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- The diaptomid fauna of Israel (Copepoda,
- ³ Calanoida, Diaptomidae), with notes on the
- systematics of Arctodiaptomus similis (Baird, 1859)
- and Arctodiaptomus irregularis Dimentman & Por,
- 1985 stat. rev
- Federico Marrone^{1*}, Adam Petrusek², Giuseppe Alfonso³ and Marco Arculeo¹

Abstract

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30 31 **Background:** To date, only scarce information is available about the diaptomid copepods of the Middle East despite the ecological and biogeographical importance of the family Diaptomidae in the inland waters of the Holarctic region. Moreover, the taxonomic status of some of the taxa occurring in the area is in need of revision. We studied crustaceans collected from temporary and permanent lentic water bodies in Israel with the aim of providing an updated census of the diaptomid copepods occurring in the country. Furthermore, we morphologically and genetically analysed samples of *Arctodiaptomus similis* s.l. to shed light on its taxonomy.

Results: Five diaptomid taxa were collected during this survey. Among these, *Phyllodiaptomus blanci* is new for the country and the whole circum-Mediterranean area and might be an allochthonous species of eastern origin. Within the collected samples, we singled out two parapatric groups of populations within *A. similis* s.l.; these consistently differ both based on morphology (chaetotaxy of the left male antennule) and molecular data (divergence over 17% at mitochondrial DNA gene for cytochrome b). We thus attribute the full species rank to *Arctodiaptomus irregularis* Dimentman & Por, 1985 stat. rev., originally described as a subspecies of the widespread species *Arctodiaptomus similis* (Baird, 1859).

Conclusions: We critically evaluated all species hitherto reported for Israeli inland waters. Considering both the confirmed literature data and the new findings, Israeli diaptomid fauna is composed of at least seven species. However, the need for further surveys in the Middle East and for detailed systematic revisions of some controversial taxa is stressed. Our results on the systematics of *A. similis* s.l. illustrate the importance of implementing molecular analyses when investigating diversity patterns of groups which are difficult to resolve based on morphology alone.

Keywords: Copepod diversity; Middle East; *Hemidiaptomus gurneyi canaanita*; *Arctodiaptomus similis*; *Arctodiaptomus irregularis* stat. rev; *Phyllodiaptomus blanci*; *Neolovenula alluaudi*

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Background

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Representatives of the copepod family Diaptomidae often dominate the lentic inland water bodies of the Holarctic, Oriental, and Afrotropical biogeographical regions, being a key taxon in many lacustrine and pond ecosystems (Dussart and Defaye 2001, 2002). Species of this family are as a rule characterised by limited distributional ranges, which are significantly constrained by the legacies of historical biogeographical events (Leibold et al. 2010). However, an ever-increasing corpus of molecular evidence suggests that our knowledge on diaptomid species distributions and biogeography is currently hampered by a gross underestimation of the actual diversity of the group (e.g., Marrone et al. 2013, and references therein). In the inland waters of the Mediterranean area, about 100 diaptomid species belonging to 14 genera are currently reported to occur (Dussart and Defaye 2002), but the information available for certain regions is far from being satisfactory (cf. Marrone 2006).

To date, uneven information is available on the inland water calanoid copepod fauna of the Middle East. While the diaptomid faunas of Turkey (e.g., Gündüz 1998; Ustaoğlu 2004; Ustaoğlu et al. 2005) and Iran (e.g., Brehm 1937; Löffler 1956, 1961) are relatively well-known, only scarce and anecdotal records are currently available for other countries of this region (e.g., Gurney 1921; Spandl 1923; Kiefer 1978b; Dumont 1979, 2009; Khalaf 2008; Mohamed and Salman 2009). The Israeli diaptomid fauna is quite a peculiar case: although several papers and reviews are available (e.g., Baird 1859; Richard 1893; Kiefer 1930; Yaron 1964; Dimentman and Por 1985, Azoulay 2001, and references therein), only five diaptomid taxa are currently listed for the country; this seems quite a species-poor fauna when compared to other circum-Mediterranean regions of comparable area, where usually about 9 to 14 diaptomid species are present (e.g., Ruffo and Stoch 2005; Marrone et al. 2005; Marrone 2006; Turki and Turki 2010; Alfonso and Belmonte 2011). Furthermore, the taxonomical validity of some 'forms' or 'subspecies' described in the past decades based on Israeli specimens is rather controversial, and the presence itself of some taxa in the country is in need of confirmation, thus casting further uncertainty on the actual composition of the Israeli diaptomid fauna.

We studied plankton samples collected in Israeli inland waters between 2003 and 2011 in order to update the available information on the composition and distribution of diaptomid copepods in the country. Furthermore, we genetically analysed several populations of *Arctodiaptomus similis* s.l. to test the hypothesis that two parapatric groups of populations differing in morphology, observed in our dataset, actually represent distinct evolutionary lineages of species rank.

