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# LONG TERM QUANTITATIVE ECOLOGICAL ASSESSMENT OF INSECTICIDES TREATMENTS IN FOUR AFRICAN RIVERS: A METHODOLOGICAL APPROACH.

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Abstract

In West Africa different insecticides had been applied in selected river areas for the reduction of the blackfly populations vectors of *Onchocerca volvulus*, a parasite causing blindness. To evaluate the possible long term effects of the larvicides on the non target fauna an aquatic monitoring programme has been up from the initial phase of the project. Addressing the attention to the invertebrates data collected in four countries during a maximum period ranging from 1977 to 1996, this paper shows and discusses the data analysis strategy for the measure and interpretation of the biological variation. In particular the application of quantitative ecological analysis methods: Principal Component Analysis, rank abundance models and the community diversity indexes, is critically discussed and comments are given to the ecological interpretation of the results.


## 1. Introduction

*Onchocerca volvulus*, a parasitic worm giving rise to skin reactions and eventually severe ocular lesions followed by blindness [1], had been a major public health problem in many fertile valleys of West African countries. The parasite is transmitted by the female blackfly of the *Simulium damnosum* complex [2] and, being difficult to control the adults, it was decided to destroy with insecticides the vector at its larval stages whose distributions is limited to rapids.

The initial phase of the project (Onchocerciasis Control Programme - OCP) covered a vast area of 764 000 $km^2$ in which up to 18 000 km of rivers had been partly weekly sprayed with larvicides. To prevent flies reinvasion, from 1989 the original programme was expanded to 1 235 000 $km^2$, controlling about 50 000 km of rivers.

Obviously such an extensive and prolonged use of larvicides could have important environmental risks, therefore an aquatic monitoring programme has been settle up from the initial phase of the program to evaluate the possible long-term effects of the larvicides on the non target fauna.

From 1975 to 1985 temephos, chlorphoxim (two organophosphorous compounds) and a biological insecticide, *Bacillus thuringiensis* var. *israelensis* [B.t. H-14] had been the larvicides used.

From 1980, the appearing of certain forest cytotypes of the vector resistant to temephos [3] and to chlorphoxim by 1982 [4] forced the search of new compounds and the implementation of a renewed treatment strategy based on the rotational use of the insecticides with suspension periods. The new compounds were: permethrin ( pyrethroid), carbosulfan (carbamate), pyraclofos (organo-phosphorus) and vectron ( pseudo-pyrethroid). After the papers of Lévêque et al. and Yaméogo et al. [5,6] that provided a comprehensive evaluation of the first ten years monitoring of fish and non target insects populations, the biological data collected within a wide area till 1998 allow to face new questions arising from the implementation of the OCP.

It is now possible a better evaluation of: i) the long-term changes of the invertebrate and fish populations with respect to their taxonomic composition as well as to their trophic structures, ii) the severity of each specific larvicide used during the programme, iii) the resistance of the communities to the induced stresses and iv) their recovery capacity.

Approaching the analysis of the invertebrate data collected in four countries during a period ranging from 1977 to 1996 for addressing the above mentioned questions, this paper presents and discusses the data collections and the applied quantitative and qualitative numerical methods of analysis used in ecology, with the major objective of commenting their different contribution and the complementary informations they provide to the evaluation and comprehension of the data variation.

Examples of the application of the discussed methods are presented, while the detailed results of the analyses will be published in a forthcoming paper which include fish data.

| RIVER | STATION | COUNTRY | MAXIMUM SAMPLING PERIOD |
|---|---|---|---|
| Entomokro | Danangoro | Ivory Coast | Dec. '77 - Feb. '96 |
| Pru | Asubende | Ghana | Gen. '80 - Apr. 95 |
| Niandan | Sansambaya | Guinea | Dec. '84 - Apr. '94 |
| Kaba | Outamba | Sierra Leone | Mar. '89 - Apr. '94 |

**Table 1: location of the sampling stations and maximum sampling periods.**

2. Available data and sampling methods

The study addresses the invertebrates collected during the dry season in four rivers located in West Africa during the time extents and within the sampling stations indicated in table 1. The rivers are savanna type with a water regime characterised by high discharges from July to November and a low water period from January to June, details on hydrological and physicochemical charactristics of the rivers are reported in Iltis and Lévêque [7] and in Moniod *et al.* [8].

