Caldasia 26(1) 2004: 275-290

SOME OBSERVATIONS OF BUCCAL DEFORMITIES IN CHIRONOMID LARVAE (DIPTERA: CHIRONOMIDAE) FROM THE CIÉNAGA GRANDE DE SANTA MARTA, COLOMBIA

Algunas observaciones sobre deformidades bucales en larvas de quironómidos (Diptera: Chironomidae) de la Ciénaga Grande de Santa Marta, Colombia.

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ABSTRACT
The mouthpart deformities of chironomid larvae are considered as indicators of environmental stress caused by water pollution such as heavy metals, pesticides, but as well by organic contamination. In the present study larvae from the side channels of the Ciénaga Grande de Santa Marta were investigated. The chironomid community of the channels are dominated by species of the genera *Goeldichironomus* and *Chironomus*. All together 21 taxa were determined. The average proportion of mouthpart deformities in all chironomid larvae was 21 percent. Incidents of mechanical abrasion reached up to 59 percent. The highest frequency of abnormalities occurred in *Goeldichironomus carus* in which more than 90 percent of all specimens were affected. These numbers found here are fairly above average frequencies observed even at polluted sites. Under natural conditions deformity frequencies are expected not to exceed 8 percent. On the one hand, distinct concentrations of heavy metals originating from Río Magdalena can be detected in the sediments of the channels. On the other, these concentrations do not reach the levels which are described as triggers of high deformation rates like those found here. The most plausible explanation of these contradictory findings assumes a non-detected synergism of several environmental stressors. Possibly, the physical structure of the deposited sediments and a pronounced nocturnal oxygen depletion owing to organic pollution contribute an important part to the crucial processes on a small spatial scale. However, as far as no further investigative efforts will be done the reason for this phenomenon of high deformity frequencies will remain conspicuous but speculative to a large extent.

Key words. Chironomidae, mouthpart deformities, heavy metals, organic pollution, sediment, Ciénaga de Santa Marta.

RESUMEN

Las deformidades bucales en larvas de quironómidos son consideradas como indicadores de estrés ambiental causado por contaminación de aguas con metales pesados, pesticidas, pero también con carga orgánica. En el estudio presentado aquí se investigaron las larvas de los canales laterales de la ciénaga grande de Santa Marta. La comunidad de los canales es dominada por las especies de los géneros *Goeldichironomus* y *Chironomus*. En total se determinaron 21 taxones. La tasa promedio de deformidades bucales, sobre todas las larvas de quironómidos, fue del 21%. Los casos de abrasión mecánica alcazaron el 59%. La frecuencia más alta de anormalidades ocurrió en *Goeldichironomus carus*, del cual más de 90% de todos los individuos estuveron afectados. Estos números encontrados aquí están bien por encima de las frecuencias promedio observadas, incluso en sitios contaminados. Bajo condiciones naturales, las tasas de deformidades normalmente no excede el 8%. Por un lado, se pueden detectar concentraciones marcadas de metales pesados en los sedimentos de los canales provenientes del río Magdalena. Por otro lado, estas concentraciones no alcanzan los niveles descritos como factores desencadenantes de altas tasas de deformidades bucales como las encontradas aquí. Una explicación plausible de estos resultados contradictorios asume un sinergismo no detectado de diferentes estresores ambientales. Posiblemente, la estructura física de los sedimentos depositados y un agotamiento nocturno de oxígeno, a causa de la contaminación orgánica, contribuyen a una parte importante de los procesos esenciales a pequeña escala espacial. Sin embargo, en tanto que no se hagan más esfuerzos investigativos, la causa de este fenómeno de altas tasas de deformidades va a quedar especulativo hasta un cierto punto.

Palabras clave. Chironomidae, deformidades de las partes bucales, metales pesados, contaminación orgánica, sedimentos, Ciénaga de Santa Marta.

INTRODUCTION

Direct effluent discharges and agricultural runoff water most often contain complex mixtures of contaminants which may produce new compounds due to break-down and transformation processes and hence contribute to the complexity of the total toxic burden. By the employment of chemical and physical measurements only, the synergistic effect of pollution on its biotic community may not be fully and easily assessed (Rosenberg & Resh 1993). In general, biological indicators provide a potential for direct observation of the overall effect of environmental contaminants by virtue of their role in aquatic ecosystems (Warwick 1988).

For some regions of Colombia, methods of biological indication of organic pollution of running waters have been developed based on community structure as evidenced by macroinvertebrate families (Roldán 1999, Riss *et al.* in press) or by chironomid (Chironomidae: Diptera) genera (Riss *et al.* subm.). The latter group of aquatic insects is eminently important ecologically, in Colombia (Riss & Ospina 1998, 2000, Ospina *et al.* 1999, Ruíz *et al.* 2000a, b) and worldwide (Pinder 1983, Armitage *et al.* 1995, Coffman & Ferrington 1996).

