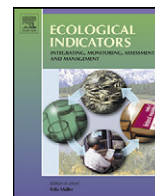




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## Influence of small-scale gold mining on French Guiana streams: Are diatom assemblages valid disturbance sensors?

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## ABSTRACT

The ongoing gold-rush in French Guiana could cause severe disturbance to ecosystems. Although illegal, small gold mining sites are rapidly expanding. Few studies have attempted to measure the consequences of the increased gold mining on the biota of small forest streams, and to date no study has dealt with primary producers. Here we measured the response of diatom assemblages to gold mining in ten sites differently affected by the mining activity (i.e., reference, formerly gold-mined and currently exploited). Our results showed that both taxonomic and functional structure of the diatom assemblages were influenced by the intensity of gold mining activity. A significant relationship between soil erosion and diatom motility ability has been demonstrated. These findings show that diatom assemblages are sensitive to gold mining disturbance and suggest that diatom communities may be used as sensors of the environmental stress caused by small-scale gold mining activities.

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### 1. Introduction

Tropical ecosystems are threatened by human activities (Cincotta et al., 2000; Lorion and Kennedy, 2009; Portillo-Quitero and Sánchez-Azofeifa, 2010), which are causing a profound disturbance of the forests and loss of biodiversity (Lawton et al., 1998). French Guiana, which has 90% of rainforest coverage and includes tropical wilderness area covering the upper Amazonia and the Guiana Shield, is no exception (Mittermeier, 1988). In French Guiana the increase in the anthropogenic pressure originates mainly from gold mining.

During the last decade, gold mining activities, including illegal mining sites, have increased exponentially, potentially leading to considerable environmental damage (Hammond et al., 2007). The extraction of gold deposits consists of washing the soil with high pressure water jets, leading to major environmental impacts, such as deforestation, soil erosion and heavy metal pollution (Cleary,

1990). The gold mining sites are located along rivers that collect all the effluents overloaded with suspended matter and toxicants, such as mercury, which is used to recover gold from the sediment (Watts et al., 2003). The mercury released into the environment impacts aquatic ecosystems, including the primary producers (Anson Moyer et al., 2002; Kelly et al., 2007), macroinvertebrates (Henny et al., 2005; Žižek et al., 2007) and fish (Barbosa et al., 2003; Sampaio da Silva et al., 2009). Little attention has been paid to other potential impacts including soil erosion, which is more visible and is probably the most pervasive and devastating consequence in the short term (Hammond et al., 2007). Experimental evidence indicates that the turbidity generated by fine sediment can affect stream productivity (Parkhill and Gulliver, 2002; Izagirre et al., 2009) and species interactions (Utne-Palm, 2002; Pekcan-Hekim and Lappalainen, 2006). In fact, siltation and the subsequent biological impairment are among the most prevalent problems in streams and rivers throughout the world (Pimentel and Kounang, 1998).

In French Guiana, the National Forestry Office estimated that 1333 km of watercourses were directly affected by gold mining, as well as 12,000 ha of forest (CIRAD-ONF, 2006; Mansillon et al., 2009). The number of illegal mining sites in French Guiana is unknown, but estimated between 500 and 900 sites. However, there is little information on the overall impact of gold mining activities (Mol and Ouboter, 2004; Mendiola, 2008; Yule et al., 2010), especially for the most abundant and widespread type of

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gold extraction in the Guiana shield, namely illegal small-scale gold mines (Hammond et al., 2007). As the rivers of the Guiana shield are characterised by some of the lowest levels of natural suspended solids in the world (Hammond et al., 2007), the potential impact of increased turbidity and siltation due to mining could therefore be profound.

Our purpose was to investigate how small mining sites affect biota in small forest streams of French Guiana. Streams with similar hydromorphological features, but that were affected by various degrees of gold mining intensity (reference, formerly gold-mined and currently exploited) were selected for the study. We focussed on diatoms that colonise almost all aquatic ecosystems (Round et al., 1990) and that are recognized as efficient bioindicators (Prygiel and Whitton, 1999; Stormer and Smol, 1999). The impact of gold mining on both the taxonomic and the functional structure of diatom assemblages was tested. In addition, we hypothesized that motile diatoms are able to tolerate the high load of suspended solids generated by gold mining, whereas non-motile life-forms may have little chance of survival under high load of sediment. This aimed to assess whether the diatoms could be valuable sensors to monitor streams impacted by small-scale gold mining activities.

