

DISTRIBUTION AND PREVALENCE OF 15 SPECIES OF EPIBIONT
PERITRICH CILIATES ON THE CRAYFISH *CAMBARELLUS*
PATZCUARENSIS VILLALOBOS, 1943 IN LAKE PÁTZCUARO,
MICHOACÁN, MEXICO

BY

ROSAURA MAYÉN-ESTRADA¹⁾ and MA. ANTONIETA ALADRO-LUBEL
Laboratorio de Protozoología, Departamento de Biología, Facultad de Ciencias, UNAM,
Ap. Postal 70-374, C.P. 04510, Mexico, D.F., Mexico

ABSTRACT

The goal of the present study was to know the distribution and prevalence of ciliated Protozoa attached to the exoskeleton and gills of the crayfish, *Cambarellus patzcuarensis*. Samples of crayfish were taken at two sites in Pátzcuaro Lake, Michoacán, Mexico. Fifteen species of ciliated Protozoa (peritrichs) were found attached as epibionts on the crayfish. Their distribution and prevalence on 14 body parts of the basibiont is presented. Community structures were analysed using Jaccard's index and principal component analysis. The results showed that the gills had no similarity with respect to the other body parts as regards peritrich attachment. The pereopods and the uropods were the body parts with the highest species richness, the gills had the lowest. *Cothurnia variabilis* is the only species with a high degree of specificity to the body region to which it is attached (gills). Of the 15 species found, only one has been reported exclusively as an epibiont on crustaceans.

RESUMEN

El objetivo del presente trabajo fue conocer la distribución y frecuencia de especies de protozoarios ciliados adheridos al exoesqueleto y branquias del acocil *Cambarellus patzcuarensis*. Las muestras de crustáceos fueron recolectadas en dos sitios del lago de Pátzcuaro, Michoacán, México. Se presenta la distribución y frecuencia de 15 especies de protozoarios ciliados (peritricos) en 14 regiones del cuerpo del basibionte. La estructura de las comunidades fue analizada utilizando el índice de similitud de Jaccard y el análisis de componentes principales. Los resultados muestran que las branquias no tuvieron ninguna similitud con el resto de las regiones del cuerpo. Los pereópodos y los urópodos mostraron una alta riqueza específica, y las branquias la menor. *Cothurnia variabilis* fue la única especie con un alto grado de especificidad respecto a la región del cuerpo donde se adhiere (branquias). De las 15 especies de ciliados, solamente una ha sido registrada exclusivamente como epibionte de crustáceos.

¹⁾ Fax: +52.56224828; e-mail: rme@hp.fciencias.unam.mx

INTRODUCTION

Crustaceans serve as hosts to symbiotic, commensal, parasitic, and pathogenic representatives of all major taxa of the Protozoa (Couch, 1983). The exoskeleton of crustaceans provides suitable organic substrates for many species of epibiont ciliates, especially suctorians, chonotrichs, and peritrichs (Fenchel, 1965; Morado & Small, 1995; Fernández-Leborans & Tato-Porto, 2000).

The impact of the populations or communities of ciliated Protozoa on their hosts has been considered both without repercussion (Foster et al., 1978), or in relation with some damage to their hosts, like some impairment of swimming efficiency of the host (Henebry & Ridgeway, 1979; Evans et al., 1979). The epibionts appeared to be involved in mortality of pond-reared shrimps (Overstreet, 1973) or in reducing subitaneous egg production and naupliar and adult survival, and, for heavily infested females, attachment of these ciliates may block the genital pore (Weissman et al., 1993). Lightner (1991) reported that the genera *Zoothamnium*, *Epistylis*, and *Vorticella*, when abundant on the surface of the gills of penaeid shrimp, can cause hypoxia and death.

