EDUCATIONAL MATERIAL

Protocol for collecting VOCs using the headspace collecting device

PURPEST - Plant pest prevention through technology-guided monitoring and site-specific control

Collaborative Project (RIA Research and Innovation action)

HORIZON EUROPE CALL – HORIZON-CL6-2021-FARM2FORK-01

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Summary

This document briefly describes the conditioning and preparing the materials for VOCs sampling using the Headspace collecting device. This educational material, developed as part of **the PurPest project** under Work Package 1 (WP1), has been extended and edited to provide a clear and practical protocol for VOC collection, enabling researchers, practitioners, and students to effectively use the device.

Acknowledgment goes to the authors, Ali Karimi and Jürgen Gross of Julius Kühn-Institut whose expertise transformed a complex process into an accessible and replicable protocol. This work, as part of the broader PurPest initiative, reinforces our commitment to advance knowledge and tools that support sustainable farming practices.

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1 INTRODUCTION TO THE HEADSPACE COLLECTION DEVICE

The headspace collecting device (HSCD), Fragrance Collector FSP-2246F, was developed by Jürgen Gross [Julius Kühn-Institut (JKI), Dossenheim, Germany] in collaboration with Ralf Kunath (FLUSYS GmbH, Offenbach, Germany). This device was specifically produced as a limited series for the PurPest project (European Commission, 2023). The HSCD is designed for the precise collection of volatile organic compounds (VOCs) in various environmental conditions, making it suitable for field sampling. The device is robust, can be transported, for example, as a piece of luggage in an aircraft, on a tractor or the loading area of a pick-up truck and is intended for outdoor use. The HSCD can be operated from a 240 V socket or, e.g. for outdoor use, easily by using a portable power station.

The HSCD is a derivative of a prototype formerly described (Rid et al. 2016; Gross et al. 2019) and consists of six fragrance collection systems ("channels") mounted in parallel, each consisting of a suction pump (KNF Neuberger GmbH, Freiburg, Germany) coupled to a digital mass flow meter (detector and controller; Bronkhorst High Tech B.V., AK Ruurlo, The Netherlands) with a measuring range of 0.1 to 1 ln/min air (Karimi and Gross, 2024), which are mounted in a trolley suitcase, connected by tubes, and wired electrically (Figures 1.1 A, B). To prevent cross contamination, there are no connections between single channels. The HSCD is adjusted to collect VOCs in a wide range of temperature (5 to 40°C). The programming of both flow rate and total collected air volume of each single channel can be done using a digital control unit with a LCD touch screen allowing to switch on or off each channel individually. Flow rate [ln/min], volume [ln], volatile load [%], temperature [°C], humidity [%], date, and time of sampling, together with all status indicator are displayed in real time (Karimi and Gross, 2024).

Furthermore, the user can select between two sampling methods, the open loop sampling (OLS) and the closed loop sampling (CLS) mode (Figure 1.1 C). An important difference between the two sampling modes is that if oven bags are used in OLS mode, they can shrink, which can cause injury to the plants. This does not happen in CLS mode. Using the 12 mechanical interfaces in the front plate of the device, tubes can just connect to the six input (suction) interfaces (OLS) or additionally to the output interfaces (CLS). For odor collection, each channel of the HSCD must be connected by mechanical interfaces to an air filter cartridge (ICAF 2X6, Sigma Scientific, Micanopy, USA) using Teflon tubing to remove VOCs from ambient air, and the stream of the new purified air is then passed over a suitable vessel containing the insect or plant sample and from there to a sample tube on which the VOCs are collected (Karimi and Gross, 2024).