## 2. Methods

### 2.1. Study area

The study area was located in the Approuague Basin (French Guiana). Nine sampling sites were located in the Nouragues Nature Reserve, in small tributaries of the Arataï river (tributary of the Approuague), and one site outside and downstream of the reserve, in a tributary of the Approuague river (Fig. 1; Table S1). These streams were low order streams of similar size (less than 5 m wide) flowing in a primary forest environment (also termed old-growth forest, see Hilbert and Wiensczyk, 2007).

To assess the effect of gold mining activities on diatom communities, we identified and selected sites with three different levels of gold mining intensity: (i) reference sites (sites R1–R4) which had never been exploited; (ii) formerly exploited sites (F1–F3) where gold mining activity had been recorded during the year before sampling, but had not been exploited for at least 6 months; and (iii) currently exploited streams (C1–C3), which were under exploitation during field sampling. In order to obtain an immediate assessment of the physical impact on the streams, we measured the turbidity with a WTW field turbidimeter. Vigouroux et al. (2005) showed that turbidity is an accurate proxy of overall gold mining intensity (heavy metals, decrease of light intensity, increase of siltation). In addition to an increase of turbidity, gold mining also causes an increase of siltation, due to the deposition of fine mineral material on the river beds (EPA, 2000; Vigouroux et al., 2005).

The sampling sites were located close to the confluence with the main river (Approuague or Arataï) and hundreds of meters downstream from the areas of illegal mining. The sites therefore correspond to the catchment outlet and drain all the effluents produced by the mining activity.

### 2.2. Sampling and treatment procedures

The samples used for diatom analysis were obtained from periphytic material collected in November, during the dry season. Diatoms can be found on most submerged surfaces but the recommended substrate/habitat combination to sample diatoms are stones obtained from a riffle with a flowing current (Kelly et al., 1998; AFNOR, 2000). Owing to the difficulty of finding the same substrate in all the sampling sites, we systematically investigated two different substrates: stones (S) if available, wood (W) and/or

submerged leaves (L). In accordance with Townsend and Gell (2005), the genera composition did not differ between substrates. The samples were coded with the station code (R, F, C), site number (1–4) and substratum (S, W or L).

The sampling protocol consisted of brushing material from hard and stable substrates into a clean container and then storing the material in a flask. Samples were then fixed with buffered formaldehyde. At the laboratory, samples were cleaned in boiling hydrogen peroxide and hydrochloric acid. After removing all traces of hydrochloric acid, the diatoms were dried onto cover slips and fixed on microscope slides using a high-refractive index resin (Naphrax<sup>®</sup>, R.I. = 1.7) to obtain permanent slides.

### 2.3. Taxonomical and ecological data

We identified and counted the relative abundance of the diatom taxa. Counting (400 valves) was carried out using an Olympus BX51 differential interference contrast microscope (DIC) with 1000× magnification under oil immersion. The identification of the diatom flora was done to the genus level according to Round et al. (1990), Krammer and Lange-Bertalot (1991–1997) and Metzeltin and Lange-Bertalot (1998, 2007). A more accurate identification (i.e., species level) was not feasible due to the current limited knowledge of the diatom flora of French Guiana.