However, the indication of organic contamination does not include toxic impacts. A promising biological indicator of toxic contamination of the water body and sediments lies in the frequency and patterns of morphological deformities occurring in chironomid larvae (Diptera: Chironomidae). Such abnormalities reflect sublethal effects and can be considered as early warning signals for environmental degradation by chemical contaminants (Warwick 1990). Deformities can be defined as morphological features that depart from the normal configuration (Warwick 1988, Madden *et al.* 1995). Effects produced by mechanical wear, breakage or abrasion are common phenomena of the mouthparts stressed by hard substrate surfaces, mineral particles and sclerotized organic material (Janssens de Bisthoven & Ollevier 1989, Vermeulen 1995). These are usually not included in deformity screening, unless their massive occurrence suggests a possible general weakening of chitinous structures due to chemical contamination. In cases with high proportions of totally worn and/or broken mouthparts the real picture of deformed larvae frequency in investigated populations can be obscured (Nazarova 1999).

The larvae of most chironomid species live on or in sediments, where they feed on organic matter (detritus) and the associated microfauna and flora. Because of their benthic feeding habits, these larvae are directly exposed to contaminants in sediments throughout their development. Results from many field studies strongly indicate a relationship between increased incidence of deformations and toxic sediment stress (Hamilton & Sæther 1971, Köhn & Frank 1980, Warwick 1980a, b, 1985, Janssens de Bisthoven & Van Speybroeck 1994, Nazarova *et al.* 2001a, b, etc.). In laboratory experiments it has been shown that chironomid deformities can be induced by heavy metals (Koswalt & Knight 1987, Janssens de Bisthoven 1995, Nazarova *et al.* 1999), organochloro-pesticides (Hamilton & Sæther 1971, Warwick 1985, Madden *et al.* 1992), cholinesterase inhibitors (Nazarova 2000), and other organic xenobiotics.

So far, no studies on chironomid deformities had been published for Colombia, and only few are available for the whole of South America (Pereira de Araújo 2000, Kuhlmann *et al.* 2001, Callisto *et al.* 2002). In general, chironomid research in this region is still a difficult task due to problems of identification, poor information on species assemblages, and the paucity of descriptions of larval morphology.

The goal of the present investigation was to study the biodiversity of the chironomid fauna in some channels around the Ciénaga Grande de Santa Marta, Colombia, and the frequency and severity of larval mouthpart deformities observed in relation to heavy metal concentration in sediments at the sampling sites.

STUDY AREA

The study sites are located along the main channel of Caño Clarín and the side branches (10°56’-11°00’ N, 74°33’-74°38’ W) which connect the río Magdalena and the two big lagoons Ciénaga Grande de Santa Marta and Ciénaga de Pajaral. These waters support rich, recently established vegetation, mostly of *Typha domingensis* and *Avicennia germinans* polycorms (Kahlheber 2000). The macrobenthic fauna associated with these macrophytes was sampled from September 1999 to February 2000. Contents of heavy metals in the sediments during the sampling period were the following (means and standard deviations, maximum values in brackets): Cu: 18 ± 7 (35) mg kg^{-1}, Cd: 1.3 ± 0.4 (2.4) mg kg^{-1}, Zn: 69 ± 18 (135) mg kg^{-1}, Pb: 11 ± 4 (18) mg kg^{-1}, Hg: 0.0024 ± 0.00005 mg kg^{-1}, and Cr 11 ± 1 mg kg^{-1} (Perdomo 1998, Cadavid & Espinosa 2001).

METHODS

A total of 27 samples of the epiphytic fauna were taken from *Typha* roots and the pneumatophores of *Avicennia*. Macroinvertebrates were washed from the plants by use of a hand net (0.1 mm mesh size) and the specimens stored in 70% alcohol. Chironomids were sorted and mounted following Warwick (1988). For identification the works of Wiederholm (1983), Epler (1995, 2001), Ospina *et al.* (1999), and Ruíz *et al.* (2000a, b) were used, as well as some other publications (Roback 1977, Roback & Coffman 1983, Cranston & Nolte 1996). Determinations were confirmed with the chironomid reference collection of the Laboratory of Invertebrates (Dept. of Biology, National University of Colombia, Bogotá). The percentage of chironomid larvae with misshapen head capsules in the total number of larvae was taken as a measure for the presence of toxic contaminants in the watercourse bed (Van Urk *et al.* 1992).

