed lots using an appropriate method. Data from interceptions of pospiviroid-infected seed presented in the draft report have been updated in this final report with data from 2008 to 2016. References are provided to analyses of this Australian data recently published in the scientific literature (Constable et al. 2019; Dall et al. 2019).

The inclusion of data from seed lots tested in other countries when estimating the prevalence of infected seed lots was considered inappropriate by the department, as complete datasets for such seed lots were not available. Although it is known that tomato seed lots contaminated with pospiviroid-infected seeds have been detected in other countries, the actual frequencies of detection are unknown.

Issue 3: Seed testing

Seed sample size

Stakeholders suggested that a sample of 3,000 seeds is sufficient to test tomato seed lots for the pathogens, and that the proposal to increase the sample that will be tested for PepMV is not based on an identified risk.

The size of a seed sample taken for testing is a critical factor that affects the effectiveness of tests. Seed lots contaminated with pospiviroid-infected seeds have been detected by Australian laboratories many times using samples of 20,000 seeds (Constable et al. 2019). Empirical evidence from this testing shows that the fraction of viroid-infected seeds in commercially prepared and traded seed lots is often very small (Constable et al. 2019; Dall et al. 2019) (Appendix C). Using samples of 20,000 seeds or more is necessary to detect such small fractions of infected seeds. Testing a sample of 20,000 seeds from a large seed lot of 100,000 seeds or more, provides a level of confidence of 99% of detecting the presence of contamination at a rate at or above 0.02% (i.e. 2 seeds in 10,000).

Based on the weight of available evidence, the department considers the increase in sample size to 20,000 seeds for testing for PepMV is justified. This evidence includes the number of detections of PepMV in crops by other countries and in traded seed lots by Australia and other countries, and the evidence of seed transmission and global transport of the virus (Chapter 3, Appendix A).

Testing or coated treated seed

A stakeholder asked that the department allow testing of treated or coated seeds.

If PCR tests are required, the department recommends that the seed sample be drawn prior to any seed treatment being applied. This is consistent with ISPM 38 in which it is acknowledged that seed treatments may adversely affect detection methods by physically or chemically inhibiting them or influencing their sensitivity (FAO 2017a). The department notes that other testing protocols also recognise the potential for adverse impacts of seed treatment and affirm that their tests are not validated for seed treated with protective chemicals or biological substances (ISF 2017b; ISHI-Veg 2020).

Bioassays

A stakeholder suggested that ELISA and PCR tests are inadequate to determine whether a seed lot carries a viable pathogen, and that bioassays should be undertaken to confirm whether a seed lot contains infectious agents.

Bioassays are not recommended by the department for routine diagnostic work on seeds for planting. Generally, tests that detect proteins or nucleic acids, such as ELISA and PCR, are required to detect pathogens in seed. Bioassays, including tests where indicator plants are inoculated, are often insensitive and unreliable (Legrand 2015).

PCR testing protocols

Stakeholders queried the validation of laboratory tests by Australia.

Substantial evidence has been published on the performance of the protocols used to detect the identified pospiviroids in tomato seeds by Australian laboratories. The published evidence shows the protocols to be sensitive, specific and robust (Constable et al. 2019; Constable et al. 2017) (Appendix C). Australian laboratories have also obtained sufficient evidence on the performance of the PCR protocols used to detect PepMV to justify continuing their use.

Validation of protocols used by overseas laboratories to detect the identified pospiviroids and PepMV are the responsibility of those laboratories and the NPPOs of those jurisdictions.

Issue 4: Heat treatment for PepMV

A stakeholder suggested a dry heat treatment of 72°C for 72 hours instead of 80°C for 72 hours, as proposed in the draft report as a treatment option for PepMV.

