

2004-015: Draft annex to ISPM 27:2006 - Genus Anastrepha

Comm	Para	Comment type	Comment	Explanation	Country
no.	no.	type			
1.	G	Substantive	I support the document as it is and I have no comments		Lao People's Democratic Republic, Georgia, Thailand, United States of America, Canada, Mexico, New Zealand, Ghana, Korea, Republic of, OIRSA, Malawi, Burundi, Belize, Gabon
2.	G	Substantive	Suggest to supplement the relevant materials for this standard is not full.	This standard describe the identification of Genus Anastrepha. But there is large difference in the research for morphological classify and molecular biology, especially the complex species of Anastrepha fraterculus is in the researching. The scientific basis is disputed.	China
3.	G	Substantive	The standard is well- written and detailed in terms of diagnostics. The keys are relevant and they work. This standard is very relevant to the Caribbean Paragraph 46: It is recommended that another clearing agent other than xy	The use of xylene is being phased out due to its carcinogenic property. The diagrams appear after the captions and usually on the other page.	Jamaica, Trinidad and Tobago, Saint Kitts And Nevis, Dominica, Barbados, Antigua and Barbuda
			lene be used. It is recommended that the labels on the Figures are consistent with the ch aracters mentioned in the keys. E.g. [204] Figure 2 The captions for the Figures should be placed beneath the relevant diagram		

Comm no.	Para no.	Comment type	Comment	Explanation	Country
4.	G	Substantive	This Diagnostic Protocol (DP) presents the characteristics of the most economically relevant species. However there are many other species, what means that we can find Anastrepha species that are not contemplated in the protocol. As it is very difficult to get key to all species, we suggest (since it was a group of experts that drew up the protocol) that should be placed as an annex, a key that includes the largest number of species as possible. For species key in the larval stage it would be recommended to include optical microscope images in addition to electron microscope photos, because they show better how the structures of the key would be seen. Observation with electron microscope is not a routine procedure and in many countries is costly, so that is not used in routine daily work. Therefore it would be convenient to have images showing the key structures under optical microscope. We are proposing to include some new figures, and if the proposal is accepted, numbering of Figures should be fixed accordingly. It would also be useful to include an identification key for adults of Anastrepha. Although the PD includes a genus description it may be useful to include a key for differentiate the Anastrepha genus from other Tephritidae genus. In this regard we propose the TPDP to consider the inclusion of Hernandez-Ortiz key.	This Diagnostic Protocol (DP) presents the characteristics of the most economically relevant species. However there are many other species, what means that we can find Anastrepha species that are not contemplated in the protocol. As it is very difficult to get key to all species, we suggest (since it was a group of experts that drew up the protocol) that should be placed as an annex, a key that includes the largest number of species as possible. For species key in the larval stage it would be recommended to include optical microscope images in addition to electron microscope photos, because they show better how the structures of the key would be seen. Observation with electron microscope is not a routine procedure and in many countries is costly, so that is not used in routine daily work. Therefore it would be convenient to have images showing the key structures under optical microscope. We are proposing to include some new figures, and if the proposal is accepted, numbering of Figures should be fixed accordingly. It would also be useful to include an identification key for adults of Anastrepha. Although the PD includes a genus description it may be useful to include a key for differentiate the Anastrepha genus from other Tephritidae genus. In this regard we propose the TPDP to consider the inclusion of Hernandez-Ortiz key. t	COSAVE, Uruguay, Brazil, Peru, Chile, Argentina
5.	G	Substantive		1) Suggest using "A. fraterculus species complex" instead of a number of different names as used in this protocol such as "A. fraterculus sensu lato" in paragraph [79], or "A. fraterculus (species complex)" in paragraph [132], or "A. fraterculus" in paragraph [134] and [136], and adding a description of the features of each known local population to appropriately reflect the recent progress in taxonomic research on A. fraterculus. 2) Add clear pictures or figures of the habitus (thorax in dorsal aspect, abdominal tergites and wings) of every species, as such pictures or figures are useful for	Japan

