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Impacts of cotton production on the diversity and distribution of benthic macroinvertebrates in the Alibori and Sota rivers in northern Bénin

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Abstract

This study carried out on two (02) streams of the Alibori river and three (03) streams of the Sota river in the cotton basin of northern Bénin aims to highlight the impact of cotton production on benthic macroinvertebrate communities. On each stream, three (03) stations (clean; moderately polluted and heavily polluted) were determined. In each station eight (08) MIB samples were collected. In situ parameters were measured while others were assayed in the laboratory. A Canonical Correspondence Analysis (CCA) was used to match the physico-chemical data with the MIB families. Diversity indices were used to determine the effects of pesticides on MIBs. The physicochemical variables revealed a significant dominance of temperature, conductivity, total ammonia and total phosphate values under the effect of agricultural pesticides. The macrofauna collected consisted of 95.15% insects, 2.23% worms, 2.03% mollusks, 0.42% hydracarans and 0.17% Crustaceans before the use of pesticides. After the use of pesticides, this macrofauna is made up of 69.70% of insects, 26.4% of worms, 2.44% of mollusks, 1.03% of crustaceans, and 0.39% of hydracarians. A high abundance of insects and hydracarans and a low abundance of molluscs, worms and crustaceans were noted before the use of pesticides than after the use of pesticides in the Beninese cotton basin. This situation is corroborated by the low abundance and taxonomic richness as well as the low values of the Shannon index and the high values of the Hilsenhoff index at the stations after the use of pesticides, especially the stations under strong agricultural disturbances.

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Introduction

Agricultural activities are the major cause of aquatic ecosystem degradation in rural areas (Leigh *et al.*, 2010). Indeed, the use of agricultural chemical pesticides affects aquatic biological communities via agricultural runoff due to rainfall or introduced irrigation water where it presents significant ecotoxicological risks to the organisms living there. Thus, it is estimated that only 0.1% of sprayed pesticides reach target pests, the rest being distributed in ecosystems where it contaminates land, water and air (Agbohessi *et al.*, 2015). In Bénin, environmental chemical aggression poses a serious threat to aquatic biodiversity, particularly in areas of northern Bénin, where up to 90% of the national cotton production is produced. Cotton production requires the use of large quantities of pesticides (Adechian *et al.*, 2015) that affects the biological integrity of impacted ecosystems (Agbohessi *et al.*, 2015). Benthic macroinvertebrates (BMI) are the most widely used bioindicators of ecological quality (Edia *et al.*, 2010). Their studies allow the assessment of impacts of pollution and alteration of aquatic habitats (Hepp *et al.*, 2010). The presence or absence and abundance of bioindicator BIM have been shown to be determinants of the chronic and episodic impact of human disturbance on biodiversity and aquatic ecosystem functioning (Piscart *et al.*, 2009, 2011). Some previous research has demonstrated the excessive use of agricultural chemical pesticides in the cotton basin of northern Bénin (Agbohessi *et al.*, 2013; Adechian *et al.*, 2015 ; Gouda *et al.*, 2018 ; Douny *et al.*, 2021 ; Orou Piami *et al.*, 2021). Others have studied the diversity and community structure of BMI in the cotton basin in northern Bénin (Agagbé, 2008 ; Adandédjan *et al.*, 2012; Imorou Toko *et al.*, 2012a ; Houelome *et al.*, 2016 ; Chikou *et al.*, 2018). Among these previous researches only Orou Piami *et al.*, (2021) evaluated the effects of pesticides on BMI on the Sota river. Therefore, there are gaps in knowledge regarding the effects of pesticides on BMI in the cotton basin in northern Bénin, which represents the area most threatened by intensity in this sector (Agbohessi *et al.*, 2015; Imorou Toko *et al.*, 2012b). The integrated use of macroinvertebrate biodiversity information in bio-monitoring can improve the management of aquatic ecosystems (Sundar, 2020). Therefore, the present study focused on the impacts of pesticides on MIBs in the cotton basin of northern Bénin.

Materials and methods

Study environment

The study was conducted in the Alibori River and the Sota river. These two rivers are located in the cotton basin in northern Bénin and are subjected to intense agricultural pressures (Houelome *et al.*, 2016, Orou Piami *et al.*, 2021). With a watershed covering an area of 13,740 km² and 427 km long, the Alibori River originates at an altitude of about 410 m in the Kita granite massif on the flank of the Atacora chain in the commune of Péhonco. While the Sota, with a surface area of 13,360 km2 and a length of about 254 km, has its source at an altitude of more than 400 m on the eastern flanks of the Kalalé sandstone plateau, which it will bypass from the north before heading in a SSW-NNE direction on the basement formations. Ninety (90) km after its source, the Sota enters the Cretaceous sandstone formations that it cuts quite deeply and joins, after 250 km of travel, the Niger river about 1 km downstream from Malanville (Le Barbé *et al.*, 1993). The Alibori receives on its right bank the tributaries of the streams: Souédarou, Sarédarou, Darou-Woka and Sanson and on its left bank the tributaries of the streams: Morokou, Kparé, Kénou, Yassikoga, Konékoga and Kpako before flowing into the Niger River upstream from Malanville. As for the Sota, it receives successively the tributaries of Souamon on its right bank, the Tassiné (102 km long with a surface area of 3031 km2), the Bouli (145 km long and 2380 km2 of surface area), the lrané (55 km long and 1832 km2 of surface area), the Gouroukpa and the cascade of Sosso on its left bank. The Alibori River extends into the department of Alibori with many tributaries scattered in the communes of the cotton basin (Kandi, Gogounou, Banikoara, Karimama and Malanville); in the department of Atakora (commune of Kérou and Péhonko) and in the department of Borgou (commune of Sinendé).

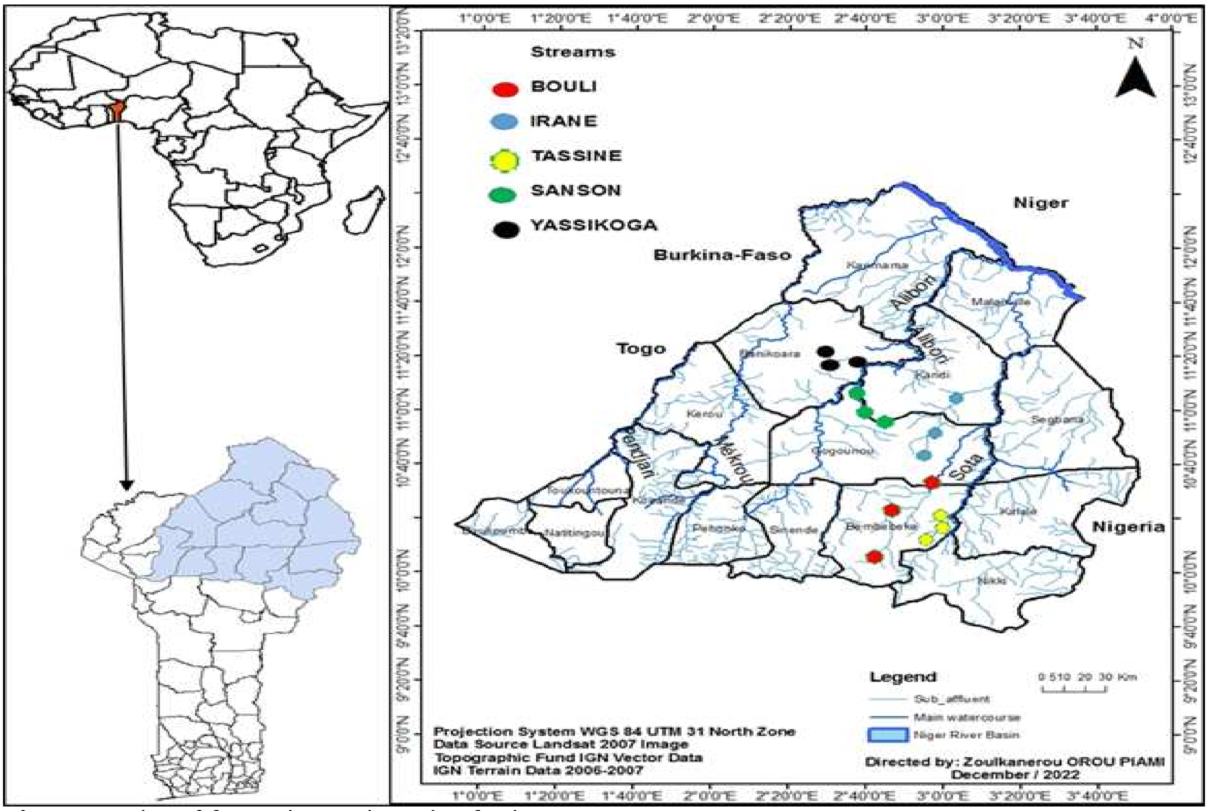

Fig. 1. Location of the Beninese Niger River basin.