The decapod, *Cambarellus patzcuarensis* Villalobos, 1943, an inhabitant of the bottom of Lake Pátzcuaro, Michoacán, Mexico, belongs to the family Cambaridae Hobbs, 1942, subfamily Cambarellinae Laguarda, 1961 (cf. Hobbs, 1988). This crayfish constitutes an important link in the ecological webs and it represents a considerable biomass for fish nutrition. Although several species of ciliated Protozoa have been reported attached to the surface of this decapod (Mayén-Estrada & Aladro-Lubel, 1998, 2000, 2001), the studies of ciliate epibionts of crayfish in wild conditions are yet scarce (López-Ochoterena & Ochoa-Gasca, 1971; Matthes & Guhl, 1973; Lahser, 1976). The goal of the present study was to know the distribution and prevalence of 15 species of ciliated Protozoa (peritrichs) attached to the exoskeleton and gills of *Cambarellus patzcuarensis*.

MATERIALS AND METHODS

The crayfish were collected at two sites in Lake Pátzcuaro, Michoacán, Mexico (19°32' to 19°41'N 101°32' to 101°43'W): Espíritu, located on the eastern shore of the lake, and Jarácuaro on the western shore (Mayén-Estrada & Aladro-Lubel, 1998). Fourteen samples were obtained during 1990-1992, and one in 1994, using a 5 mm mesh net. One part of each sample was fixed with 5% formaldehyde for staining procedures, and with 1% glutaraldehyde for standard scanning electron microscopy (Postek et al., 1980); the other part was maintained alive in the laboratory, in aquaria with oxygen supply. We examined 174 decapods, 109 from Espíritu and 65 from Jarácuaro. Fourteen body parts were considered:

rostrum, antennules, antennae, scale, carapace, chela, mouthparts, pereopods, pleopods, abdominal somites, telson, uropods (endopod and exopod), and gills, i.e., where living epibionts were observed using light microscopy. After fixation, staining techniques with Harris' haematoxylin and protargol (Lee et al., 1985) were used to reveal major characteristics of the epibionts. The distribution of each species over the exoskeleton, as well as the prevalence and also the species richness, were recorded for each body region of the hosts. The Jaccard index was used to estimate the degree of similarity between communities of epibionts of Espíritu, Jarácuaro, and Espíritu-Jarácuaro. The similarity values obtained were summarized by clustering, using the UPGMA method (program NTSYSpc v. 1.8). The principal component analysis (PCA) ordination method (Sneath & Sokal, 1973) was employed, taking into account the data of prevalence of ciliate species on each body part of the crayfish, again using NTSYS (pc v. 1.8).

RESULTS

Fifteen species of epibiont peritrich ciliates were identified attached to the exoskeleton of *Cambarellus patzcuarensis*: *Cothurnia variabilis* Kellicott, 1883, *Platycola decumbens* (Ehrenberg, 1830), *Thuricola folliculata* Kent, 1881, *Carchesium polypinum* (Linnaeus, 1758), *Vorticella campanula* Ehrenberg, 1831, *V. communis* Fromentel, 1874, *V. fromenteli* (Fromentel, 1874), *V. infusionum* Dujardin, 1841, *V. latifunda* Nenninger, 1948, *V. microstoma* Ehrenberg, 1830, *V. natans* Fauré-Fremiet, 1924, *V. striata* Dujardin, 1841, *Pseudovorticella quadrata* Foissner, 1979, *Zoothamnium dichotomum* Wright-Kent, 1882, and *Z. simplex* Kent, 1881 (fig. 1, table I). All 15 species are new records for the crustacean host *Cambarellus patzcuarensis*.

At Espíritu, 73% of the crayfish presented at least one species of epibiont, while at Jarácuaro, 78% of the crayfish had epibionts attached. The decapods of the Espíritu site hosted 15 species of ciliates, those of Jarácuaro hosted 13 species (table I).

