MARINE POLLUTION BY HYDROCARBONS AND ITS IMPACT ON MICROFAUNA: THE CASE OF BENTHIC FORAMINIFERA IN THE CONGOLESE ATLANTIC BASIN

ABSTRACT

This work traces the negative impact that microfauna suffers during the discharge of petroleum products in the Congolese Atlantic basin. It shows how much pollution in the marine environment poses a threat to the ecosystem. This study is based on sediments taken from several boreholes. The research work undertaken has highlighted the pollution of marine ecosystems by hydrocarbons. Geochemical analysis of the sediments shows heavy metal concentrations higher than natural standards. Micropaleontological analysis shows that benthic foraminifera receive and absorb most of the pollutants released into the marine environment. This leads to disturbances in the metabolic system, species malformations, and a numerical and qualitative reduction in biodiversity that is a hindrance to interpretation. Indeed, these results show that 29.16% of polluted benthic foraminifera are unidentifiable. Spread from the infralittoral environment to the bathyal environment, the families Ammoniidae, Bolivinidae, Buliminidae, Cassidulinidae, Cibicididae, Eponididae, Nonionidae, Rhabdamminidae, Textulariidae and Uvigerinidae are the most affected by this pollution. Thus,the seabed is exposed to all types of anthropogenic degradation, in particular to the full spectrum of toxic pollutants. Oil pollution of marine waters poses a real threat to the health of marine ecosystems.

Keywords: pollution, benthic foraminifera, hydrocarbons, Congolese Atlantic basin

I. INTRODUCTION

The Congolese Atlantic basin is full of a high concentration of organic matter, which justifies the presence of a rich and very varied biodiversity. Its ecosystem, thanks to the good geographical and climatic conditions, is favourable to the development of marine fauna and flora. The Congolese Atlantic basin also has significant hydrocarbon resources that have been the subject of intense oil exploitation since the 1960s. These various oil activities are at the origin of the hydrocarbon discharges recorded on the ocean floor. These oil discharges constitute marine pollution that disturbs marine organisms and their ecological environment. This negative impact on the marine environment prompts an assessment of marine ecosystems. In the context of this study, this assessment is made using foraminifera.

Benthic foraminifera, marine microorganisms, are a category of species with precise ecological requirements that depend on all the variables of the marine environment (salinity, temperature, depth, pH, oxygen content, concentration of organic matter). These environmental variables define their distribution in the marine environment. Benthic foraminifera have been present in all marine environments since the early Cambrian (Culver, 1991; Sen Gupta, 1999). They provide a great deal of information about geological time. They are widelyused in oceanography as paleoclimatic and paleoenvironmental bio-indicators (Debenay, 1990; Collins *et al.*, 1995 ; Culver and Buzas, 1985; Hayward *et al.*, 1996; Blais-Stevens and Patterson, 1998; Ernst *et al.*, 2002) because of their sensitivity to environmental changes and their great capacity for fossilization. Benthic foraminifera are also used for the ecological quality of polluted or unpolluted ecosystems (Debenay et *al.*, 2000; Scott et *al.*, 2001; Hallock et *al.*, 2003; Burone et *al.*, 2006).

Recent micropaleontological studies carried out on sediments of Cretaceous and Miocene ages have shown evidence of pollution on microfossils (benthic foraminifera) by hydrocarbons. The present work consists of assessing the impact of marine pollution caused by oil. It provides, for the first time, precise information on the modifications and deformations undergone by benthic foraminifera under the effect of this pollution.

II. GEOGRAPHICAL AND GEOLOGICAL CONTEXT

II.1. Geographical context

The Congolese Atlantic basin is located in the Gulf of Guinea, on the passive margin of the West African coast. It is located between the meridians 11° and 13° East and the parallels 4° and 5° South, then extends from the southwest of Gabon to the northeast of Cabinda over 250 km. Its seafront oriented SE-NW is largely open to the south. In its western part, the Congolese coastal basin extends over about 108 km on the Atlantic. The area of our study extends from the subtidal part to the bathyal environment (Figure 1).

