SOME DATA CONCERNİNG THE APPLICATION OF WEBB METHOD TO SEDIMENT SAMPLES FROM THE MURAT RIVER (AĞRI REGION – TURKEY)

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Abstract. The Murat River (in Turkish Murat Nehri, Murat Suyu or Murat Irmağı) is the eastern branch of the Euphrates. The documentation activity about the Murat River started in September 2011. This direction of research is new and original, as there have not been published any scientific papers in Turkey so far, especially dedicated to this ecosystem. Webb method is applied for the separation of ciliates from the Murat River sediments and there were obtained good results. The slides were analysed regularly for five days to establish the qualitative and quantitative composition of ciliate fauna. There were identified individuals belonging to 13 ciliate species on the slides during the observation days.

Keywords: Ciliates, the Murat River.

Rezumat. Câteva date privind aplicarea metodei Webb la probe de sediment provenite din Râul Murat (Regiunea Ağrı - Turcia). Râul Murat (în limba turcă Murat Nehri, Murat Suyu ori Murat Irmağı) reprezintă ramura estică a Eufratului. Activitatea de documentare privind râul Murat a început în septembrie 2011. Această direcţie de cercetare este nouă, în Turcia nu sunt multe articole ştiinţifice dedicate acestui ecosistem. Metoda Webb a fost aplicată cu rezultate bune pentru separarea ciliatelor de sedimentele râului Murat. Lamelele au fost analizate cu regularitate timp de patru zile în scopul stabilirii compoziţiei calitative şi cantitative a faunei de ciliate. În timpul observaţiilor au fost identificaţi indivizi aparţinând unui număr de 13 specii.

Cuvinte cheie: ciliate, Râul Murat.

INTRODUCTION

The observation and diagnosis of benthic ciliates is sometimes difficult taking into account their thigmotaxis capacity. In order to separate ciliates from origin sediments, there were discovered different methods, which present much or less advantages concerning the application time and distinct action according to the species fragility degree.

The separation method proposed by Uhlig (DRAGESCO & DRAGESCO-KERNÉIS, 1986b; UHLIG, 1964) is very advantageous if referring to the short time necessary to separate ciliates from sediments (only two hours), but aggressive for cells (especially fragile ciliates).

Although the duration of application is longer (maximum five days), Webb method consists in ciliates migration on the slides disposed at the surface of sediments looking for food and oxygen. We tested this method and the results are encouraging in case of the samples of sediment taken from the median part of the seashore and paramarine lakes (DUMITRACHE-KERKMANN, 2005). Our scientific paper presents results of Webb method application of sediments from the Murat River (Ağrı Region, Turkey). The Murat River (in Turkish Murat Nehri, Murat Suyu or Murat Irmağı) is the eastern branch of the Euphrates and it springs near the small town Doğubeyazıt. Agri city is the capital of the region with the same name and is located in eastern Anatolia; in year 2008, the population of this city was officially estimated to 91.817 inhabitants (WIKIPEDIA, 2012).

MATERIAL AND METHODS

The water and sediment samples collected from the second station of the Murat River (established in the area where the city sewerage system discharges into the Murat river; water temperature was 23.5 ºC) (KERKMANN et al., 2012; KERKMANN, 2012) were left to rest into laboratory for a few hours; after the removal of the water from the sediment, the first centimetres of the samples were distributed in three Petri Dishes noted WI (Photo 1).

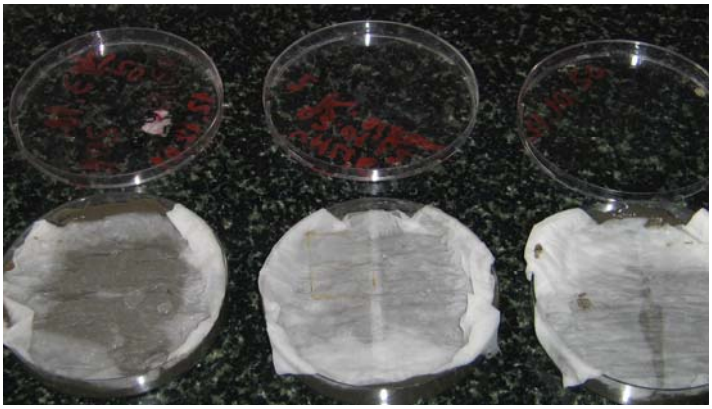

Photo 1 – Webb Method (original).