Concerning the distribution and species richness of ciliates on the body parts of the hosts, the pereopods and uropods had the greatest number of species (13 and 12 species, respectively), and the gills the lowest (one species at both sites) (fig. 1). Nine species were attached to 1-10.7% of the hosts, three species to 11-20% and only one species was found attached to more than 30% of the hosts: *Cothurnia variabilis* attached to 45.8% on the gills of the hosts at Espíritu and 43% at Jarácuaro (table I). Examination of the communities of epibiont ciliates revealed that five species (*Vorticella campanula*, *V. infusionum*, *V. latifunda*, *Pseudovorticella quadrata* and *Zoothamnium simplex*), were attached to 13 body

TABLE I (part I)
Distribution and frequency of epibiont ciliates found on the body parts of *Cambarellus patzcuarensis* Villalobos, 1943

| Species | Body parts | | | | | | | | | | | | | | | | | |
|--|------------|------|-----|-----|-----|-----|-----|-----|-----|-----|------|-----|-----|-----|---|---|---|--|
| | ro | | an | | at | | sc | | ca | | ch | | mp | | | | | |
| | E | J | E | J | E | J | E | J | E | J | E | J | E | J | | | | |
| <i>Cothurnia variabilis</i> Kellcott, 1833 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | |
| <i>Platycola decumbens</i> (Ehrenberg, 1830) | - | - | - | - | - | - | - | - | 0.9 | 4.6 | - | - | - | - | - | - | - | |
| <i>Thuricola folliculata</i> Kent, 1881 | - | - | - | 1.5 | - | - | - | - | 0.9 | 1.5 | - | - | 1.5 | - | - | - | - | |
| <i>Carchesium polypinum</i> (Linnaeus, 1758) | 6.4 | 7.6 | 1.8 | 4.6 | - | - | - | - | - | 3.0 | 0.9 | - | - | - | - | - | - | |
| <i>Vorticella campanula</i> Ehrenberg, 1831 | 9.1 | 3.0 | 8.2 | 4.6 | 6.4 | 1.5 | 3.6 | 1.5 | 7.3 | 4.6 | 8.2 | 4.6 | 4.5 | 4.6 | - | - | - | |
| <i>V. communis</i> Fromentel, 1874 | - | - | 1.8 | 1.5 | - | - | 0.9 | - | 0.9 | 1.5 | 0.9 | - | - | - | - | - | - | |
| <i>V. fromenteli</i> (Fromentel, 1874) | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | |
| <i>V. infusioenum</i> Dujardin, 1841 | 8.2 | - | 3.6 | - | 5.5 | 1.5 | 6.4 | - | 7.3 | - | 10.0 | - | 1.8 | - | - | - | - | |
| <i>V. latifunda</i> Nenninger, 1948 | 3.6 | 1.5 | 1.8 | 1.5 | - | 1.5 | 3.6 | 1.5 | 5.5 | 3.0 | 3.6 | 1.5 | 0.9 | 4.6 | - | - | - | |
| <i>V. microstoma</i> Ehrenberg, 1830 | 1.8 | 4.6 | 2.7 | 1.5 | - | - | 2.7 | 1.5 | 6.4 | 1.5 | 2.7 | 1.5 | 0.9 | 1.5 | - | - | - | |
| <i>V. natans</i> Fauré-Fremiet, 1924 | 1.8 | - | - | - | - | - | 0.9 | - | 0.9 | - | 1.8 | - | - | - | - | - | - | |
| <i>V. striata</i> Dujardin, 1841 | 1.8 | - | 3.6 | 1.5 | 2.7 | 1.5 | 2.7 | - | 1.8 | 1.5 | 1.8 | 3.0 | 1.8 | 3.0 | - | - | - | |
| <i>Pseudovorticella quadrata</i> Foissner, 1979 | 1.8 | - | 0.9 | 6.1 | 0.9 | 4.6 | 1.8 | 3.0 | 1.8 | 6.1 | 2.7 | 4.6 | 1.8 | 1.5 | - | - | - | |
| <i>Zoothamnium dichotomum</i> Wright-Kent, 1882 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | |
| <i>Z. simplex</i> Kent, 1881 | 11.0 | 12.3 | 4.5 | 7.6 | 4.5 | 6.1 | 2.7 | 9.2 | 2.7 | 6.1 | 2.7 | 6.1 | 1.8 | 3.0 | - | - | - | |

