2017 First consultation

1 July – 30 September 2017

Compiled comments for Draft diagnostic protocol for *Bactrocera dorsalis* Complex

Summary comments

Name	Summary
Cuba	No hay comentarios para el PD
EPPO Σ	Finalised by the EPPO Secretariat on behalf of its 51 Member Countries.
European Union	Comments finalised by the European Commission on behalf of the EU and its 28 member States on 29/09/2017.
Samoa	no further comments
South Africa	No comments from the National Plant Protection Organisation of South Africa.

#	Para	Text	Comment
1	G	(General Comment)	**Cameroon** Ce protocole est complet, détaillé et richement illustré. Il apportera un outil supplementaire pour soutenir le travail des ONPV qui font face à ce fléau, notamment le Cameroun. *Category : TECHNICAL*
2	G	(General Comment)	**Myanmar** Myanmar has B.dorsalis & B.carambolae only, the rest spp. are absent in Myanmar. *Category : SUBSTANTIVE*
3	G	(General Comment)	**Peru** We agree with the Draft Annex to ISPM 27 – Bactrocera dorsalis complex (2006-026) *Category : TECHNICAL*
4	G	(General Comment)	**United States of America** The United States has no comments on this draft standard. *Category : SUBSTANTIVE*
5	G	(General Comment)	**European Union** Bactrocera dorsalis (Tephritidae). Systematic Entomology DOI: 10.1111/syen.12250. *Category : SUBSTANTIVE*
6	G	(General Comment)	**Swaziland** the diagnostic protocol (DP) is appropriate *Category : SUBSTANTIVE*
7	G	(General Comment)	**Canada** Canada supports the draft annex to ISPM 27 - Bactrocera dorsalis complex

#	Para	Text	Comment
			Category : SUBSTANTIVE
8	G	(General Comment)	**Nepal** It is perfect. I have no any comment *Category : EDITORIAL*
9	G	(General Comment)	**Guyana** Guyana has no objection to this Annex and considers it an important one. *Category : SUBSTANTIVE*
10	G	(General Comment)	**Panama** Panama has no comments on this document. *Category : EDITORIAL*
11	G	(General Comment)	**EPPO** Bactrocera dorsalis (Tephritidae). Systematic Entomology DOI: 10.1111/syen.12250 *Category : SUBSTANTIVE*
12	G	(General Comment)	**Tajikistan** no comments *Category : SUBSTANTIVE*
13	G	(General Comment)	**Tajikistan** No comments *Category : SUBSTANTIVE*
14	G	(General Comment)	**Bahamas** The draft demonstrates that a comprehensive multidisciplinary approach has been applied to resolve the proposed new classification of B. dorsalis complex. The Bahamas therefore supports the adoption of this diagnostic protocol. *Category : SUBSTANTIVE*
15	G	(General Comment)	**Thailand** agree with the proposed draft DP for Bactrocera dorsalis complex *Category : SUBSTANTIVE*
16	G	(General Comment)	**Lao People's Democratic Republic** Lao PDR agreed with this drafted annex ISPM 27. *Category : SUBSTANTIVE*
17	G	(General Comment)	**Honduras** HONDURAS NO TIENE COMENTARIOS *Category : TECHNICAL*
18	G	(General Comment)	**Nicaragua** Nicaragua considera que es necesario que para la definición del Protocolo de Diganóstico sobre clasificación taxonómica de Bactrocera dorsalis se consideren todas las variantes del insecto para no hablar de complejo B. dorsalis, sino designar características propias de cada individuo que permita su identificación por separado. *Category : TECHNICAL*

#	Para	Text	Comment
19	G	(General Comment)	**China** Up to now, the taxonomic status of Bactrocera dorsalis complex is still a scientific issue with obvious arguments between traditional morphological diagnosis and current genetic diagnosis. Among the 6 species of Bactrocera dorsalis complex (B. dorsalis, B. carambolae, B. caryeae, B. kandiensis, B. occipitalis and B. pyrifoliae) in this Draft Annex, only B. pyrifoliae can be trapped by CUE and with different morphological characters, B. dorsalis and the other 4 species can be trapped by ME and with so similar morphological characteristics. How about the exactly taxonomic status of the 6 species of B. dorsalis complex? Are they different species or some of them are the synonym of B. dorsalis? This scientific issue has been paid more attention in the Tephritidae field in the world and some teams are working on it. In the meantime, the morphological diagnostic characters among the 6 species of Bactrocera dorsalis complex are very difficult to operate practically. The current version of Draft Annex is not practical especially for most members of IPPC. *Category : SUBSTANTIVE*
20	G	(General Comment)	**Algeria** No figure is illustrated on the conventional protocol of identification of the flie *Category : TECHNICAL*
21	G	(General Comment)	**PPPO** I have no comments to make on this draft ISPM *Category : EDITORIAL*
22	1	**Draft Annex to ISPM 27 – *Bactrocera dorsalis* complex (2006-026)** **Comment: We agree with the draft.**	**Nigeria** *Category : TECHNICAL*
23	28	Given that a new classification has been proposed but not adopted by all experts, synonyms are currently treated as subjective (ICZN rules). As IPPC is supposed to develop DPs for recognized species, the DP is not intended to instruct on revision debates.	**European Union** Whether a junior synonym is objective or subjective has nothing to do with general adoption of the synonymization or not. ICZN states that objective synonyms are those for which the name bearing type is the same. Even if there is general consensus that a particular name is a junior synonym, it remains a subjective synonym if the types are different (which is the case for B. dorsalis, B. papayae, B. philippinensis and B. invadens). *Category : SUBSTANTIVE*
24	28	Given that a new classification has been proposed but not adopted by all experts, synonyms are currently treated as subjective (ICZN rules). As IPPC is supposed to develop DPs for recognized species, the DP is not intended to instruct on revision debates.	**EPPO** Synonyms are currently treated as subjective (ICZN rules)" Whether a junior synonym is objective or subjective has nothing to do with general adoption of the synonymization or not. ICZN states that objective synonyms are those for which the name bearing type is the same. Even if there is general consensus that a

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			particular name is a junior synonym, it remains a subjective synonym if the types are different (which is the case for B. dorsalis, B. papayae, B. philippinensis and B. invadens) *Category : SUBSTANTIVE*
25	39	Fruit flies of the family Tephritidae represent an economically important insect group with a worldwide distribution. The biology of these fruit flies is dependent on host plants that can serve as mating locations, oviposition sites for eggs, and nutrient resources for developing larvae. The genus *Bactrocera* Macquart consists of over 650 described species that are distributed mostly in regions of Asia and Australasia and subtropical islands of the southern Pacific Ocean (Drew and Romig, 2013). Within the genus is a group of flies named the *Bactrocera dorsalis* complex (Drew and Hancock, 1994; Drew, 2004; Clark *et al*., 2005). This complex comprises 85 described species (Vargas *et al.*, 2015) that share a very similar appearance, but the complex as a whole does not represent a monophyletic lineage and is merely a group of convenience (Leblanc *et al.*, 2015). The complex is named after one of its member species, *Bactrocera dorsalis* (Figure 1) which is a polyphagous pest of commercial fruits. Several other species in the complex are also recognized as pests, based on plant host use and pest records (White and Elson-Harris, 1992; Clarke *et al*., 2005; Vargas *et al.*, 2015; Plant Health Australia, 2016).	**Kenya** Include Africa in the distibution as we have several species of Bactrocera are already established in Africa e.g. B. zonata, B. dorsalis (invadens), B. curcurbitacea *Category : TECHNICAL*
26	39	Fruit flies of the family Tephritidae represent an economically important insect group with a worldwide distribution. The biology of these fruit flies is dependent on host plants that can serve as mating locations, oviposition sites for eggs, and nutrient resources for developing larvae. The genus *Bactrocera* Macquart consists of over 650 described species that are distributed mostly in regions of Asia and Australasia and subtropical islands of the southern Pacific Ocean (Drew and Romig, 2013)<u>, and Africa</u>. Within the genus is a group of flies named the *Bactrocera dorsalis* complex (Drew and Hancock, 1994; Drew, 2004; Clark *et al*., 2005). This complex comprises 85 described species (Vargas *et al.*, 2015) that share a very similar appearance, but the complex as a whole does not represent a monophyletic lineage and is merely a group of convenience (Leblanc *et al.*, 2015). The complex is named after one of its member species, *Bactrocera dorsalis* (Figure 1) which is a polyphagous pest of commercial fruits. Several other species in the complex are also recognized as pests, based on plant host use and pest records (White and Elson-Harris, 1992; Clarke *et al*., 2005; Vargas *et al.*, 2015; Plant Health Australia, 2016).	**Kenya** General comment *Category : TECHNICAL*
27	40	The scope of the current protocol is to diagnose adult fruit flies for ~~six~~ <u>some</u> species of the *Bactrocera dorsalis* complex that are found in commercial fruits and vegetables	**Viet Nam**