On the sediments wetted with distilled water (to prevent ciliates inflow from the water of the original ecosystem), it was applied a laboratory tissues and four slides. The lower sediment layer of the samples was put in another Petri dishes noted WII (see Table 1, legend) and subjected to the same procedure.

Table 1. Evidence of ciliates species on the slides of first sample.

Species	HP	W.I.I.1	W.I.I.2	W.I.I.3	WI.I.4	W.I.II.1	W.I.II.2	W.I.II.3	W.I.II.4	W.I.III.1	W.I.III.2	W.I.III.3	W.I.III.4
***Holophrya* sp.**	R	-	4	-	3	3,4	-	-	-	-	-	-	-
Urotricha globosa CLAPAREDE et LACHMANN 1857	Ba,Al	3	3	-	-	-	-	-	3	3	2,3	3	3
***Urotricha* sp.**	Ba,Al	4	4	4	4	4	4	4	4	4	4	4	4
***Prorodon* sp.**	R	4	-	-	-	-	-	-	3,4	-	-	-	-
***Didinium* sp.**	R	-	-	-	-	-	-	-	3	3	-	3	-
Plagiocamp arouxi KAHL 1932	Ba,Al	-	-	-	-	-	1,2	-	-	-	-	-	-
Plagioyla nasuta STEIN 1860	Ba,Sb, Al,Fl	3,4	4	-	3,4	-	3,4	-	3	-	-	-	-
Colpidium colpoda (LOSANA 1829) STEIN 1860	Ba,Fl, Al	4	4	4	4	-	-	-	-	-	-	-	-
Paramecium cf. aurelia EHRENBERG 1838	Ba	-	-	-	-	4	-	4	-	-	-	-	-
Uronema nigricans (MÜLLER 1786) FLORENTIN 1901	Ba,Fl	1,3,2,4	3,4	3,4	3,4	2,3,4	1,2,3,4	2,3,4	2,3,4	2,3,4	2,3,4	1,3,4	2,3,4
Vorticella campanula EHRENBERG 1833	Ba,Al	3,4	3,4	3,4	3,4	2,3,4	2,3,4	1,2,3,4	2,3,4	2,3,4	1,3,4	1,2,4	2,3,4
***Oxytricha* sp. 1**	Ba,Fl	-	-	-	4	-	3	-	-	-	-	-	1,3
***Stylonychia* sp.**	Ki,Fl	-	3,4	-	4	4	4	4	4	4	4	-	-
Forms under 10 µm	?	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3	1,2,3,4	1,2,4	2,3,4	1,2,3.4	1,2,3 4	1,2,3 4	1,2,3 4	1,2,3 4
Flagellata	-	1	-	1	1	-	-	1	-	-	-	-	-
Nematoda	-	1	-	-	1	-	2	1	-	1	1	-	1

Legend of Table 1 and 2: 1,2,3,4 – days of samples examination; **W I.I.1.** = sample I, Petri dish I, slide 1; W I.I.2 = sample I, Petri dish I slide 2; W I.I.3 = sample I, Petri dish I, slide 3; W I.I.4 = sample I, Petri dish I, slide 4; W I.II.1 = sample I, Petri dish II, slide 1; W I.II.2 = sample I, Petri dish II, slide 2; W I.II.3 = sample I, Petri dish II, slide 3; W I.II.4 = sample I, Petri dish II, slide 4; W I.III.1 = sample I, Petri dish III, slide 1; W I.III.2 = sample I, Petri dish III, slide 2; W I.III.3 = sample I, Petri dish III, slide 3; W I.III.4 = sample I, Petri dish III, slide 4; W II.I.1. = sample II, Petri dish I, slide 1; WII.I.2 = sample II, Petri dish I, slide 2; WII.I.3 = sample II, Petri dish I, slide 3; WII.I.4 = sample II, Petri dish I, slide 4; WII.II.1 = sample II, Petri dish II, slide 1; WII.II.2 = sample II, Petri dish II, slide 2; WII.II.3 = sample II, Petri dish II, slide 3; WII.II.4 = sample II, Petri dish II, slide 4; WII.III.1 = sample II, Petri dish III, slide; WII.III.2 = sample II, Petri dish III, slide 2; WII.III.3 = sample II, Petri dish III, slide 3; WII.III.4 = sample II, Petri dish III, slide 4; Al = algae (except of diatoms but inclusive autotrophic flagellates); Ba = bacteria; Fl = heterotrophic flagellates; Ki = diatoms; R = predator; Sb = sulphur bacteria (FOISSNER & BERGER, 1986).