TABLE I (part 2)

| Species | Body parts | | | | | | | | | | | | | | | | | | | |
|--|------------|------|-----|-----|-----|------|------|-----|-----|------|-----|-----|------|-----|-----|-----|---|---|------|------|
| | pe | | | pl | | | as | | | te | | | uen | | | uex | | | gi | |
| | E | J | | E | J | | E | J | | E | J | | E | J | | E | J | | E | J |
| <i>Cothurnia variabilis</i> Kellicott, 1833 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 45.8 | 43.0 |
| <i>Platycola decumbens</i> (Ehrenberg, 1830) | - | 4.6 | - | - | - | - | 3.0 | - | - | 1.5 | 0.9 | 1.5 | 0.9 | 1.5 | 0.9 | - | - | - | - | - |
| <i>Thuricola folliculata</i> Kent, 1881 | - | - | - | - | - | - | 1.5 | - | - | - | - | - | - | - | - | - | - | - | - | - |
| <i>Carchesium polypinum</i> (Linnaeus, 1758) | 0.9 | 1.5 | - | 1.5 | 2.7 | 10.7 | 10.7 | - | 1.5 | 0.9 | 0.9 | - | 0.9 | 0.9 | 3.0 | - | - | - | - | - |
| <i>Vorticella campanula</i> Ehrenberg, 1831 | 11.9 | 6.1 | 2.7 | 4.6 | 6.4 | 3.0 | 3.0 | 8.2 | 4.6 | 7.3 | 6.1 | 6.1 | 7.3 | 6.1 | 8.2 | 7.6 | - | - | - | - |
| <i>V. communis</i> Fromentel, 1874 | 1.8 | 1.5 | - | - | 0.9 | - | - | 1.8 | - | 3.6 | 1.5 | 2.7 | 3.6 | 1.5 | 3.0 | - | - | - | - | - |
| <i>V. fromenteli</i> (Fromentel, 1874) | 0.9 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - |
| <i>V. infusionum</i> Dujardin, 1841 | 11.0 | - | 2.7 | - | 4.5 | 1.5 | 1.5 | 7.3 | 3.0 | 10.0 | 1.5 | 9.1 | 10.0 | 1.5 | 9.1 | - | - | - | - | - |
| <i>V. latifunda</i> Neminger, 1948 | 9.1 | 1.5 | 2.7 | - | 0.9 | 1.5 | 1.5 | 2.7 | 4.6 | 3.6 | 6.1 | 4.5 | 3.6 | 6.1 | 7.6 | - | - | - | - | - |
| <i>V. microstoma</i> Ehrenberg, 1830 | 9.1 | 10.7 | - | 1.5 | 1.8 | 1.5 | 1.5 | 3.6 | - | 4.5 | 6.1 | 5.5 | 4.5 | 6.1 | 6.1 | - | - | - | - | - |
| <i>V. natans</i> Fauré-Fremiet, 1924 | 0.9 | 1.5 | - | - | - | - | - | - | - | 2.7 | - | 1.8 | 2.7 | - | - | - | - | - | - | - |
| <i>V. striata</i> Dujardin, 1841 | 4.5 | 1.5 | - | - | 1.8 | 1.5 | 1.5 | 3.6 | 3.0 | 2.7 | 1.5 | 4.5 | 2.7 | 1.5 | 1.5 | - | - | - | - | - |
| <i>Pseudovorticella quadrata</i> Foissner, 1979 | 5.5 | 6.1 | 2.7 | 1.5 | 1.8 | 4.6 | 4.6 | 1.8 | 4.6 | 5.5 | 4.6 | 6.4 | 5.5 | 4.6 | 6.1 | - | - | - | - | - |
| <i>Zoothamnium dichotomum</i> Wright-Kent, 1882 | 0.9 | - | - | - | - | - | - | - | - | 0.9 | - | 0.9 | 0.9 | - | - | - | - | - | - | - |
| <i>Z. simplex</i> Kent, 1881 | 5.5 | 4.6 | 2.7 | 3.0 | 0.9 | 9.2 | 9.2 | 3.6 | 1.5 | 6.4 | 6.1 | 1.8 | 6.4 | 6.1 | 4.6 | - | - | - | - | - |

E = Espirito; J = Jarácuaro; ro = rostrum; an = antennules; at = antennae; sc = scale; ca = carapace; ch = chela; mp = mouth parts; pe = pereopods; pl = pleopods; as = abdominal somites; te = telson; uen = uropods (endopod); uex = uropods (exopod); gi = gills.

