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THE DETECTION OF AN ALIEN PEST, THE COTTON LEAF ROLLER *HARITALODES DECORATA* (LEPIDOPTERA: CRAMBIDAE), ON THE BLACK SEA COAST OF RUSSIA

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Summary. Here we report about the first finding of an alien pest, the cotton leaf roller *Haritalodes derogata* (Fabricius, 1775) (Lepidoptera: Crambidae), on the Russian Black Sea Coast. In summer of 2021, the significant damage caused by *H. degorata* to *Hibiscus* spp. was documented in the ornamental plantations of Sochi. The current distribution data and the bionomics of the species in the invaded region are provided. The DNA barcoding data obtained for the specimens from Sochi are analyzed comparing to the species representatives from the other parts of the world.

Key words: Crambid moth, invasion, important pest, host plants, *Hibiscus, Tilia*, Sochi, European part of Russia.

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Резюме. В статье сообщается о первой находке на Черноморском побережье России опасного вредителя – хлопковой огневки *Haritalodes derogata* (Fabricius, 1775) (Lepidoptera: Crambidae). Летом 2021 г. в декоративных насаждениях г. Сочи зафиксированы значительные повреждения, наносимые вредителем гибискусам *Hibiscus* spp. В работе приведены сведения о распространении и биоэкологии чужеродного вида во вторичном ареале. ДНК-баркоды, полученные для образцов огневки.

INTRODUCTION

The cotton leaf roller, *Haritalodes derogata* (Fabricius) (Lepidoptera: Crambidae), is a polyphagous species known as an important pest of crop (especially cotton, cashew, jujube and others), herbacious and tree plants in ornamental plantings in many countries of the world, in particularly of Africa, Asia and Oceania (Dhindsa *at al*., 1980; Byun *at al*., 2008; Yamanaka, 2008; CAB International, 2022). In Russia for the first time *H. derogata* was found in Southern Primorye in the second half of the 19th century. The species was identified by the entomologist Otto Bremer (Saint Petersburg), who provided the detailed information about the moths in the Russian Far East (Bremer, 1864). In 2013, *H. derogata* was recorded in Transbaikalia (Korsun, 2017), where its origin remains questionable.

Here we report about the first detection of *H. derogata* in the European part of Russia, on the territory of the Black Sea Cost, where the pest has already established its population and presently provides noticeable damage to ornamental plants. The paper discusses bionomics and genetically characterizes the novel pest in the invaded region of Southern Russia.

MATERIAL AND METHODS

Sampling and rearing. The study was carried out in the ornamental plantations of Sochi in June–September 2021–2022. The survey was also done in Anapa, Tuapse and Red Polyana (Sochi) in September 2022. In Sochi, the plants of *Hibiscus* and *Tilia* spp. were examined on the territory of sanatorium-resort, parks, squares and street plantings across the territory from the Psou River on the south (from the border with Abkhazia) to the village Lazarevskoe on the north. The damage level was documented, especially when reaching up to 100% (estimated as a relative number of damaged leaves on the plants). Larvae found in the rolled leaves were randomly collected with their food substrate for rearing. Few larvae were preserved in 95% ethanol, and stored in the freezer at –20^{o}C prior DNA barcoding.

Feeding larvae were kept in the plastic boxes (500 ml volume) lined with filter paper and covered with mesh at a temperature of +26±2^{o}C and a relative humidity of 65±5%. The boxes were monitored every second day and the number of larval instars, the duration of larval stage, the date of pupation, and adult emergence were recorded. Additionally, adults were attracted to the light using led lamp (Thomson E14 8W 3000K 640 lm) in late evening (from 21:00 till 24:00) in ornamental plantings in two districts of Sochi: Khostinskiy and Lazarevskiy. The adults reared in laboratory and those attracted to the light were set and stored in the entomological collection of the Subtropical Scientific Centre, Sochi.