The daily control of the slides aimed at establishing the qualitative and quantitative composition of ciliates (forms larger than 10 µm) migrated on the slides. The experiment finished after only three days considering that a complete and diversity ciliate fauna installed on the slides. The maximum recommended duration is five days. Some ciliates were studied "*in vivo*", while in case of others, there were applied colorations such as methyl green or haematoxylin.

Some quantitative data were obtained by daily counting of the forms larger than 10 µm. During the experiment, the Petri dishes were maintained in laboratory conditions, temperature ranging between 18 and 21ºC. There were also taken pictures. The systematic arrangement of the determined forms respects the systematics proposed by Puytorac et al. (DRAGESCO & DRAGESCO-KERNÉIS, 1986a).

RESULTS AND DISCUSSIONS

After the systematic examination of the slides disposed on the sediment surface, 13 species of Ciliata were found; 6 of them only to genus. During the experiment there were observed small forms (up to 10 µm), unidentifiable with the available equipment, as well as the presence of representative groups of protozoans or metazoans installed on the organic pellicle, which covers the exposed slides (Table 1).

The ciliate fauna from the sediments of the Murat River (Ağrı Region) interested us since September 2011, the first scientific results already making the subject of a scientific paper. Two ciliate forms completed the initial list proposed by us, *Prorodon* sp. and *Didinium* sp. (KERKMANN et al., 2012).

The ciliates migration on the slides disposed at the surface of the sediments is a dynamic phenomenon, migration being triggered by the search for food; most of the identified forms are bacterivorous, microalgivorous, while others feed on zooflagellates (FOISSNER & BERGER, 1986). When the parameters of one or many abiotic factors do not

correspond to ciliates requirements, they return into the first millimetres of the sediments or transform into cysts until favourable conditions return (personal observations).

In terms of ciliates fauna installed on the slides with sediments of the first sample, after analysing table 1, one may notice that in the first examination day, the number of identified species was small, except small forms (up to 10 µm), which were identified constantly during the experiment and were probably bacterivorous.

From the total of identified species in the four days of the experiment only two species seem to have adapted to the new conditions imposed by the presence of the artificial substrate. Thus, the individuals of *Vorticella campanula* (EHRENBERG 1833 and *Uronemanigricans* (MÜLLER 1786; FLORENTIN 1901) were identified on 3 of the 12 slides from the first examination day, representing the constant presence throughout the observations.

Other individuals of other species appeared on the slides in the second (*Urotricha globosa* CLAPAREDE et LACHMANN 1857), third (*Prorodon* sp., *Holophrya* sp., *Stylonychia* sp.) and even in the last day of the experiment *Urotricha* sp., *Colpidium colpoda* (LOSANA 1829) STEIN 1860, *Paramecium cf. aurelia* EHRENBERG 1838 respectively *Stylonychia* sp. Among the identified species, there are some which even if they appeared in the last day of observations, they were found on all the slides of sample 1 or on more than half of them (*Stylonychia* sp.). A possible explanation could be their ecological plasticity and the species evolution through the differentiation of cilia.

In terms of the evolution of the species number during the four observation days (Fig. 1), the situation is relatively balanced, their number increasing with the time passed from the initial moment.

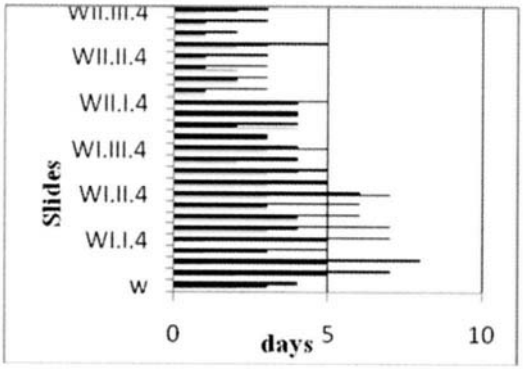

Figure 1. WEBB – Daily number of species on each slide.

Regarding the frequency of ciliates species developed on the slides of the second sample of sediments (WII), the situation is the same; the ciliates belonging to *Urotrycha* and *Stylonychia* genus appeared on the majority of the exposed slides in the last experiment day (Table 2).

Table 2. Evidence of the ciliates species on the slides of second sample.

