National Diagnostic Protocol

Meadow Spittlebug (*Philaenus spumarius*)

NDP 54 V1

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Purpose

National Diagnostic Protocols (NDPs) are diagnostic protocols for the unambiguous taxonomic identification of plant pests. NDPs:

- are a verified information resource for plant health diagnosticians
- are consistent with ISPM No. 27 – Diagnostic Protocols for Regulated Pests
- provide a nationally consistent approach to the identification of plant pests enabling transparency when comparing diagnostic results between laboratories; and,
- are endorsed by regulatory jurisdictions for use (either within their own facilities or when commissioning from others) in a pest incursion.

Where an International Plant Protection Convention (IPPC) diagnostic protocol exists it should be used in preference to NDPs, unless it is shown that the NDP has improved procedures for Australian conditions. NDPs may contain additional information to aid diagnosis. IPPC protocols are available on the IPPC website:

https://www.ippc.int/core-activities/standards-setting/ispms

Process

NDPs are facilitated and endorsed by the Subcommittee on Plant Health Diagnostics (SPHD). SPHD reports to Plant Health Committee and is Australia's peak technical and policy forum for plant health diagnostics.

NDPs are developed and endorsed according to Reference Standards developed and maintained by SPHD. Current Reference Standards are available at https://www.plantbiosecuritydiagnostics.net.au/resources/?category=national-diagnostic-protocols

NDPs are living documents. They are updated every 5 years or before this time if required (i.e. when new techniques become available).

Document status

This version of the National Diagnostic Protocol (NDP) for Meadow Spittlebug (*Philaenus spumarius*) is current as at the date contained in the version control box below.

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Further information

Inquiries regarding technical matters relating to this protocol should be sent to: sphd@aff.gov.au

Endorsement and publication details
The NDP was endorsed by the Subcommittee on Plant Health Diagnostic (SPHD) in 2025.
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Contents

1 INTRODUCTION

Xylem-feeding Hemiptera (true bugs) can be vectors of the plant bacterium *Xylella fastidiosa. Xylella fastidiosa* can utilize over 359 host plant species (CABI 2020b) and is the causative agent of many plant diseases including Pierce's disease of grapevine, bacterial leaf scorch, oleander leaf scorch, coffee leaf scorch, mulberry leaf scorch, alfalfa dwarf, phony peach disease, and citrus variegated chlorosis (Hopkins 1988; Hopkins & Purcell 2002; Almeida & Nunney 2015). This diagnostic protocol treats the most prominent *Xylella* vector within the family Aphrophoridae, the Meadow Spittlebug, *Philaenus spumarius* (L.).

Philaenus spumarius is polyphagous, with a host range of over 1000 plant species (Yurtsever 2000; Thompson et al. 2023; Appendix 8.1). Nitrogen-fixing plants are preferred (Thompson 1994), with nymphs favouring herbaceous Asteraceae and Fabaceae (Bodino et al. 2019). Although the feeding damage of *P. spumarius* is typically minor, except in certain crops (e.g., strawberries and alfalfa - Weaver & King 1954; Zajac & Wilson 1984), serious plant damage is caused through vectoring *X. fastidiosa* (Cornara et al. 2016, 2017a, b, 2018). *Philaenus spumarius* is the primary vector of all *X. fastidiosa* subspecies (i.e. *pauca*, *fastidiosa* and *multiplex*), affecting a range of plants; indeed, individuals can harbour multiple *X. fastidiosa* subspecies at once (Cruaud et al. 2018). The Meadow Spittlebug has been identified as the dominant insect contributing to the spread of *X. fastidiosa* across Europe (Cornara et al. 2019), which is having detrimental impacts on olives (Saponari et al. 2014), oleander and periwinkle (Cornara et al. 2017a). When first detected in olive groves, Italy spent €13.5 million in six months attempting to contain *Xylella* (Stokstad 2015). Schneider et al. (2020) has predicted an economic loss of billions of dollars over the next 50 years in Italy alone caused by *Xylella*. In the United States, Pierce's disease in grapevines costs California approximately $104.4 million per annum (Tumber et al. 2014). Although transmission is predominantly attributed to the Glassy-winged sharpshooter (*Homalodisca vitripennis*) in the USA (Hopkins & Purcell 2002), *P. spumarius* is also capable of transmitting *Xylella* in grapevines (Cornara et al. 2016). Given the wide host range of both *P. spumarius* and *X. fastidiosa*, there are likely many cryptic insect-vector transmissions.

Numerous resources treat *P. spumarius*, but none separates it from Australian species of Aphrophoridae. Information on Australian Aphrophoridae is sparse, and the most recent taxonomic review does not consider exotic genera (Liang & Fletcher 2003). This diagnostic protocol collates information on *P. spumarius*, in an easy-to-use diagnostic guide. It provides keys to distinguish the native Aphrophoridae of the Australian region from exotic genera that are known *Xylella* vectors and describes the male genitalia of *P. spumarius* to separate this species from others.

2 TAXONOMIC INFORMATION

This protocol provides the diagnostic capabilities to separate *P. spumarius* (Aphrophoridae) from other Auchenorrhyncha known to occur within Australia and surrounding regions, including other families of spittlebug (Hemiptera: Cercopoidea). Higher taxonomy follows Cryan and Svenson (2010), while tribal and generic taxonomy follows Hamilton (2012, 2013, 2014).

Taxonomic Position

Philaenus spumarius (Linnaeus, 1758)

Phylum:	Arthropoda
Class:	Insecta
Order:	Hemiptera
Suborder:	Auchenorrhyncha
Superfamily:	Cercopoidea
Family:	Aphrophoridae
Subfamily:	Aphrophorinae Amyot & Serville, 1843
Tribe:	Philaenini Metcalf, 1955
Genus:	*Philaenus* Stål, 1864
Species:	*Philaenus spumarius* (Linnaeus, 1758)

Synonyms

Aphrophora spumaria (L.)
Aphrophora vittata (Fabricius)
Cicada spumarius (L.)
Cercopis spumaria (L.)
Philaenus graminis (DeGeer)
Philaenus leucophthalmus (L.)
Philaenus lineatus (Fabricius)
Philaenus xanthocephalus (Schrank)
Ptyelus lineatus (Fabricius)
Ptyelus vittatus (Fabricius)

Common names

Meadow Spittlebug
Common Meadow Spittlebug
Meadow Froghopper
Cuckoo-Spit Insect

3 DETECTION

Philaenus spumarius is widely polyphagous (Appendix 8.1), thus it has the potential to enter Australia on a very wide range of plant species. Furthermore, if *P. spumarius* enters Australia undetected, it is likely capable of utilising native plants and weeds given evidence from other countries where it has extended its range (New Zealand: Larivière et al. 2010). *Philaenus spumarius* has been intercepted at Australian borders on cut flowers and foliage (Department of Agriculture, Water and the Environment 2021). In terms of surveillance and detection for *P. spumarius* post-border, particular sentinel plants near points of entry, such as shipping ports, warehouses and airports, are an efficient means of intercepting *P. spumarius*, and detecting *Xylella* if they do enter the country (Mansfield et al. 2019). Lavender is an ideal candidate for sentinel plantings for multiple reasons (Larivière et al. 2010; Metin & Karaca 2017; Broadmeadow et al. 2019; Ganassi et al. 2020), and is not known to host native Australian Cercopoidea, including the highly polyphagous *Bathyllus albicinctus*. However, the use of sentinel plants is only effective in early detection if regular surveillance is conducted, making this option potentially non-viable in Australia which has dozens of entry ports.

In Australia it is likely that *P. spumarius* would occur as nymphs or adults throughout spring to late autumn in temperate and cool regions. This is inferred from the biology of *P. spumarius* in New Zealand, which is the only recorded locality of the spittlebug in the Southern Hemisphere (CABI 2020a), having been introduced to this region circa 1960 (Hamilton & Morales 1992). In New Zealand, entomologists have noted that the meadow spittlebug is present as nymphs or adults from October to March, which is spring, summer and autumn in the Southern Hemisphere (Hamilton & Morales 1992; Larivière et al. 2010; Sandanayaka et al. 2017). Thus, bubble nests of the species are likely to be obvious from spring through to early summer in cool and temperate regions of Australia.

Philaenus spumarius is sensitive to environmental conditions; high humidity coupled with temperatures of 15.6 ˚C are optimal (Weaver & King 1954). Chmiel and Wilson (1979) demonstrated that *P. spumarius* nymphal temperature thresholds for development and survival were between 2.8 °C to 26.7 °C, while Morente et al. (2018a) had success at maintaining a laboratory population of *P. spumarius* through varying the temperature between monthly means of 5.4 °C (February) to 26.77 °C (July) and maintaining humidity close of 100%. At lower humidity or greater temperature variations, egg hatch viability decreases, egg oviposition is reduced, and mortality of adults and nymphs increases, as they are prone to desiccation during drier, hotter conditions (Weaver & King 1954). For example, in Turkey adult bugs disappear early in summer due to the dry conditions (Yurtsever 2000) but return with rain suggesting that they have moved to a more suitable microhabitat such as tree canopy during drier conditions (e.g., Chartois et al. 2023). A similar trend would likely occur in temperate regions of Australia. A warming climate in southern California has been implicated in the northward shift of *P. spumarius* to cooler regions (Karban & Strauss 2004). These environmental tolerances suggest that *P. spumarius* may not survive the warmer conditions in Australian subtropical to tropical regions, nor the drier regions in the semi-arid or arid interior of Australia. This hypothetical, thermal distribution boundary is supported by *P. spumarius* being restricted to cool, higher altitudes in Hawaii, the only tropical occurrence of the spittlebug (Yurtsever 2000). In drier countries, *P. spumarius* is known to occur in the more mesic habitats, for example within Hyrcanian forests and Steppes in Iran (Mozaffarian & Wilson 2015).

3.1 Symptoms

Philaenus spumarius can utilise much of the host plant, with the possible exceptions of seed and root material (Table 1). The life cycle of *P. spumarius* first involves 'packets' of up to 20 eggs laid along stems or plant litter in a frothy cement, followed by five instars of nymphs on stems and leaves in bubble nests. Adults then emerge after the 5th instar moult, feeding on stems, leaves, new shoots, fruits and flowers (Yurtsever 2000; Table 2).

Table 1. Pattern of damage and activity of *Philaenus spumarius* on host plants.

Plant part	**Attacked**	**Insect Life Stage**
Root	No	Not reported
Stem	Yes	Eggs, nymphs, adults
Leaves	Yes	Eggs, nymphs, adults
Growing tips	Yes	Nymphs, adults
Buds	Yes	Nymphs, adults
Flowers	No	Not reported, but general feeding on plant can cause reduced flowering
Fruit and Seeds	Yes	Nymphs, adults on fruit

Table 2. Life cycle of *Philaenus spumarius* on host plants.

Life stage	**Site of Activity**
Eggs	'packets' of up to 20 eggs laid along stems and dry plant material close to or on the ground
Nymphs (5 instars)	Feed within a white spittle mass on leaves and stems, often at stem/shoot nodes
Adults	Feed on stems, leaves, fruit, flowers (usually at base) and new shoots

Nymphs of *P. spumarius* are encased in a spittle-like foam or froth, hence the common name "spittlebug". *Philaenus spumarius* infestations are usually detected by the presence of nymphs in these bubble nests along the stems, at leaf nodes and on fresh shoots (Fig. 1). Media in the United Kingdom has engaged the public to record spittlebugs (noting bubble nests as an indicator of presence) as a form of surveillance for *Xylella* (BBC 2019). The nymphs and bubble nests of *P. spumarius* cannot be distinguished from the bubble nests of native Australian Aphrophoridae spittlebugs (Fig. 1), therefore the detection of bubble nests on a potential host plant in Australia should prompt further investigation through molecular work or rearing of nymphs to adults. It should also be noted that bubble nests are likely to be seasonal. The main symptom of severe infestation on host plants by *P. spumarius* is plant dwarfism (Horsfield 1978; Yurtsever 2000; Hambäck 2001; CABI 2020a) and loss of plant biomass (Carson & Root 1999). However, more extensive and obvious damage is attributed to infection of plants by *Xylella* than by

the feeding damage caused directly by the spittlebugs. Symptoms of *Xylella* infestation vary depending on the plant species affected, but in many cases include severe dieback (see Fig. 2). A range of symptoms displayed by different host plant species is provided by Vos et al. (2019).

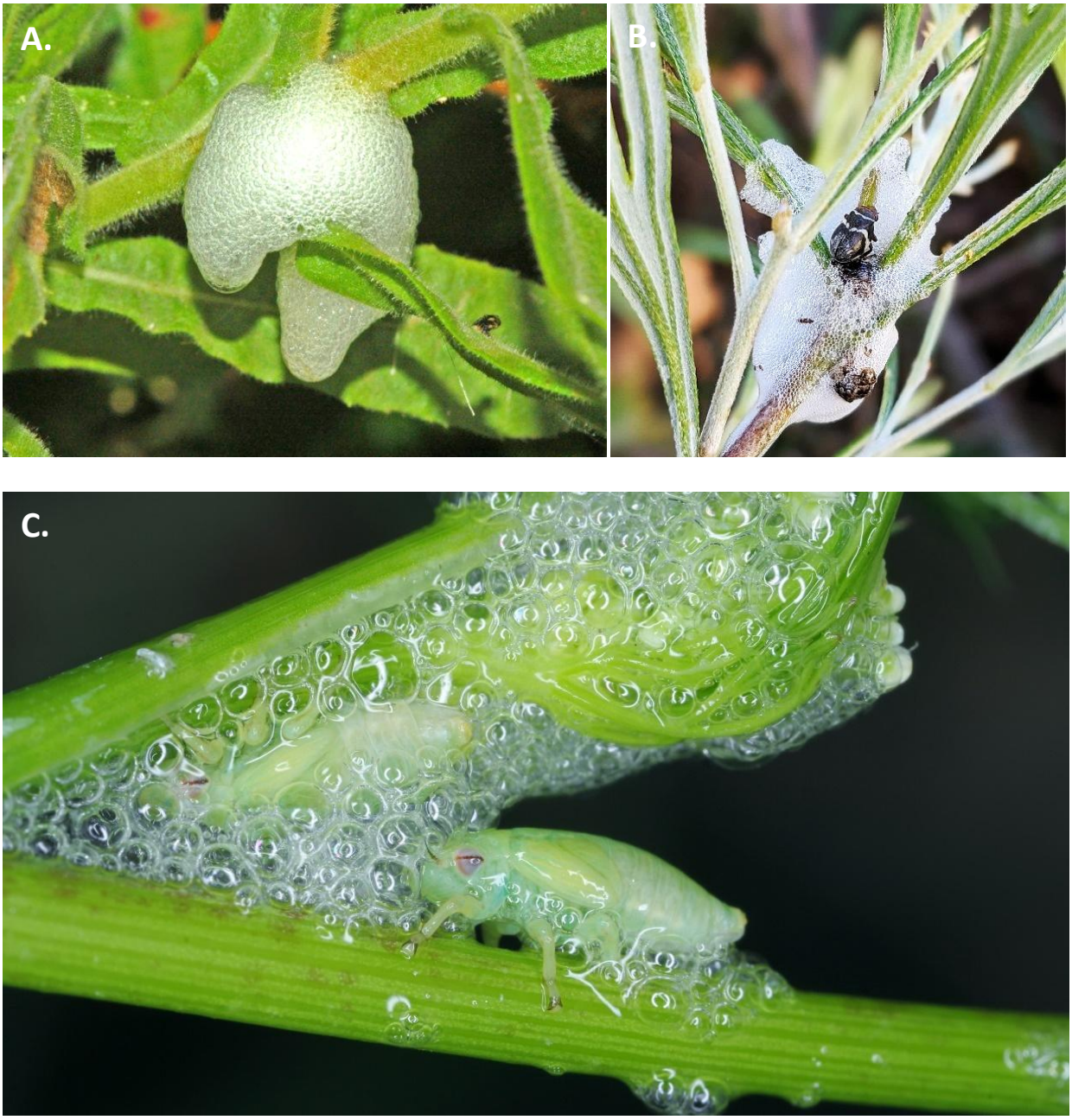

Figure 1. A. *Philaenus spumarius* nymph bubble nest © Hectonichus 2010 https://commons.wikimedia.org/w/index.php?curid=15061231. **B.** the bubble nest of the common Australian species *Bathyllus albicinctus* (Erichson) [Hemiptera: Aphrophoridae] © Pia Scanlon, Western Australian Agricultural Authority, Perth, Australia 2020. **C.** *Philaenus spumarius* nymphs in bubble nest © Tomasz Klejdysz, shutterstock.com

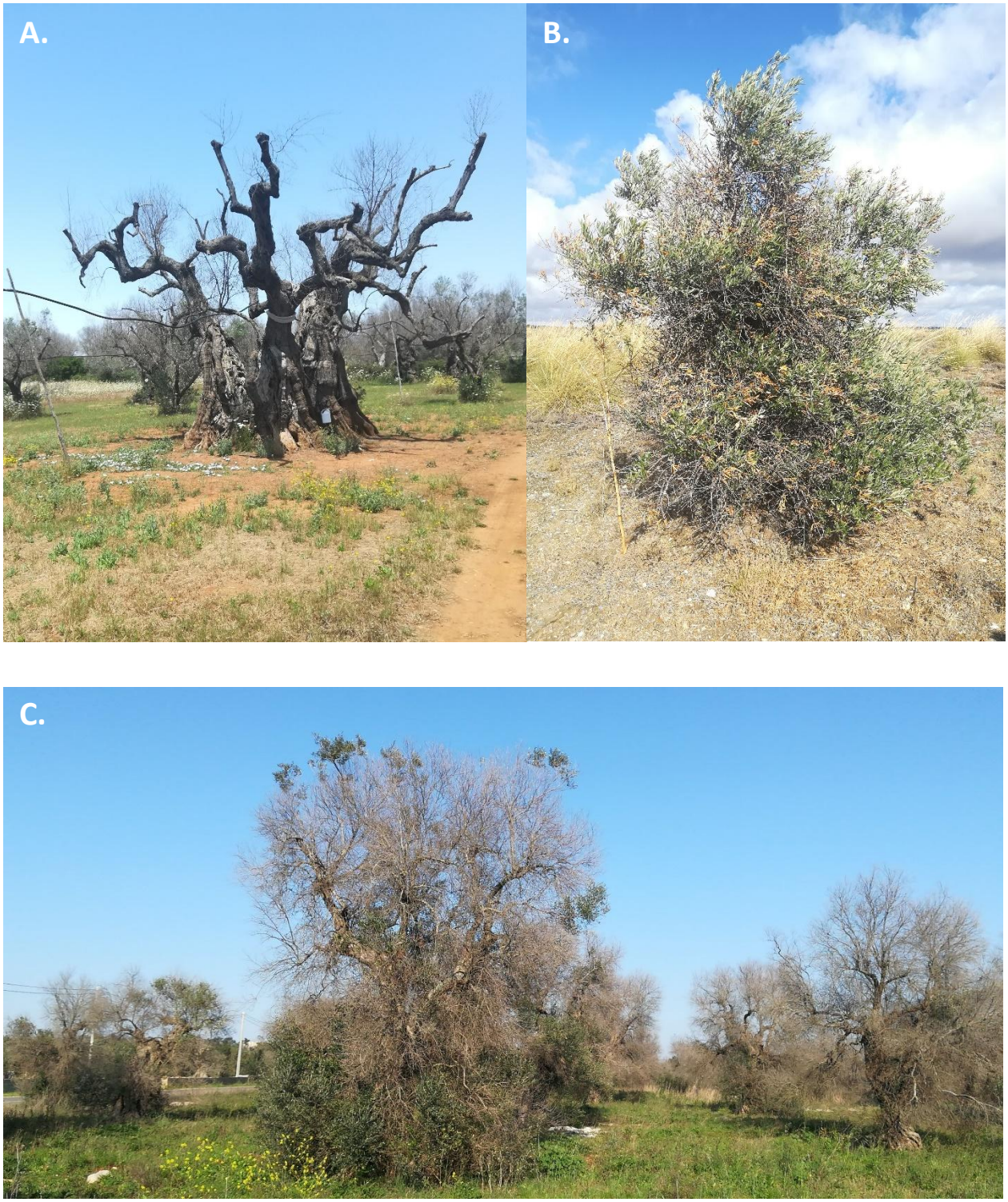

Figure 2. *Xylella* affected olive trees in **A.** Spain (top right) and **B., C.** Italy (top left, bottom). Adapted from Cornara et al. 2018, used with permission April 2020.

3.2 Sampling

3.2.1 Methods

Eggs of *P. spumarius* are difficult to detect, but if found are best stored in 95-100% ethanol for molecular work. Hand collecting nymphs is the simplest method to sample instars as their mobility is poor. Nymphs can be removed from plants and their foam bubble nests using forceps or a fine paintbrush. They should be collected into 95-100% ethanol in a vial to facilitate molecular determination. The spittle can be washed from the nymphs first, using a weak detergent solution as required.

Adults are not encased in such froth and are highly mobile, moving freely on plants by either walking, or jumping. When they are disturbed, spittlebugs will often jump a significant distance (Burrows et al. 2008) and can be difficult to catch. Sweeping a net over vegetation (particularly low vegetation: Cruaud et al. 2018; Morente et al. 2018b) or beating plants over a net / beating sheet is often the most efficient method to capture adult spittlebugs. As adults are very active and can be difficult to capture even while in the net, a pooter (or aspirator), either battery operated or manual, assists with collecting individuals from the beating or sweeping net. A comprehensive video tutorial on field collection is given by the European Food Safety Authority (2017).