Species was identified based on external morphology of adults (Sinev & Streltsov, 2019). The identification was confirmed by S.Yu. Sinev (ZIN RAS, Saint Petersburg). The photographs of typical damage, immature stages and adults were taken with a Fujifilm X30 camera (Japan). The maps were built using ArcGIS 9.3 (Release 9.3. New York St., Redlands, CA. Environmental Systems Research Institute).

DNA barcoding. The two larvae of *H. derogata* sampled from *Hibiscus syriacus* and *Tilia caroliniana* were DNA barcoded to genetically characterize the species, define the nearest neighbors and estimate intraspecific divergence. DNA barcoding was performed at the Canadian Center for DNA barcoding (CCDB, Canadian Center for DNA barcoding) at the University of Guelph (Canada). The mitochondrial cytochrome oxidase I gene (mtDNA COI, 658 bp) was sequenced in the larvae using the primer set C_LepFolF/C_LepFolR using the standard protocol (de Waard *et al*., 2008). The sequences, trace files, biogeographic data and images of the vouchers were deposited in the Barcode of Life Data Systems (BOLD) (Ratnasingham & Hebert, 2007) and NCBI (National Centre for Biotechnology Information). All data are accessible in BOLD via the link dx.doi.org/10.5883/DS-HARDER.

For phylogenetic analysis, eight sequences of the specimens indicated in BOLD as *H. derogata*, originating from China (1 sequence), Japan (1), Papua New Guinea (1), Philippines (1), South Africa (2), and Australia (2) were involved to the analysis (Table 1). Barcode Index

Numbers (BINs) of the analyzed specimens were retrieved from BOLD (Ratnasingham & Hebert, 2013). The alignment was done in BioEdit 7.2.5 (Jeanmougin *et al*., 1998). A maximum likelihood tree was constructed in MEGA X (Kumar *et al*., 2018) using the maximum likelihood method, the Kimura two-parameter model, and a bootstrap method (1000 iterations). Intraspecific distances were estimated using the same approaches. The DNA barcode of a leaf mining moth *Phyllocnistis messaniella* (Zeller, 1846) (process ID CAMRU150-21) was used to root the phylogenetic tree (specimen data see in Table 1).

Table 1. The specimens of *Haritalodes derogata* involved into DNA barcoding and the related sequences borrowed from BOLD for comparison.

No.	**Process ID**	**Country**	**Locality**	**Collection Date**	**Collectors**	**GenBank Accession**
1	ANICN897-10	Australia	Bucasia	03.02.2008	K.J. Sandery	HQ953084
2	ANICN898-10	Australia	Bucasia	05.12.2004	K.J. Sandery	HQ953084
3	GBMNA10303-19	China	–	–	–	KF390987
4	CAMRU156-21	Russia	Sochi	08.08.2021	N. Kirichenko, N. Karpun, E. Zhuravleva	KR233479
5	CAMRU157-21	Russia	Sochi	12.08.2021		OP443583
6	GBGL12102-13	Japan	Ibaraki	28.08.2012	R. Nakano	OP443582
7	GBMNA27238-19	Philippines	–	–	–	AB751248
8	LPNGM338-07	Papua New Guinea	Misima Island	11.10.2002	Binatang Research Centre	MK459704
9	GWOTI530-12	South Africa	–	–	A. Hausmann	JX970307
10	LSAFR620-12	South Africa	Hekpoort	14.II.2012	P. Hebert, J. deWaard	OP443581
			Outgroup			
11	CAMRU150-21	Russia	Sochi	08.08.2021	N. Kirichenko, N. Karpun, E. Zhuravleva	OP443580

Remark. For each specimen, Process ID is provided linking the records in the BOLD database with the voucher specimen data; (–) – no data available.