RESULTS

A total of 238 specimens was processed, and 21 taxonomic units belonging to 16 genera were identified. The most frequent genus was *Goeldichironomus*, including 2 species and one morphospecies, which accounted for 35 % of the total abundance. The genus *Chironomus*, with 3 morphospecies, made up 20 % of the total number. The remaining 45 % included the following taxa (in decreasing dominance): *Larsia* sp., Tanypodinae Genus A, *Cladopelma* sp., 2 morphospecies of *Polypedilum* sp., *Fissimentum desiccatum*, *Lauterborniella* sp., *Tanytarsus* sp., *Tanypus* sp., *Dicrotendipes* sp., *Coelotanypus* sp., *Labrundinia* sp., *Apedilum* sp., *Beardius* sp., and *Clinotanypus* sp. The conspicuous larva of the species *Fissimentum desiccatum* (Fig. 1) was first described from Brasil by Cranston & Nolte (1996) as an inhabitant of temporary standing water bodies and is the first record of this species for Colombia.

All larvae were examined for presence of deformities and other morphological abnormalities, such as excessive or and missing pigmentation, and abrasion of mouthparts (Table 1).

The highest frequencies of all types of abnormalities was found in the genera *Goeldichironomus* (Figs 2, 4-7) and *Chironomus* (Fig. 3). Species in the latter genus are frequently mentioned in the literature as morphologically quickly reacting to environmental stress (Warwick 1988, Wiederholm 1984). In contrast, no references could be found for deformities in species of *Goeldichironomus*. The most frequently met abnormalities in this genus were abrasion and breakages of the mentum (Fig. 4) and/or mandibles, which reached 96.5 % of all specimens in *Goeldichironomus carus*. In the most severe cases we observed completely and unevenly worn menta or mandibles (Fig. 5). Among the deformities in this species the most peculiar form were splits of the outermost lateral teeth which in most cases were developed asymmetrically (Fig. 6). In some cases additional asymmetrical teeth and splits of mentum were observed (Fig. 7). In all other taxa such specific morphological responses were not found. There the amount of abnormalities was relatively low, or the number of larvae was too small to be taken into account. In most cases the abnormalities concerned lacking or misshapen teeth owing to high abrasion (Fig. 8) Abnormalities observed in Tanypodinae were apparent on the hypopharyngal structures (Fig. 9 and 10). The overall proportion of larvae with buccal deformations was determined as 21%.