While the Sota covers the communes of Malanville, Ségbana, Kandi and Gogounou in the Alibori department and the communes of Bembèrèkè, Kalalé and Nikki in the Borgou department. These rivers are aquatic ecosystems located in the Sudanian zone with a dry tropical climate. The Alibori is located between 10°30' and 12° North latitude and 1°32' and 3°50' East longitude. The Sota is located between 9°54' and 11°95' North latitude and 2°28' and 3°52' East longitude (Koumassi, 2014) (Fig.1).

Selection of sampling stations

The collection of benthic macroinvertebrate samples and physico-chemical parameters was carried out in the same stations in two (2) periods: in June (beginning of the rainy season) and in September (full rainy season). During the beginning of the rainy season (June), data were collected before any pesticide use in the cotton basin of northern Bénin. These data represent the initial state of biological and physico-chemical parameters. The data were also collected during the rainy season, during the intensive use of pesticides (September), which compared to the data from the first collection, allows to see the effects of agricultural chemical pesticides on the community structure of benthic macroinvertebrates and physico-chemical parameters. During each campaign, two (02) rivers (Alibori and Sota) of the Beninese cotton basin were considered. During the sampling, five (05) streams (Bouli, Irané, Tassiné, Sanson and Yassikoga) tributaries of the Alibori and Sota rivers of the Beninese cotton basin were considered, two (02) streams (Sanson and Yassikoga) tributaries of the Alibori river and three (03) streams (Bouli, Irané, Tassiné) tributaries of the Sota river. At each tributary stream, three (03) sampling stations were determined using land use measurements within a 2 km radius of the stream.

These are: one (01) station upstream of the cotton crops and which serves as a control; one (01) intermediate station with a low land occupation by cotton crops and less than 30% of the land within a radius of 2 km; and one (1) potentially highly impacted station where most of the surrounding land (>50%) is occupied by cotton crops. These stations are determined according to depth (less than or equal to 1m); current speed (low); and turbidity (low), and length (the length of a determined station is equal to 10 times the width of the wetted bed).

Macroinvertebrate sampling method

At each determined station, benthic macroinvertebrates were sampled using a Surber net with a mesh size of 500µm and a metal frame with an area of 1/20 of m^2 i.e. 0.05 m^2. At each sampling site, this Surber net is placed in the bed against the direction of the water flow. Once the Surber is placed, the inside of the metal frame is scraped and washed to a depth of about 0.5 cm and pushed into the net, under the help of the water current, the samples are collected using the net. The sampling consists of eight benthic macroinvertebrate samples from different types of substrates or habitats. At each station, four (04) samples were taken on the dominant substrates and four (04) samples on marginal substrates. The substrates or habitats considered.

Measurement of physico-chemical parameters

Prior to the collection of benthic macroinvertebrate samples, physico-chemical parameters (Temperature, pH, Transparency, Depth, Conductivity and Total Dissolved Solids) were measured in situ in the morning at each study station. The pH was measured with a pH meter (HANNA HI 98107).

The Conductivity, Temperature and TDS were measured with a Conductivimeter (HANNA HI 99300). The water depth was measured with a graduated ruler. The transparency was measured with a Secchi disk. The length and width of the wetted bed were measured with a decameter. The water sample was taken using sterilized boxes and the chemical parameters (ammonia nitrogen and total phosphate) were measured in the laboratory. Finally a GPS, Garmin GPS 72 navigator is used to determine the geographical coordinates of the sampling stations.

Sorting, observation and identification of macroinvertebrates

In the laboratory, the samples were washed and rinsed to reduce waste and sorted station by station. The sorting and observation took place under a binocular magnifying glass. After this operation, the invertebrates were separated according to their morphological appearance and grouped by class, order, and family. The taxonomic determination was made up to the families.

The identification of the macroinvertebrates is done using the following identification keys: The benthic macroinvertebrates of New Caledonian rivers (Mary, 2017), the simple determination key of freshwater macroinvertebrates for the use of the "Petit gardien des rivières" (Leclerc, 2010), the Guide d'identification des principaux macroinvertébrés benthiques d'eau douce du Québec (Moisan, 2010), freshwater invertebrates, systematics, biology, ecology (Tachet *et al.*, 2000) and aquatic entomology (McCafferty, 1981). At the end of the determination of these organisms, they were preserved in the pillboxes containing 70% alcohol.

Data processing

The analysis of data on physico-chemical parameters and benthic macroinvertebrates of the tributary streams of the Alibori and Sota rivers was done through metrics such as taxonomic abundance, taxonomic richness, frequency of observation, Shannon's diversity index and Piélou's equitability index and Hilsenhoff index.

The Shannon diversity index (H') was calculated according to the Shannon and Weaver, (1963) formula: $\mathbf{H'=-\Sigma pi.log2pi}$ (1) with pi the relative abundance of species i in the sample. Piélou's Equitability Index (E) (Pielou, 1969), which is the ratio of true diversity to maximum diversity was calculated by the formula: $\mathbf{E=H'/log2S}$ (2) with S the species richness. The family biotic index (Hilsenhoff index) was used to determine the water quality of the different stations. This index was calculated by multiplying the number of individuals of taxon F (n_i) by its tolerance score (t_i), which is then divided by the total number of macroinvertebrates in the sample (N): $FBI = \frac{\sum_i^f n_i t_i}{N}$, Where F is the family name, n_i is the number of family individuals, t_i is the tolerance value of family i and N is the total number of individuals. The tolerance values for benthic macroinvertebrate families are from Hilsenhoff.

After testing for normality of all biological parameters ($p < 0.05$), variabilities of biological data and physico-chemical parameters were evaluated using the Wilcoxon test at the 5% threshold with R3.4.2 software (R Core, 2021), Rcmd and Factominer Package).The frequency of observation (FO) of the families was determined. It is the ratio between the numbers of stations where the family is present by the total number of stations studied. Three groups were defined (Dajoz, 1985): "very frequent" families have an observation frequency greater than or equal to 50%; "frequent" families have an observation frequency between 25 and 50% and "rare" families have an observation frequency of less than 25%. The Hierarchical clustering was used to group stations based on the similarity of association of macroinvertebrate families. Also, a Canonical Correspondence Analysis (CCA) was performed using Past software (Hammer *et al.*, 2001), in order to match the data of physico-chemical parameters with those of biological parameters obtained during sampling.EPT and EPT/C ratios were calculated before and after pesticide use to determine the impact of pollutants on the structure and diversity of pollutant-sensitive families (Table 1).

Results

Physico-chemical characteristics of water in the Beninese cotton basin

The mean values, standard deviations and coefficients of variation of the physico-chemical parameters of the water at each of the sampling stations in the Alibori and Sota watersheds are presented in Table 2. The values of these physico-chemical parameters recorded at the intermediate stations and those under high agricultural pressure varied significantly between before and after pesticide use (Wilcoxon paired test, $p<0.05$).