RESULTS AND DISCUSSION

***Haritalodes derogata* (Fabricius, 1775)**
Figs 1–4

MATERIAL EXAMINED. **Russia:** Sochi, Nagornaya str., 09.VII 2021, larvae from *Hibiscus syriacus*, 7♂, 9♀, adults emerged 10-15.VIII 2021, N. Karpun, E. Zhuravleva, E. Shoshina coll.; Sochi, Yana Fabritsiusa str., 12-13.VIII 2021, larvae from *H. syriacus,* 14♂, 17♀, adults emerged 23-29.VIII 2021, 1 larva DNA barcoded (Sample ID: NK1072; Process ID: CAMRU157-21), E. Zhuravleva, E. Shoshina coll.; ibidem, 13.VIII 2021, larvae from *Abutilon hybridum*, 4♂, 5♀, adults emerged 23-30.VIII 2021, N. Karpun, E. Zhuravleva, coll.; ibidem, 15.VIII 2021, larvae from *H. syriacus*, 4♂, 5♀, adults emerged 25-30.VIII 2021,

N. Karpun, E. Zhuravleva, coll.; Sochi, park "Southern Cultures", 08.VIII 2021, larvae from *Tilia caroliniana*, 5♂, 6♀, adults emerged 20-27.VIII 2021, 1 larva DNA barcoded (Sample ID: NK1071; Process ID: CAMRU156-21), N. Karpun, E. Zhuravleva, N. Kirichenko coll.; Sochi, Zeleny alleyway, 14.VIII 2021, larvae from *H. syriacus*, 4♂, 5♀, adults emerged 31.VIII 2021, E. Zhuravleva coll.; ibidem, same collection date, larvae from *A. theofrasti*, 3♂, 4♀, adults emerged 31.VIII-5.IX 2021, E. Zhuravleva coll.; Sochi, Lazarevskoe, 29.VIII 2021, larvae from *H. syriacus*, 5♂, 5♀, adults emerged 12-19.IX 2021, E. Zhuravleva, E. Shoshina coll.; ibidem, 13.VIII.2022, larvae from *A. hybridum*, 1♂, 2♀, adults emerged 23-30.VIII 2022, E. Zhuravleva; Sochi, Kurortniy av., 21.VIII 2021, larvae from *H. mutabilis*, 5♂, 5♀, adults emerged 05-11.IX 2021, E. Zhuravleva, E. Shoshina coll.; Sochi, Gagarina str., 28.VIII 2021, larvae from *A. hybridum*, 4♂, 5♀, adults emerged 10-16.IX 2021, E. Zhuravleva, E. Shoshina coll.; ibidem, Gagarina str., 08.VIII 2022, larvae from *T. begoniifolia*, 5 larvae, 1 pupae, E. Zhuravleva coll.; Anapa, Shevchenko str., *H. syriacus*, 07.IX 2022, 1 pupa, E. Shoshina coll.

ADULTS. The morphology of the specimens collected in Sochi (Fig. 1) well corresponds to the detailed description of the moth habitus given in Kumar *et al*. (2013). The body length of the specimens from Sochi was 12,6±0,05 (♂) and 13±0,03 (♀) mm, the wingspan 25,6±0,18 (♂) and 27,4±0,14 (♀) mm.