The department reconsidered the research data presented by Ling (2010) and concluded the data on treatment at 72°C for 72 hours did not provide sufficient evidence of efficacy as a phytosanitary measure. The data for 72°C were derived from 12 replicates of 250 seeds, whereas 48 replicates were used at 80°C. Data were also presented on treatments at both temperatures for 24 and 48 hours; two positive infections were observed when seeds were treated at 80°C for 24 hours and one positive infection was detected when seeds were treated at 80°C for 48 hours. In contrast, only one positive infection was observed from seeds treated at 72°C for 24 hours. Given the differences in the numbers of replicates at the two temperatures, these results of positive infections cast doubt on the negative results from treatments at 72°C,

and further support the need for a higher temperature and longer duration to deactivate PepMV. The department also noted that Ling (2010) stated that some virus deactivation results observed in other trials could be due to an 'uneven distribution of PepMV-infested seed in a replicate or a variation in the inoculation [of the bioassay]'.

The final report retains 80°C for 72 hours as the recommended treatment option for PepMV. It is acknowledged that this treatment was reported to reduce germination rate by 2% to 4%, and noted that it may be an appropriate option only under some circumstances.

Issue 5: Alternative measures

Chemical treatment for PepMV

Stakeholders suggested chemical treatments as potential alternative measures to mitigate the risk posed by PepMV.

The department considered the available information about chemical treatments intended to remove PepMV from tomato seeds, and concluded that there is insufficient evidence of the efficacy of the proposed treatments. For example, Córdoba-Sellés et al. (2007) reported treatment of tomato seeds carrying PepMV with a solution of 10% tri-sodium phosphate, following which the researchers did not detect transmission of the virus from seeds to seedlings. However, only 100 seeds were used in the experiment with this treatment. The department concluded that this research did not provide sufficient evidence that the treatment would be effective for large seed lots, as too few seeds had been tested and there were too few experimental replicates.

Field inspections and pest free areas

Stakeholders suggested that field inspections and pest free areas (PFAs) be accepted as alternative phytosanitary measures to replace testing for the pathogens.

Visual inspections of seed production crops (field inspection) may be an appropriate phytosanitary measure to detect some arthropod pests when they are easily recognised, or to detect other pests that produce characteristic visible symptoms during their life cycle. In contrast, in many cases it is impossible to identify plant pathogens from symptoms on crop plants. For this reason, the department does not accept field inspection as a phytosanitary measure for seed-borne pathogens. Furthermore, field inspection was previously found not to be an effective measure for PSTVd and it was withdrawn in 2012 (Chapter 1.2.3).

NPPOs that propose to use pest free areas as a measure for managing risks posed by the quarantine pests identified in this report should provide the department with an appropriate submission following the requirements outlined in Chapter 4.4 of the final report.

Good Seed and Plant Practices (GSPP)

A stakeholder proposed use of a combination of seed testing and the GSPP system as an alternative phytosanitary measure.

The department is supportive and agrees in principle to broader consideration of systems approach frameworks as potential options for the seeds for sowing pathway. The department is

supporting, along with industry and other NPPOs, the development of an annex to ISPM 38, which will consider this issue. The department notes that the GSPP system is likely to be considered in the development of this annex, and that the annex is expected to provide further guidance and a framework for consideration of commercial practices as components of a systems approach.

The GSPP system was specifically designed to manage *Clavibacter michiganensis* spp. *michiganensis* in tomato seed and plant production. The department acknowledges that this system provides elements likely to be required in a broader systems approach framework. However, further research is required to determine if the GSPP system will adequately mitigate the risks from the broad range of quarantine pests associated with the tomato seed pathway to a level that achieves the ALOP for Australia.