Methods

Plankton samples were collected throughout the country, 88 focusing on small- to medium-sized freshwater habitats; 89 both permanent and temporary water bodies were investi- 90 gated, and some sites were sampled multiple times on different dates. Altogether, we analysed samples from 60 92 different sites (Figure 1). Samples were collected using a 93 200- μ m mesh hand net along the water bodies' shorelines; 94 open waters were sampled using 80-to-200- μ m mesh- 95 sized towing nets. Collected crustaceans were fixed *in situ* 96

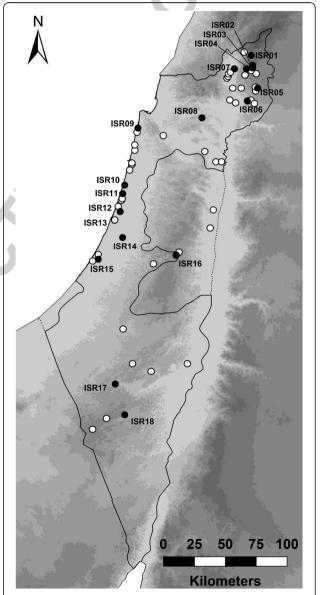


Figure 1 Map of the sampling sites. Samples collected between 2003 and 2011 from both temporary and permanent water bodies were analysed to improve knowledge on diversity and distribution of diaptomid copepods of Israel. Black dots represent sites where at least one diaptomid species was collected; white dots are sites without diaptomids.

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with 96% ethanol. They were then sorted in the laboratory 97 under a dissecting microscope, and diaptomid specimens 98 were prepared according to Dussart and Defaye (2001). 99 Morphological identification was performed according 100 to Kiefer (1974, 1978a), Borutzky et al. (1991), and 101 Ranga-Reddy (1994). Line drawings were prepared 102 using a compound microscope equipped with a camera 103 104 lucida.

Samples are stored in the crustacean collection of Federico Marrone and are available for loan on request. 106 Voucher specimens from the type localities of Arctodiaptomus similis (Baird, 1859) and Arctodiaptomus irregularis Dimentman & Por, 1985 stat. rev. have been deposited in 109 the collection of the Smithsonian Institution (National 110 Museum of Natural History) in Washington DC, USA, with the catalogue numbers USNM #1226919 and USNM

Thirteen specimens of A. similis s.l. from various regions 114 115 of Israel and two congeneric species to be used as outgroups (Arctodiaptomus cf. stephanidesi (Pesta, 1935) and Arctodiaptomus alpinus (Imhof, 1885), both collected in 117 Italy) were analysed genetically by amplifying and sequen-118 cing a 329-bp long fragment of the mitochondrial gene for 119 the cytochrome b (Cyt-b), a molecular marker which is known to be informative on the phylogeography and the 121 molecular systematics of closely related diaptomid taxa (e.g., Staton et al. 2003; Thum and Derry 2008; Thum and Harrison 2009; Marrone et al. 2010, 2013). Genetic analyses were performed following the protocols and 125 procedures described in Marrone et al. (2010, 2013), with the aim of investigating the molecular structuring of the taxon and to compare it with the observed patterns 128 of morphological variation. Chromatograms were imported 129 130 and edited with Chromas Lite 2.01 (Technelysium Pty. Ltd., South Brisbane, Australia) and aligned with BioEdit 131 (Ibis Biosciences, Carlsbad, CA, USA) (Hall 1999). The 132 quality of the obtained sequences was checked with Sequence Scanner v1.0. Only sequences with continuous 134 135 reads of high quality bases were used; when the sequences were not of sufficient quality, the reverse complement se-136 quences were also obtained. MEGA 5 (Tamura et al. 2011) was used to translate the Cyt-b sequences to amino acids 138 in order to check for the possible presence of frameshifts 139 or stop codons, which would indicate the presence 140 of sequencing errors or pseudogenes. The sequences 141 were deposited in GenBank under Accession Numbers 142 143 KM488608 to KM488622.

Bayesian inference (BI) of phylogeny and maximum likelihood (ML) analyses were performed on the Cyt-b dataset as implemented by MrBayes 3.2.1 (Ronquist et al. 2012) and PhyMl v.3 (Guindon and Gascuel 2003), respectively. Both analyses were performed using a Hasegawa, Kishino, and Yano model of sequence evolution for molecular data with a proportion of invariable sites (HKY + I), as selected by the Akaike information 151 criterion in MrModeltest 2.2 (Nylander 2004). Node supports were evaluated by their posterior probabilities in the BI tree and with 1,000 bootstrap replicates in the ML analysis. The BI analysis was performed with two independent runs of 2,000,000 generations and four Markov chains using default heating values. Trees and parameter values were sampled every 100 generations resulting in 20,000 saved trees per analysis. An initial fraction of 5,000 trees (20%) was conservatively discarded as 'burn-in'. For all analyses, standard deviation of split frequencies reached values lower than 0.0065, and values of the potential scale reduction factor (PSRF) were between 1.0 and 1.004 for all parameters, indicating convergence of the runs.