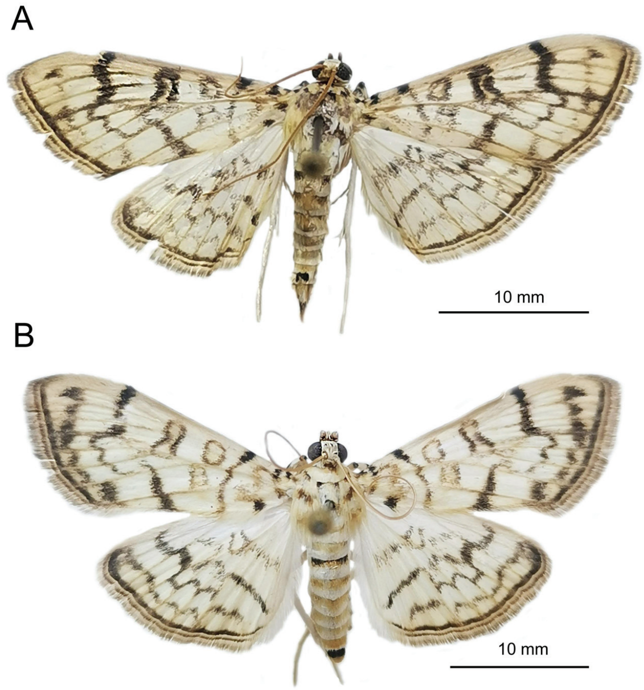

Fig. 1. Adults of *Haritalodes derogata.* A – male, B – female. Reared from *Hibiscus syriacus*, Sochi, Yana Fabritsiusa str., 25.VIII 2022 (the adult emergence date).

DNA BARCODING. DNA barcoding data obtained for two larvae sampled from *H. syriacus* and *T. caroliniana* in Sochi (Russia) allowed identifying *H. derogata* with 100% confidence in BOLD, where the closest neighbor originated from China (Fig. 2). No divergence was recorded between two specimens from Sochi. They, together with the specimen from China, formed one cluster with the specimen from Philippines (0,47% divergence) corresponding to one BIN: BOLD:AAB8317 (Fig. 2; Table 2). Interestingly, the other specimens identified as *H. derogata* in BOLD and/or GenBank formed four other divergent clusters, with each cluster corresponding to a different BIN. Among them, two specimens from South Africa were assigned to BIN: BOLD:AAP9908 and two specimens from Australia to BIN: BOLD:AAB8315 (Fig. 2). The specimens from Japan and Papua New Guinea corresponded to the BINs: BOLD:AAB8318 and BOLD:AAB8316 accordingly (Fig. 2).

Table 2. Intra- and interspecific divergences in COI mtDNA gene among *Haritalodes derogata* specimens originating from different countries.

Country	**Country**						
	Russia (Sochi)	China	Philippines	South Africa	Australia	Papua New Guinea	Japan
Russia (Sochi)	[0]						
China	0	[–]					
Philippines	0,47	0,47	[–]				
South Africa	4,15	4,15	4,65	[0]			
Australia	6,14	6,14	6,32	5,48	[0,15]		
Papua New Guinea	6,47	6,47	6,65	5,63	5,0	[–]	
Japan	7,0	7,0	0,47	7,36	6,49	6,47	[–]

Remark. Minimal pairwise distances are given for each species pair; values in square brackets represent maximal intraspecific distances; [—] no data because a single specimen was sequenced.

Minimal pairwise distances estimated for each species pair varied from 4,15% (for the specimen from South Africa and Russia (Sochi) to 7,36% (for the specimens from South Africa and Japan) (Table 2). Such pronounced genetic distances between the specimens of *H. derogata* originating from different parts of the world may suggest the presence of cryptic species.

BIOLOGY. In Sochi, the larvae caused characteristic damage to the leaves (Fig. 3A–C). Larvae made long cut in the upper part of the leaf blade and rolled the cut part downwards (Fig. 3C) binding the roll with silk. Larvae fed inside such constrictions during whole larval stage. At the beginning, they stayed together (i.e. the larvae from the 1st to the 5th instar) inside a leaf roll; later they dispersed on a host and fed singly in new leaf rolls (Fig. 3D-F). Young larvae skeletonized, the older ones rudely gnawed leaf lamina inside rolled leaves. Matured larvae left their shelters and made new leaf rolls in which they pupated. Matured larva were 25–27 mm long. Pupae were 13-15 mm long, brown, cremaster with an awl-shaped projection (Fig. 3G).