Comm no.	Para no.	Comment type	Comment	Explanation	Country	
				identification. 3) Point out the names of parts using arrows in the pictures or figures.		
6.	1	Substantive	Draft Annex to ISPM 27:2006 – <u>Major economic significance species</u> of genus AnastrephaGenus Anastrepha (2004-015)	The text was written only including 7 economic significance species of genus Anastrepha.	China	
7.	6	Editorial	The family Tephritidae, members of which are commonly known as true fruit flies, comprises about 4 450 species in 500 or so genera (Norrbom <i>et al.</i> , 1999a, 1999b; Norrbom, 2004a) (the figure is about 4 700 species currently, A.L. Norrbom, pers. comm., XXXX2014). The Tephritidae are distributed worldwide in temperate, tropical and subtropical regions. <i>Anastrepha</i> Schiner (Tephritidae: Toxotrypanini) is the largest genus of Tephritidae in the Americas, and is represented by more than 250 species that occur from the southern United States (Texas and Florida) to northern Argentina (Foote <i>et al.</i> , 1993; Hernández-Ortiz, 1992; Hernández-Ortiz and Aluja, 1993; Norrbom, 2004a; Norrbom <i>et al.</i> , 2012). At least <u>sevensix</u> species of <i>Anastrepha</i> are considered major economic pests because of the great importance of the cultivated fruits they attack (e.g. mango and citrus) and their wide host range; for example, the Mexican fruit fly, <i>A. ludens</i> (Loew); the West Indian fruit fly, <i>A. obliqua</i> (Macquart); the Caribbean fruit fly, <i>A. suspensa</i> (Loew), the guava fruit fly, <i>A. striata</i> Schiner, the sapodilla fruit fly, <i>A. serpentina</i> (Wiedemann); the melon fruit fly, <i>A. grandis</i> (Macquart); and the South American fruit fly, <i>A. fraterculus</i> (Wiedemann). The latter has been recognized as a cryptic species complex (Hernández- Ortiz <i>et al.</i> , 2004, 2012). This diagnostic protocol for <i>Anastrepha</i> covers morphological identification of the genus and the species of major economic concern. For further general information about species of Tephritidae, see Norrbom (2010).	 Superfluous text. 2. Date needed for pers comm. Seven species: A. ludens, A. obliqua, A. suspensa, A. striata, A. serpentina, A. grandis and A. fraterculus, or refer to A fraterculus separately. 4. Reference to common names is not necessary, complicates text and may generate difficulties in translation. 	EPPO, European Union, Georgia, Serbia	
8.	6	Editorial	The family Tephritidae, members of which are commonly known as true fruit flies, comprises about 4 450 species in 500 or so genera (Norrbom <i>et al.</i> , 1999a, 1999b; Norrbom, 2004a) (the figure is about 4 700 species currently, A.L. Norrbom, pers. comm., XXXX). The Tephritidae are distributed worldwide in temperate, tropical and subtropical regions. <i>Anastrepha</i> Schiner (Tephritidae: Toxotrypanini) is the largest genus of Tephritidae in the Americas, and is represented by more than 250 species that occur from the southern United States (Texas and Florida) to northerm Argentina (Foote <i>et al.</i> , 1993; Hernández-Ortiz, 1992; Hernández-Ortiz and Aluja, 1993; Norrbom, 2004a; Norrbom <i>et al.</i> , 2012). At least sixseven species of <i>Anastrepha</i> are considered major economic pests because of the great importance of the cultivated fruits they attack (e.g. mango and citrus) and their wide host range; for example, the Mexican fruit fly,	This protocol explains 7 species.	Japan	

Comm no.	nm Para Comment . type no.		Comment	Explanation	Country	
			A. ludens (Loew); the West Indian fruit fly, A. obliqua (Macquart); the Caribbean fruit fly, A. suspensa (Loew); the guava fruit fly, A. striata Schiner; the sapodilla fruit fly, A. serpentina (Wiedemann); the melon fruit fly, A. grandis (Macquart); and the South American fruit fly, A. fraterculus (Wiedemann). The latter has been recognized as a cryptic species complex (Hernández-Ortiz et al., 2004, 2012). This diagnostic protocol for Anastrepha covers morphological identification of the genus and the species of major economic concern. For further general information about species of Tephritidae, see Norrbom (2010).			
9.	7	Technical	The length of the tephritid life cycle varies according to genusetype as well as environmental and climatic conditions(Basso, 2003). Female Anastrepha deposit their eggs inside fruits. The number of eggs deposited per fruit is variable, and depends mainly on features of the host fruit such as size and ripeness (Malavasi <i>et al.</i> , 1983), but each species also seems to have innate limits on the number of eggs laid (Aluja <i>et al.</i> , 1999). Within several days, deposited eggs hatch and larvae emerge. Larvae usually feed on fruit pulp, but in some cases also or exclusively on seeds. Mature larvae usually leave the fruit to pupate in the ground, but in certain cases pupation can take place within the fruit. Adults usually emerge after a pupal period of 16– 25 days, and they require a period of sexual maturation of 5–20 days after emergence. During this process the flies obtain food from homopteran secretions, bird faeces, and juice produced by ripe fruits (Prokopy and Roitberg, 1984).	"Genotype" seems too specific - there are many gentoypes in a species. Definitely life cycles vary between genera.	EPPO, European Union, Georgia, Serbia	
10.	9	Editorial	The introduction of some cultivated exotic species such as <i>Mangifera indica</i> and <i>Citrus</i> spp. have allowed some pest species of <i>Anastrepha</i> to expand their original areas of distribution and enhance their reproductive potential. However, they still have marked preferences for certain native hosts, which is probably indicative of their original host relationships. In this regard, the species <i>A. suspensa, A. fraterculus</i> and <i>A. striata</i> breed mainly in hosts belonging to the family Myrtaceae, <i>A. ludens</i> in the Rutaceae, <i>A. obliqua</i> in the Anacardiaceae, <i>A. serpentina</i> in the Sapotaceae, and <i>A. grandis</i> in the Cucurbitaceae (Norrbom, 2004b).	"A. triata" to be replaced by "A. striata" (cf. paragraphs [6] ret [19]).	EPPO, European Union, Georgia, Serbia	
11.	9	Editorial	The introduction of some cultivated exotic species such as <i>Mangifera indica</i> and <i>Citrus</i> spp. have allowed some pest species of <i>Anastrepha</i> to expand their original areas of distribution and enhance their reproductive potential. However, they still have marked preferences for certain native hosts, which is probably indicative of their original host relationships. In this regard, the species <i>A. suspensa, A. fraterculus</i> and <i>A. Striata</i> breed mainly in hosts belonging to the family Myrtaceae, <i>A. ludens</i> in the Rutaceae, <i>A. obliqua</i> in the Anacardiaceae, <i>A. serpentina</i> in the Sapotaceae, and <i>A. grandis</i> in the	The correct scientific name is "Striata".	COSAVE, Uruguay, Brazil, Peru	