Table 1. Interpretation scale for the SurVol Benthos (FBIv3) biological health index (Hilsenhoff, 1988).

Hilsenhoff values (FBI)	Degree of pollution	Degree of organic pollution
0,00 à 3,75	Excellent	No organic pollution
3,76 à 4,25	Very good	Slight organic pollution possible
4,26 à 5,00	Good	Probable organic pollution
5,01 à 5,75	Average	Average: fairly substantial organic pollution
5,76 à 6,50	Somewhat poor	Somewhat poor: substantial organic pollution
6,51 à 7,25	Poor	Bad: very substantial organic pollution
7,26 à 10,00	Very bad	Very bad: severe organic pollution

The analysis of the average values of the physico-chemical parameters showed that the high values of temperature, conductivity, total ammonia and total phosphate and then the low values of pH and transparency were recorded after the use of pesticides especially at the level of the heavily polluted stations.

The coefficients of variation were also higher at the medium and heavily polluted stations than at the reference stations. Among the study stations, the Yassikoga station under the strong agricultural disturbances presented the highest values of temperature, conductivity, total ammonia and total phosphate and then the low values of pH and transparency.

Abundance of macrofaunal order classes collected in the five streams

The main taxa sampled consist of: 95.15% insects, 2.23% glass, 2.03% mollusks, 0.42% hydracarans, and 0.17% Crustaceans before pesticides. While after the use of pesticides, it is constituted by 69,70% of insects, 26,40% of glasses, 2,44% of mollusks, 1,03% of Crustaceans and 0,39% of hydracarians. We note a high abundance of insects and hydracarans and a low abundance of molluscs, worms and Crustaceans before the use of pesticides than after the use of pesticides in the Beninese cotton basin. Of these classes recorded before and after pesticide use, insects were the most dominant at all study stations (Fig. 2).

Table 2. Physicochemical parameters of the waters of the Alibori and Sota rivers.

Rivers	Stations	Season	Temp °C	pH	Transp (cm)	Prof (cm)	Cond (µS/cm)	TDS (mg/l)	NH_4^+ (mg/l)	P_2O_5 (mg/l)	*p-value*	Coeff. var
Bouli	Ref	BP	20,8±0,99	7±0,14	22±1,41	20±1,41	37±3,54	18,5±4,95	0,02	0,05	0.2582	46,26801
		AP	23,75±0,05	6,95±1	16±1,5	21,5±1	53±0,5	26,5±	0,42	0,1		63,0894
	MP	BP	23,65±0,25	6,8±0,21	12±2,83	12±2,83	82±0,71	41±0,71	0,75	0,16	0.0391	65,60392
		AP	25,8±0,71	6,6±1,41	11±1,41	32±0,71	102±2,35	51±0,71	1,05	1,09		81,36037
	HP	BP	26,5±1,56	6,1±0,14	8,5±2,12	19,5±0,71	181±1,41	90,5±1,41	1,15	0,66	0.0065	82,39246
		AP	30,8±2,32	5,1±0,71	6,5±1,41	21±1,41	200±0,71	100±3,50	3,11	1,03		89,92086
Irané	Ref	BP	19,35±0,49	7,1±0,28	24±4,24	34,5±10,61	60±14,14	30±7,07	0,01	0,001	0,0761	61,63637
		AP	22,5±1,22	6,9±0,35	13±1,41	20±0,71	65±0,71	32,5±2,35	0,02	0,12		41,12955
	MP	BP	23±0,71	7,2±0,35	12,5±3,54	13,5±4,95	87±4,24	43,5±2,12	1,05	0,05	0,0144	97,1187
		AP	26,1± 1,14	6,15±0,07	11±1,4	11±1,4	93±1,4	46,5±0,7	2,06	1,16		84,59016
	HP	BP	25,95±0,07	6,6±0,28	7±1,41	10,5±3,54	244±56,66	122±2,82	2,25	0,48	0.008	114,0041
		AP	29,5±0,42	5,3±0,14	5,5±0,70	15±1,41	320±2,83	160±1,41	3,04	1,17	4	102,8555
Tassiné	Ref	BP	22,95±0,21	7,5±0,64	20±9,9	20±9,9	46±1,41	23±0,71	0,075	0,02	0,1271	55,23117
		AP	23±1,41	7±0,41	17±1,41	49,5±30,40	53±1,41	26,5±0,71	0,16	0,20		76,754
	MP	BP	26,1±0,14	6,35±0,21	10±0,00	17±7,07	64±0,71	32±0,71	1,3	0,12	0,003	79,14247
		AP	28,9±1,41	6±0,07	13±1,41	38,5±4,50	77±2,12	38,5±1,41	2,08	0,9	0	70,35809
	HP	BP	29,05±1,48	5,95±0,49	11±15,56	24±15,56	66±4,95	33±2,12	2,12	0,45	0,0101	70,76042
		AP	31,5±2,12	5,4±0,00	7±1,41	15,5±0,71	495±0,71	247,5±0,71	4,01	2,76		98,47917
Sanson	Ref	BP	22,35±0,64	6,75±0,35	27±12,73	44,5±26,16	52±11,31	26±5,66	0,05	0,01	0,225	54,54361
		AP	23,5±2,12	7±0,00	25,5±0,71	13,5±0,71	62±071	31±0,71	0,25	0,35		50,63375
	MP	BP	23,95±0,92	6,15±0,35	17,5±2,12	8,5±2,12	116±11,31	58±5,66	1,10	0,14	0,0076	122,7347
		AP	27,75±0,35	6,85±0,07	12±0,00	14,5±0,54	150±5,66	75±2,83	2,10	0,5		86,52647
	HP	BP	28,15±0,49	6,6±0,28	10±1,41	17±12,73	195±84,15	97,5±42,43	3,25	0,35	0,0096	126,4136
		AP	33,65±0,50	5±0,28	7,5±1,41	14±1,41	304±8,49	152±4,24	4,7	2,12		102,7668
	Ref	BP	21,1±0,42	7,2±1,27	19,5±3,54	16,5±2,12	61±4,24	30,5±2,12	0,10	0,02		53,91103
		AP	22,6±0,85	6,9±0,00	17,5±0,71	29,5±0,71	83±0,71	41,5±0,71	0,14	0,07	0,5326	59,64608
Yassikoga	MP	BP	23,8±0,57	5,15±0,35	14±1,41	16,5±2,12	147±7,07	73,5±3,54	1,27	0,16	0,0011	115,9644
		AP	27,65±0,14	4,9±1,14	11±1,41	11±0,71	287±1,41	143,5±0,71	3,08	1,02		98,36888
	HP	BP	26,5±0,85	5,3±0	12±2,83	14±5,66	227±1,41	113,5±0,71	3,6	0,96	0.0019	123,1588
		AP	35,5±2,12	4±0,42	6,5±0,71	13±2,83	632±3,54	316±2,12	5,03	3,19		112,7547

BP: Before Pesticides, AP: After Pesticides, Ref: Reference, MP: Medium Polluted, HP: Heavily Pollute.

In all study stations, the highest taxonomic abundances of insects were recorded at the reference stations and the lowest abundances were obtained at the stations under cotton pollution.

In the total macrofauna collected, 19 orders of MIBs were recorded. According to the Fig. 2, the comparison of orders according to their contribution to taxonomic richness shows that Diptera, Odonata, Ephemeroptera and Oligochaetes are the most dominant, especially at the reference stations of all study streams. From the results obtained from the study, we note a total absence of plecopterans in the Beninese cotton basin of northern Benin.

Table 3. Observation frequency of benthic macroinvertebrate families collected.

Observation Frequency Intervals	F ≥ 50%		25% ≤ F < 50%		F < 25%	
Seasons	BP	AP	BP	AP	BP	AP
Total taxonomic richness	78	78	78	78	78	78
Relative taxonomic richness	20	15	25	15	33	48
% Frequency of occurrence	25,64	19,23	32,05	19,23	42,31	61,54

BP: Before Pesticides, AP: After Pesticides.

