www.biodicon.com Biological Diversity and Conservation

ISSN 1308-8084 Online; ISSN 1308-5301 Print 11/1 (2018) 139-142

Research article/Araştırma makalesi

Isolate algae population on *Simulium ornatum sp. comp*. from four different streams in Eskişehir and Kütahya/Turkey

Ebru Ceren FİDAN [*1], Betül YILMAZ ÖZTÜRK [2] and Ümit ŞİRİN [1]

[1] EskişehirOsmangazi University, Department of Arts and Sciences, Meşelik Campus, Eskişehir, Turkey.
[2] Eskişehir Osmangazi University, Central Research Laboratory Application and Research Center, Meşelik Campus, Eskişehir, Turkey

Abstract

This study presents 14 algae species belonging to three different phylum. Algae species are isolated from thorax and abdomen part of *Simulium ornatum sp. comp*. larvae. Specimens are collected from 4 streams from Eskişehir and Kütahya provinces. These algae species are isolated for the first time from a *Simulium ornatum sp. comp*. larvae. All of the species are new record for study sites. Taxonomic list and pictures of algae species are given in the text.

Key words: isolate, Algae, *Simulium ornatum sp. comp*, Turkey

---------- * ----------

Eskişehir ve Kütahya illerinde bulunan dört farklı akarsudan elde edilen *Simulium ornatum sp. comp*. üzerinden izole edilmiş alg populasyonları

Özet

Bu çalışma ile üç farklı filuma ait 14 alg türü sunulmaktadır. Alg türleri *Simulium ornatum sp. comp*. larvasının torax ve abdomen bölgelerinden izole edilmiştir. *Simulium ornatum sp. comp*. türleri Eskişehir ve Kütahya illerinde bulunan dört farklı akarsudan toplanmıştır. Tespit edilen alg türleri bir *Simulium ornatum sp. comp*. larvası üzerinden ilk kez izole edilmektedir. Alg türlerinin hepsi çalışma alanı için yeni kayıttır ve tüm türlerin taksonomik listesi ve fotoğrafları metin içinde verilmiştir.

Anahtar kelimeler: izole, Alg, *Simulium ornatum sp. com*, Türkiye

1. Introduction

Simuliidae (Diptera) larvae and pupae are inhabits freshwaters, their habitats and altitude can range from mean sea level to nearly 5000 m altitude. The larvae are similar to its pupae, live attached to plants, rocks and smooth surface in water. This family play an important role in the life cycle of freshwaters. Adults are strong flyers, and females are vectors of a lot of pathogens because of their biting habits, sothey are important for public health(Coscarón & Arias, 2007).

Simuliidae larvae are using their cephalic head fans to catch solid particles from running waters. Larvae are also able to scrape periphyton from the substrate to which they are attached (Werner & Pont, 2003). Larvae is generally feed on algae, bacteria, organic particles of dead organisms and inorganic material, showing a great capacity to metabolize different materials (Coscarón & Arias, 2007).The importance of algae in the diet of aquatic insects is highly variable. Because of their high fat and protein content Simuliidae is consuming lots of algae specimens. While there are many researches about algae and bacteria composition in guts of some Simuliidae species (Alencar, Ludwig, Soares, & Hamada, 2001; Gíslason & Jóhannsson, 1991; Moore, 1977; Schröder, 1988; Thompson, 1989) there is no research about exterior surface of Simulidae specimens.

In this study *Simulium ornatum sp. comp*. had chosen because of its wide distribution in different streams and high tolerance to temperature, pollution and organic content, larvae and pupae of *Simulium ornatum sp. comp*. can be

* *Corresponding author* / Haberleşmeden sorumlu yazar: Tel.: +902222393750; Fax.: +902222291418; E-mail: ebruceren@ogu.edu.tr
 BioDiCon. 672-0317

found in very different types of running water. The *Simulium ornatum sp. comp*. is known to have five species in Turkey. In larval stages of this group has similar morphology. Larvae of different instars can be found nearly all year (Bernotien, 2010). The purpose of the study cultivate the algae specimens from exterior surface of *Simulium ornatum sp. comp.* and determinate them. It's thought to be Algaes can be use larvaes as habitat, besides larvaes use algaes as nutrients.

