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First report of folliculinid ciliates affecting Caribbean scleractinian corals

Received: 20 January 2005 / Accepted: 24 October 2005 / Published online: 17 March 2006
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Abstract This is the first report of a ciliate of the genus *Halofolliculina* infecting hard coral species of six families (Acroporidae, Agaricidae, Astrocoeniidae, Faviidae, Meandrinidae and Poritidae) and milleporids in the Caribbean. Surveys conducted during 2004–2005 in Venezuela, Panama and México confirmed that this ciliate affects up to 25 scleractinian species. The prevalence of this ciliate at the coral community level was variable across sites, being most commonly found at Los Roques, Venezuela, and at Bocas del Toro, Panama (prevalence 0.2–2.5%), but rarely observed in the Mexican Caribbean. Ciliates were more prevalent within populations of acroporids (*Acropora palmata*, *Acropora cervicornis* and *Acropora prolifera*) in Los Roques. Recent observations also corroborate the presence of these ciliates in Curacao and Puerto Rico. Our observations indicate that ciliates affecting corals have a wider distribution than previously thought, and are no longer exclusively found in the Indo-Pacific and Red Sea.

Keywords Ciliates · Caribbean corals · Emergent syndromes · Coral diseases

Communicated by Biological Editor K. Sullivan Sealey

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Introduction

Coral diseases have emerged among the most important factors contributing to coral reef decline world wide (Harvell et al. 1999; Rosenberg and Ben-Haim 2002). The more rapid spread of marine pathogens compared to terrestrial parasites, the introduction of microorganism and invasion of exotic species, the global warming and the increasing frequency of human disturbances upon coral reefs have been highlighted as critical factors in enhancing the virulence and rate of transmission of pathogens and compromising the mechanisms of host resistance (Ruiz et al. 2000; Harvell et al. 2002; McCallum et al. 2003; Torchin et al. 2003; Mullen et al. 2004).

During the past few decades reports of new coral diseases and syndromes have increased, especially in the Caribbean region where 66% of all syndromes/diseases have been observed (Green and Bruckner 2000; Weil 2004). While most coral pathogens identified to date are microparasites (i.e., cyanobacteria, bacteria or fungi), only two out of the 22 described coral syndromes (Sutherland et al. 2004), have been associated with the presence of the macroparasites. The skeletal eroding band (SEB) and the brown band disease (BrB) have been associated with the ciliates *Halofolliculina corallasia* (Antonius 1999; Willis et al. 2004) and *Helicostoma nonatum* (Borneman 2001), respectively. These ciliates have been considered exclusive to the Indo-Pacific and Red Sea corals (Winkler et al. 2004), even though *H. corallasia* has been actively looked for in the Caribbean (Antonius and Lipscomb 2001).

This is the first report of a protozoan ciliate of the genus *Halofolliculina* affecting 25 species of Caribbean scleractinian corals of six families (Acroporidae, Agaricidae, Astrocoeniidae, Faviidae, Meandrinidae and Poritidae). Our observations included oceanic, barrier and coastal fringing reefs off the coasts of Venezuela, Panama, Mexico, Curacao and Puerto Rico.

Results and discussions

The ciliate was first observed in three colonies of *Eusmilia fastigiata* (Fig. 1a) during a CARICOMP survey (April 2004) in Sombrero Key, Morrocoy National Park, Venezuela (10°52'N–69°16'W). We observed small black dots adjacent to the border between healthy and recently dead tissues, and after examination under a dissecting microscope we confirmed the presence of these ciliates. One month later we conducted a survey in Dos Mosquises Sur key at Los Roques National Park (Venezuela, 11°48'03"N–66°53'30"W) on a barrier and a coastal fringing reef. Coral colonies in either quadrats (1 m²) or belt-transects (20×2 m) were checked for the presence of the ciliate, which was found infecting species of *Acropora palmata*, *Acropora cervicornis*, *Acropora prolifera*, *Colpophyllia natans*, *Montastraea annularis*, *Montastraea faveolata* (Fig. 1b), *Montastraea franksi*, *Madracis mirabilis* and *Diploria labyrinthiformis* (Table 1).

