Contribution of macroinvertebrates to leaf breakdown in the Okpara River, Bénin.

Tchaou C. Madina, Gouissi F. Modeste*, Abahi K. Simon, Adje D. Darius, Orou Piami Zoulkanerou, Okoya J. Antoine and Gnohossou M. Pierre
Department of Management of Natural Resources (AGRN), Water and Soil Engineering (IES), Laboratory of Ecology, Health and Animal Production (LESPA), Faculty of Agronomy (FA) University of Parakou (UP) BP 123 Parakou Benin
*Corresponding author: gouissi@yahoo.fr

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ABSTRACT
The decomposition of organic matter is a key process in the functioning of rivers. This organic matter is an important source of energy and nutrients in food webs. The objective of this work is to evaluate the degradation of the litter of three plant species in the water of the Okpara River. The degradation of the three leaves (Piliostigma thonningii, Terminalia avicennioides and Flueggea virosa) was made during the rainy season by using the litterbag technique of different mesh sizes which are large mesh (LM) of 5 mm and fine mesh (FM) of 0.5 mm. A total of 96 litterbags, each containing 3.5 g of dried litter were used; 32 litterbags (16 LM litterbags and 16 FM litterbags) per species. These litterbags were placed in the Okpara River waters during 28 days and 24 litterbags are recovered at four different dates: one week, two weeks, three weeks and four weeks. At each recovery date, four (04) physico-chemical parameters (temperature, pH, conductivity and TDS) were measured and the macroinvertebrates were harvested. The study found that Fluggea leaves decompose and lose their weight faster than those of Terminalia and Piliostigma, a very rapid loss since it reaches the value of 58.29% of the initial weight in 28 days only. This loss of initial weight could be related to the structure of the leaf, the action of microorganism and the moderate rise in temperature (26 °C) favoring a biological activation. The study also revealed the dominance of Chironomidae family and functional collector groups.

INTRODUCTION
Leaf litter decomposition is the important source of energy for aquatic communities (Swift et al., 1979). These processes contribute to the degradation and the consumption of the litter materials by decomposers, scavengers and detritus feeders (Couteaux et al., 1995).

Thus, the decomposition of organic matter is a fundamental process that directs the functioning of rivers and the functional integrity of these ecosystems can be apprehended in a relevant way by an assessment of litter decomposition rates (Tenkiano, 2017).

In considering this importance, the litter’s decomposition has been experimented in some African regions. For instance we have Tenkiano (2017) who has worked on the decomposition of leaves species of: Albizia Zygia, Milletia Zechiana, Pterocarpus Santalinoides, Alchornea Cordifolia in the watercourses of
Guinea and Dobson et al. (2003) who have conducted their research in the decomposition of leaves of *Dombey Goetzii*, *Rhus Natalensis*, *Syzygium cordatum*, *Vanguera madagascariensis* on the Njoro River in Kenya. Despite their results, very little informations are available on the decomposition of the litters in west Africa.

In the same vain, our knowledge in Benin Republic is still limited, because no previous study has highlight the decomposition of litter and the organisms involved in watercourses.

It is therefore be necessary in this study to overcome this shortcoming by providing a first database on the decomposition of three leaf species in the Okpara River.

**MATERIALS AND METHODS**

**Study area**

The study was conducted on the Okpara River, which is one of the tributaries of the Ouémé River. The Okpara River is located between 8°14'-9°45'N latitude and 2°35'-3°25' longitude in the department of Borgou and originates at 450 m altitude to southwest from Nikki to Péèrè. The Okpara River has an irregular flow rate ranging between 0.0001 and 150 m³/s. It is approximately 362 km long with a basin of a total area of 10,000 km². Its basin largely covers the communes of Péèrè in the east, Nikki in the north-east, N'dali in the west, Parakou and Tchaourou in the south and finally a small part of the commune of Bembèrèkè in North. The study area is influenced by the southern Sudanese climate, characterized by a long dry season (mid-October to April) followed by a wet period (May to mid-September). The average annual rainfall over the last ten years is 1028.90 mm with maximum rainfall amounts during the months of August and September.

**Choice, harvest and leaves treatment**

For this study, we have chosen the leaves of three litters which are all widely spread species all along the upper part of Ouémé River. There are the leaves of *Piliostigma Thonningii*, the leaves of *Terminalia avicennioides* and the leaves of *Flueggea virosa*.

The three species leaves were harvested in the dry season using the gardener's pruning shears in order to avoid the contact of the leaves with the soil and to avoid the beginning of decomposition's process before the experience.

