First record of *Alainites sadati* Thomas, 1994 (Ephemeroptera: Baetidae) in Tunisia, description of the larval stage and ecology

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Abstract

*Alainites sadati* Thomas, 1994 is recorded for the first time in Tunisia. The first description of the larval stage of *A. sadati* Thomas, 1994 is provided based on material from Algeria, near the type locality, and Tunisia. This species can be separated from the other Palaearctic species by the number of gills, the setation of the legs, the prolongation of the paraproct and the reticulation of the tergites. Based on the new data, the ecology of *A. sadati* is discussed.

Key words: Ephemeroptera, Baetidae, *Alainites sadati*, larva, North Africa

Introduction

In her revision of the European species of *Baetis* Leach, 1815, Müller-Liebenau (1969) divided this genus in eleven species groups. *Baetis muticus* Linnaeus, 1758 was first included in the *gracilis* group (two species) (Müller-Liebenau, 1969), and subsequently to the *muticus* group (Müller-Liebenau, 1974). The author considered clearly this classification provisional as far as she wrote (Müller-Liebenau, 1973): “I feel that after many more years of describing and distinguishing species within the Baetidae, our concept of what a genus is in the family and our classification of the genera will be much different that what they are today”. In fact, the concept of *Baetis* as proposed by Müller-Liebenau has been proved to be paraphyletic (Waltz et al., 1994; Waltz & McCafferty, 1997; Fujitani et al., 2003; Fujitani, 2008; Gattolliat et al., 2008; Novikova & Kluge, 1987); new genera were established for part of these species groups (McCafferty & Waltz, 1995; Novikova & Kluge, 1987; Waltz et al., 1994). The species previously assigned to the *muticus* group were reassigned to the genus *Alainites* Waltz & McCafferty, 1994 (Waltz et al., 1994). The villopore is a valuable apomorphy uniting *Baetis* and related genera. As *Alainites* does not possess this character, it can not belong to this complex of genera (Waltz et al., 1994). At the larval stage, the distinctive characters to separate this genus were: the body (and especially the thorax) laterally compressed, the glossae with abundant thin setae on the dorsal side, the prostheca of the right mandible reduced to two bristles-like and feathered appendages, the absence of villopore and the paraproct with prolongation. At the imaginal stage, these characters were: hindwings, when present, with three longitudinal veins, the second being bifurcated, the segment III of the male forceps spherical to slightly elongated and curved (Waltz et al., 1994). In our opinion, these characters, specially the absence of villopore, clearly indicate that *Alainites* does not belong to the *Baetis* complex and the tentative of Jacob (2003) and Bauernfeind & Soldán (2012) gathering again all the species groups in *Baetis sensu lato* is no more tenable (Fujitani, 2003).
Eighteen species are presently included in *Alainites*: the Palaearctic realm encompassing 13 species and the Oriental five (Barber-James et al., 2010). Nine species are described at both larval and imaginal stages, five only at the larval stage and four only at the imaginal stage. Three species of *Alainites* were reported from North Africa: *Alainites muticus* (Linnaeus, 1758) from the Moroccan Middle Atlas and Rif -this identification needing to be confirmed- (Thomas, 1998), *A. oukaimeden* (Thomas & Sartori, 1992) from the Moroccan High Atlas known at both imaginal and larval stage (Thomas, 1998; Thomas et al., 1992), and *A. sadati* Thomas, 1994 from Algeria described only at the imaginal stage (Thomas & Gagneur, 1994). The genus was also mentioned from Tunisia but without specific designation (Boumaïza & Thomas, 1995; Thomas, 1998).

Intensive samplings (57 localities from 2005 to 2010) were carried out in several streams of Tunisia (Zrelli et al., 2006; Zrelli et al., 2011a; Zrelli et al., 2011b), allowing the collection of several larvae of *Alainites*. Morphological comparison with the larvae from the typical river of *A. sadati* in Algeria reveals that the material is conspecific. This species is cited for the first time in Tunisia and was originally described only at the imaginal stage. So far *A. sadati* is the only species recorded in Algeria and Tunisia, over a longitudinal range exceeding 1,100 km. Moreover, as far as it was recorded at only 40 km from Morocco, without natural frontier, it is probably present also in the North Eastern part of this country.