Algae are extremely various and found almost all around the world. They are producing nearly 70 percent of the air we breathe and they are very important in food chains of ocean an inland water (Altunöz, Obalı, Atıcı, & Arru, 2008; David M John, Whitton, & Brook, 2002).

They are quite different organisms due to their morphological structures. They can range from the microalgae, to macroalgae; large seaweeds Microalgae include both cyanobacteria, and green, brown and red algae. Cyanobacterias are also considered in algae because of their pigment content (Fabregas & Herrero, 1985; Hellio, Berge, Beaupoil, Le Gal, & Bourgougnon, 2002).

Algae can be found in freshwaters and seas, besides they can live in extreme industrial wastes backwaters. They can help the abstersion of heavy metals in water. It has been proven that the heavy metal absorption of microalgae is superior to the widespread physicochemical processes used to remove toxic heavy metals (Kumar, Dahms, Won, Lee, & Shin, 2015).

Algae are similar to high-order plant groups (with photosynthetic pigments) and, it is a group of organisms which is quite privileged because of its immature formation, cell wall structure, developmental stages, colonial lifestyle. At the same time, they make a significant contribution to the chemical change of oxygen, carbon dioxide and water cycle with their photosynthesis properties (Larkum, 2016; Prescott, 1973; Ramanan, Kim, Cho, Oh, & Kim, 2016).

2. Materials and methods

Simulium ornatum sp. comp. (Fig-1) was collected from four different streams which is located in Eskişehir and Kütahya provinces (Kargın, Sökmen, İncik and Akkaya streams) (Table-1;Fig-1) at April 2016. This four streams have almost the same feature and same species.

Table-1: Site names, Provinces Coordinates and Altitude information of Localities

Site Names	Provinces	Coordinates		Altitude
		N	E	
Kargın	Eskişehir	39 35 19	30 16 53	916 m
Akkaya	Eskişehir	39 37 14	30 18 41	817 m
İncik	Kütahya	39 32 40	30 15 30	998 m
Sökmen	Kütahya	39 30 13	30 11 27	1026 m

Figure-1: a. General view of *S. ornatum sp. comp.* b. Satellite image of localities

Four specimens were chosen from each sampling sites. The samples was washed with distilled water. The larvae obtained from their natural habitat were washed up in pure water three times. Then sweepings from the surface, particularly surfaces closer to the head by scratching extracted and added into the medium. To obtain pure culture, BG-11 (Table 2) medium was first prepared, followed by mixing with 2% agar to make solid medium (Rippka, 1988). BG 11 is evenly distributed in sterile petri dishes.

Table-2: BG-11 medium used in the purification of species

BG-11 Medium	**g/L**		
$NaNO_3$	15	**Trace elements**	**mg/L**
K_2HPO_4	0.4	H_3BO_3	61
$MgSO_4$. $7H_2O$	0.75	$MnSO_4$. H_2O	169
$CaCl_2$. $2H_2O$	0.36	$ZnSO_4$.$7H_2O$,	287
Citric acid	0.06	$CuSO_4$. $5H_2O$	2.5
Iron (III) ammonium citrate	0.06	$(NH_4)_6Mo_7O_{24}$. $4H_2O$	12.5
Na_2-EDTA	0.01		
Na_2CO_3	0.2		

Algae samples taken from larvae were transferred to these medium. The medium was incubated for 20 days at 25 °C under light of 3000 lux for 12 hours at night and 12 hours at light (Figure 3a). Subsequently, 100 ml of sterile liquid was transferred to the BG-11 medium (solutions containing various salts and trace elements) by taking a single cell from the colonies formed on the medium (Katırcıoğlu, Aslım, Türker, Atıcı, & Beyatlı, 2008; Rippka, 1988). Liquid medium were left to incubate for 20 days as in solid medium (Figure 3b).

The sources used in the diagnosis of the algae (D. M. John, Whitton, & Brook, 2005; Prescott, 1973; Seckbach & Kociolek, 2011). The algae Base, algal database was used to control the current systematic categories of the identified species.