In order to compare the observed molecular distances with those available in literature for other diaptomid taxa, uncorrected molecular distances among specimens and between groups were calculated in PAUP 4.0b10 (Swofford 1998).

Results Morphological identification

We analysed samples originating from 60 different sampling sites. In 18 sites, diaptomid copepods were present (Figure 1, Table 1). Most of these habitats were temporary rain pools, diaptomids were only rarely encountered in permanent water bodies such as reservoirs and fishponds, although these were well-represented in our sample set. 178 Altogether, five diaptomid taxa were collected (Table 2). Among these, Hemidiaptomus gurneyi canaanita, two 'forms' of A. similis s.l., and Neolovenula alluaudi were already known for the fauna of the country. Conversely, the finding of Phyllodiaptomus blanci in a reservoir of a city park in Tel Aviv is the first one for Israel and for the whole circum-Mediterranean area. In spite of previously published findings, no Eudiaptomus species were collected during the present survey.

Studied A. similis s.l. populations differed in the antennular chaetotaxy, which consistently presents one versus two setae on the 15th and 17th segments of the left male antennule (Figure 2). Based on this character, these could be ascribed to two parapatric groups, one restricted to water bodies of the Golan Heights only, the other found in other regions of the country (Table 3).

Molecular analyses

The BI and ML trees based on a 329-bp long fragment of the Cyt-b showed a concordant topology, with a clear division of the investigated A. similis s.l. samples into two groups, hereafter referred to as the 'Golan' and the 'Israeli' clades (Figure 3). These clades are in good accordance with the two groups singled out based on morphology (Table 3) and show a sharp geographical segregation.

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Table 1 List of the sampled localities with diaptomid presence

t1.2	Code	Site name - district	Sampling date(s)	Latitude (N)	Longitude (E)	Hydroperiod
t1.3	ISR01	Lake Ram - Golan Heights	14 March 2011	33.23639	35.76583	Permanent
t1.4	ISR02	Pool at El Rom - Golan Heights	9 March 2010	33.16472	35.77694	Temporary
t1.5	ISR03	Bab El Hawa pool - Golan Heights	9 March 2010	33.14333	35.77417	Temporary
t1.6	ISR04	Orvim reservoir - Golan Heights	7 March 2011; 11 March 2011	33.13972	35.73194	Permanent
t1.7	ISR05	Khusniya pool - Golan Heights	8 March 2010	32.99583	35.81056	Temporary
t1.8	ISR06	Vernal pools close to Daliyot Reservoir, Golan Heights	2 January 2003	32.89972	35.77589	Temporary
t1.9	ISR07	Lehavot Habashan - Northern District	15 March 2011	33.13833	35.64389	Permanent
t1.10	ISR08	Mashkena pool - Northern District	6 March 2010; 17 March 2011	32.78111	35.4075	Temporary
t1.11	ISR09	Ditch at Atlit - Haifa District	3 January 2003	32.70444	34.94194	Temporary
t1.12	ISR10	Brechat Reserve - Haifa District	7 March 2011	32.29111	34.84583	Temporary
t1.13	ISR11	Ga'ash pond - Haifa District	7 March 2011	32.22889	34.83083	Temporary
t1.14	ISR12	Pond in Yarkon park - Tel Aviv District	29 January 2003	32.09722	34.81278	Permanent
t1.15	ISR13	Pond in Holon - Tel Aviv District	26 January 2004	32.03472	34.77194	Temporary
t1.16	ISR14	Moshav Ishrash - Center District	Not available.	ca 31.90	ca 34.83	Temporary
t1.17	ISR15	Rainwater-flooded field near Tel Ashdod - Southern District	22 January 2004	31.75167	34.65194	Temporary
t1.18	ISR16	Mamilla pool - Jerusalem District	4 March 2011	31.77801	35.22058	Temporary
t1.19	ISR17	Small pools in Sde Boker, Vadi Zin - Southern District	9 March 2011	30.84167	34.77722	Temporary
t1.20	ISR18	Pool in Makhtesh Ramon crater - Southern District	7 March 2011; 12 March 2011	30.61694	34.84611	Temporary

t1.21 Decimal geographical coordinates (WGS84) are provided.

The mean intra-clade uncorrected molecular distance

204 was 1.6% (ranging from 0.9% to 3%) within the Israeli

clade and 0.7% (ranging from 0% to 0.9%) within the 205

Golan clade. The average uncorrected molecular distance 206

between the two clades was 17.3%.