The most effective and common passive surveillance method is employing yellow sticky traps, alongside sweep-netting (Markheiser et al. 2021; Dongiovanni et al. 2023). More passive, less commonly used, methods of capturing *P. spumarius* include Malaise traps (e.g., Koufakis et al. 2019) and light trapping using UV light or mercury vapour lamps. Common factors influencing the success rate of trap catches, particularly with light trapping, are humidity, temperature, wind, surrounding habitat and the mobility of the insect itself (Bowden 1982).

Adults should be collected alive and subsequently be killed either by freezing or with a small amount of ethyl acetate in a killing jar. Once dead, specimens can be pinned dry or glued onto cardboard using a water-soluble glue. If adults of the suborder Auchenorrhyncha are collected and stored in ethanol, they lose colour and the structural integrity of the exoskeleton, but they can still be identified. Both curation methods (ethanol and dry) are suitable for genitalia dissection. If facilities or resources to pin or card mount material are unavailable, then specimens can be stored dry in vials, along with paper towel to prevent damage. If numerous specimens have been collected, retaining at least one specimen in 95-100% ethanol will assist with molecular analysis. All specimens should be labelled with the date of collection, locality, the host plant, the collector, and, if possible, the origin of the material. Pinned material should have individual labels on acid-free card under the pinned insects (Fig. 3). Small insects, such as *P. spumarius,* are often staged on micropins in dense foam so as not to overly damage the specimen with the standard pins (Fig. 3).

Figure 3. Curated insects showing micro-pinned specimen of *Philaenus spumarius* from the New South Wales Department of Primary Industries and Regional Development Biosecurity Collections (ASCTHE027080). The labels are either written in Indian ink or printed.

Specimens in ethanol should have labels written in pencil or Indian ink, on acid free card, placed in plastic or glass leak-proof vials with the insect (refer to Löcker et al. (2018) for further details on curation). Workers should collect as many specimens as they can, as only males will provide a definitive morphological identification.

4 IDENTIFICATION

The Meadow Spittlebug *Philaenus spumarius* is a true bug (Hemiptera) within the spittlebug and froghopper superfamily Cercopoidea, suborder Auchenorrhyncha. It belongs to the family Aphrophoridae, which contains approximately 157 described genera and 990 species globally (Soulier-Perkins 2020). The genus *Philaenus* is European and western Asian in origin (Maryańska-Nadachowska et al. 2015) containing 46 species. *Philaenus spumarius* is the most widespread and common, found throughout the Palaearctic region from Japan and China to the United Kingdom, USA, Canada, northern Africa, New Zealand. There are no species of *Philaenus* represented in Australia (Fletcher 2009 and updates). Previous reports suggesting that *P. spumarius* is present in Australia are erroneous (e.g., Syrett et al. 1999; Memmott et al. 2000). No specimen with Australian locality data has been uncovered to date, and Dr Murray Fletcher, Australia's preeminent Auchennorrhyncha expert for the past 40 years confirms that he has no knowledge of the species being detected within Australian borders (M. Fletcher pers. comm. Jan 2023).

Figure 4. *Philaenus spumarius* collected in Kentucky, United States of America, showing **A.** lateral and **B.** dorsal views (New South Wales Department of Primary Industries and Regional Development Biosecurity Collection, specimen ASCTHE027696). © Pia Scanlon, Western Australian Agricultural Authority (Department of Primary Industries and Regional Development).

Adult *P. spumarius* are 5 - 7 mm in length and are generally grey, brown, or black in colouration (Figs 3, 4), although in rarer cases some specimens may appear to have orange, white or yellow colouration, while nymphs tend to be green (Fig. 1C). Adult *P. spumarius* have at least five distinct colour morphs ranging from solid brown, black or grey, through to varying combinations of stripes, patches and mottling (see Hamilton 2006; Fig. 5). Colour and pattern are not reliable features on which to base an identification. Reliable morphological identification of *P. spumarius* eggs and nymphs is not possible.

Figure 5. The variety of colour morphs of *Philaenus spumarius*. All images have had backgrounds and legs removed, and have been rotated to similar angles for comparative purposes. The following pertains to copyright of the original images (for full details and image sources, refer to Appendix 8.4): **A., F., J.** © Sandy Rae; **B.** © Pia Scanlon; **C.** © Laurence Livermore; **D., H., L., M.** © AfroBrazilian; **E.** © Walwyn; **G.** © gailhampshire; **I.** © Sanja565658; **K.** © Hectonichus; **N.** © Judy Gallagher; **O.** © Syrio.

4.1 Morphological identification

4.1.1 Preparation of adult specimens for morphological identification

Specimens of Aphrophoridae can be either stored in ethanol or dry pinned (Figs 3 and 4). For long-term storage, dry pinning is preferred, however this must occur relatively soon after collection to preserve the colour and shape of the cuticle of the specimen. For detailed accounts of curating leafhoppers and spittlebugs, see "Preservation and preparation of Auchenorrhyncha for identification" pages 24-28 in Löcker et al. (2018). Male genitalia must be dissected for a definitive identification of *P. spumarius.* Female specimens cannot be used to differentiate between species.

Equipment and chemicals required for dissection and mounting

Dissection:

- Hotplate or preferably dry block heater
- Dissecting/Stereo microscope
- Ethanol (~70%)

- 10% KOH (potassium hydroxide) solution
- Distilled water
- Fine dissection forceps, preferably Dumont jeweller's forceps #4
- Micropin mounted onto end of toothpick or similar
- Cavity glass block (embryo glass)
- Fine brush

Curation:

- Insect pins
- Micropins 0.015 mm diameter
- Fine dissection forceps, preferably Dumont jeweller's forceps #4
- Glycerol (to store genitalia in)
- Genitalia tubes (to store genitalia in)
- Acid-free card (for labelling)
- Indian Ink pens or pencils (for labelling)
- Dense foam for mounting micropinned specimens
- Pinning block
- Ethanol (~70%)

Dissection of male genitalia

Male genitalia examination provides definitive identification of *P. spumarius*. When male specimens have been curated in lower concentrations of ethanol (60-85%), specimens are flexible and can be examined *in situ*. The last segments of the abdomen and the genital capsule should be gently squeezed to reveal the male genitalia. Although this is a quick method of morphological examination and species identification, better clarity, and therefore more certainty, is provided through full genitalia dissection as outlined below.

Genitalia must first be dissected from the body of the specimen, either using forceps or small dissection scissors to remove the last segments of the abdomen prior to the genital capsule. The segment is then cleared using 10% KOH in a cavity glass block, over a heating block at approximately 50-60 ˚C. Care should be taken as KOH is corrosive and can cause burns if in contact with skin. Clearing allows easier viewing of the sclerotized male segments. The segment should be checked intermittently to ensure that it does not over-clear. Over-clearing is indicated by the removal of all colour, the structures become transparent, and identification is very difficult.

Once the segment is cleared to a satisfactory degree, wash in distilled water to remove all KOH. The segment is then placed in a genitalia tube with glycerol to keep the genitalia capsule malleable until required. The genitalia tube is placed under the remainder of the pinned specimen, with the pin piercing the rubber cap of the genitalia tube. If the specimen is curated in ethanol, then the genitalia can be placed within a microvial in ethanol, and within the larger vial containing the remainder of the *P. spumarius* individual.

4.1.2 Morphological Identification of spittlebugs

Philaenus spumarius has the following combination of morphological characters that differentiates it from all other Australian spittlebugs in the family Aphrophoridae. These characters may be used for quick, presumptive indications of identity, but any suspect specimens must be subsequently confirmed with molecular analysis and genitalia dissections.

- Adults approximately 5 - 7 mm in length

- Adult colour any of the following, or a combination: yellow, white, brown, grey, black, orange (e.g., Figure 5)
- Ocelli equidistant from each other and eyes (Figure 9d)
- Vertex shorter in length than pronotum (Figure 9d)
- Antennal ridge sulcate (a concave groove present) between two carinae (Figure 11d)
- Formula of distal (=apical) spines on the tibia and tarsi of the hind leg typically = 8:7:11 (tibia, 1st tarsal segment, 2nd tarsal segment). Note that aberrant specimens could vary with specimens observed with the following ranges 8-10:6-7:7-12 spines.

The native species of Aphrophoridae from Australia and surrounding regions including Christmas Is., Lord Howe Is., Norfolk Is., New Guinea, the Solomon Islands and New Zealand can be separated from *Philaenus* by differences in external morphology, such as the globular *Bathyllus,* the very large *Amarusa*, and the elongated vertex of *Philagra* and *Interocrea*. Other Australian and New Guinean genera such as *Clovia* and *Liorhina* feature bold and colourful patterns. Superficially, Australian *Anyllis* species are perhaps the most likely to be confused with *Philaenus*, as they are a similar size, shape and tan in colour. The single lateral spine on hind tibia of *Anyllis* and two spines on *Philaenus* however, easily separate these genera. Finally, the genus *Basilioterpa*, which is present on Lord Howe Island and islands of New Zealand, may also be confused with *Philaenus* based on colouration and spinal formations on the hind tibia. *Basilioterpa* has a single antennal carina and ocelli that are closer to each other than to the eyes, which separates it from *Philaenus*, which possesses double antennal carinae and ocelli equidistant to each other and to the eyes.

There are further exotic spittlebug vectors of *Xylella,* such as *Neophilaenus campestris,* which are more difficult to distinguish from *Philaenus*. The key provided here separates *Philaenus* from all other known genera. Given that there are 46 species of *Philaenus*, species identification of *P. spumarius* must involve male genitalia dissections. Molecular analysis may be used to refine species identity. This protocol provides diagrams and photographs of *P. spumarius* male genitalia and other characters, which should allow a field or laboratory technician to distinguish a specimen of *P. spumarius* from other species. However, secondary confirmation from a taxonomist or molecular specialist (as given in section 5 of this protocol) is recommended.

Key

This key distinguishes *Philaenus* species from the Auchenorrhyncha of Australia (and its territories), New Guinea and New Zealand (adapted from Hamilton 1980; Hamilton & Morales 1992; Liang & Fletcher 2003; Hamilton 2012). Some characters may be ambiguous (where possible these characters are highlighted in the key), therefore all identifications made using this key should be validated by experienced Auchenorrhyncha taxonomists who are familiar with the Australian fauna and using molecular methods to genus level (to discount the presence of other exotic taxa).

1a. Tegula present (Figure 6A).....not Cercopoidea (Fulgoroidea)

1b. Tegula absent (Figure 6B) 2

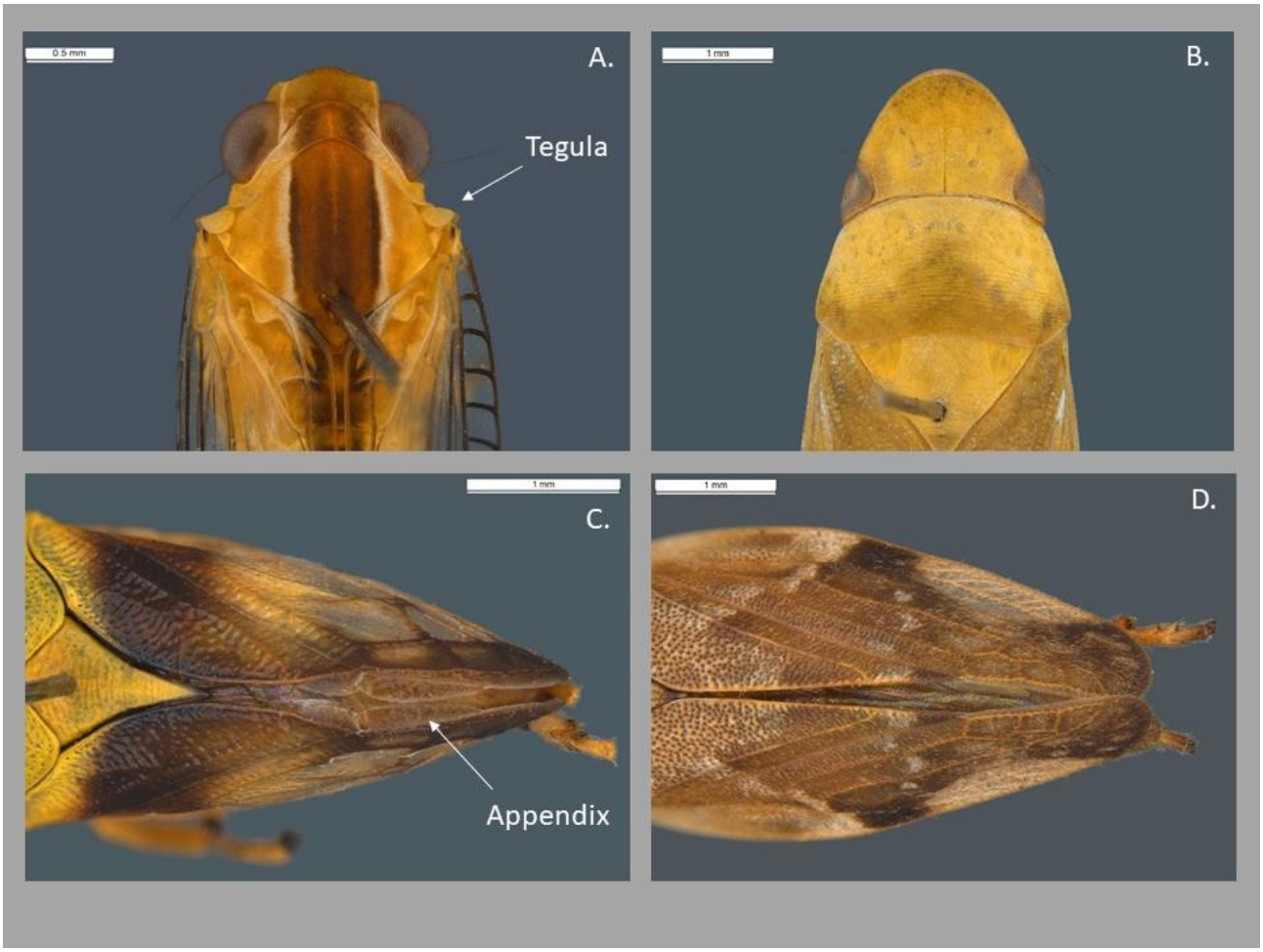

Figure 6. Differences between Membracoidea, Fulgoroidea and Cercopoidea. **A.** Tegula covering wing joint of *Gaetulia chrysopoides*, Tropiduchidae (Fulgoroidea), **B.** no tegula present, showing *Reuplemmeles hobartensis*, Cicadellidae (Membracoidea), **C.** appendix present on forewing of *Chaetophyes* sp., Machaerotidae (Cercopoidea), and **D.** large appendix lacking on *Anyllis leiala*, Aphroporidae (Cercopoidea). Plate adapted from photographs by © Pia Scanlon, Western Australian Agricultural Authority 2020

2a. Hind tibia with >2 lateral spines, tibia and tarsi not noticeably flared (Figure 7B)..... .....not Cercopoidea … (Cicadellidae, Cicadidae)

2b. Hind tibia with 1-2 lateral spines, distal end flared, tarsi flared (Figure 7A, C, D)…… ……Cercopoidea … 3

Figure 7. Hind legs of Auchenorrhyncha showing **A.** tibia with 1 lateral spine and double row of distal tibial spines (*Anyllis leiala*, Aphrophoridae), **B.** tibia and tarsi of Cicadellidae with many lateral tibial spines and narrow tarsi, **C.** tibia with 2 lateral spines and two rows of distal (apical) tibial spines (*Philaenus spumarius,* Aphrophoridae), and **D.** tibia with 1 lateral spine and single (indistinct) row of distal tibial spines (*Chaetophyes* sp., Machaerotidae). Plate adapted from photographs by © Pia Scanlon, Western Australian Agricultural Authority 2020.

3a. Obvious appendix on forewing (Figure 6C). Single or double row of spines on distal end of hind tibia (Figure 7D).....not Aphrophoridae (Machaerotidae)

3b. Very small or no appendix on forewing (Figure 6D). Double row of spines on distal end of hind tibia (Figure 7A)..... 4

4a. Posterior margin of pronotum straight or slightly sinuate (Figure 8A, B).....not Aphrophoridae (Cercopidae)

4b. Posterior margin of pronotum in distinct 'W' shape (Figure 8C, D).....Aphrophoridae 5

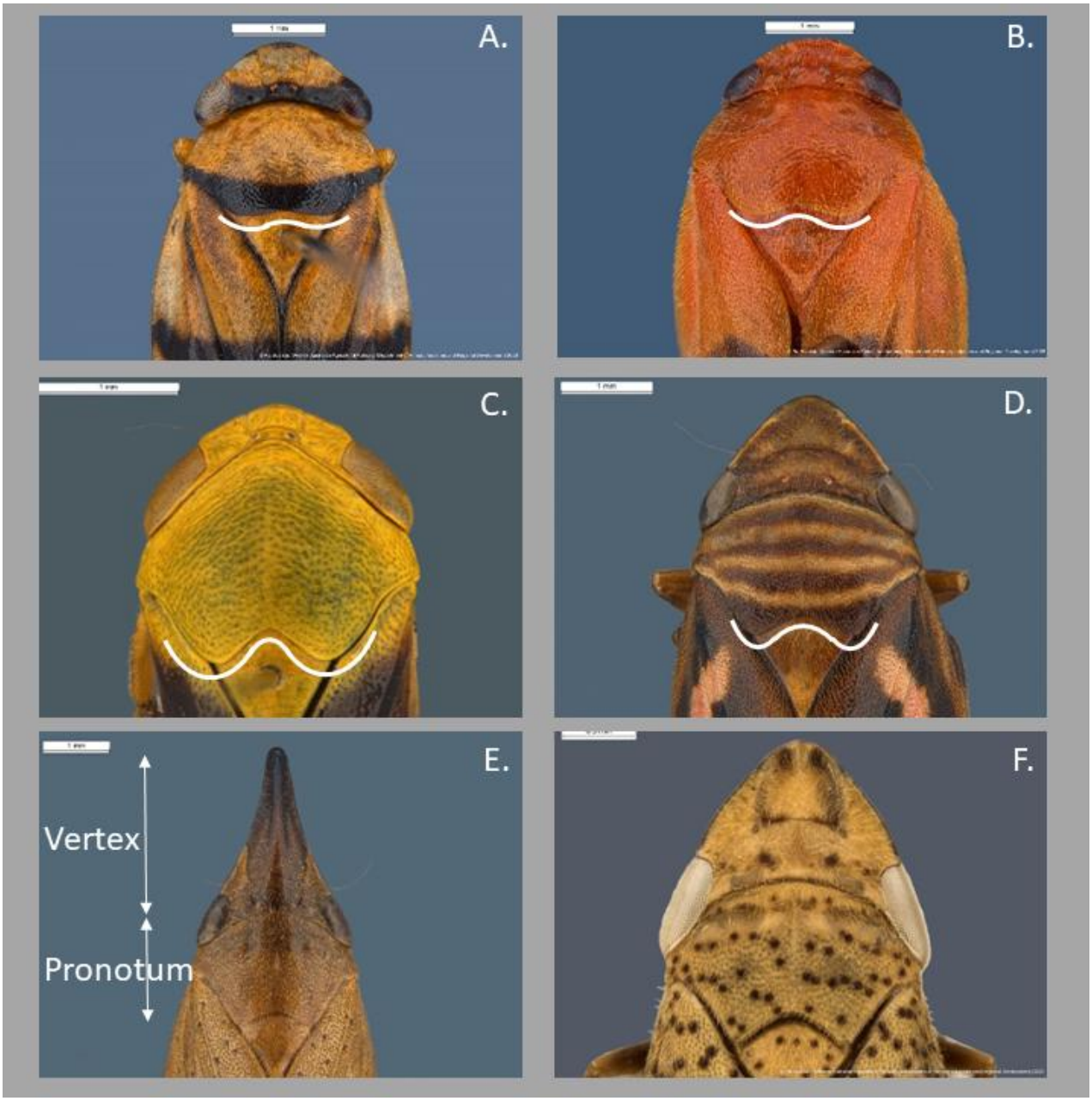

Figure 8. Head, vertex and pronotum of Cercopoidea showing hind margin of pronotum straight or sinuate (indicated by white line) for **A.** *Aufidus trifasciata*, Cercopidae, and **B.** *Euryaulax carnifex*, Cercopidae, and hind margin of pronotum in distinct 'W' shape for **C.** *Chaetophyes* sp., Machaerotidae, **D.** *Liorhina loxosema*, Aphrophoridae, **E.** *Philagra* sp., Aphrophoridae and **F.** *Interocrea* sp., Aphrophoridae. Also note the vertex versus pronotum length (as demonstrated in **E.**) for each species. Plate adapted from photographs by © Pia Scanlon, Western Australian Agricultural Authority 2020.