Frequency of occurrence of families of macroinvertebrates collected

The Table 3 presents the classification of macroinvertebrate families collected in the tributary streams of the Alibori River and the Sota River according to the season and their frequency of occurrence. Before the use of pesticides, the very frequent families occupy 25.64% of the total wealth.

Frequent families occupy 32.05% of the total wealth. The rarely frequent families occupy 42.31% of the total wealth. While after the use of pesticides, 19.23% of the total wealth are very frequent families, 19.32% of the total wealth are frequent families and 61.54% of the wealth represented the rarely observed families. It is noted that the majority of the very frequent (F ≥ 50%) and frequent (25% ≤ F < 50%) families were recorded before the treatment of cotton fields with pesticides while the majority of the rare families (F < 25%) were observed after the treatment of cotton fields with agricultural pesticides in the northern Benin cotton basin (Table 4).

Table 4. Frequency of observation of each benthic macroinvertebrate family collected.

F ≥ 50%				25% ≤ F < 50%				F < 25%					
BP		AP		BP		AP		BP		AP			
Hydropsychidae	80	Baetidae	87	Lepidostomatidae	33	Mesoveliidae	26,7	Brachycentridae	7	Brachycentridae	0	Corduliidae	13
Hydroptilidae	60	Caenidae	60	Ephemerellidae	27	Veliidae	33,3	Ecnomi dae	7	Ecnomi dae	0	Platycnemididae	0
Leptoceridae	53	Corixidae	73	Leptophlebiidae	40	Carabidae	26,7	Helicopsychidae	7	Helicopsychidae	0	Mymaridae	7
Baetidae	80	Dytiscidae	87	Potamanthidae	40	Elmidae	40	Philopotamidae	20	Hydropsychidae	7	Pyralidae	7
Caenidae	87	Ceratopogonidae	87	Pléidae	40	Gyrinidae	47	Ephemeridae	20	Hydroptilidae	20	Tridactylidae	13
Corixidae	80	Chironomidae	100	Gerridae	27	Hydraenidae	40	Notonectidae	20	Lepidostomatidae	7	Aphididae	13
Naucoridae	67	Culicidae	60	Elmidae	40	Hydrophilidae	40	Carabidae	13	Leptoceridae	7	Cercopidae	20
Mesoveliidae	53	Simuliidae	80	Gyrinidae	27	Tipulidae	27	Chrysomelidae	7	Philopotamidae	0	Cicadellidae	0
Veliidae	73	Tabanidae	53	Helodidae	27	Sialidae	27	Hydroscaphidae	0	Ephemerellidae	0	Delphacidae	13
Dytiscidae	80	Gomphidae	53	Hydraenidae	27	Noctuidae	27	Heteroceridae	0	Ephemeridae	7	Atyidae	7
Hydrophilidae	60	Lestidae	53	Culicidae	27	Ceropidae	27	Limnebiidae	13	Leptophlebiidae	20	Cambaridae	20
Ceratopogonidae	93	Libelludiae	73	Stratiomyidae	40	Copepodes	33	Spercheidae	0	Potamanthidae	7	Sphaeriidae	0
Chironomidae	100	Oligochètes	100	Cordulegasteridae	47	Physidae	40	Scirtidae	13	Pléidae	7	Unionidae	0
Simuliidae	67	Nemathelmintes	67	Corduliidae	40	Planorbidae	40	Staphylinidae	7	Gerridae	7	Limnaeidae	13
Tabanidae	93	Trombidiformes	60	Pyralidae	40	Glossiphoniidae	33	Noteridae	0	Naucoridae	13	Neritidae	0
Gomphidae	80			Tridactylidae	33			Athericidae	7	Notonectidae	0		
Lestidae	100			Aphididae	40			Dixidae	0	Chrysomelidae	0		
Libelludiae	100			Cercopidae	27			Psychodidae	0	Haliplidae	0		
Oligochètes	73			Cicadellidae	33			Rhagionidae	0	Helodidae	0		
Trombidiformes	67			Copepodes	40			Thaumaleidae	0	Hydroscaphidae	0		
				Unionidae	33			Tipulidae	0	Heteroceridae	7		
				Physidae	47			Platycnemididae	7	Limnebiidae	13		
				Planorbidae	40			Sialidae	0	Spercheidae	20		
				Glossiphoniidae	33			Mymaridae	7	Scirtidae	13		
				Nemathelmintes	27			Delphacidae	20	Staphylinidae	0		
								Ceropidae	13	Noteridae	7		
								Atyidae	0	Athericidae	0		
								Cambaridae	0	Dixidae	0		
								Sphaeriidae	0	Psychodidae	20		
								Limnaeidae	13	Rhagionidae	13		
								Neritidae	13	Stratiomyidae	0		
								Haliplidae	20	Thaumaleidae	13		
								Noctuidae	20	Cordulegasteridae	20		

BP: Before Pesticides, AP: After Pesticides.

Spatial and temporal variation in abundance and taxonomic richness in the Alibori and Sota sub-basins

Abundance (number of individuals) and taxonomic richness (number of families) in the Alibori and Sota sub-basins varied significantly ($p < 0.05\%$) between the pre-pesticide use and post-pesticide use periods in the study area. Across all study stations, the highest taxonomic abundances (number of individuals) were obtained at the reference stations and the lowest were observed at the stations under agricultural chemical pesticide pressure. Among the study stations, the Bouli station showed a higher abundance before the use of pesticides while the Yassikoga station showed a lower abundance during the phytosanitary treatment of agricultural fields. Regarding taxonomic richness, in all study streams, the highest richness was observed in the reference station and the lowest richness was recorded in the stations under agricultural pressure. The Yassikoga reference station showed the highest richness before the phytosanitary treatment, while the lowest taxonomic richness was recorded in the Yassikoga station under agricultural pressure during the period of phytosanitary treatments of agricultural crops by the farmers (Fig. 3).

Table 5. Composition of benthic invertebrate communities under the effect of cotton production.

Indexes	Definition	Predicted response according to the increase of disturbances	Sampling period	Values
%EPT	Percentage of composition in families of Ephemeroptera, Plecoptera, and Trichoptera	Decrease	BP	17,97%
			AP	11,54%
% EPT (abundance)	Percentage of total number (abundance) of Ephemeroptera, Plecoptera and TIichoptera	Decrease	BP	23,79%
			AP	10,82%
% EPT without Hydropsychidae	% of Ephemeroptera, Plecoptera and Trichoptera without Hydropsychidae	Decrease	BP	21,40%
			AP	1075%
% Chironomidae	Percentage of total number of Chironomidae	Increase	BP	35,93 %
			AP	34,22%
Ratio EPT/Chironomidae	Sum of the total number (abundance) of EFA divided by the total number of Chironomidae	Decrease	BP	0,66
			AP	0,32
% of Insects	Percentage of total number of insects	Decrease	BP	95,15%
			AP	69,71%
% d'Insectes non Diptères	Percentage of total number of non-dipteran insects	Decrease	BP	52,94%
			AP	25,47%
% of non-insect taxa	% of total non-insect abundance	Increase	BP	4,85%
			AP	30,29%
% of 2 Dominant taxa	% of total number of 2 taxa (Chironomidae and Oligochaetes) dominant.	Increase	BP	37,85%
			AP	58,93%

BP: Before Pesticides, AP: After Pesticides,

Spatial and temporal variation in Shannon's diversity and Piélou's equitability indices

The Fig. 4 presents the degree of organization of the benthic macroinvertebrate community collected before and after the use of agricultural pesticides. The Shannon diversity and Piélou equitability indices are identical and follow a proportional evolution at all sampling stations. According to this figure, at all 5 study streams, the highest values of these two indices were recorded at the reference stations and the lowest, at the stations under agricultural pollution, in addition, these two indices presented higher values before the use of phytosanitary products than those during the phytosanitary treatment. Among all the study stations, the reference station of Tassiné presented the highest value of Shannon's diversity index (2.91 bits) and Piélou's equitability index (0.82 bits), while the lowest value of these two indices was obtained at the heavily polluted station of Tassiné (0.49 bits for Shannon and 0.21 bits for Piélou).