In August 2004, the ciliate was observed in the northern Mexican Caribbean only in *A. palmata* and with a low prevalence (5 out of 5,087 colonies). These observations were made during a survey of eight back-reef sites along a 20 km reef tract in the 'Parque Nacional Arrecife de Puerto Morelos'. By May 2005, the ciliate was observed with a higher frequency in colonies of *A. palmata*, *Dichocoenia stokesi*, *M. annularis* and *C. natans* in one of the back-reef sites of Puerto Morelos. In May 2005 the ciliate was observed in other reefs of the northern Mexican Caribbean (Akumal), on two fore-reef sites (depth: 7 and 13 m), in a few large colonies of *M. faveolata* and *M. annularis* often affected by yellow band disease. In the summer of 2005 ciliates were also observed in the southern Mexican Caribbean at 10–15 m deep sites; but only in few reefs.

In October 2004, the ciliate was observed on a single colony of *C. natans* in Bocas del Toro (BDT), Panama, after a two-hour dive whilst actively searching for signs of infection. The discovery of *Halofolliculina* sp. in BDT prompted us to conduct more extensive surveys at this site. In March 2005, we surveyed corals for diseases at six permanent annual monitoring sites established by the Environmental Science Program (ESP) in 1997 in Cayo Roldan (09°13'11"N, 82°19'31"W), Isla Colon (09°20'58"N, 82°15'48"W), Cayo Agua (09°08'01"N, 82°02'25"W), Isla Solarte (09°18'29"N, 82°12'26"W), Isla Cristobal (09°15'21"N, 82°14'06"W) and Isla Bastimento (09°16'03"N, 82°07'11"W). We checked for the presence of the ciliate on all coral colonies within three replicate 20 m² permanent belt transects (10×2 m) at each of five depth ranges (18–16 m, 15–13 m, 12–10 m, 4–3 m, and < 3 m). For branching species such as *Porites furcata* we counted each physically independent unit as a colony.

Overall prevalence of the ciliate at BDT was 1.6% (371 out of 23,869 colonies), and it was commonly observed in five out of the six monitoring sites (Roldan,

Agua, Colon, Cristobal and Solarte), with prevalence ranging from 0.2% (Agua) to 2.5% (Roldan). At each site most colonies affected by ciliates occurred between 3 and 4 m (52–82%), and only 0.86–8.33% occurred in the deepest intervals (13–18 m). However, in Cayo Agua, 60% of infections occurred at 10–12 m and the other 40% at 13–15 m. Of the 371 colonies infected with ciliates at all sites, 31.3±29.4% (±SD) were *P. furcata* (Fig. 1c), 28.7±30.7% (±SD) were *Agaricia tenuifolia* (Fig. 1d) and 20.6±25.6% (±SD) were *Agaricia agaricites*. We also found the ciliate on thirteen other coral species, but at a lower prevalence.

In BDT we observed for the first time *Halofolliculina* sp. infections in *Porites furcata*, *P. astreoides*, *A. agaricites*, *A. tenuifolia*, *Agaricia fragilis*, *Agaricia lamarcki*, *Siderastrea siderea*, *Scolimia cubensis*, *Leptoseris cuculata*, *Madracis decactis*, *Stephanochoenia intersepta* and the milleporid *Millepora alcicornis* (Table 1). Recent observations (summer and fall of 2005) in Curacao and Puerto Rico corroborate the presence of these ciliates in *A. palmata* (Fig. 1e), *A. cervicornis* (Fig. 1f), *Montastraea cavernosa*, *D. labyrinthiformis* (Fig. 1g) and *E. fastigiata* in these localities.

In all affected species, the infection appeared as a dark band flanked with healthy tissue on one side and skeleton recently devoid of tissue on the other (Fig. 1b–f). On closer inspection the band had a dark spotted appearance due to the presence of individual ciliates (Fig. 1b, g). Both *H. corallasia* and the Caribbean form are sessile in a lorica with the cell body attached at its pointed posterior end (Fig. 1j), showing two conspicuous peristomial wings bearing feeding cilia at the anterior end (Fig. 1h, i) (Antonius 1999; Antonius and Lipscomb 2001).

We initially proposed these ciliates might have recently invaded from the Indo-Pacific region (Cróquer et al. in press). However, closer examination and comparison of the body structure of these ciliates with *H. corallasia*; indicate that both the Caribbean and Indo-Pacific ciliates are indeed different species (D. Lipscomb et al. unpublished data). It is possible that this ciliate has been unnoticed to date because it has a superficial resemblance to black-band disease when found at high densities (Fig. 1b, g). It is also possible that ciliates might have been in lower prevalence and are now becoming more ubiquitous because of the increasing frequency and severity of stress factors (e.g increasing sea temperatures, bleaching, eutrophication, diseases, etc.) that have affected corals across the Caribbean region lately (Hughes 1994; Hughes et al. 2003; Santavy et al. 2005).

