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Nota/Note

SOLF – System for data acquisition in olfactometry bioassays

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SUMMARY

Olfactometry bioassays are useful for studying the behavioral responses of insects to volatiles of different sources: host or non-host plants, other insects or to synthetic blends. Data collection must reflect the insect behavior as it is affected by the sources of odor. The software SOLF (System for data acquisition in olfactometry bioassays) was developed at Vegetal Ecophysiology Laboratory – "Embrapa Mandioca e Fruticultura" as a demand of a project titled "Biological control and alternative technologies for the management of the citrus psyllid", part of the research activities inserted in an arrangement of Embrapa focused at the Huanglongbing citrus management. This software is applied to collect and manage the results obtained in olfactometry bioassays conducted with *Diaphorina citri* Kuwayama and also with other insect species. The routine of SOLF was developed in Visual Basic for Application (VBA) language version 7.0. The excel spreadsheet interacts with VBA in order to recognize and record each numeric keyboard typed. As response variables, the software provides information on residence time and number of entries. Before the implementation of SOLF, data collection in olfactometry bioassays was manually transcript in lab notebooks. The main benefits of SOLF are the automatization and optimization of data acquisition process during the olfactometry bioassays, which turns the acquisition of data faster, more efficient and less subject to error, leading to a better control of the process.

Index terms: software, VOC's, insect behavior.

SOLF – Sistema para aquisição de dados em bioensaios de olfatometria

RESUMO

Os bioensaios de olfatometria são úteis para estudar as respostas comportamentais de insetos a voláteis de diferentes fontes: plantas hospedeiras ou não-hospedeiras, outros insetos ou misturas sintéticas. A coleta de dados deve refletir o comportamento do inseto, pois é afetada pelas fontes de odor. O *software* SOLF (Sistema para aquisição de dados em bioensaios de olfatometria) foi desenvolvido no Laboratório de Ecofisiologia Vegetal - "Embrapa Mandioca e Fruticultura" como uma demanda de um projeto intitulado "Controle biológico e tecnologias alternativas para o gerenciamento do psílideo dos citros", parte das atividades de pesquisa inseridas em um arranjo da Embrapa focado no gerenciamento do *huanglongbing*. Este *software* é aplicado para coletar e gerenciar os resultados obtidos em bioensaios de olfatometria realizados com *Diaphorina citri*

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Kuwayama e também com outras espécies de insetos. A rotina do SOLF foi desenvolvida no Visual Basic for Application (VBA) versão 7.0. A planilha do Excel interage com o VBA para reconhecer e gravar cada teclado numérico digitado. Como variáveis de resposta, o *software* fornece informações sobre o tempo de residência e o número de entradas. Antes da implementação do SOLF, a coleta de dados em bioensaios de olfatometria foi transcrita manualmente em cadernos de laboratório. Os principais benefícios do SOLF são a automatização e otimização do processo de aquisição de dados durante os bioensaios de olfatometria, o que torna a aquisição de dados mais rápida, eficiente e menos sujeita a erros, levando a um melhor controle do processo.

Termos de indexação: *software*, VOC´s, comportamento de insetos.

Diaphorina citri Kuayama, 1908 (Hemiptera, Liviidae) is the vector of Huanglongbing (HLB), the most severe citrus disease worldwide (Halbert & Manjunath, 2004). As there is no known cure for this disease, HLB management is mostly depending on the vector control (Belasque et al., 2010).

The manipulation of insect behavior is considered one of the most promising strategies for pest management (Pickett et al., 1997; Turlings & Ton, 2006; Pickett et al., 2007; Li et al., 2009; Pickett, 2012). Plant attractants and repellents can be used to repel insect pests or attract and keep natural enemies in the agroecosystem (Birkett & Pickett, 2003; Pickett et al., 2006, Bruce et al., 2005; Webster et al., 2008; Li et al., 2009; Padmaja et al., 2010).

Studies on chemical ecology has providing some good results on sustainable methods for pest management through application of push-pull systems and conservative biological control (Cook et al., 2007; Sandhu et al., 2010; Yan et al., 2015). These studies are useful for getting data on insect preferences when different sources of odor or synthetic blends are compared. Behavioral assays can be used for evaluating the effects of chemical compounds found in host plants and explain the role they can play on *D. citri* attraction, repellency, settling or growth (Robbins et al., 2012).