In the laboratory, the harvested leaves were air-dried, sorted to remove the already deteriorated leaves and oven-dried at 65 °C for 48 hours (Allen, 2013).

The leaves were then chilled, weighed to 0.1 mg by lots of 3.5 g (Allen, 2013) and finally stored in plastic bags for use.

**Study of leaves degradation in water**

The lots of 3.5 g of air-dried litter were used to fill each of the 96 litterbags. The two types of litterbags used were differentiated by the size of the space between the meshes. The large mesh (LM) litterbags have a mesh gap of 5 mm, which allows macroinvertebrates to have access to the leaves.

The fine mesh (FM) litterbags have a mesh gap of 0.5 mm to exclude invertebrates shredders without disturbing colonization by microorganisms.

The litterbags were placed during the rainy season and collected at four different dates: One week, two weeks, three weeks and four weeks, for a total of 28 days spent in the river.
The litterbags are attached to the bottom of the watercourses with chains. On each chain are positioned a LM litterbag and a FM litterbag for each species of leaf, ie (06) six litterbags per chain.

At each date, four chains were recorded, making four repetitions for each type of mesh. In total, 96 bags were used: 48 litterbags of large mesh and 48 litterbags of fine mesh. Each bag was recovered individually, while retaining all the macroinvertebrates with the litterbag.

**In situ measurement of physical parameters of water**

The measurements of physical parameters (temperature, conductivity, pH and TDS) were performed using field materials *in situ* early in the morning, during the establishment of chains and at each date of extraction of the chains. Thus, a portable electronic conductivity meter (HANNA HI 99300) was used for measuring the conductivity (Cond in µS/cm), the TDS (mg/L) and the temperature (Temp °C) and a portable pH meter (HANNA HI 98107) was used to measure the pH of the water.

**Litterbags recovery and treatment**

In the laboratory, the samples were processed following the removal of the litterbags. The litterbags were emptied individually. The leaves contained in the litterbags were cleaned under tap on a 100 μm sieve; the invertebrates associated with these litters were sorted and conserved in the bowls in an alcohol solution of final concentration of 70 ° to be identified later under binocular dissecting microscope. The leaves contained in the litterbags are also cleaned and ridded of sediment.

**Identification of macroinvertebrates**

In the laboratory, the captured macroinvertebrates were identified under a binocular dissecting microscope. This operation was done according to the methodology as defined by other authors (Abahi, 2018, Abahi *et al.*, 2018) who worked on macroinvertebrates in northern Benin. The taxonomic determination was made using the following keys: Benthic Macroinvertebrates of the streams of "la Nouvelle-Calédonie" (Mary, 2017), "Identification guide of the main benthic macroinvertebrates of freshwater from Quebec written by Moisan (2010), Freshwater invertebrates: Systematic, biology, ecology (Tachet *et al.*, 2000) and aquatic entomology (McCafferty, 1981). After the identification, the samples were stored in 70% alcohol in the laboratory in pill containers and the faunistic list was established.

**Determination of mass loss and decomposition rate**

The recovered litter was dried in the oven at 65 °C to constant mass and weighed at 0.1 mg to determine the remaining mass. The decomposition rate k will be calculated using the following formula:

\[ M_t = M_0e^{-kt} \]

\( M_t \) is the mass remaining at time t, \( M_0 \) the initial mass at \( t_0 \), k the decomposition rate and t the exposure time in days.

Depending on the value of k, there are three types of decomposition (Petersen and Cummins, 1974):

- Slow decomposition (k <0.005 days -1)
- Average decomposition (0.005 < k < 0.002 days -1)
- Fast decomposition (k >0.002 days -1)

**Statistical processing and data analysis**

By using the Excel spreadsheet, macroinvertebrate inventory data and physico-chemical parameters were processed. The test of Kruskal-Wallis to the step of 5% with R3.4.2 software was used to evaluate the variability of Kinetics of decomposition and physico-chemical parameters.
RESULTS

Environmental Variable

The average value of (04) four physico-chemical parameters of water quality during the experimentation is presented in Table 1.