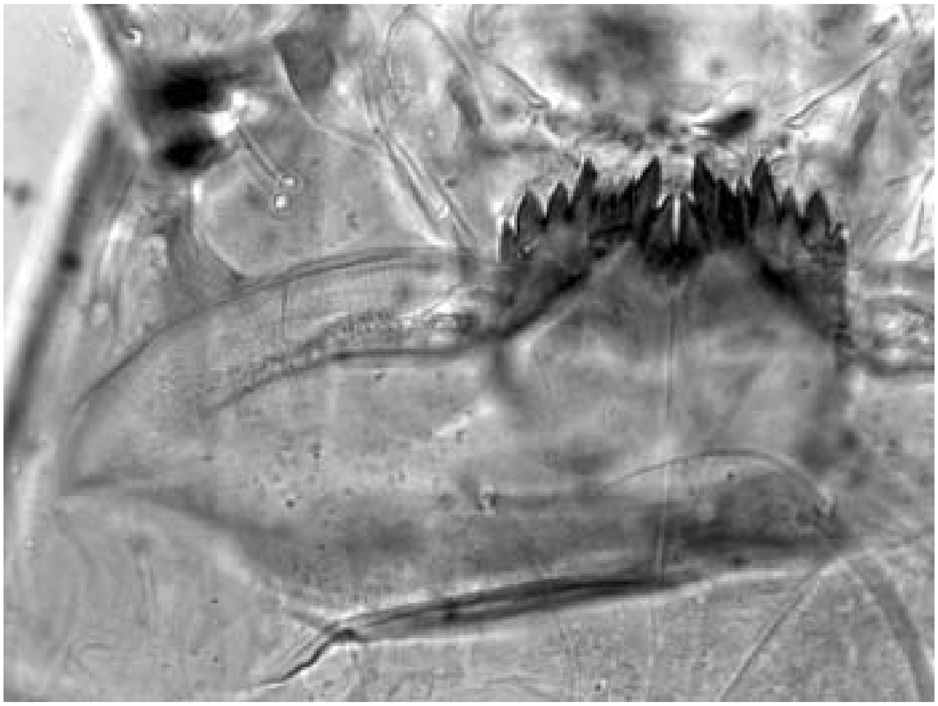

Fig. 1. *Fissimentum desiccatum,* larval mentum

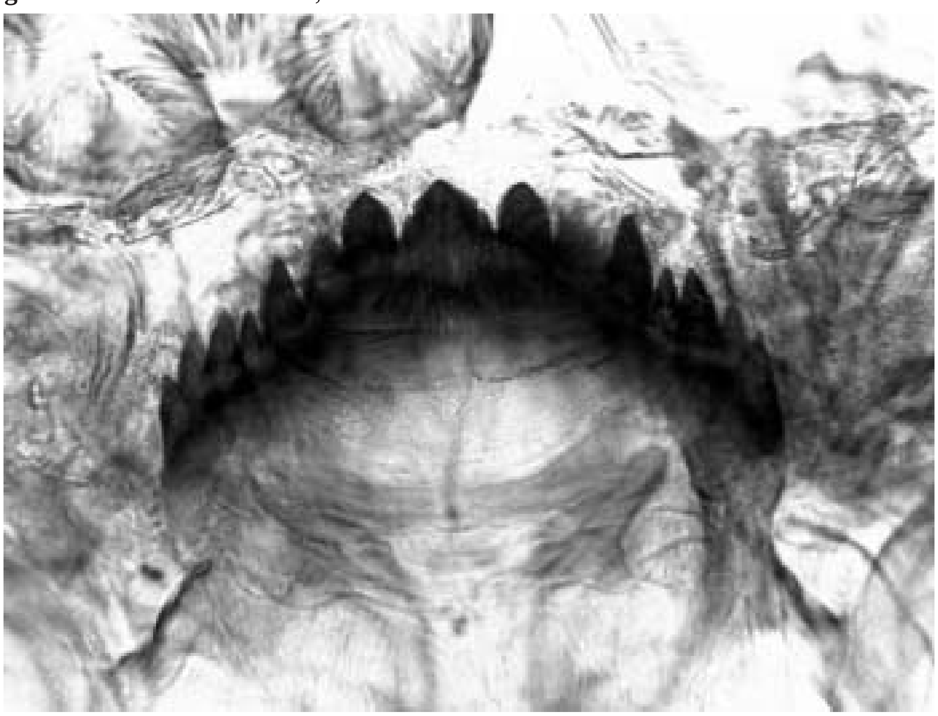

Fig. 2. *Goeldichironomus* sp., normally developed larval mentum

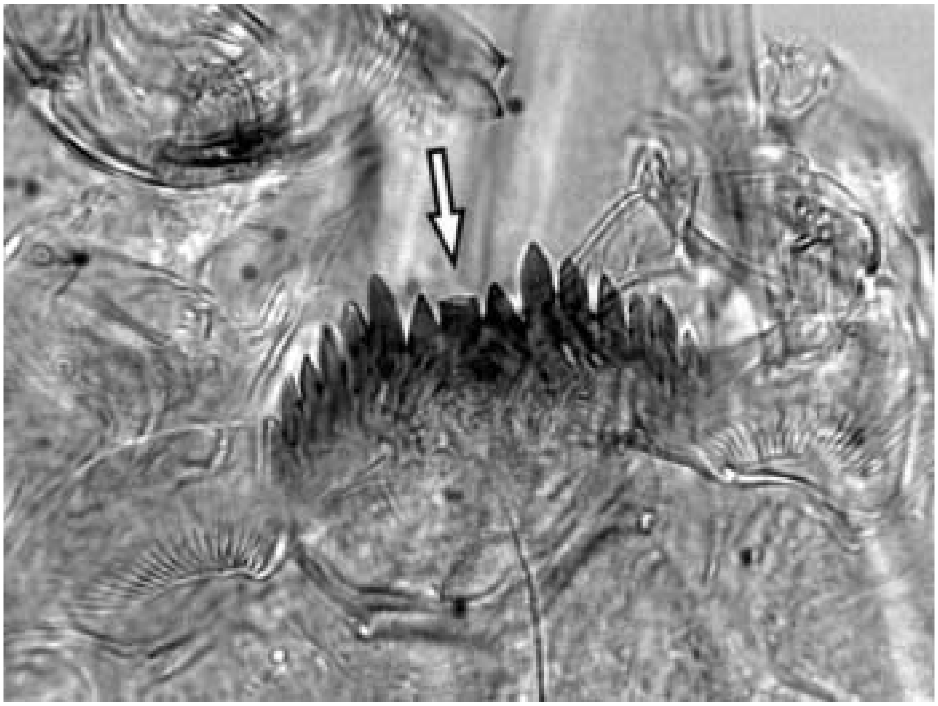

Fig. 3. *Chironomus* sp., larva (3rd instar), mentum with broken central tooth (arrow)

Fig. 4. *Goeldichironomus* sp., worn larval mentum with broken first lateral tooth (arrow)