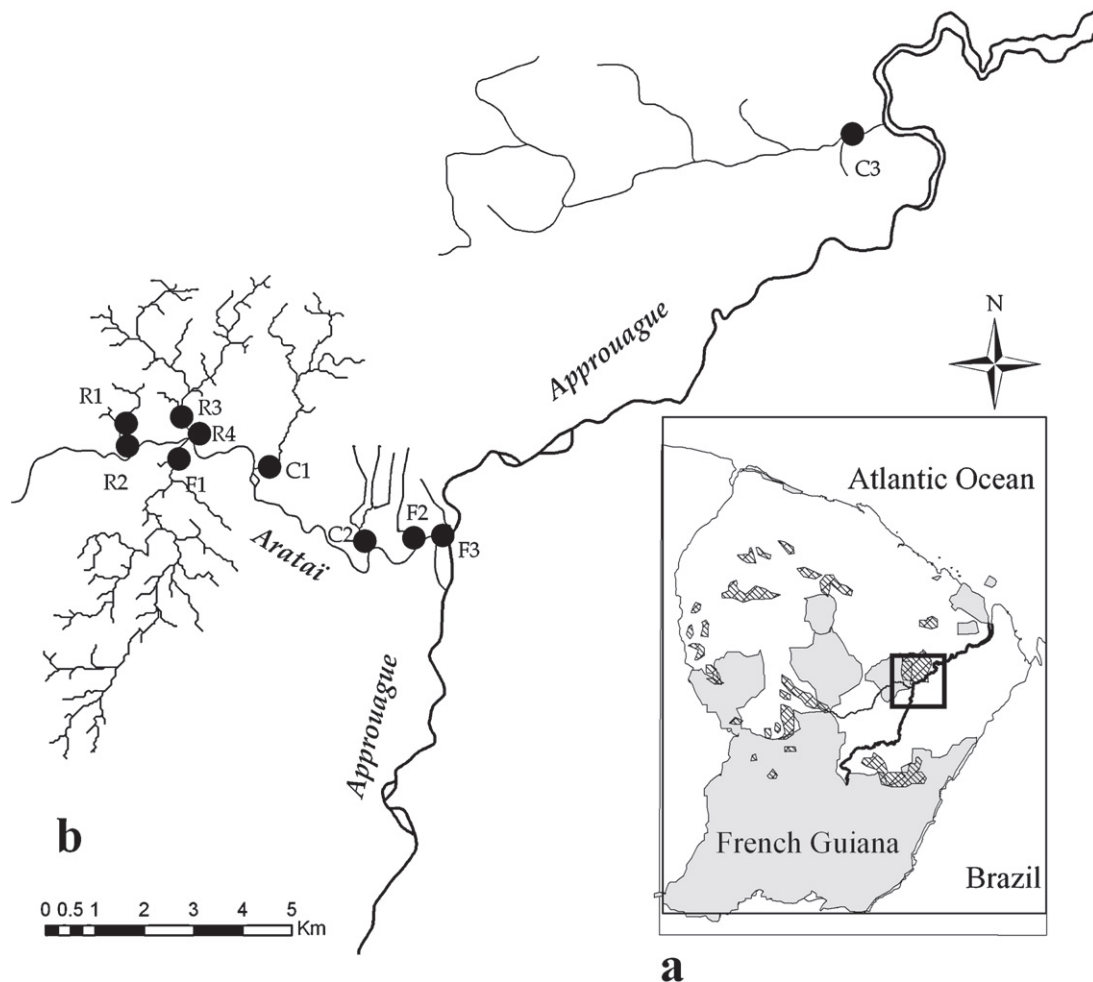
The genus level was nevertheless sufficient to sort genera according to motility. Indeed, the taxonomical dichotomies leading to the identification of the genus level are based on the cell symmetry and the presence or not of a raphe system and its course. These generic features determine the diatom life-forms and throw light on their motility. Many studies have concluded that the raphe system accounts for diatom motion (Edgar, 1982; Bertrand, 1990, 1992, 2008; Round et al., 1990), and Coste (in Cemagref, 1982) and Hill et al. (2001) demonstrated that the genus level is sufficient to determine the motility ability of diatoms. A distinction has thus been made between three life-forms of periphytic diatoms mainly according to Denys (1994) and completed by Round et al. (1990): (i) epontic life-form referring to taxa that are not motile and firmly attached to any kind of substrate; (ii) benthic life-form that are motile and live on the substrate; and (iii) euplanktonic life forms that are not motile and live in the water column.

### 2.4. Data analysis

The data were analyzed using R statistical software (R Core Team Development, 2008). To determine taxonomical similarities between the samples and to promote site typology, sampling sites have been classified via a hierarchical cluster analysis using Ward's linkage method with Bray and Curtis distance measure. The Mean Split Silhouette (MSS) criterion (Pollard and van der Laan, 2002) and the Multiple Response Permutation Procedure (MRPP) (Mielke and Berry, 1976) were used to validate the clustering relevance. The MSS value determines the optimal level of the classification tree where the clusters are the most homogenous and the MRPP analysis tests if the differences between the clusters are significant.

An indicator genus for each group of site typology was identified according to Dufrêne and Legendre (1997) Indicator Value (IndVal). Taxonomical units having the highest IndVal score were used to discriminate the clusters. The statistical significance of the indicator values was evaluated using a randomization procedure with 500 permutations. Processing was performed using the function 'duleg' of the package 'labdsv' (Dufrêne and Legendre, 1997).