Discussion 208

t2.1

t1.1

Checklist and distribution

- Five diaptomid taxa were collected in the present survey;
- in addition to these, Eudiaptomus drieschi and Arctodiap-
- tomus (Rhabdodiaptomus) salinus have to be considered

part of the Israeli diaptomid checklist based on bibliograph- 213 ical evidence. The reports of Arctodiaptomus (Arctodiapto- 214 mus) wierzejskii and Eudiaptomus gracilis in the country 215 are in need of being substantiated and are possibly errone- 216 ous (see comments below). At the present state of know- 217 ledge, the checklist of Israeli diaptomid copepods is thus 218 composed of seven taxa (Table 2).

The paradiaptomid N. alluaudi, which was previously 220 known in the country only from the Sinai and Negev de- 221 serts (Dimentman and Por 1985), was collected in our survey both in the Negev desert and on the Golan Heights, 223

Table 2 Checklist of Israeli Diaptomidae (Copepoda, Calanoida)

t2.2	Taxa	Sites of occurrence (present study)	Sources
t2.3	Diaptominae		
t2.4 t2.5	Hemidiaptomus (Hemidiaptomus) gurneyi canaanita Dimentman & Por, 1985	ISR02, ISR05, ISR06, ISR11, ISR14	2, 6, 8, 9, 11, 13
t2.6	^a Eudiaptomus drieschi (Poppe & Mrázek 1895)	n.d.	10
t2.7	Arctodiaptomus (Arctodiaptomus) similis (Baird, 1859)	ISR07, ISR08, ISR09, ISR10, ISR11, ISR13, ISR15, ISR16	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 13
t2.8	Arctodiaptomus (Arctodiaptomus) irregularis Dimentman & Por, 1985	ISR02, ISR03, ISR04, ISR05, ISR06	
t2.9	^a Arctodiaptomus (Rhabdodiaptomus) salinus (Daday, 1885)	n.d.	2
t2.10	^b Phyllodiaptomus blanci (Guerne & Richard, 1896)	ISR12	13
t2.11	Paradiaptominae		
t2.12	Neolovenula alluaudi (Guerne & Richard, 1890)	ISR01, ISR03, ISR17, ISR18	8, 9, 12, 13

^aNot found during the present survey; ^bfirst record for Israel. The actual occurrence of Arctodiaptomus wierzejskii and Eudiaptomus gracilis in Israel is doubtful,

t2.14 and the species are not included in the table (see 'Discussion' section). The codes of sampling sites refer to Table 1. n.d., not detected. Sources: 1: Baird (1859);

^{2:} Richard (1893); 3: Kiefer (1930); 4: Fischer (1953); 5: Petkovski (1961); 6: Yaron (1964); 7: Kiefer (1974); 8: Por (1984); 9: Dimentman & Por (1985); 10: Azoulay

^{(2001); 11:} Marrone et al. (2010); 12: Alfonso & Belmonte (2013); 13: Present work.

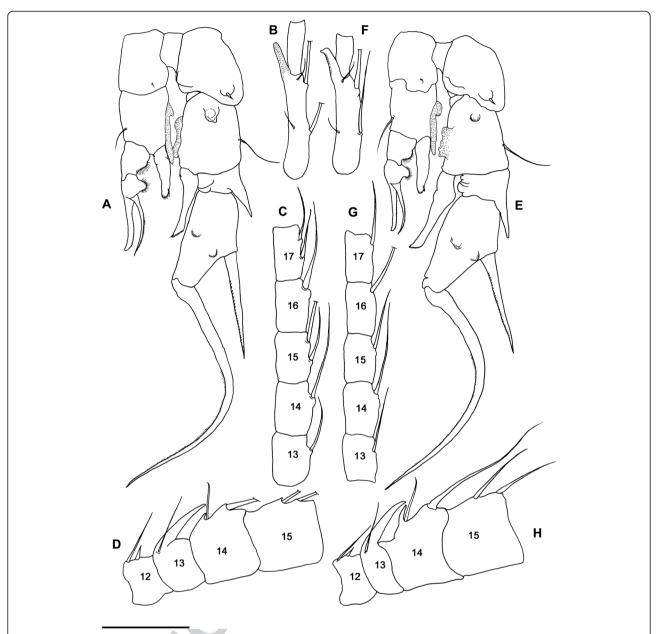


Figure 2 *Arctodiaptomus similis* (Baird, 1859) and *Arctodiaptomus irregularis* Dimentman & Por, 1985. Anatomical features of *A. irregularis* collected in Bab El Hawa (ISR03) on 9 March 2010 (A-D) and *A. similis* collected in Birket Mamilla pool (ISR16) on 4 March 2011 (E-H). A and E: male fifth pair of the legs (posterior view). B and F: antepenultimate article of the male right antennula. C and G: detail of the chaetotaxy on the articles 13 to 17 of the male left antennula. D and H: articles 12 to 15 of the male right antennula. Scale bar 100 μm.

thus widening to the north the distribution of this species in the country. This finding was recently anticipated by Alfonso and Belmonte (2013) in a review on *N. alluaudi* distribution.