#	Para	Text	Comment
		associated with international trade. These species are: *B. dorsalis*, *B. carambolae*, *B. caryeae*, *B. kandiensis*, *B. occipitalis* and *B. pyrifoliae*. Distributions of these species are mapped with their pest status and invasion history by Vargas *et al*. (2015).	*Category : TECHNICAL*
28	41	A lack of characters that can be used reliably to distinguish *B. dorsalis* from two other species (i.e. *B. papayae* Drew and Hancock, 1994, and *B. invadens* Drew *et al*., 2005) has resulted in debate regarding the valid taxonomy of the species (Clarke *et al*., 2005; Chen and Hui, 2007; Schutze *et al*., 2015a, 2015b; Drew & Romig, 2016~~)~~, Schutze et al. 2017). These three species have been treated as members of a sibling species complex, not to be confused with the *Bactrocera dorsalis* complex (Clarke and Schutze, 2014). It is not possible to reliably distinguish among these three species because an accurate identification requires both evaluation of species distribution information and analysis of morphological characters that are not discrete for the species. Species distribution information may not be reliable when examining specimens collected outside its known range. Published molecular data cannot distinguish these species (Schutze *et al*., 2015a). In a review of available evidence, Schutze *et al*. (2015a) concluded that these three species are in fact a single biological species called *Bactrocera dorsalis*. Drew and Romig (2016) disagree with that revision. In this protocol, the three species are collectively treated as *B. dorsalis sensu lato*.	**European Union** Drew and Romig (2016) disagree with that revision." But see Schutze et al. 2017. *Category : TECHNICAL*
29	41	A lack of characters that can be used reliably to distinguish *B. dorsalis* from two other species (i.e. *B. papayae* Drew and Hancock, 1994, and *B. invadens* Drew *et al*., 2005) has resulted in debate regarding the valid taxonomy of the species (Clarke *et al*., 2005; Chen and Hui, 2007; Schutze *et al*., 2015a, 2015b; Drew & Romig, 2016). These three species have been treated as members of a sibling species complex, not to be confused with the *Bactrocera dorsalis* complex (Clarke and Schutze, 2014). It is not possible to reliably distinguish among these three species because an accurate identification requires both evaluation of species distribution information and analysis of morphological characters that are not discrete for the species. Species distribution information may not be reliable when examining specimens collected outside its known range. Published molecular data cannot distinguish these species (Schutze *et al*., 2015a). In a review of available evidence, Schutze *et al*. (2015a) concluded that these three species are in fact a single biological species called *Bactrocera dorsalis*. Drew and Romig (2016) disagree with that revision. In this protocol, the three species are collectively treated as *B. dorsalis sensu lato*.	**Swaziland** a clarity is sought whether we still regard B. invadens and B. dorsalis as two separate species on just as one spp *Category : TECHNICAL*
30	41	A lack of characters that can be used reliably to distinguish *B. dorsalis* from two other	**EPPO**

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		species (i.e. *B. papayae* Drew and Hancock, 1994, and *B. invadens* Drew *et al*., 2005) has resulted in debate regarding the valid taxonomy of the species (Clarke *et al.*, 2005; Chen and Hui, 2007; Schutze *et al.*, 2015a, 2015b; Drew & Romig, 2016, Schutze et al. 2017). These three species have been treated as members of a sibling species complex, not to be confused with the *Bactrocera dorsalis* complex (Clarke and Schutze, 2014). It is not possible to reliably distinguish among these three species because an accurate identification requires both evaluation of species distribution information and analysis of morphological characters that are not discrete for the species. Species distribution information may not be reliable when examining specimens collected outside its known range. Published molecular data cannot distinguish these species (Schutze *et al.*, 2015a). In a review of available evidence, Schutze *et al*. (2015a) concluded that these three species are in fact a single biological species called *Bactrocera dorsalis*. Drew and Romig (2016) disagree with that revision. In this protocol, the three species are collectively treated as *B. dorsalis sensu lato*.	Drew and Romig (2016) disagree with that revision." But see Schutze et al. 2017. *Category : TECHNICAL*
31	41	A lack of characters that can be used reliably to distinguish *B. dorsalis* from two other species (i.e. *B. papayae* Drew and Hancock, 1994, and *B. invadens* Drew *et al*., 2005) has resulted in debate regarding the valid taxonomy of the species (Clarke *et al.*, 2005; Chen and Hui, 2007; Schutze *et al.*, 2015a, 2015b; Drew & Romig, 2016). These three species have been treated as members of a sibling species complex, not to be confused with the *Bactrocera dorsalis* complex (Clarke and Schutze, 2014). It is not possible to reliably distinguish among these three species because an accurate identification requires both evaluation of species distribution information and analysis of morphological characters that are not discrete for the species. Species distribution information may not be reliable when examining specimens collected outside its known range. Published molecular data cannot distinguish these species (Schutze *et al.*, 2015a). In a review of available evidence, Schutze *et al*. (2015a) concluded that these three species are in fact a single biological species called *Bactrocera dorsalis*. Drew and Romig (2016) disagree with that ~~revision~~revision and reversed the synonymy; however, Schutze et al. (2017) published a rebuttal to Drew & Roming (2016) that supports the synonymy by Schutze et al. (2015). In this protocol, the three species are collectively treated as *B. dorsalis sensu lato*.	**Australia** *Category : TECHNICAL*
32	45	~~*Bactrocera*~~ *B. dorsalis s.l.* attacks over 270 plant species (Vargas *et al*. 2015) in over 50 families of commercial fruits and wild fruits (CABI, 2016). It has the largest species range of the six pests included in the protocol, and is found on some islands in	**European Union** Please see end of paragraph 41. *Category : EDITORIAL*

#	Para	Text	Comment
		the Pacific Ocean, and most of continental Africa (sub-Saharan countries) in addition to its original Asian range (Drew and Hancock, 1994; Drew *et al*., 2005; White, 2006; Drew and Romig, 2013; Schutze *et al*., 2015a, b).	
33	45	~~*Bactrocera*~~ *B. dorsalis s.l.* attacks over 270 plant species (Vargas *et al*. 2015) in over 50 families of commercial fruits and wild fruits (CABI, 2016). It has the largest species range of the six pests included in the protocol, and is found on some islands in the Pacific Ocean, and most of continental Africa (sub-Saharan countries) in addition to its original Asian range (Drew and Hancock, 1994; Drew *et al*., 2005; White, 2006; Drew and Romig, 2013; Schutze *et al*., 2015a, b).	**EPPO** Please see end of paragraph 41. *Category : EDITORIAL*
34	52	**Taxonomic position:** Insecta, Diptera, Tephritidae, Dacinae, *Bactrocera*	**Colombia** Se requiere incluir la subfamilia en la posición taxonómica. *Category : TECHNICAL*
35	53	The species included in the *Bactrocera dorsalis* complex are in the subgenus *Bactrocera* (*Bactrocera*). According to ICZN (1999), three species are treated as subjective synonyms under *Bactrocera dorsalis s.l.*: *Bactrocera papayae*, *Bactrocera invadens* and *Bactrocera philippinensis*. Drew and Romig ~~(2013) placed~~ (2013), *Bactrocera (B) philippinensis* and *Bactrocera conformic* are synonym~~*B. philippinensis* as a synonym of *B. papayae*~~. Revision by Schutze *et al*. (2015a) places *B. invadens* and *B. papayae* as junior synonyms of *B. dorsalis*. Drew and Romig (2016) provide an argument for treating these as separate species. Note that *Bactrocera invadens* was not formally placed into the *Bactrocera dorsalis* complex by Drew *et al*. ~~(2013)~~(2013) and Tsuruta &White referred to species *Bactrocera invadens* as to *Bactrocera dorsalis* (Drew and Romig, 2013), but based on Schutze *et al*. (2015a) is considered a sibling species of, or synonym of, *Bactrocera dorsalis*. The current protocol treats these names (*B. papayae*, *B. invadens* and *B. philippinensis*) as part of *Bactrocera dorsalis s.l.*	**Viet Nam** Tsuruta &White referred to species B. invadens as to B. dorsalis *Category : TECHNICAL*
36	53	The species included in the *Bactrocera dorsalis* complex are in the subgenus *Bactrocera* (*Bactrocera*). According to ICZN (1999), three species are treated as subjective synonyms under ~~*Bactrocera*~~*B. dorsalis s.l.*: ~~*Bactrocera*~~*B. papayae*, ~~*Bactrocera*~~*B. invadens* and ~~*Bactrocera*~~*B. philippinensis*. Drew and Romig (2013) placed *B. philippinensis* as a synonym of *B. papayae*. Revision by Schutze *et al*. (2015a) ~~places~~ placed *B. invadens* and *B. papayae* as junior synonyms of *B. dorsalis*. Drew and Romig (2016) provide an argument for treating these as separate species. Note that *Bactrocera invadens* was not formally placed into the *Bactrocera dorsalis* complex by Drew *et al*. (2013), but based on Schutze *et al*. (2015a) is considered a	**European Union** *Category : EDITORIAL*