The material examined is housed in the Collection of the Laboratory of Environment Biomonitoring (LBE), Unit of Hydrobiology, Faculty of Sciences of Bizerta, Tunisia, in Alain Thomas personal collection (AT) and in the Museum of Zoology, Lausanne, Switzerland (MZL).

*A. sadati* Thomas, 1994
(Figs 1–6)

**Specimens examined**

**Algeria** (J. Gagneur coll.).

Tlemcen : Riv. *Khemis* K0, 34°34'21"N 1°37'08"W, alt. 1045 m, 15 to 25-X-1981, 9 larvae and 1 last instar larva on microscopic slide (AT), 10 larvae and 1 last instar larva on microscopic slide (MZL); same locality, 11 to 25-XI-1981, 3 last instar larvae on microscopic slides (AT); same locality, 4-IV-1984, 8 larvae (AT); same locality, 2-IV-1985, 1 last instar larva on microscopic slide (AT). Riv. *Khemis* K1, 34°37'03"N 1°35'33"W, alt. 950 m, 4 to 22-V-1981, 1 larva (AT). Riv. *Khemis* K2, 34°40'39"N 1°30'29"W, alt. 650 m (*Locus typicus*), 4 to 22-V-1981, 3 larvae (AT). Riv. *Tafna* T4, 34°44'32"N 1°35'29"W, alt. 470 m, 4 to 22-V-1981, 2 larvae (MZL). Riv. *Bou Messaoud* B1, 34°57'57"N 1°21'42"W, alt. 375 m, 4 to 27-V-1981, 1 larva (AT), 9 larvae and 1 last instar larva on microscopic slide (MZL). Riv. *Saf Saf* SS, 34°53'14"N 1°14'51"W, alt. 700 m, 25-IV-1986, 4 larvae (MZL); same locality, 24-V-1986, 1 last instar larva on microscopic slide (AT). Riv. *Chouly* C1, 34°50'18"N 1°49'46"W, alt. 800 m, 4 to 22-V-1981, 1 larva (MZL); same locality, 17-X-1982, 2 larvae (MZL). Riv. *Chouly* C2, 34°55'22"N 1°01'40"W, alt. 353 m, 4 to 22-V-1981, 4 larvae (MZL); same locality, 11 to 25-XI-1981, 4 larvae (AT); same locality, 22/23-XII-1981, 2 larvae and 1 last instar larva on microscopic slide (MZL); same locality, 24-II to 11-III-1983, 10 larvae and 2 last instar larvae on microscopic slides (AT), and 1 larva (MZL).

**Tunisia** (S. Zrelli coll.).

Aïn Draham : Riv. *Ennour*, 36°48'02"N 8°39'31"E, alt. 418 m, 27-VII-2005, 4 larvae (LBE). *Brensia*, 36°46'51"N 8°39'31"E, alt. 588 m, 31-III-2006, 1 larva (LBE); Beja : Riv. *Beja*, 36°45'38"N 6°51'39"E, alt. 190 m, 27-III-2009, 1 larva on microscopic slide (MZL). Riv. *Ksar Mezouar*, 36°46'58"N 8°20'11"E, alt. 236 m, 01-XI-2005 (LBE); same locality, 28-V-2005, 2 larvae (LBE); same locality, 29-VII-2005, 15 larvae (LBE); same locality, 02-X-2005, 18 larvae (LBE); same locality, 01-XI-2005, 4 larvae (LBE); same locality, 16-II-2006, 18 larvae (LBE); same locality, 30-IV-2006, 1 larva (LBE). Fernana : Riv. *Ellil*, 36°34'35"N 8°44'02"E, alt. 23 m, 28-VII-2005, 7 larvae (LBE + MZL); same locality, 13-IX-2005, 4 larvae (LBE); same locality, 12-XII-2005, 2 larvae (LBE); same locality, 02-IV-2006, 14 larvae (LBE); same locality, 30-IV-2006, 2 larvae (LBE). Riv. *Ghrib*, 36°22'10"N 8°24'41"E, alt. 260 m 30-IV-2006, 1 larva (LBE).
FIGURE 1–6. Larval structures of *Alainites sadati* from Khemis (Algeria) except Fig. 3b from Beja (Tunisia): 1. Labrum (left: ventral, right: dorsal). 2. Right mandible. 3a. Left mandible. 3b. Left prostheca and incisors. 4. Hypopharynx. 5. Right maxilla. 6. Labium (left: ventral, right: dorsal).
Diagnosis of *Alainites sadati*