To assess the effects of environmental characteristics on variation in diatom composition, we carried out a non-parametric MANOVA (McArdle and Anderson, 2001), an analysis of variance using distance matrices performed by the function 'adonis' of the package 'vegan' (Oksanen et al., 2008). This function partitions



**Fig. 1.** Location of sampling sites. (a) Map of French Guiana indicating the location of the study area (black square). The biodiversity reserves (grey) and the gold mining areas (hatched) are also indicated. Note that some mining areas are located inside the biodiversity reserves. (b) Detailed map of the study area indicating the sampling sites: R1–R4: reference sites; F1–F3: formerly exploited sites; C1–C3: currently exploited sites.

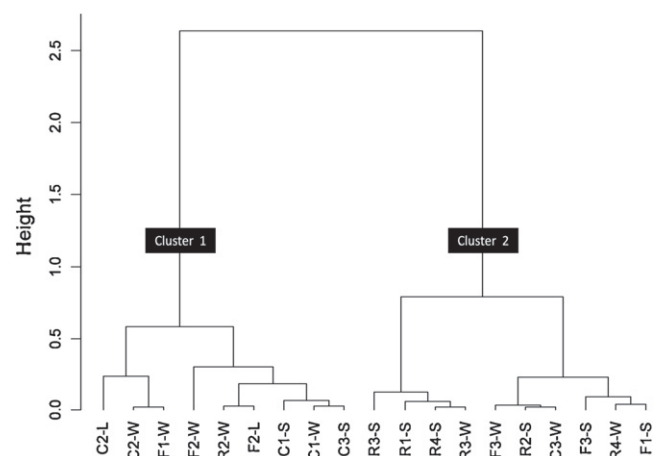
sums of squares using metric or semimetric distance matrices. The significance of the test was given by *F*-tests based on sequential sums of squares from 1000 permutations of the raw data.

### 3. Results

A total of 43 genera were identified. The number of genera identified at each site ranged from 11 to 27 (Table 1). The following genera had greater than 20% abundance in at least one sample: *Achnanthis* Kützing, *Aulacoseira* Thwaites, *Encyonema* Kützing, *Eunotia* Ehrenberg, *Fragilaria* Lyngbye, *Gomphosphenia* Lange-Bertalot, *Navicula* Bory de Saint Vincent, *Nupela* Vyverman & Compère, *Nitzschia* Hassall and *Surirella* Turpin. The average composition of the diatom communities was of 55% epontic life-forms and 44% benthic life-forms, whereas the remaining 1% represented eu planktonic forms. Differences in diatom taxonomic and life-form composition between samples were significantly explained by stream status (R, F or C) (non-parametric MANOVA,  $P < 0.002$  and  $P = 0.01$ , respectively, Table 2), but not clearly by substratum ( $P = 0.58$  and  $P = 0.027$ , respectively).

The percentage of benthic forms was greatest in currently exploited sites ( $70.0\% \pm 13.83$  SD), whereas the reference and formerly exploited sites had values below 50%. Furthermore, the relative abundance of the benthic forms was lower in the reference sites ( $27.1\% \pm 13.11$  SD) than in the formerly exploited sites ( $31.3\% \pm 12.3$  SD).

The hierarchical clustering based on life-forms distinguished two clusters (Fig. 2), whereas clustering based on taxonomy discriminated five distinct subsets (Fig. 3). These typologies, based on taxonomical and biological traits clearly separated the sites

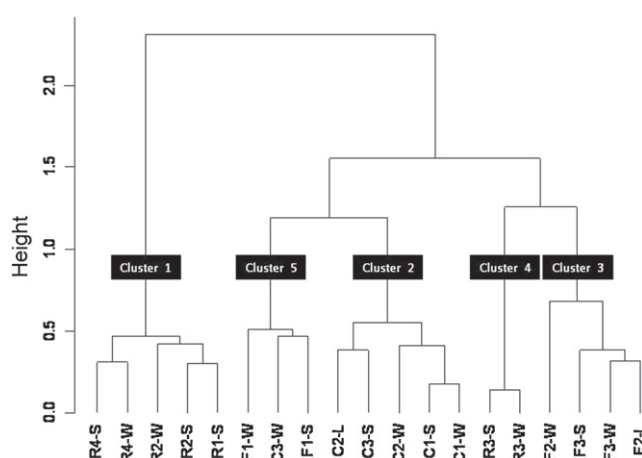


**Fig. 2.** Hierarchical clustering (Euclidean distance; Ward algorithm) of the sampling sites based on diatom life-forms. R1–R4: reference sites; F1–F3: formerly exploited sites; C1–C3: currently exploited sites. The substrata are indicated after the site labels: W: wood; S: stone; L: leaves.