5a. Ocelli distinctly closer to each other than eyes (Figure 9A).....(not ***Philaenus***) … 6

5b. Ocelli distinctly closer to eyes than each other (Figure 9B).....(not ***Philaenus***) … 10

5c. Ocelli equidistance from each other and eyes (Figure 9D).....11

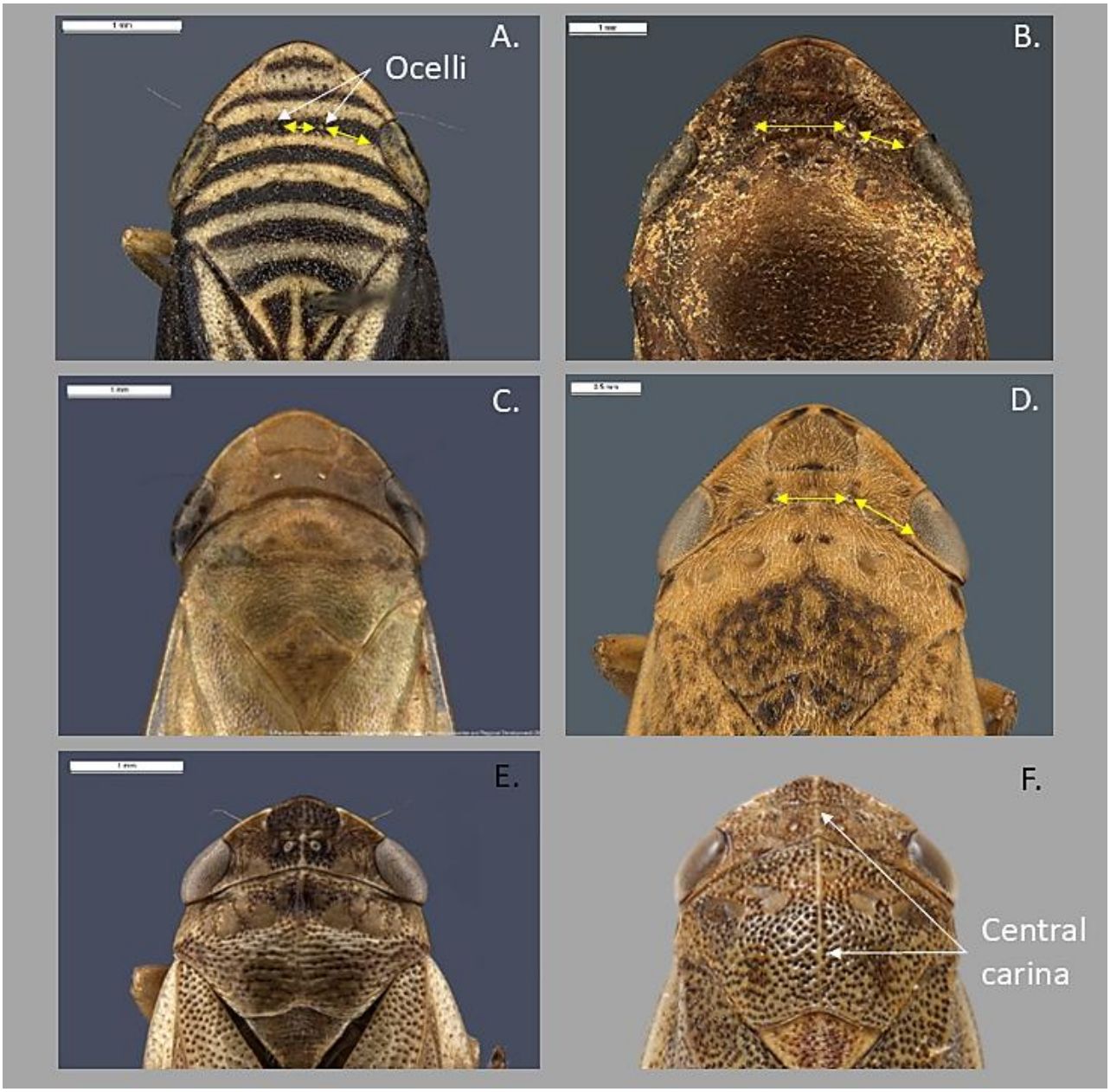

Figure 9. Head, vertex and pronotum of Aphrophoridae showing ocelli distance to each other and eyes. **A.** *Clovia eximia*, **B.** *Amarusa australis*, **C.** *Basilioterpa pallida*, **D.** *Philaenus spumarius*, **E.** *Anyllis spinostylus*, and **F.** *Aphrophora alni*. Plate adapted from photographs A., B., C., D. & E. by © Pia Scanlon, Western Australian Agricultural Authority 2020. Figure F. adapted from © Mozaffarian & Wilson 2015, used with permission Sept 2020.

6a. One lateral spine on hind tibia (Figure 7A).....***Anyllis*** (TRIBE Aphrophorini) Australia

6b. Two lateral spines on hind tibia (Figure 7C).....7

7a. Frons greatly expanded, bulbous. Insects rounded, coleopterous (Figure 10A).....***Bathyllus*** (TRIBE Cloviini) Australia, New Zealand and New Guinea

7b. Frons not greatly expanded, either flat or convex. Insects elongate (Figure 10B, C, D, E, F).....8

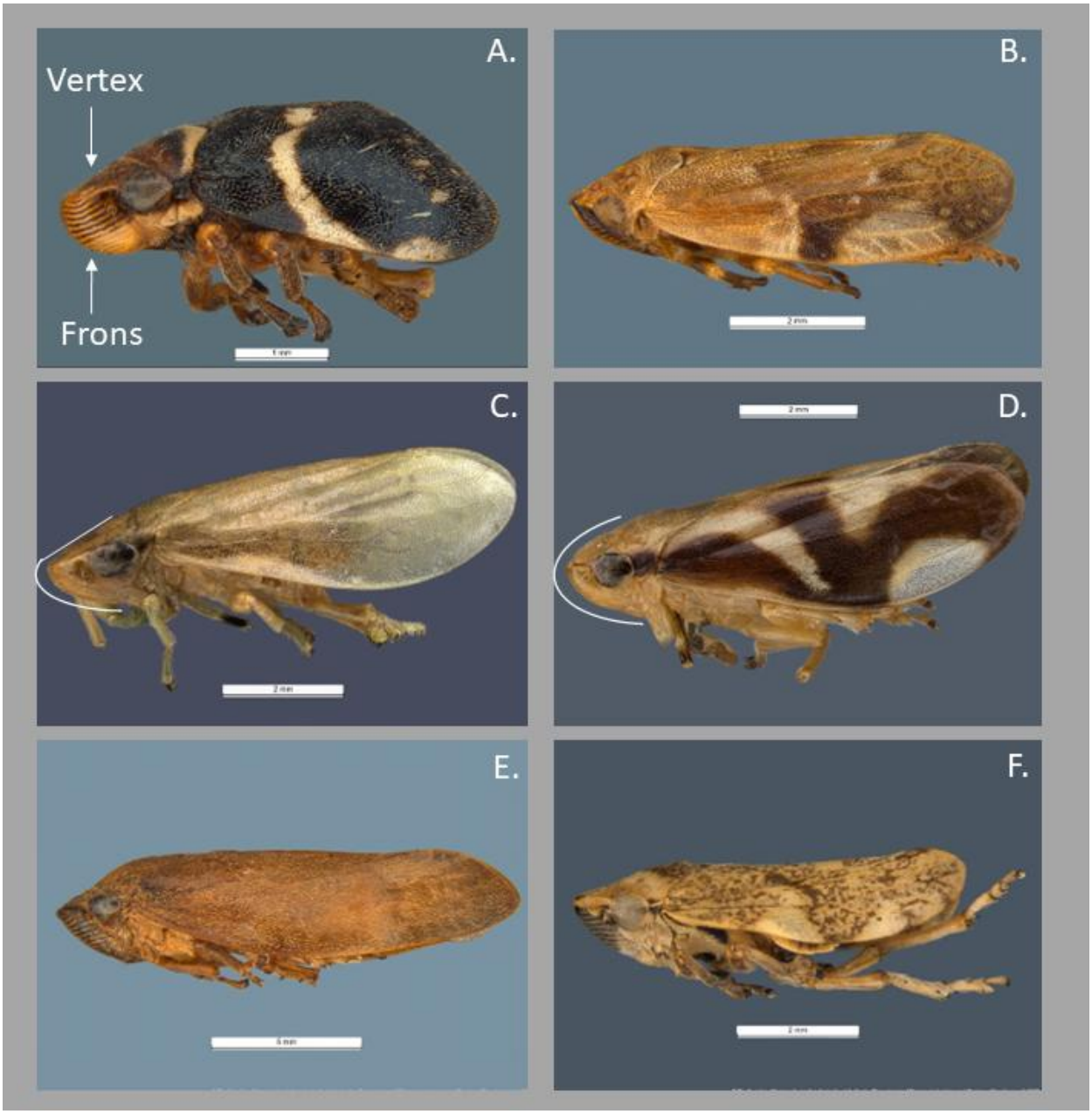

Figure 10. Lateral images of Aphrophoridae showing size and differences between frons and vertex. **A.** *Bathyllus albicinctus*, **B.** *Anyllis leiala*, **C.** *Basilioterpa pallida*, **D.** *Carystoterpa vagans*, **E.** *Amarusa australis*, and **F.** *Philaenus spumarius*. Plate adapted from photographs by © Pia Scanlon, Western Australian Agricultural Authority 2020.

8a. Frons and/or vertex with prominent stripe/s (Figures 7C, 9A). Spines on distal end of hind tibia ≤10.....***Clovia*** (TRIBE Cloviini) Christmas Island, Madagascar, PNG, West Papua, Bangladesh, Indonesia; or ***Cloviana*** (TRIBE Orthorhaphini) West Papua, PNG, Solomon Is.

8b. Frons and vertex without patterns (Figures 7A, 9C). Spines on distal end of hind tibia ≥10.....9

9a. Angle of frons from dorsal to ventral convex (strongly inflated), vertex slightly inflated, giving the appearance of a rounded head when viewed laterally (Figures 10D, 11A).....***Carystoterpa*** (Tribe Orthoraphini) New Zealand

9b. Frons slightly inflated and relatively flat, vertex sloping down to meet frons at a sharp angle when viewed laterally (Figures 10C, 11B).....***Basilioterpa*** (Tribe Philaenini) Australia (Lord Howe Island), New Zealand

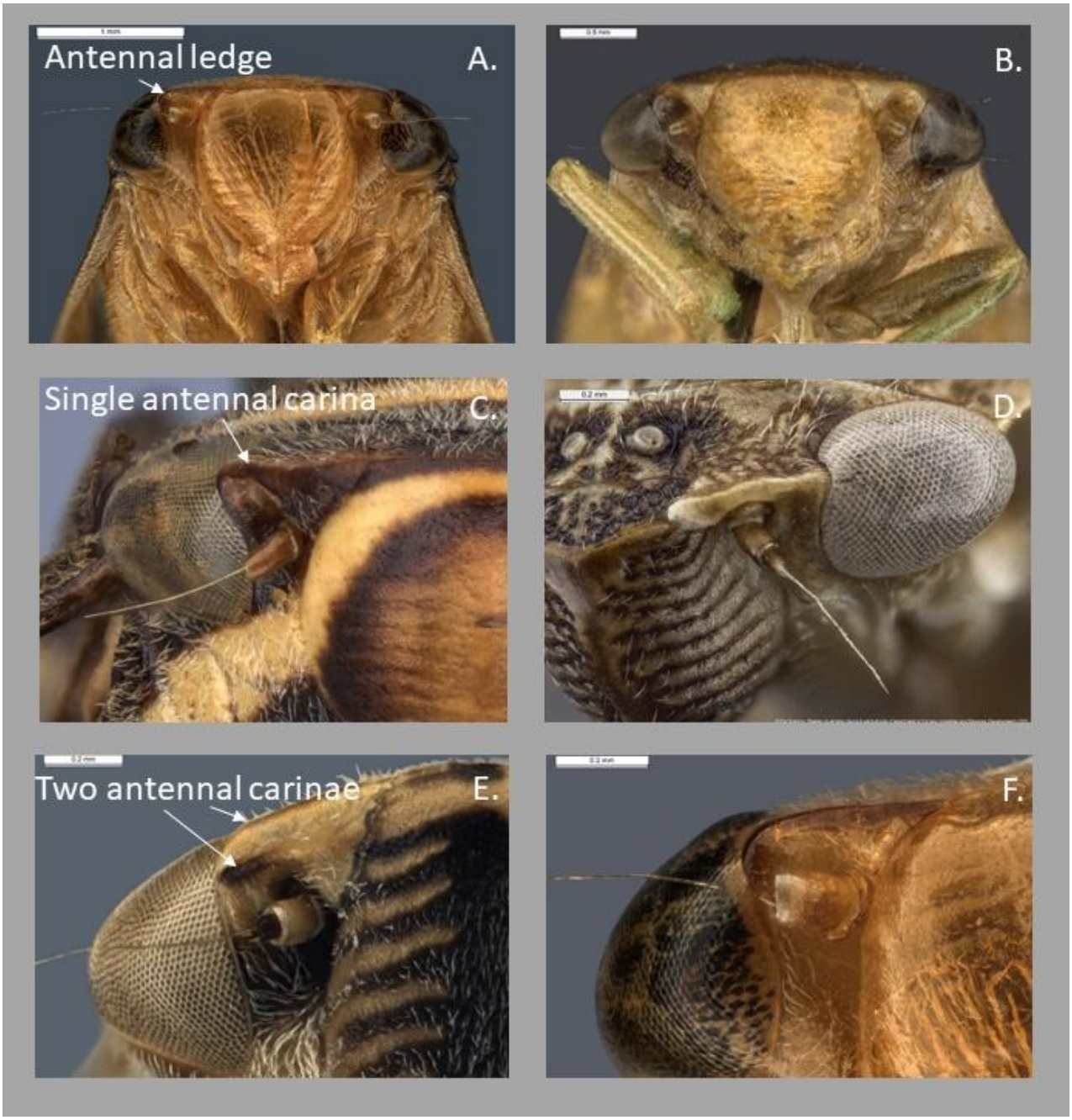

Figure 11. The face and antennal structures of Aphrophoridae. **A.** Frons and carinate antennal ledge of *Carystoterpa vagans*, **B.** frons and rounded antennal ledge of *Basilioterpa pallida*, **C.** single antennal carina of *Clovia borneensis*, and **D.** *Anyllis spinostylus*, **E.** Double antennal carinae of *Philaenus spumarius*

marked with grooves between carinae, and **F.** close up of antennal segments of *Carystoterpa vagans*. Plate adapted from photographs by © Pia Scanlon, Western Australian Agricultural Authority 2020.

10a. Large, length greater than 10 mm (Figure 10E)……..……..***Amarusa*** (Tribe Ptyelini) Australia, Indonesia, Papua New Guinea, Vanuatu, Solomon Islands, Philippines

10b. Smaller, less than 10 mm in length (Figure 10D).....Genera not present in Australia (e.g., ***Ptyelus, Plinia***)

11a. Vertex expanded apically, vertex as long as pronotum or longer (Figure 8E). Tegmen may have scattered indented pits (Figure 8E,F) .....(not ***Philaenus***) 12

11b. Vertex less than length of pronotum, although may appear similar in length, measurements must be made using a microscope (Appendix 8.3). Tegmen relatively smooth, lacking pits (Figure 8D)……..13

12a. Pronotum wider than head (including eyes) (Figure 8E)…….....***Philagra*** (Tribe Aphrophorini) China, Japan, Taiwan, Cambodia, Vietnam, Laos, India, Australia

12b. Pronotum narrower than head (including eyes) (Figure 8F).....***Interocrea*** (Tribe Orthoraphini) Australia, New Guinea, Solomon Islands, New Caledonia

13a. Third antennal segment retracted within second segment (e.g., Figure 11F)…….....***Liorhina*** (Tribe Orthoraphini) Australia, New Guinea, Solomon Islands, Philippines.

13b. Third antennal segment visible outside of second antennal segment, two antennal carinae (Figure 11E),.....(Genera not present in Australia) 14

14a. Central carinae on frons plate, vertex and/or pronotum (Figure 9E, F).....Genera not present in Australia (e.g., ***Aphrophora, Neophilaenus***)

14b. No central carinae on vertex or pronotum (Figure 9D), antennal ledge sulcate (concave groove exists) between carinae (Figure 11E), base of male anal tube (segment X) expanded laterally (Figure 12A).....***Philaenus*** (Tribe Philaenini)

Male genitalia

The above key separates the genus *Philaenus* from other genera. All *Philaenus* species are exotic to Australia, thus must be reported if detected. To determine species level identification, male genitalia must be examined in detail, and molecular methods could be utilised (see below). Note that current molecular methods cannot reliably separate all species, so results should be interpreted alongside morphology. Females, nymphs and freshly emerged males (that have not 'hardened' and coloured up) cannot provide as definitive an identification as mature males.

The key features of male *Philaenus* genitalia are an anal tube with the basal segment (also called segment X) having expanded lateral edges and an upright sturdy rod-like aedeagus with small hooks or spines on the terminus (Figure 12) (Archibald et al. 1979). *Philaenus spumarius* may be separated from mostother species of *Philaenus* by the presence of six obvious spines on the aedeagus, with the apical pair long and curved (Figure 12). The aedeagi of Australian species of Aphrophoridae are distinctly different from this shape (Appendix 8.2).

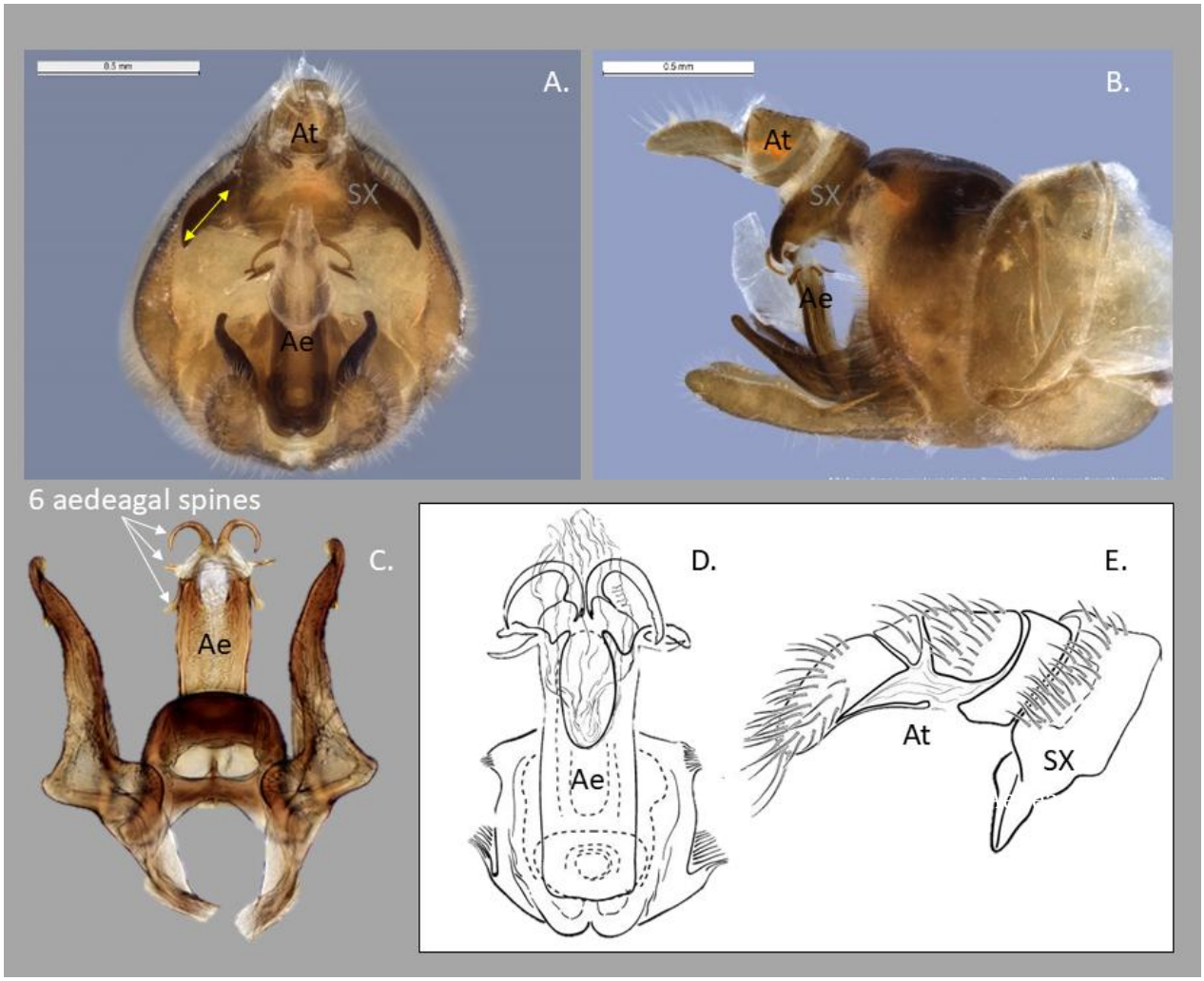

Figure 12. Male genitalia of *Philaenus spumarius.* **A.** image of cleared genital capsule posterior view (ASCTHE027384) with yellow arrow highlighting lateral extensions of anal tube, **B.** image of cleared genital capsule lateral view (ASCTHE027384), **C.** dissected male genitalia (posterior view), **D.** dorsal drawing of the male genitalia and **E.** lateral drawing of the anal tube. Key: Ae - aedeagus topped by six spines, At - anal tube, SX – Segment X with lateral expansions visible. Plate adapted from the following contributors: Figures A, B. © Pia Scanlon, Western Australian Agricultural Authority 2020; Figure C. © Janet Graham 2020 https://www.flickr.com/photos/149164524@N06/49515258862/in/photolist-2iruDnG-QP1aYE-WyLBBj-TC2wFJ-2j8nHGM-225m9S8-5dtmXW-26rTYyf-ro1tNF-2jr29zy-2jsJSHT-5dtmRs-puYh8r-5dtn3y-2jCtH15-XrHaea-JaADbJ-rpLusL-rpLusf-26VSXtP-2hd9AuU-2gjTzJr-2gjTydb-VViEL5-uFVLD7-ePWv6T-29fuHga-9U4Lf4-9U7GWh-f6v7cq-2j6Ytph-29bgF6j-2hM3oTA-2b2u7zQ-2irtuRK-8bcRM7-29fuH8p-2d76Fhc-2g2WvW2-Zmrvnh-2jcahU7-ouLgw1-26N2XeL-9Wugwu-5mJhS8-VSu1Fm-5Kmzfs-8kx1uq-2gJpSw1-2gJpawp; Figures D, E. © Boukhris Bouhachem et al. (2019).