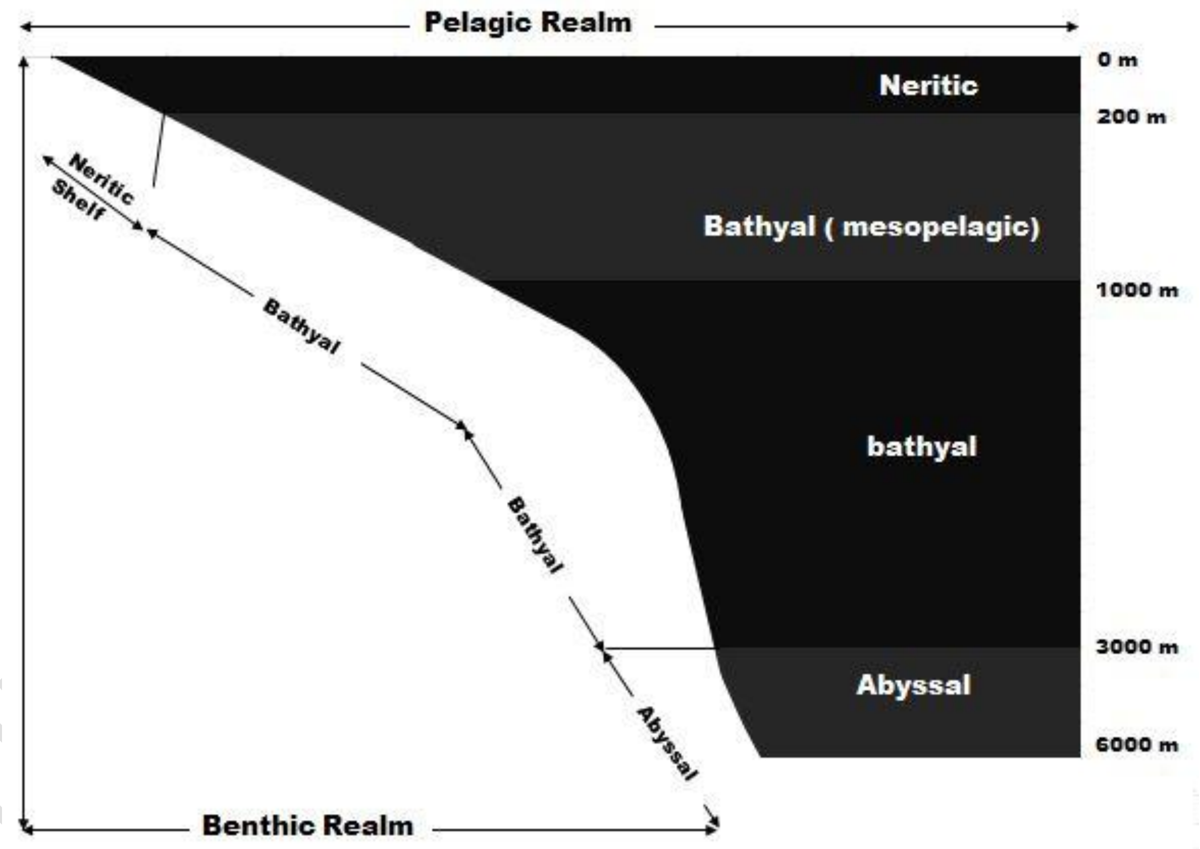

Figure 1 : marginal-coastal area (Kobawila*et al*., 2021)

II.2. Geological context

The history of the Congolese Atlantic basin is linked to the Cretaceous opening of the South Atlantic between Africa and South America. A salt-bearing episode testifies to this opening and makes it possible to separate two major periods of sedimentation: a fluvio-lacustrine period corresponding to the ante-salt-bearing period and a marine period corresponding to the post-salt-flowering period. The fluvio-lacustrine period from the Neocomian to the Aptian marked the first filling of the collapse ditches (Micholet, 1970). The Aptian recorded a general collapse which was at the origin of the first marine incursions and the deposition of a powerful salt series (Reyre, 1984; Guiraud *et al.*, 1992). In addition, the post-salt period presents two large superimposed units: an aggradation unit of Aptian-Eocene age, and a progradation unit of Miocene to present age. These two large units are separated by major erosion of Oligocene age (Seranne *et al.*, 1992). Our study area is located in the Atlantic basin south of the city of Pointe-Noire (Figure 2).