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		sibling species of, or synonym of, *Bactrocera dorsalis*. The current protocol treats these names (*B. papayae*, *B. invadens* and *B. philippinensis*) as part of *Bactrocera dorsalis s.l.*	
37	53	The species included in the *Bactrocera dorsalis* complex are in the subgenus *Bactrocera* (*Bactrocera*). According to ICZN (1999), three species are treated as subjective synonyms under *Bactrocera dorsalis s.l.*: *Bactrocera papayae*, *Bactrocera invadens* and *Bactrocera philippinensis*. Drew and Romig (2013) placed *B. philippinensis* as a synonym of *B. papayae*. Revision by Schutze *et al*. (2015a) places *B. invadens* and *B. papayae* as junior synonyms of *B. dorsalis*. Drew and Romig (2016) provide an argument for treating these as separate species. Note that *Bactrocera invadens* was not formally placed into the *Bactrocera dorsalis* complex by Drew *et al*. (2013), but based on Schutze *et al*. (2015a) is considered a sibling species of, or synonym of, *Bactrocera dorsalis*. The current protocol treats these names (*B. papayae*, *B. invadens* and *B. philippinensis*) as part of *Bactrocera dorsalis s.l.*	**European Union** Note that Bactrocera invadens was not formally placed into the Bactrocera dorsalis complex by Drew et al. (2013)," This is a recent replacement as Drew et al 2005, 2008 did place it in the dorsalis complex. *Category : SUBSTANTIVE*
38	53	The species included in the *Bactrocera dorsalis* complex are in the subgenus *Bactrocera* (*Bactrocera*). According to ICZN (1999), three species are treated as subjective synonyms under *Bactrocera dorsalis s.l.*: *Bactrocera papayae*, *Bactrocera invadens* and *Bactrocera philippinensis*. Drew and Romig (2013) placed *B. philippinensis* as a synonym of *B. papayae*. Revision by Schutze *et al*. (2015a) places *B. invadens* and *B. papayae* as junior synonyms of *B. dorsalis*. Drew and Romig (2016) provide an argument for treating these as separate species. Note that *Bactrocera invadens* was not formally placed into the *Bactrocera dorsalis* complex by Drew *et al*. (2013), but based on Schutze *et al*. (2015a) is considered a sibling species of, or synonym of, *Bactrocera dorsalis*. The current protocol treats these names (*B. papayae*, *B. invadens* and *B. philippinensis*) as part of *Bactrocera dorsalis s.l.*	**EPPO** Note that Bactrocera invadens was not formally placed into the Bactrocera dorsalis complex by Drew et al. (2013)," This is a recent replacement as Drew et al 2005, 2008 did place it in the dorsalis complex. *Category : SUBSTANTIVE*
39	53	The species included in the *Bactrocera dorsalis* complex are in the subgenus *Bactrocera* (*Bactrocera*). According to ICZN (1999), three species are treated as subjective synonyms under ~~*Bactrocera*~~*B.* ~~*-dorsalis*~~dorsalis *s.l.*: ~~*Bactrocera*~~*B.* *-papayae*, ~~*Bactrocera*~~*B.* *-invadens* and ~~*Bactrocera*~~*B.* *-philippinensis*. Drew and Romig (2013) placed *B. philippinensis* as a synonym of *B. papayae*. Revision by Schutze *et al*. (2015a) ~~places~~ placed *B. invadens* and *B. papayae* as junior synonyms of *B. dorsalis*. Drew and Romig (2016) provide an argument for treating these as separate species. Note that *Bactrocera invadens* was not formally placed into the *Bactrocera dorsalis* complex by Drew *et al*. (2013), but based on Schutze *et al*. (2015a) is considered a sibling species of, or synonym of, *Bactrocera dorsalis*. The current protocol treats	**EPPO** *Category : EDITORIAL*

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40	53	The species included in the *Bactrocera dorsalis* complex are in the subgenus *Bactrocera* (*Bactrocera*). According to ICZN (1999), three species are treated as subjective synonyms under *Bactrocera dorsalis s.l.*: *Bactrocera papayae*, *Bactrocera invadens* and *Bactrocera philippinensis*. Drew and Romig (2013) placed *B. philippinensis* as a synonym of *B. papayae*. Revision by Schutze *et al*. (2015a) places *B. invadens* and *B. papayae* as junior synonyms of *B. dorsalis*. Drew and Romig (2016) provide an argument for treating these as separate species; however Schutze et al. (2017) published a rebuttal of this arguement. Note that *Bactrocera invadens* was not formally placed into the *Bactrocera dorsalis* complex by Drew *et al*. (2013), but based on Schutze *et al*. (2015a) is considered a sibling species of, or synonym of, *Bactrocera dorsalis*. The current protocol treats these names (*B. papayae*, *B. invadens* and *B. philippinensis*) as part of *Bactrocera dorsalis s.l.*	**Australia** *Category : TECHNICAL*
41	66	*Dacus* (*Bactrocera*) *caryeae* Kapoor, 1971; Hardy, 1977 *Chaetodacus ferrugineus incises* Bezzi, 1916	**Viet Nam** Chaetodacus ferrugineus incises Bezzi, 1916 (Drew and Romig, 2013) *Category : TECHNICAL*
42	67	*Bactrocera* (*Bactrocera*) *dorsalis s.l.* (Hendel, 1912)	**Viet Nam** According to para 53 *Category : TECHNICAL*
43	81	*Bactrocera philippinensis* Drew and Hancock, 1994 (subjective) *Bactrocera conformic*	**Viet Nam** Following to Drew and Romig 2013 Bactrocera (B) philippinensis and Bactrocera conformic are synonym *Category : TECHNICAL*
44	107	Fruit flies of the genus *Bactrocera* are detected mainly by male lure trap or in fruits. Only male adult fruit flies are captured by male lure trapping, while all immature stages such as eggs (Figure 2(a)), early to final instar larvae (Figures 2(b) to (d)), and ~~pupae and~~ puparia and pupae (Figures 2(e) to (f)) can be found during inspection of fruits.	**European Union** Proper order (please see figures 2(e) and (f)). *Category : EDITORIAL*
45	107	Fruit flies of the genus *Bactrocera* are detected mainly by male lure trap or in fruits. Only male adult fruit flies are captured by male lure trapping, while all immature stages such as eggs (Figure 2(a)), early to final instar larvae (Figures 2(b) to (d)), and ~~pupae and~~ puparia and pupae (Figures 2(e) to (f)) can be found during inspection of fruits.	**EPPO** Proper order (please see figures 2(e) and (f)). *Category : EDITORIAL*
46	107	Fruit flies of the genus *Bactrocera* are detected mainly by male lure trap or in fruits. Only male adult fruit flies are captured by male lure trapping, while all immature	**Philippines**

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		stages such as eggs (Figure 2(a)), early to final instar larvae (Figures 2(b) to (d)), and pupae and puparia (Figures 2(e) to (f)) can be found during inspection of fruits.	*Category : TECHNICAL*
47	109	Guidance on trapping *Bactrocera* fruit flies is given in Appendix 1 of ISPM 26 (*Establishment of pest free areas for fruit flies (Tephritidae)*). Additional information on trapping methods is provided by Drew (1982), Drew and Romig (2010), and FAO and IAEA (2003). The *Bactrocera dorsalis* complex includes species that respond to different male lures. When the lure responsiveness information is available, it can be used as supporting information for species identification. Five of the target species in this diagnostic protocol are methyl eugenol responding species. The only exception is *B. pyrifoliae*, which has been reported to respond to an alternative lure: cue lure (Drew and Romig, 2013).	**European Union** Additional information on trapping methods is provided by Drew (1982), Drew and Romig (2010), and FAO and IAEA (2003)." See note in references. Revised publication in 2013. *Category : EDITORIAL*
48	109	Guidance on trapping *Bactrocera* fruit flies is given in Appendix 1 of ISPM 26 (*Establishment of pest free areas for fruit flies (Tephritidae)*). Additional information on trapping methods is provided by Drew (1982), Drew and Romig (2010), and FAO and IAEA (2003). The *Bactrocera dorsalis* complex includes species that respond to different male lures. When the lure responsiveness information is available, it can be used as supporting information for species identification. Five of the target species in this diagnostic protocol are methyl eugenol responding species. The only exception is *B. pyrifoliae*, which has been reported to respond to an alternative lure: cue lure (Drew and Romig, 2013).	**EPPO** Additional information on trapping methods is provided by Drew (1982), Drew and Romig (2010), and FAO and IAEA (2003)." See note in references. Revised publication in 2013 *Category : EDITORIAL*
49	112	Fruits with soft areas, dark stains, dark pin spots, rot, orifices or injuries that might have originated from female oviposition or larval feeding activities are targeted for inspection. In order to detect punctures made by female flies during oviposition, fruits should be examined under a microscope by an expert. If larval exit holes are observed, the fruit containers should be inspected for pupae. Second and third instar larvae and pupae are not likely to occur when unripe fruits are collected and packed; however, these fruits might host eggs and first instar larvae, which are more difficult to detect. Potentially infested fruits that show typical punctures made by ovipositioning female flies should be cut open to search for eggs or larvae inside. The success of detection depends on careful sampling and examination of fruits.	**Ghana** *Category : SUBSTANTIVE*
50	115	Larvae can be reared to adults by placing infested fruits in cages containing a pupation medium (e.g. damp vermiculite, sand or sawdust) at the bottom..... The cages are covered with cloth or fine mesh. Once the larvae emerge from the fruit, they will move to the pupation medium. It is recommended that each fruit be incubated separately.	**New Zealand** Suggested add temperature range for rearing adnf L:D ratio...Foodsugar/protein. need these to be added to these guidelines for rearing fruit flies. *Category : TECHNICAL*