Six spines on the aedeagus are also present in *P. tesselatus*, Melichar, 1899. This species was in 1959 reduced to a geographic subspecies of *P. spumarius,* with which it was synonymized in 1972, but later resurrected from synonymy based on differences in body size and male genitalia structure. In general, appendages of the male aedeagus are larger in *P. tesselatus* (Figs 13B, 13D-13F) than in *P. spumarius* (Figs 12A-12D), but both species show some variation in their aedeagal structures, which partly overlap. Seabra et al. (2021) detected some admixture (hybridisation) which suggests some level of recent or ongoing gene-flow between *P. spumarius* and *P. tesselatus*. Nevertheless, their results point towards these species being closely related but independent gene-pools (separate species), probably early in the speciation 'continuum'. Both species share the same mitochondrial COI haplotypes, making DNA barcoding with this gene ineffective. However, Seabra et al. (2021) found divergence in RAD sequencing markers.

Male genitalia of *P. maghresignus* (Figs 13J-13P), *P. ebursianus* (Figs 13V-13X) and *P. italosignus* (Fig. 13Q) show some resemblance with *P. spumarius* but have a reduced number of spines on the apex of the aedeagus. There are other species within *Philaenus* that may have similar male genitalia, however, there are no comprehensive keys published and availability of genitalia illustrations is limited.

Figure 13. *Philaenus* species. **A.-H.** *Philaenus tesselatus*: **A.** habitus dorsal, **B.-H.** male genitalia. **I.-P.** *Philaenus maghresignus*: **I.** habitus dorsal, **J.-P.** male genitalia. **Q.** *Philaenus italosignus* male genitalia. **R.**-**U.** *Philaenus spumarius*: male genitalia. **V.-X.** *Philaenus ebursianus* male genitalia. Plate adapted from the following contributors: Figures A.-P. © Boukhris Bouhachem et al. (2019); Figures Q.-R. © JC Streito, INRA CBGP in EPPO (2020); Figures S.-X. Mozaffarian & Wilson (2015).

Other features of P. spumarius

To confirm that the suspect individual is *P. spumarius*, the morphology of other body parts should also be examined.

Wings: The wings are not diagnostic as the venation of many Aphrophoridae are similar (Figure 14A, B). Note that the outer margin (top margin in Figure 14B) of the forewing is flat to broadly rounded, and not concave.

Legs: There are 8 distal spines on the hind tibia, arranged in two indistinct rows. There are 7 to 8 distal spines on the first tarsal segment and typically 11 distal spines on the second tarsal segment (Figure 14C).

Head: The vertex, head and pronotum ratios should be assessed, along with the placement of the ocelli, the appearance of the vertex plate and the antennal ledge. No median carinae are present on the vertex, pronotum or frons (Figures 9D, 14D). The ocelli are approximately equidistant between each other and the eyes (Figure 9D). The vertex length is approximately half that of the pronotum (Figure 9D). The antennal ledge features two antennal carinae with parallel grooves between them (Figure 11E). The shape and form of the frons and the angle that it meets the vertex should be compared to Figure 10F; the frons is not expanded and meets the vertex at an angle of 70-75˚.

Colour and pattern: should only be considered if it is bright (orange, red, yellow, green) and in contrast to previous descriptions of colour morphs (e.g., Figure 5). Colour and pattern should only receive secondary emphasis given the possibility of undescribed colour morphs.

Nymphs: consistently bright green through the different instars with darker antennae, and eyes red to pink, occasionally very dark (Figure 1). Dead specimens will change colour. As noted above however, colour is not considered a reliable identification tool and should be only used as a guide to possible species.

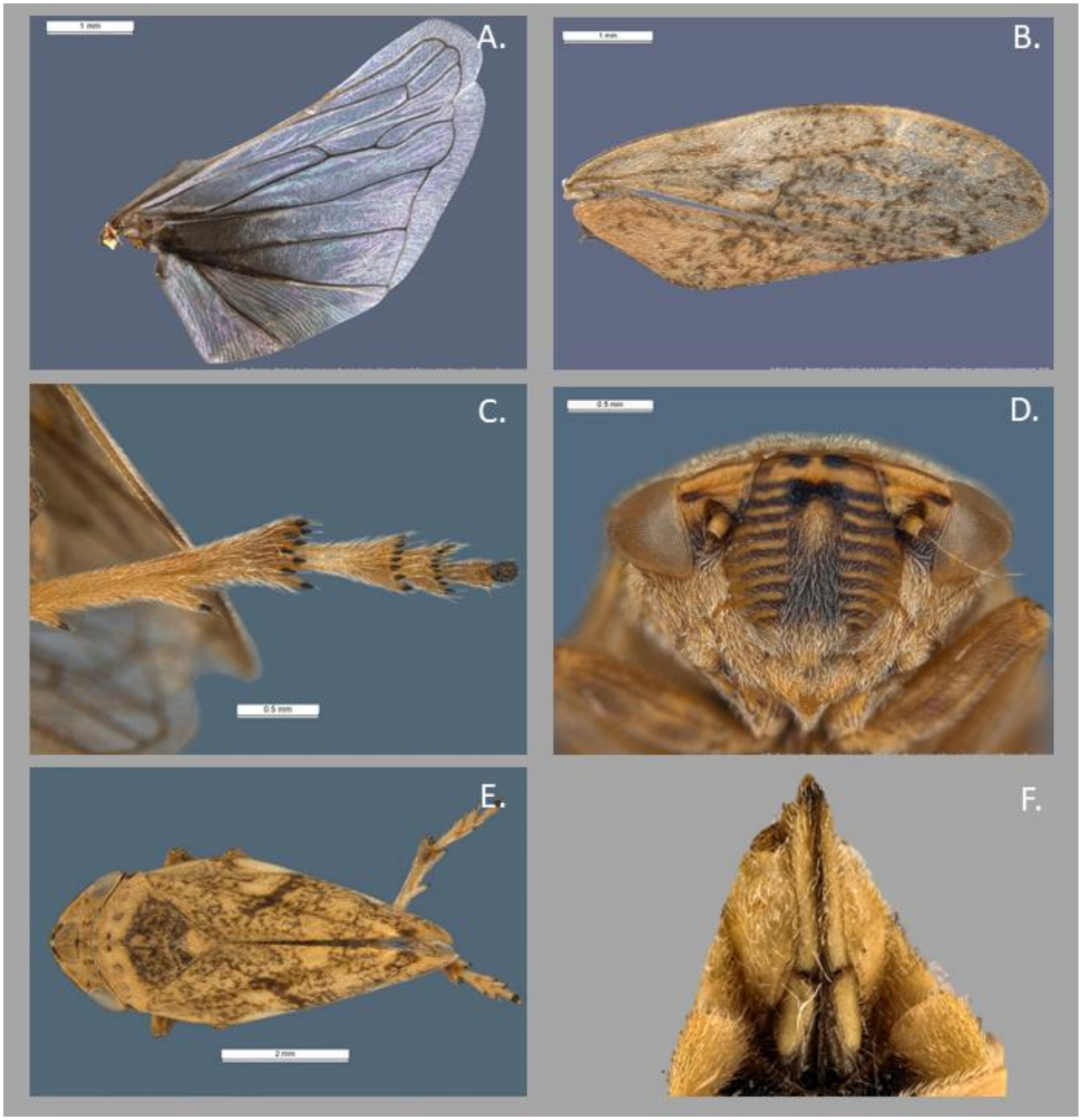

Figure 14. *Philaenus spumarius*, Aphrophoridae. **A.** hindwing (ASCTHE028060), **B.** forewing (ASCTHE027781), **C.** hind leg (ASCTHE027781), **D.** face (ASCTHE027696), **E.** dorsal habitus (ASCTHE027696), and **F.** female genitalia, ventral view (ASCTHE028060). Plate adapted from photographs by © Pia Scanlon, Western Australian Agricultural Authority 2020.

Reference material within Australia

Department of Primary Industries and Regional Development, Orange Agricultural Institute (ASCU), New South Wales:

- 1 male, 2 females, United States of America: Kentucky, Metcalfe Co., N of Center, Wooded area off Hwy 218. At U.V. light. 12.vi.1997. Carl Cook. Dry pinned, genitalia dissected ASCTHE027384 (♂), Dry pinned ASCTHE027385 (♀), Dry pinned ASCTHE027386 (♀)

- 2 females, United States of America: Kentucky, Hart Co., Green River, Stovall Park, Munfordville 29.vi.1991. Carl Cook. Dry pinned ASCTHE028059, ASCTHE028060
- 1 female, United States of America: Kentucky, Hart Co., Munfordville, Stovall Park, u.v. light at Green River, 26.vi.1994. C. Cook. Dry pinned ASCTHE027781
- 1 female, United States of America: Tennessee, Lewis Co., Gordonsburg 13.vii.1994. Carl Cook. Dry pinned ASCTHE027566
- 1 female, United States of America: Kentucky, Laurel Co., Lily, 03.vi.1989. Carl Cook. Dry pinned ASCTHE027696
- 1 female, United States of America: Kentucky, Mammoth Cave Nat. Pk. Dennison Ferry. 17.vii.2001. C. Cook & Judy Malone. MCNP ATBI Permit #0164. Dry pinned ASCTHE027263
- 1 female, United States of America: Kentucky, Green Cty, Legion Pk & Green River, Greensburg, Hwy #417. 25.v.2002, Carl Cook. Dry pinned ASCTHE027080
- 2 females, Austria: Styria, Siegersdorf near Herberstein, 47°12’34"N, 15°47’50”E, 515m coll. Thomas Friess, Georg Derbuch 13.viii.2018. Dry mounted on cardboard ASCT00237993, ASCT00237992
- 1 male, 2 females, Belgium, Liege province, Buetgenbach, 50°30’42"N, 6°10’00”E, coll. Jerome Constant & Linda Semeraro 8.ix.2021 Dry mounted on cardboard. ASCT00237986 (♂), ASCT00237984 (♀), ASCT00237985 (♀).

South Australian Museum (SAMA), South Australia:

- 1 female, United States of America: Palo Alto, Ca. 21 Nov. 1980 N.B. Tindale. {On separate label: For Gordon Gross} Dry pinned SAMA 20-18005

Department of Energy, Environment and Climate action (Victorian Agricultural Invertebrate Collection, VAIC), Victoria:

- 1 female, United States of America: Williamson St Pk. Yamhill Co. ORE, vi-9-1962, C.W. O’Brien. Dry pinned, VAIC number: 081239

Department of Primary Industries and Regional Development (DPIRD, Western Australia:

- 1 female, 2 males, Belgium: Louvain-La-Neave, 50.6862° N, 4.6062° E, 19-Aug-2019, J. Constant. In ethanol ICDB147108 (1♀), 147109 (1♂), 147110 (1♂)

4.2 Molecular identification

4.2.1 Preparation of specimens for molecular analysis

Specimens of *P. spumarius* designated for molecular analysis should be freshly captured and stored in ethanol at concentrations higher than 95%. Retaining specimens below 5 ˚C also improves the success of sequencing. Dried material can be viable for molecular work, but sequencing success depends on several factors such as how the material has been stored (i.e. whether it was in a solution previously and then dried), age of the specimen, and how the specimen was captured (e.g., water bath, chemical knockdown, sweeping, vacuum samples, etc).

4.2.2 Molecular identification of spittlebugs

General information

There have been little molecular advancements in the identification of native Australian species of Aphrophoridae. In contrast, *P. spumarius* has received much attention as an important economic pest species globally, particularly in its role as a vector of *Xylella*. Despite this, there have been numerous obstacles in defining an adequate molecular protocol, and future work is required to develop an accurate test that can separate related species.

There are 2,932 entries in GenBank for *P. spumarius*, including multiple whole genome records. Within the genus *Philaenus* there has been much splitting and synonymy, some of which has been based on molecular work (e.g. Maryańska-Nadachowska et al. 2011, 2013). It is noted that 185 CO1 DNA barcode compliant sequences of *P. spumarius* released at the BOLD online repository (accessed 20/02/2025) are consistent as a single cluster of related sequences (BIN AAB1850) available for query-based sequence identification of *P. spumarius* (BOLD Systems 2014). There is a paucity of reference sequences available for other *Philaenus* species.

It is noted that COI sequences have been previously assigned to incorrectly identified specimens because authors neglected to use morphological analysis of male genitalia for identification (Seabra et al. 2020). In particular, BOLD Process ID GBMH5383-09 (GenBank FJ516394) identified as *P. maghresignus* should be corrected to *P. spumarius*.

Conventional PCR methods

Refinements to molecular methods for identifying *P. spumarius* and related species are continually occurring. EPPO (2020) details a conventional PCR method, adapted from Lantero et al. (2018) to identify *P. spumarius*. This method does not distinguish between *P. spumarius and P. italosignus*. The authors also caution that it may not differentiate *P. spumarius and P. tesselatus.* The amplicon size for this method is 128bp which is likely insufficient for species identification and differentiation by conventional PCR.

Lantero et al. (2018) and Rodrigues et al. (2022) used conventional PCR techniques to detect *P. spumarius* DNA in predator gut content. However, neither method is shared here because this research did not sufficiently assess whether methods were species-specific to *P. spumarius*, particularly in relation to other species of *Philaenus*.

It is difficult to differentiate species of *Philaenus* molecularly. For example, it is claimed that mitochondrial COI alone cannot be used to differentiate between *P. spumarius* and *P. tesselatus,* and a comparison of other gene regions is required (Maryańska-Nadachowska et al. 2011; Seabra et al. 2020; EPPO, 2020). Although Rodrigues et al. (2022) suggested that there is a good likelihood of their method being species-specific based on "the Primer-BLAST results", much work remains to validate this statement. Identification of *Philaenus* at the species level may not be feasible by using small CO1 fragments alone (Seabra et al. 2010). Lester et al. (2020) suggests that other *Philaenus* species, and indeed other closely related genera (particularly *Cercopis*, *Neophilaenus*, *Aphrophora* and *Mesoptyelus*), should be analysed in further detail including producing reference sequences for other gene regions such as ITS2 and ELF to aid with future identification. Thus, currently there is no established and validated conventional PCR method to confidently identify *P. spumarius* molecularly and establishing such a procedure is beyond the scope of this diagnostic protocol.

Quantitative PCR methods

EPPO (2020) described a quantitative PCR protocol (developed by Lester et al., unpublished) targeted to the nuclear ribosomal internal transcribed spacer 2 (ITS2) region, to distinguish *P. spumarius* from several other *Philaenus* species and related genera. It is reproduced below with permission.

The Western Australian Department of Primary Industries and Regional Development (WA DPIRD) Diagnostic and Laboratory Services (S. Bertazzoni and Dr. M. Kehoe) tested the EPPO (2020) quantitative PCR protocol by successfully identifying pinned *P. spumarius* that were 23 years old, and fresh *P. spumarius* material (~1 year old) in ethanol. However, their testing also indicated a positive detection of *P. spumarius* for a specimen that was later identified by CO1 barcoding as *Aphrophora salicina,* and an indeterminant result for a specimen known to be *Aphrophora salicina*. Five specimens representing five different genera native to Australia were tested using the EPPO protocol and were found to be negative: *Amarusa australis*, *Anyllis leiala, Bathyllus albicinctus*, *Liorhina loxosema* and *Philagra recurva.* In addition, further verification of this molecular assay by another laboratory (NSW DPIRD) revealed inconsistent detection: two out of three replicates for *Aphrophora alni* and one out of three replicates for each of *Neophilaenus exclamationis, Cercopis arcuata, Cercopis sanguinolenta* and *Anyllis leiala*. These inconsistent detections need to be considered before using this assay.

Quantitative PCR for the detection of* P. spumarius *DNA (EPPO, 2020; Lester et al, unpublished)

General information

- The target gene is the internal transcribed spacer 2 (ITS2) region, amplicon length is 157bp.
- Oligonucleotides:

Primer name	Sequence
PhiSpu_ITS2_F	5′-TCA TAA CCC CAC GTT TGT CC-3′
PhiSpu_ITS2_R	5′-CAA TTG TTC CGC ATC GTA CG-3′
PhiSpu_ITS2_P (HPLC purified)	5′-FAM-GCC CAC AAC CGC CAC GAC CA BHQ1-3′

- This test was successfully performed on QuantStudioTM Flex 6 machine.
- Analyses were performed using QuantStudioTM quantitativePCR software.

Method

1. DNA extraction from both adult and immature insects can be performed using commercially available kits such as the Blood & Tissue kit (Qiagen); an extraction protocol is described in PM 7/129 (EPPO, 2021). The extracted DNA should be used immediately or stored at approximately –20 °C until use.
2. Quantitativepolymerase chain reaction
    a. Master Mix

Reagent	Working concentration	Volume per reaction (µL)	Final concentration
Molecular grade water*	NA	5.75	NA
QuantitativePCR buffer JumpStart™ Taq ReadyMix™	2×	10	1×
Forward Primer (PhiSpu_ITS2_F)	10 µM	0.5	250 nM

Reagent	Working concentration	Volume per reaction (µL)	Final concentration
Reverse Primer (PhiSpu_ITS2_R)	10 µM	0.5	250 nM
Probe 1 (PhiSpu_ITS2_P)	10 µM	0.25	125 nM
Eukaryotic 18S Endogenous control (Applied Biosystems ®)	20×	1	1×
Subtotal		18	
DNA template		2	
Total		20	

[a] *Molecular grade water should be used preferably or prepared purified (deionized or distilled), sterile (autoclaved or 0.22 µm filtered) and nuclease-free.
[b] NA, not applicable.

b. PCR cycling conditions: 2 min at 95 °C, followed by 40 cycles of denaturation and annealing/elongation for 15 s at 95 °C and 1 min at 60 °C, respectively.

Essential procedural information

1. Obtain controls for a reliable test result. The following (external) controls should be included for each series of nucleic acid extractions and amplifications of the target organism and target nucleic acid, respectively:
    - Negative isolation control (NIC) to monitor contamination during nucleic acid extraction: nucleic acid extraction and subsequent amplification of clean extraction buffer.
    - Positive isolation control (PIC) to ensure that nucleic acid of sufficient quantity and quality is isolated: nucleic acid extraction and subsequent amplification of the target organism.
    - Negative amplification control (NAC) to rule out false positives due to contamination during the preparation of the reaction mix: amplification of molecular grade water that was used to prepare the reaction mix.
    - Positive amplification control (PAC) to monitor the efficiency of the amplification: amplification of nucleic acid of the target organism. This can include nucleic acid extracted from the target organism, whole genome amplified DNA or a synthetic control (e.g. cloned PCR product).

2. **Interpretation of results:**
   To assign results from PCR-based tests the controls require verification: The PIC and PAC amplification curves should be exponential. It is suggested that interpretation of the results from the quantitative PCR protocol be restricted to four categories as follows:
    - Positive: Detection of DNA consistent with *Philaenus spumarius* and replicated at least twice.
    - Negative*:* No detection of DNA consistent with *Philaenus spumarius* and replicated at least twice.

- Inconclusive: Replicates in *P. spumarius* specific assay give inconsistent results.
- Indeterminant: No result was obtained, sample contaminated or of poor quality or quantity for testing.

Sequences from reference specimens of *P. spumarius* from BOLD, particularly BOLD Process ID NZIND020-22 which is from New Zealand, and therefore of the closest proximity to Australia, should be compared.

From the information available from GenBank and BOLD, *P. spumarius* and *P. tesselatus* sequences are identical within the ITS2 region. The test may not discriminate between the two species (EPPO 2020). Theoretically, there are further species which may amplify with this qPCR such as *P. arslani* and *P. loukasi* (Roslyn Mirrington pers. comm. Sept 2023).

4.2.3 Xylella testing in spittlebugs

The pest status of *P. spumarius* is linked to its ability to act as a vector for the bacterium *Xylella*. If a suspect *P. spumarius* individual is detected in Australia, or indeed any suspect exotic Aphrophoridae, it should be tested for *Xylella*. Nymphs of *P. spumarius* have not, however, been linked to transmission and are unlikely to vector *Xylella* (Cornara et al. 2019). To test for *Xylella* in spittlebugs the CTAB-based protocol (Loconsole et al. 2014) is recommended. The head is removed from the rest of the insect, and the eyes are excised using a scalpel. The remaining parts of the head consisting of the vertex, frons and labium are used for testing, see a video of the dissection here https://www.ponteproject.eu/protocols-xylella/preparation-philaenus-spumarius-samples-xylella-fastidiosa-detection/ . Further methods are available to determine the specific strain of *Xylella* present (e.g., Elbeaino et al. 2020, Xylella draft NDP). As of April2025, an improved *Xylella* diagnostic and preparedness NDP is being developed through HIA (MT17006) involving a collaboration between Victoria, New South Wales, Western Australia, Queensland and New Zealand, which is currently being progressed for endorsement.

4.2.4 Additional information related to molecular methods

The LCO1490/HCO2198, C1J1718m/C1N2191m (Dallas et al. 2003), and Cerco-F/Cerco-R primers (Martoni et al., 2024) can also be used to aid in the pest diagnostics. The verifying lab (NSW DPIRD) has tested the COI primers LCO1490 (5′-GGTCAACAAATCATAAAGATATTGG-3′) and HCO2198 (5′-TAAACTTCAGGGTGACCAAAAAATCA-3′) on eight *P. spumarius* specimens. All eight specimens failed to amplify using these standard primers but were successfully amplified in other Cercopidae/Aphrophoridae genera, suggesting that these primers may still be broadly effective within the family. However, six out of the eight *P. spumarius* tested specimens were successfully amplified and sequenced using the primers C1-J-1718m (5′-GGAGGATTTGGAAATTGATTAGT-3′) and C1-N-2191m (5′-CAGGTAAAATTAAAATATAAACTTCTGG-3′) (Dallas et al., 2003), generating a 472 bp fragment.