Spatial and Temporal Variation of the Hilsenhoff Index under Cotton Production

The Fig. 5 shows the spatial and temporal variation of the Hilsenhoff index values for each study station.

Table 6. Percentages of information distributed in a system of dimensions of the axes defined by the ACC.

Axis	Eigenvalue	%
1	0.12759	63.15
2	0.074414	36.83
3	3.2387E-05	0.01603

The lowest Hilsenhoff index values were obtained at the reference stations before the use of pesticides and the highest values were recorded at the stations under cotton pollution. Before the use of pesticides, the reference stations of Irané and Yassikoga presented the lowest values of the Hilsenhoff index (4), while the highest values (10) were obtained at the stations under heavy cotton pollution. Overall, it can be seen that the Hilsenhoff values obtained at the reference stations varied between 4 and 7. These values, compared to the water quality interpretation scales, vary between very good, good, average and poor quality water. As for the values of the Hilsenhoff indexes for the stations under agricultural pollution, they varied between 8 and 10 and correspond to waters of poor and very poor quality.

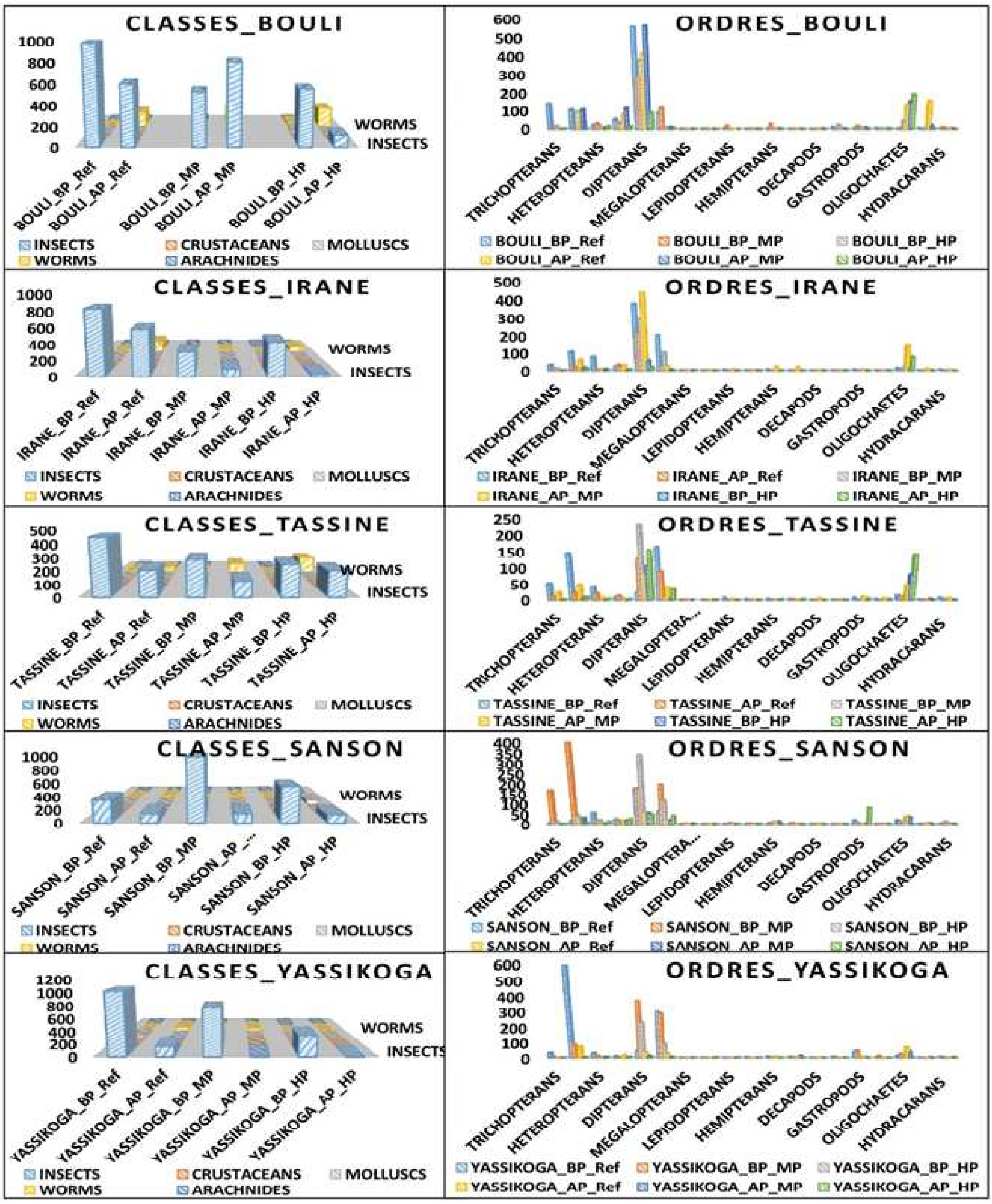

Fig. 2. Abundance of classes and orders of macrofauna collected in the five streams.

BP: Before Pesticides, AP: After Pesticides, Ref: Reference, MP: Medium Polluted, HP: Heavily Pollute.

Indices measuring benthic invertebrate community composition before and after pesticide use

Benthic macroinvertebrate community variables representing pre- and post-pesticide use water quality indices were calculated and presented in Table 5. It is noted that based on the predicted response according to the increase in disturbance, the percentage of EFA, EFA without Hydropsychidae, EFA/Chironomidae ratio, insects and non-dipteran insects is higher before pesticide use than after agricultural pesticide use. While the percentage of the 2 dominant taxa (Chironomidae and Oligochaetes) and that of all non-insect taxa is higher after pesticide use than before pesticide use in the cotton basin of northern Benin. Thus, there is a large difference between the biotic index values obtained before and after pesticide use.

Dendrogram of the sampling stations

This dendrogram shows that the sampling stations are grouped into 6 classes. The first class includes the stations after the use of pesticides, namely the reference and moderately polluted stations of Bouli and the reference station of Irané. The second, third and sixth classes included only the stations before the use of pesticides. The fourth and fifth classes included all the stations after pesticide use, except for the reference station at Tassiné. From the analysis of this dendrogram, we note the overall grouping of stations before and after pesticide use in the North Benin cotton basin (Fig. 6).