The organisms were sampled, by means of Surber net and as day and night drift, during the application of the larvicides as well as during suspension and pre-treatment periods. Details of monitoring and sampling methods can be found in Lévêque et al. [9] and Dejoux et al. [10].

The three sampling strategies, Surber samples, day and night drift, had been employed in order to collect biological data showing different informations.

Night drift, which is supposed to be mainly voluntary, reflects an active period while the number of the day drift organisms is related with their health condition.

As regards the use of the Surber net, this technique allows to sample, in a quantitative way, the organisms living in specific river areas and for this reason the collections reflect the structure of the benthic communities, where this term indicate an assemblage of interacting individuals sharing at the same time the same space. Obviously this concept of community is less applicable to the drift samples which represent collections of organism turning up from a wider area located upstream the sampling site and thus mainly controlled by factors external than those related to the sample location.

Because of this possible source of bias the presented examples of the numerical methods have been applied to the Surber samples.

All the sampled individuals were classified according to their taxonomic level as well as their trophic role, this second classification method being based on the association between a limited set of feeding adaptations found in freshwater invertebrates and their basic nutritional resource categories: gathering and filtering collectors, predators, shredders and scrapers [11]. To avoid the presence of rare taxa only the principal systematic units belonging to the Ephemeroptera, Trichoptera and. Chironomidae were used for the analyses.

## 3. Aims and Protocols

The data analysis strategy has proceeded along two relatively distinct paths, the first considered the evaluation of the changes in the invertebrate taxonomic and functional structures, the second has been oriented towards the examination of the main fractions of the taxonomic variation by means of multivariate techniques of analysis.

On the basis of the results of a preliminary data inspection, the following analysis techniques were selected for the study.

- *Invertebrate taxonomic diversity.* Three aspects of the taxonomic diversity have been estimated: taxa richness, the distribution of the individuals among those taxa (equitability or evenness) and the heterogeneity, a measure that encompasses the first two diversity measures. The nonparametric indexes utilised were, respectively, the Margalef richness index: (S-1)/ln N, the Pielou evenness index: H'/H'max and the Shannon heterogeneity index: $H' = -\sum p_i \ln (p_i)$.

For the representation of the invertebrate taxonomic structure, occurring during the pre-treatment, treatment and suspension periods, the median values of the indexes was preferred to the average values, for that the median values are less outliner sensitive.

- *Relative abundance of the functional groups.* The relative abundance of the invertebrates classified as functional groups were estimates for each treatment period. On the fact that the trophic structure is a property of the living organisms emerging at community level, only the Surber samples have been used for this analysis.

- *Rank abundance models.* This graph-approach to the analysis of the biological structures consists in a conventional form of presenting the importance of each taxon as abundance ($y$-axis) with the different taxa concerned arranged in rank order along the $x$-axis from the commonest to the rarest. The pattern of the line connecting the taxa of each sample allows a visual examination of the invertebrates structures: S-shaped curves are related to high heterogeneity values, on the contrary high slopes indicate more dominated structures. The comparison of the curves permits to detect the changes that can take place in the invertebrates structures during the different sampling periods.

In the example graphs the species abundance are represented by means of the median values occurring during the pre-treatment, treatment and suspension periods.

- *Multivariate analysis.* Because of the linear response of the invertebrates abundance Principal Components Analysis (PCA) was preferred to the unimodal multivariate analysis approaches (i.e. Correspondence Analysis sensu ter Braak [12]). The PCA was performed to the log-transformed abundance of the taxa collected by means of Surber net.

The analysis has been applied separately to each river communities; an all rivers data matrix has not been analysed because a preliminary ordination of this matrix showed a wide variation in the invertebrate communities among the rivers.

## 4. *Discussion and Conclusions*

For the selection of the appropriate analysis methodology, the following problems, other than the different sources of bias detailed by Lévêque [5] were considered:

i) insufficiencies of pre-treatments samples and replicates that constrained the use of non parametric analyses;

ii) the incremental biological variation because of the rotational use of the larvicides;

iii) the presence of outliner values and

iv) the rivers peculiarities that outlines the importance of considering the different antropic pressure that have been taking place, with different time, in the treated rivers.