		Comment type	Comment	Explanation	Country	
			Cucurbitaceae (Norrbom, 2004b).			
12.	9	Editorial	The introduction of some cultivated exotic species such as <i>Mangifera indica</i> and <i>Citrus</i> spp. have allowed some pest species of <i>Anastrepha</i> to expand their original areas of distribution and enhance their reproductive potential. However, they still have marked preferences for certain native hosts, which is probably indicative of their original host relationships. In this regard, the species <i>A. suspensa, A. fraterculus</i> and <i>A. <u>striata</u> breed mainly in hosts belonging to the family Myrtaceae, <i>A. ludens</i> in the Rutaceae, <i>A. obliqua</i> in the Anacardiaceae, <i>A. serpentina</i> in the Sapotaceae, and <i>A. grandis</i> in the Cucurbitaceae (Norrbom, 2004b).</i>	The correct scientific name is "striata".	Chile	
13.	9	Editorial	The introduction of some cultivated exotic species such as <i>Mangifera indica</i> and <i>Citrus</i> spp. have allowed some pest species of <i>Anastrepha</i> to expand their original areas of distribution and enhance their reproductive potential. However, they still have marked preferences for certain native hosts, which is probably indicative of their original host relationships. In this regard, the species <i>A. suspensa, A. fraterculus</i> and <i>A. triatastriata</i> breed mainly in hosts belonging to the family Myrtaceae, <i>A. ludens</i> in the Rutaceae, <i>A. obliqua</i> in the Anacardiaceae, <i>A. serpentina</i> in the Sapotaceae, and <i>A. grandis</i> in the Cucurbitaceae (Norrbom, 2004b).	Editorial correction	Japan	
11.	9	Editorial	The introduction of some cultivated exotic species such as <i>Mangifera indica</i> and <i>Citrus</i> spp. have allowed some pest species of <i>Anastrepha</i> to expand their original areas of distribution and enhance their reproductive potential. However, they still have marked preferences for certain native hosts, which is probably indicative of their original host relationships. In this regard, the species <i>A. suspensa, A. fraterculus</i> and <i>A. Striata</i> breed mainly in hosts belonging to the family Myrtaceae, <i>A. ludens</i> in the Rutaceae, <i>A. obliqua</i> in the Anacardiaceae, <i>A. serpentina</i> in the Sapotaceae, and <i>A. grandis</i> in the Cucurbitaceae (Norrbom, 2004b).	The correct scientific name is "Striata".	Argentina	
15.	9	Technical	The introduction of some cultivated exotic species such as <i>Mangifera indica</i> and <i>Citrus</i> spp. have allowed some pest species of <i>Anastrepha</i> to expand their original areas of distribution and enhance their reproductive potential. However, they still have marked preferences for certain native hosts, which is probably indicative of their original host relationships. In this regard, the species <i>A. suspensa, A. fraterculus</i> and <i>A. striata</i> breed mainly in hosts belonging to the family Myrtaceae, <i>A. ludens</i> in the Rutaceae, <i>A. obliqua</i> in the Anacardiaceae, <i>A. serpentina</i> in the Sapotaceae, and <i>A. grandis</i> in the Cucurbitaceae (Norrbom, 2004b).	The 's' is missing from A. striata	Jamaica, Trinidad and Tobago, Saint Kitts And Nevis, Dominica, Barbados, Antigua and Barbuda	
16.	10	Editorial	Among native hosts in the American tropics, there seems to be an ancestral association with plants that produce latex and particularly the	The term "group" is not defined. Or replace "group" by "specles complex" in the last sentence.	EPPO, European	