*Imago*: Forewing hyaline with hyaline venation and double intercalary veins. Hindwing with a broad and short costal spur; three longitudinal veins, second vein bifurcated in proximal half, additional veinlets present. Abdominal colouration uniformly dark brown. Male gonopods segment III long and proximally narrowed.

**Description**

**Larva**

**Length.** Last instar female: general body 4.7–6.7 mm; cerci 3.3 mm; median caudal filament 2.1 mm. Last instar male: general body 3.9–6.0 mm; cerci 2.9 mm; median caudal filament 1.5 mm.

**Colouration.** General colouration medium brown. Head uniformly medium brown, slightly darker between ocelli and at insertion of antennae. Turbinate eyes in male larvae orange. Legs ecru. Thorax median brown without mark or pattern. Abdominal tergites medium brown without any pattern. Abdominal sternites light brown. Cerci and median caudal filament ecru to light brown without bands or pattern.

**Head.** Antennae close to each other, with an interantennal carina. Dorsal surface of labrum (Fig. 1) with five long and stout setae not arranged in a row in the distal half and numerous small to medium setae scattered on the surface; ventral surface with four small pointed setae near lateral margin; distal margin with feathered setae. Hypopharynx (Fig. 4): lingua slender and trilobed covered with short thin setae; superlinguae with short triangular spines apico-laterally. Right mandible (Fig. 2) with incisors composed of eight denticles, the fifth smaller and inward, inner margin of incisors with a row of very thin setae; prostheca bifid with thin setae; margin between prostheca and mola with tiny denticles, tuft of setae at apex of mola present. Left mandible (Fig. 3a): incisors with seven denticles (sometimes presence of an eighth small denticle, in outer position (Fig. 3b)); prostheca with ten to twelve (generally eleven) denticles and a comb-shaped structure; margin crenulated between prostheca and mola but without setae; tuft of setae at apex of mola absent. Maxillae (Fig. 5) with four elongated teeth; lacinia with two rows of setae, one with abundant small setae ending with stouter and longer setae towards the outer margin, second row with two long stout dentisetae; a single thin seta at base of teeth, frequently broken off, row of four stout setae at base of lacinia; palp two-segmented; segment II apically rounded, covered with thin setae. Labium (Fig. 6) with glossae slightly shorter than paraglossae; margins of glossae with medium setae, ventral surface with a row of medium setae, dorsal surface with abundant thin setae; paraglossae falcate, with three rows of long, stout setae apically; labial palp three-segmented; segment II with a dorsal oblique row of five fairly long pointed setae; segment III subconical, with short to medium setae.

**Thorax.** Forelegs (Fig. 7a). Surface of trochanter with numerous pointed setae. Femora not reticulated, dorsally with one row of stout setae, dorsoapical setal patch formed by three stout and pointed setae; ventral margin with very abundant stout setae. Tibiae not reticulated, dorsal margin with a patch of 6–9 pointed setae apically; tibiopatellar suture faintly visible; mesotibia with 9 to 13 pointed setae along dorsal margin (Fig. 7b). Tarsi: dorsal margin with a very few thin setae, ventral margin with about 15 pointed setae. Tarsal claws (Fig. 8) with a single row of teeth increasing in size towards the apex (considering 35 legs of 8 last-instar larvae: range = 11 to 16 teeth; mean = 13.4; SD = 1.2). Hindwing pads present.