#	Para	Text	Comment
		Each sample should be observed and pupae gathered daily. The pupae are placed in containers with the pupation medium, and the containers are covered with a tight lid that enables proper ventilation. Once the adults emerge, they must be kept alive for several days to ensure that the tegument and wings acquire the rigidity and characteristic coloration of the species. Flies can be fed with honey (sugar) and water. The adults are then killed by freezing, or by exposure to ethyl acetate or other killing agents appropriate for morphological examination, and then mounted on pins. Prior to mounting (before they harden), it is useful to gently squeeze the apical part of the preabdomen with forceps, then squeeze the base and apex of the oviscape to expose the aculeus tip for females, and to pull out the aedeagus for males. Alternatively, this will need to be dissected later in flies.	
51	115	Larvae can be reared to adults by placing infested fruits in cages containing a pupation medium (e.g. damp vermiculite, sand or sawdust) at the bottom. The cages are covered with cloth or fine mesh. Once the larvae emerge from the fruit, they will move to the pupation medium. It is recommended that each fruit be incubated separately<u>.....</u>. Each sample should be observed and pupae gathered daily. The pupae are placed in containers with the pupation medium, and the containers are covered with a tight lid that enables proper ventilation. Once the adults emerge, they must be kept alive for several days to ensure that the tegument and wings acquire the rigidity and characteristic coloration of the species. Flies can be fed with honey (sugar) and water. The adults are then killed by freezing, or by exposure to ethyl acetate or other killing agents appropriate for morphological examination, and then mounted on pins. Prior to mounting (before they harden), it is useful to gently squeeze the apical part of the preabdomen with forceps, then squeeze the base and apex of the oviscape to expose the aculeus tip for females, and to pull out the aedeagus for males. Alternatively, this will need to be dissected later in flies.	**New Zealand** Suggest that provide a reason why they are incubated separately. *Category : TECHNICAL*
52	117	Identification at the level of the species or the *Bactrocera dorsalis* complex requires morphological examination of adult flies. It is generally difficult and not reliable to morphologically identify eggs, larvae or pupae to the species level. It is not possible to identify a fly to the *Bactrocera dorsalis* complex using immature life stages.	**Russian Federation** We consider it necessary either to develop identification of larvae stage as it is the stage that mostly spreads on plant products, e.g. tropical fruits, or to adopt this draft, adding information on larvae identification during further revision of the standard. *Category : SUBSTANTIVE*
53	118	Molecular methods of *Bactrocera* species identification have been reported and provide additional information to support morphological identifications of specimens. DNA sequencing of the cytochrome oxidase I DNA barcode does not provide adequate resolution to identify many species in the *B. dorsalis* complex (details in	**Japan** Editorial *Category : EDITORIAL*

#	Para	Text	Comment
		section 4.3~~)~~4). Other molecular methods lack the specificity data needed to demonstrate that a test is accurate for species identification. For example, the molecular profiles of all six pest species targeted in the protocol are not known using rDNA analysis (section 4.3). DNA can be used to distinguish *B. carambolae* from *B. dorsalis s.l.* and this test is provided in the protocol (section 4.3.2).	
54	118	Molecular methods of *Bactrocera* species identification have been reported and provide additional information to support morphological identifications of specimens. DNA sequencing of the cytochrome oxidase I DNA barcode does not provide adequate resolution to identify many species in the *B. dorsalis* complex (details in section 4.3). Other molecular methods lack the specificity data needed to demonstrate that a test is accurate for species identification. For example, the molecular profiles of all six pest species targeted in the protocol are not known using rDNA analysis (section 4.3). DNA can be used to distinguish *B. carambolae* from *B. dorsalis s.l.* and this test is provided in the protocol (section 4.3.2).	**Kenya** We propose addition of a statement on appropriate molecular identification technique to species level since those mentioned in the paragraph have been termed as inadequate. *Category : TECHNICAL*
55	120	Proper preparation of specimens is essential for accurate morphological identification. General instructions on preparation of adult fruit fly specimens are given by Drew (1991) and White and Elson-Harris (1992).	**European Union** We assume you are considering dry preservation here only. Nowadays, a lot of material is preserved in ethanol;a.o. for better DNA preservation. This has some consequences for recognition of certain characters (like the medial presutural stripe in B. occipitalis). Perhaps this should be stressed. *Category : TECHNICAL*
56	120	Proper preparation of specimens is essential for accurate morphological identification. General instructions on preparation of adult fruit fly specimens are given by Drew (1991) and White and Elson-Harris (1992).	**EPPO** We assume you are considering dry preservation here only. Nowadays, a lot of material is preserved in ethanol;a.o. for better DNA preservation. This has some consequences for recognition of certain characters (like the medial presutural stripe in B. occipitalis). Perhaps this should be stressed. *Category : TECHNICAL*
57	123	Structures of the ovipositor such as oviscape, eversible membrane and aculeus have been used as important taxonomic characters at species level (Hardy, 1949, 1969; Hardy and Adachi, 1954; Drew and Hancock, 1994). Since the review by Drew and Hancock (1994), aculeus length has been used in particular for distinguishing some of the fruit fly species within the *Bactrocera dorsalis* complex, and male aedeagus length, which is highly correlated with aculeus length, has also been used because only males are trapped in lure trapping surveys. . Care must be taken when interpreting genitalic morphometric information for species diagnostics, as some members of the complex (e.g., Bactrocera dorsalis) exhibit a wide range of aedeagus lengths over their geographic distribution (Krosch et al., 2013; Schutze et al., 2015)”. Preparation	**Australia** *Category : TECHNICAL*

#	Para	Text	Comment
		methods for male genitalia are included in section 4.1.1.	
58	125	Examination of the costal band below the R2+3 vein will be made easier by putting white paper underneath the wing or by using transmitted light.	**Colombia** Falta referencia a las figuras, el texto no posee una figura explicativa asociada. *Category : TECHNICAL*
59	131	Preparation of the abdomen for dissection and examination of genitalia can be accomplished by first removing the abdomen from the specimen and soaking it in a 10% solution of KOH at 95 °C for 10 to 20 minutes depending on the condition of the specimen. Once the KOH soak is complete, the digested abdomen can be transferred to a spot of glycerol.	**European Union** Preparation of the abdomen can be combined with DNA extraction by using a tissue lysis buffer that will also clear the abdomen. This is an invasive but not destructing method of DNA extraction. Using KOH renders the abdomen useless for DNA extraction. *Category : TECHNICAL*
60	131	Preparation of the abdomen for dissection and examination of genitalia can be accomplished by first removing the abdomen from the specimen and soaking it in a 10% solution of KOH at 95 °C for 10 to 20 minutes depending on the condition of the specimen. Once the KOH soak is complete, the digested abdomen can be transferred to a spot of glycerol.	**EPPO** Preparation of the abdomen can be combined with DNA extraction by using a tissue lysis buffer that will also clear the abdomen. This is an invasive but not destructing method of DNA extraction. Using KOH renders the abdomen useless for DNA extraction. *Category : TECHNICAL*
61	137	Methods to identify fly specimens to the genus *Bactrocera* are not within the scope of the current protocol. However, proper screening of specimens is important to ensure that flies being diagnosed are within the subgenus *Bactrocera* (*Bactrocera*). The work of White and Elson-Harris (1992) provides a useful resource for those general identifications. Characters used to identify fruit flies to the tribe Dacini, including the genus *Bactrocera*, are useful in the identification of flies to the subgenus *Bactrocera* (*Bactrocera*). These flies have reduced chaetotaxies on the head, with ocellar (Figure ~~8(b))~~ 8(c)) and postocellar (Figure ~~8(b))~~ 8(c)) bristles absent (atrophied); the first flagellomere (Figure 8(a)) is at least three times as long as broad; and wing cell cup extension is very long (Figure 9(a)). In addition to these characteristics, fruit flies of the genus *Bactrocera* have separate abdominal tergites ~~(Figures 6(a) and~~ ~~(~~(Figure 6(a)~~d)~~) (except for first and second tergites). In addition to the above characteristics of the genus *Bactrocera*, the subgenus *Bactrocera* also has the characteristics listed below.	**European Union** Correct figures ? *Category : EDITORIAL*
62	137	Methods to identify fly specimens to the genus *Bactrocera* are not within the scope of the current protocol. However, proper screening of specimens is important to ensure that flies being diagnosed are within the subgenus *Bactrocera* (*Bactrocera*). The work of White and Elson-Harris (1992) provides a useful resource for those general identifications. Characters used to identify fruit flies to the tribe Dacini, including the genus *Bactrocera*, are useful in the identification of flies to the subgenus *Bactrocera*	**Japan** Editorial *Category : EDITORIAL*