Glossary

Term or abbreviation	Definition
Additional declaration	A statement that is required by an importing country to be entered on a phytosanitary certificate and which provides specific additional information on a consignment in relation to regulated pests (FAO 2019b).
Appropriate level of protection (ALOP)	The level of protection deemed appropriate by the Member establishing a sanitary or phytosanitary measure to protect human, animal or plant life or health within its territory (WTO 1995).
Appropriate level of protection (ALOP) for Australia	The <i>Biosecurity Act 2015</i> defines the appropriate level of protection (or ALOP) for Australia as a high level of sanitary and phytosanitary protection aimed at reducing biosecurity risks to very low, but not to zero.
Area	An officially defined country, part of a country or all or parts of several countries (FAO 2019b).
Australian territory	Australian territory as referenced in the Biosecurity Act 2015 refers to Australia, Christmas Island and Cocos (Keeling) Islands.
Biosecurity	The prevention of the entry, establishment or spread of unwanted pests and infectious disease agents to protect human, animal or plant health or life, and the environment.
Biosecurity Australia	An agency of the Australian government responsible for biosecurity regulation that was subsumed into the department in 2012.
Biosecurity measures	The <i>Biosecurity Act 2015</i> defines biosecurity measures as measures to manage any of the following: biosecurity risk, the risk of contagion of a listed human disease, the risk of listed human diseases entering, emerging, establishing themselves or spreading in Australian territory, and biosecurity emergencies and human biosecurity emergencies.
Biosecurity import risk analysis (BIRA)	The <i>Biosecurity Act 2015</i> defines a BIRA as an evaluation of the level of biosecurity risk associated with particular goods, or a particular class of goods, that may be imported, or proposed to be imported, into Australian territory, including, if necessary, the identification of conditions that must be met to manage the level of biosecurity risk associated with the goods, or the class of goods, to a level that achieves the ALOP for Australia. The risk analysis process is regulated under legislation.
Biosecurity risk	The <i>Biosecurity Act 2015</i> refers to biosecurity risk as the likelihood of a disease or pest entering, establishing or spreading in Australian territory, and the potential for the disease or pest causing harm to human, animal or plant health, the environment, economic or community activities.
Consignment	A quantity of plants, plant products or other articles being moved from one country to another and covered, when required, by a single phytosanitary certificate (a consignment may be composed of one or more commodities or lots) (FAO 2019b).
Control (of a pest)	Suppression, containment or eradication of a pest population (FAO 2019b).
The department	The Australian Government Department of Agriculture, Water and the Environment.
ELISA	A laboratory test using the Enzyme-linked immunosorbent assay method.
Endangered area	An area where ecological factors favour the establishment of a pest whose presence in the area will result in economically important loss (FAO 2019b).
Endemic	Belonging to, native to, or prevalent in a particular geography, area or environment.
Entry (of a pest)	Movement of a pest into an area where it is not yet present, or present but not widely distributed and being officially controlled (FAO 2019b).
Equivalence (of phytosanitary terms)	The situation where, for a specified pest, different phytosanitary measures achieve a contracting party's appropriate level of protection (FAO 2019b).
Establishment (of a pest)	Perpetuation, for the foreseeable future, of a pest within an area after entry (FAO 2019b).