In addition, Martoni et al. (2024) have recently developed a new set of Cercopidae-specific COI primers targeting a 642 bp fragment: Cerco-F (5′-TTYGGDATTTGATCAGGAATAATTGG-3′) and Cerco-R (5′-GAATAAATGTTGRTATAAAATWGGRTC-3′). These primers were successful in amplifying the Australian species in their study.

Please note that the individual lab should validate these primers before using them in their own laboratory.

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8 APPENDICES

8.1 Appendix 8.1 Host plant records for *Philaenus spumarius* (Aphrophoridae: Hemiptera)

Appendix 8.1. Host plant records for *Philaenus spumarius* (Aphrophoridae: Hemiptera). Superscript numbers in 'Geographic region' indicate the reference/s for the observation which are listed after the table. Further information, including additional host plant species for this pest, can be found in Thompson et al., (2023) and DAFF (2022).

Family	Genus	Species	Common name	Geographic region
Adoxaceae	*Sambucus*	*canadensis*	Common elder	USA[57]
Adoxaceae	*Sambucus*	*nigra*	European elder	Czech Republic[30] Netherlands[15] USA[57]
Adoxaceae	*Sambucus*	*racemosa*	Red elderberry	Germany[27]
Adoxaceae	*Sambucus*	*sp.*	Elderberry	USA (Hawaii)[11]
Adoxaceae	*Viburnum*	*opulus*	guelder rose	Netherlands[15]
Aizoaceae	*Carpobrotus*	*edulis*	Pig-face	Portugal[63], Spain[63]
Aizoaceae	*Mesembryanthemum*	*sp.*	Fig marigold	USA (Hawaii)[10]
Aizoaceae	*Sesuvium*	*portulacastrum*	Sea purslane	USA (Hawaii)[11]
Amaranthaceae	*Amaranthus*	*retroflexus*	Pigweed amaranth	Slovakia[49]
Amaranthaceae	*Beta*	*vulgaris*	Beet	USA[57]
Amaranthaceae	*Beta*	*vulgaris subsp. vulgaris*	Swiss chard	USA (Hawaii)[11]
Amaranthaceae	*Chenopodium*	*album*	Lambsquarter	England[58] USA[57]
Amaranthaceae	*Spinacia*	*oleracea*	Spinach	USA[57]
Amaryllidaceae	*Allium*	*cepa*	Onion	USA[57]
Amaryllidaceae	*Allium*	*schoenoprasum*	Chives	Finland[21]
Amaryllidaceae	*Allium*	*sp.*		Italy[16]
Amaryllidaceae	*Allium*	*vineale*	Wild garlic	USA[57]
Amaryllidaceae	*Pancratium*	*maritimum*	Sea daffodil	Portugal[63], Spain[63]
Anacardiaceae	*Rhus*	*glabra*	Smooth sumac	USA[57]
Anacardiaceae	*Rhus*	*sp.*	Sumac	USA[57]
Anacardiaceae	*Toxicodendron*	*diversilobum*	Pacific poison oak	USA [13]
Anacardiaceae	*Toxicodendron*	*pubescens*	Poison ivy	USA[57]
Apiaceae	*Aciphylla*	*sp.*		New Zealand[33]
Apiaceae	*Aegopodium*	*podagraria*	Goutweed, Ground elder	Europe[57]

Apiaceae	*Ammi*	*visnaga*	Toothpick-plant	Turkey[48]
Apiaceae	*Angelica*	*litoralis*	Garden angelica	Finland[21]
Apiaceae	*Angelica*	*silvestris*	Wild angelica, Wild chervil	Europe[57] Finland[21] UK[3, 58]
Apiaceae	*Apium*	*graveolens*	Celery	USA (Hawaii, Ohio)[57]
Apiaceae	*Apium*	*graveolens var. dulce*	Celery	USA[13]
Apiaceae	*Berula*	*erecta*	lesser water-parsnip or cutleaf waterparsnip	Netherlands[15]
Apiaceae	*Carum*	*carvi*	Caraway	Europe[57]
Apiaceae	*Caucalis*	*platycarpos*	Carrot bur parsley	Turkey[48]
Apiaceae	*Chaerophyllum*	*temulum*	rough chervil	Netherlands[15]
Apiaceae	*Conium*	*maculatum*	Poison hemlock	USA[13]
Apiaceae	*Conopodium*	*majus*	pignut	England[58]
Apiaceae	*Daucus*	*carota*	Carrot	France[57] Portugal[63], Spain[63] New Zealand[23] USA [13, 29, 57] USA (Hawaii)[10] Greece[64]
Apiaceae	*Daucus*	*sp.*		Italy[16]
Apiaceae	*Eryngium*	*sp.*	Eryngo, Seaholly	France[57]
Apiaceae	*Eryngium*	*sp.*	Eryngo, Seaholly	USA[35]
Apiaceae	*Falcaria*	*vulgaris*	Sickleweed	Europe[57]
Apiaceae	*Foeniculum*	*sp.*		Italy[16]
Apiaceae	*Foeniculum*	*vulgare*	Fennel	Portugal[63], Spain[63]
Apiaceae	*Heracleum*	*sphondylium*	Cow parsnip	Europe[57]
Apiaceae	*Pastinaca*	*sativa*	Parsnip	England[58] Europe, USA[57] USA (Hawaii) [10]
Apiaceae	*Petroselinum*	*crispum var. neapolitanum*	Italian parsley	USA[35]
Apiaceae	*Petroselinum*	*crispum*	Parsley	France[57] USA [13, 57] USA (Hawaii) [57]
Apiaceae	*Peucedanum*	*palustre*	Milk-parsley	Finland[21] UK[3]
Apiaceae	*Pimpinella*	*major*	Greater burnet saxifrage	USA[57]
Apiaceae	*Pimpinella*	*nigra*	Solidstem burnet saxifrage	USA[57]
Apiaceae	*Pimpinella*	*saxifraga*	Burnet saxifrage	USA[57]
Apiaceae	*Sanicula*	*liberta*		USA[13]
Apiaceae	*Scandix*	*pecten - veneris*		Greece[64]
Apiaceae	*Scandix*	*sp.*		Italy[16]
Apiaceae	*Seseli*	*tortuosum*	Mediterranean cicely	Portugal[63], Spain[63]
Apiaceae	*Tordylium*	*sp.*		Italy[16]
Apiaceae	*Torilis*	*japonica*	Erect hedgeparsley	Europe[57]
Apiaceae	*Torilis*	*nodosa*	knotted hedgeparsley	Portugal[63], Spain[63]
Apiaceae	*Torilis*	*sp.*		Italy[16]

Apocynaceae	*Alstonia*	*boonei*	cheese wood	Nigeria[17]
Apocynaceae	*Alyxia*	*oliviformis*	Maile (N)	USA (Hawaii)[11]
Apocynaceae	*Apocynum*	*cannabinum*	Indian hemp	USA[61]
Apocynaceae	*Catharanthus*	*roseus*	Madagascar periwinkle	Italy[8]
Apocynaceae	*Nerium*	*oleander*	Oleander	Italy[8]
Apocynaceae	*Vinca*	*major*		USA[13]
Apocynaceae	*Vinca*	*sp.*	Periwinkle	USA (Hawaii)[10]
Aquifoliaceae	*Ilex*	*anomala*	Hawai'i holly (N)	USA (Hawaii)[10]
Araceae	*Arum*	*italicum*	Italian arum	France[57]
Araceae	*Zantedeschia*	*aethiopica*	Calla lily	USA[13]
Araliaceae	*Hedera*	*canariensis*	Canarian ivy	USA[13]
Araliaceae	*Hedera*	*helix*	English ivy	USA[13]
Asparagaceae	*Asparagus*	*acutifolius*	Wild asparagus	Italy
Asparagaceae	*Chlorogalum*	*pomeridianum*	California soaproot	USA[13]
Asparagaceae	*Cordyline*	*fruticosa*	Cordyline	USA (Hawaii)[10]
Asparagaceae	*Muscari*	sp.		Italy[16]
Asphodelaceae	*Asphodelus*	Sp.		Greece[64]
Asphodelaceae	*Hemerocallis*	*fulva*	Daylily	USA[57]
Asphodelaceae	*Simethis*	*mattiazzii*	Kerry lilly	Portugal[63], Spain[63]
Asteraceae	*Acanthospermum*	*australe*	Spiny-bur, Paraguayan starburr	USA (Hawaii)[11]
Asteraceae	*Achillea*	*millefolium*	Yarrow	England[58] Europe[57] Finland[21] France[57] USA[50, 57] Italy[65]
Asteraceae	*Achillea*	*ptarmica*	Sneezewort	Europe[57]
Asteraceae	*Ambrosia*	*elatior*	Fine cut ragweed	USA[57]
Asteraceae	*Ambrosia*	*sp.*	Ragweed	USA[61]
Asteraceae	*Ambrosia*	*sp.*	Ragweed	USA[57]
Asteraceae	*Anaphalis*	*margaritacea*	Western pearly everlasting	Canada[60] USA[13, 25]
Asteraceae	*Andryala*	*integrifolia*		USA[57]
Asteraceae	*Antennaria*	*plantaginifolia*	Plantain leaf pussytoes	USA[57]
Asteraceae	*Anthemis*	*sp.*	Chamomile	France[57] Italy[16]
Asteraceae	*Anthemis*	*chia*		Greece[64]
Asteraceae	*Arctium*	*lappa*	Greater burdock	Europe[57]
Asteraceae	*Arctium*	*minus*	Common burdock	Netherlands[15] USA[29]
Asteraceae	*Arctium*	*sp.*	Burdock	USA[57]
Asteraceae	*Artemisia*	*absinthium*	Wormwood	[5]
Asteraceae	*Artemisia*	*campestris*	Field wormwood	Portugal[63], Spain[63]
Asteraceae	*Artemisia*	*dracunculus*	Tarragon	USA[57]

Asteraceae	*Artemisia*	*sp.*		[5]
Asteraceae	*Artemisia*	*vulgaris*	Mugwort	Europe[57] Finland[42] USA (Hawaii)[10] USA[13, 57]
Asteraceae	*Aster*	*sp.*		USA[57]
Asteraceae	*Baccharis*	*pilularis*	Coyote brush	USA [3]
Asteraceae	*Bellis*	*perennis*	Common daisy	USA[57]
Asteraceae	*Bellis*	*sp.*		Italy[16]
Asteraceae	*Bidens*	*pilosa var. minor*	Cobbler's pegs	USA (Hawaii)[10]
Asteraceae	*Calendula*	*arvensis*	Field marigold	Portugal[31]
Asteraceae	*Calendula*	*officinalis*	Common Marigold	Turkey[48]
Asteraceae	*Calendula*	*sp.*	Calendula	Italy[16] Greece [64] USA[57]
Asteraceae	*Carduus*	*nutans*	Musk thistle, Nodding thistle	England[58] USA[2, 19, 54]
Asteraceae	*Carduus*	*pycnocephalus*	Italian thistle	USA[13]
Asteraceae	*Carduus*	*sp.*		Italy[16]
Asteraceae	*Celmisia*	*sp.*	New Zealand aster	New Zealand[34]
Asteraceae	*Centaurea*	*sp.*		Italy[16]
Asteraceae	*Chondrilla*	*juncea*		Greece[64]
Asteraceae	*Chrysanthemum*	*maximum*	Shasta daisy	USA[13] USA (Hawaii)[10]
Asteraceae	*Chrysanthemum*	*sp.*	Chrysanthemum	USA[57] USA (Hawaii)[11]
Asteraceae	*Cichorium*	*inthybus*	Chicory	USA[57] Turkey[48]
Asteraceae	*Cichorium*	*sp.*		Italy[16]
Asteraceae	*Cirsium*	*arvense*	Creeping thistle	Canada[1, 60] Europe[57] New Zealand[23,33] Portugal[31] USA [14, 57] Italy[65]
Asteraceae	*Cirsium*	*eriophorum*	Wooly thistle	England[58]
Asteraceae	*Cirsium*	*lanceolatum*	Bull thistle	USA[13, 57]
Asteraceae	*Cirsium*	*palustre*	Marsh plume	Canada[1]
Asteraceae	*Cirsium*	*sp.*	Thistle	Europe[57] France[57] Italy[16] USA [57]
Asteraceae	*Cirsium*	*vulgare*	Bull thistle	Canada[1]
Asteraceae	*Coleostephus*	*myconis*	Corn marigold	Portugal[31]
Asteraceae	*Conyza*	*sumatrensis*	Tall fleabane	USA (Hawaii)[10]
Asteraceae	*Crepis*	*capillaris*	Smooth hawksbeard	Europe[57]
Asteraceae	*Crepis*	*paludosa*	Marsh hawk's-beard	Germany[27]
Asteraceae	*Crepis*	*sp.*	hawksbeard	Italy[16] Greece [64]
Asteraceae	*Crepis*	*tectorum*	Narrowleaf hawksbeard	Finland[21]
Asteraceae	*Cynara*	*scolymus*	Artichoke	USA[13]
Asteraceae	*Dahlia*	*sp.*	Dahlia	Europe[57] USA[13] USA (Hawaii)[10]
Asteraceae	*Delairea*	*odorata*	Cape ivy	USA (Hawaii)[10]
Asteraceae	*Dubautia*	*scabra*	Kupaoa (N)	USA (Hawaii)[10]

Asteraceae	*Echinops*	*ritro*	southern globethistle	Netherlands[15]
Asteraceae	*Erigeron*	*annuus*	Daisy fleabane	USA[57]
Asteraceae	*Erigeron*	*canadensis*	Canadian horseweed	Germany[57] Portugal[63] Spain[63]
Asteraceae	*Erigeron*	*glaucus*	Seaside fleabane	Slovakia[49]
Asteraceae	*Erigeron*	*philadelphicus*	Philadelphia fleabane, Robins plantain	USA[57, 61]
Asteraceae	*Erigeron*	*sp.*		Italy[16]
Asteraceae	*Eupatorium*	*cannabinum*	hemp-agrimony	Netherlands[15]
Asteraceae	*Felicia*	*petiolata*	Blue felicia	USA[13]
Asteraceae	*Galactites*	*sp.*		Italy[16]
Asteraceae	*Gazania*	*rigens*	Treasure flower	USA[13]
Asteraceae	*Glebionis*	*sp.*		Italy[16]
Asteraceae	*Glebionis*	*segetum*	Corn marigold/corn daisy	Portugal[31]
Asteraceae	*Gnaphalium*	*purpureum*	Purple cudweed	USA (Hawaii)[10]
Asteraceae	*Helianthus*	*sp.*	Sunflower	France[57] USA[57]
Asteraceae	*Helichrysum*	*italicum*	Curry plant	Portugal[63], Spain[63]
Asteraceae	*Helminthotheca*	*echioides*	Bristly ox-tongue	USA[13] Greece[64]
Asteraceae	*Hieracium*	*aurantiacum*	Orange hawkweed	USA[57]
Asteraceae	*Hieracium*	*caespitosum*	Meadow hawkweed	New Zealand[33]
Asteraceae	*Hieracium*	*laevigatum*	Smooth hawkweed	Europe[57]
Asteraceae	*Hieracium*	*lepidulum*	hawkweed	New Zealand[33]
Asteraceae	*Hieracium*	*pilosella*	Mouse-ear hawkweed	New Zealand[33]
Asteraceae	*Hieracium*	*piloselloides subsp. praealta*	Tall hawkweed	New Zealand[33]
Asteraceae	*Hieracium*	*praealtum*	Tall mouse-ear-hawkweed, King devil	New Zealand[34]
Asteraceae	*Hieracium*	*pratense*	King devil	USA[57]
Asteraceae	*Hieracium*	*tridentatum*	Threetooth hawkweed	Europe[57]
Asteraceae	*Hieracium*	*umbellatum*	hawkweed	Netherlands[15]
Asteraceae	*Hypochaeris*	*glabra*	Smooth catsear	USA[13]
Asteraceae	*Hypochaeris*	*radicata*	Catsear, false dandelion	USA (Hawaii)[10]
Asteraceae	*Hypochaeris*	sp.		Portugal[63], Spain[63]
Asteraceae	*Jacobaea*	*vulgaris*	ragwort	England[58]
Asteraceae	*Lactuca*	*sativa*	Lettuce*	USA (Hawaii)[10]
Asteraceae	*Lactuca*	*sp.*		Italy[16]
Asteraceae	*Lactuca*	*virosa*	Prickly lettuce	USA [57]
Asteraceae	*Lapsana*	*communis*	Common nipplewort	Europe[57] Netherlands[15]

Asteraceae	*Leontodon*	*hispida*	Bristly hawkbit	Europe[57]
Asteraceae	*Leontodon*	sp.	hawkbits	Portugal[63], Spain[63]
Asteraceae	*Leucanthemum*	*sylvaticum*		Portugal[63], Spain[63]
Asteraceae	*Leucanthemum*	*vulgare*	Oxeye daisy	Europe[57] Finland[21] UK[3] USA[57]
Asteraceae	*Madia*	*elegans*	Common madia	USA[13]
Asteraceae	*Matricaria*	*sp.*		France[57] Italy[16]
Asteraceae	*Petasites*	*officinalis*	Butterbur	Germany[27]
Asteraceae	*Petasites*	*sp.*	Butterburs	Europe[57]
Asteraceae	*Picris*	*sp.*		Italy[16]
Asteraceae	*Pluchea odorata*	*Pluchea odorata*	Sweetscent	USA (Hawaii)[10]
Asteraceae	*Psephellus*	*dealbatus*	Persian cornflower	Netherlands[15]
Asteraceae	*Reichardia*	*sp.*	Rudbeckia	Italy[16]
Asteraceae	*Rudbeckia*	*laciniata*	Rudbeckia	Netherlands[15]
Asteraceae	*Rudbeckia*	*laciniata var. hortensia*	Rudbeckia, Goldenglow	USA[13]
Asteraceae	*Scorzoneroides*	*autumnalis*	Fall dandelion, Autumn hawkbit	Europe[57] Finland[21] UK[3]
Asteraceae	*Senecio*	*ovatis (fuchsii)*	Wood ragwort	Germany[27]
Asteraceae	*Senecio*	*sp.*		Italy[16]
Asteraceae	*Senecio*	*vulgaris*		Portugal[63], Spain[63]
Asteraceae	*Silphium*	*perfoliatum*	cup plant	Netherlands[15]
Asteraceae	*Silybum*	*marianum*	Milk thistle	Greece[64]
Asteraceae	*Solidago*	*altissima*	Canada goldenrod*	USA (Hawaii)[10] USA[59]
Asteraceae	*Solidago*	*canadensis*	Canada goldenrod	USA[57]
Asteraceae	*Solidago*	*gigantea*	Giant goldenrod	Italy[65]
Asteraceae	*Solidago*	*rugosa*	Goldenrod	Canada[60]
Asteraceae	*Solidago*	*sp.*	Goldenrod	Netherlands[15] USA[53, 57, 61]
Asteraceae	*Solidago*	*virgaurea*	Solidago	Finland[21]
Asteraceae	*Sonchus*	*asper*	Spiny sowthistle	USA[13]
Asteraceae	*Sonchus*	*oleraceus*	Common sowthistle	USA[13] USA (Hawaii)[10] Portugal[63], Spain[63] Greece[64]
Asteraceae	*Sonchus*	*sp.*	Sonchus	Italy[16, 41] New Zealand[34]
Asteraceae	*Sonchus*	*tenerrimus*	Slender sowthistle	Portugal[31]
Asteraceae	*Symphyotrichum*	*tradescantii*	Shore aster (Michaelmas daisy?) [57]	
Asteraceae	*Tanacetum*	*vulgare*	Tansy	Finland[37] Netherlands[15] USA [13]
Asteraceae	*Taraxacum*	*officinale*	Dandelion	Turkey[48] USA[13,57] Italy[65]
Asteraceae	*Taraxacum*	*sp.*	Dandelion	Finland[21] France[57] Portugal[63], Spain[63] Greece[64]
Asteraceae	*Telekia*	*speciosa*	Bull's eye	Netherlands[15]
Asteraceae	*Tragopogon*	*porrifolius*	Salsify	USA[13]