Our results suggest that ciliates may constitute a new coral syndrome that adds to the 22 plus diseases reported for the Caribbean (Green and Bruckner 2000; Weil 2004; Sutherland et al. 2004), and might represent an additional potential threat to the prominent reef building corals of the Caribbean. Protozoans are common and wide-spread pathogens in both terrestrial and marine ecosystems (Harvell et al. 1999;

Fig. 1 General aspect of ciliate bands with different densities on **a** *E. fastigiata*, **b** *M. faveolata*, **c** *P. furcata*, **d** *A. tenuifolia*, **e** *A. palmata*, and **f** *A. cervicornis*. **g** Highly-dense and thick ciliate band resembling black band disease on *D. labyrinthiformis*. **h** Detail of ciliates clustering on *Porites astreoides*: note large peristomial wings out of the lorica. **i** Morphology of the ciliate showing peristomial wings bearing feeding cilia, and **j** Posterior body attached to the lorica

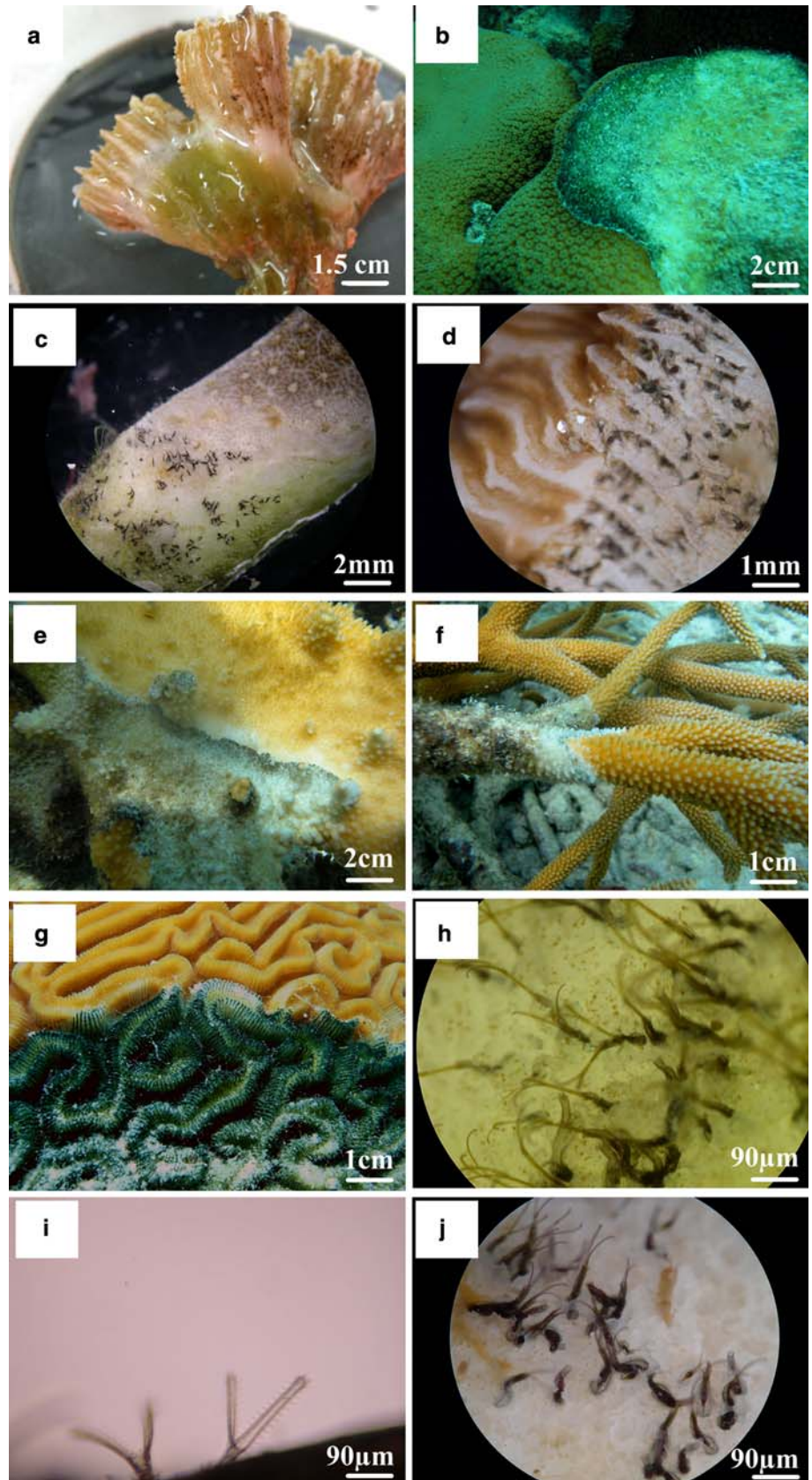


Table 1 Total number of coral and hydrocoral species affected by the ciliate *Halofolliculina* sp. at each locality and sites surveyed during 2004–2005

Species	Localities						
	Venezuela		Panama	Mexico		Curacao	Puerto Rico
	Los Roques	Morrococoy	Bocas del Toro	Akumal	Puero Morelos	Sea Aquarium	La Parguera
<i>A. cervicornis</i>	x		x	x	x		x
<i>A. palmata</i>	x				x	x	
<i>A. prolifera</i>	x						
<i>A. agaricites</i>			x		x		
<i>A. fragilis</i>			x				
<i>A. tenuifolia</i>			x				
<i>A. lamarcki</i>			x				x
<i>C. natans</i>	x	x	x				
<i>Dichochoenia stockesi</i>					x		
<i>E. fastigiata</i>	x	x					x
<i>L. cuculata</i>				x			
<i>M. alvicornis</i>			x				
<i>M. decactis</i>			x				
<i>M. mirabilis</i>	x						
<i>M. annularis</i>	x			x	x		
<i>M. faveolata</i>	x			x	x	x	x
<i>M. franksi</i>	x		x				
<i>M. cavernosa</i>						x	
<i>Diploria strigosa</i>				x			
<i>D. labyrinthiformis</i>	x						x
<i>P. furcata</i>			x				
<i>P. astreoides</i>			x				
<i>S. siderea</i>			x				
<i>S. cubensis</i>			x				
<i>S. intersepta</i>			x				
Total	10	2	14	5	6	3	5