Species	W.II.I.1	W.II.I.2	W.II.I.3	WII.I.4	W.II.II.1	W.II.II.2	W.II.II.3	W.II.II.4	W.II.III.1	W.II.III.2	W.II.III.3	W.II.III.4
***Holophrya* sp.**	-	-	-	-	-	-	-	-	-	-	-	-
Urotricha globosa CLAPAREDE et LACHMANN 1857	2	2	2,3	-	-	-	-	-	-	-	-	-
***Urotricha* sp.**	4	4	4	4	4	4	4	4	4	-	-	-
***Prorodon* sp.**	-	-	-	-	-	-	-	-	-	-	-	-
***Didinium* sp.**	-	-	-	-	-	-	-	-	-	-	-	-
Plagiocamp arouxi KAHL 1932	-	-	-	-	-	-	-	-	-	-	-	-
Plagiopyla nasuta STEIN 1860	-	-	-	-	-	-	-	-	-	-	-	-
Colpidium colpoda (LOSANA 1829) STEIN 1860	-	-	-	-	-	-	-	-	-	-	-	-
Paramecium cf. aurelia EHRENBERG 1838	-	-	-	-	-	-	-	-	-	-	-	-
Uronema nigricans (MÜLLER 1786) FLORENTIN 1901	1,2,3,4	2,3,4	1,2,3,4	2,3,4	-	-	-	-	4	-	3,4	4
Vorticella campanula EHRENBERG 1833	3	2	1,2,3	2,4	4	2,3,4	2,4	4	2,3,4	4	3,4	3,4
***Oxytricha* sp. 1**	-	-	-	-	-	-	-	-	3	-	-	4
***Stylonychia* sp.**	-	4	4	4	-	-	-	-	4	-	-	-
Forms under 10 µm	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3,4	1,2,3
Flagellata	-	-	-	-	-	-	-	-	-	-	-	-
Nematoda	-	4	4	-	4	4	-	4	-	-	-	-

Regarding the qualitative composition of the ciliates fauna (Figs. 2-4), during the first day of observation, there were identified only small forms (up to 10 μm); the same situation was observed in the second day, when on the slides surfaces there were identified the first larger ciliates forms (number variation between 1 and 7 individuals). The most abundant forms were observed in the third day of the experiment, the maximum value reaching 315 individuals (W.I.II.4), while in the fourth day, the number increased to 2.510.

Regarding the qualitative variation of the ciliate individuals belonging to different species, in the second day of the experiment, there were identified only two individuals of *Vorticella campanula* (EHRENBERG 1833) on slide W.I.III.I.; in the third day, the number reached 42, while in the fourth, it increased to 341; for *Uronema nigricans* (MÜLLER 1786; FLORENTIN 1901), we registered a similar evolution - 3 individuals (first day), 18 (second day), respectively 401 (fourth day). A possible explanation of their quantitative evolution could be the abundance of food - small algae and bacteria (Table 1).

CONCLUSIONS

1. During the four days of the experiment there were identified 13 ciliate species on the slides, most of them feeding on bacteria and small algae.
2. If in the first two days of the experiment the small forms (up to 10 μm) dominated, the number of larger forms increased in the next two days (Figs. 2-4), because most of them consume small algae and bacteria (Table 1).
3. Despite the longer time of observations and difficulty of work consisting in daily control of the slides, compared to other methods (UHLIG, 1964), the present method is recommended especially for fragile forms because it is not so brutal.

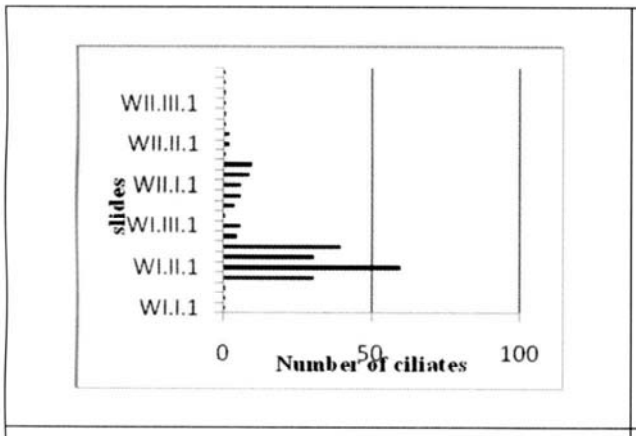

Figure 2. WEBB – Quantitative variation of the ciliates during the second day of the experiment.