**Table 1**

Inventory at the genus level of the diatoms and their life-form. Sample site codes: R=reference; F=formerly exploited; C=currently exploited; 1–4 correspond to sites.

GENERA	Life form	R1	R2	R3	R4	F1	F2	F3	C1	C2	C3
<i>Adlafia</i> Moser, Lange-Bertalot & Metzeltin	Benthic		X			X					
<i>Amphora</i> Ehrenberg	Benthic						X				
<i>Brachysira</i> Kutzing	Benthic				X		X	X	X	X	X
<i>Caloneis</i> Cleve	Benthic				X						
<i>Capartogramma</i> Kufferath	Benthic		X			X	X				
<i>Chamaepinnularia</i> Lange-Bertalot & Krammer	Benthic							X	X	X	
<i>Craticula</i> Grunow	Benthic			X						X	
<i>Cymbopleura</i> Krammer	Benthic						X				
<i>Diademsis</i> Kutzing	Benthic	X	X		X		X	X			
<i>Diploneis</i> Ehrenberg	Benthic				X	X	X	X			
<i>Eolimna</i> Lange-Bertalot & Schiller	Benthic		X		X		X			X	
<i>Faiiacia</i> Stickle & Mann	Benthic		X		X		X				
<i>Frustulia</i> Rabenhorst	Benthic	X		X	X	X	X	X	X	X	X
<i>Geissleria</i> Lange-Bertalot & Metzeltin	Benthic				X						
<i>Germainiella</i> Lange-Bertalot & Metzeltin	Benthic		X								
<i>Gyrosigma</i> Hassall	Benthic					X		X	X		X
<i>Hippodonta</i> Lange-Bertalot, Metzeltin & Witkowski	Benthic		X			X					
<i>Kobayasiella</i> Lange-Bertalot	Benthic								X		
<i>Luticola</i> Mann	Benthic		X	X		X	X	X		X	
<i>Naviculadicta</i> Lange-Bertalot	Benthic		X		X	X			X		
<i>Navicula</i> Bory de Saint Vincent	Benthic	X	X	X	X	X	X	X	X	X	X
<i>Neidium</i> Pfitzer	Benthic									X	
<i>Nitzschia</i> Hassall	Benthic	X	X	X	X	X	X	X	X	X	X
<i>Orthoseira</i> Thwaites	Benthic		X					X			
<i>Pinnularia</i> Ehrenberg	Benthic	X	X		X	X	X	X		X	X
<i>Placoneis</i> Mereschkowsky	Benthic		X								
<i>Stauroneis</i> Ehrenberg	Benthic	X	X	X			X		X		
<i>Stenopterobia</i> Brebisson	Benthic			X		X	X	X	X	X	
<i>Surirella</i> Turpin	Benthic			X	X	X	X	X	X	X	X
<i>Staurosira</i> Williams & Round	Epontic		X				X				
<i>Achnanthes</i> Bory de Saint Vincent	Epontic		X	X		X		X			
<i>Achnantheidium</i> Kutzing	Epontic	X	X	X	X		X	X	X	X	X
<i>Cocconeis</i> Ehrenberg	Epontic		X		X	X					
<i>Encyonema</i> Kutzing	Epontic	X	X	X		X	X		X	X	X
<i>Eunotia</i> Ehrenberg	Epontic	X	X	X	X	X	X	X	X	X	X
<i>Fragilaria</i> Lyngbye	Epontic		X	X	X		X	X	X	X	X
<i>Gomphonema</i> Ehrenberg	Epontic	X	X	X	X	X	X	X	X	X	X
<i>Gomphosphenia</i> Lange-Bertalot	Epontic		X		X						
<i>Nupela</i> Vyvermann & Compere	Epontic	X	X		X	X	X	X			
<i>Terpsinoe</i> Ehrenberg	Benthic		X								
<i>Aulacoseira</i> Thwaites	Euplanktonic						X		X		
Centrophyceidae undetermined	Euplanktonic							X			
<i>Cydotella</i> Kutzing	Euplanktonic						X				
Number of genera		11	27	14	20	19	26	20	17	17	12