Asteraceae	*Tragopogon*	*sp.*		Italy[16]
Asteraceae	*Tripleurospermum*	*inodorum*	Scentless chamomile	Europe[57]
Asteraceae	*Tripleurospermum*	*maritimum*	False mayweed	Finland[21]
Asteraceae	*Urospermum*	*sp.*		Italy[16]
Asteraceae	*Vernona*	*amygdalina*	Bitter Leaf	Nigeria[17]
Asteraceae	*Wyethia*	*amplexicaulis*	Mule ears	USA[61]
Balsaminaceae	*Impatiens*	*noli-tangere*	Touch-me-not balsam	Europe[57]
Balsaminaceae	*Impatiens*	*parviflora*	Touch-me-not balsam	Netherlands[15]
Berberidaceae	*Berberis*	*thunbergii*	Japanese barberry	USA[57]
Betulaceae	*Alnus*	*glutinosa*	European alder	Finland[20,22, 39] USA[57]
Betulaceae	*Alnus*	*incana*	Grey alder	Finland[20, 22, 39] USA[47]
Betulaceae	*Alnus*	*sp.*	Alder	USA[61]
Betulaceae	*Betula*	*pendula (verrucosa)*	Silver birch	[57]
Betulaceae	*Betula*	*pubescens*	Downy birch	[57]
Betulaceae	*Betula*	*sp.*	Birch	USA[61]
Betulaceae	*Betula*	*tremula*	Birch	[57]
Betulaceae	*Carpinus*	*betulus*	European hornbeam	Italy[65]
Betulaceae	*Carpinus*	*sp.*	Hornbeam	France[57]
Betulaceae	*Corylus*	*avellana*	Common hazel	Europe[57] Italy[65] Netherlands[15]
Boraginaceae	*Anchusa*	*officinalis*	common bugloss or alkanet [57]	
Boraginaceae	*Borago*	*sp.*		Italy[16]
Boraginaceae	*Cerinthe*	*sp.*		Italy[16]
Boraginaceae	*Echium*	*sp.*		Italy[16] Portugal[63], Spain[63]
Boraginaceae	*Echium*	*vulgare*	Viper's bugloss	Turkey[48]
Boraginaceae	*Echium*	*vulgare*	Viper's bugloss	
Boraginaceae	*Heliotropium*	*arborescens*	Garden heliotrope	USA[13]
Boraginaceae	*Lithospermum*	*officinale*	common gromwell	England[58]
Boraginaceae	*Myosotis*	*azorica*	Azorean mouse-ear	USA (Hawaii) [10]
Boraginaceae	*Myosotis*	*scorpioides*	True forget-me-not	USA[13]
Boraginaceae	*Myosotis*	*sp.*		Italy[16]
Boraginaceae	*Symphytum*	*officinale*	Common comfrey	France[57]
Brassicaceae	*Alliaria*	*petiolata*	Garlic Mustard	Canada[12] Netherlands[15]
Brassicaceae	*Armoracia*	*rusticana*	Horseradish	Europe[57] USA[61]
Brassicaceae	*Aurinia*	*saxatilis*	basket of gold	Netherlands[15]
Brassicaceae	*Barbarea*	*stricta*	Small-flowered winter-cress	Finland[21]
Brassicaceae	*Barbarea*	*verna*	American cress	USA[57]

Brassicaceae	*Barbarea*	*vulgaris*	Winter rocket cress	USA[57]
Brassicaceae	*Brassica*	*napus*	Rape, Canola	Europe[57]
Brassicaceae	*Brassica*	*nigra*	Black mustard	USA[57]
Brassicaceae	*Brassica*	*oleracea var. botrytis*	Cauliflower	USA (Hawaii)[10] USA[57]
Brassicaceae	*Brassica*	*oleracea var. gemmifera*	Brussels sprouts	USA (Hawaii)[11]
Brassicaceae	*Brassica*	*rapa*	turnip, field mustand, bird rape, etc	Netherlands[15]
Brassicaceae	*Brassica*	*sp.*		Italy[16]
Brassicaceae	*Cakile*	*maritima*	Sea rocket	Portugal[63], Spain[63]
Brassicaceae	*Capsella*	*bursa-pastoris*	Shepherd's purse	Europe[57] USA [57]
Brassicaceae	*Capsella*	*sp.*		Italy[16]
Brassicaceae	*Cardamine*	*pratensis*	Cuckooflower, Lady's smock[57]	
Brassicaceae	*Diplotaxis*	*sp.*		Italy[16]
Brassicaceae	*Erysimum*	*cheiranthoides*	Wormseed wallflower	Europe[57]
Brassicaceae	*Erysimum*	*cheiri*	Wallflower	USA[13]
Brassicaceae	*Hesperis*	*matronalis*	Dame's rocket	Europe[57]
Brassicaceae	*Isatis*	*sp.*		Italy[16]
Brassicaceae	*Isatis*	*tinctoria*	Woad	Finland[21]
Brassicaceae	*Mathiola*	*incana*	Hoary stock	USA[13]
Brassicaceae	*Myagrum*	*sp.*		Italy[16]
Brassicaceae	*Raphanus*	*sp.*		Italy[16]
Brassicaceae	*Raphanus*	*sativus*	Wild radish	USA[13]
Brassicaceae	*Raphanus*	*sativus var. longipinnatus*	Daikon	USA (Hawaii)[10]
Brassicaceae	*Rapistrum*	*sp.*		Italy[16]
Brassicaceae	*Sisymbrium*	*altissimum*	Tall hedge mustard	USA[57]
Brassicaceae	*Thlaspi*	*arvense*	Pennycress	Europe[57] USA[57]
Buddlejaceae	*Buddleja*	*davidii*	Butterfly bush	UK[3]
Buddlejaceae	*Buddleja*	*sp.*	Butterfly bush	USA[57]
Campanulaceae	*Campanula*	*medium 'Calycanthema'*	Canterbury bells	USA[57]
Campanulaceae	*Campanula*	*persicifolia*	Peach-leaved bellflower	USA[13]
Campanulaceae	*Campanula*	*trachelium*	nettle-leaved bellflower	Netherlands[15]
Campanulaceae	*Jasione*	*maritima*		Portugal[63], Spain[63]
Campanulaceae	*Legousia*	*sp.*		Italy[16]
Caprifoliaceae	*Centranthus*	*calcitrapae*		Portugal[63], Spain[63]
Caprifoliaceae	*Centranthus*	*ruber*	Red valerian	USA[13]
Caprifoliaceae	*Dipsacus*	*fullonum*	Wild teasle	England[58] USA[57]

Caprifoliaceae	*Knautia*	*arvensis*	Field scabious	England[58]
Caprifoliaceae	*Knautia*	*arvensis*	Field scabious	Europe[57]
Caprifoliaceae	*Knautia*	*sp.*		Italy[16]
Caprifoliaceae	*Lonicera*	*japonica*	Japanese honeysuckle	USA (Hawaii)[11]
Caprifoliaceae	*Lonicera*	*sp.*	Honeysuckle	USA[57]
Caprifoliaceae	*Sambucus*	*caerulea*	Blue elder	USA[13]
Caprifoliaceae	*Succisa*	*pratensis*	devil's-bit scabious	UK[18]
Caprifoliaceae	*Symphoricarpos*	*sp.*	Snowberry	France[57]
Caprifoliaceae	*Weigela*	*sp.*		France[57]
Caprifoliaceae	*Weigela*	*sp.*		USA[57]
Caryophyllaceae	*Cerastium*	*arvense*	Starry grasswort, field chickweed	Europe[57]
Caryophyllaceae	*Cerastium*	*glomeratum*		Greece[64]
Caryophyllaceae	*Cerastium*	*sp.*		Italy[16]
Caryophyllaceae	*Cerastium*	*vulgatum*	Mouse-ear chickweed	USA[57]
Caryophyllaceae	*Dianthus*	*barbatus*	Sweet william	USA[13]
Caryophyllaceae	*Dianthus*	*caryophyllus*	Carnation	USA[13]
Caryophyllaceae	*Dianthus*	*chinensis*	China pinks	USA (Hawaii)[10]
Caryophyllaceae	*Dianthus*	*sp.*	Carnation	USA[13, 57]
Caryophyllaceae	*Lychnis*	*coronaria 'Alba'*	White-flowered rose campion	USA[57]
Caryophyllaceae	*Lychnis*	*flos-cuculi*	Ragged-robin	UK[57]
Caryophyllaceae	*Sagina*	*procumbens*	Procumbent pearlwort	Finland[21]
Caryophyllaceae	*Saponaria*	*officinalis*	Common soapwort	USA[13, 29, 57]
Caryophyllaceae	*Scleranthus*	*annuus*	Annual knawel	Europe[57]
Caryophyllaceae	*Silene*	*dioica*	red campion	Netherlands[15]
Caryophyllaceae	*Silene*	*latifolia*	Bladder campion	Europe[57] UK[57]
Caryophyllaceae	*Silene*	*noctiflora*	Night-flowering catchfly	USA[57]
Caryophyllaceae	*Silene*	*sp.*	Campion	France[57]
Caryophyllaceae	*Silene*	*sp.*		Italy[16]
Caryophyllaceae	*Silene*	*uniflora*	sea campion	Portugal[63], Spain[63]
Caryophyllaceae	*Silene*	*vulgaris*	Bladder campion	England[58] Finland[21]
Caryophyllaceae	*Stellaria*	*graminea*	Grass-leaved stitchwort or starwort	Europe[57] Finland[21] England[58]
Caryophyllaceae	*Stellaria*	*media*	Common chickweed	Finland[21] USA [13, 57] Greece [64]
Caryophyllaceae	*Stellaria*	*nemoreum*	Wood stitchwort	Europe[57]
Caryophyllaceae	*Stellaria*	*palustris*	Meadow starwort	Europe[57]

Caryophyllaceae	*Stellaria*	*sp.*		Italy[16] Portugal[63], Spain[63]
Celastraceae	*Euonymus*	*japonicus cv. Microphyllus*	Japanese euonymus	Turkey[48]
Chenopodiaceae	*Chenopodium*	*sp.*		Italy[16]
Cistaceae	*Cistus*	*monspeliensis*	Montpellier cistus	France[7]
Cistaceae	*Cistus*	*salviifolius*	Gallipoli rose	Portugal[63], Spain[63]
Cistaceae	*Helianthemum*	*nummularium*	Rock rose	USA[13] UK[3]
Cistaceae	*Helianthemum*	*sp.*	Rock rose	France[57]
Combretaceae	*Terminalia catapa*	*Terminalia catapa*	Indian or Country almond	Nigeria[17]
Combretaceae	*Terminalia ivorensis*	*Terminalia ivorensis*	Ivory Coast almond	Nigeria[17]
Commelinaceae	*Commelina*	*diffusa*	Native wandering jew	USA (Hawaii)[10]
Commelinaceae	*Tradescantia*	*fluminensis*	Wandering Jew	USA [13]
Convolvulaceae	*Convolvulus*	*arvensis*	Field bindweed	England[58] UK[3] USA[13, 57] Italy[65]
Convolvulaceae	*Convolvulus*	*sp.*		[57] Italy[16]
Convolvulaceae	*Dichondra*	*sp.*	Dichondra	USA[61]
Convolvulaceae	*Ipomoea*	*batatas*	Sweet potato	USA (Hawaii)[10]
Convolvulaceae	*Ipomoea*	*indica*	Blue morning glory (N)	USA (Hawaii)[11]
Convolvulaceae	*Ipomoea*	*muricata*		USA[13]
Convolvulaceae	*Ipomoea*	*sp.*	Morning glory	USA (Maryland)
Coriariaceae	*Coriaria*	sp.		New Zealand[33]
Cornaceae	*Cornus*	*suecica*	Dwarf cornel, Bunchberry	Finland[21]
Crassulaceae	*Sedum*	*acre*	Goldmoss stonecrop	Finland[21]
Crassulaceae	*Sedum*	*telephium*	Orpine, Livelong	Finland[21]
Crassulaceae	*Sedum*	sp.	Stonecrops	Portugal[63], Spain[63]
Crassulaceae	*Umbilicus*	*rupestris*	Navelwort	Portugal[63], Spain[63]
Cucurbitaceae	*Bryonia*	*dioica*	ed bryony, white bryony	Netherlands[15]
Cucurbitaceae	*Ecballium*	*sp.*		Italy[16]
Cucurbitaceae	*Marah*	*fabaceus*	California manroot	USA[13]
Cupressaceae	*Juniperus*	*communis*	Juniper	Finland[21]
Cupressaceae	*Thuja*	*occidentalis*	American Arborvitae	USA[50]
Cyperaceae	*Carex*	*canescens*	Hoary sedge	Finland[21]
Cyperaceae	*Carex*	*nigra*	Common sedge	Finland[21] UK[3]
Cyperaceae	*Cyperus*	*brevifolius*	Mullumbimby couch	USA (Hawaii)[10]
Cyperaceae	*Eleocharis*	*mamillata*	Soft-stem spikerush	Finland[21]
Cyperaceae	*Schoenoplectus*	*tabernaemontani*	River clubsedge	UK[3]
Dennstaedtiaceae	*Pteridium*	*aquilinum*	Austral bracken, Eastern bracken	UK[3] ? [57]

Elaeagnaceae	*Hippophae*	*rhamnoides*	Common sea buckthorn	Europe[47]
Equisetaceae	*Equisetum*	*arvense*	Common horsetail	Europe[57] USA [57] Italy[65]
Ericaceae	*Calluna*	*vulgaris*	Common heather	Europe[57] UK[24]
Ericaceae	*Styphelia*	*tameiameiae*	Pūkiawe (N)	USA (Hawaii)[11]
Ericaceae	*Vaccinium*	*myrtillus*	Bilberry	[57]
Ericaceae	*Vaccinium*	*reticulatum*	Ohelo 'Ai, Hawaiian Blueberry (N)	USA (Hawaii)[11]
Ericaceae	*Vaccinium*	*sp.*	Blueberry	USA[55]
Euphorbiaceae	*Euphorbia*	*sp.*		Italy[16] Greece[64]
Euphorbiaceae	*Euphorbia*	*terracina*	Geraldton carnation weed	Portugal[63], Spain[63]
Euphorbiaceae	*Mercurialis*	*perennis*	Dog's mercury	UK[3]
Euphorbiaceae	*Mercurialis*	*sp.*		Italy[16]
Euphorbiaceae	*Ricinus*	*comminis*	Castor bean	Nigeria[17]
Fabaceae	*Acacia*	*dealbata*	Silver wattle	Portugal[63], Spain[63]
Fabaceae	*Acacia*	*longifolia*	Long-leaf wattle	Portugal[63], Spain[63]
Fabaceae	*Acacia*	*melanoxylon*	Australian blackwood	Portugal[63], Spain[63]
Fabaceae	*Acacia*	*saligna*	Orange wattle	Italy[8]
Fabaceae	*Argyrolobium*	*biebersteinii*		Turkey[48]
Fabaceae	*Astragalus*	*sp.*		Italy[16] Iran [68]
Fabaceae	*Coronilla*	*sp.*		Italy[16]
Fabaceae	*Cytisus*	*multiflorus*	White broom	Portugal[63], Spain[63]
Fabaceae	*Cytisus*	*scoparius*	Scotch broom	Portugal[63], Spain[63]
Fabaceae	*Delonix*	*regia*	Royal Poinciana	Nigeria[17]
Fabaceae	*Galega*	*officinalis*	Goat's rue	Turkey[48]
Fabaceae	*Lathyrus*	*odoratus*	Sweet pea	USA [57]
Fabaceae	*Lathyrus*	*palustris*	Marsh pea	UK [57]
Fabaceae	*Lathyrus*	*sp.*		Italy[16]
Fabaceae	*Lonchocarpus*		vine	Nigeria[17]
Fabaceae	*Lotus*	*corniculatus*	Birdsfoot trefoil	USA [53, 57] Portugal[63], Spain[63]
Fabaceae	*Lotus*	*pedunculatus*	Big trefoil	New Zealand[33]
Fabaceae	*Lotus*	*sp.*	Trefoil	Italy[16] USA[55, 57]
Fabaceae	*Medicago*	*littoralis*	shore medick	Portugal[63], Spain[63]
Fabaceae	*Medicago*	*lupulina*	Black medic	England[58] Europe[57]
Fabaceae	*Medicago*	*polymorpha*	Burclover	USA (Hawaii)[57] Greece[64]
Fabaceae	*Medicago*	*polymorpha subsp. hispida*	Burclover	USA (Hawaii)[10]
Fabaceae	*Medicago*	*sativa*	Lucerne, Alfalfa	New Zealand[23, 28, 33] USA [13, 25, 40, 51, 57, 61] Italy[65] Iran[68]

Fabaceae	*Medicago*	*sp.*		Italy[16] USA[61]
Fabaceae	*Melilotus*	*alba*	White sweet clover	USA[57]
Fabaceae	*Melilotus*	*indica*	Yellow melilot	USA[13]
Fabaceae	*Melilotus*	*officinalis*	Yellow sweet clover	Turkey[48] USA[57]
Fabaceae	*Melilotus*	*sp.*	Sweet clover	Italy[16] USA[52]
Fabaceae	*Onobrychis*	*viciifolia*	Sainfoin	France
Fabaceae	*Parkia*	*biglobosa*	African locust bean	Nigeria[17]
Fabaceae	*Pisum*	*sativum*	Garden pea	USA[54, 57]
Fabaceae	*Pisum*	*sp.*		Italy[16]
Fabaceae	*Psorolea*	*bituminosa*	Arabian pea	Turkey[48]
Fabaceae	*Robinia*	*pseudoacacia*	Black locust	Italy[65]
Fabaceae	*Samanea*	*saman*	Monkey pod tree	Nigeria[17]
Fabaceae	*Scorpiurus*	*sp.*		Italy[16]
Fabaceae	*Spartium*	*junceum*	Spanish broom	Italy[8]
Fabaceae	*Trifolium*	*campestre*	Hop trefoil	USA (Hawaii)[10]
Fabaceae	*Trifolium*	*hybridum*	Alsike clover	Europe[57] USA[57]
Fabaceae	*Trifolium*	*incarnatum*	Crimson clover	USA[54]
Fabaceae	*Trifolium*	*medium*	Zigzag clover, Mammoth clover	USA (Ohio)
Fabaceae	*Trifolium*	*pratense*	Red clover	UK[3] USA[52, 57, 61] Italy[65]
Fabaceae	*Trifolium*	*repens*	Ladino clover, White Dutch	England[58] USA[57] Greece [64]
Fabaceae	*Trifolium*	*sp.*	Clover	France[57] New Zealand[33, 34] USA [52, 57, 61] Portugal[63], Spain[63] Italy[16] Greece[64]
Fabaceae	*Vicia*	*sp.*	vetch	France[57] Italy[16] Portugal[63], Spain[63]
Fabaceae	*Vicia*	*cracca*	Tufted vetch	Finland[21]
Fabaceae	*Vicia*	*hirsuta*	Hairy vetch	Europe[57]
Fabaceae	*Vicia*	*sativa*	Common vetch	USA[57]
Fabaceae	*Vicia*	*sativa ssp. angustifolia*	Narrow-leaf vetch	USA[55]
Fabaceae	*Vicia*	*villosa*	Winter vetch	USA
Fagaceae	*Quercus*	*rubra*	Northern red oak	Italy[65]
Fumariaceae	*Fumaria*	*sp.*		Italy[16]
Gentianaceae	*Centaurium*	*erythraea*	Common centaury	England[58] USA (Hawaii) [10]
Gentianaceae	*Centaurium*	*pulchellum*	Lesser centaury	France[57]
Geraniaceae	*Erodium*	*cicutarium*		Greece[64]
Geraniaceae	*Erodium*	*sp.*		Italy[16] Greece[64]
Geraniaceae	*Geranium*	*carolinianum var. australe*	Carolina geranium	USA (Hawaii)[10]
Geraniaceae	*Geranium*	*dissectum*	cut-leaved crane's-bill	Portugal[63], Spain[63]
Geraniaceae	*Geranium*	*molle*		Greece[64]

Geraniaceae	*Geranium*	*robertianum*	Robert's geranium	Portugal[63], Spain[63]
Geraniaceae	*Geranium*	*sanguineum*	bloody crane's-bill	Europe[57]
Geraniaceae	*Geranium*	*sp.*		Italy[16] Portugal[63], Spain[63]
Geraniaceae	*Geranium*	*pratense*	meadow crane's-bill	Netherlands[15]
Geraniaceae	*Pelargonium*	*× hortorum*	Fish geranium	USA[13]
Geraniaceae	*Pelargonium*	*× domesticum*	Regal geranium	USA[13]
Geraniaceae	*Pelargonium*	*graveolens*	Rose geranium	USA[13] USA (Hawaii)[11]
Geraniaceae	*Pelargonium*	*peltatum*	Ivy geranium	USA[13]
Grossulariaceae	*Ribes*	*silvestre? (sativum)*	Garden or Woodland currant	USA[13]
Grossulariaceae	*Ribes*	*sp.*	Currant	UK[57]
Hydrangeaceae	*Hydrangea*	*macrophylla*	Bigleaf hydrangea	Europe[57]
Hydrangeaceae	*Hydrangea*	*paniculata var. grandiflora*	Peegee hydrangea	USA[13]
Hydrangeaceae	*Hydrangea*	sp.		USA[57]
Hypericaceae	*Hypericum*	*hirsutum*	hairy St John's-wort.	England[58]
Hypericaceae	*Hypericum*	*maculatum or tetrapterum*	St. John's Wort	Europe[57]
Hypericaceae	*Hypericum*	*moserianum*	St. John's Wort	USA [13] USA (Hawaii)[10]
Hypericaceae	*Hypericum*	*perforatum*	St. John's Wort	England[58] Europe[57] UK[3]
Iridaceae	*Crocosmia*	*× crocosmiiflora*	Montbretia	USA (Hawaii)[57]
Iridaceae	*Gladiolus*	sp.	Gladiolus	USA[13]
Iridaceae	*Iris*	*germanica*	Bearded iris or German iris	Turkey[48]
Iridaceae	*Iris*	sp.		USA[29, 57]
Iridaceae	*Tritonia*	*x crocosmiiflora*	Tritonia	USA (Hawaii)[10]
Juglandaceae	*Hicoria*	*ovata*	Shagbark hickory	USA[57]
Juglandaceae	*Juglans*	sp.	walnut	Turkey[66]
Juncaceae	*Juncus*	*gerardii*	Blackgrass	UK[3]
Juncaceae	*Juncus*	sp.		Iran [68]
Juncaginaceae	*Triglochin*	*maritimum*	Coriander grass	UK[3]
Lamiaceae	*Ajuga*	*reptans*	bugle, blue bugle, bugleherb	England[58]
Lamiaceae	*Dracocephalum*	*parviflorum*	American dragonhead	USA[57]
Lamiaceae	*Galeopsis*	*bifida*	Common hemp-nettle	Finland[21]
Lamiaceae	*Glechoma (Nepeta)*	*hederacea*	Ground-ivy	USA[57]
Lamiaceae	*Lamium*	*amplexicaule*	Common henbit	USA[57]
Lamiaceae	*Lamium*	*maculatum*	spotted dead-nettle	France[57] Portugal[63], Spain[63]
Lamiaceae	*Lamium*	*sp.*		Italy[16]
Lamiaceae	*Lavandula*	*angustifolia*	English lavender	[5]
Lamiaceae	*Lavandula*	*sp.*	Lavender	New Zealand[23, 34] Bulgaria[67]