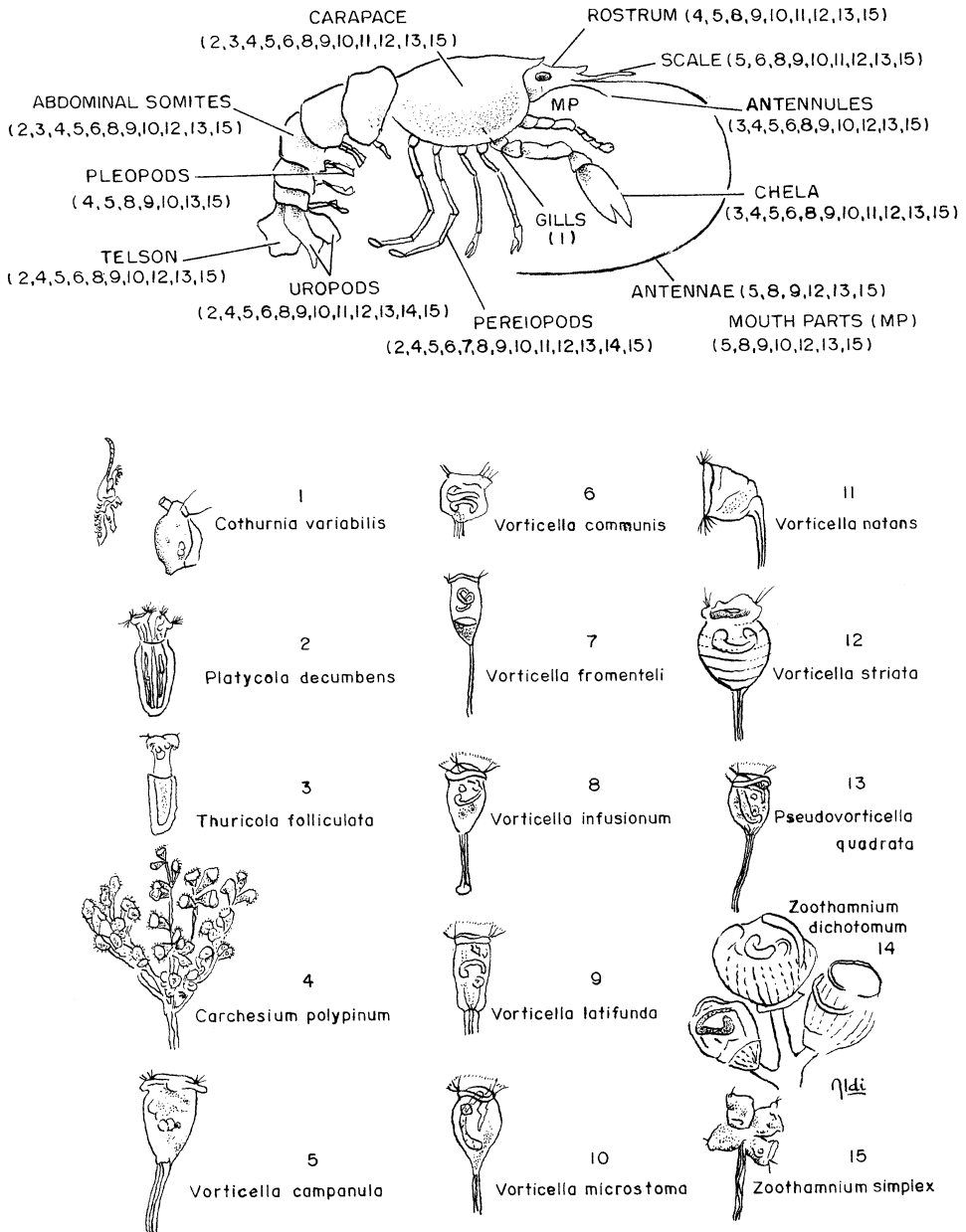


Fig. 1. Schematic representation of the distribution of the 15 epibiont ciliates attached to the body parts of *Cambarellus patzcuarensis* Villalobos, 1943. The numbers in the parentheses correspond to the species of ciliates.