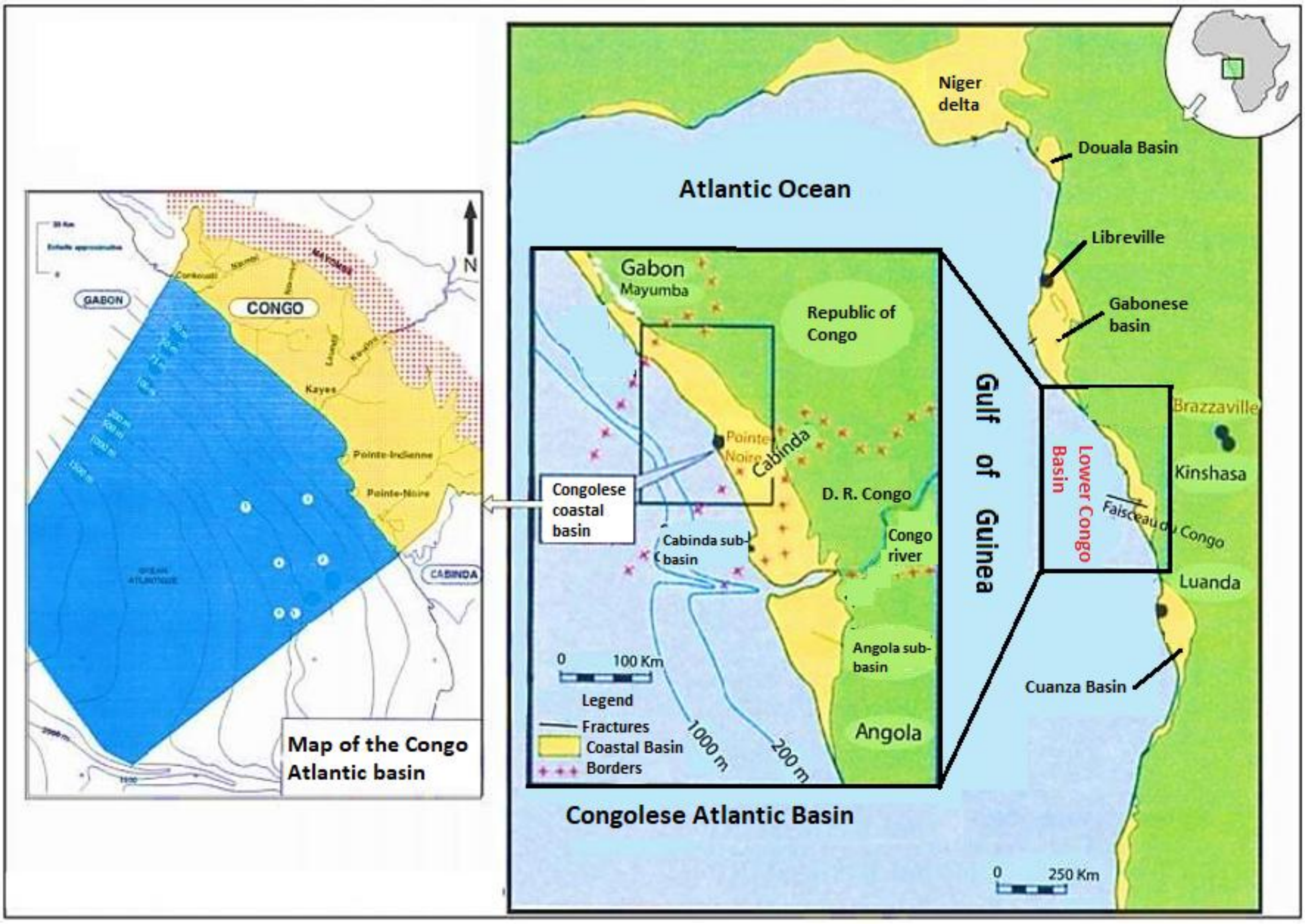

Figure 2: Location maps of the Congolese Atlantic basin (Vernet *et al.*, 1996)

III. MATERIALS AND METHODS

This study is carried out on a set of 252 samples (core samples and cuttings) from six boreholes carried out by two oil companies in the Congolese Atlantic basin. Paleogeography, in relation to the gradually increasing bathymetry on the Congolese Atlantic margin, contributed to the choice of the location of these wells.

Sediment samples were taken to analyze the heavy metal content. The purpose of heavy metal analysis is to assess and monitor the ecological quality of the marine environment. Samples submitted for heavy metal analysis are taken every 3 m. The heavy metals selected in this study are lead (Pb), chromium (Cr), zinc (Zn), cadmium (Cd) and nickel (Ni). The analysis is performed by inductively coupled plasma emission spectrometry (ICP). A tri-acid attack (nitric acid, hydrofluoric acid and perchloric acid) is applied to the samples in order to eliminate the organic matter and quartz contained in the sediment and to dissolve the metal elements to be assayed. The determination of organic matter in sediments is based on the knowledge of the TOC analyzed by carbograph.

The bathymetric zone in which benthic foraminifera are collected extends from the continental slope to the bathyal environment. The micropaleontological preparations of the cores are selected to study and evaluate their micropaleontological content, in particular benthic foraminifera. The sampling step adopted is 10 cm. We consider, in agreement with the hypothesis of Murray (2000), that only living foraminifera are more significant in this study since it is living individuals that react to environmental variations. In order to distinguish between live foraminifera and dead foraminifera, the samples are quickly processed within 24 hours of collection. The Walton method (1952) was then used, as recommended by Murray and Bowser (2000). This method (Walton, 1952) is based on the Bengal Rose solution which tints the organic matter bright pink. It quickly differentiates between living and dead individuals.