Comm no.	Para no.	. type			Explanat	ion	Country	
			family Sapotaceae. Sapotaceous fruits are frequent hosts of species of the <i>dentata, leptozona, serpentina, daciformis, robusta</i> and <i>cryptostrepha</i> groups (cryptic species complexes). The Myrtaceae are also very important hosts: about 26 <i>Anastrepha</i> species, in particular belonging to the <i>fraterculus</i> group, have been reported in plants belonging to this family (Norrbom and Kim, 1988; Norrbom <i>et al.</i> , 1999c).					Union, Georgia, Serbia
17.	19	Editorial					te (1942) is missing for the description of es A. lathana which is a synonym of A.	EPPO, European Union, Georgia, Serbia
			Common name	Anastrepha species	Synonyms			
			South American fruit fly	Anastrepha fraterculus (Wied emann, 1830)	Tephritis mellea Walker, 1837Trypeta unicolor Loew, 1862Anthomyia frutalis Weyenbergh, 1874Anastrepha fraterculus var. soluta Bezzi, 1909Anastrepha peruviana Townsend, 1913Anastrepha braziliensis Greene, 1934Anastrepha costarukmanii Capoor, 1954Anastrepha scholae Capoor, 1955Anastrepha pseudofraterculus			

Comm no.	Para no.	Comment type	Comment			Explanation	Country
					Capoor, 1955 Anastrepha lambayecae Korytkowski and Ojeda, 1968		
			Melon fruit fly	Anastrepha grandis (Macquart, 1846)	Anastrepha schineri Hendel, 1914 Anastrepha latifasciata Hering, 1935		
			Mexican fruit fly	Anastrepha Iudens (Loew, 1873)	Anastrepha lathana Stone <u>1942</u>		
			West Indian fruit fly	Anastrepha obliqua (Macquart, 1835)	Anastrepha mombinpraeoptans Sein, 1933 Anastrepha fraterculus var. ligata Lima, 1934 Anastrepha trinidadensis Greene, 1934		
			Sapodilla fruit fly	Anastrepha serpentina (Wiedemann, 1830)	<i>Urophora vittithorax</i> Macquart, 1851		
			Guava fruit fly	Anastrepha striata Schiner, 1868	<i>Dictya cancellaria</i> Fabricius, 1805 (see Norrbom <i>et al.</i> , 1999b)		
			Caribbean fruit fly	Anastrepha suspensa (Loew, 1862)	Anastrepha unipuncta Sein, 1933 Anastrepha longimacula		

Comm no.	Para no.	Comment type	Comment			Explanation
					Greene, 1934	
8.	19	Substantive		names and synonym ing to the genus <i>Ana</i>	is of fruit fly species of major econor astrepha	nic Based on circumstances and h economic significance evaluati Anastrepha will be great differe
			Common name	Anastrepha species	Synonyms	
					Tephritis mellea Walker, 1837	
					Trypeta unicolor Loew, 1862	
					Anthomyia frutalis Weyenbergh, 1874	
					Anastrepha fraterculus var. soluta Bezzi, 1909	
					Anastrepha peruviana Townsend, 1913	
			South American fruit fly	Anastrepha fraterculus(Wiede mann, 1830)	Anastrepha braziliensis Greene, 1934	
					Anastrepha costarukmanii Capoor, 1954	
					Anastrepha scholae Capoor, 1955	
					Anastrepha pseudofraterculus Capoor, 1955	
					Anastrepha lambayecae Korytkowski and Ojeda, 1968	
			Melon fruit fly	Anastrepha	Anastrepha schineri Hendel,	

Comm no.	Para no.	Comment type	nt Comment			Explanation	Country
				grandis (Macquart, 1846)	1914 <i>Anastrepha latifasciata</i> Hering, 1935		
			Mexican fruit fly	Anastrepha Iudens (Loew, 1873)	Anastrepha lathana Stone <u>1942</u>		
			West Indian fruit fly	Anastrepha obliqua (Macquart, 1835)	Anastrepha mombinpraeoptans Sein, 1933 Anastrepha fraterculus var. ligata Lima, 1934 Anastrepha trinidadensis Greene, 1934		
			Sapodilla fruit fly	Anastrepha serpentina (Wiedemann, 1830)	<i>Urophora vittithorax</i> Macquart, 1851		
			Guava fruit fly	Anastrepha striata Schiner, 1868	<i>Dictya cancellaria</i> Fabricius, 1805 (see Norrbom <i>et al.</i> , 1999b)		
			Caribbean fruit fly	Anastrepha suspensa	<i>Anastrepha unipuncta</i> Sein, 1933		
				(Loew, 1862)	Anastrepha longimacula Greene, 1934		

omm o.	Para no.	Comment type	Comment			Explanation	Country
			genus Anastrepha,	and to consider whe	ether this protocol should add the		
_					ia, A. antunesi, A. antunesi etc.		
19.	19	Technical		names and synonym jing to the genus <i>An</i> a		mic The coorect scientific name is "Anastrepha fraterculus var mombinpraeoptans Sein". We propose to add other synonyms.	ulus COSAVE, Uruguay, Brazil, Peru, Chile, Argentina
			Common name	Anastrepha species	Synonyms		
			South American fruit fly	Anastrepha fraterculus (Wiede mann, 1830)	Tephritis mellea Walker, 1837 Trypeta unicolor Loew, 1862 Anthomyia frutalis Weyenbergh, 1874 Anastrepha fraterculus var. soluta Bezzi, 1909 Anastrepha peruviana Townsend, 1913 Anastrepha braziliensis Greene, 1934 Anastrepha costarukmanii Capoor, 1954 Anastrepha scholae Capoor, 1955 Anastrepha pseudofraterculus Capoor, 1955		
					Anastrepha lambayecae Korytkowski and Ojeda, 1968		
			Melon fruit fly	Anastrepha	Anastrepha schineri Hendel.		