The pest is commonly known to develop three generations; exceptionally, in warmer regions, up to four generations were documented (Dhindsa *et al*., 1980; Uematsu, 1986). In the humid subtropics of the Black Sea Coast in 2021–2022, two generations were recorded per year: after overwintering, adults emerged in the second half of June, whereas the adults of the second generation were emerging during most of August. Adults lived 7-10 days. They were active in late evening and night and were attracted by the light. Outdoor, the larvae of the first generation pupated inside the rolled leaves on plants, whereas the larvae of the second generation pupated in rolled leaves on the litter or under the bark of host plants. Under natural conditions, the entire life cycle of one generation took 30–42 days. In laboratory conditions, the females laid up to 300 eggs singly on the lower surface of host plant leaves. The eggs were small, greenish, elliptical. Under laboratory conditions, larval stage took about 20 days, pupation took 8-10 days. These data are consistent with the biology of *H. derogata* described in other countries, in particularly, India and Azerbaijan (Badiyala, 2011; Sontakke *et al*., 2015; Gahramanova *et al*., 2020).

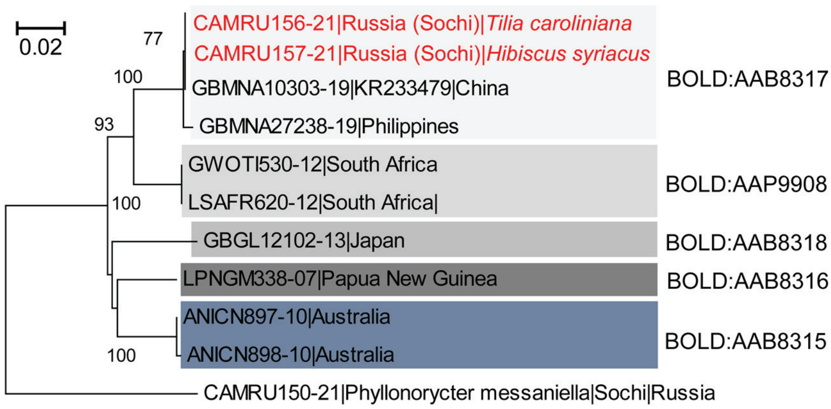

Fig. 2. Maximum likelihood COI tree illustrating intraspecific divergence of *Haritalodes derogata* in its modern range. The clusters are indicated by different colors correspond to different BINs. Each specimen is indicated by the BOLD process ID (begins with CAMRU. GBMNA etc.), followed the country of origin (and the locality, in case of Russia), and a host plant (where known). Bootstrap values > 70 are given next to the corresponding branches.

HOST PLANTS: Polyphagous species on Malvaceae, Fabaceae, Rhamnaceae, Anacardiaceae, Amaranthaceae, and Rosaceae. In South East Asia, Africa and Oceania, larvae feed on leaves of *Hibiscus, Gossypium*, *Sida, Pavonia, Anacardium occidentale, Glycine max, Zizyphus mauritiana, Z. jujuba, Abelmoschus esculentus*, and others (Mariselvi & Manimegalai, 2016; Roychoudhury *et al*., 2017). In the Russian Far East, larvae feed on herbaceous, subshrubs, and woody plants from several families (Sinev & Streltsov, 2019).

On the Black Sea Coast of Russia, *Haritalodes derogata* was initially recorded in plantings of *Hibiscus syriacus*. Further surveys of 2021 and 2022 showed that it also attacks *H. sinensis, H. mutabilis, H. lasiocarpos* in all districts of Sochi. In the beginning of August 2021, *H. derogata* larvae were documented on the *Tilia caroliniana* and *T. begoniifolia* in the Imeretinskaya Lowland (the dendropark "Southern Cultures"). In late August 2021, larvae were found also on *Abutilon* × *hybridum* in ornamental plant, and on *Tilia platyphyllos.* In the same period, the characteristic damage was recorded on the weedy plant *Abutilon theophrasti.*