Indeed, the sample is placed for 24 hours in the fixative (2% formalin), then, after washing with fresh water, placed in a solution of Bengal Rose (1 g/L of ethanol), for 10 minutes. A second wash removes the excess dye. Bengal Rose has the advantage of not attaching itself to shell debris and mineral particles. Live foraminifera must have their protoplasm reduced to a bright red pellet usually located in the last compartment. We note as some authors (Jorissen et *al*., 1995, Gupta, 2002, Murray, 2006), in the applicability of Walton's method (1952), that some dead foraminifera with a preserved protoplasm can be interpreted as alive, which can lead to an overestimation of the number of living foraminifera.
However, the criteria for determining Bengal pink are strict and the following (Geslin et *al*., 2004):

- homogeneous colouration: at least one coloured compartment;
- bright pink color;
- presence of protoplasm evidenced by "wet-picking" (i.e., wetting of the test to make the calcite less opaque);
- absence of signs of perforation (linked to predation by nematodes for example);
- presence of sediment near the foramen.

After drying in an oven at 50°C, the residues obtained pass through a set of sieves between 500 μ and 63 μ. The foraminifera are then analysed and sorted with a binocular magnifying glass, at a magnification of between 20 and 100 times, with a needle or a very fine moistened brush.
The determination of benthic foraminifera is based on the classification described by Loeblich and Tappan (1987). The benthicforaminifera tests obtained are classifiedintotwo groups: calcarehyalinebenthics (B-CH) and agglutinatedbenthics (B-AG). To study the behaviour of thesetwo groups, weadopted the

analyticalapproach of Koutsoukos and Hart (1990) whichdivides the benthicgenerainto 7 morphogroups. This approach correlates the associations of benthic genera to a morphotype, a habitat, a biotope, a degree of resistance to stress and a paleoenvironment. Finally, the biostratigraphic division is based on the zonation established by Olsson *et al.* (2006), Berggren *et al.* (1995) and Berggren and Pearson (2005).

IV. RESULTS

The analysis of the concentrations of the selected heavy metals (Pb, Cr, Zn, Cd, Ni) in the sediments of the Congolese Atlantic basin revealsresults (table 1 and figure 3) above the natural concentrations defined by Illou (1999) and OSPAR (2019).

Table 1: Heavy metal content in sediments in the Congolese Atlantic basin

Nature	Lead (Pb)	Chrome (Cr)	Zinc (Zn)	Cadmium (Cd)	Nickel (Ni)
Study Contents	67.7 mg/kg	105 mg/kg	510 mg/kg	58 mg/kg	40 mg/kg
Natural levels (OSPAR, 2019)	47 mg/kg	81 mg/kg	150 mg/kg	1,2 mg/kg	20,9 mg/kg
Illou Contents (1999)	30 mg/kg	45 mg/kg	61 mg/kg	3 mg/kg	7 mg/kg