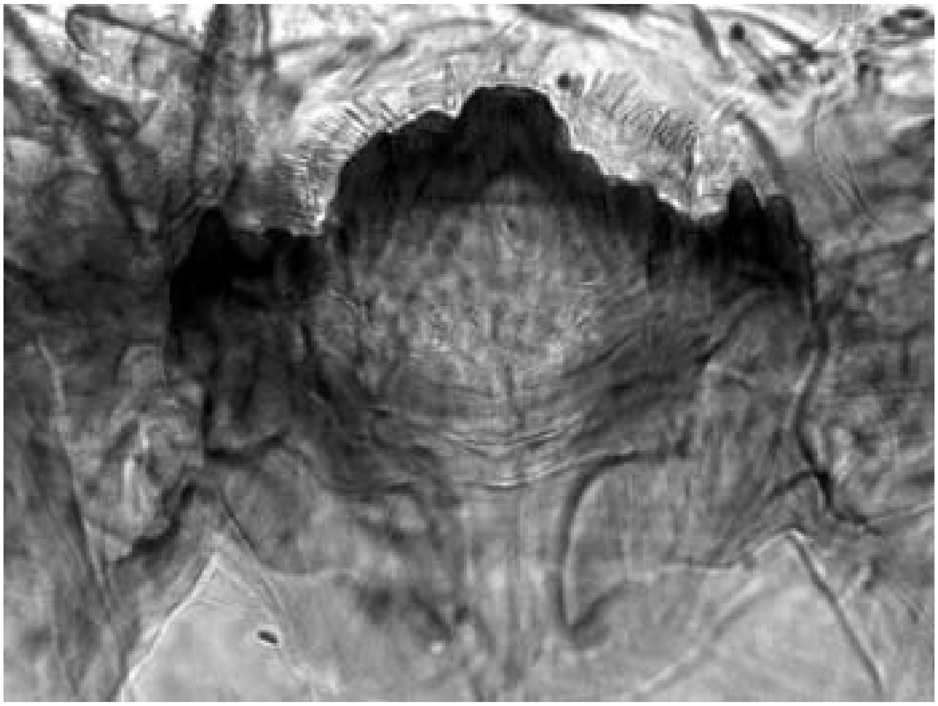

Fig. 5. *Goeldichironomus* sp., severely and unevenly worn larval mentum

Fig. 6. *Goeldichironomus* sp., asymmetrical splits in outermost lateral teeth (arrows) of larval mentum