parts, two species (*Vorticella microstoma* and *V. striata*) were found on 12 body parts, and only *Vorticella fromenteli* and *C. variabilis* were found attached to only a single body part (pereopods and gills, respectively) (table I, fig. 1). The values obtained with Jaccard's index for comparison between the assemblages of species at both sites, was 0.86. The data resulting from comparisons with this index, i.e., between communities of peritrichs attached to the various body parts of the crayfish, are shown in fig. 2. For Espíritu (fig. 2A), the antennules-abdominal somites and uropods exopod-endopod clustered at the maximum value of similarity (1.0). At Jarácuaro (fig. 2B), maximum similarity was obtained between the antennules-carapace (0.9), followed by chela-mouth parts (0.8). For Espíritu-Jarácuaro (fig. 2C), all body regions (except the gills) had a similarity value of 0.5. The maximum values of similarity (1.0) were for antennules-abdominal somites and uropods endopod-exopod of Espíritu (ane-ase and uene-uexe) followed by rostrum-chela of Espíritu (roe-che), and antennules-carapace of Jarácuaro (anj-caj) (0.9). In the three cases, the gills had no similarity with the rest of the body regions. The results of PCA, taking into account the prevalence of the 15 peritrich ciliates attached to the 14 body parts of the crayfish, are shown in fig. 3. For Espíritu (fig. 3A), component 1 separates the body regions into two groups, with the gills situated on the far right side of the diagram; to the left, the pereopods and uropods are located. The other body parts, except the rostrum, are grouped according to the two components. For Jarácuaro (fig. 3B), in relation to component 1, the gills are placed on the right side, followed by the group of pleopods, rostrum and scale; the pereopod and uropod-exopod, are placed on the left side. All the other body parts are grouped according to the two components. For Espíritu-Jarácuaro pooled data, component 1 separates the body regions into two groups (fig. 3C): the gills of Espíritu-Jarácuaro (gie and gij) on the right side; and the pereopods, uropods (endopod and exopod) from Espíritu (pee, uene, uexe) on the left side; component 2 separates the antennules, carapace, and abdominal somites of Jarácuaro (anj, caj and asj) in the lower part, and the rest of the body parts are grouped according to both components, 1 and 2.

DISCUSSION

The epibiont ciliates were found attached to the various body parts of *Cambarellus patzcuarensis*, the crayfish considered in this study, and occupied any region of the exoskeleton without preference (except *Cothurnia variabilis*). This included setae of the appendages, which demonstrates that the species have a low degree of specificity as regards recognition of the body parts, and the exoskeleton is apparently suitable for their attachment. The data obtained from PCA ordination support