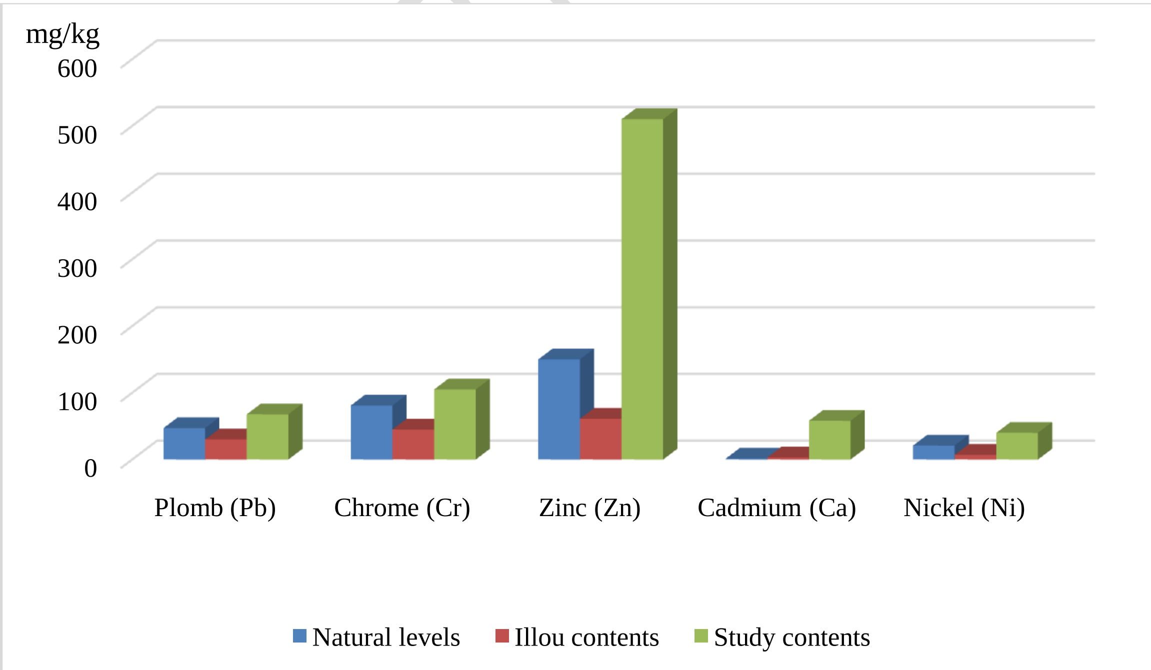

Figure 3:evolution of heavymetal concentrations in the marine sedimentsstudied

The micropaleontologicalanalysiscarried out gave a total of 1749 benthic foraminifera. The results obtained on benthic foraminifera reveal a taxonomically diverse fauna. From these results, 1063 agglutinated species (60.78%, divided into 20 genera and 33 species) and 686 calcareous test species (39.22%, divided into 39 genera and 69 species) can be distinguished. The micropaleontological collection of benthic foraminifera is very significant in terms of quantity and biodiversity. Agglutinated benthic foraminifera are more abundantthancalcareousbenthicforaminifera (figure 4). Calcareous benthic foraminifera are the most diverse (Tables 2 and 3). The assemblage of agglutinatedbenthicspecies (figure 5) isricher in *Rhabdamminaabyssorum*, *Textulariapanamensis*, *Ammoniabeccarii* and *Trochamminaglobigeriniformis*. The species*Eponidesspp*., *Bolivinatenuicostata*, *Anomalinoidesspp*., *Nonionasterizans* and *Uvigerinaperegrina*are the mostabundant in the calcareousbenthicspecies assemblage (figure 6). The number of living benthic foraminifera is preponderant for a meaningful ecological analysis. In addition, 29.16%, i.e. 360 agglutinated benthic foraminifera and 150 calcareous benthic foraminifera, have not been identified because of traces of pollution by hydrocarbon discharges. The monitoring of the level of oil pollution is exclusively based on the biological and morphological aspects observed on benthic foraminifera. Growth defects, morphological abnormalities, calcification defects and the presence of parasitism are abnormalities observed in these undetermined species. The tests present a number of anomalies that affect all species in fairly large proportions. There is a biotope that is not very conducive to the development of certain species. These deformations are related to the high concentration of heavy metals released by hydrocarbons. The families Ammoniidae, Bolivinidae, Buliminidae, Cassidulinidae, Cibicididae, Eponididae, Nonionidae, Rhabdamminidae, Textulariidae and Uvigerinidae show pronounced traces of pollution.

Following the direction of the deepening of the sea level, the pollution of the seabed by hydrocarbons has made it possible to note:

- the infralittoral zone: itisdominated by the Textulariidae assemblage, in particular by the species*Textulariapanamensis*. Several individuals suffer from a morphological malformation. The elongated test presents torsion and irregularities with a reduced number of compartments. Traces of pollution are also observed on several unidentified species.
- the circalittoral zone: this environment is marked by the abundance of the Ammoniidae assemblage, including the species*Ammoniabeccarii*. This species has several individuals polluted by hydrocarbon discharges. Some tests *of Ammonia beccarii* show growth defects and morphological irregularities. Several unidentified benthic foraminifera bearing traces of pollution are recorded.
- the bathyal zone: this environment is dominated by the assemblages of Bolivinidae, Buliminidae, Cassidulinidae, Cibicididae, Eponididae, Nonionidae, Rhabdamminidae, Textulariidae and UvigerinidaeUvigerinidae. The speciesbelonging to these assemblages are listed in Tables 3 and 4. The Uvigerinidaefamilyis the most important and seems to characterize the biotope. Severalspeciescollected carry morphological malformations and parasites. Oil pollution marks are alsorecorded on manybenthicforaminifera, makingthemdifficult to identify. In general, thereisalso a reduction in abundance, a decrease in the size and diversity of benthicforaminifera.