Data acquisition usually is performed using expensive systems or through manual collection and transcription of data in laboratory notebooks, which may cause some errors that interfere with the results of the experiments.

The objective of this work was to develop and evaluate a software for data acquisition and data management as an innovative methodology in olfactometry bioassays conducted for *D. citri*.

The system for data acquisition in olfactometry bioassays (SOLF) was developed at Vegetal Ecophysiology Laboratory – "Embrapa Mandioca e Fruticultura". It was first demanded for the project titled "Biological control and alternative technologies for the management of the citrus psyllid". This project is part of the research activities inserted in an arrangement of Embrapa focused at the Huanglongbing citrus management. Although this program has been developed for applying to bioassays involving *D. citri*, it can also be used for other insects.

For *D. citri*, a multiple choice olfactometer has been used (Figure 1A), but the system can be applied to two-arm olfactometers. The insect is inserted through a small hole in the central area of the olfactometer, called non-response area (area 5). The number of arms treated (areas 1 to 4) depends on the type of the bioassay. Charcoal-filtered air is pushed through the odor source chambers in direction to the olfactometer (Figure 1B). The flow rate was kept at 0.25 L/min at each arm.

The procedure used for *D. citri*, once the insect is inserted in the olfactometer, is to record its position systematically for a time interval previously determined (10 minutes in our conditions). It must be considered that the insect can move very fast inside the olfactometer (Figure 1A), requiring attention from the observer.

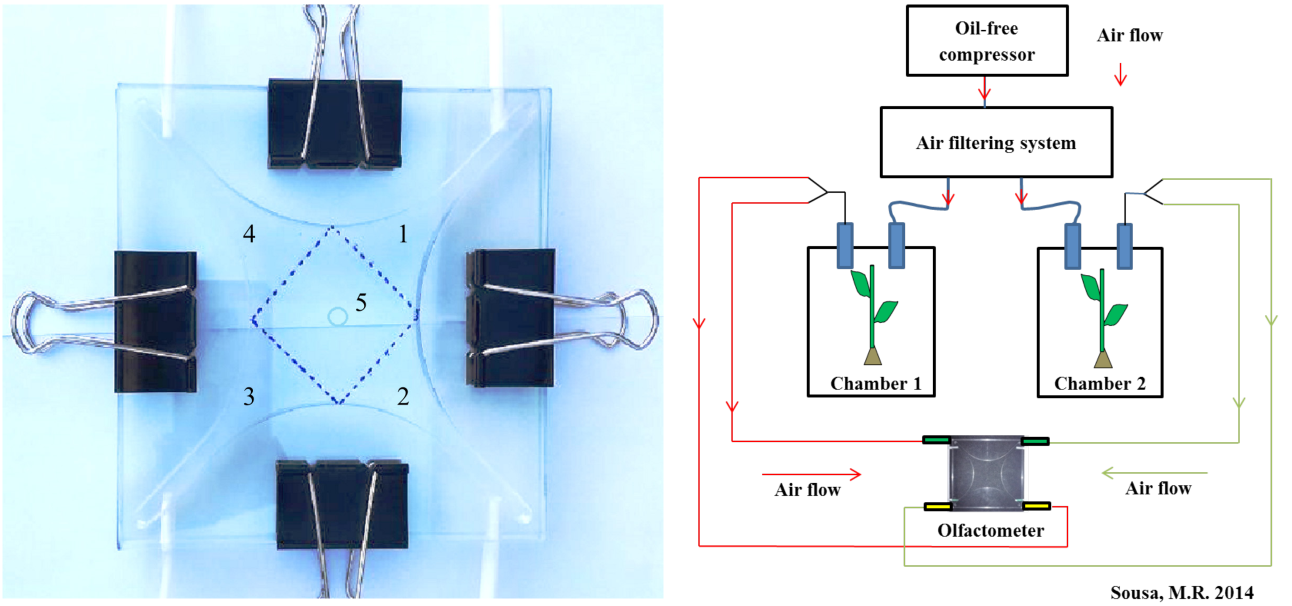
The time spent and the number of entries in the different areas of the olfactometer by *D. citri* are the variables usually collected in olfactometry bioassays.

The previous system (before software implementation) was based on manual registration of data obtained in olfactometry bioassays on a laboratory notebook followed by typing them in a computer spreadsheet for processing the results. This approach was time-demanding for processing the results. Besides that, as the data were manually registered and transferred to an electronic spreadsheet, the process became vulnerable to human error.