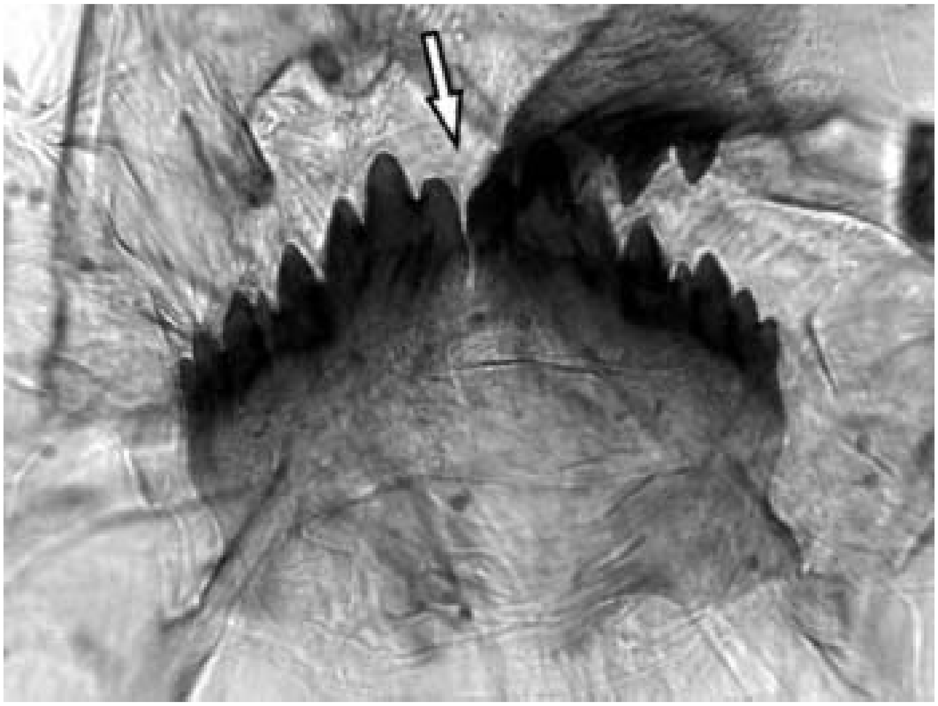

Fig. 7. *Goeldichironomus* sp., central split (arrow) in larval mentum

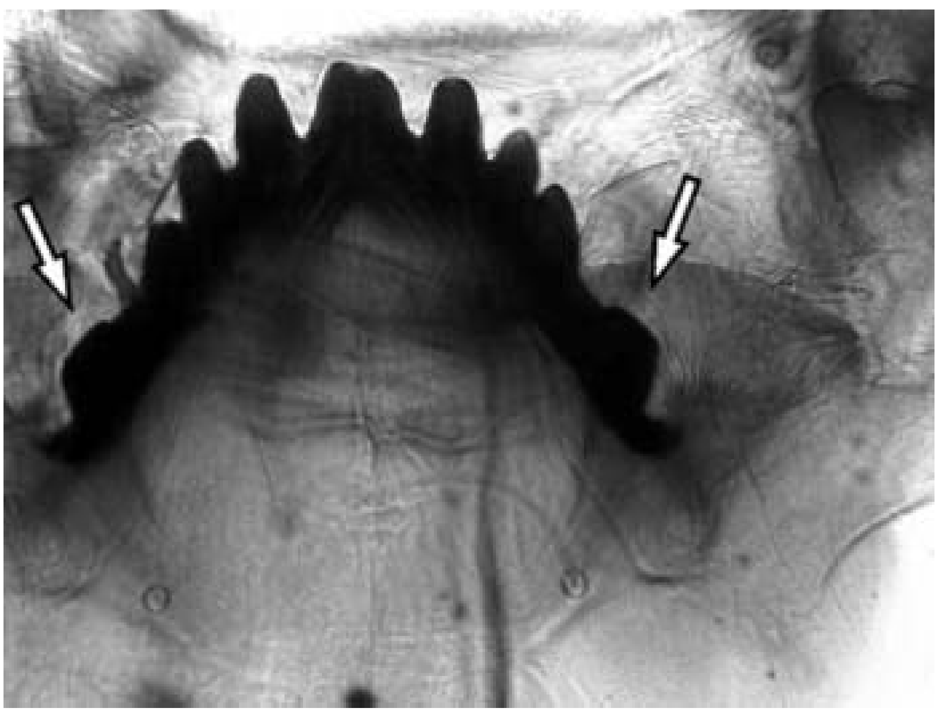

Fig. 8. *Dicrotendipes* sp., mechanically worn (arrows) larval mentum

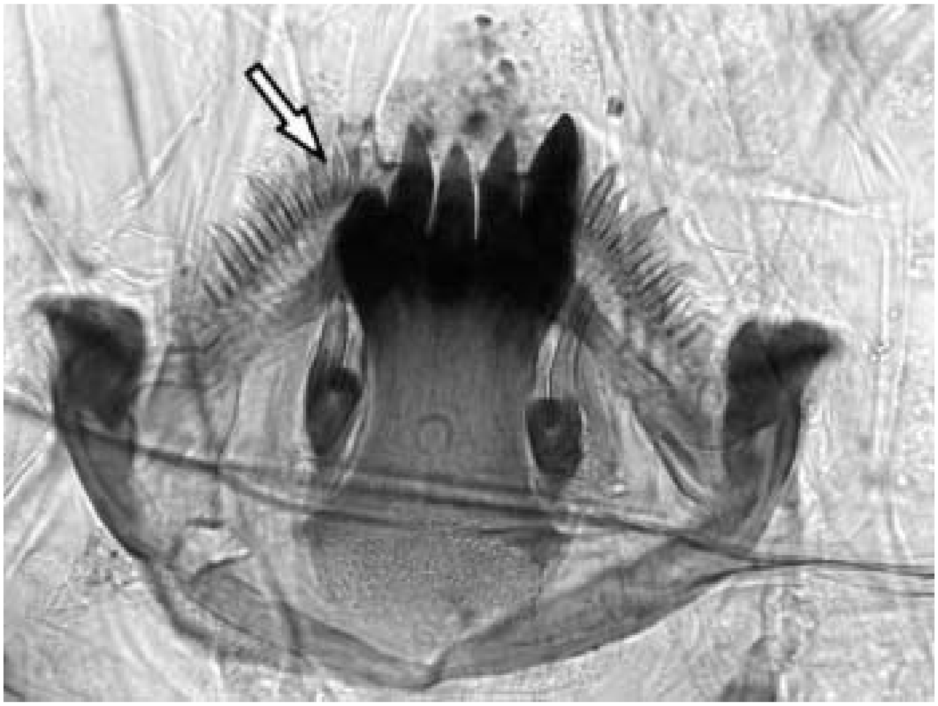

Fig. 9. Tanypodinae Genus A, deformed ligula with stunted outer tooth (arrow)