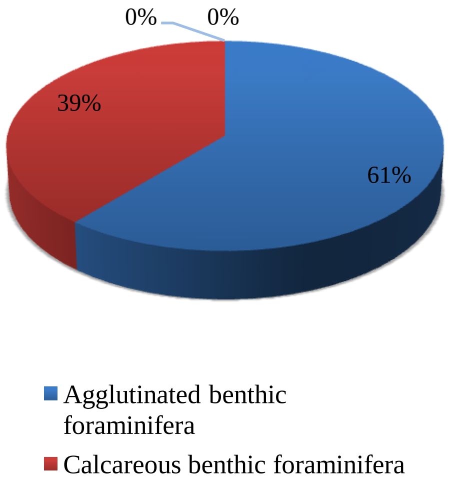

figure 4: distribution of benthic foraminifera

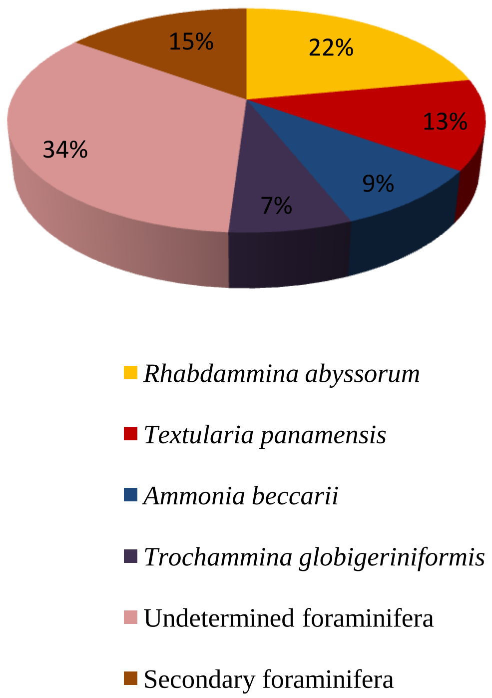

figure 5: distribution of agglutinated benthic foraminifera

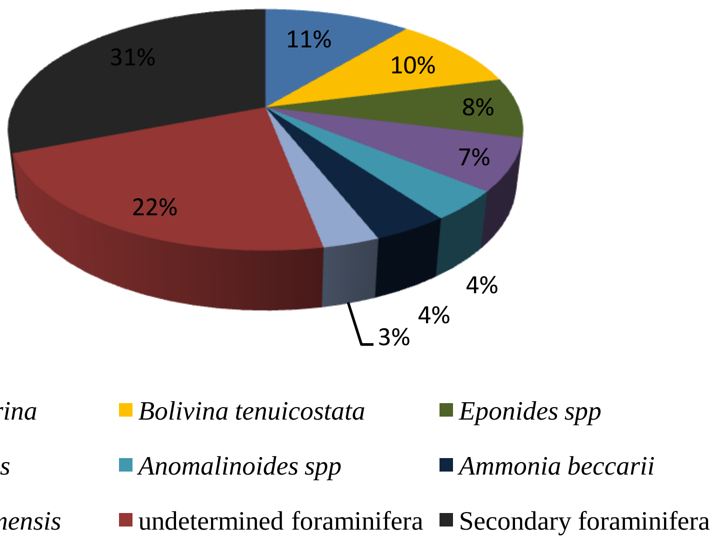

figure 6: distribution of calcareous benthic foraminifera

Table 2: List of calcareous benthic foraminifera

Alabaminatangentialis	*Gavelinella*spp.
Allomorphinatrigona	*Globobuliminaovata*
*Altistoma*spp.	*Globocassidulinasubglobosa*
Ammoniabeccarii	*Gyroidinaneosoldanii*
*Anomalina*cf. *alazanensisspissiformis*	*Gyroidinoides*spp.
*Anomalinoides*cf. *helicinus*	*Lenticulin* spp.
*Anomalinoides*spp.	*Megastomellaafricana*
Bolivina antiqua	*Melonis affinis*
Bolivinabeyrichi	*Melonispompilioides*
Bolivina cf. *Imperatrix*	*Melonis*spp.
Bolivinadifformis	*Miocene neobulimin*
Bolivinainterjunctamandarovensis	*Neobulimina* spp.
Bolivina spp.	*Nodosaria*spp.
Bolivinatenuicostata	*Nodosaria/Dentalina*spp.
*Brizalina*aff.*dilatata*	*Nonion centrosulcatum*
*Brizalina*cf. *mexicana*	*Nonionasterizans*
Brizalinadilatata	*Nonionella*cf. *turgida*
*Brizalina*spp.	*Osangularia*spp.
Bulimina elongata	*Planulinaariminensis*
Bulimina elongata group	*Planulina*spp.
Buliminaexilis	*Planulinawuellerstorfi*
*Bulimina*spp.	*Polymorphinid*spp.
*Buliminella*spp.	*Pseudocassidulinoidesgaloa*
Cassidulinella pliocenica	*Quinqueloculina*spp.
Cassidulinoidesbradyi	*Sphaeroidina bulloides*
Chilostomellaovoidea	*Spiroloculina*spp.
Cibicidoides pseudoungerianus	*Uvigerina* cf. *Mexican*
*Cybicidoids*spp.	*Uvigerina* cf. *pilgrim*
Cibicidoides ungerianus	*Uvigerinamantaensis*
*Dentalina*spp.	*MexicanUvigerina*
Elphidiumcrispum	*Uvigerinaperuviana*
*Eponides*spp.	*Uvigerina* spp.
*Eponides*spp. *(small)*	*Uvigerina* spp. (pustulate)
*Fursenkoina*spp.	*Valvulineria gigantea*

	*Valvulineria*spp.