Term or abbreviation	Definition
Fresh	Living; not dried, deep-frozen or otherwise conserved (FAO 2019b).
Fumigation	A method of pest control that completely fills an area with gaseous pesticides to suffocate or poison the pests within.
Genus	A taxonomic category ranking below a family and above a species and generally consisting of a group of species exhibiting similar characteristics. In taxonomic nomenclature the genus name is used, either alone or followed by a Latin adjective or epithet, to form the name of a species.
Goods	The <i>Biosecurity Act 2015</i> defines goods as an animal, a plant (whether moveable or not), a sample or specimen of a disease agent, a pest, mail or any other article, substance or thing (including, but not limited to, any kind of moveable property).
Host	An organism that harbours a parasite, mutual partner, or commensal partner, typically providing nourishment and shelter.
Host range	Species capable, under natural conditions, of sustaining a specific pest or other organism (FAO 2019b).
IGB	Inspector General Biosecurity
IIGB	Interim Inspector General Biosecurity
Import permit	Official document authorising importation of a commodity in accordance with specified phytosanitary import requirements (FAO 2019b).
Incursion	An isolated population of a pest recently detected in an area, not known to be established, but expected to survive for the immediate future (FAO 2019b).
Infection	The internal 'endophytic' colonisation of a plant, or plant organ, and is generally associated with the development of disease symptoms as the integrity of cells and/or biological processes are disrupted.
Infestation (of a commodity)	Presence in a commodity of a living pest of the plant or plant product concerned. Infestation includes infection (FAO 2019b).
Inspection	Official visual examination of plants, plant products or other regulated articles to determine if pests are present or to determine compliance with phytosanitary regulations (FAO 2019b).
Intended use	Declared purpose for which plants, plant products, or other regulated articles are imported, produced or used (FAO 2019b).
Interception (of a pest)	The detection of a pest during inspection or testing of an imported consignment (FAO 2019b).
International Plant Protection Convention (IPPC)	The IPPC is an international plant health agreement, established in 1952, that aims to protect cultivated and wild plants by preventing the introduction and spread of pests. The IPPC provides an international framework for plant protection that includes developing International Standards for Phytosanitary Measures (ISPMs) for safeguarding plant resources.
International Standard for Phytosanitary Measures (ISPM)	An international standard adopted by the Conference of the Food and Agriculture Organization, the Interim Commission on Phytosanitary Measures or the Commission on Phytosanitary Measures, established under the IPPC (FAO 2019b).
Introduction (of a pest)	The entry of a pest resulting in its establishment (FAO 2019b).
ISTA	International Seed Testing Association
Lot	A number of units of a single commodity, identified by its homogeneity of composition, origin etc., forming part of a consignment (FAO 2019b). Within this report a 'lot' refers to a quantity of seed of a single variety, harvested from a single production site during a season and packed at one time.
National Plant Protection Organization	Official service established by a government to discharge the functions specified by the IPPC (FAO 2019b).
Non-regulated risk analysis	Refers to the process for conducting a risk analysis that is not regulated under legislation (DAWR 2016).

Term or abbreviation	Definition
Official control	The active enforcement of mandatory phytosanitary regulations and the application of mandatory phytosanitary procedures with the objective of eradication or containment of quarantine pests or for the management of regulated non-quarantine pests (FAO 2019b).
Outbreak	A recently detected pest population, including an incursion, or a sudden significant increase of an established pest population in an area (FAO 2019b).
Pathogen	A biological agent that can cause disease to its host.
Pathway	Any means that allows the entry or spread of a pest (FAO 2019b).
Pest	Any species, strain or biotype of plant, animal, or pathogenic agent injurious to plants or plant products (FAO 2019b).
Pest categorisation	The process for determining whether a pest has or has not the characteristics of a quarantine pest or those of a regulated non-quarantine pest (FAO 2019b).
Pest free area (PFA)	An area in which a specific pest does not occur as demonstrated by scientific evidence and in which, where appropriate, this condition is being officially maintained (FAO 2019b).
Pest free place of production	Place of production in which a specific pest does not occur as demonstrated by scientific evidence and in which, where appropriate, this condition is being officially maintained for a defined period (FAO 2019b).
Pest free production site	A defined portion of a place of production in which a specific pest does not occur as demonstrated by scientific evidence and in which, where appropriate, this condition is being officially maintained for a defined period and that is managed as a separate unit in the same way as a pest free place of production (FAO 2019b).
Pest risk analysis (PRA)	The process of evaluating biological or other scientific and economic evidence to determine whether an organism is a pest, whether it should be regulated, and the strength of any phytosanitary measures to be taken against it (FAO 2019b).
Pest risk assessment (for quarantine pests)	Evaluation of the probability of the introduction and spread of a pest and of the magnitude of the associated potential economic consequences (FAO 2019b).
Pest risk assessment (for regulated non-quarantine pests)	Evaluation of the probability that a pest in plants for planting affects the intended use of those plants with an economically unacceptable impact (FAO 2019b).
Pest risk management (for quarantine pests)	Evaluation and selection of options to reduce the risk of introduction and spread of a pest (FAO 2019b).
Pest risk management (for regulated non-quarantine pests)	Evaluation and selection of options to reduce the risk that a pest in plants for planting causes an economically unacceptable impact on the intended use of those plants (FAO 2019b).
Pest status (in an area)	Presence or absence, at the present time, of a pest in an area, including where appropriate its distribution, as officially determined using expert judgement on the basis of current and historical pest records and other information (FAO 2019b).
Phytosanitary certificate	An official paper document or its official electronic equivalent, consistent with the model of certificates of the IPPC, attesting that a consignment meets phytosanitary import requirements (FAO 2019b).
Phytosanitary certification	Use of phytosanitary procedures leading to the issue of a phytosanitary certificate (FAO 2019b).
Phytosanitary measure	Phytosanitary relates to the health of plants. Any legislation, regulation or official procedure having the purpose to prevent the introduction and/or spread of quarantine pests, or to limit the economic impact of regulated non-quarantine pests (FAO 2019b). In this risk analysis the term 'phytosanitary measure' and 'risk management measure' may be used interchangeably.
Phytosanitary procedure	Any official method for implementing phytosanitary measures including the performance of inspections, tests, surveillance or treatments in connection with regulated pests (FAO 2019b).