#	Para	Text	Comment
		(*Bactrocera*). These flies have reduced chaetotaxies on the head, with ocellar (Figure 8(b)) and postocellar (Figure 8(b)) bristles absent (atrophied); the first flagellomere (Figure 8(a)) is at least three times as long as broad; and wing cell cup extension is very long (Figure 9(a)). In addition to these characteristics, fruit flies of the genus *Bactrocera* have separate abdominal tergites (Figures 6(a) and ~~((~~b~~d))~~) (except for first and second tergites). In addition to the above characteristics of the genus *Bactrocera*, the subgenus *Bactrocera* also has the characteristics listed below.	
63	137	Methods to identify fly specimens to the genus *Bactrocera* are not within the scope of the current protocol. However, proper screening of specimens is important to ensure that flies being diagnosed are within the subgenus *Bactrocera* (*Bactrocera*). The work of White and Elson-Harris (1992) provides a useful resource for those general identifications. Characters used to identify fruit flies to the tribe Dacini, including the genus *Bactrocera*, are useful in the identification of flies to the subgenus *Bactrocera* (*Bactrocera*). These flies have reduced chaetotaxies on the head, with ocellar (Figure ~~8(b))~~ 8(c)) and postocellar (Figure ~~8(b))~~ 8(c)) bristles absent (atrophied); the first flagellomere (Figure 8(a)) is at least three times as long as broad; and wing cell cup extension is very long (Figure 9(a)). In addition to these characteristics, fruit flies of the genus *Bactrocera* have separate abdominal tergites ~~(Figures 6(a) and~~ ~~(~~(Figure 6(a)~~d)~~) (except for first and second tergites). In addition to the above characteristics of the genus *Bactrocera*, the subgenus *Bactrocera* also has the characteristics listed below.	**EPPO** Proper figures? *Category : EDITORIAL*
64	137	Methods to identify fly specimens to the genus *Bactrocera* are not within the scope of the current protocol. However, proper screening of specimens is important to ensure that flies being diagnosed are within the subgenus *Bactrocera* (*Bactrocera*). The work of White and Elson-Harris (1992) provides a useful resource for those general identifications. Characters used to identify fruit flies to the tribe Dacini, including the genus *Bactrocera*, are useful in the identification of flies to the subgenus *Bactrocera* (*Bactrocera*). These flies have reduced chaetotaxies on the head, with ocellar (Figure 8(b)) and postocellar (Figure 8(b)) bristles absent (atrophied); the first flagellomere (Figure 8(a)) is at least three times as long as broad; and wing cell cup extension is very long (Figure 9(a)). In addition to these characteristics, fruit flies of the genus *Bactrocera* have separate abdominal tergites (Figures 6(a) and (d)) (except for first and second tergites). In addition to the above characteristics of the genus *Bactrocera*, the subgenus *Bactrocera* also has the characteristics listed below.	**Colombia** La figura no hace referencia a esta letra (9a), no está claro a que hacen referencia los asteriscos en la figura. *Category : TECHNICAL*
65	138	The presence of diagnostic characters of other *Bactrocera* subgenera is useful in	**European Union**

#	Para	Text	Comment
		diagnosing flies as not being members of the *Bactrocera dorsalis* complex via exclusion. For example, flies in the subgenus *Bactrocera* (*Afrodacus*) lack anterior supra-alar bristles (Figure 10) and flies in the subgenus *Bactrocera* (*Gymnodacus*) lack pectens on tergite 3 (Figure ~~6)~~6(a)). The characters listed below are used for defining the subgenus *Bactrocera*. In starting identification, it is important to confirm that the fruit flies in question meet the definition. At this stage of identification, superficially similar species in other subgenera such as *Afrodacus* or *Gymnodacus* that could be intercepted during plant inspection can be excluded.	Please see figure 6(a) and paragraph 142. *Category : EDITORIAL*
66	138	The presence of diagnostic characters of other *Bactrocera* subgenera is useful in diagnosing flies as not being members of the *Bactrocera dorsalis* complex via exclusion. For example, flies in the subgenus *Bactrocera* (*Afrodacus*) lack anterior supra-alar bristles (Figure 10) and flies in the subgenus *Bactrocera* (*Gymnodacus*) lack pectens on tergite 3 (Figure ~~6)~~6(a)). The characters listed below are used for defining the subgenus *Bactrocera*. In starting identification, it is important to confirm that the fruit flies in question meet the definition. At this stage of identification, superficially similar species in other subgenera such as *Afrodacus* or *Gymnodacus* that could be intercepted during plant inspection can be excluded.	**EPPO** Please see figure 6(a) and paragraph 142. *Category : EDITORIAL*
67	138	The presence of diagnostic characters of other *Bactrocera* subgenera is useful in diagnosing flies as not being members of the *Bactrocera dorsalis* complex via exclusion. For example, flies in the subgenus *Bactrocera* (*Afrodacus*) lack anterior supra-alar bristles (Figure 10) and flies in the subgenus *Bactrocera* (*Gymnodacus*) lack pectens on tergite 3 (Figure ~~6)~~6(a)). The characters listed below are used for defining the subgenus *Bactrocera*. In starting identification, it is important to confirm that the fruit flies in question meet the definition. At this stage of identification, superficially similar species in other subgenera such as *Afrodacus* or *Gymnodacus* that could be intercepted during plant inspection can be excluded.	**Colombia** La figura correcta es la 6a. *Category : EDITORIAL*
68	142	abdominal ~~stern~~tergite ~~ite~~ 5 of male with pecten (Figure 6(a))~~5 of male with pecten (Figure 6(a))~~	**Colombia** No es sternite, es tergite *Category : TECHNICAL*
69	146	one pair of scutellar (sc.) bristles present (Figure 10).	**European Union** Perhaps indicate which ones? (apical) *Category : TECHNICAL*
70	146	one pair of scutellar (sc.) bristles present (Figure 10).	**EPPO** Perhaps indiquate which ones? (apical) *Category : TECHNICAL*
71	148	Characters useful for the identification of adult flies following the terminology of Drew and Romig (2013) are listed in Table 2. The ~~definition~~ description of the	**European Union** More appropriate term?

#	Para	Text	Comment
		Bactrocera dorsalis complex in this protocol follows Drew and Romig (2013) except for scutum colour. Scutum colour in Drew and Romig (2013) is black, but herein black and red-brown are included in the description of the complex. A specimen must have characters that match the descriptions provided in Table 2 to confidently identify the fly as a *B. dorsalis* complex species.	*Category : EDITORIAL*
72	148	Characters useful for the identification of adult flies following the terminology of Drew and Romig (2013) are listed in Table 2. The ~~definition~~ description of the *Bactrocera dorsalis* complex in this protocol follows Drew and Romig (2013) except for scutum colour. Scutum colour in Drew and Romig (2013) is black, but herein black and red-brown are included in the description of the complex. A specimen must have characters that match the descriptions provided in Table 2 to confidently identify the fly as a *B. dorsalis* complex species.	**EPPO** More appropriate term? *Category : EDITORIAL*
73	153	Distinct facial spots present (Figures 8(a), 8(b), 11)	**European Union** Perhaps: face yellow with distinct black facial spots present. *Category : TECHNICAL*
74	153	Distinct facial spots present (Figures 8(a), 8(b), 11)	**EPPO** Perhaps: face yellow with distinct black facial spots present *Category : TECHNICAL*
75	157	Lateral vittae present (Figure 10) and yellow (Figures 10 and 13)	**Colombia** La figura 10 no muestra el color mencionado. *Category : TECHNICAL*
76	161	~~Yellow~~ Yellowish colour (Figures 1 and 12)	**Colombia** En la figura 12 los escutelos aparecen más amarillentos (yellowish) que simplemente amarillos (yellow). *Category : TECHNICAL*
77	165	Never with other dark patterns (Figure ~~11)~~12)	**European Union** Correct figure ? *Category : EDITORIAL*
78	165	Never with other dark patterns (Figure ~~11)~~12)	**Japan** Editorial *Category : EDITORIAL*
79	165	Never with other dark patterns (Figure ~~11)~~12)	**EPPO** Proper figure? *Category : EDITORIAL*
80	165	Never with other dark patterns ~~(Figure 11)~~	**Colombia** La imágenes de la figura 11 hacen referencia a la cabeza en vista antero-lateral, las cuales no son adecuadas para mostrar ausencia de patrones torácicos dorsales. Se debe relacionar les a vista dorsal del tórax. *Category : TECHNICAL*
81	177	With a “T” pattern on tergites 3–5 (Figures 6 (a) and 16)	**Colombia**