Lamiaceae	*Lycopus*	*europaeus*	Gypsywort	UK[3]
Lamiaceae	*Mentha*	*sp.*	Mint	USA[57] USA (Hawaii) [10]
Lamiaceae	*Mentha*	*spicata*	Spearmint	USA[13]
Lamiaceae	*Monarda*	*fistulosa*	Wild bergamot	USA[57]
Lamiaceae	*Monarda*	*sp.*	Wild bergamot, Beebalm	USA[35]
Lamiaceae	*Monarda*	*didyma*	Bee Balm	Netherlands[15]
Lamiaceae	*Nepeta*	*cataria*	Catnip	USA[57]
Lamiaceae	*Nepeta*	*mussinii*	Catmint	USA[13]
Lamiaceae	*Origanum*	*majorana*	Sweet marjoram	USA[13, 57]
Lamiaceae	*Origanum*	*vulgare*	Wild marjoram	USA[13]
Lamiaceae	*Phlomis*	*sp.*		Italy[16]
Lamiaceae	*Prunella*	*vulgaris*		England[58]
Lamiaceae	*Rosemarinus*	*officinalis*	Rosemary	USA[13, 57]
Lamiaceae	*Rosmarinus*	*sp.*		Italy[16]
Lamiaceae	*Salvia*	*coccinea*	Texas sage	USA[13]
Lamiaceae	*Salvia*	*officinalis*	Narrowleaf sage	USA[13]
Lamiaceae	*Salvia*	sp.	Sage	USA[57]
Lamiaceae	*Satureia*	sp.	Savory	France[57]
Lamiaceae	*Scutellaria*	*galericulata*	Common skullcap	Finland[21]
Lamiaceae	*Stachys*	*ajugoides*	Bugle hedgenettle	USA[13]
Lamiaceae	*Stachys*	*bullata*	California hedgenettle	USA[13]
Lamiaceae	*Thymus*	*vulgaris*	Common thyme	USA[13]
Lamiaceae	*Westringia*	*fruticosa*	coastal rosemary	Italy[8]
Lythraceae	*Lythrum*	*maritimum*	Pukamole, Loosestrife	USA[57] USA (Hawaii)[10]
Lythraceae	*Lythrum*	*salicaria*	Purple loosestrife	Europe[57] Finland[21, 42] UK[3]
Lythraceae	*Punica*	*granatum*	pomegranate	Iran[68]
Malvaceae	*Alcea (Althaea)*	*rosea*	Hollyhock	USA[13, 57]
Malvaceae	*Hibiscus*	*tiliaceus*	Coastal hibiscus (N)	USA (Hawaii)[10]
Malvaceae	*Kokia*	*drynarioides*	Hawaiian tree cotton (N)	USA (Hawaii)[11]
Malvaceae	*Malva*	*moschata*	musk mallow	England[58]
Malvaceae	*Malva*	*parviflora*	Cheese weed	USA[13]
Malvaceae	*Malva*	*sp.*		Italy[16]
Malvaceae	*Modiola*	*caroliniana*	Carolina mallow	USA (Hawaii)[10]
Malvaceae	*Tilia*	*platyphyllos*	Large leaved linden	Europe[57]
Malvaceae	*Tilia*	*sp.*	American linden tree	USA[61]
Melastomataceae	*Tibouchina*	*semidecandra*	Lasiandra	USA (Hawaii)[10]

Menyanthaceae	*Menyanthes*	*trifoliata*		Netherlands[15]
Montiaceae	*Claytonia*	*virginica*	Virginia springbeauty	USA (Ohio)
Myricaceae	*Myrica*	*gale*	Bog-myrtle	Ireland[36] UK[3]
Myrtaceae	*Eucalyptus*	*sp.*	Eucalypt	[5]
Myrtaceae	*Leptospermum*	*laevigatum*	Coastal tee tree	USA[13]
Myrtaceae	*Metrosideros*	*excelsa*	pōhutukawa	New Zealand[33]
Myrtaceae	*Metrosideros*	*polymorpha*	ʻōhiʻa lehua	USA (Hawaii)[10]
Oleaceae	*Fraxinus*	*excelsior*	European ash	[57]
Oleaceae	*Fraxinus*	*pennsylvanica var. lanceolata*	Green ash	USA[57]
Oleaceae	*Ligustrum*	*ovalifolium*	California privet	USA[57]
Oleaceae	*Ligustrum*	*sp.*	Privet	USA[57]
Oleaceae	*Ligustrum*	*sp.*	Privet	USA[29]
Oleaceae	*Ligustrum*	*vulgare*	Common privet	USA[57]
Oleaceae	*Olea*	*europaea cv. Coratina*	Olive	Italy[8]
Oleaceae	*Syringa*	*vulgaris*	Lilac	USA[29, 57]
Onagraceae	*Chamaenerion*	*angustifolium*	Fireweed, Rosebay willowherb	Europe[57] Finland[21, 37]
Onagraceae	*Epilobium*	*palustre*	Marsh willowherb	Finland[21]
Onagraceae	*Epilobium*	*sp.*	Willow-herb	Europe[57] France[57]
Onagraceae	*Fuchsia*	*arborescens*	Tree fuchsia	USA (Hawaii)[11]
Onagraceae	*Fuchsia*	*magellanica*	Hummingbird fuchsia	USA (Hawaii)[10]
Onagraceae	*Fuchsia*	*triphylla*	Fuchsia	USA[13]
Onagraceae	*Oenothera*	*biennis*	Common evening primrose*	Europe[57] UK[3] USA[57]
Onagraceae	*Oenothera*	*lamarckiana*	Evening primrose	Netherlands[15]
Onagraceae	*Oenothera*	*odorata*	Evening Primrose	USA[57]
Onagraceae	*Oenothera*	*stricta*	Common Evening Primrose *	USA (Hawaii)[10]
Orchidaceae	*Dactylorhiza*	*maculata*	Heath spotted-orchid	Finland[21]
Orobanchaceae	*Bellardia*	*sp.*		Italy[16]
Orobanchaceae	*Melampyrum*	*sp.*		Finland[37]
Orobanchaceae	*Rhinanthus*	*angustifolius*	narrow-leaved rattle or greater yellow-rattle	Netherlands[15]
Orobanchaceae	*Rhinanthus*	*minor*	Yellow rattle	Finland[21]
Oxalidaceae	*Oxalis*	*corniculata*	Creeping oxalis	USA (Hawaii)[11]
Oxalidaceae	*Oxalis*	*sp.*	Wood sorrel	Italy[16] USA[57]
Papaveraceae	*Eschscholtzia*	*californica*	California poppy	USA[13]
Papaveraceae	*Papaver*	*argemone*	Long pricklyhead poppy	[57]
Papaveraceae	*Papaver*	*dubium*	Long-headed poppy	[57]

Papaveraceae	*Papaver*	*orientale*	Oriental poppy	USA[13, 57]
Papaveraceae	*Papaver*	*rhoeas*	Flanders poppy, Corn poppy	USA[13]
Papaveraceae	*Papaver*	*somniferum*	Opium poppy	[57]
Papaveraceae	*Papaver*	*sp.*	Poppy	Italy[16] USA[57]
Phrymaceae	*Erythranthe*	*guttata (guttatus)*	common yellow monkeyflower	USA[6]
Phyllanthaceae	*Phyllanthus*	*sp.*		Nigeria[17]
Pinaceae	*Picea*	*pungens*	Blue spruce	Italy[65]
Pinaceae	*Pinus*	*strobus*	White pine	USA (Hawaii)[57]
Pinaceae	*Pinus*	*sylvestris*	Scotch pine	USA[56]
Plantaginaceae	*Hebe*	*salicifolia*	Willow-leaf hebe	USA (Hawaii)[10]
Plantaginaceae	*Hebe*	*sp.*	Hebe	New Zealand[34]
Plantaginaceae	*Linaria*	*vulgaris*	Common toadflax	Europe[57]
Plantaginaceae	*Plantago*	*lagopus var. genuina*	Plantain	Europe[57]
Plantaginaceae	*Plantago*	*lanceolata*	Ribwort plantain	England[58] Europe[57] USA [13, 57] USA (Hawaii) [10] Portugal[63], Spain[63]
Plantaginaceae	*Plantago*	*major*	Common plantain	Europe[57] Finland[21] USA [57]
Plantaginaceae	*Plantago*	*maritima*	Sea plantain	UK[3]
Plantaginaceae	*Plantago*	*rugelii*	Purple stemmed plantain	USA[57]
Plantaginaceae	*Plantago*	*sp.*	Plantain	Italy[16] USA[52]
Plantaginaceae	*Veronica*	*arvensis*	Wall speedwell, Corn speedwell	Europe[57]
Plantaginaceae	*Veronica*	*chamaedrys*	Bird's-eye speedwell	Europe[57] England[58]
Plantaginaceae	*Veronica*	*longifolia*	Garden speedwell	Finland[21]
Plantaginaceae	*Veronica*	*peregrina*	Neckweed, Purslane speedwell	USA[57]
Plantaginaceae	*Veronica*	*persica*	Creeping speedwell	England[58]
Plantaginaceae	*Veronica*	*plebeia*	Creeping speedwell	USA (Hawaii)[10]
Plantaginaceae	*Veronica*	*sp.*	Speedwell	Italy[16] France[57]
Plantaginaceae	*Veronica*	*stricta var. stricta*	Koromiko	New Zealand[33]
Platanaceae	*Platanus*	*occidentalis*	American plane	USA[57]
Plumbaginaceae	*Limonium*	*sp.*	Statice	[57]
Poaceae	*?*	*?*	"Introduced genera/species"	New Zealand[33]
Poaceae	*Agropyron*	*repens*	Quack grass	USA[57]
Poaceae	*Agrostis*	*alba*	Redtop	USA[57]
Poaceae	*Agrostis*	*alba var. stolonifera*	Creeping bent	USA[57]
Poaceae	*Agrostis*	*alba var. vulgaris*	Fine bent	Europe[57]
Poaceae	*Agrostis*	*capillaris*	Common Bent	Turkey[48]
Poaceae	*Agrostis*	*sp.*		France[57]
Poaceae	*Agrostis*	*stolonifera*	Creeping bentgrass	Finland[21]

Poaceae	*Anthoxanthum*	*odoratum*	Sweet vernal grass	USA[57]
Poaceae	*Apera*	*spica-venti*	Loose silky-bent	Europe[57]
Poaceae	*Arrhenatherum*	*elatius*	Tall oat-grass	USA[57] Italy[65]
Poaceae	*Avena*	*barbata ssp. barbata*	Slender wild oat	Turkey[48]
Poaceae	*Avena*	*sativa*	Oats	USA[57]
Poaceae	*Avena*	*sp.*		Italy[16]
Poaceae	*Brachiaria*	*mutica*	Para grass	USA (Hawaii)[10]
Poaceae	*Briza*	*sp.*		Italy[16]
Poaceae	*Bromus*	*hordeaceus ssp. hordeaceus*	Soft brome	Turkey[48]
Poaceae	*Bromus*	*sp.*	Brome grass	Italy[16] USA[57]
Poaceae	*Bromus*	*tectorum*	Brome grass	Europe[57]
Poaceae	*Coix*	*lacryma-jobi*	Job's tears	USA (Hawaii)[10]
Poaceae	*Corynephorus*	*canescens*	Grey hair-grass	Europe[57]
Poaceae	*Cynodon*	*dactylon*	Couch grass	USA (Hawaii)[10]
Poaceae	*Dactylis*	*glomerata*	Cock's-foot	Europe[57] USA (Hawaii)[10] USA[57] Portugal[63], Spain[63]
Poaceae	*Dactylis*	*sp.*		Italy[16]
Poaceae	*Deschampsia*	*flexuosa*	Wavy hairgrass	Finland[21]
Poaceae	*Digitaria*	*setigera*	East Indian crabgrass	USA (Hawaii)[10]
Poaceae	*Elymus*	*arenarius*	Blue lyme grass	Finland[21]
Poaceae	*Eriogonum*	*saxatile*	Hoary buckwheat	USA [13]
Poaceae	*Festuca*	*glauca*	Blue fescue	Europe[57]
Poaceae	*Festuca*	*ovina*	Sheep's fescue	Europe[57]
Poaceae	*Festuca*	*rubra*	Red fescue	Europe[57] Finland[21] UK[3]
Poaceae	*Holcus*	*lanatus*	Common velvetgrass	USA (Hawaii)[10]
Poaceae	*Holcus*	*sp.*	Velvetgrass	Germany[57]
Poaceae	*Hordeum*	*bulbosum*	Bulbous barley	Turkey[48]
Poaceae	*Hordeum*	*murinum*	Wild barley, Wall barley	USA[13, 57]
Poaceae	*Hordeum*	*murinum ssp. leporinum*	Barley grass	Turkey[48]
Poaceae	*Hordeum*	*sp.*		Italy[16]
Poaceae	*Hordeum*	*vulgare*	Barley	USA[55, 57]
Poaceae	*Lagurus*	*ovatus*	hare's-tail	Portugal[63], Spain[63]
Poaceae	*Lepidium*	*campestre*	Field peppergrass	USA[57]
Poaceae	*Lepidium*	*ruderale*	Roadside peppergrass	Europe[57]
Poaceae	*Lepidium*	*virginicum*	Virginia peppergrass	USA[57]
Poaceae	*Lolium*	*multiflorum*	Italian ryegrass	USA[57]
Poaceae	*Lolium*	*perenne*	Perennial ryegrass	Europe[57] USA[57]

Poaceae	*Lolium*	*sp.*		Italy[16]
Poaceae	*Panicum*	*purpurascens*	Para grass	USA (Hawaii)[57]
Poaceae	*Panicum*	*virgatum*	Switchgrass	USA[26]
Poaceae	*Pennisetum*	*clandestinum*	Kikuyu grass	New Zealand[44]
Poaceae	*Phalaris*	*arundinacea*	Reed canary grass	Finland[21]
Poaceae	*Phalaris*	*sp.*		Italy[16]
Poaceae	*Phleum*	*pratense*	Timothy grass	Europe[57] USA[57, 61]
Poaceae	*Phleum*	*sp.*		France[57] Italy[16]
Poaceae	*Poa*	*palustris*	Fowl bluegrass	Europe[57] Finland[21] USA[57]
Poaceae	Poaceae spp.	*sp.*		Italy[16]
Poaceae	*Sacciolepis*	*contracta*	Glenwood grass	USA (Hawaii)[10]
Poaceae	*Sorghum*	*sp.*		Italy[16]
Poaceae	*Triticum*	*aestivum*	Wheat	USA[55,57] Iran[68]
Poaceae	*Zea*	*mays*	Corn	USA [57, 61]
Polemoniaceae	*Phlox*	*panieulata*		Netherlands[15]
Polemoniaceae	*Phlox*	*sp.*	Phlox	France[57] USA [57]
Polemoniaceae	*Polemonium*	*caeruleum*	Jacob's-ladder	Europe[57]
Polygonaceae	*Polygala*	*myrtifolia*	myrtle-leaf milkwort	Italy[8]
Polygonaceae	*Polygonum*	*amphibium*	longroot smartweed, water knotweed	Netherlands[15]
Polygonaceae	*Polygonum*	*coccineum*	Water smartweed	Europe[57]
Polygonaceae	*Rheum*	*rhaponticum*	False rhubarb	USA (Hawaii)[10] USA [57]
Polygonaceae	*Rumex*	*acetosa*	Common sorrel	Europe[57] Finland[21] UK[58] USA[57] Portugal[63], Spain[63]
Polygonaceae	*Rumex*	*acetosella*	Sheep sorrel	Europe[57] UK[3, 46] USA [13, 29, 57] USA (Hawaii)[10]
Polygonaceae	*Rumex*	*bucephalophorus*		Greece[64]
Polygonaceae	*Rumex*	*conglomeratus*	Clustered dock, Green dock	USA[13]
Polygonaceae	*Rumex*	*crispus*	Curly dock	Europe[57] Finland[21] USA[13, 57]
Polygonaceae	*Rumex*	*obtusifolius*	Broad-leaf dock	Europe[57] Netherlands[15] UK[3] USA[57]
Polygonaceae	*Rumex*	*occidentalis*	Western dock	USA (Hawaii)[57]
Polygonaceae	*Rumex*	*sp.*		Italy[16]
Polypodiaceae	*Polypodium*	*vulgare*	Common polypody	Finland[21]
Primulaceae	*Anagallis*	*arvensis*	Scarlet pimpernel	USA (Hawaii)[10]
Primulaceae	*Lysimachia*	*foemina*	Blue pimpernel	Greece[64]
Primulaceae	*Lysimachia*	*sp.*		Italy[16]
Primulaceae	*Lysimachia*	*thyrsiflora*	Tufted loosestrife	UK[3]
Primulaceae	*Lysimachia*	*vulgaris*	Yellow garden loosestrife	Finland[21, 42] UK[3]

Primulaceae	*Primula*	*sp.*		USA[57]
Primulaceae	*Primula*	*veris*		England[58]
Ranunculaceae	*Adonis*	*sp.*		Italy[16]
Ranunculaceae	*Aquilegia*	*canadensis*	Wild columbine	USA [57]
Ranunculaceae	*Aquilegia*	*chrysantha*	Golden columbine	USA [13]
Ranunculaceae	*Caltha*	*palustris*	Marsh marigold	England[58] Europe[57] Netherlands[15] UK[3]
Ranunculaceae	*Clematis*	*vitalba*	Old man's beard	France[57] Turkey[48]
Ranunculaceae	*Nigella*	*sp.*		Italy[16]
Ranunculaceae	*Ranunculus*	*abortivus*	Littleleaf buttercup	USA (Ohio)
Ranunculaceae	*Ranunculus*	*acris*	Meadow buttercup	Europe[57] France[57] Netherlands[15] UK[3] USA[57]
Ranunculaceae	*Ranunculus*	*bulbosus*	Bulbous buttercup	Europe[57]
Ranunculaceae	*Ranunculus*	*flammula*	Lesser spearwort	Europe[57]
Ranunculaceae	*Ranunculus*	*lingua*	Greater spearwort	Netherlands[15]
Ranunculaceae	*Ranunculus*	*muricatus*		Greece[64]
Ranunculaceae	*Ranunculus*	*repens*	Creeping buttercup	England[58] Europe[57] Portugal[63], Spain[63]
Ranunculaceae	*Ranunculus*	*repens var pleniflorus*	Double-flowered creeping buttercup	USA[13]
Ranunculaceae	*Ranunculus*	*sp.*	Buttercups	France[57] Italy[16] USA[57]
Ranunculaceae	*Thalictrum*	*minus*	lesser meadow-rue	Netherlands[15]
Resedaceae	*Reseda*	*sp.*		Italy[16]
Rhamnaceae	*Ceanothus*	*sanguineus*	Redstem ceanothus	USA [61]
Rosaceae	*Alchemilla*	*glabra*	smooth ladies mantle	England[58]
Rosaceae	*Agrimonia*	*eupatoria*	common agrimony	England[58]
Rosaceae	*Amelanchier*	*sp.*	Serviceberry	USA[57]
Rosaceae	*Chaenomeles*	*sp.*	Japanese quince, Flowering quince	USA[57]
Rosaceae	*Chaenomeles*	*speciosa (lagenaria)*	Japanese quince, Flowering quince	USA[13]
Rosaceae	*Comarum*	*palustre*	Marsh cinquefoil	Finland[21] UK[3]
Rosaceae	*Crataegus*	*laevigata*	Midland hawthorn	UK[46]
Rosaceae	*Crataegus*	*monogyna*	Hawthorn	UK[46]
Rosaceae	*Filipendula*	*ulmaria*	Meadowsweet	Europe[57] Finland[21, 37, 42] Netherlands[15] UK[3]
Rosaceae	*Fragaria*	*ananassa*	Strawberry	Finland[42] New Zealand[28] USA (Hawaii) [29] USA [4,29, 45, 51, 61, 62]
Rosaceae	*Fragaria*	*ananassa cv. Hood*	Strawberry	USA[55]
Rosaceae	*Fragaria*	*chiloensis*	Chilean strawberry (N)	USA[13] USA (Hawaii)[11]
Rosaceae	*Fragaria*	*sp.*	Strawberry	Canada [38] USA[57]