Table 1: Physical parameters values of Okpara River water during the experimentation

<table>
<thead>
<tr>
<th>Dates</th>
<th>Temperature (°C)</th>
<th>TDS (ppm)</th>
<th>pH</th>
<th>Conductivity (µS/cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 Date</td>
<td>26.10 ± 0.10</td>
<td>180.3 ± 1.71</td>
<td>8.02 ± 0.27</td>
<td>367.11 ± 1.14</td>
</tr>
<tr>
<td>07 Dates</td>
<td>26.15 ± 0.07</td>
<td>184.5 ± 0.70</td>
<td>8.05 ± 0.07</td>
<td>369.0 ± 1.44</td>
</tr>
<tr>
<td>14 Dates</td>
<td>26.15 ± 0.21</td>
<td>151.0 ± 5.65</td>
<td>8.45 ± 0.07</td>
<td>302.0 ± 11.31</td>
</tr>
<tr>
<td>21 Dates</td>
<td>32.15 ± 7.42</td>
<td>166.5 ± 3.53</td>
<td>8.80 ± 0.15</td>
<td>339.5 ± 2.12</td>
</tr>
<tr>
<td>28 Dates</td>
<td>26.15 ± 0.22</td>
<td>163.5 ± 0.70</td>
<td>8.40 ± 0.14</td>
<td>327.0 ± 1.41</td>
</tr>
</tbody>
</table>

The analysis of the measured temperatures reveals that the highest temperature was observed on the 21st day of the experimentation and the smallest temperature value was recorded on the day of the launching of the litterbags.

The conductivity values observed range between 302.0 ± 11.31 μS / cm and 369.0 ± 1.44 μS/cm. The highest conductivity was recorded on the 7th day of the experimentation and the lowest was obtained on the 14th day.

The dissolved solids content described a similar evolution as that of the conductivity. In addition, the highest pH values were observed on the 21st day of the experimentation and the lowest pH value was recorded on the 7th day.

In the same way, the physico-chemical parameters of water quality subjected to the Kruskal-Wallis test did not reveal any significant difference between the stations (P=0.3916).

Decomposition of leaves

Kinetics of leaf decomposition of different species

Whatever the mesh of the litterbags, the *Flueggea* leaves decomposition speed (0.059) is slightly higher than that of the leaves of *Terminalia* and *Piliostigma* (Figure 1 and 2). But this difference is neither significant in FM (P = 0.1928) nor in LM (P = 0.2231) at 5%.

In addition, all the leaves have undergone a rapid decomposition because their decomposition rate \(k\) is greater than 0.002 day \(-1\) (Table 2).

![Decomposition rate of the leaves contained in the fine mesh litterbags after 28 days of immersion](image)

Fig. 1: Decomposition rate of the leaves contained in the fine mesh litterbags after 28 days of immersion
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Table 2: Value and characterization of the decomposition of different species

<table>
<thead>
<tr>
<th>Different species</th>
<th>Average value of K</th>
<th>Characterization of decomposition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Piliostigma</td>
<td>0.032</td>
<td>Fast</td>
</tr>
<tr>
<td>Terminalia</td>
<td>0.041</td>
<td>Fast</td>
</tr>
<tr>
<td>Flueggea</td>
<td>0.047</td>
<td>Fast</td>
</tr>
</tbody>
</table>

Kinetics of decomposition of leaves according to different meshes

Figure 3 presents the decomposition rate of the leaves according to different mesh. It reveals that the decomposition rate of the leaves contained in the litterbags of large meshes is high. But between the different meshes there is no significant difference at the 5% (p = 0.7436).

Evolution of leaf mass according to time

It appears that as the days pass the initial mass of leaf decreases. Thus, the initial mass which was 3.5 g gradually decreases to take the value of 1.46 g on the twenty-eighth day. Whatever the date, the mass of the Flueggea species is small more than the mass of the other species. Thus, leaf decomposition of Flueggea is faster than for other species; a very fast loss since it reaches the value of 58.29% of the initial weight in 28 days only (Figure 4).
Benthic macroinvertebrates of Okpara River water

The study has helped to capture 461 macroinvertebrates that belongs to 11 families and 07 orders.

Abundance of macroinvertebrates classes

The harvested macrofauna is dominated by insects (79.83%), followed by worms (10.41%) and molluscs is the smallest (9.76%) (Figure 5).

![Figure 5: Relative abundance of different classes of captured macroinvertebrates](image)

Abundance of macroinvertebrates orders

Figure 6 illustrates the relative abundance of different orders of captured macroinvertebrates. Diptera constituted the first group of the macrofauna with 77.22% of the richness. They are followed by gastropods (9.76%), Oligochaetae (6.94%), Nemathelminths (3.25%) and Ephemeroptera (2.17%). The other orders: Plecoptera and Leeches each has less than 1% of the total richness.