#	Para	Text	Comment
			La figura correcta es la 6a *Category : EDITORIAL*
82	178	***4.2.3 Morphological identification of six economically important species of ~~Bactrocera dorsalis~~Bactrocera dorsalis complex***	**Viet Nam** *Category : TECHNICAL*
83	201	Medium-sized, oval (Figure 11a)	**Viet Nam** should be more detail, "oval" shaped faces are not specific for classification *Category : TECHNICAL*
84	201	Medium-sized, oval (Figure ~~11a)~~11(a))	**Japan** Editorial *Category : EDITORIAL*
85	202	Large, elongate oval (Figure 11(b))	**Viet Nam** should be more detail, "elongate" or "oval" shaped faces are not specific for classification *Category : TECHNICAL*
86	203	Medium to large, circular to oval (inter-regionally variable) (Figure 11(c))	**Viet Nam** should be more detail, "oval" shaped faces are not specific for classification *Category : TECHNICAL*
87	204	Large, oval (Figure 11(d))	**Viet Nam** should be more detail, "oval" shaped faces are not specific for classification *Category : TECHNICAL*
88	205	Large, oval (Figure 11e)	**Viet Nam** should be more detail, "oval" shaped faces are not specific for classification *Category : TECHNICAL*
89	205	Large, oval (Figure ~~11e)~~11(e))	**Japan** Editorial *Category : EDITORIAL*
90	261	Dull black (Figure 12(a))	**Viet Nam** Should be more detail. Should be as follows: "The black spot at the top of scutelum occupies a large area of the scutelum" *Category : TECHNICAL*
91	262	Pure black (Figure 12(b))	**Viet Nam** Should be more detail. Should be as follows: "The black spot at the top of scutelum occupies a large area of the scutelum" *Category : TECHNICAL*
92	262	~~Pure~~ Entirely black (Figure 12(b))	**European Union** *Category : EDITORIAL*
93	262	~~Pure~~ Entirely black (Figure 12(b))	**EPPO**

#	Para	Text	Comment
			Category : EDITORIAL
94	263	Black to red-brown (inter or intra-regionally variable) (Figure 12(c))	**Viet Nam** Should be more detail. Should be as follows: "The black spot at the top of scutelum occupies a large area of the scutelum" *Category : TECHNICAL*
95	264	Black (Figure 12(d))	**Viet Nam** Should be more detail. Should be as follows: "The black spot at the top of scutelum occupies a large area of the scutelum" *Category : TECHNICAL*
96	265	Black with clear central stripe (Figure 12(e))	**Viet Nam** Should be more detail. Should be as follows: "The black spot at the top of scutelum occupies a large area of the scutelum" *Category : TECHNICAL*
97	265	Black with clear central stripe (Figure 12(e))	**European Union** A better figure is needed to demonstrate this, the central stripe is not visible. *Category : TECHNICAL*
98	265	Black with clear central stripe (Figure 12(e))	**EPPO** A better figure is needed to demonstrate this, the central stripe is not visible *Category : TECHNICAL*
99	266	Pure black (Figure 12(f))	**Viet Nam** Should be more detail. Should be as follows: "The pure spot at the top of scutelum occupies a large area of the scutelum" *Category : TECHNICAL*
100	266	~~Pure~~ Entire black (Figure 12(f))	**European Union** *Category : EDITORIAL*
101	266	~~Pure~~ Entirely black (Figure 12(f))	**EPPO** *Category : EDITORIAL*
102	274	Anterior margin of mesopleural stripe (Figures 4(a) and 13)	**European Union** Aluja & Norrbom 1999 set the standard terminology for tephritid morphology. Preference was given to anepisternum in favour of mesopleuron. If Aluja & Norrbom is to be followed, this should be changed throughout (including reference to anepisternal bristles, anepisternal stripe, etc). At least it should be mentioned that the alternative term exists and is used commonly in a number of publications (White & Elson Harris, White 2006 revision of African Dacina, etc). *Category : SUBSTANTIVE*
103	274	Anterior margin of mesopleural stripe (Figures 4(a) and 13)	**EPPO** Aluja & Norrbom 1999 set the standard terminology for tephritid morphology. Preference was given to anepisternum in favour of

#	Para	Text	Comment
			mesopleuron. If Aluja & Norrbom is to be followed, this should be changed throughout (including reference to anepisternal bristles, anepisternal stripe, etc). At least it should be mentioned that the alternative term exists and is used commonly in a number of publications (White & Elson Harris, White 2006 revision of African Dacina, etc) *Category : SUBSTANTIVE*
104	355	Aedeagus length (mm) (Figure ~~7)~~7(d))	**Japan** Editorial *Category : EDITORIAL*
105	375	2. Scutum entirely~~pure~~ black (Figure 12(b)), abdominal tergites 3–5 with broad black dorsolateral markings (Figures 16(b) and 17(b)); lateral vittae very narrow (Figure 3(b))……..*B. caryeae*	**European Union** *Category : TECHNICAL*
106	375	2. Scutum entirely~~pure~~ black (Figure 12(b)), abdominal tergites 3–5 with broad black dorsolateral markings (Figures 16(b) and 17(b)); lateral vittae very narrow (Figure 3(b))……..*B. caryeae*	**EPPO** *Category : TECHNICAL*
107	376	– Scutum mostly black (Figure 12(d)), abdominal tergites 3–5 with "T" pattern and tergites 4–5 with very narrow anterolateral black marking (Figures 16(d) and 17(d)); lateral vittae narrow (Figure 3(d))…….*B. kandiensis*	**European Union** *Category : TECHNICAL*
108	376	– Scutum mostly black (Figure 12(d)), abdominal tergites 3–5 with "T" pattern and tergites 4–5 with very narrow anterolateral black marking (Figures 16(d) and 17(d)); lateral vittae narrow (Figure 3(d))…….*B. kandiensis*	**EPPO** *Category : TECHNICAL*
109	378	– Costal band widening slightly to moderately around apex of wing……4	**Colombia** No hay figura asociada que explique el carácter. Se requiere incluir figura para mayor entendimiento. *Category : TECHNICAL*
110	385	DNA sequencing of either the internal transcribed spacer 1 (ITS1) or 2 (ITS2) nuclear DNA regions has been proposed as a reliable test to distinguish between the species *B. carambolae* and *B. dorsalis s.l.* (Boykin *et al*., 2014; Schutze *et al*., 2015a). The internal transcribed spacer 1 (ITS1) test as described by Boykin *et al*. (2014) for distinguishing between the two species is included in the current protocol. This test is designed to diagnose a fly as *B. carambolae* based on the presence of a unique DNA insertion. Specificity of the test for *B. carambolae* has been examined using four additional species in the *Bactrocera dorsalis* complex: *B. dorsalis s.l.*, *B. occipitalis*, *B. opiliae* and *B. cacuminata.*	**European Union** Is 4 species a wide enough panel? *Category : TECHNICAL*
111	385	DNA sequencing of either the internal transcribed spacer 1 (ITS1) or 2 (ITS2) nuclear DNA regions has been proposed as a reliable test to distinguish between the species	**EPPO** Is 4 species a wide enough panel? *Category : TECHNICAL*

#	Para	Text	Comment
		B. carambolae and *B. dorsalis s.l.* (Boykin *et al*., 2014; Schutze *et al*., 2015a). The internal transcribed spacer 1 (ITS1) test as described by Boykin *et al*. (2014) for distinguishing between the two species is included in the current protocol. This test is designed to diagnose a fly as *B. carambolae* based on the presence of a unique DNA insertion. Specificity of the test for *B. carambolae* has been examined using four additional species in the *Bactrocera dorsalis* complex: *B. dorsalis s.l.*, *B. occipitalis*, *B. opiliae* and *B. cacuminata.*	
112	386	In this diagnostic protocol, methods (including reference to brand names) are described as published, as these define the original level of sensitivity, specificity and reproducibility achieved. ~~The use of names of reagents, chemicals or equipment in these diagnostic protocols implies no approval of them to the exclusion of others that may also be suitable.~~ Laboratory procedures presented in the protocols may be adjusted to the standards of individual laboratories, provided that they are adequately validated.	**Uruguay** Text deleted for consistency with other DP *Category : TECHNICAL*
113	388	Boykin *et al.* (2014) and Ball and Armstrong (2008) provide protocols for DNA extraction using commercial kits that are useful because small starting material such as one fruit fly leg can give enough DNA yield and quality for PCR reactions. The methods used to preserve fruit flies for morphological and molecular examination are not the same. Ethanol is a common preservative for fruit fly DNA. Although fruit fly specimens can be preserved in ≥95% ethanol at −20 °C or colder for long-term storage, ethanol can alter the colouring of adult specimens, which can hinder morphological identification. All identifications performed using this protocol require morphological examination. In cases where molecular methods are to be used, it is therefore recommended that a leg be removed and stored in ethanol for DNA extraction and that the remaining specimen be prepared for morphology work. Further examples of methods are provided by Plant Health Australia (2016).	**European Union** All identifications performed using this protocol require morphological examination. In cases where molecular methods are to be used, it is therefore recommended that a leg be removed and stored in ethanol for DNA extraction and that the remaining specimen be prepared for morphology work. Further examples of methods are provided by Plant Health Australia (2016). Dry versus ethanol preservation is mentioned here. It is suggested to also mentioned this earlier for adult specimen preservation. *Category : TECHNICAL*
114	388	Boykin *et al.* (2014) and Ball and Armstrong (2008) provide protocols for DNA extraction using commercial kits that are useful because small starting material such as one fruit fly leg can give enough DNA yield and quality for PCR reactions. The methods used to preserve fruit flies for morphological and molecular examination are not the same. Ethanol is a common preservative for fruit fly DNA. Although fruit fly specimens can be preserved in ≥95% ethanol at −20 °C or colder for long-term storage, ethanol can alter the colouring of adult specimens, which can hinder	**EPPO** All identifications performed using this protocol require morphological examination. In cases where molecular methods are to be used, it is therefore recommended that a leg be removed and stored in ethanol for DNA extraction and that the remaining specimen be prepared for morphology work. Further examples of methods are provided by Plant Health Australia (2016). Dry versus ethanol preservation is mentioned here. It is suggested to also mentioned this earlier for adult specimen preservation.