**Abdomen.** Tergites slightly reticulated; distal margin of tergite IV with medium triangular spines (Fig. 9). Gills on segments II to VII, all well developed, elliptic and serrated all along margins; tracheation well visible but poorly divided (Fig. 10). Paraproct (Fig. 11) with abundant scale bases, without setae, prolongation covered with numerous small spines, margin with 7 broad, triangular spines inner to prolongation and numerous small spines outer to prolongation; postero-lateral extension covered with scale bases, margin with 15 small to medium spines.

Discussion

The type species of *Alainites*, *A. muticus* (Linnaeus, 1758), differs from all the other Western Palearctic species by having seven pairs of gills (versus six in other species including *A. sadati*). Characters used to separate the different species of *Alainites* were discussed in detail by Sartori & Thomas (1991). Using the same set of characters (table 1), *A. sadati* can be distinguished from *A. oukaimeden* and *A. navasi* (Müller-Liebenau, 1974) by the degree of reticulation of the tergites and mandibles, from *A. kars* (Thomas & Kazanci, 1989) by the number of stout setae on the dorsal margin of femora and tarsi, from *A. albinatii* (Sartori & Thomas, 1989) by the spination of the surface of the extension of the paraprocts. Characters such as the setation of the ventral margin of femora and the shape of the third segment of the labial palp are not always illustrated for the different species, but are certainly of interest for the specific identification. Despite being separated by about 1000 km, Algerian and Tunisian populations appear extremely similar morphologically.

Distribution and habitat

The distribution of *Alainites sadati* appears rather restricted; the species is only reported from the North West of Algeria and from the North of Tunisia. Despite important samplings covering a great part of Tunisia (Zrelli et al., 2006; Zrelli et al., 2011a; Zrelli et al., 2011b), it was collected only in five different wadis in the humid mountains of North Western Tunisia; it seems completely absent from the South of Tunisia, confirming the preliminary observations of Boumaïza & Thomas (1995) s. n. *Alainites sp*. In North Western Algeria, its frequency of occurrence was 22.5 % over 40 sites covering the Tafna catchment, and this species is notably absent from the eight small coastal wadis prospected in the same time in the vicinity of this large wadi. Larvae live in mountains brooks and streams with permanent water and stony bottom preferring mainly cobble and pebble. Nymphs were often found in vascular hydrophytes and plant debris.
Table 1: Some discriminant characters between *Alainites sadati* and five related species: *A. albinatii*, *A. kars*, *A. muticus*, *A. navasi* and *A. oukaimeden*. D.M. = dorsal margin