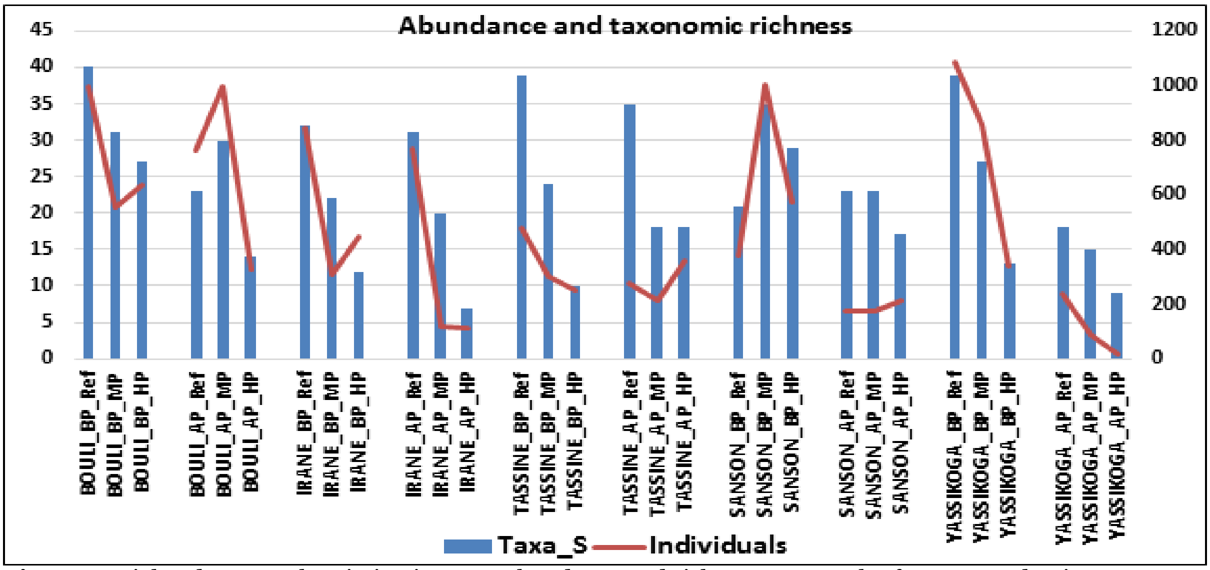

Fig. 3. Spatial and temporal variation in taxon abundance and richness as a result of cotton production. BP: Before Pesticides, AP: After Pesticides, Ref: Reference, MP: Medium Polluted, HP: Heavily Pollute.

Relationships between benthic macroinvertebrates and physico-chemical parameters under the effect of cotton production disturbances

A canonical correspondence analysis (CCA) was carried out between the physico-chemical parameters, the families of benthic macroinvertebrates and the different sampling stations, in order to see the difference between the reference stations and those that were most affected by disturbances of agricultural origin. This analysis reveals that the first two axes express 99% (axis 1: 63.15% and axis 2: 36.83%) of the information (Table 6). Axis 1 is strongly and positively correlated with the high values of transparency and depths. These parameters (transparency and depth) are strongly related to the pre-pesticide stations especially most of the pre-pesticide reference stations. These stations are strongly associated with the families Ephemerellidae, Limnebiidae, Ecnomidae, Platycnemididae, Chrysomelidae, Naucoridae, Corixidae, Brachycentidae and Hydropsychidae (Fig. 7) Axis 2 is positively correlated with high pH, conductivity and TDS values and is associated with the moderately polluted stations from before the use of pesticides, with the exception of the highly polluted station of Bouli and the reference station of Yassikoga. These stations are associated with the families Helicopsychidae, Philopotamidae, Haliplidar, Unionidae, Staphylinidae, Gerridae, Cicadellidae, Libilludae, Baetidae, Aphididae, Helodidae and Hydroptilidae. This same axis strongly and negatively correlated with high values of temperature, ammonia nitrogen ($NH4^+$) and total phosphorus (P_2O_5) and it is associated with all the stations of the post pesticide use. These degraded stations are strongly related to the families Mymaridae, Taumaleidae, Sialidae, Tipulidae, Copepods, Oligochaetes, Nemathelminthes, Chironomidae, Ceratopogonidae, Pleidae and Planorbidae.

It can be seen that these parameters (temperature, ammonia nitrogen and total phosphorus) of axis 2 had the greatest influence on the distribution of benthic macroinvertebrate families recorded in the five tributary streams of the Alibori and Sota sub-basins in northern Benin (Fig 7).

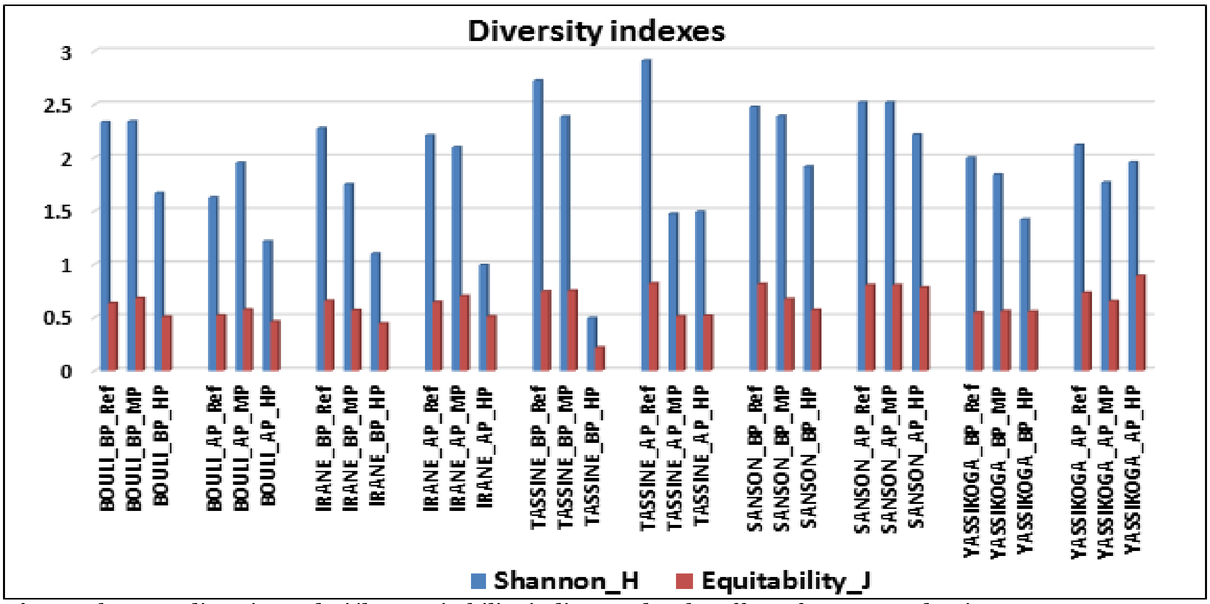

Fig. 4. Shannon diversity and Piélou equitability indices under the effect of cotton production.
BP: Before Pesticides, AP: After Pesticides, Ref: Reference, MP: Medium Polluted, HP: Heavily Polluted.

Discussion

The present study recorded eight physico-chemical parameters (temperature, pH, transparency, depth, conductivity, TDS, total ammonia and total phosphate). All these recorded parameters varied significantly before and after the use of agricultural chemical pesticides (Wilcoxon test, $p < 0.05$).

The average recorded temperature values ranged from 19.35°C to 29.05°C before pesticide use and from 22.5%°C to 35.5°C after pesticide use. The highest temperature values were measured at the stations under high agricultural pressure in Yassikoga and Tassiné. This high water temperature at the highly polluted stations could be explained by the effects of solar radiation on the water due to the deforestation of the river bank for agricultural production. These observations are similar to those of Toumi *et al.*, (2016) who revealed that temperature fluctuations are related to local climatic conditions and more specifically to air temperature and water evaporation phenomena.

The average transparency values recorded varied between 7 cm and 27 cm before the use of pesticides and between 5.5 cm and 25 cm after the use of agricultural pesticides. For both sampling periods, the lowest transparency values were measured at the heavily disturbed Irané and Tassiné stations, while the highest transparency values were observed at the Sanson reference stations. The low transparency values recorded at the stations under agricultural disturbance could be explained by the presence of organic and mineral wastes as well as sludge drained by runoff from the fields into the aquatic environment. These results are similar to those of (Hamid *et al.*, 2014). These authors revealed that the high turbidities are due to suspended solids which represent all the mineral and organic particles contained in the wastewater. These low water transparencies prevent the penetration of sunlight into the water and decreases photosynthesis leading to low dissolved oxygen (O_2) and increased temperature and carbon dioxide (CO_2). (Dimane *et al.*, 2016) revealed that suspended solids modify the turbidity of waters and reduce light penetration thus limiting photosynthesis. The pH expresses the state of acidity or alkalinity of water in relation to the logarithmic scale from 0 to 14. Thus, water with pH = 7 is said to be neutral; water with pH < 7 is said to be acidic; and water with pH > 7 is said to be basic. Overall, the lowest pH values ranged from 4 to 7.50. The low pH values (4 and 5.50) were recorded at the moderately polluted and heavily polluted stations.