Comm no.	Para no.	Comment type	Comment			Explanation	Country
				grandis (Macquart, 1846)	1914 <i>Anastrepha latifasciata</i> Hering, 1935		
			Mexican fruit fly	Anastrepha Iudens (Loew, 1873)	Anastrepha lathana Stone <u>1942</u>		
			West Indian fruit fly	Anastrepha obliqua (Macquart, 1835)	Anastrepha mombinpraeoptans Sein, 1933 Anastrepha fraterculus var. ligata Lima, 1934 Anastrepha trinidadensis		
			Sapodilla fruit fly	Anastrepha serpentina (Wiedemann, 1830)	Greene, 1934 Urophora vittithorax Macquart, 1851		
			Guava fruit fly	Anastrepha striata Schiner, 1868	<i>Dictya cancellaria</i> Fabricius, 1805 (see Norrbom <i>et al.</i> , 1999b)		
			Caribbean fruit fly	Anastrepha suspensa (Loew, 1862)	Anastrepha unipuncta Sein, 1933 Anastrepha longimacula		
20.	22	Technical	shipments, in bagg vehicles. Fruits with have originated fro	s. Infested fruits car age, and even on a soft areas, dark sta m female ovipositior	Greene, 1934 a be found in imported or exported eroplanes or terrestrial transportation ains, rot, orifices or injuries that might a or larval feeding activities are targeted ures made by female flies during	1. Cf. ISPM 5. 2. Clarity. The paragraph is specifically on detection.	EPPO, European Union, Georgia, Serbia

Comm no.	Para no.	Comment type	Comment	Explanation	Country
			oviposition, the <u>visual examination</u> test-should be done under a microscope by a specialist. 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 opened to search for eggs or larvae inside. The success of <u>detection phytosanitary measures</u> -depends on careful sampling and examination of fruits.		
21.	23	Editorial	Inspection of traps. Guidance on trapping <i>Anastrepha</i> fruit flies for establishment of pest free areas is given in Appendix 1 of ISPM 26:2006. In general, monitoring systems established for the detection of fruit fly adults in trees, either in fruit-growing regions or in border areas between countries, require the utilization of McPhail traps baited with food attractants or synthetic lures. The baits, often with rich sources of ammonium, should be recognized and approved internationally (e.g. ISPM 26:2006). The specific methods of trap deployment and time of service of the traps must be in agreement with the national phytosanitary regulations in use by each country.	Simpler.	EPPO, European Union, Georgia, Serbia
22.	23	Technical	establishment of pest free areas is given in Appendix 1 of ISPM 26:2006. In general, monitoring systems established for the detection of fruit fly adults in	for trapping procedures for fruit fly species (Tephritidae) of economic importance under different pest statuses	COSAVE, Uruguay, Brazil, Peru, Chile, Argentina
23.	28	Technical	To study this idea further, the International Atomic Energy Agency (IAEA) is coordinating an international research project to describe the cryptic species in the <i>Anastrepha fraterculus</i> complex. As part of this project, molecular methods are being examined for diagnostic utility within the genus. Based on	Database (BOLD), with 82 x species currently with	Australia
24.	32	Editorial	The fruits to be examined are placed in cages covered with cloth or fine mesh and that have a sterile pupation medium (e.g. damp vermiculite, sand or sawdust) at the bottom. Once the larvae emerge from the fruit, they will move	Simpler wording	EPPO, European Union, Georgia, Serbia

Comm no.	Para no.	Comment type	Comment	Explanation	Country
			to the substratum for pupation. It is recommended to incubate each fruit separately. Each sample must 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 48–72 h to ensure that the tegument and wings acquire the rigidity and characteristic coloration of the species. The adults are then killed and preserved by placing them in 70% ethanol, or they are killed with ethyl acetate or another agent and then mounted on pins. For female flies, immediately after killing them (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 (so that it does not need to be dissected later).		
25.	32	Technical	The fruits to be examined are placed in cages covered with cloth or fine mesh	For molecular studies the percentage of ethanol used is 96.	COSAVE, Uruguay, Brazil, Peru, Chile, Argentina
26.	36	Technical	It is preferable to cut off the whole abdomen from a female to dissect the	Potassium hydroxide is also used to remove the internal contents.	Japan