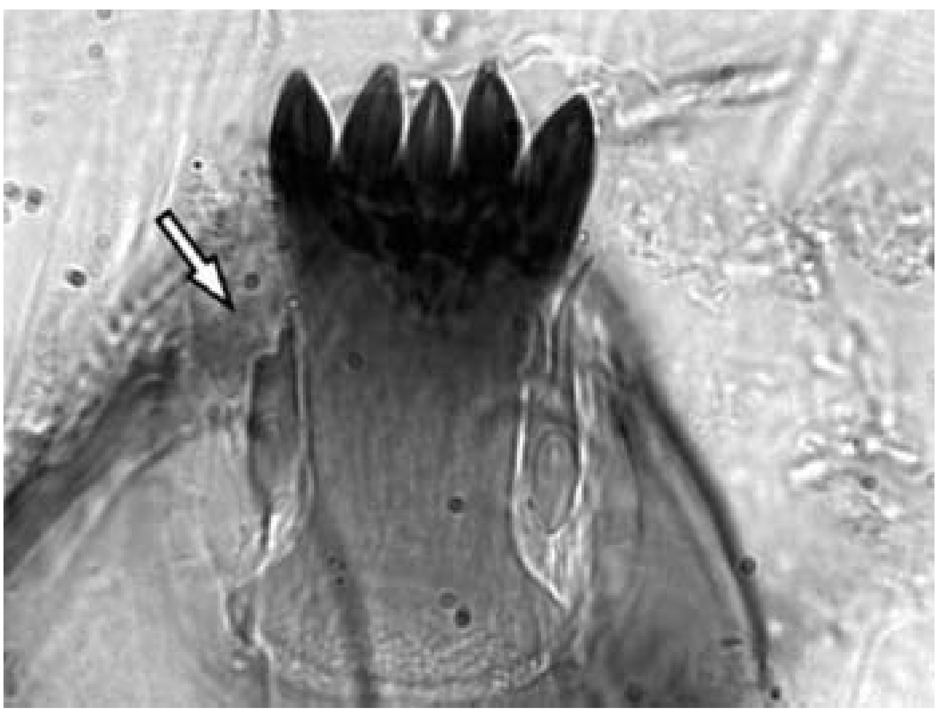

Fig. 10. Tanypodinae Genus A, deformed paraligula with stunted outer tooth (arrow)

Table 1. Relation of normal (norm) to abnormal (abn) development of mouthparts in species and morphospecies of chironomid larvae found in the channels Clarín and side branches of the Ciénaga Grande de Santa Marta.

species/morphospecies	larvae number	pigmentation		abrasion mentum		abrasion mandible		deformities	
	[n]	norm	abn	norm	abn	norm	abn	norm	abn
Clinotanypus sp.	1	1	0	1	0	1	0	1	0
Coelotanypus sp.	2	2	0	2	0	2	0	2	0
Labrundinia sp.	2	2	0	2	0	2	0	2	0
Larsia sp.	37	36	1	25	12	26	11	35	2
Tanypodinae Genus A	24	23	1	16	8	13	11	24	0
Tanypus sp.	3	3	0	2	1	3	0	2	1
Apedilum sp.	1	1	0	1	0	1	0	1	0
Beardius sp.	1	1	0	1	0	1	0	1	0
Chironomus sp. I	12	11	1	2	10	4	8	12	0
Ch. sp. II	25	20	5	6	19	9	16	21	4
Ch. gr. *dorsalis*	11	10	1	4	7	7	4	9	2
Cladopelma sp.	13	13	0	8	5	13	0	13	0
Dicrotendipes sp.	2	1	1	1	1	1	1	2	0
Fissimentum desiccatum	5	4	1	4	1	5	0	5	0
Goeldichironomus carus	58	34	24	2	56	2	56	20	38
G. devineyae	19	14	5	4	15	1	18	17	2
G. sp.	6	5	1	2	4	1	5	5	0
Lauterborniella sp.	5	3	2	4	1	4	1	4	1
Polypedilum sp. I	1	1	0	1	0	1	0	1	0
P. gr. *nubeculosum*	6	6	0	6	0	6	0	6	0
Tanytarsus sp.	4	4	0	4	0	4	0	4	0
total number [n]	238	195	43	98	140	107	131	187	50
percent [%]	100	82	18	41	59	45	55	79	21

Table 2. Percentages of abnormalities in taxa of chironomid larvae found in the channels Clarín and side branches of the Ciénaga Grande de Santa Marta.

species/morphospecies	larvae number	pigmentation	abrasion	deformities
	[n]	[%]	[%]	[%]
Larsia sp.	37	3	32	5
Tanypodinae Genus A	24	4	33	0
Chironomus spp.	48	15	75	12.5
Cladopelma sp.	13	0	0	0
Goeldichironomus carus	58	41	97	67
Goeldichironomus cet.	25	24	74	8
ceteri	33	12	12	6
total number	238	18	61	21

DISCUSSION

Heavy metal concentrations in sediments at the sampling sites were rather within the range described for the middle and lower río Magdalena and the Ciénaga Grande (Perdomo *et al.* 1998, Cadavid & Espinosa 2001, Riss *et al.* 2002). They did not differ considerably from concentrations found in several other tropical and subtropical water bodies: concentrations of Cu, Cd, and Zn were sightly above or below, Pb and Cr slightly below such reference levels; only Hg showed the overall lowest concentration (Abu-Hilal 1987, Wallner-Kersanach 1994, Callisto *et al.* 2000, Rosales-Hoz *et al.* 2000, Toledo *et al.* 2000). However, in comparison, heavy metal contents in sediments at the sampling sites reflect a low to medium degree of pollution due to residual water influence from río Magdalena (Perdomo *et al.* 1998, Marín *et al.* 2000).