Rosaceae	*Fragaria*	*vesca*	European wood strawberry [57]	
Rosaceae	*Fragaria*	*virginiana*		USA[57]
Rosaceae	*Geum*	*rivale*	Purple avens	England[58] [57]
Rosaceae	*Geum*	*urbanum*	Cloveroot, Avens	Netherlands[15] [57]
Rosaceae	*Malus*	*sp.*	Apple	USA[57] Iran[68]
Rosaceae	*Potentilla*	*anserina*	Silverweed cinquefoil	UK[3]
Rosaceae	*Potentilla*	*canadensis*	Five finger, Dwarf cinquefoil	USA[57]
Rosaceae	*Potentilla*	*erecta*	Tormentil, Erect cinquefoil	Finland[21]
Rosaceae	*Potentilla*	*monspeliensis*	Rough cinquefoil	[57]
Rosaceae	*Potentilla*	*reptans*	creeping cinquefoi	England[58]
Rosaceae	*Prunus*	*armeniaca*	Apricot	USA[13]
Rosaceae	*Prunus*	*avium*	Wild cherry	Italy[8,65] USA[57]
Rosaceae	*Prunus*	*cerasus*	Sour cherry	USA[57]
Rosaceae	*Prunus*	*domestica*	European plum	USA[57]
Rosaceae	*Prunus*	*ducis*	Almond	[5]
Rosaceae	*Prunus*	*pensylvanica*	Fire cherry, Bird cherry	[57]
Rosaceae	*Prunus*	*persica*	Peach	USA[57]
Rosaceae	*Prunus*	*persica × dulcis*	rootstock	Italy[8]
Rosaceae	*Prunus*	*sp.*	Plum	USA[13, 29, 57]
Rosaceae	*Prunus*	*virginiana*	Bitter-berry, Chokecherry, Virginia bird cherry and Western chokecherry	USA[29, 57]
Rosaceae	*Rosa*	*(polyantha) multiflora*	Baby rambler	USA[29, 57]
Rosaceae	*Rosa*	*amblyotis*		Russia[43]
Rosaceae	*Rosa*	*canina*	Dog rose	[57]
Rosaceae	*Rosa*	*chinensis*	China rose	USA[29, 57]
Rosaceae	*Rosa*	*corymbifera*		Russia[43]
Rosaceae	*Rosa*	*davidii*		Russia[43]
Rosaceae	*Rosa*	*davurica*		Russia[43]
Rosaceae	*Rosa*	*rugosa*	Rugosa rose	Russia[43] USA [50]
Rosaceae	*Rosa*	*sicula*		Russia[43]
Rosaceae	*Rosa*	*sp.*	Rose	New Zealand[28] USA (Hawaii)[11] USA [57]
Rosaceae	*Rosa*	*sp. X*	Hybrid tea rose	USA[57]
Rosaceae	*Rosa*	*xanthina*		Russia[43]
Rosaceae	*Rubus*	*× loganobaccus*	Loganberry	[57]

Rosaceae	*Rubus*	*alleghaniensis*	High blackberry	USA[57]
Rosaceae	*Rubus*	*argutus*	Hawaiian blackberry*	USA (Hawaii)[10]
Rosaceae	*Rubus*	*canadensis*	Millspaugh's blackberry	USA[57]
Rosaceae	*Rubus*	*fruiticosus*	Blackberry	[5]
Rosaceae	*Rubus*	*idaeus*	Red raspberry	Finland[21] [5]
Rosaceae	*Rubus*	*idaeus var. aculeatissimus*	Wild raspberry	[57]
Rosaceae	*Rubus*	*occidentalis*	Black raspberry	USA[29,57]
Rosaceae	*Rubus*	*parviflorus*	Thimbleberry	USA[13]
Rosaceae	*Rubus*	*penetrans*	Prickly Florida blackberry	USA (Hawaii)[57]
Rosaceae	*Rubus*	*procerus*	Himalaya berry	USA [13]
Rosaceae	*Rubus*	*sp.*	Rubus	Netherlands[15] Portugal[63], Spain[63]
Rosaceae	*Rubus*	*sp.*	Himalaya berry	UK[57]
Rosaceae	*Rubus*	*sp.*	Blackberry	[57]
Rosaceae	*Rubus*	*sp.*	Raspberry	[57]
Rosaceae	*Rubus*	*ulmifolius*	Elmleaf blackberry	Italy[65]
Rosaceae	*Rubus*	*vitifolius*	California blackberry	USA[13]
Rosaceae	*Sanguisorba*	*minor*	Salad burnet	France[57]
Rosaceae	*Sanguisorba*	*sp.*		Italy[16]
Rosaceae	*Sorbaria*	*pallasii*		Russia[43]
Rosaceae	*Sorbaria*	*sorbifolia*	False spiraea	Russia[43]
Rosaceae	*Sorbus*	*aucuparia*	European mountain ash	Europe[57] Netherlands[15]
Rosaceae	*Spirea*	*alba*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*betulifolia*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*cana*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*chamaedrifolia*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*densiflora*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*douglasii*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*japonica*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*latifolia*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*lucida*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*menziesii*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*nipponica*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*salicifolia*	Meadowsweet	Russia[43]
Rosaceae	*Spirea*	*sp.*	Meadowsweet	Germany[57] USA [29, 57]
Rosaceae	*Spirea*	*Spiraea × bumalda*	Meadowsweet	Russia[43]

Rosaceae	*Spirea*	*Spiraea × bumalda Burv. "Shraederii"*	Meadowsweet	Russia[43]
Rubiaceae	*Coprosma*	*ernodeoides var. typica*	Black fruited coprosma (N)	USA (Hawaii)[10]
Rubiaceae	*Coprosma*	*rhynchocarpa*	Woodland mirrorplant (N)	USA (Hawaii)[10]
Rubiaceae	*Crucianella*	*maritima*		Portugal[63], Spain[63]
Rubiaceae	*Galium*	*album*	White bedstraw	UK[3]
Rubiaceae	*Galium*	*aparine*	Cleavers	UK[3] USA[13, 57]
Rubiaceae	*Galium*	*asprellum*	Rough bedstraw	UK[3]
Rubiaceae	*Galium*	*mollugo*	Whorled bedstraw	Europe[57] UK[3]
Rubiaceae	*Galium*	*palustre*	Marsh bedstraw	Finland[21] UK[3]
Rubiaceae	*Galium*	*sp.*	Bedstraw	[57] Italy[16]
Rubiaceae	*Galium*	*syilvaticum*	Wood bedstraw	USA[57]
Rubiaceae	*Galium*	*verum*	Yellow bedstraw	USA[13, 57]
Rubiaceae	*Houstonia*	*caerulea*	Azure bluet, Quaker ladies	USA[57]
Rubiaceae	*Sherardia*	sp.		Italy[16]
Rutaceae	*Citrus*	*sinensis*	Sweet orange	Italy[8] Nigeria[17]
Rutaceae	*Citrus*	*unshiu*	Satsuma mandarin	USA (Hawaii)[10]
Salicaceae	*Populus*	*nigra*	Lombardy poplar	Turkey[48] USA[61]
Salicaceae	*Populus*	*tremula*	European aspen	[57]
Salicaceae	*Populus*	sp.	poplar	Iran[68]
Salicaceae	*Salix*	*alba*	White willow	[57]
Salicaceae	*Salix*	*babylonica*	Weeping willow	Turkey[48]
Salicaceae	*Salix*	*caprea*	Goat willow	[57]
Salicaceae	*Salix*	*cinerea*	Grey willow	[57]
Salicaceae	*Salix*	*fragilis*	Brittle willow	[57]
Salicaceae	*Salix*	*repens*	Creeping willow	[57]
Salicaceae	*Salix*	*sp.*	Willow	Germany[29] [57]
Salicaceae	*Salix*	*triandra*	Almond-leaved willow	Europe[57]
Salicaceae	*Salix*	*viminalis*	Basket willow	Europe[57] UK[3]
Sapindaceae	*Acer*	*campestre*	Field maple	Italy[65]
Sapindaceae	*Acer*	*sp.*	Maple	USA [57]
Sapindaceae	*Blighia*	*sapida*	Ackee	Nigeria[17]
Sapotaceae	*Sideroxylon*	*sp.*	Bully trees (N)	USA (Hawaii)[11]
Sarraceniaceae	*Darlingtonia*	*sp.*	Pitcherplant	USA[61]
Saxifragaceae	*Heuchera*	*sanguinea*	Coral bells	USA[13]
Scrophulariaceae	*Scrophularia*	*californica*	Figwort	USA[13]

Scrophulariaceae	*Verbascum*	*sp.*	Mullen	Italy[16] USA[57, 61]
Scrophulariaceae	*Verbascum*	*thapsus*	Great mullen	USA[57]
Solanaceae	*Capsicum*	*sp.*	Cayenne pepper	France[57]
Solanaceae	*Cestrum*	*elegans (purpureum)*	Bastard Jasmine	USA[13]
Solanaceae	*Nicotiana*	*tabacum*	Tobacco	[5]
Solanaceae	*Physalis*	*peruviana*	Cape gooseberry	USA (Hawaii)[10]
Solanaceae	*Solanum*	*dulcamara*	Woody nightshade	Netherlands[15] USA[61]
Solanaceae	*Solanum*	*lycopersicum*	Tomato	USA (Hawaii)[11]
Solanaceae	*Solanum*	*nigrum*	Woody nightshade	Netherlands[15]
Solanaceae	*Solanum*	*sp.*	Solanum	Italy[16] USA[61]
Solanaceae	*Solanum*	*tuberosum*	Potato	[57] Iran[68]
Tamaricaceae	*Tamarix*	Sp.		Iran[68]
Thymelaeaceae	*Wikstroemia*	*phillyreifolia*	False ohelo (N)	USA (Hawaii)[10]
Tropaeolaceae	*Tropaeolum*	*sp.*	Nasturtium	USA (Hawaii)[11]
Ulmaceae	*Ulmus*	*glabra*	Wych elm, Scotch elm or Scots elm	Europe[57]
Ulmaceae	*Ulmus*	*sp.*	Dwarf elm	USA[57]
Urticaceae	*Parietaria*	*sp.*	Parietaria	France[57]
Urticaceae	*Pipturus*	*sp.*	Mamaki? (N)	USA (Hawaii)[11]
Urticaceae	*Urtica*	*dioica*	Nettle	England[58] Italy[9]
Urticaceae	*Urtica*	*sp.*	Nettle	USA[57]
Verbenaceae	*Lantana*	*camara*	Lantana	Nigeria[17]
Verbenaceae	*Stachytarpheta*	*sp.*	Porterweeds	USA (Hawaii)[57]
Verbenaceae	*Verbena*	*litoralis*	Seashore vervain	USA (Hawaii)[10]
Verbenaceae	*Verbena*	*rigida*	Slender vervain	USA[13]
Violaceae	*Melicytus*	*ramiflorus*	Māhoe	New Zealand[33]
Violaceae	*Viola*	*odorata*	Sweet violet	USA[13]
Violaceae	*Viola*	*palustris*	Marsh Violet	Netherlands[15]
Violaceae	*Viola*	*tricolor*	Garden pansy	USA[57]
Vitaceae	*Parthenocissus*	*quinquefolia*	Virginia creeper	Netherlands[15]
Vitaceae	*Vitis*	*sp.*	Grapevine	Germany[57] New Zealand[44]
Vitaceae	*Vitis*	*vinifera*	Grapevine cv. Cabernet Sauvignon	Italy[8, 32, 65]
Vitaceae	*Vitis*	*vinifera cv. Emperor*	Grapevine	USA[13]
Zingiberaceae	*Hedychium*	*coronarium*	White garland-lily	USA (Hawaii)[11]

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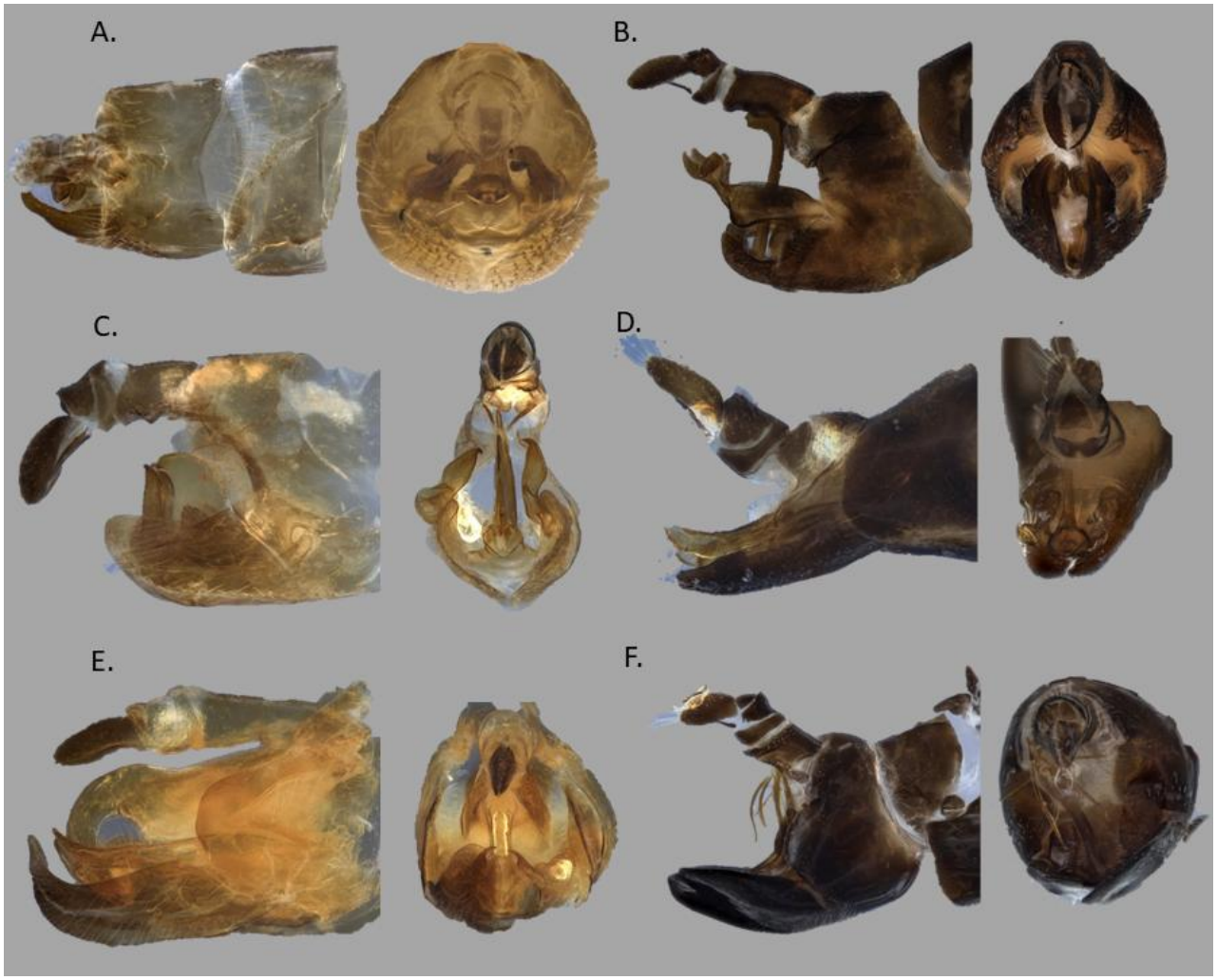
8.2 Appendix 8.2 Male genitalia of select native Australian genera of Aphrophoridae

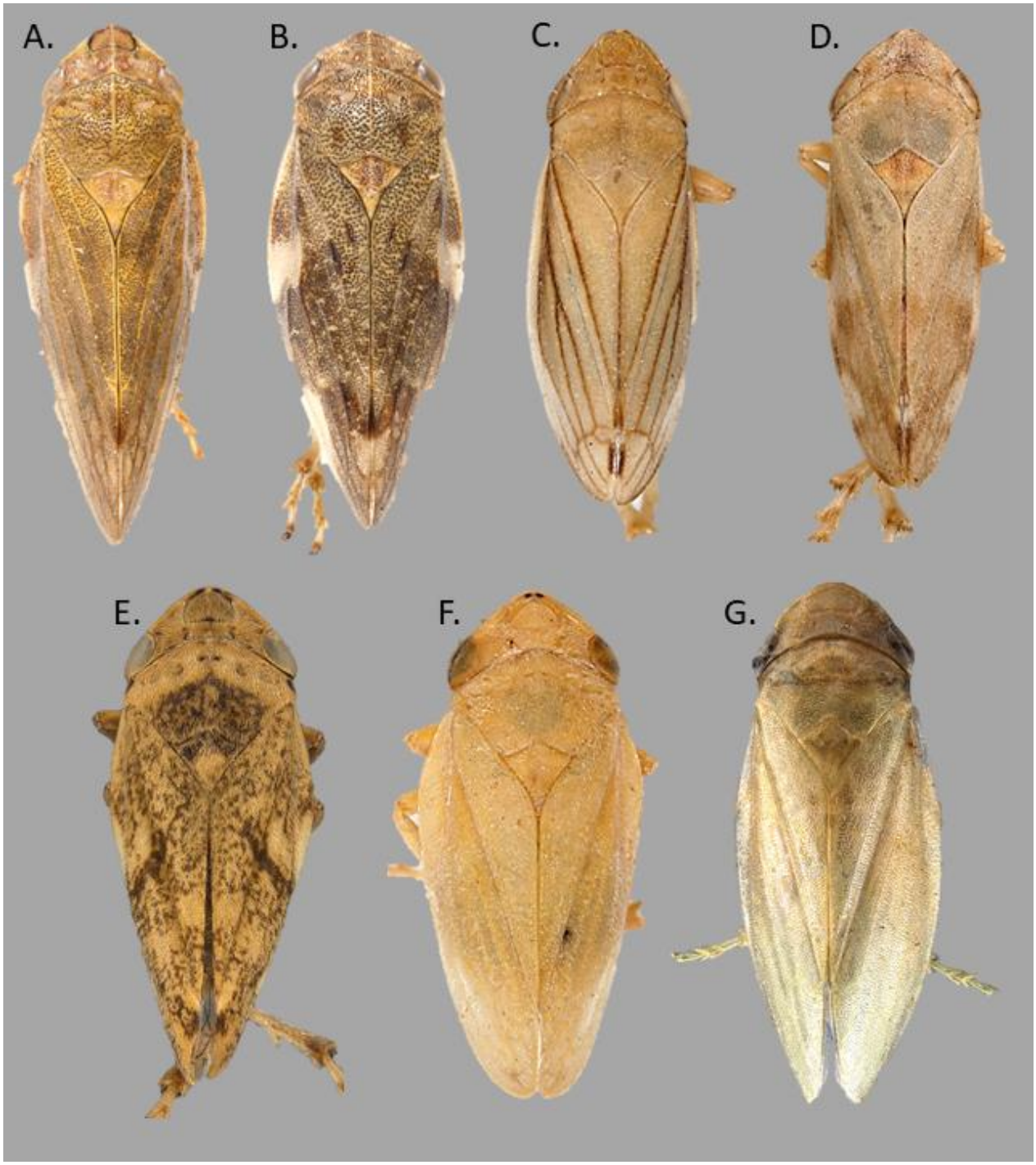
Appendix 8.2: Male genitalia of some native Australian genera of Aphrophoridae. **A.** *Anyllis sp.,* **B.** *Amarusa australis*, **C.** *Philagra* sp., **D.** *Clovia eximia*, **E.** *Carystoterpa vagans*, and **F.** *Bathyllus albicinctus*.

Plate adapted from photographs by © Pia Scanlon, Western Australian Agricultural Authority 2020.



8.3 Appendix 8.3 Dorsal habitus images of predominantly exotic Aphrophoridae

Appendix 8.3: Dorsal habitus images of predominantly exotic species of Aphrophoridae. **A.** *Aphrophora salicina,* **B.** *Aphrophora alni*, **C.** *Neophilaenus lineatus*, **D.** *Neophilaenus campestris*, **E.** *Philaenus spumarius*, **F.** *Philaenus elbursianus*, and **G.** *Basilioterpa pallida*. Plate adapted from © Pia Scanlon, Western Australian Agricultural Authority 2020 (Figures E. & G.), and © Mozaffarian & Wilson 2015 (Figures A, B, C, D. & F.), used with permission Sept 2020.



8.4 Appendix 8.4 The full copyright and source details of images from Figure 5

Figure 5. The variety of colour morphs of *Philaenus spumarius*. All images have had backgrounds and legs removed, and have been rotated to similar angles for comparative purposes. The following pertains to copyright and the sources of the original images:
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M. © AfroBrazilian 2019 - (free under creative commons) Available at https://commons.wikimedia.org/wiki/File:Philaenus_spumarius_26.JPG
N. Judy Gallagher 2019, (free under creative commons) Available at BugGuide.Net
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9 DIAGNOSTICS PROCEDURES TO SUPPORT SURVEILLANCE

9.1 Introduction

There are limited in-field diagnostic procedures to support specific surveillance for *Philaenus spumarius*. In Australia it is likely that *P. spumarius* would occur as nymphs or adults throughout spring to late autumn in temperate and cool regions, as inferred through the biology of *P. spumarius* in New Zealand, which is the only recorded locality of the spittlebug in the Southern Hemisphere (CABI 2020a). This timing is, however, similar to many native temperate Cercopoidea species. Given the wide host range of *P. spumarius,* viable options for surveillance include using sentinel plants at entry points (as described in Section 3). For targeted surveillance, efforts would be more efficiently employed by training primary producers of known host plants, such as olives and grapes to identify both the signs of *Xylella*, and the bubble nests of spittlebug nymphs both on these crops, and on the weeds under, or bordering, crops. *P. spumarius* infestations are usually detected by the presence of nymphs in these bubble nests along the stems, at leaf nodes and on fresh shoots (see Fig. 1 within NDP).

9.2 Sampling

A variety of in field sampling techniques are detailed in section 3.2. A lure for *P. spumarius* has not been developed to date. In the event of a suspected incursion, sweeping and sticky traps are the best collection methods as the primary objective for diagnosis is to capture a male *P. spumarius*. An indication of the potential presence of *P. spumarius* is the presence of bubble nests on host species that are not favoured by native Australian Cercopoidea such as lavender, grapes, onion and carrots. Nymphs should be carefully removed from the bubble nest and placed in 95-100% ethanol for further molecular analysis in the laboratory. The nymphs and bubble nests of *P. spumarius* cannot be distinguished from the bubble nests of native Australian Aphrophoridae spittlebugs, therefore if nests are detected, they should be investigated further through molecular work or rearing the nymphs to adults. It should also be considered that *P. spumarius* bubble nests will not be present year-round but are most likely present in spring and summer in temperate regions.

9.3 In-field Tests

There are currently no available in-field tests for *P. spumarius* and, given the number of colour morphs (see Fig. 5) and the presence of similar looking Australian native Cercopoidea, field diagnosis to species should not be attempted. Bubble nests of both native and introduced spittlebugs cannot be identified to species, and are likely to be present around the same time of year in temperate regions (spring-summer).