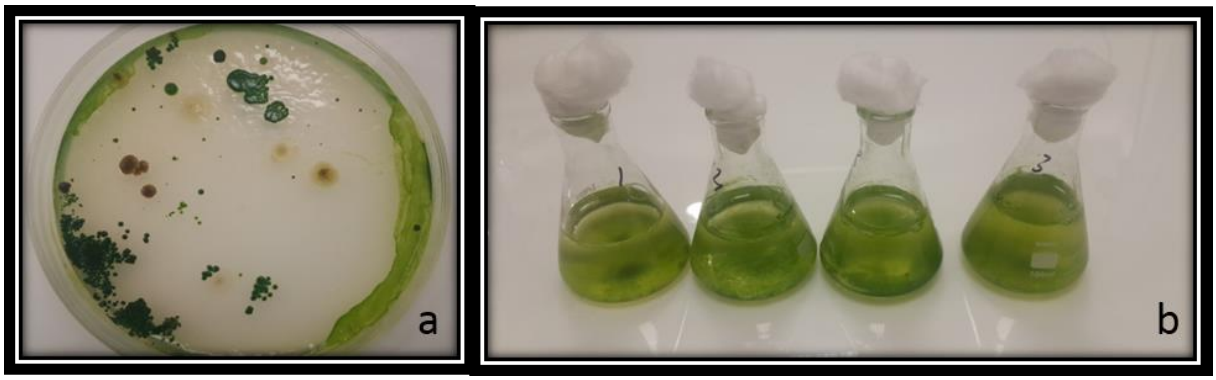

Figure 3. Images of cultures a. growing species in the solid medium b. growing species in the liquid medium.

3. Results

As a result of this study 14 different algae specimens were isolated with from all of the cultures. Taxonomical status and images of each species were given below**.**

1. ***Phylum*** Chlorophyta
   Subphylum Chlorophytina
   Class Chlorophyceae
   Order Sphaeropleales
   Family Scenedesmaceae
   ***Species Scenedesmus incrassatulus* Bohlin, 1897 (Figure 4-a)**

2. ***Phylum*** Chlorophyta
   Subphylum Chlorophytina
   Class Chlorophyceae
   Order Sphaeropleales
   Family Scenedesmaceae
   ***Species Scenedesmus incrassatulus var. mononae* G. M. Smith, 1914 (Figure 4-b)**

3. ***Phylum*** Chlorophyta
   Subphylum Chlorophytina
   Class Chlorophyceae
   Order Sphaeropleales
   Family Scenedesmaceae
   ***Species Scenedesmus bijugatus* Kützing, 1834 (Figure 4-c)**

4. ***Phylum*** Chlorophyta
   Subphylum Chlorophytina
   Class Chlorophyceae

Order Sphaeropleales
Family Scenedesmaceae
Species Scenedesmus quadricauda **Chodat, 1926 (Figure 4-d)**

5. ***Phylum*** Chlorophyta
Subphylum Chlorophytina
Class Chlorophyceae
Order Chlamydomonadales
Family Haematococcaceae
Species Haematococcus lacustris **(Girod-Chantrans) Rostafinski, 1875 (Figure 4-e)**

6. ***Phylum*** Chlorophyta
Subphylum Chlorophytina
Class Trebouxiophyceae
Order Chlorellales
Family Oocystaceae
Species Oocystis pusilla **Hansgirg, 1890 (Figure 4-f)**

7. ***Phylum*** Chlorophyta
Subphylum Chlorophytina
Class Chlorophyceae
Order Sphaeropleales
Family Scenedesmaceae
Species Coelastrum microporum **Nägeli, 1855 (Figure 4-g)**

8. ***Phylum*** Chlorophyta
Subphylum Chlorophytina
Class Trebouxiophyceae
Order Chlorellales
Family Chlorellaceae
Species Chlorella vulgaris **Beyerinck [Beijerinck], 1890 (Figure 4-h)**

9. ***Phylum*** Chlorophyta
Subphylum Chlorophytina
Class Chlorophyceae
Order Chlamydomonadales
Family Chlamydomonadaceae
Species Chlamydomonas mucicola **Schmidle, 1897 (Figure 4-i)**

10. ***Phylum*** Chlorophyta
Subphylum Chlorophytina
Class Chlorophyceae
Order Sphaeropleales
Family Selenastraceae
Species Selenastrum minutum **(Nägeli) Collins, 1907 (Figure 4-j)**

11. ***Phylum***Cyanobacteria
Class Cyanophyceae
Subclass Oscillatoriophycideae
Order Chroococcales
Family Chroocccaceae
Species Chroococcus turgidus **(Kützing) Nägeli, 1849 (Figure 4-k)**