#	Para	Text	Comment
		morphological identification. All identifications performed using this protocol require morphological examination. In cases where molecular methods are to be used, it is therefore recommended that a leg be removed and stored in ethanol for DNA extraction and that the remaining specimen be prepared for morphology work. Further examples of methods are provided by Plant Health Australia (2016).	*Category : TECHNICAL*
115	441	The size of ITS1 is different for *B. carambolae* and *B. dorsalis* because of a 44-bp insertion in *B. carambolae* located near one end of the gene located near the ITS7 primer. The inserted DNA is identical in all *B. carambolae* studied. The sequence of the insertion is: 5´- GAAAAATTAATAAAAAGTTAAATGATCTTTTTATAAAAAAT-3´.	**European Union** Note that B. tryoni also has a 44bp insertion in the same place (sequence 5'-AAAAAATTTTATAAAAAGTTAAATGATCTTTTTATAGTAAAT-3'). *Category : TECHNICAL*
116	441	The size of ITS1 is different for *B. carambolae* and *B. dorsalis* because of a 44-bp insertion in *B. carambolae* located near one end of the gene located near the ITS7 primer. The inserted DNA is identical in all *B. carambolae* studied. The sequence of the insertion is: 5´- GAAAAATTAATAAAAAGTTAAATGATCTTTTTATAAAAAAT-3´.	**EPPO** Note that B. tryoni also has a 44bp insertion in the same place (sequence 5'-AAAAAATTTTATAAAAAGTTAAATGATCTTTTTATAGTAAAT-3'). *Category : TECHNICAL*
117	442	The ITS1 sequence is variable between conspecific specimens of these two species (Boykin *et al.*, 2014). Consequently, an identical match for sites outside of the insertion region is not expected. However, the test sequence should be at least 99% similar to one of the reference sequences for the interpretation to proceed. It is possible to distinguish between *B. carambolae* and *B. dorsalis s.l.* after comparing the DNA sequence of the tested specimen with a representative sequence of each species: GenBank KC446737 for *B. carambolae* and KC446776 for *B. dorsalis*. If the tested sequence is most similar to *B. carambolae* and has the 44-bp insertion region, then it can be diagnosed as *B. carambolae*. If the tested sequence is most similar to *B. dorsalis* and lacks the insertion region, then it is diagnosed as not *B. carambolae*. Several other species in the *B. dorsalis* complex lack the insertion and a match with *B. dorsalis s.l.* cannot exclude those as a possible identification.	**European Union** Sequences KC446981 (B papayae) and KC446898 (B. dorsalis) from Boykin et al both have this insertion. Samples KC446930, KC446861 and KC446910 (B. carambolae) do not have this insertion. As it stands, the 44 bp insertion is therefore not fully diagnostic for B. carambolae. It is possible that these are misidentified samples; One of these samples has been querried by one EPPO Lab before with the authors but no response as been received yet. *Category : TECHNICAL*
118	442	The ITS1 sequence is variable between conspecific specimens of these two species (Boykin *et al.*, 2014). Consequently, an identical match for sites outside of the insertion region is not expected. However, the test sequence should be at least 99% similar to one of the reference sequences for the interpretation to proceed. It is possible to distinguish between *B. carambolae* and *B. dorsalis s.l.* after comparing the DNA sequence of the tested specimen with a representative sequence of each species: GenBank KC446737 for *B. carambolae* and KC446776 for *B. dorsalis*. If the tested sequence is most similar to *B. carambolae* and has the 44-bp insertion region,	**EPPO** *Category : TECHNICAL*

#	Para	Text	Comment
		then it can be diagnosed as *B. carambolae*. If the tested sequence is most similar to *B. dorsalis* and lacks the insertion region, then it is diagnosed as not *B. carambolae*. Several other species in the *B. dorsalis* complex lack the insertion and a match with *B. dorsalis s.l.* cannot exclude those as a possible identification.	
119	443	**4.4 Other molecular methods of identification**	**Japan** *Muraji and Nakahara (2002) Discrimination among pest species of Bactrocera (Diptera: Tephritidae) based on PCR-RFLP of the mitochondrial DNA. Applied Entomology and Zoology 37(3): 437–446. *Category : SUBSTANTIVE*
120	444	Plant Health Australia (2016) has compiled a resource for identification of *Bactrocera* species using DNA methods. That resource summarizes three molecular options for identification: conventional PCR and restriction fragment length polymorphism (RFLP) of the ITS1 region (Plant Health Australia, 2016), PCR-RFLP analysis of a segment of rRNA array including the ITS1 and 18S gene regions (Armstrong *et al*., 1997; Armstrong and Cameron, 2000), and DNA barcoding of the *cytochrome oxidase subunit I* (*COI*) gene (Armstrong and Ball, 2005) based on the Barcode of Life Data Systems (BOLD) resource (Ratnasingham and Hebert, 2007). The species *B. caryeae*, *B. kandiensis*, *B. occipitalis* and *B. pyrifoliae* do not have molecular profiles available for either of the PCR-RFLP tests described in the Plant Health Australia resource, precluding their use as a diagnostic test for the pests. For ~~the species~~ *B. ~~dorsalis~~dorsalis s.l.*, the resource provides expected PCR product sizes of ITS1 and the expected fragment sizes of digested PCR products of the rDNA fragment including ITS1+18S. These rDNA tests lack specificity data to support diagnosis of a fly as *B. dorsalis s.l.* using genetic profiles alone. However, rDNA profiles that do not match recorded results of *B. dorsalis s.l.* can be used to reject diagnosis of a fly as *B. dorsalis s.l.*	**European Union** ? (please see the last two sentences of the paragraph and paragraph 41). *Category : TECHNICAL*
121	444	Plant Health Australia (2016) has compiled a resource for identification of *Bactrocera* species using DNA methods. That resource summarizes three molecular options for identification: conventional PCR and restriction fragment length polymorphism (RFLP) of the ITS1 region (Plant Health Australia, 2016), PCR-RFLP analysis of a segment of rRNA array including the ITS1 and 18S gene regions (Armstrong *et al*., 1997; Armstrong and Cameron, 2000), and DNA barcoding of the *cytochrome oxidase subunit I* (*COI*) gene (Armstrong and Ball, 2005) based on the Barcode of Life Data Systems (BOLD) resource (Ratnasingham and Hebert, 2007). The species *B. caryeae*, *B. kandiensis*, *B. occipitalis* and *B. pyrifoliae* do not have molecular profiles available	**EPPO** ? (please see the last two sentences of the paragraph and paragraph 41). *Category : TECHNICAL*

#	Para	Text	Comment
		for either of the PCR-RFLP tests described in the Plant Health Australia resource, precluding their use as a diagnostic test for the pests. For ~~the species~~ *B.* ~~*dorsalis*~~*dorsalis s.l.*, the resource provides expected PCR product sizes of ITS1 and the expected fragment sizes of digested PCR products of the rDNA fragment including ITS1+18S. These rDNA tests lack specificity data to support diagnosis of a fly as *B. dorsalis s.l.* using genetic profiles alone. However, rDNA profiles that do not match recorded results of *B. dorsalis s.l.* can be used to reject diagnosis of a fly as *B. dorsalis s.l.*	
122	445	DNA barcode records are not available for *B. pyrifoliae*. The *cytochrome oxidase I* (*COI*) DNA barcode records for the other five species cannot distinguish at the species level (Armstrong and Ball, 2005). To date, no study has provided information on how to use *COI* sequence data to accept or reject a diagnosis of a specimen as part of the *Bactrocera dorsalis* complex or as one of the 85 species within the complex. The work by Leblanc *et al*. (2015) demonstrates that this complex is not a monophyletic group and a molecular diagnosis of the complex is not possible. The standard DNA Barcode *COI* region cannot be used reliably to differentiate *B. dorsalis s.l.* from other species in the *Bactrocera dorsalis* complex including *B. carambolae* (Armstrong and Ball, 2005).	**Philippines** We are confused as to the inconsistency of the above statements. *Category : SUBSTANTIVE*
123	449	~~**6. Contact points for further information**~~	**Viet Nam** This section move to Appendix 1 *Category : EDITORIAL*
124	450	~~Further information on this protocol can be obtained from:~~	**Viet Nam** move to Appendix 1 *Category : EDITORIAL*
125	451	~~Pest Identification and Diagnostics Section, Yokohama Plant Protection Station, Ministry of Agriculture, Forestry and Fisheries, Japan (Kenji Tsuruta; e-mail: tsurutak@pps.maff.go.jp; tel.: +81-45-622-8940; fax: +81-45-621-7560).~~	**Viet Nam** This para move to Appendix 1 *Category : EDITORIAL*
126	452	~~Regional R&D Training Center for Insect Biotechnology (RCIB), Department of Biotechnology, Mahidol University, 272 Rama VI Road, Ratchathewee, Bangkok 10400, Thailand (Sujinda Thanaphum; e-mail: sujinda.tha@mahidol.ac.th; tel.: +66814333963; fax: +6623547160).~~	**Viet Nam** This para move to Appendix 1 *Category : EDITORIAL*
127	453	~~William F. Barr Entomological Museum, Department of Plant, Soil and Entomological Sciences, University of Idaho, 875 Perimeter Drive MS 2339, Moscow, Idaho, 83844-2339, United States of America (Luc Leblanc; e-mail: leblancl@uidaho.edu; tel.: +1 208-885-6274; fax: +1 208-885-7760).~~	**Viet Nam** This para move to Appendix 1 *Category : EDITORIAL*