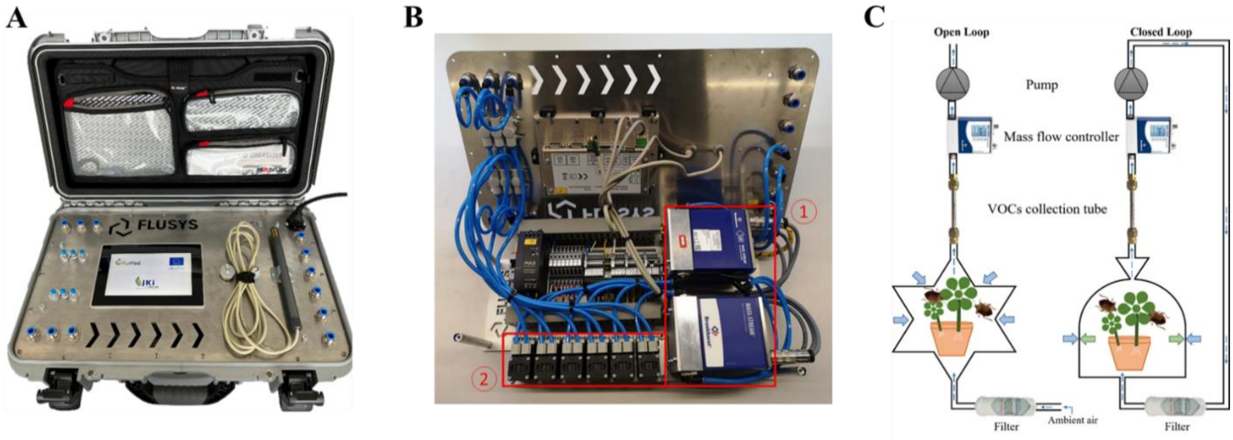


Figure 1.1: Portable digital 6-channel headspace collecting device (HSCD) for collecting VOCs in standardized manner. In the center is the touch screen, each left and right are six mechanical interfaces (blue). To the right of the screen are the sensors for temperature and rH on an external probe, which are connected to the device with a 2 m cable (A). Device opened to show the mass flow meters (1), suction pumps (2), tubing and wiring (B). The circuit diagrams of open loop system (OLS) and closed loop system (CLS) for sampling (C) (Karimi and Gross, 2024).

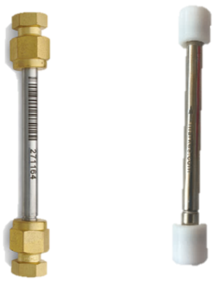
1.1 HEADSPACE SAMPLING

To detect and identify plant-pest specific VOCs, single branches of plants or individual pests are wrapped in oven plastic bags or glass bottles as described by Rid et al. (2016). Using clean air filter cartridges, stream of ambient air are purified and pumped, controlled by the HSCD, through each bag/bottle until it reached the final volume. Volatile organic compounds from headspace sampling are trapped in stainless steel, prepacked sample tubes with Tenax TA35/60 sorbent (Markes, Neu-Isenburg, Germany). Karimi and Gross (2024) reported how to sample VOCs by the HSCD and the chemical analysis of the sampled VOCs using an automated thermal desorption system (ATD) connected to a gas chromatography–mass spectrometry (GC–MS).

2 MATERIAL FOR COLLECTION VOCS

Materials and devices which need to connect the headspace collecting device pieces are listed in Table 2.1 and Table 2.2.

Table 2.1: Materials which are needed for collection VOCs.

Materials	Information	Figure
Teflon Tube	Teflon® tubes (6 mm and 8 mm) for connecting of the individual parts. **6 mm tube:** inside 4 mm and outside 6 mm diameter **8 mm tube:** inside 6 mm and outside 8 mm diameter	
Sample tubes	Tubes filled with sorbent (e.g., with Tenax® TA35/60; appropriate for many common plant volatiles) with Teflon®-, coated brass compression caps (e.g., Swagelok, PerkinElmer). Volatiles are trapped on stainless steel, prepacked sample tubes containing 200-mg Tenax TA35/60 sorbent (Markes, Neu-Isenburg, Germany).	
Opener	The opener is used to open coated brass compression caps.	
Clean air filter cartridges	Using filter cartridges (ICAF 2X6, Sigma Scientific, Micanopy, USA), ambient air is purified, and stream with defined air flow through the bag until having reached a specified final volume.	
Oven plastic bag	Bags made of polyethylene terephthalate (PET) (e.g., Toppits®, Melitta, Minden, Germany).	

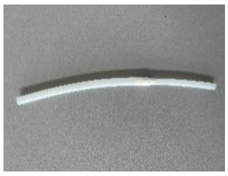
Glass bottle	For in vivo experiments glass bottles with the polybutylene terephthalate (PBT) high temperature caps can be used. These delivery caps have a GL45 thread, can be dry heat sterilized at up to 250°C.	
PTFE Thread Seal Tape	It is used to connect Teflon tubes (6 and 8 mm), most effective on high-pressure lines and temperature (120 °C).	 

Table 2.2: The auxiliary devices which are needed for collection VOCs.

Materials	**Information**	**Figure**
External power supply	An external power supply is needed for field experiment. For example, a power station with a 1000 Wh capacity will provide approximately 30 hours of runtime for the HSCD.	Yard Force Power Station LX  PS1200
Oven	Use for cleaning oven plastic bags, Teflon tubes, glasses, etc. Depend on what material is needed to be clean, temperature is different (e.g. plastic bag at 60 °C and glass bottle at 120 °C).	
Condition Oven	TurboMatrix TC 220 Condition Oven (max oven temperature of 400 °C) is used to condition the Tenax tubes.	