Comm no.	Para no.	Comment type	Comment	Explanation	Country
			transferred to a microvial with glycerine and pinned under the mounted dry specimen. For permanent slides, proceed as described in section 4.1.2. Mounting the aculeus permanently in the ventral position prevents the observation of some characters better seen in lateral view. For this reason, preservation in glycerine in a microvial is often preferable.		
27.	38	Substantive	For permanent slides, proceed as described in section 4.1.2.1, avoiding the NaOH solution. Wing characters can usually be observed without mounting, so mounting is not recommended as a general practice. It may be necessary for morphometric studies, but it is not necessary for observation of the characters used in the key in section 4.3.2. If permanent mounts are made, it is recommended to cut off one of the wings from its base (the right wing is preferred because it facilitates comparison with images reported in the literature and this diagnostic protocol).	Section 4.1.2.1 does not have any information applicable to the preparation of wings for microscopic examination.	Jamaica, Trinidad and Tobago, Saint Kitts And Nevis, Dominica, Barbados, Antigua and Barbuda
28.	42	Technical	Morphological examination of larvae (section 4.2.2) can be performed on unmounted larvae using a stereomicroscope, on slide-mounted larvae using a <u>compound</u> microscope, or on critical-point dried larvae using a scanning electron microscope (SEM). Slide mounting larvae can preclude subsequent analysis of morphological characters. On slide-mounted larvae it is possible to examine external morphology (e.g. anterior and posterior spiracles, oral ridges) as well as internal structures such as the cephalopharyngeal skeleton (Figures 21–44), using an optical microscope with objective 20x, 40x or higher. Detailed, high resolution observation of the external morphology of larvae is only possible using an SEM (Figures 45–61). It is therefore not recommended to slide mount all specimens representing a sample or the only larva available for diagnosis; unmounted larvae should be kept for future analysis.		Jamaica, Trinidad and Tobago, Saint Kitts And Nevis, Dominica, Barbados, Antigua and Barbuda
29.	44	Editorial	To prepare specimens for examination _⊥ the larvae must be treated in hot water, which can be accomplished by placing live larvae in water <u>atef</u> approximately 65° C for 2–4 min. The larvae are cooled to room temperature and then immersed in 50% alcohol for 15–30 min. The specimens are transferred to a hermetic vial (15–25 ml) filled with 70% alcohol. It is advisable to include a label on the vial with all sampling information. These samples are ready for examination under a stereomicroscope or subsequent preparation for slide mounting or examining under an SEM.		EPPO, European Union, Georgia, Serbia
30.	45	Technical	To prepare specimens for slide mounting, it is necessary to remove (clean) all the internal contents to allow observation of the cuticle, oral opening, cephalopharyngeal skeleton and anterior spiracles, as well as the posterior spiracular plate and anal lobes. This can be accomplished by making two transverse incisions in the larva, one behind the cephalic region and the anterior spiracles, and one before the caudal segment. The incised larva then		Japan

Comm no.	Para no.	Comment type	Comment	Explanation	Country
			needs to be immersed in a test tube containing 10% NaOH <u>or KOH</u> solution and heated in a boiling water bath for 10–15 min. The internal contents can then be carefully removed from the specimen using forceps and distilled water under a stereomicroscope (45x magnification or greater).		
31.	46	Technical	Before doing this, cleaned structures must be dehydrated by placing them for	more) rather than in xylene.	EPPO, European Union, Georgia, Serbia
32.	46	Technical	Permanent slide mounts can be made using Canada balsam or Euparal. Before doing this, cleaned structures must be dehydrated by placing them for 25 min in each of 50%, 75% and 100% ethanol. For mounting with Canada balsam, the specimens should be transferred to absolute xylene for 3–5 min to clear them and then immediately mounted on a slide with one or two drops of Canada balsam. When Euparal is used as the mounting medium, structures should be transferred from 100% ethanol to clove oil for about 30 min to clear them before mounting. In both cases, slides must be allowed to dry for several days (the time can be reduced by using an oven), but they can be examined under the microscope at low magnification immediately after mounting. Slides should be labelled.		Jamaica
33.	48	Substantive	4.3 Morphological identification of adults	As diagnostic protocol of genus Anastrepha, approximate genus morphology characters of Tephritidae should be provided.	China
34.	51	Substantive		Capital letters are used for both longitudinal and cross veins in Fig. 4. Lowercase letters are used for cells of wings in Fig. 4.	Japan