Hemidiaptomus (Hemidiaptomus) gurneyi canaanita is an endemic taxon whose distinct status was confirmed by molecular analyses, in spite of the morphological identity of Israeli populations with those collected in the central Mediterranean, including the type locality of the species (Marrone et al. 2010). Hemidiaptomus gurneyi s. l. is characterised by a sharply disjointed distribution, with the populations belonging to *Hemidiaptomus gur-neyi gurneyi* occurring in the central Mediterranean area (from eastern Algeria to the Balkan Peninsula), and 237 those belonging to the subspecies *Hemidiaptomus gur-neyi canaanita* occurring in Israel (Marrone et al. 2010). 239 The report of the presence of the species in Hungary (Dussart and Defaye 2002) is not supported by any reference or sample (cf. Kiefer 1978a; Petkovski 1983; Stella 242 1984; Dussart 1989; Borutzky et al. 1991; Marrone et al. 243

t3.1

Table 3 A. similis s.l. populations based on setae number on 13th, 15th, and 17th segments of left male antennule

t3.2	on 13th, 15th, and 17th	segments o	of left male ar	tennule
t3.3	Population	N	1-1-1	1-2-2
t3.4	ISR01 - Golan Heights	3		√
t3.5	ISR02 - Golan Heights	3		$\sqrt{}$
t3.6	ISR03 - Golan Heights ^a	5		$\sqrt{}$
t3.7	ISR04 - Golan Heights	1		$\sqrt{}$
t3.8	ISR05 - Golan Heights	3		$\sqrt{}$
t3.9	ISR06 - Golan Heights	5		$\sqrt{}$
t3.10	ISR07 - Northern District	1	$\sqrt{}$	
t3.11	ISR08 - Northern District	2	\checkmark	
t3.12	ISR11 - Haifa District	1	$\sqrt{}$	
t3.13	ISR13 - Tel Aviv District	3	$\sqrt{}$	
t3.14	ISR15 - Southern District	4	$\sqrt{}$	
t3.15	ISR16 - Jerusalem District ^b	6	\checkmark	

No males were available from populations ISR09 and ISR10. N, number of male specimens studied. ^aType locality of A. similis irregularis Dimentman & Por,

t3.18 1985 (now A. irregularis); btype locality of A. similis (Baird, 1859). 2010); it has thus to be considered a lapsus calami of 244 the authors. In our survey, H. gurneyi canaanita was 245 collected in water bodies of central and northern areas 246 of Israel, which is in good accordance with the distribu- 247 tion pattern described for the species by Dimentman 248 and Por (1985). Based on the available drawing, the re- 249 port of the occurrence of a female Hemidiaptomus spe- 250 cimen in 'Birket de Banias' (Richard 1893), originally 251 attributed to Hemidiaptomus amblyodon (Marenzeller, 252 1873) by the author, can in fact be unequivocally as- 253 cribed to *H. gurneyi canaanita*.

Two species belonging to the genus Eudiaptomus are 255 to date reported for the country: Eudiaptomus gracilis (G.O. Sars, 1863) and Eudiaptomus drieschi (Poppe & 257 Mrázek, 1895), both of them for Lake Kinneret (see 258 Azoulay 2001, and references therein). As it has already 259 been stressed (e.g., Dimentman and Por 1985; Azoulay 260 2001), it is possible that the early reports of E. gracilis 261 for Lake Kinneret should be in fact ascribed to a mis- 262 identification of E. drieschi, and that E. gracilis should 263 thus be excluded from Israeli diaptomid fauna. However, 264 conclusive evidence is not available, and the report of 265 occurrence of this euryecious diaptomid species in the 266

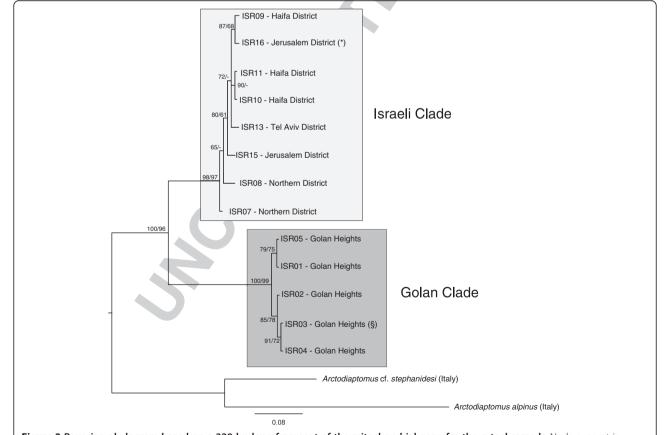


Figure 3 Bayesian phylogram based on a 329-bp long fragment of the mitochondrial gene for the cytochrome b. Node support is reported as nodal posterior probabilities/ML bootstrap; values below 50 are indicated by a dash. Asterisk (*): type locality of A. similis (Baird, 1859); Section sign (§): type locality of A. similis irregularis Dimentman & Por, 1985 (now A. irregularis).