**Fig. 3.** Hierarchical clustering (Euclidean distance; Ward algorithm) of the sampling sites based on diatom genera. R1–R4: reference sites; F1–F3: formerly exploited sites; C1–C3: currently exploited sites. The substrata are indicated after the site labels: W: wood; S: stone; L: leaves.

according to their level of impairment. Reference sites (two clusters), former (two clusters) and current sites (one cluster) were clearly separated according to taxonomical typology, whereas formerly exploited sites were mixed in the life-form typology.

Species indicator values (IndVal) were calculated for each diatom genus. Among them, ten had a significant score ( $P < 0.05$ ) and were significant representatives of each set of samples defined by the taxonomical typology (Table 3). Concerning the two clusters of reference sites (cluster 1 and 4), indicator taxa were the *Nupela*, *Caloneis* Cleve and *Achnantheidium* genera respectively. In the case of the formerly exploited sites (clusters 3 and 5) the main representative genera were *Eunotia*, *Pinnularia* Ehrenberg, *Encyonema* and *Hippodonta* Lange-Bertalot, Metzeltin & Witkowski. *Surirella* pooled with *Navicula* and *Brachysira* Kutzing was clearly characteristic of currently exploited sites (cluster 2).

In the reference sites, the diatom assemblages were dominated by non-motile diatoms (epontic taxa; i.e., *Achnantheidium*, *Nupela*, *Eunotia*, *Gomphonema* Ehrenberg, *Gomphosphenia* genera). Conversely, the motile benthic life-forms were dominant in the currently exploited sites. The diatom communities of formerly exploited sites combined benthic and epontic forms, although benthic diatoms remained dominant. Among these communities, we identified the *Achnantheidium*, *Eunotia*, *Encyonema*,

**Table 2**

Non-parametric MANOVA assessing the effects of substratum and gold mining (stream status: reference; formerly exploited; currently exploited) on variations in taxonomic and life-form similarity between sites. The significance of the tests was checked using *F*-tests based on sequential sums of squares from 1000 permutations of the raw data.

	MS	<i>F</i>	<i>R</i> <sup>2</sup>	<i>P</i>	
<b>Taxonomy</b>					
Substratum	0.161	0.858	0.078	0.580	<i>ns</i>
Status	0.596	3.178	0.288	0.002	**
Residuals	0.188		0.634		
<b>Life-form</b>					
Substratum	0.153	4.743	0.260	0.027	*
Status	0.210	6.483	0.356	0.010	**
Residuals	0.032		0.384		

*ns*: non-significant.

\* *P* < 0.05.

\*\* *P* < 0.01.

*Frustulia* Rabenhorst, *Gomphonema*, *Navicula* and *Nitzschia* genera.

#### 4. Discussion

The current knowledge of the overall impact of gold mining activities on the aquatic fauna is limited and, to our knowledge, our study is the first reporting the effect of gold mining on diatom flora. Almost all studies in neotropical environments have been on fish assemblages (Mol and Ouboter, 2004; Mendiola, 2008). Of these studies, only Mol and Ouboter (2004) have dealt with the impacts of small-scale gold mines in the neotropics. The results of these fish studies are consistent with our findings on diatoms, as both studies demonstrated that gold mining does not induce a sudden decline in genera richness, but profoundly affects assemblage composition (Mol and Ouboter, 2004; Mendiola, 2008). Such a consistent response across trophic levels (i.e., diatoms and fish) suggests that gold mining may affect the entire food web (Yule et al., 2010). Indeed, gold mining has been reported to induce a decline in the taxonomic and functional diversity of fish (Tarras-Walshberg et al., 2001), macroinvertebrates (Milner and Piorowski, 2004), and primary producers (Quinn et al., 1992).

Although the mining activities we considered here were small-scale operations involving only a few workers and no heavy equipment (the miners cross the forest on foot carrying their equipment), the resulting impact was sufficient to exceed the resistance threshold of the stream. As a direct consequence, we observed that current, former, and reference sites host distinct diatom assemblages. We thus confirmed that stream recovery was still incomplete several months after the cessation of gold mining activities. Our results are consistent with those of Yule et al. (2010) on an Indonesian river, where the extent of the recovery 10 months after the mining ceased depended on the duration

**Table 3**

Significant indicator genera (IndVal, *P* < 0.05) for each cluster.