It has to be firstly noted that the use of different analysis methods on different biological informations (taxonomical and functional) allows itself to face the possible source of bias because of the problems previously listed. Considering that no standard analysis procedures are available for answering the questions addressed by the monitoring programme or, more in general, in the long-term impact assessment studies, the use of different analytic techniques applied on different biological aspects are necessary to corroborate a comprehensive evaluation of the biological data.

Among the questions posed in the introduction are the measure of the biological variation because of the larviciding and the evaluation of the severity of each specific larvicide. For addressing these questions the commonly used analysis is PCA (or equivalent ones depending of the data property i.e. Correspondence Analysis for biological variation following unimodal pattern). In Fig. 1 an example of its application to Niandan samples is shown. PCA detects two biological gradients oriented 45° with respect the first two principal components. The first gradient explains the biological variation occurring because of the treatments irrespectively of the applied larvicide (bold arrow in the figure), the second one is related to the biological variation specifically related to each larvicide (dotted arrow in the figure), roughly opposing the data sampled during B.t. treatment and the ones sampled during the application of permethrin.

While these gradients demonstrate both a general effect of the larviciding and a different response of the communities to the applied larvicides, it is however difficult from the ordination diagram to define the severity of each specific larviciding (i.e. the taxonomic structures most affected are the ones related to the use of B.t. or to permethrin?).

Fig 2a answer to this question, clearly showing the previous severity gradient with the additional graphic information of the levels of the taxonomic structural changes related to each larvicide. Normally structured communities, at equilibrium, generally are composed by taxa having similar abundance and when these are plotted according to

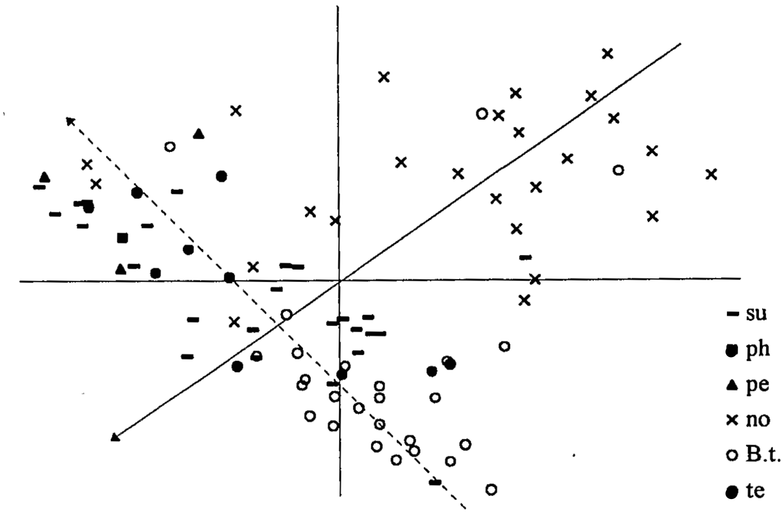


Fig 1. Scatterplot of the samples position according the first two components of the PCA explaining, respectively 43 and 22 % of the total data variation. The bold and the dotted arrows show, respectively, the biological variation because of the treatments and the biological variation related to each specific larvicide. Su = suspension, ph = phoxim, pe = permethrin, no = pre-treatment, B.t. = *Bacillus thuringiensis*, te = temephos.