12. ***Phylum*** Cyanobacteria
Class Cyanophyceae
Subclass Oscillatoriophycideae
Order Chroococcales
Family Microcystaceae
Species Microcystis aeruginosa **(Kützing) Kützing, 1846 (Figure 4-l)**

13. ***Phylum***Bacillariophyta
Subphylum Bacillariophytina
Class Bacillariophyceae
Subclass Bacillariophycidae
Order Naviculales
Family Naviculaceae
Species Navicula capitatoradiota **H.Germain, 1981 (Figure 4-m)**

14. ***Phylum***Bacillariophyta
Subphylum Bacillariophytina
Class Bacillariophyceae
Subclass Bacillariophycidae
Order Bacillariales
Family Bacillariaceae
Species Nitzschia perminuta **(Grunow) M.Peragallo, 1903 (Figure 4-n)**

Feeding behavior of aquatic beetles can be classified with morphology of mouthparts and functional feeding groups (Cummins, 1973). These groups is generally using for provide a great convenience on complication of organization but not specify the trophic roles of individual species (Mihuc, 1997). There are filter feeders, predators, foragers and scrappers in freshwater systems (Cummins, 1973). *Simulium* species are generally living in freshwater streams and rivers while in larval and pupal stages. Several studies have found negative effects of lowered species richness on ecosystem functioning depend on anthropogenic stress (Feld, Kiel, & Lautenschläger, 2002; Malmqvist, Zhang, & Adler, 1999). But also, they can show great adaptability on different habitats. They feed on organic particles of dead organisms, algae and bacteria besides inorganic material. They can show both positive and negative react to physical and chemical degradation as acidification and organic pollution (Glötzel, 1973). These conditions are effective factors for algaes, they are can outbreak in such places (Yakup, Ertürk, & Akkayunlu, 2010). Algeas, as nutritional source of blackflies, are contains starches, proteins, lipids, minerals and sugars that occur in photosynthesis. Besides, algal derived coproducts such as, carotenoids, omega 3 polyunsaturated fatty acids, β-carotene, (docosahexaenoic acid and eicosahexaenoic), astaxanthin, squalene, phycobiliproteins are important materials (Mehta et al., 2018).