Chironomid samples analysed here, although on a rather limited scale, indicate the presence of a rich community associated with the roots of *Typha domingensis* stands. 9 of the 16 genera found belong to the subfamily Chironominae, 7 to the Tanypodinae. The larvae of the first group are considered as sediment- or filter-feeders, some of them scrapers like *Beardius* sp. and *Fissimentum* sp., those of the second group as predators or omnivorous (Armitage *et al.* 1995). Species in the majority of the genera found are considered eurytopic ubiquists and mostly colonizers of soft littoral sediments of standing water bodies (Wiederholm 1983, Epler 2001). Some (*Apedilum*, *Lauterborniella*, *Labrundinia*, and *Dicrotendipes*) may occur more frequently on submerged vegetation. Members of the dominant genera (*Goeldichironomus*, *Chironomus*, *Larsia*) are known to also colonize hypertrophic and sewage ponds (Mazzeo *et al.* 1999, Ebeling 1978) or even animal carcasses (Hicks *et al.* 1997). On the onå hand, the community composition, especially the total absence of the more oxybiont representatives of the subfamily Orthocladiinae, can be attributed to biogeographic reasons, on the other hand to the conditions at the sampling sites which may show pronounced oxygen depletions caused by high bacterial and macrophyte respiration rates.

Whether or not larvae of certain chironomid species are present at any freshwater site is in principle dependent on its microhabitat structure and physico-chemical conditions. Toxic contaminants can influence this strongly. Simultaneously they are responsible for the occurrence of malformations in these larvae. So, the combination of the two parameters – community composition and conditions of the larval mouthparts – can be an important indicator characterizing the ecological state of a study area.

It is assumed that a frequency of deformities in a chironomid community exceeding 8% of total larvae number can be taken as an indicator of some unfavourable impact (Warwick 1988). Thus, taking into account the rather high rate of deformities (21 %) and a community entirely lacking oxygen-demanding species, one may conclude that the ecological state at the investigated sites of the Ciénaga Grande de Santa Marta is rather critical from a general point of view.

However, at first glance, this result does not conform to the relatively low heavy metal concentrations measured directly at the sampling sites. High deformity rates of more than 8 % had been mostly found associated with higher pollutant concentrations (Janssens de Bisthoven *et al.* 1992, Pettigrove 1989, van Urk *et al.* 1992, Warwick, 1990, 1991, 1992). Under conditions of heavy metal impact comparable to our sites the deformity rates were observed not to exceed 3 % (Callisto *et* *al*. 2000). Consequently we assume synergistic or undetected effects on the benthic coenosis in our study, especially since it is known that the particular expression of mouthpart abnormalities is strongly influenced by a variety of environmental stressors, such as pesticides or high deposition rates of fine mineral particles. The latter mentioned factor could be the cause of the conspicuously high rate of mentum abrasion found in the species and morphospecies of *Chironomus* and *Goeldichironomus*. But even organic pollution can be considered as a possible cause of deformities (Servia *et al.* 2000).

However, to obtain a clearer picture of process trends taking place here it is recommendable and necessary to continue this screening investigation. Future investigations on deformities should include larger sample sizes, which could yield more detailed results. Series of quantitative samples from the Ciénagas and the adjacent channels, followed by a qualitative and quantitative registration of deformity types, could provide far more conclusive information about the anthropogenic impact on the major habitats than presented here. Nevertheless, one of the serious problems which still considerably complicate ecological investigations in this biogeographic region is the limited identifiability of the aquatic invertebrate fauna due to the lack of taxonomic knowledge. Most of the aquatic taxa found in the Neotropical region are still not described at the species level (Riss *et al.* 2002), and in Chironomidae the situation is even worse (Spies & Reiss 1996). Nevertheless, deformity rates can be calculated on the basis of reproducible taxonomic units, regardless of their definite systematic position. Thus, in spite of the mentioned restrictions the deformity criteria as well as other physiology-based reactions can be applied as a useful indicator of environmental quality.

ACKNOWLEDGEMENTS

We thank Elisabeth Meyer, Department of Limnology, University of Muenster, and Dirk de Beer, Max-Planck Institute of Marine Microbiology (Bremen) for providing technical equipment. Many thanks to Martin Spies for language improvements and the critical revision of the manuscript. Thanks are due as well to Larisa Frolova and Venera Latypova, Kazan State University, for technical assistance and revision of the manuscript, respectively. This work was supported by Giessen University within the partnership agreement with Kazan State University.

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Recibido: 22/12/2002
Aceptado: 19/01/2004