3 PREPARING AND CONDITIONING THE MATERIALS

3.1 Teflon Tubes

For collecting VOCs with the HSCD, Teflon® tubes (6 mm and 8 mm) are needed. In the closed loop system (Figure 3.1), six long 8 mm Teflon tubes (ca. 2 meters) are used to connect the HSCD inputs to the Tenax Tubes and then Tenax Tubes are connected to the sampling oven bags and/or glass bottles. In addition, twelve Teflon tubes (consist of 6 and 8 mm Teflon tubes which are sealed together by PTFE Thread Seal Tape; Figure 3.2) are required, which six of them are used to connect the HSCD output (8 mm) to the filter cartridges (6 mm) and the other six tubes for connecting the filter cartridges (6 mm) to the sampling oven bags or bottles (8 mm). In the open loop system, there is no need to connect the HSCD output to the filters.
Before sampling VOCs, Teflon tubes should be cleaned. In this case, inside of Teflon tubes should be rinsed by Ethanol 70% and then heat them in an oven at 120 °C for at least 6 h, ensuring the complete evaporation of the solvent (Figure 3.3).

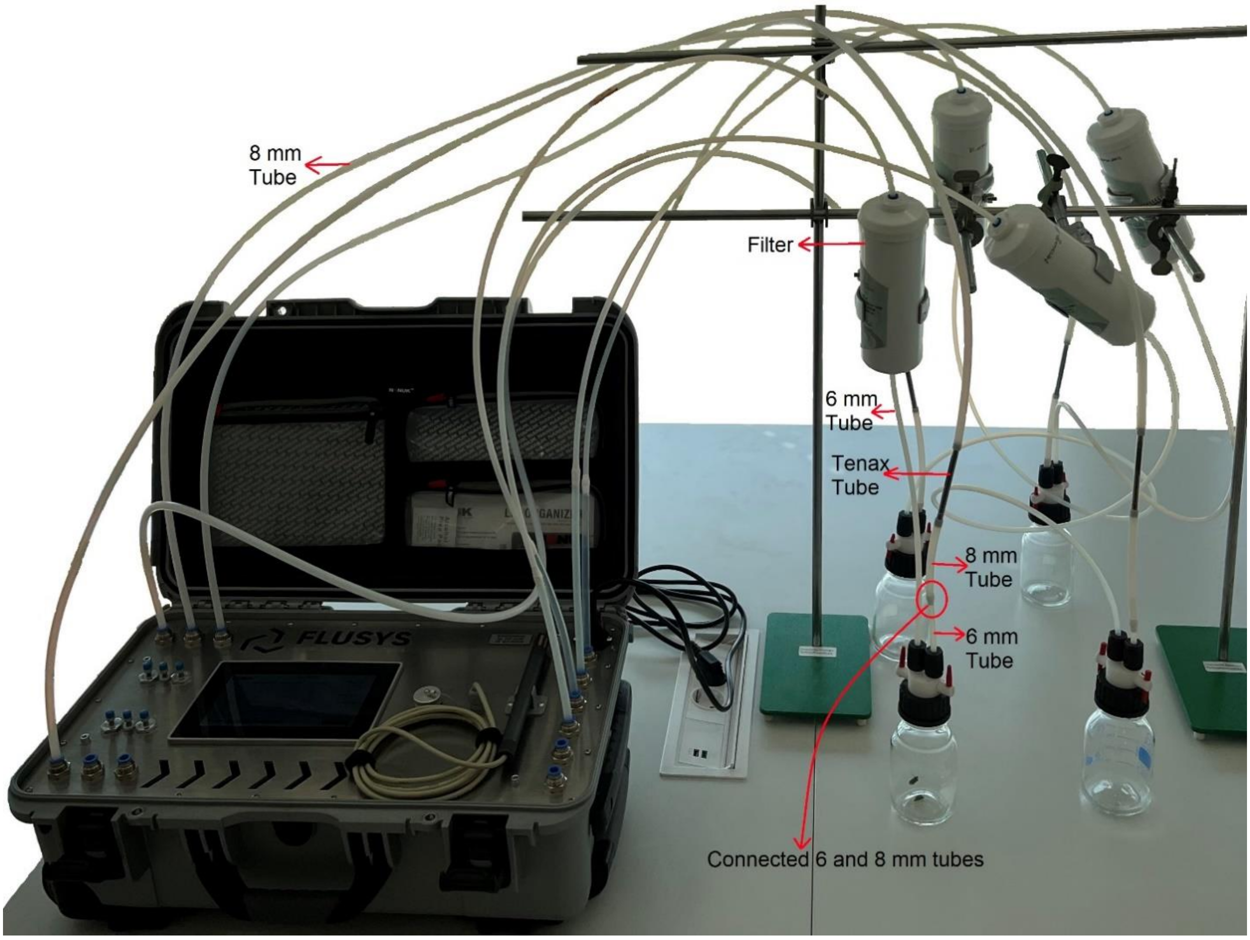


Figure 3.1: Preparing the headspace collecting device for collecting VOCs in CLS mode.