Comm no.	Para no.	Comment type	Comment	Explanation	Country
			to apex of wing; and V-band forming an inverted V shape, comprising the proximal arm (subapical band) along vein $\frac{dm-cuDM-CU}{dm-cu}$ and the distal arm (posterior apical band) arising from cell <i>m</i> , both are convergent in cell $\frac{R_{I_{4+5}}}{dm}$; distal arm frequently incomplete or absent. The typical wing pattern is modified in some economically important species (see key to species).		
35.	68	Editorial	4- Anterior apical band of wing (=distal section of S-band) narrow to moderately broad, never reaching apex of vein M ; V-band with arms separated anteriorly or if joined, with large hyaline mark between them and vein $M_{\tau_{i}}$ Secuto-scutellar suture with or without brown spot medially; aculeus variable.	1) Cf. paragraph [70]. 2) Cf. paragraph [70].	EPPO, European Union, Georgia, Serbia
36.	70	Editorial	Anterior apical band of wing (=distal section of S-band) extremely wide, reaching apex of vein <i>M</i> ; V-band broad and complete, with arms widely connected anteriorly, hyaline mark between them and vein <i>M</i> small or absent (Figure 10); scuto-scutellar suture usually with large rounded brown spot medially; female aculeus 1.4–1.6 mm long (Figure 17), tip 0.19–0.23 mm long, 0.10–0.13 mm wide, lateral margins serrate on distal 0.50–0.65 (Figure 17).		EPPO, European Union, Georgia, Serbia
37.	78	Editorial	Both mediotergite and subscutellum with broad dark brown to black markings on sides (Figure 3A); brown spot on scuto-scutellar suture usually present _{τ_{1}} aculeus 1.4–1.9 mm long ₁ ; aculeus tip 0.20–0.28 mm long ₁ ; lateral margins with 8 to14 teeth on distal two-fifths to three-fifths (Figure 20); wing pattern variable (Figure 13).	Consistency with paragraph 70	EPPO, European Union, Georgia, Serbia
38.	81	Substantive		The Dacus genus is one of important quarantine fruit fly groups in the world.	China
39.	100	Editorial		Why is there a "*"?	EPPO, European Union, Georgia, Serbia
40.	105	Substantive	opening, or dental sclerite conspicuous (Figures 45, 47); and/or caudal tubercles strongly developed; or larva taken from papaya with caudal tuberclesridges lacking and caudal sensilla strongly reduced.	The term "caudal ridges" is used in Figs. 59 and 60. Change "caudal tubercles" to"caudal ridges" based on White et al. (2000) Glossary. pp. 881-924. M. Aluja & A.L. Norrbom (ed.), Fruit Flies (Tephritidae): Phylogeny and Evolution of Behavior.	Japan
41.	111	Editorial	Dorsal spinules absent on all abdominal segments, or if present, only in segment A1 (Abdominal segment)(some specimens of <i>A. ludens</i>).	For more clarity.	Japan
42.	121	Editorial		Clearer.	EPPO, European Union, Georgia, Serbia

Comm no.	Para no.	Comment type	Comment	Explanation	Country
			<i>indica</i> ; distribution: southern Texas, (USA) to Panama.)		
43.		Editorial	anterior spiracles with 9 to 15 tubules (Figure 41); posterior spiracular slits 2.5–3.5 times longer than wide (Figure 42). Cephalopharyngeal skeleton as in Figure 29. (Main hosts: larvae breed in fruits of Myrtaceae; distribution: Florida, (USA) and Antilles.)		EPPO, European Union, Georgia, Serbia
44.	125	Substantive	(average, 8) (Figure 36); oral ridges usually in 12 to 14 rows; anterior spiracle with 13 to 19 tubules in a single row (Figure 35); anal lobes usually bilobed (as in Figure 57). Cephalopharyngeal skeleton as in Figure 30. (Main hosts: larvae breed in fruits of Sapotaceae; distribution: tropical Americas.)	basal and apical branched parts.	
45.	153	Editorial	 Allen L. Norrbom (Systematic Entomology Laboratory <u>(SEL)</u>, United States Department of Agriculture (USDA), <u>Smithsonian Institution</u>, Washington, DC, USA) 	1) More precise (cf. paragraph [2]). 2) More precise (cf. paragraphs [2] et [156]).	EPPO, European Union, Georgia, Serbia
46.	157	Editorial	- Gary Steck (Florida Department of Agriculture and Consumer Services, Division of Plant Industry-, Gainesville, FL, USA)	Consistency	EPPO, European Union, Georgia, Serbia
47.	159	Editorial	Division, Department of Primary Industries, Knoxfield Centre, Melbourne, Victoria, Australia) .	New paragraph [159]bis suggested if relevant: please see paragraph [2], "Consultation on technical level": has Ms Valérie Balmès been forgotten in the section "7. Acknowledgements" or has she commented to a lesser extent than the other experts?	EPPO, European Union, Georgia, Serbia
			 Valérie Balmès (Anses, Laboratoire de la santé des végétaux, Unité entomologie et plantes invasives, Montpellier, France). 		
48.	160	Editorial	8. References	Check the order of references (alphabetical) e.g. rows 175 and 176 should be after row 181. Also check the order of rows 186, 189,190.	EPPO, European Union, Georgia, Serbia
49.	190	Editorial	Norrbom, A.L., Korytkowski, C.A., Zucchi, R.A., Uramoto, K., Venable, G.L., McCormick, J. & Dallwitz, M.J. 2012. Onwards. <i>Anastrepha</i> and <i>Toxotrypana</i> : descriptions, illustrations, and interactive keys. Version 31 August 2012. Available at <u>http://delta-intkey.com</u> (last accessed XXX_date ???).	Last accessed at which date?	EPPO, European Union, Georgia, Serbia
50.	198	Editorial		The legend of the figures should be below and not above the figures.	EPPO, European Union, Georgia, Serbia