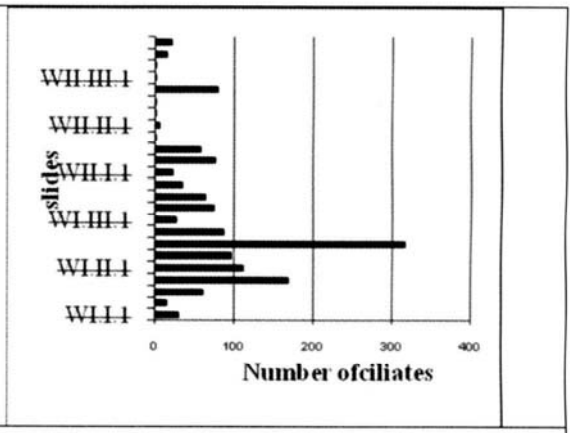

Figure 3. WEBB – Quantitative variation of the ciliates during the third day of experiment.

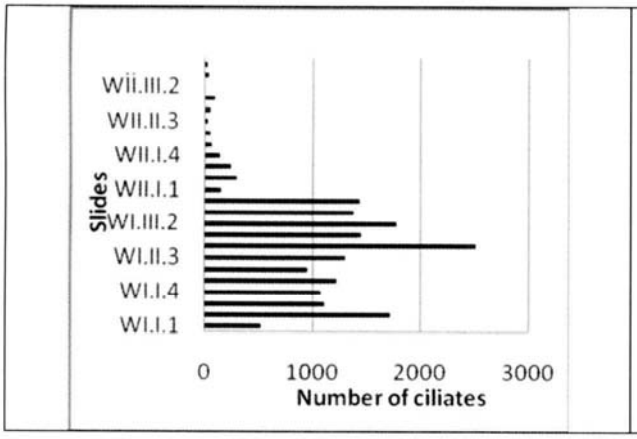

Figure. 4 -WEBB – Quantitative variation of the ciliates during the fourth day of experiment

REFERENCES

DRAGESCO J. & DRAGESCO-KERNÉIS ARMELLE. 1986a. *Classification de Phylum Ciliophora DOFLEIN, 1901*. În: Écologie des Ciliés. Écologie des Ciliés libres de l'Afrique intertropical (introduction à la connaissance et à l'étude des Ciliés) Faune Tropical. ORSSTOM. Paris. **26**: 71-75.

DRAGESCO J. & DRAGESCO - KERNÉIS ARMELLE. 1986b. *Récoltes, cultures, techniques de préparation et d'observation des Ciliés.* Écologie des Ciliés. Ciliés libres de l'Afrique intertropical (introduction à la connaissance et à l'étude des Ciliés) Faune Tropical. ORSSTOM. Paris. **26**: 83-85.

DUMITRACHE - KERKMANN GINA RALUCA. 2005. *WEBB - metoda eficientă de separare a ciliatelor din sedimentele marine si lacustre.* Argesis - Studii şi comunicări. Seria Ştiinţele Naturii. Muzeul Judeţean Argeş. Piteşti. **13**: 145-150.

FOISSNER W. & BERGER H. 1986. *A user-friendly guide to the Ciliates (Protozoa, Ciliophora) commonly used by hydrobiologists as bioindicators in rivers lakes and waste waters with nptes of theirecology.* Freshwater Biology. Paris. **35**: 377-385.

KERKMANN GINA RALUCA. 2012. *Preliminary data regarding the qualitative composition of ciliates from the Murat River (Agri Region, Turkey).* Oltenia. Studii şi comunicări. Ştiinţele Naturii. Muzeul Olteniei Craiova. **28**(1): 41-44.

KERKMANN GINA RALUCA, HATUNOĞLU AŞKIM, HATUNOĞLU Y. 2012. *Preliminary data regarding anthropogenic impact on the Murat River (Ağrı Region).* International Journal of Ecosystems and Ecology Science (IJEES). Agricultural University of Tirana. **2**(2): 55-60.

UHLIG G. 1964. *Eine einfache Methode zur Extraction der vagilen, mesopsammalen Mikrofauna.* Helgol. Wiss. Meeresunters. **11**: 178-185.

WEBB M. G. 1956. *An ecological study of brackish water ciliates*. Journal of Animal Ecology, published by British Ecological Society. London. **25**: 148-175.

WIKIPEDIA. 2012. Available online at: http://en.wikipedia.org/wiki/A%C4%9Fr%C4%B1_Province (Accessed: February 7, 2013).

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