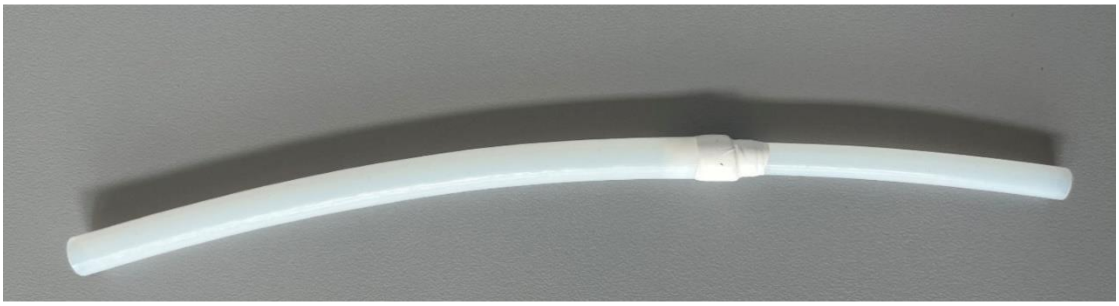


Figure 3.2: Connected 6 and 8 mm Teflon tubes together.

Figure 3.3: Cleaned Teflon tubes using an Oven.

3.2 Sample tubes (Tenax)

Before sampling VOCs (max. 2 week), the sample tubes should be conditioned using the condition Oven (TurboMatrix TC 220, max oven temperature of 400 °C). The Tenax tubes should be conditioned at 280 °C for 45 min. For conditioning the Tenax tubes, the gas (nitrogen) should be passed through the tubes from the top, which minimizes chance for tube contamination. After conditioning the tubes as well as sampling VOCs, the Tenax tubes should be sealed with coated brass compression caps (Figure 3.4).

Figure 3.4: Tenax tubes sealed with coated brass compression caps.

3.2.1 Teflon®-, coated brass compression caps

3.2.1.1 Coated brass compression caps

First, unscrew the caps (Figure 3.5), and then the caps should be sonicated in isopropanol for 15 min. After removing isopropanol, the caps are sonicated again in methanol for 15 min. After sonication, methanol should be removed, and let the caps under the clean bench to dryness for 15 min. After dryness, the caps should be screwed together again, placed in petri dishes and with the lid open, heated in an oven for at least 1 h at 60 °C. The solvents, isopropanol and methanol, can be recollected and used at least 5 times to clean the caps.

Figure 3.5: Unscrewed Coated brass compression caps (right). The unscrewed caps screwed together again, and prepared for heating in an oven (left).

3.2.1.2 Teflon caps

For regular use, Teflon caps should be rinsed with ethanol 70 % for a few minutes, placed on a petri dish and then heat them for at least 1 h at 100 °C using an oven (Figure 3.6).

Figure 3.6: The rinsed Teflon caps with ethanol 70 %, preparing for heating in an oven.

3.3 Oven plastic bag and Glass bottle

To collect VOCs, parts of or whole target plants should be wrapped in oven plastic bags made of PET. The oven bags are sealed using cable ties.

Before using the plastic bag, they should be heated in an oven for at least 6 h at 60 °C (Figure 3.7). First, cut the bag into the exact size that is needed (e.g. plant branches, approximately 50-60 cm, depending on the size of the branches), and before putting them in oven, the bags should be opened, because the inside of the bags should be cleaned by the heated air. The plastic bag is used just once for sampling.

Glass bottles are useable many times. However, after each trial, the bottles and their caps should be rinsed by Ethanol 70% and then heat them in an oven at 120 °C for at least 6 h.

Figure 3.7: The cleaned plastic bag using an oven.

4 HINTS

4.1) To avoid contamination, the Teflon tubes, oven bags, glasses, etc. should be brought out from the oven a few minutes before the experiment.

4.2) Samples should be analyzed within two weeks of collection using a thermal desorption device (TD) connected to a GC-MS (gas chromatograph mass spectrometer).

4.3) Before starting GC-MS analysis, internal standard (1 µl) should be injected into the Tenax tubes directly.

4.4) The coated brass compression caps of Tenax tubes should be replaced by the Teflon caps (Figure 4.1) when preparing the tubes for GC-MS analysis.

Figure 4.1: Tenax tubes sealed with Teflon caps.

5 REFERENCE

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