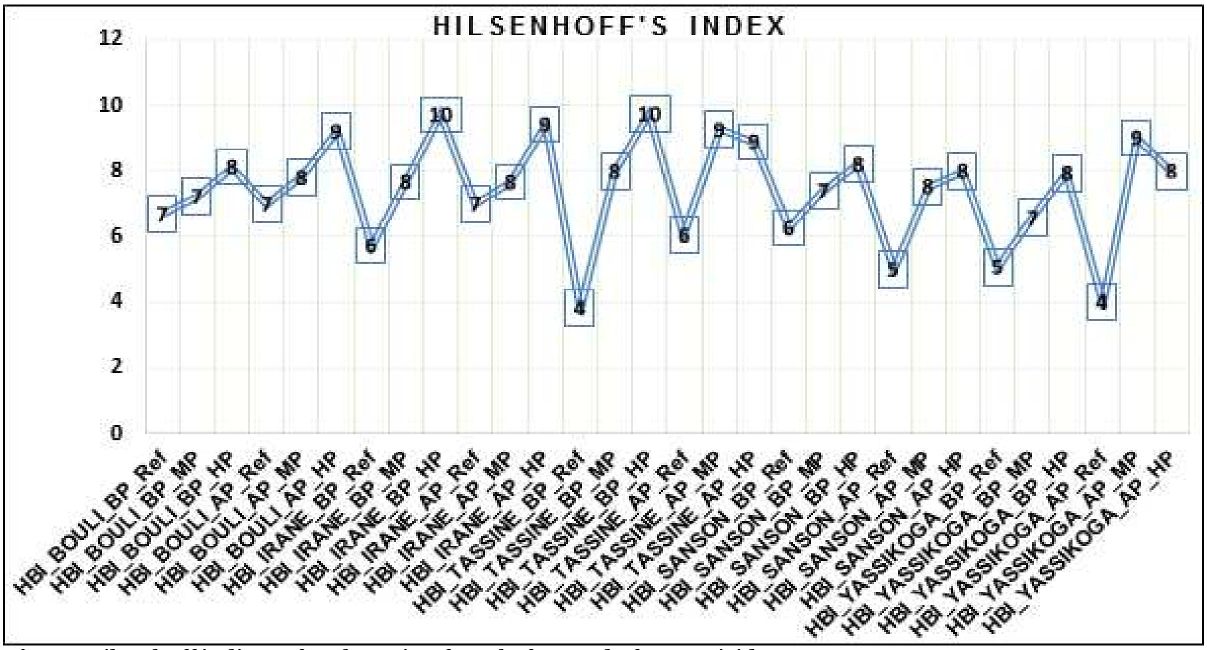

Fig. 5. Hilsenhoff indices of each station from before and after pesticide treatment.

BP: Before Pesticides, AP: After Pesticides, Ref: Reference, MP: Medium Polluted, HP: Heavily Pollute.

These pH values are lower than the norm (6.5 and 8.5) recommended by the Brussels Institute for Environmental Management (IBGE, 2005), which represents the characteristics of waters where life develops optimally. These low pH values could be explained by runoff loaded with more toxic acidic ionic form solutions (Tfeil *et al.*, 2018). The average conductivity and TDS values recorded are proportional and ranged from 37 µS/cm to 632 µS/cm (conductivity) and 18 mg/l to 316 mg/l (TDS) respectively. Electrical conductivity measures the ability of water to conduct electrical current, as most dissolved materials in water are in the form of electrically charged ions (Derwich *et al.*, 2010). The highest values of conductivity and TDS were recorded at stations under heavy agricultural disturbance. High conductivity values resulting in high mineralization. Ammonia nitrogen ($NH4^+$) values ranged from 0.01 mg/l to 2.13 before the use of pesticides to 0.02 mg/l to 3.19 mg/l after the phytosanitary treatments of cotton plants. Based on preliminary criteria used to designate reference stations, $NH4^+$ < 0.75 mg/l (Waite *et al.*, 2000).

The highest values of ammonia nitrogen ($NH4^+$) were recorded at stations receiving high agricultural pressures. Ammonium ions are an excellent indicator of water pollution by organic discharges of agricultural origin (Mihoub, 2017). Total phosphate ($P2O5$) values ranged from 0.001 mg/l to 0.96 mg/l before the period of agricultural pesticide use, while after the application of agricultural pesticides, these phosphate values ranged from 0.07 mg/l to 3.19 mg/l. Preliminary criteria used to designate reference stations indicated that P_2O_5 < 0.02 mg/l (Major *et al.*, 2000). The low values of total phosphate (P_2O_5) were recorded at the stations before the use of pesticides, especially the reference stations. The high P_2O_5 values were obtained at stations under heavy agricultural disturbance. Les valeurs de P_2O_5 des stations de l'après l'utilisation de pesticides sont supérieures à la valeur optimale (0,2 mg/l) de l'Etablissement Public Territorial du Bassin de la Sèvre Nantaise (Moulin de Nid d' Oie). Ainsi les valeurs élevées de P_2O_5 seraient dues à l'utilisation des terres autour des zones fluviales dans le bassin cotonnier béninois. Cette utilisation est principalement destinée aux terres agricoles et pourrait être une explication possible des niveaux élevés de phosphate provenant du ruissèlement pendant l'épandage des engrais comme observé dans cette étude. From the point of view of taxonomic composition, the organisms collected are constituted of 95.15% insects, 2.23% worms, 2.03% mollusks, 0.42% hydracarans and 0.17% Crustaceans before the use of pesticides. Whereas after the use of pesticides, it is constituted by 69,70% of insects, 26,4% of worms, 2,44% of mollusks, 1,03% of Crustaceans, and 0,39% of hydracarians. We note a high abundance of insects and hydracarans and a low abundance of molluscs, worms and crustaceans before the use of pesticides than after the use of pesticides in the Beninese cotton basin. This difference in taxonomic abundance would be due to the effects of cotton production on the diversity of benthic macroinvertebrates in the tributary streams of the Alibori and Sota rivers. The canonical correspondence analysis (CCA) showed in axis 1, a strong positive correlation between high transparency and depth, pre-pesticide use stations especially most of the pre-pesticide use reference stations as well as the families Ephemerellidae, Limnebiidae, Ecnomidae, Platycnemididae, Chrysomelidae, Naucoridae, Corixidae, Brachycentidae and Hydropsychidae.