lake cannot be excluded. *Eudiaptomus* species were observed in Lake Kinneret only since the 1960s onwards, although the Lake Kinneret zooplankton was investigated by frequent sampling since the early 20th century. This suggests a relatively recent colonization of the lake from an unknown source area, possibly corresponding with various anthropogenic changes of the Lake Kinneret system which

took place since the 1950s, including intensive stocking of 274 fish exotic to the lake (e.g., Gophen 1979). No *Eudiaptomus* 275 species were collected during our survey; however, we did 276 not sample this large freshwater lake. 277

A thriving population of the diaptomid *P. blanci* was 278 observed in a permanent reservoir within a city park in 279 Tel Aviv (Figure 4). This species is new for Israel and 280 **F4**

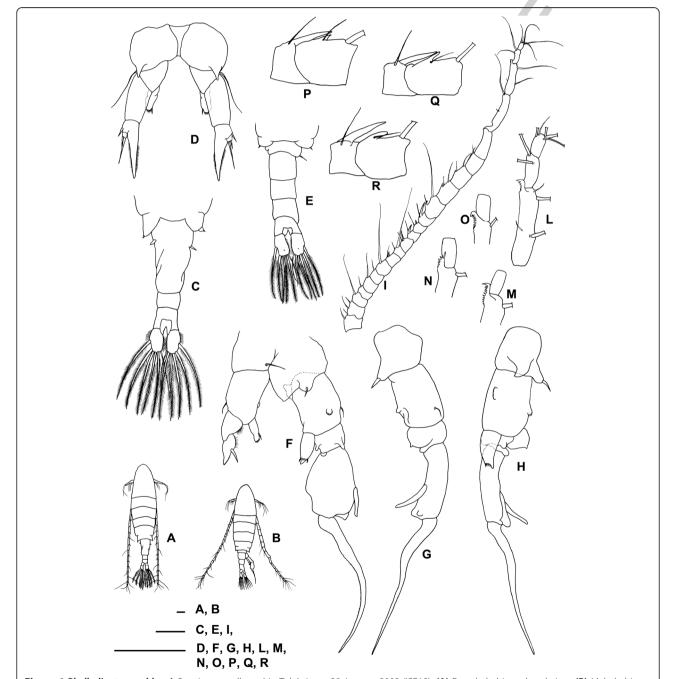


Figure 4 *Phyllodiaptomus blanci*. Specimens collected in Tel Aviv on 29 January 2003 (ISR12). **(A)** Female habitus, dorsal view. **(B)** Male habitus, dorsal view. **(C)** Female urosome, dorsal view. **(D)** Female fifth pair of legs, posterior view. **(E)** Male urosome, dorsal view. **(F)** Male fifth pair of legs, posterior view. **(G)** Male right leg, outer lateral view. **(H)** Male right leg, inner lateral view. **(I)** Male right antennula. **(L-O)** Variability of the antepenultimate segment of the male right antennula. **(P-R)** Variability of 13° and 14° articles of the male right antennula. Scale bar 100 μm.

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the whole circum-Mediterranean area. Phyllodiaptomus is a diaptomid genus whose distribution area 282 encompasses Central and Eastern Asia, from India to Iraq 283 (Borutzky et al. 1991; Ranga-Reddy 1994; Sanoamuang 284 and Teeramaethee 2006; Khalaf 2008). P. blanci and 285 *Phyllodiaptomus irakiensis* are the westernmost species 286 of the genus, both of them being known to occur in Iraq 287 288 (Khalaf 2008, and references therein). It is not clear 289 whether the presence of a P. blanci population in Israel is to be ascribed to an anthropogenic introduction to 290 the country or whether the species has to be considered autochthonous. However, the man-made origin of the 292 single known Israeli locality of this species suggests that, 293 294 pending further findings in the area, P. blanci is an alien copepod species in Israel. 295

Richard (1893) reported the occurrence of Arctodiaptomus (Rhabdodiaptomus) salinus from 'Birket Abo-Zeineh', a brackish marsh close to the northern coast of Lake Kinneret. The species is considered part of the Israeli fauna by Ranga-Reddy (1994) and Dussart and Defaye (2002). Dimentman and Por (1985) considered that this finding needed to be substantiated; however the presence of A. salinus in Israel is rather likely as this halophilous taxon is characterised by a broad circum-Mediterranean and Palaearctic distribution, including several countries in the Middle East (Dussart and Defaye 2002; Dumont 2009). Inland endorheic water bodies and salty coastal marshes and lagoons are the habitats to be sampled in order to find new evidence for the presence of this taxon