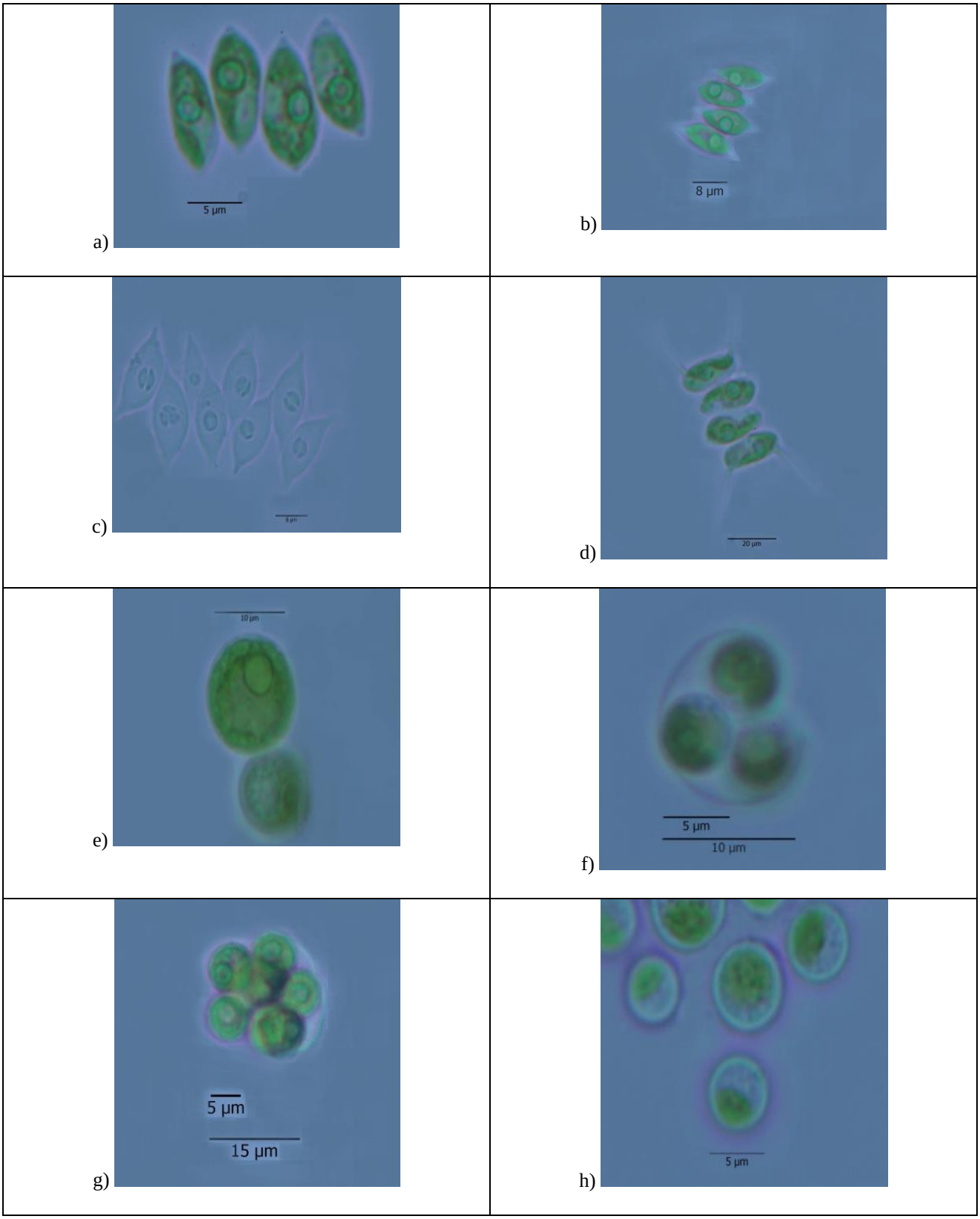

Figure 4. Images of Algae species which isolated from *S. ornatum sp. comp.* specimens