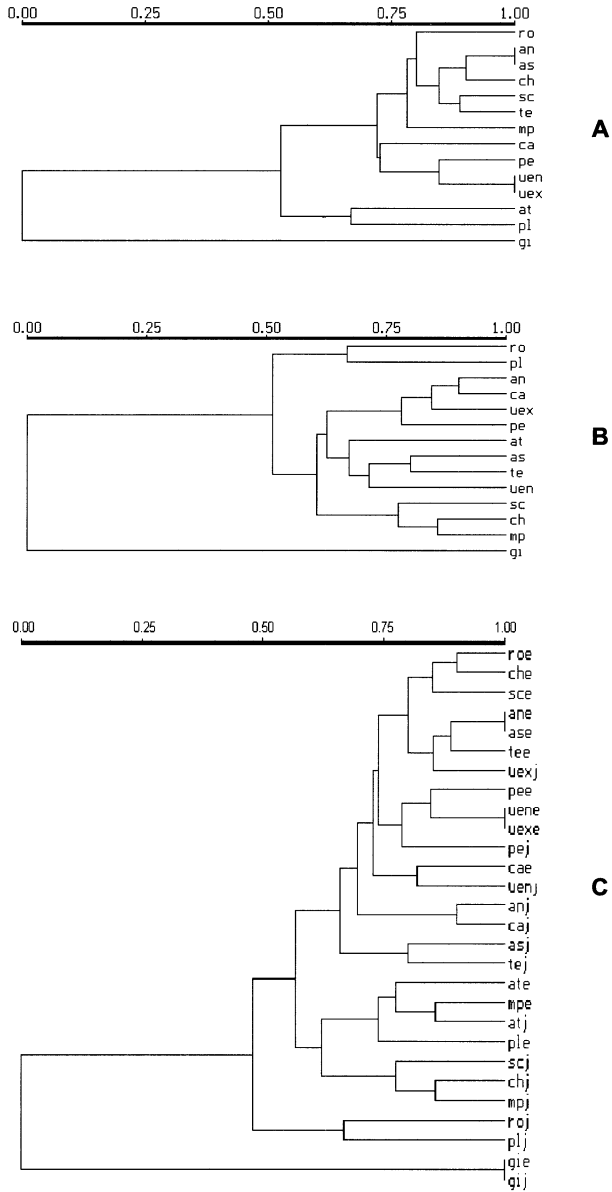


Fig. 2. Phenogram of the cluster analysis (UPGMA) of Jaccard's index, showing the similarities between body parts of *Cambarellus patzcuarensis* Villalobos, 1943, according to the peritrich ciliate species attached. A, Espiritu; B, Jarácuaro; C, Espiritu-Jarácuaro.

this idea (fig. 3). Henebry & Ridgeway (1979), working with epibionts of copepods and cladocerans, reported that, since peritrichs are primarily bacterial feeders, there would seem to be no great advantage to their being located in particular areas on their hosts, where they would experience no damage from the organism's swim-