Term or abbreviation	Definition
Phytosanitary regulation	Official rule to prevent the introduction and/or spread of quarantine pests, or to limit the economic impact of regulated non-quarantine pests, including establishment of procedures for phytosanitary certification (FAO 2019b).
PRA area	Area in relation to which a pest risk analysis is conducted (FAO 2019b).
Production site	In this report, a production site is a continuous planting of tomato plants treated as a single unit for pest management purposes. If a growing area is subdivided into one or more units for pest management purposes, then each unit is a production site. If the growing area is not subdivided, then it is also the production site.
Quarantine	Official confinement of regulated articles for observation and research or for further inspection, testing or treatment (FAO 2019b).
Quarantine pest	A pest of potential economic importance to the area endangered thereby and not yet present there, or present but not widely distributed and being officially controlled (FAO 2019b).
Regulated article	Any plant, plant product, storage place, packaging, conveyance, container, soil and any other organism, object or material capable of harbouring or spreading pests, deemed to require phytosanitary measures, particularly where international transportation is involved (FAO 2019b).
Regulated non-quarantine pest	A non-quarantine pest whose presence in plants for planting affects the intended use of those plants with an economically unacceptable impact and which is therefore regulated within the territory of the importing contracting party (FAO 2019b).
Restricted risk	Restricted risk is the risk estimate when risk management measures are applied.
Risk management measure	Conditions that must be met to manage the level of biosecurity risk associated with the goods or the class of goods, to a level that achieves the ALOP for Australia. In this risk analysis, the term 'risk management measure' and 'phytosanitary measure may be used interchangeably.
RT-PCR	A laboratory test using the reverse transcription polymerase chain reaction method.
Spread (of a pest)	Expansion of the geographical distribution of a pest within an area (FAO 2019b).
SPS Agreement	WTO Agreement on the Application of Sanitary and Phytosanitary Measures.
Stakeholders	Government agencies, individuals, community or industry groups or organizations, whether in Australia or overseas, including the proponent/applicant for a specific proposal, who have an interest in the policy issues.
Surveillance	An official process which collects and records data on pest occurrence or absence by surveying, monitoring or other procedures (FAO 2019b).
Systems approach(es)	The integration of different risk management measures, at least two of which act independently, and which cumulatively achieve the appropriate level of protection against regulated pests.
Treatment	Official procedure for the killing, inactivation or removal of pests, or for rendering pests infertile or for devitalisation (FAO 2019b).
Unrestricted risk	Unrestricted risk estimates apply in the absence of risk management measures.
Vector	An organism that does not cause disease itself, but which causes infection by conveying pathogens from one host to another.
Ware potato	Potatoes and potato crops grown for consumption rather than propagation from seed.
WTO	World Trade Organisation

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