Table 3: List of agglutinated benthic foraminifera

*Ammobaculites*spp.	*Haplophragmoides*cf. *kirki*
Ammodiscusincertus	*Haplophragmoides Church*
*Ammodiscus*spp.	*Haplophragmoides*spp.
*Ammomarginulina*spp.	*Hormosinapilulifera*
Ammosphaeroidinapseudopauciloculata	*Karreriellabradyi*
Bathysiphoncapillaris	*Marssonella*spp.
Conglophragmiumcoronatum	*Recurvoides*spp.
Cyclamminaacutidorsata	*Reophax*spp.
Cyclamminaamplectens	*Rhabdamminaabyssorum*
Cyclammina placenta	*DiscreetRhabdammina*
Cyclamminarotundidorsata	*Rhabdammina robusta*
*Cyclammina*spp.	*Rhabdammina*spp.
Eggerellabradyi	*Saccammina placenta*
*Eggerella*spp.	*Saccammina*spp.
*Evolutinella*spp.	*Textularia*spp.
Unidentified benthic foraminifera	*Trochammina globigeriniformis*
*Gravellina*spp.	*Trochammina spp.*

V. DISCUSSION

The micropaleontological and geochemical results obtained show signs of pollution on benthic foraminifera and their living environment.

The heavy metal content obtained (Pb: 67.7 mg/kg dw; Cr: 105 mg/kg dw; Zn: 510 mg/kg dw; Cd: 58 mg/kg dw; Ni: 40 mg/kg dw) in sediments are higher than the natural levels. Overall, these concentrations are higher in the subtidal zone than in the bathyal zone. Despite the decline in heavy metal concentrations towards the bathyal zone, the values still remain very high. These results, which are well above the standards, prove that the marine environment is highly polluted. The impact of this pollution is remarkable for the multiple deformations and malformations presented by benthic foraminifera. These results are comparable to those obtained by Illou (1999) on the Tunisian coast. All these data, in agreement with Illou (1999), can be correlated with the results obtained by Lei *et al.* (2016) in the Bohai Sea in China, after the Penglai oil spill in 2011. This shows that the effluent levels are often higher than the discharge standards accepted in the maritime public domain (Illou, 1999).

The infralittoral zone, characterized by low hydrodynamics, is a very calm environment. It has a high concentration of dissolved nutrients and a biotope rich in various species. This area has a high proliferation of microfossils and an abundance of the species*Textulariapanamensis* and *Ammoniabeccarii*. The subtidal zone exposed to anthropogenic oil pollution has a high percentage of polluted benthic foraminifera. It should be noted that the proximity of the infralittoral to the coast makes this environment particularly sensitive to anthropogenic disturbances (Barbier, 2017; Gasiunaite, *et al.*, 2021). The presence of hydrocarbon (heavy metals) discharges in this area makes the environment cloudy, sometimes anoxic. Given the calm environment, a large number of benthic foraminifera ingest hydrocarbons that impact their metabolic systems. They react quickly and negatively by developing morphological abnormalities: torsion of the test, greatly reduced size of the test, reduced number of compartments. Some benthic foraminifera disappear by suffocation, others try to migrate to the least polluted areas. In this context, many polluted foraminifera become difficult to identify. This abnormal situation could explain the decline in diversity in the environment. Schwing*et al.* (2015, 2018) reached similar conclusions in the northern Gulf of Mexico.

In the circalittoral zone, hydrodynamics are very low from 50m, and sediments are deposited according to this energy gradient. The pollution of hydrocarbons of anthropogenic origin on all the benthic foraminifera of the Congolese Atlantic basin caused abnormal torsions of the test, an anarchic development of the compartments, the appearance of abnormal protrusions and the appearance of additional openings. These anomalies recorded by the benthicforaminifera*Textulariapanamensis* and *Ammoniabeccarii*maketheir identification difficult, sometimes impossible. Alve (1991) and Yanko *et al* (1999) demonstrate that the presence of pollutants (heavy metals) profoundly affects the morphology of benthic foraminifera. Some benthic foraminifera that are not resistant to pollution are disappearing, resulting in a decrease in diversity. In addition, the death of these microfossils allows opportunistic species to settle in the environment.