<table>
<thead>
<tr>
<th></th>
<th>PAIRS GILLS</th>
<th>SURFACE TERGITES</th>
<th>PARAPROCT EXTENSION</th>
<th>DISTAL MARGIN TERGITE IV: SPINES</th>
<th>MARGIN BETWEEN RIGHT PROSTHECA AND MOLA</th>
<th>SURFACE MANDIBLES</th>
<th>SETAE D.M. FORE-FEMUR</th>
<th>SETAE D.M. FORE-TIBIA</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>albinatii</em> (Sartori and Thomas, 1989) Corsica</td>
<td>6</td>
<td>slightly reticulated</td>
<td>half covered by spines</td>
<td>long triangular pointed</td>
<td>~ 10 teeth</td>
<td>scale bases + reticulated</td>
<td>~ 15</td>
<td>~ 7</td>
</tr>
<tr>
<td><em>kars</em> (Thomas and Kazanci, 1989) Turkey</td>
<td>6</td>
<td>slightly reticulated</td>
<td>spines only on border</td>
<td>wide, rather short, pointed</td>
<td>no teeth</td>
<td>just a few blunt spines + setae</td>
<td>&gt; 40</td>
<td>~ 9</td>
</tr>
<tr>
<td><em>muticus</em> (Linnaeus, 1758) Europa</td>
<td>7</td>
<td>slightly reticulated</td>
<td>spines only on border</td>
<td>short triangular, broad basally, acute</td>
<td>~ 10 teeth</td>
<td>rare scale bases</td>
<td>~ 14</td>
<td>~ 8</td>
</tr>
<tr>
<td><em>navasi</em> (Müller-Liebenau, 1974) West Mediterranean</td>
<td>6</td>
<td>nearly smooth</td>
<td>covered by few spines</td>
<td>short triangular</td>
<td>~ 10 teeth</td>
<td>scale bases + numerous scales</td>
<td>~ 26</td>
<td>~ 21</td>
</tr>
<tr>
<td><em>oukaimeden</em> (Thomas and Sartori, 1991) Morocco</td>
<td>6</td>
<td>strongly reticulated</td>
<td>covered by spines</td>
<td>long relatively narrow</td>
<td>~ 10 teeth</td>
<td>reticulated</td>
<td>~ 19</td>
<td>~ 8</td>
</tr>
<tr>
<td><em>sadati</em> Thomas, 1994 Algeria/Tunisia</td>
<td>6</td>
<td>slightly reticulated</td>
<td>covered by spines</td>
<td>medium triangular</td>
<td>~ 10 teeth</td>
<td>just a few setae</td>
<td>23–25</td>
<td>6–7</td>
</tr>
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Alainites sadati was found in streams from 2 m (Ksar Mezouar) to 10 m width (Ellil), and water depth from 30 to 90 cm in Tunisia, and similarly from < 1 m (Bou Messaoud and Chouly) to 6 m (Khemis) in NW Algeria. The altitudinal range is comprised between 235 and 590 m in Tunisia, and between 270 and 1045 m in NW Algeria where A. sadati shows the second highest mean elevation of colonized sites (709 m) after Baetis maurus Kimmins, 1938 (759 m) among ten Baetidae species over 48 study sites. Considering the maximal temperature of streams, A. sadati exhibits also the second lowest value among Baetidae (mean = 21.1°C, SD = 3.8) after Baetis maurus (mean = 20.7°C, SD = 3.2).

These Tunisian streams are characterized by very low conductivity (< 553 µs/cm, SD = 216). Like Baetis punicus Thomas, Boumaïza & Soldán, 1983 and Centroptilum luteolum (Müller, 1776) but contrary to other Tunisian Baetidae, A. sadati is not adapted to high mineralized waters (Boumaïza & Thomas, 1995). In NW Algeria, A. sadati shows the lowest mean value of water conductivity at sites (738 µs/cm, SD = 141), together with two rare species: Baetis numidicus Soldán & Thomas, 1983 and Paraleptophlebia cincta (Retzius, 1783). Its limited ecological range probably explains its restricted distribution in Northern Tunisia and its absence from South of Tunisia.

Due to their very large ecological range, Baetis rhodani (Pictet, 1843) and Cloeon cognatum Stephens, 1835 were always collected together with A. sadati. Nigrobaetis rhithralis (Soldán & Thomas, 1983) also often occurs with A. sadati (Zrelli et al., 2011a). The two species of Leptophlebiidae, Choroterpes (Euthraulus) lindrothi Peters, 1980 and Habrophlebia consigloii Biancheri, 1959 are among the most stenotopic and most sensitive species of Tunisian mayflies, they were also often collected with A. sadati.

Acknowledgments

The authors thank Mr James Gagneur of University of Tlemcen, then University of Toulouse now retired, for the donation of his material from NW Algeria. The first author is very grateful to Dr Michel Sartori, Director of the Museum of Zoology in Lausanne for hosting her and to conduct a stage research in his laboratory; to Dr Mustapha Bejaoui for his help and advice. Dr Robert D. Waltz (USA) and an anonymous reviewer are thanked for their comments.

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