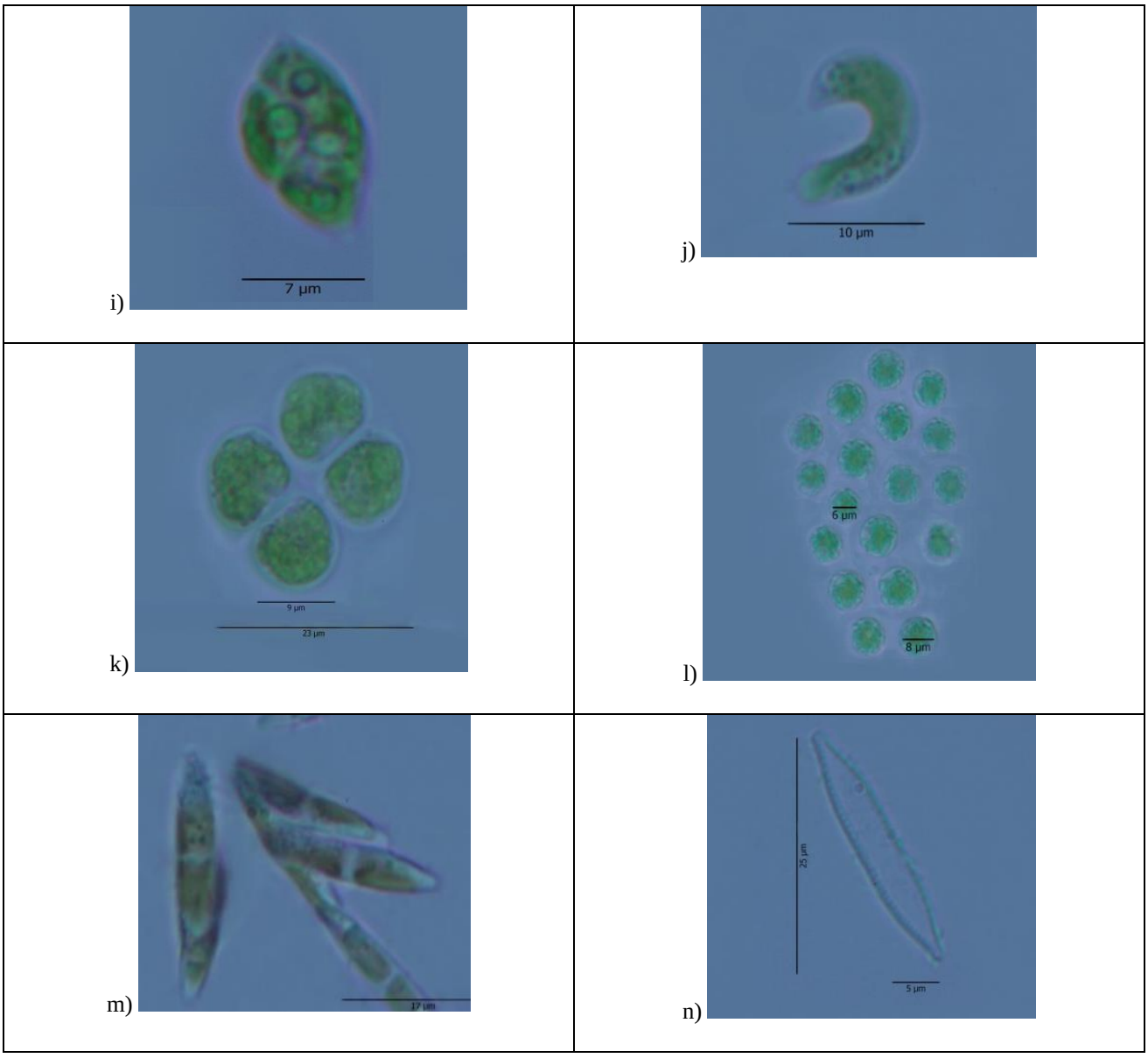

Figure 4. (continued)

4. Conclusions and discussion

Algae consumption from Simulidae has investigated from several studies (Burton, 1973; Schröder, 1988), but study about isolate algae community from external parts of Simuliidae larvae is not exist up until now. All of these species are first record for the study sites. With this primary study we had chance to observe that external surface of *Simulium ornatum sp. comp.* larvae, hosts algaes with quite diversity (10 species from Chlorophyta and two species for each Bacillariophyta and Cyanobacteria) and assume that it related to nutrition of *Simulium ornatum sp. comp.*

Algae species, which are isolated and cultivated, are all the same in four streams. So we cannot make comment about effect of streams ecology on biodiversity of algae community. Besides, a comparison study must be done from the internal and external algae composition of *S. ornatum sp. comp*. for understanding the roles of algaes and Simulidae species with each other.

References

Alencar, Y. B., Ludwig, T. A. V., Soares, C. C., Hamada, N. (2001). Stomach content analyses of Simulium perflavum Roubaud 1906 (Diptera: Simuliidae) larvae from streams in Central Amazonia, Brazil. *Memórias do Instituto Oswaldo Cruz, 96*(4), 561-576.

Altunöz, M., Obalı, O., Atıcı, T., & Arru, L. (2008). Seasonal monitoring of algal flora in the Pamukkale travertines and thermal springs (Denizli/Turkey).

Bernotien, R. (2010). On the biology of Simulium ornatum group (Diptera: Simuliidae), new and rare for Lithuania. . *Insect, Species*(22), 11-16.

Burton, G. J. (1973). Feeding of Simulium hargreavesi Gibbins larvae on Oedegonium algal filaments in Ghana. *Journal of medical entomology, 10*(1), 101-106.

Coscarón, S., Arias, C. L. C. (2007). *Aquatic Biodiversity in Latin America Volume 3: Neotropical Simuliidae (Diptera: Insecta)* (Vol. 3): Pensoft publishers.

Cummins, K. W. (1973). Trophic relations of aquatic insects. *Annual review of entomology, 18*(1), 183-206.

Fabregas, J., Herrero, C. (1985). Marine microalgae as a potential source of single cell protein (SCP). *Applied microbiology and biotechnology, 23*(2), 110-113.

Feld, C. K., Kiel, E., Lautenschläger, M. (2002). The indication of morphological degradation of streams and rivers using Simuliidae. *Limnologica-Ecology and Management of Inland Waters, 32*(3), 273-288.