Arctodiaptomus (Arctodiaptomus) wierzejskii was erroneously cited to be present in Israel by Yaron (1964), who misidentified A. similis s.l. for this species (this erroneous datum was then unfortunately perpetuated by Dussart and Defaye 2002). The mistake was possibly due to the use of the trinomen Diaptomus wierzejskii palaestinensis by Kiefer (1930) for the diaptomid population inhabiting 'Birket Mamilla' and by Fischer (1953) for the populations inhabiting some fish ponds throughout the country. D. wierzejskii palaestinensis is in fact a synonym of Arctodiaptomus similis (Kiefer 1932, 1974).

The diaptomin taxon Arctodiaptomus (Arctodiaptomus) similis s.l. (Baird, 1859) inhabits both permanent and temporary water bodies of central and northern parts of the country, proving to be the commonest calanoid copepod occurring in Israeli inland waters. In spite of its abundance, the taxonomy of A. similis s.l. is to date controversial. Currently, two taxa of subspecific rank are ascribed to A. similis s.l., but the taxonomical arrangement of the species is in need of revision (cf. Ranga-Reddy 1994; Dussart and Defaye 2002). The species was described from Israel by Baird (1859) and later reported to occur throughout Southwestern Asia and Eastern Europe (e.g., Richard 1893; Petkovski 1961; Kiefer 1930, 1974, 1978; Dimentman and

Por 1985, Azoulay 2001, Dussart and Defaye 2002). Some 335 authors reported the presence of the species in Sardinia (Kiefer 1978a; Dussart and Defaye, 2002) without providing any drawing or precise locality data; furthermore, extensive limnological surveys carried out in the island (e.g., Stella 1970; Stella et al. 1972; Stella and Margaritora 1975/1976; Fadda et al. 2011; Marrone and Stoch, unpublished data) only recorded the presence of the congeneric species A. salinus and A. wierzejskii, and no evidence of the presence of A. similis s.l. was ever collected. Thus, the report of the 344 presence of the species in Sardinia is almost certainly erroneous, and the species is currently not considered part of 346 the Italian copepod fauna (Stella 1984; Ruffo and Stoch 2005). The Israeli populations of *A. similis* s.l. studied by us could be split into two parapatric groups (Table 3), differing in the chaetotaxy of the left male antennule (Figure 2C,G) and in the mtDNA Cyt-b sequences (Figure 3).

Taxonomical notes on Arctodiaptomus (Arctodiaptomus) similis (Baird, 1859) and Arctodiaptomus (Arctodiaptomus) irregularis Dimentman & Por, 1985 stat. rev

A. similis was described by Baird (1859) based on specimens raised from dried mud collected in the pool of Gihon in Jerusalem (now Birket Mamilla pond). Later on, Kiefer (1930) described D. wierzejskii palaestinensis based 358 on specimens collected in Birket Mamilla, but this taxon was later synonymised, by the author himself, with A. similis (see Kiefer 1974 for an annotated list of the synonyms of the species). In the same work, Kiefer (1974) stressed the variability of the ornamentation and chaetotaxy of the male antennule and established two infrasubspecific formae based on the presence of two versus one 365 setae on the 13th, 15th, and 17th segments of the left male 366 antennule (forma saetosior) and on the presence of a rodlike instead of claw-like process of the antepenultimate segment of the right male antennule (forma irregularis). Based on this last character, and on the presence versus 370 absence of a tooth on the 14th segment of the right male second antenna, Dimentman and Por (1985) established the subspecies Arctodiaptomus similis irregularis, whose 373 type locality lies in Birket Bab el Hawa, on the Golan Heights. However, it has to be stressed that several authors (Richard 1893; Petkovski 1961; Kiefer 1974, 1978a) reported the presence of a pronounced intra-populational 377 morphological variability for the morphological characters which, according to Dimentman and Por (1985), should allow unequivocal assignment of each population to one or the other subspecies.

In good accordance with Richard (1893) and Petkovski 382 (1961), the A. similis s.l. populations studied by us showed a noteworthy variability in the morphology of the right 384 male antennule; although the populations from the Golan Heights show, as a rule, a smaller tooth on the 14th segment (Figure 2D) and a blunter process on the 387

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antepenultimate segment of the antennule (Figure 2B), 388 exceptions and intermediate forms were observed. Con-389 versely, the number of setae on the 15th and 17th seg-390 ments of the left male antennule allowed us to soundly 391 distinguish the specimens collected throughout the 392 country in two groups (Table 3). This observation is in 393 sharp contrast with Kiefer (1974, 1978a), who stressed a 394 significant variability for the chaetotaxy of the segments 395 13th, 15th, and 17th both at intra- and inter-population 396 level. 397

The two groups of populations singled out based on the number of setae present on the 15th and 17th segments of the left male antennule (Figure 2C,G) are parapatric and in good accordance with the observed molecular clustering in two clades of the studied populations (Figure 3). The molecular distance observed between the two groups is of the same order of magnitude of those observed among different lineages deserving the status of cryptic species within other diaptomid species, like Skistodiaptomus pallidus (14.3% to 17.2%, Thum and Harrison 2009), Hemidiaptomus ingens (18%, Marrone et al. 2010), and Onychodiaptomus sanguineus (22%, Thum and Derry 2008); furthermore, a high sequence similarity was observed within each of the two clades.