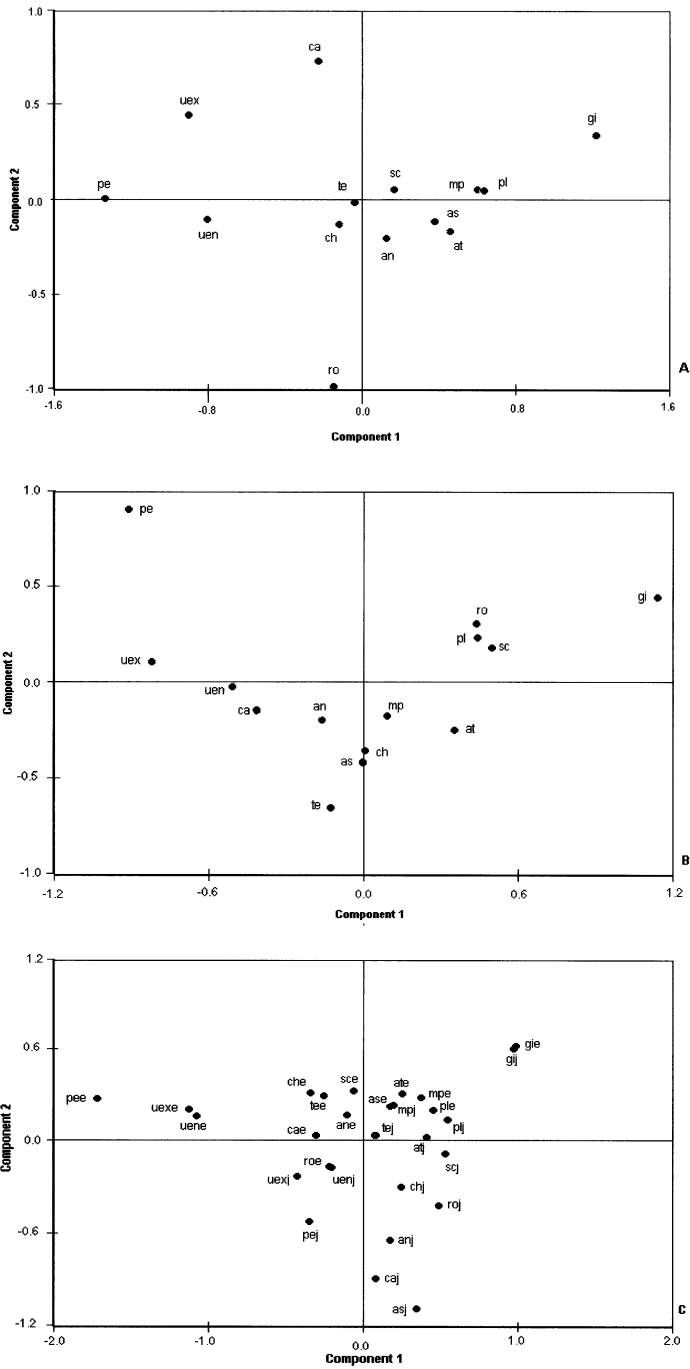


Fig. 3. Ordination plot from Principal Component Analysis. The diagrams show the positions on components 1 and 2 of the body parts of *Cambarellus patzcuarensis* Villalobos, 1943, from: A, Espíritu (61.4% cumulative); B, Jarácuaro (51.7% cumulative); and, C, Espíritu-Jarácuaro pooled (53.5% cumulative).

ming and feeding activities. This is consistent with the present results, where five species were found attached to 13 body parts, eight species attached to 3-12 body parts, and only two species (*Vorticella fromenteli* and *Cothurnia variabilis*) exclusively attached to pereopods and gills, respectively. In the case of *V. fromenteli*, its prevalence was low (0.9%). The species with the highest prevalence (43-45.8%) was *Cothurnia variabilis*, which settled on the gills. This can be explained, because the branchial chamber is protected by an overlying carapace, and the gill bailer provides a constant flow of water across the gill surface. The high species richness on pereopods (13 species) and uropods (12 species) might be explained on the basis of the shape of these body parts, and also by the presence of projections such as setae, which create many microhabitats providing shelter, food, and oxygen for the epibionts, in the same manner as do seagrasses (Aladro-Lubel & Martínez-Murillo, 1999). This is consistent with the values of community similarity obtained for these appendages, which ranges from 0.78 to 1.0 (fig. 2C).

Three species have not previously been reported as epibionts: *Vorticella fromenteli*, *V. natans* and *Pseudovorticella quadrata* (cf. Foissner et al., 1992; Warren, 1986). Five species have been observed attached to several organic substrates other than crustaceans (algae, aquatic phanerogams, leeches, insects): *Thuricola folliculata*, *Vorticella communis*, *V. infusionum*, *V. latifunda*, and *Zoothamnium dichotomum* (cf. Wailes, 1943; Nenninger, 1948; Stiller, 1971; González, 1979; Trueba, 1980; Foissner et al., 1992; Martínez-Murillo & Aladro-Lubel, 1994). Seven species have been reported as epibionts of crustaceans before: *Cothurnia variabilis*, *Platycola decumbens*, *Carchesium polypinum*, *Vorticella campanula*, *V. microstoma*, *V. striata*, and *Zoothamnium simplex* (Morado & Small, 1995; Fernández-Leborans & Tato-Porto, 2000). From these, *Cothurnia variabilis*, has been reported to be attached exclusively to crustaceans (Warren & Paynter, 1991). It has been documented as a peritrich with a high degree of specificity in relation to the body part to which it attaches (i.e., gills) (Nenninger, 1948; Stiller, 1971; Hamilton, 1952), which is confirmed by the present results.

The role of crustaceans as basibionts of ciliates includes providing a proper substrate for the attachment, areas sheltered from predators, and the host's free movement creates a flow of water that brings oxygen and food for the ciliates, and also acts as a dispersal mechanism to new habitats. The assemblage of 15 species of ciliated Protozoa in Lake Pátzcuaro (15 at Espíritu and 13 at Jarácuaro), indicates that the exoskeleton was suitable for their attachment, fully utilizing the available surface. This is consistent with the average values of the Jaccard index obtained at a level of overall comparison (between the two communities: Espíritu vs. Jarácuaro), which shows a high similarity (0.86).

The present findings allow us to consider these ciliated Protozoa as mere epibionts, with an ecological rather than symbiotic relationship with their hosts.

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