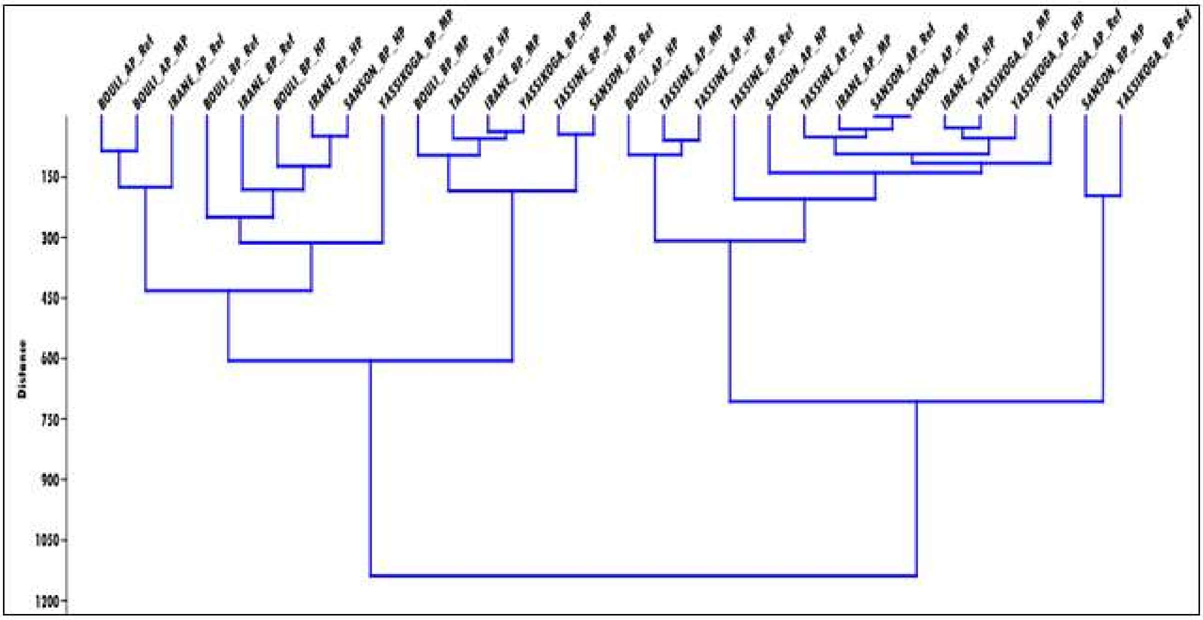

Fig. 6. Dendrogram of the stations of the before and after pesticides treatments.
BP: Before Pesticides, AP: After Pesticides, Ref: Reference, MP: Medium Polluted, HP: Heavily Pollute.

This situation is similar to that of (Moisan and Pelletier, 2008) in Quebec who revealed that in a river, the distribution of macroinvertebrates depends on the amount of incident light, and the transparency of the water.

These stations met the criteria of the Ministry of Sustainable Development, Environment, Wildlife and Parks whose reference stations are those least disturbed with the absence of point source discharge of pollutants. The higher the index score, the more likely the study environment is to be affected by a stressor. The Hilsenhoff index values at the reference stations (4 and 7) compared to the water quality interpretation scales varied between very good, good, fair and somewhat poor.

These good water qualities at the pre-pesticide reference stations will be the cause of the proliferation of the pollution-sensitive families of Ephemeroptera and Trichoptera taxa. The presence of Ephemeroptera would reflect good water quality as these organisms are known to live in well oxygenated and good quality environments (Diomandé *et al.*, 2009). These results were corroborated by the taxonomic richness and the high values of the Shannon and Piélou equitability indices, which reveal that the benthic macroinvertebrate communities in the pre-pesticide use stations, mainly those of the references, are much more diversified, more organized and more stable than those of the stations under high pollution of the post-pesticide use agricultural. Axis 2 is strongly and negatively correlated with high values of temperature, ammonia nitrogen ($NH4^+$) and total phosphorus (P_2O_5) and is associated with all stations of the post-pesticide use. These degraded stations are strongly related to the families Mymaridae, Taumaleidae, Sialidae, Tipulidae, Copepods, Oligochaetes, Nemathelminthes, Chironomidae, Ceratopogonidae, Pleidae and Planorbidae. It is noted that the majority of these families are those resistant to pollution.

The high values of T°C, $NH4^+$ and P_2O_5 at the stations with high pollution would be due to the fertilizers used by farmers in the cotton crops. In surface waters, ammonia nitrogen and total phosphorus come mainly from agricultural land leaching and municipal and industrial wastewater.

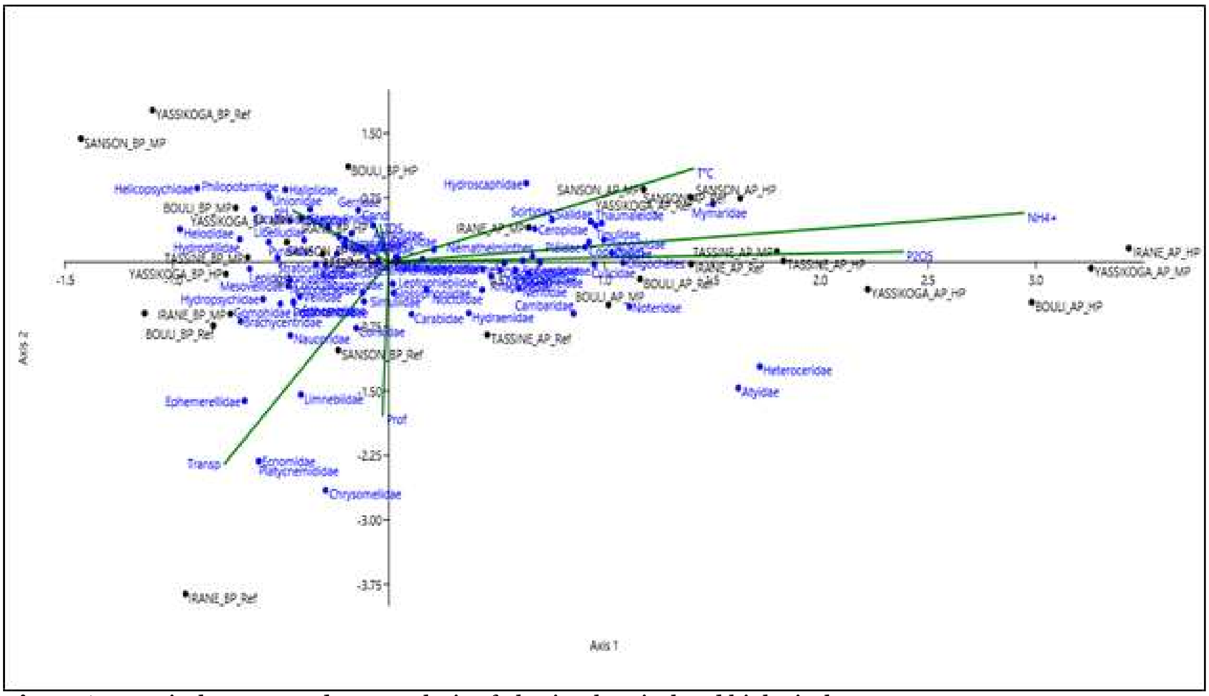

Fig. 7. A canonical correspondence analysis of physicochemical and biological parameters.
BP: Before Pesticides, AP: After Pesticides, Ref: Reference, MP: Medium Polluted, HP: Heavily Pollute.

The high concentration of total phosphorus (P_2O_5) can promote eutrophication through algal blooms, making the water undrinkable and causing a lot of damage to aquatic fauna (Allain, 2001). The pollution tolerance of benthic macroinvertebrates varies according to the taxonomic level of interest (orders, families, genera, etc.); while some are very sensitive to external stressors, others are more tolerant. A pollution tolerance rating is thus associated with each group of benthic macroinvertebrates. This rating is between 0 and 10, with 0 meaning that the individual is intolerant and 10 meaning is very tolerant. As for the Hilsenhoff index values for stations under agricultural pollution, they ranged from 8 to 10 and correspond to poor and very poor water quality. This situation was corroborated on the one hand by the low abundances and taxonomic richness at the polluted stations before the use of agricultural pesticides, and on the other hand by the low values of Shannon's diversity and Piélou's equitability indices at the polluted stations after the use of pesticides. The low values of these two indices could be explained by the fact that the benthic macroinvertebrate communities at the polluted stations after the use of pesticides are poorly structured.

Conclusion

This study used two relatively simple methods (physico-chemical and biological) to analyze the pressure of agricultural activities on the water quality of the five tributary streams of the Alibori and Sota rivers in northern Benin during the period of pre-pesticide use and the period of phytosanitary treatment of cotton crops. The physico-chemical method revealed that temperature, ammoniacal nitrogen and phosphate are parameters that have much more influence on the structure and distribution of benthic macroinvertebrate communities in the five tributary streams of the Alibori and Sota rivers of the Beninese cotton basin. This situation is corroborated by the low taxonomic richness, the low values of the Shannon diversity and Piélou equitability indices as well as the high values of the Hilsenhoff indices after the use of agricultural inputs at the stations exposed to cotton production pressure in northern Benin.

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