![Figure 6: Relative abundance of different orders of captured macroinvertebrates](image)

Abundance of macroinvertebrates families

Eleven (11) macroinvertebrate families were captured (Table 3). The families of Chironomidae collected during the study constitute 77.01% of the total richness with 355 individuals. They are followed by Limnaeidae (8.68%), Lumbriculida (6.94%), Nematodes (3.25%) and Leptohyphidae (1.95%). The other families are the most marginal with percentages varying between 0.22% and 0.87% of the total richness.
Contribution of macroinvertebrates to leaf breakdown in the Okpara River in Bénin

Table 3: Macroinvertebrates collected in the Okpara River waters

<table>
<thead>
<tr>
<th>Classes</th>
<th>Orders</th>
<th>Families</th>
<th>Total effective</th>
<th>Percentage of families</th>
</tr>
</thead>
<tbody>
<tr>
<td>Insects</td>
<td>Diptera</td>
<td>Chironomidae</td>
<td>355</td>
<td>77.01</td>
</tr>
<tr>
<td>Insects</td>
<td>Diptera</td>
<td>Culicidae</td>
<td>1</td>
<td>0.22</td>
</tr>
<tr>
<td>Insects</td>
<td>Ephemeroptera</td>
<td>Leptophyidae</td>
<td>9</td>
<td>1.95</td>
</tr>
<tr>
<td>Insects</td>
<td>Ephemeroptera</td>
<td>Ephemereillida</td>
<td>1</td>
<td>0.22</td>
</tr>
<tr>
<td>Insects</td>
<td>Plecoptera</td>
<td>Perlodidae</td>
<td>2</td>
<td>0.43</td>
</tr>
<tr>
<td>Molluscs</td>
<td>Gastropods</td>
<td>Physidae</td>
<td>4</td>
<td>0.87</td>
</tr>
<tr>
<td>Molluscs</td>
<td>Gastropods</td>
<td>Planorbidiae</td>
<td>1</td>
<td>0.22</td>
</tr>
<tr>
<td>Molluscs</td>
<td>Gastropods</td>
<td>Limnaeidae</td>
<td>40</td>
<td>8.68</td>
</tr>
<tr>
<td>Worms</td>
<td>Leeches</td>
<td>Glossiphoniida</td>
<td>1</td>
<td>0.22</td>
</tr>
<tr>
<td>Worms</td>
<td>Nemathelmintes</td>
<td>Nematodes</td>
<td>15</td>
<td>3.25</td>
</tr>
<tr>
<td>Worms</td>
<td>Oligochaeta</td>
<td>Lumbriculidae</td>
<td>32</td>
<td>6.94</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td></td>
<td></td>
<td><strong>461</strong></td>
<td><strong>100</strong></td>
</tr>
</tbody>
</table>

Abundance of macroinvertebrates according to plant species

The distribution of absolute abundances by species is quite variable (Table 4). Absolute abundances range from 196 to 121 individuals. *Terminalia* represents the lowest abundance and *Flueggea* the highest one.

Table 4: Abundance of macroinvertebrates according to plant species

<table>
<thead>
<tr>
<th>Orders</th>
<th>Families</th>
<th><em>Flueggea</em></th>
<th><em>Pilostigma</em></th>
<th><em>Terminalia</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Diptera</td>
<td>Chironomidae</td>
<td>154</td>
<td>117</td>
<td>84</td>
</tr>
<tr>
<td>Diptera</td>
<td>Culicidae</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Gastropods</td>
<td>Physidae</td>
<td>1</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Gastropods</td>
<td>Planorbidiae</td>
<td>0</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Nemathelmintes</td>
<td>Nematodes</td>
<td>0</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>Oligochaeta</td>
<td>Lumbriculidae</td>
<td>17</td>
<td>0</td>
<td>15</td>
</tr>
<tr>
<td>Ephemeroptera</td>
<td>Leptophyidae</td>
<td>1</td>
<td>1</td>
<td>7</td>
</tr>
<tr>
<td>Gastropods</td>
<td>Limnaeidae</td>
<td>20</td>
<td>7</td>
<td>13</td>
</tr>
<tr>
<td>Leeches</td>
<td>Glossiphoniida</td>
<td>0</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Plecoptera</td>
<td>Perlodidae</td>
<td>2</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Ephemeroptera</td>
<td>Ephemereillida</td>
<td>0</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td></td>
<td>196</td>
<td>144</td>
<td>121</td>
</tr>
</tbody>
</table>

Abundance of functional feeding group of macroinvertebrates

From a functional point of view, collectors were the most considerable groups in the environment of study constituted respectively of 86.12 of the total number. The scraper and shredder taxa occupy respectively the second and third place with 8.89% of the number of functional feeding groups. Predators were the lowest groups in the watercourse during the study period with an abundance of 0.65% (Figure 7).