Gómez-Gutierrez et al. 2002), and this is the first report of a protozoan that may cause tissue mortality on scleractinian corals in the Caribbean Region; where coral diseases have been traditionally linked to bacteria, fungi and other microorganisms (Ramos-Flores 1983; Smith et al. 1998; Denner et al. 2003).

Alternatively, these ciliates might simply be opportunistic saprophytes or secondary parasites feeding on dead tissues as they are abundant between dead or necrotic tissues and recently exposed skeletons (Fig. 1a–g). All ciliates found in Venezuela, Mexico and Panama had zooxanthellae within their bodies (Fig. 1h), suggesting that they are ingesting or somehow acquiring zooxanthellae. Using Scanning Electron Microscopy (SEM), we have already confirmed the presence of zooxanthellae within ciliate vacuoles (D. Lipscomb et al. unpublished data).

In the Red Sea, *H. corallasia* produces rates of tissue mortality ranging from millimeters to a few centimeters per day, but the mechanisms of transmission of this ciliate remain unclear (Winkler et al. 2004). In the Caribbean, we still do not know the mechanism of transmission of these ciliates nor the rate of tissue mortality they produce on different species of corals. It is possible that transmission occurs directly from the water column or by direct contact between infected and susceptible colonies, and susceptibility may depend on the presence of previous damage/injury on the colonies. We are conducting experiments to test these hypotheses.

Whether they are primarily pathogens or just opportunist saprophytes, our observations indicated that ciliates affecting corals have a wider distribution than previously thought, and are no longer exclusive for the Indo-Pacific and Red Sea.

Acknowledgements We would like to thank E Weil, L.M. Márquez, G.W. Smith and D. Lute for their comments and suggestions on this manuscript, as well as D. Bone for providing us with laboratory facilities. To Marine Environmental Science Program of STRI and R. Collin, for providing us the logistic support at Bocas del Toro and to Carlos Guevara and Arcadio Castillo for their valuable help during disease surveys in Panama.

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