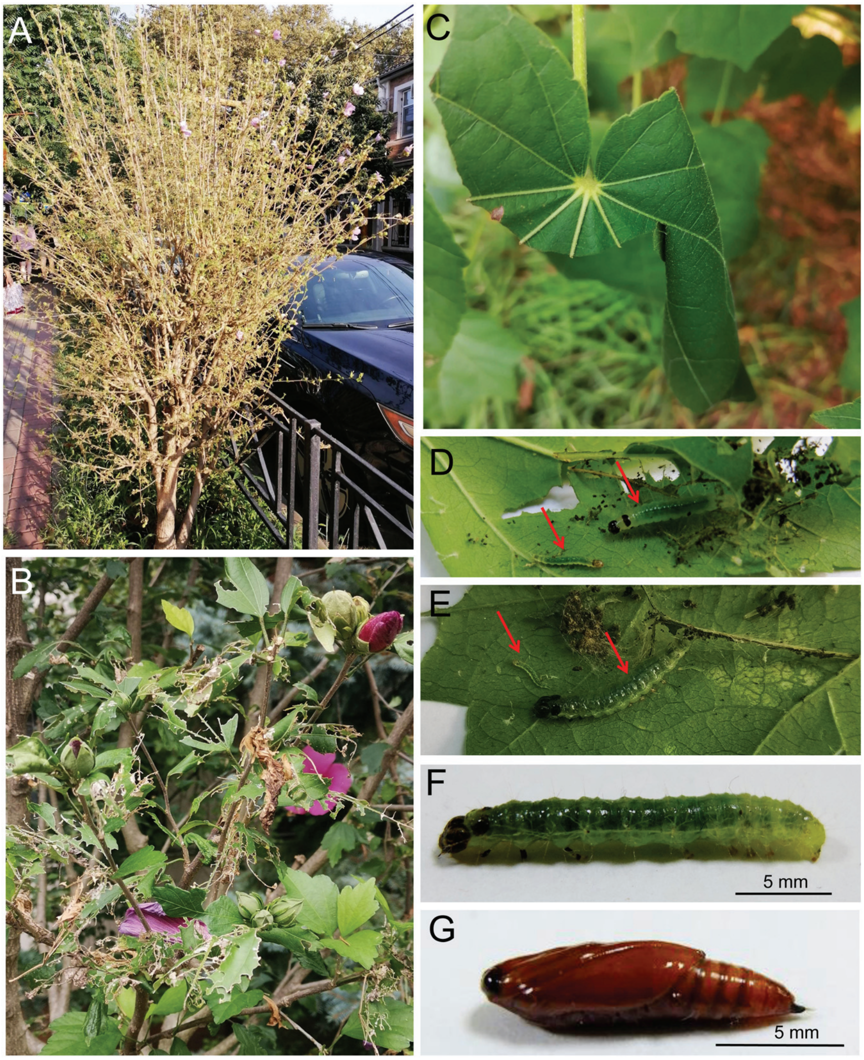

Fig. 3. Biology of *Haritalodes derogata* in Sochi, Yana Fabritsiusa str., 22–27.VIII 2021. A – severe damage caused to *Hibiscus syriacus*; B – close up of the damaged plant; C – characteristic leaf damage; D, E – unrolled leaf with young and late instar larvae (indicated by red arrows) and black frass; F – larva of the 5th instar (lateral view); G – pupa (lateral view).

DISTRIBUTION. Africa, Asia, Oceania (Fig. 4); the detailed list of the countries where the pest occurs is given in Byun *at al*. (2008) and CAB International (2022). Russia: South Primorye (Bremer, 1864), Transbaikalia (Korsun, 2017), Sochi (discovered in July 2021) (present paper), Gelendzhik (September 2021) (Krylenko, 2022), Anapa, Tuapse, Sochi (Krasnaya Polyana, 960 m above sea level) (September 2022) (present paper).