Fig. 7: Relative abundance of functional feeding group.
DISCUSSION

The temperatures recorded during this study are relatively close to those obtained by (Tenkiano, 2017) in Guinea (26°C to 36°C). They are included among the range of favorable temperatures (24°C to 35°C) which are good for the pisciculture. (Pouomogne, 1998). According to the grid of Beau (1998), the calculated pH values reflect a good quality water and adequate for aquaculture because they are between 6.5 and 9 (IBGE, 2005). The pH values observed in our study corroborate the observations of Zinsou et al. (2016) and those of N'Diaye et al. (2013) on the Senegal River. Moreover, the high values of observed electrical conductivity oscillating between 302 and 369 μS/cm in the Okpara River waters could be explained by the strong degradation of the organic matter present in the environment and would reflect the polluted nature of these waters. Such observations have already been identified in various works in African rivers (Liwouwou et al., 2018, Abahi, 2018).

In the Okpara River, the three litter species studied in this study decomposed very quickly (k> 0, 002 day⁻¹). This fast decomposition of all litter species could be due to the variation of the temperature between 26°C and 30°C. Our observations confirm those of Tenkiano (2017) and Mathuriau and Chauvet (2002); which attributed to fast decompositions the highest values of temperature.

These authors asserted that in tropical zone the decomposition of the leaves is often very fast. In addition, a temperature varying between 10°C and 30°C would be very favorable to the decomposition process (Barbe, 2017). The fast decomposition of leaves observed in this study is attributed to the quality of leaf species studied. The fast decomposition of the Flueggea leaves compared to the leaves of Terminalia and Piliostigma is related to its low lignin content. Those observations confirm those of Mathuriau and Chauvet (2002) and Goncabes et al. (2007). So, the lignin content is a good indicator of the decomposition of leaves (Gessner and Chauvet, 1994).

In addition, the decomposition in the litterbags of large meshes is faster than the decomposition in the litterbags of fine meshes. That great decomposition observed in litterbags of large meshes is logical and could be due to the combined action of organisms and some present shredders while in the litterbags of fine meshes the decomposition is made by only micro-organisms. This result was found by Tenkiano (2017) and Allen (2013) during the study of the functioning of ecosystem of watercourses of Guinea. The study of leaf decomposition in the Okpara River waters was carried out with the 461 macroinvertebrates captured. This number of individuals observed in our study is low and very distant to those obtained in the watercourse of Guinea (Tenkiano, 2017). This difference in richness observed in our study is due to the mass of the litter in the litterbags, the duration of immersion and the different mesh used. The captured benthic macrofauna is highly represented by the insects. Within those insects, the order of diptera, mainly the family of Chironomidae is the largest. The previous studies made on the decomposition of litter (Tenkiano, 2017) shows the class of insects and the family of chironomidae are the most predominant taxa. But the dominance of the Diptera orders observed in our study is contrary to that of Tenkiano (2017), which observed rather a dominance of the order of the Heteroptera and Coleoptera. On the other hand, the proliferation of Chironomidae, which are collectors, explains an abundance of organic matter accumulated in the Okpara River waters. Adandedjan et al. (2012) showed that when the organic load becomes important the pollution-sensitive species disappear to give place to pollution-tolerant species such as Chironomidae. This observation justifies the proliferation of Chironomidae observed in the Okpara River waters. The situation
goes perfectly with the shortage of shredders recorded in the watercourse during the study that expresses the low participation of macroinvertebrates in the decomposition of litters and explains easily the importance of micro-organisms in the decomposition of litters in Benin and in Africa. Hence, microorganisms (aquatic Hyphomycetes) are responsible for the degradation of litter leaves (Tenkiano, 2017).

CONCLUSION

In this comparative study of the leaves deterioration of Piliostigma, Terminalia and Flueggea, it appears that Flueggea leaves decompose and lose weight more quickly than the leaves of the other species studied. This loss is very fast because it reaches the value of 29% of the initial weight in 28 days. The study also revealed a richness of 461 macroinvertebrates belonging to eleven families whose dominants are the family of Chironomidae. The analysis of functional feeding group highlights the predominance of collectors against the inferiority of shredders, which are actually the major agents of decomposition. This rarity of the shredders observed reveals that the decomposition of the litter is ensured by the micro-organisms. In addition, the high recorded decomposition values could be related to the lignin composition of the leaves and the moderate rise in temperature (26°) thus promoting biological activation. This study is a starting point for assessing the decomposition rate of litter in the Okpara River waters.

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