Indicator genus	Cluster	IndVal	Probability
<i>Nupela</i>	1	0.900	0.002
<i>Caloneis</i>	1	0.600	0.034
<i>Surirella</i>	2	0.779	0.002
<i>Brachysira</i>	2	0.547	0.026
<i>Navicula</i>	2	0.403	0.036
<i>Eunotia</i>		0.620	0.002
<i>Pinnularia</i>	3	0.567	0.022
<i>Achnanthydium</i>	4	0.692	0.004
<i>Encyonema</i>	5	0.608	0.028
<i>Hippodonta</i>	5	0.515	0.042

of the mining activities and on the severity of the perturbation.

Gold mining generates two main disturbances due to soil erosion: increase of the load of suspended solids (i.e., water turbidity) and heavy metal release (Hammond et al., 2007; Yule et al., 2010). In order to disentangle the impact of toxicant and of suspended solid load, we paid particular attention to diatom deformity. It is known that under long-term and/or high exposure to heavy metals, diatom communities present abnormal forms (Ruggiu et al., 1998; Falasco et al., 2009; da Silva et al., 2009; Duong et al., 2010). As no diatom deformities were recorded, we concluded that the changes of the diatom flora mainly responded to suspended solids load rather than heavy metals load.

Diatom assemblages are recognized to be relevant indicators of sediment stability and sediment hydrodynamics (Sylvestre et al., 2004; Méléder et al., 2007). Consequently, the distribution of diatom genera according to the degree of impairment was strongly influenced by their differential motility ability which determines their ability to deal with substratum stability and is closely linked to siltation dynamics. This gradient is limited at one end by non-motile attached diatoms and at the other end by extremely motile diatoms, such as *Surirella*, able to adapt to very unstable substrates. The literature presents details of variable motility performance observed among benthic forms. For instance, the *Gyrosigma* Hassall, *Nitzschia*, and *Surirella* genera with high motility (Round et al., 1990; Bertrand, 1992) reflect significant siltation or sediment instability, whereas *Navicula* and *Frustulia* indicate moderate siltation and stable sediment (Sylvestre et al., 2004). The *Encyonema* and *Eunotia* genera with moderate mobility would testify for transitional conditions indicative of recovery dynamics.

These findings suggest that diatoms may be of great value in measuring the impact of current and past gold mining activities on streams. In the rivers of Montana USA, Bahls (1993) used a siltation index based on the proportion of motile species adapted to hold their position in unstable substrates. This index only takes into account the percentage of the dominant motile *Navicula* and *Nitzschia* genera. Our findings, combined with those of Bahls (1993), suggest that the proportion of benthic versus epontic diatoms might be a useful tool to assess the degree of siltation. An increase in benthic life-forms in any samples therefore means an increase in siltation, and as a consequence, a downgrading of biogenous water quality. Periphytic diatom communities thus provide evidence of the various degrees of impairment due to gold mining activity and in many ways could testify to resilience (Harrison, 1979) and to the recovery process (Neubert and Caswell, 1997). They are therefore good candidates for a “diatom tool” to assess and monitor the degree and the dynamics of disturbance on neotropical stream ecosystems subjected to soil erosion caused by gold mining activities.

#### 5. Conclusions

Our results showed that the use of the diatom genera was efficient to assess the effect of gold mining activity, acting both on diatom assemblages and functional traits. Diatom motility ability appeared to be of prime importance in order to determine soil erosion intensity due to gold mining. We concluded that the relative abundance of the motile benthic life-forms vs. non-motile epontic forms would be a significant indicator to quantify the degree of environmental impairment.

In addition, taking into account the motility ability of significant indicative benthic genera would be a promising approach to assess the temporal dynamics of siltation. Considering life-form distribution among assemblages, and motility ability of dominant benthic genera, make it possible to estimate quantitative effect and

recovery vs. impairment dynamics respectively. Consequently, the diatom flora has proved to be a valuable sensor to monitor rivers of French Guiana subject to past or present small-scale gold mining activity.

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### Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.ecolind.2011.07.018.

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