SOLF's routine was developed in Visual Basic for Application (VBA) language version 7.0. The excel spreadsheet interacts with VBA in order to recognize and record each numeric key pressed on the keyboard.

The input variables are the title of the experiment and structure of the bioassays, order of bioassays and treatments evaluated. As response variables, the software provides information on residence time, number of entries and final choice.

Every time the insect moves out of an area, a numeric key is typed according the new area to which it moved.



(A) (B)

Figure 1. A four-arm olfactometer used in the olfactometry bioassays (A). Diagram of the olfactometry system used at "Embrapa Mandioca e Fruticultura" (B).

The elapsed time between each numeric character typed is counted in milliseconds and represents the amount of time that the insect stays in any part of arena. After 10 minutes (or another interval set) the routine is finished and all records are reported in another spreadsheet for further analysis.

The first screen presented by the software basically provides information on the experiment and a regressive counter (Figure 2). When the start button is pressed, two spreadsheets are automatically fullfilled. One of them shows the steps followed by the insect (Figure 3) and the other presents the summary for the response variables (Figure 4).

The results released by the software are presented as a summarized report of the experiment (Figure 4), allowing a classification according to the treatments evaluated and a previous analysis of them, by using statistical tools available in the spreadsheet.

Thus, SOLF implementation promotes the following benefits: i) innovative automatization and optimization of data acquisition process in olfactometry bioassays; ii) lower probability of occurrence of errors due to manual data processing, ensuring the real responses of insect behavior; iii) accessibility to data in electronic spreadsheets with a standard dataset output for further analysis; iv) higher control of the process.

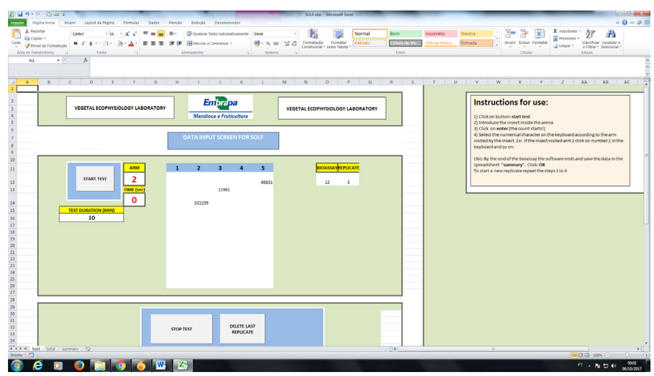


Figure 2. Initial screen for data acquisition in olfactometry bioassays.

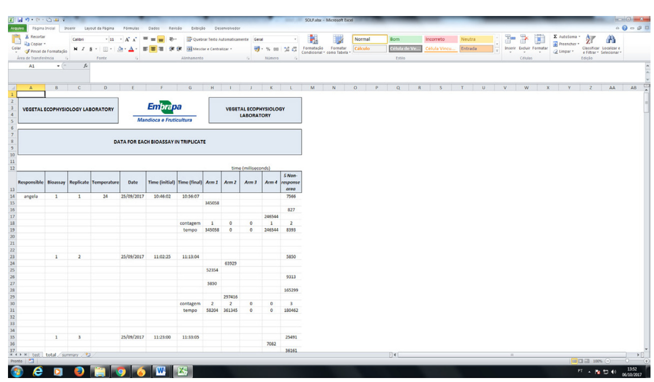


Figure 3. General report for data obtained in olfactometry bioassays through SOLF.

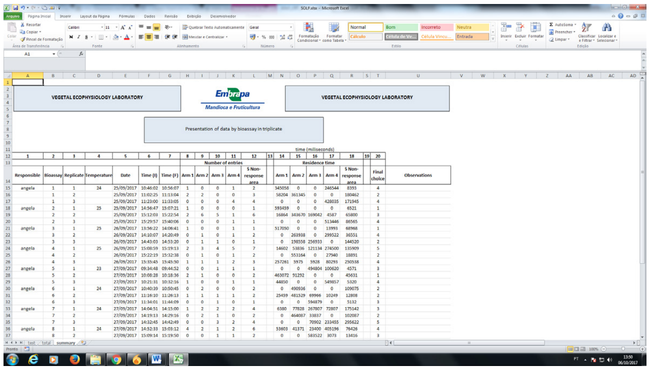


Figure 4. Summarized report for response variables generated in olfactometry bioassays through SOLF.

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