The spillage of polycyclic aromatic hydrocarbons in the bathyal zone leads to changes in environmental parameters (salinity, pH, oxygenation, luminosity) with various consequences, often interdependent and difficult to distinguish. Pollutants also interact with other chemical stressors (Scholz N. L. *et al*., 2012). Thus the conditions of the biotope become very difficult and unfavourable to the development of benthic foraminifera. This anthropogenic pollution first affects the metabolic system of benthic foraminifera, which leads to morphological disturbances, which testifies to the negative impact of their normal growth. In this bathyal environment, species belonging to the families Bolivinidae, Buliminidae, Cassidulinidae, Cibicididae, Eponididae, Nonionidae, Rhabdamminidae, Textulariidae and UvigerinidaeUvigerinidae are the mostaffected (deformations and malformations) by oildischarges. In addition, it has been observed that benthic foraminifera react rapidly and negatively in a distinctive way to metabolic disturbances: the composition of associations changes, diversity decreases, the size of tests decreases very sharply, and abnormalities multiply. According to Muncaster *et al*. (2016), exposure of microfossils to oil damages their reproductive system, alters their growth rate, and alters their behaviour. Heavy metals therefore limit the activity of microorganisms and their multiplication, as confirmed by Martens *et al.* (1994), Kandeler, Tscherko *et al*. Sandaa, Torsvik et al. (2000). (2000). The environment sometimes becomes anoxic, leading either to the disappearance of certain foraminifera or to the displacement of other species to less polluted places.

Generally speaking, from the infralittoral zone to the bathyal zone, several polluted foraminifera (29.16%) are not determined because of the multiple malformations observed. The deformation of the tests and the abnormally high diversity of benthic foraminifera would therefore be a good detector of pollution. The spill of polycyclic aromatic hydrocarbons at the bottom of the Congolese Atlantic basin leads to two main actions. The first, called the lethal effect or direct toxic effect, causes the rapid death of benthic foraminifera after absorption of harmful compounds. The second, called the sublethal effect or indirect toxic effect, leads to an accumulation of harmful effects and generates very serious disorders that can lead to the death of the individual.

Petroleum pollution by polycyclic aromatic hydrocarbons is one of the most noticed forms of marine pollution, an observation agreed with Landos *et al*. (2021) who showed that polycyclic aromatic hydrocarbons do not readily dissolve in water and tend to accumulate or bind to sediment particles over several decades. Oil discharges into the marine environment release heavy metals (lead, chromium, cadmium, nickel, zinc) that have harmful effects on marine ecosystems. These metals are potentially toxic contaminants, they accumulate in sediments, bio-concentrated by organisms (Daby, 2006). Heavy metals are among the most polluting (Baize, 2000). These heavy metals, dissolved in sediments, have a great lasting influence on the seabed environment (Wenbin *et al*., 2017). The usefulness of benthic foraminifera as sensitive indicators of marine pollution caused by oil discharges remains supported by many authors [Danovaro *et al*. (2008), Levin and Dayton (2009), Ramirez-Llodra *et al*. (2010 and 2011) and Thurber *et al.* (2014)].

VI. Conclusion

The objective of this study was to assess the impact of oil pollution on benthic foraminifera and their ecosystems. Anthropogenic pollution of the seabed by hydrocarbons has a toxic effect on the microfauna, particularly benthic foraminifera, of the Congolese Atlantic basin. The discharge of hydrocarbons into the marine environment releases heavy metals which, when dissolved in sediments, have a direct harmful effect and a great lasting influence on foraminifera and their ecosystem. Geochemical analysis of heavy metals reveals concentrations higher than natural standards. Benthic foraminifera receive and absorb most of the pollutants released into the marine environment. This results in the disruption of their metabolic system, growth and morphology, which testifies to the negative impact on their development. This pollution leads to numerical and qualitative reduction. The decline in biodiversity makes several foraminifera indeterminable, constituting an obstacle for the interpretation and reconstruction of paleoenvironments. Micropaleontological analysis shows that 29.16% of benthic foraminifera affected by pollution could not be identified. This study shows that benthic foraminifera are excellent bio-indicators for the assessment of marine oil pollution. Marine oil pollution therefore poses a real threat to the health of these ecosystems in general, and to benthic foraminifera in particular. It is therefore necessary to avoid the transfer of hydrocarbon discharges into the marine environment and to regulate the use of hazardous substances on oil platforms. The reuse and recycling of hydrocarbondischarges on oil platforms is a key preventivemeasure.

Thanks

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