Gíslason, G. M., Jóhannsson, V. (1991). Effects of food and temperature on the life cycle of Simulium vittatum Zett.(Diptera: Simuliidae) in the River Laxá, N-Iceland. *Internationale Vereinigung für theoretische und angewandte Limnologie: Verhandlungen, 24*(5), 2912-2916.

Glötzel, R. (1973). Populationsdynamik und Ernährungsbiologie von Simuliidenlarven in einem mit organischen Abwässern verunreinigten Gebirgsbach. *Arch. Hydrobiol. Suppl, 42*, 406-451.

Hellio, C., Berge, J. P., Beaupoil, C., Le Gal, Y., & Bourgougnon, N. (2002). Screening of marine algal extracts for anti-settlement activities against microalgae and macroalgae. *Biofouling, 18*(3), 205-215.

John, D. M., Whitton, B. A., Brook, A. J. (2002). *The freshwater algal flora of the British Isles: an identification guide to freshwater and terrestrial algae* (Vol. 1): Cambridge University Press.

John, D. M., Whitton, B. A., Brook, A. J. (2005). *The freshwater algal flora of the British Isles : an identification guide to freshwater and terrestrial algae*. Cambridge: Cambridge University Press.

Katırcıoğlu, H., Aslım, B., Türker, A. R., Atıcı, T., Beyatlı, Y. (2008). Removal of cadmium (II) ion from aqueous system by dry biomass, immobilized live and heat-inactivated Oscillatoria sp. H1 isolated from freshwater (Mogan Lake). *Bioresource Technology, 99*(10), 4185-4191.

Kumar, K. S., Dahms, H.-U., Won, E.-J., Lee, J.-S., Shin, K.-H. (2015). Microalgae–A promising tool for heavy metal remediation. *Ecotoxicology and environmental safety, 113*, 329-352.

Larkum, A. W. (2016). Photosynthesis and light harvesting in algae. In *The Physiology of Microalgae* (pp. 67-87): Springer.

Malmqvist, B., Zhang, Y., Adler, P. H. (1999). Diversity, distribution and larval habitats of North Swedish blackflies (Diptera: Simuliidae). *Freshwater Biology, 42*(2), 301-314.

Mehta, P., Singh, D., Saxena, R., Rani, R., Gupta, R. P., Puri, S. K., Mathur, A. S. (2018). High-Value Coproducts from Algae—An Innovational Way to Deal with Advance Algal Industry. In *Waste to Wealth* (pp. 343-363): Springer.

Mihuc, T. B. (1997). The functional trophic role of lotic primary consumers: generalist versus specialist strategies. *Freshwater Biology, 37*(2), 455-462.

Moore, J. (1977). Some factors effecting algal consumption in subarctic Ephemeroptera, Plecoptera and Simuliidae. *Oecologia, 27*(3), 261-273.

Prescott, G. (1973). Algae of the Western Qreat Lakes area Otto Koeltz Science Publishers, 977 p. In: Germany.

Ramanan, R., Kim, B.-H., Cho, D.-H., Oh, H.-M., Kim, H.-S. (2016). Algae–bacteria interactions: evolution, ecology and emerging applications. *Biotechnology advances, 34*(1), 14-29.

Rippka, R. (1988). [1] Isolation and purification of cyanobacteria. In *Methods in Enzymology* (Vol. 167, pp. 3-27): Academic Press.

Schröder, P. (1988). Distribution patterns of blackfly (Diptera: Simuliidae) associations in two Irish river systems. *Hydrobiologia, 164*(2), 149-160.

Seckbach, J., Kociolek, P. (2011). *The diatom world* (Vol. 19): Springer Science & Business Media.

Thompson, B. H. (1989). *The ingestion and digestion of algal and other foods by larval black flies (Diptera: simuliidae) of Newfoundland.* Memorial University of Newfoundland,

Werner, D., & Pont, A. (2003). Dipteran predators of Simuliid blackflies: a worldwide review. *Medical and Veterinary Entomology, 17*(2), 115-132.

Yakup, K., Ertürk, F., Akkayunlu, A. (2010). Journal of Engineering and Natural Sciences Mühendislik ve Fen Bilimleri Dergisi. *Sigma, 28*, 110-123.

*A part of this study was presented as a poster presentation at the 7th International Simuliidae Symposium, Zaragoza, Spain. September, 5th-8th 2016.

(Received for publication 29 March 2017; The date of publication 15 April 2018)