Our morphological and genetic data thus support a classification of Israeli A. similis s.l. into two main lineages of species rank. Accordingly, two species are present in Israel: A. similis (Baird, 1859) (Figure 2E,F,G,H), inhabiting the whole country with the exception of the Golan Heights, and A. irregularis Dimentman & Por, 1985 stat. rev. (Figure 2A,B,C,D), apparently confined to the Golan 418 419 Heights.

Arctodiaptomus similis (Baird, 1859) 420

- Type locality: Mamilla pool (Jerusalem District; ISR16) 421 syn.: Diaptomus wierzejskii palaestinensis Kiefer, 1930 422
- Arctodiaptomus similis similis Dimentman & Por, 1985 423
- 424 A. irregularis Dimentman & Por, 1985 stat. rev.
- Type locality: Bab el Hawa pool (Golan Heights; ISR03) 425
- syn.: Arctodiaptomus similis forma irregularis Kiefer, 426
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- Arctodiaptomus similis irregularis Dimentman & Por, 428
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430 The two taxa Arctodiaptomus similis var. smirnovi Brehm, 1938 and Arctodiaptomus spectabilis Mann, 1940 431 are junior synonyms of A. similis s.l. (cf. Kiefer, 1974), but 432 at the current state of knowledge, it is impossible to 433 soundly ascribe them to, or differentiate from, either A. 434 similis s.s. or A. irregularis. The distribution of these two taxa in the Middle East and Eastern Europe is in fact to date unknown. Interestingly, a parapatric distribution of two distinct lineages similar to that within A. similis 438 has also been observed in Israeli populations of the cladoceran Daphnia (Ctenodaphnia) chevreuxi Richard, 440 1896: one lineage was only detected in lowland pools 441 in the Mediterranean coastal plain, while a genetically distinct lineage was widespread in the Golan Heights but not found elsewhere (A. Petrusek, *unpublished data*).

Conclusions

In the light of a critical review of the existing and new data, the calanoid family Diaptomidae proved to be 447 better represented in Israel than previous literature 448 data suggested, with seven species certainly occurring in the country. This value is close to that observed in 450 other circum-Mediterranean countries of comparable size (e.g., Marrone 2006). Furthermore, it also needs to be stressed that representatives of the genus Metadiaptomus, known to occur throughout the arid regions of 454 the Mediterranean and Black Sea (e.g., Kiefer 1978b; 455 Jaume 1989; Rayner 1999; Marrone and Naselli-Flores 2005: Samchyshyna 2011, and references therein), although 457 never recorded to date in Israel, are likely to also occur 458 in the country; for instance, Metadiaptomus chevreuxi (de Guerne & Richard, 1894) is known from Jordan, and Metadiaptomus mauretanicus Kiefer & Roy, 1942 from Egypt (Dumont 1979, 2009).

Based on the updated checklist of Israeli diaptomids 463 (Table 2), counts of endemic (i.e., H. gurneyi canaanita, A. irregularis) and eastern taxa (A. similis, E. drieschi, P. blanci) in this country are comparable to counts of taxa with wider distribution areas (i.e., Arctodiaptomus salinus and Neolovenula alluaudi). Unfortunately, due to a lack of sound checklists for most of the other Middle East countries, it is currently difficult to understand the biogeographic affinities of the Israeli calanoid copepod fauna.

The possible presence of an allochthonous species in the 473 country is in accordance with the ever-growing number of successful biological invasions affecting the freshwater zooplankton in the last decades. Such phenomenon is becoming increasingly important in the Western Palaearctic 477 region, where the records of allochthonous calanoid copepods are increasing at a fast pace (e.g., Ferrari et al. 1991; Rossetti et al. 1996; Alfonso and Belmonte 2008; Brandorff 2011; Alfonso et al. 2014).

Competing interests

The authors declare that they have no competing interests

Authors' contributions

FM carried out the morphological identification of the samples, carried out the molecular analyses, and drafted the manuscript. AP carried out the samplings in Israel and helped to draft the manuscript. GA helped with the bibliographical research, realised the figures, and provided important comments to a first draft of the manuscript. MA supervised and coordinated the research and helped with the molecular analyses. All authors read and approved the final manuscript.

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