Comm no.	Para no.	Comment type	Comment	Explanation	Country
51.	203	Editorial	Source: Figure 1(<u>A)</u> adapted from Hernández-Ortiz <i>et al.</i> (2010); Figures 2 and 3 adapted from Hernández-Ortiz (1992).	See paragraph [202].	EPPO, European Union, Georgia, Serbia
52.	214	Substantive	Figures 21–26. (21) Morphology of the cephalopharyngeal skeleton of third in third instar larvae, lateral view: (22) Ceratitis capitata; (23) Anastrepha obliqua; (25) Rhagoletis tomatis; (26) Toxotrypana sp. At, apical tooth; DC, dorsal cornu. HS, hypopharyngeal sclerite; MD, mandible; Mn, mandibular neck; PB, parast Va, ventral Apodeme; VC, ventral cornu. Source: All figures adapted from Frías <i>et al.</i> (2006).	(1987), hypotheratorsand in paragraphs [105] and [1987], hypotheratorsand	Japan

Comm no.	Para no.	Comment type	Comment	Explanation	Country
53.	221	Technical			COSAVE, Uruguay, Brazil, Peru
54.	221	Technical	Figures 45–5051. (45, 47, 48) Cephalic segment of third instar larvae. (46,	To adjust numeration and because a new figure is proposed to be added.	Chile
53.	221	Technical		To adjust numeration and because a new figure is proposed to be added.	Argentina

Comm no.	Para no.	Comment type	Comment	Explanation	Country
56.	223	Technical	<image/> <section-header></section-header>	New figure 51 is being proposed to be added.	COSAVE, Uruguay, Brazil, Peru, Chile, Argentina

Comm no.	Para no.	Comment type	Comment	Explanation	Country
57.	224	Technical	Figures 51–56. Anterior spiracles of the first thoracic segment, third instar larvae: (51)Anastrepha ludens; (52)Anastrepha fraterculus; (53)Toxotrypana curvicauda; (54)Rhagoletis conversa; (55) Ceratitis capitata; (56)Bactrocera cucurbitae. <u>New Figure: Anastrepha fraterculus under optical microscope</u>	New figure is proposed to be added. It is necessary to adjust the numbers of figures.	COSAVE, Uruguay, Brazil, Peru

Comm no.	Para no.	Comment type	Comment	Explanation	Country
58.		Technical	curvicauda; (54) Rhagoletis conversa; (55) Ceratitis capitata; (56) Bactrocera cucurbitae. New Figure: Anastrepha fraterculus under optical microscope	New figure is proposed to be added. It is necessary to adjust the numbers of figures.	Chile
57.	224	Technical	Figures 51–56. Anterior spiracles of the first thoracic segment, third instar larvae: (51)Anastrepha ludens; (52)Anastrepha fraterculus; (53)Toxotrypana curvicauda; (54)Rhagoletis conversa; (55) Ceratitis capitata; (56)Bactrocera cucurbitae. New Figure: Anastrepha fraterculus under optical microscope	New figure is proposed to be added. It is necessary to adjust the numbers of figures.	Argentina
60.	226	Technical	<image/>	number of figures.	COSAVE, Uruguay, Brazil, Peru, Chile, Argentina

Comm no.	Para no.	Comment type	Comment	Explanation	Country
61.	227	Technical	Figures 57–61. (57) Anal lobes bifids, <i>Anastrepha striata</i> ; (58) Anal lobes entire, <i>Anastrepha obliqua</i> ; (59) caudal ridges absent, <i>Anastrepha suspensa</i> ; (60) caudal ridges present, <i>Bactrocera carambolae</i> ; (61) <i>Anastrepha striata</i> , dorsal view of third instar larva showing rows of dorsal spinules. <u>New Figure:</u> <u>Anastrepha fraterculus</u> , <u>dorsal view of third instar larva showing dorsal</u> spinules	New figure was added. It is necessary to adjust the numbers of figures.	COSAVE, Uruguay, Brazil, Peru
62.	227	Technical	Figures 57–61. (57) Anal lobes bifids, Anastrepha striata; (58) Anal lobes	New figure was added. It is necessary to adjust the numbers of figures.	Chile
61.	227	Technical	Figures 57–61. (57) Anal lobes bifids, Anastrepha striata; (58) Anal lobes	New figure was added. It is necessary to adjust the numbers of figures.	Argentina

Comm no.	Para no.	Comment type	Comment	Explanation	Country
64.	229	Technical	<image/>	New figure was added. It is necessary to adjust the numbers of the figures.	COSAVE, Uruguay, Brazil, Peru, Chile, Argentina

Comm no.	Para no.	Comment type	Comment	Explanation	Country