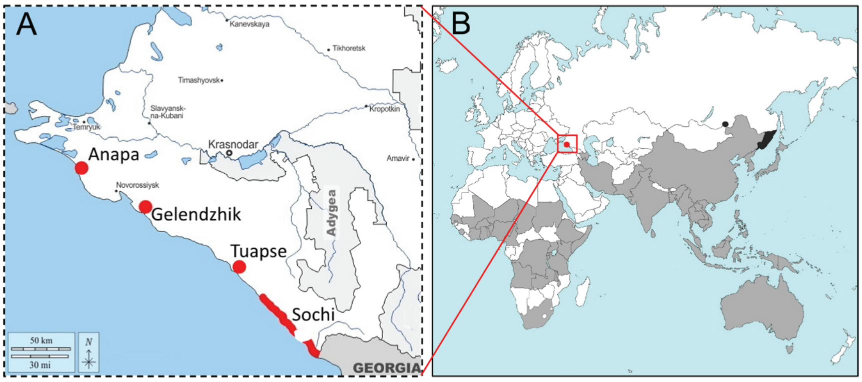

Fig. 4. The localities on the territory of the Russian Black Sea Cost where *Haritalodes derogata* was discovered in 2021–2022 (A) and the modern range of the species (B). Present distribution of *H. derogata* is shaded in gray; the regions colored in black show the presence of the species in Russia, the red dot indicates the recent species invasion to the South of Russia.

DAMAGE LEVEL. In summers 2021–2022, the species caused severe damage to *Hibiscus* spp. in Sochi: the defoliation level reached nearly 100% in Yana Fabritsiusa str. and Zeleniy line. In the other localities, the damage level was around 50%. The most damage was caused in the end of July – early August. The bushes of hibiscus severely defoliated in 2021, withstood and refoliated next summer 2022. In July–August 2022, hibiscus bushes experienced similar level of damage as the treatment procedures were delayed. In a contrast to hibiscus, the damage to *Tilia* spp. was occasional and not intense (less than 25% of leaves were damaged in the dendropark "Southern Cultures" and Centralniy district of Sochi).

CONTROL MEASURES. Chemical control is used in extreme cases to localize outbreaks in the regions of cultivation of cotton, soybean, cashew, and other crops. The insecticides based on thiomethoxam, imidacloprid, cypermethrin, lambda-cyhalothrin, and dimethoate showed satisfactory efficiency (Misra *et al*., 2002; Badiyala, 2011; Silvie *et al*., 2013; Shakya & Saxena, 2016). Due to many restrictions, the use of insecticides in ornamental plantings, especially in the resort area (such as Sochi), is very limited or impossible. Presently, no effective cure is developed to control the pest populations on Russian Black Sea Coast.

There are data on parasitoids from the families Braconidae, Ichneumonidae and Eulophidae attacking larvae and pupae of *H. degorata,* and larval pathogens, i.e. *Bacillus thuringiensis* var. Kurstaki and *Beauveria bassiana* (Odebiyi, 1982; Kumar *et al*., 2013; Gahramanova *et al*., 2020; Dannon *et al*., 2021).

PROGNOSIS. In the absence of effective measures by the relevant authorities, the forecast of further expansion of *H. derogata* in the Southern Russia cannot be optimistic. The two-year observations run in Sochi suggests a high risk of *H. derogata* invasion across Southern Russia. Furthermore, the accidental introduction of this polyphagous pest to the European regions with the subtropical climate and its rapid acclimatization with following damage to crops and ornamental plants are not ruled out.

CONCLUSIONS. The vector of *H. derogata* invasion to the Russian Black Sea Cost remains unknown. We suspect that the introduction of the pest to this region could be human-mediated. The species could be brought here accidently with plants for planting or transported with other goods. Bearing in mind that the pest already provides noticeable damage in Sochi, it has no doubts that it penetrated the region some years ago. As a matter of fact, it was overlooked here and detected only when the damaged became pronounced.

Urgent actions are required to localize the foci of the alien pest species on the territory of the Russian Black Sea Coast and prevent its further dispersal to neighboring regions and countries. Further studies would be needed to explore the genetic diversity of *H. derogata* across its wide range in Asia, Africa and Oceania to resolve the question about the existence of sister species, as well as determine the regions from where the pest could be introduced to the Black Sea Cost of Russia. These data would be of a high importance to understand the invasion history, define the distribution routes and develop the adequate management methods for controlling the pest.

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