#	Para	Text	Comment
128	454	~~A request for a revision to a diagnostic protocol may be submitted by national plant protection organizations (NPPOs), regional plant protection organizations (RPPOs) or Commission on Phytosanitary Measures (CPM) subsidiary bodies through the IPPC Secretariat (ippc@fao.org), which will in turn forward it to the Technical Panel on Diagnostic Protocols (TPDP).~~	**Viet Nam** This para move to Appendix 1 *Category : EDITORIAL*
129	455	~~**7. Acknowledgements**~~	**Viet Nam** This section move to Appendix 2 *Category : EDITORIAL*
130	456	~~The original draft of this protocol was written by Kenji Tsuruta (Ministry of Agriculture, Forestry and Fisheries, Japan (see preceding section)), Sujinda Thanaphum (Mahidol University, Thailand (see preceding section)), Luc Leblanc (University of Idaho, United States of America (see preceding section)) and Norman Barr (United States Department of Agriculture, United States of America). The following experts provided comments on earlier versions that improved the quality of the protocol: Jane Royer (Queensland Department of Agriculture and Fisheries, Australia), Mark Schutze (Queensland University of Technology, Australia), Josephine Moraa Songa (Kenya Agricultural & Livestock Research Organization, Kenya), George Momanyi (Kenya Plant Health Inspectorate Service, Kenya), Sharon Reid (Fera Science Ltd., Sand Hutton, York, United Kingdom), Yuji Kitabara (Ministry of Agriculture, Forestry and Fisheries, Japan), Eddy Dijkstra (Plant Protection Service, Netherlands), and Ken Hong Tan (Tan Hak Heng, Penang, Malaysia).~~	**Viet Nam** This para move to Appendix 2 *Category : EDITORIAL*
131	456	The original draft of this protocol was written by Kenji Tsuruta (Ministry of Agriculture, Forestry and Fisheries, Japan (see preceding section)), Sujinda Thanaphum (Mahidol University, Thailand (see preceding section)), Luc Leblanc (University of Idaho, United States of America (see preceding section)) and Norman Barr (United States Department of Agriculture, United States of America). The following experts provided comments on earlier versions that improved the quality of the protocol: Jane Royer (Queensland Department of Agriculture and Fisheries, Australia), Mark Schutze (Queensland University of Technology, Australia), Josephine Moraa Songa (Kenya Agricultural & Livestock Research Organization, Kenya), George Momanyi (Kenya Plant Health Inspectorate Service, Kenya), Sharon Reid (Fera Science Ltd., Sand Hutton, York, United Kingdom), Yuji ~~Kitabara~~ Kitahara (Ministry of Agriculture, Forestry and Fisheries, Japan), Eddy Dijkstra (Plant Protection Service, Netherlands), and Ken Hong Tan (Tan Hak Heng, Penang,	**Japan** Editorial *Category : EDITORIAL*

#	Para	Text	Comment
		Malaysia).	
132	479	**FAO & IAEA** (International Atomic Energy Agency). 2003. Trapping Guidelines for Area-Wide Fruit Fly Programmes. Vienna, IAEA. 48 pp. Available at http://www-pub.iaea.org/MTCD/publications/PDF/TG-FFP_web.pdf (last accessed 25 April, 2017).	**European Union** http://www-naweb.iaea.org/nafa/ipc/public/FruitFlyTrapping.pdf . *Category : TECHNICAL*
133	479	**FAO & IAEA** (International Atomic Energy Agency). 2003. Trapping Guidelines for Area-Wide Fruit Fly Programmes. Vienna, IAEA. 48 pp. Available at http://www-pub.iaea.org/MTCD/publications/PDF/TG-FFP_web.pdf (last accessed 25 April, 2017).	**EPPO** http://www-naweb.iaea.org/nafa/ipc/public/FruitFlyTrapping.pdf *Category : TECHNICAL*
134	494	**Schutze, M.K., Mahmood, K., Pavasovic, A., Bo, W., Newman, J., Clarke, A.R., Krosch, M.N. & Cameron, S.L.** 2015b. One and the same: Integrative taxonomic evidence that *Bactrocera invadens* (Diptera: Tephritidae) is the same species as the oriental fruit fly *Bactrocera dorsalis*. *Systematic Entomology*, 40: 472–486. Add publication...	**New Zealand** add ref Schutze et al 2017 . Systemic Entomology DOI: 10.111/syem 12250. Is latest publication in favour of B Dorsalis synonymies. *Category : TECHNICAL*
135	500		**Colombia** Debido a que todas las figuras relacionadas deben ser comparables, se requiere incluir una escala. *Category : TECHNICAL*
136	501	**Figure 1.** *Bactrocera ~~dorsalis~~dorsalis s.l.*, female (habitus)	**Colombia** Se recomienda colocar s.l. (Sensu lato) debido a que se refiere a tres especies (B. dorsalis, B. papayae, B. philippinensis) en una. *Category : TECHNICAL*
137	504		**Colombia** Las figuras 2b y 2c no son lo suficientemente nítidas y tampoco cuentan con un buen contraste (fondo) que facilite su visualización. *Category : TECHNICAL*
138	523		**Japan** Show the site of "ocellar triangle" in Figure8-(b) such as Figure8-(a) and (c) to make it easier to understand. *Category : TECHNICAL*
139	523		**Colombia** *Category : EDITORIAL*
140	524	**Figure 8.** (a) Lateral view of Dacinae head. (b) Frontal view of Dacinae head. (c) Dorsal view of Dacinae head (vertex). i. or. b, inferior fronto-orbital bristles; s. or. b, superior fronto-orbital bristles see comment.	**New Zealand** Other comment on figures - could illustrate the reproductive system of a mature female B dorslis. Adescription how to prepare a slide to locate sperm in the spermathecal. This assists in the determining the mating status of the female. *Category : TECHNICAL*
141	527		**Colombia** Se requiere indicar el significado del asterisco (*) en la explicación

#	Para	Text	Comment
			de la figura. *Category : TECHNICAL*
142	528	**Figure 9.** Wing of Dacinae. Veins: A_1, branch of anal vein; C, costa; CuA_1, CuA_2, anterior branches of cubitus; M, media; R_1, anterior branch of radius; R_{2+3}, R_{4+5}, combined posterior branches of radius; Sc, subcosta; bm-cu = basal medial-cubital crossvein; dm-cu, discal medial-cubital crossvein; r-m, radial-medial crossvein. Cells: bc, basal costal; c, costal; sc, subcostal; bm, basal medial; br, basal radial; cup, posterior cubital; dm, discal medial. Anal streak, areas around cup and cup extension indicated red outline. * Detail of c and bc cells.	**Colombia** Mencionar el significado del asterisco en la explicación de la figura. *Category : TECHNICAL*
143	547		**Colombia** La figura 15d presenta el ala rota, se requiere cambiarla por una que se encuentre en perfecto estado, para evitar confusiones. *Category : TECHNICAL*
144	552	**Figure 16.** Abdomen in dorsal view: (a) *Bactrocera carambolae*; (b) *Bactrocera caryeae*; (c) *Bactrocera dorsalis s.l.*; (d) *Bactrocera kandiensis*; (e) *Bactrocera occipitalis*; (f) *Bactrocera pyrifoliae*.	**Colombia** Figura no citada ni utilizada en el texto, solo en la clave. Se sugiere citarla también en el texto. *Category : TECHNICAL*
145	558	**Figure 18.** Postpronotal lobes in dorsal view: (a) *Bactrocera carambolae*; (b) *Bactrocera caryeae*; (c) *Bactrocera dorsalis;* (d) *Bactrocera kandiensis*; (e) *Bactrocera occipitalis*; (f) *Bactrocera pyrifoliae*.~~s~~	**European Union** Typo. *Category : EDITORIAL*
146	558	**Figure 18.** Postpronotal lobes in dorsal view: (a) *Bactrocera carambolae*; (b) *Bactrocera caryeae*; (c) *Bactrocera dorsalis;* (d) *Bactrocera kandiensis*; (e) *Bactrocera occipitalis*; (f) *Bactrocera pyrifoliae*.~~s~~	**EPPO** Typo. *Category : EDITORIAL*