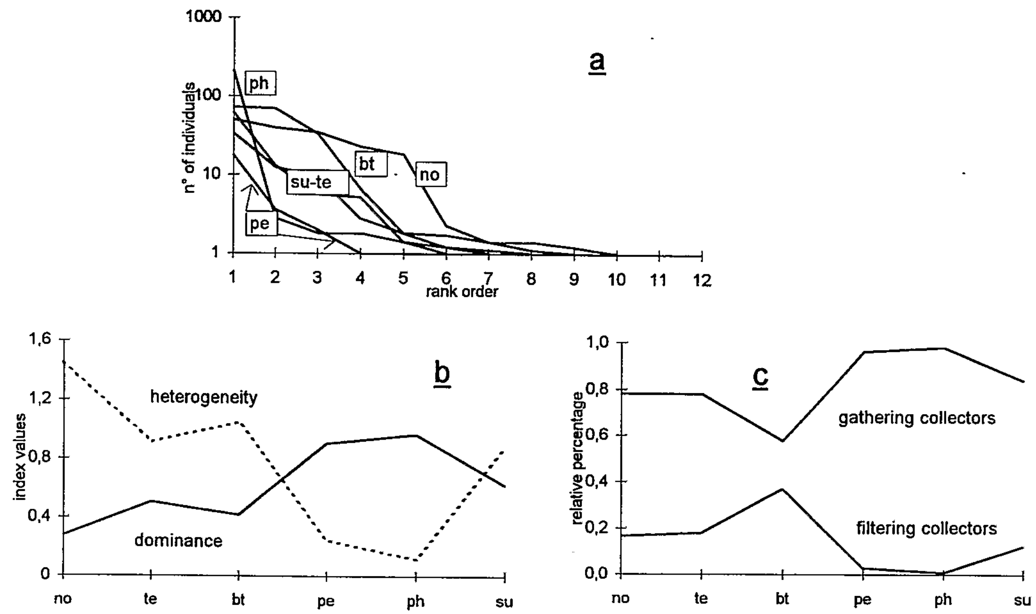


Fig 2. a) rank abundance models showing the taxonomic structure of the invertebrates communities sampled during the different treatments; b) diversity indexes variation according the treatments and c) relative percentage variation of the two most abundant functional guilds. Labels as in Fig. 1.

their decreasing abundance, the resulting curve is characterised by a clear "plateau". This S-shaped pattern, that in ecological literature is named "broken stick" [13], is clearly evident for the communities sampled during the pre-treatment periods (Fig. 2a) and can be used as reference to compare the communities structure models of the treatment periods.

From the visual examination of the rank abundance models it is possible to suggest that the biological communities sampled during the application of permethrin present the highest taxonomical changes, both in terms of abundance (y-axis) and taxa richness (x-axis). On the contrary the model of the invertebrates communities sampled during the use of B.t. shows a pattern similar to the one of the pre-treatment samples. This information it can be now of help in answering to the question about the direction of the severity gradient outlined by the PCA (dotted line).

Similar communities response is corroborated by the diversity indexes (Fig. 2b) while Fig. 2c, showing the variation of the two dominant trophic guilds, adds further informations. From this figure a different biological response occurring during B.t. treatment is evident, compared to permethrin and phoxim that it is not clearly outlined by the previous analyses. With reference to the pre-treatment observations, during B.t. treatment, the gathering collectors show a reduction in their relative abundance, on the contrary during the use of permethrin and phoxim the filtering collectors result more affected. This different response of the invertebrate trophic structures can be partially justified by the larvicide properties and the feeding habits of the invertebrates; for example the gathering collectors feed on surface deposit and thus can be more affected by B.t. because to its rapid adsorption to soil particles [14].

Considering the third question, the resistance of the communities to the larvicides is measurable, with reference to the pre-treatment data, by the bold arrow in the PCA ordination diagram and by the slope of the rank abundance models.

Finally the invertebrates' resilience, can be addressed by inspecting the communities structure variation occurring during the suspension periods. For this, all the analyses applied show that during these suspension periods the communities still present changes in their taxonomic structures; it has to be noted however that these changes do not affect the relative abundance of the trophic guilds.

In choosing the analysis strategy, we adopted the concept of "significant ecological change" used by OCP in assessing the ecological impact of larviciding [9]. Whitin the mandate of the Ecological Group two criteria were indicated "the vector control activities should not reduce the number of invertebrate species, nor causing a marked shift in the relative abundance of species" and "temporary and seasonal variations in invertebrate populations other then Simulium could be accepted."

For this we think that, other than considering the social benefit of having oncho free valley in the operational area, the range of biological variation that would normally occur in these river systems should be taken in account. In fact in the natural situation these river invertebrate communities would rarely be in equilibrium (constant in taxonomic and trophic composition) because of the natural stresses, like drought and spate events, which would occur with great frequency and regularity. When these factors are considered, it can be suggested that the biological variation outlined in the discussed examples can be positively considered and it can be concluded that the OCP has produced limited damages and never caused irreversible canges on the non-target invertebrate populations.

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