

Review of the dissertation thesis

## Chemical composition and in vitro antibacterial effects of vapours of essential oils from plants recommended by the European Medicines Agency against respiratory infections

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Lower respiratory tract infections are still significant threats to humans, especially children. Antibiotics have reduced the number of deaths, but their effectiveness is decreasing due to the rise of resistance. Also, the method of their application is not without problems. For these reasons, using volatile antimicrobials that can be directly inhaled appears to be a prospective method. The thesis combines this idea with the preselection of essential oils based on the inclusion of their monographs in the European Pharmacopoeia or EU list of traditional herbal medicines by the European Medicines Agency. This is a smart approach because legislative and regulation aspects always play a crucial role in the final evaluation of the potential product.

The present work is a traditionally formatted thesis. The thesis is written clearly and logically. For the **Introduction** part, I have only two comments:

1. In chapter 2.2.5.2 EO chemical composition, there could be a note on the sulphur-containing compounds. They are often overlooked because they are not marketed in the fragrance industry, I guess. However, for antibacterial applications, they have particular potential.
2. I am afraid that the regulatory aspect is not correctly described. There is a difference between the Pharmacopoeia monograph and the EMA herbal monograph (chapter 2.3.).

**Hypothesis, research questions and Objectives** are clear, as well as most parts of the **Material and Methods**. I have two minor comments for the GC-MS and Headspace analysis parts.

1. The concentration of the injected sample in hexane was 20 mg/ml. Even though the split ratio was 1:30, the system could be overloaded. That is also visible on the chromatograms in Figure 9, where peaks are tailing and are not separated on the baseline.

2. The sampling by SPME or GTS could be done in heated conditions to minimize cold zones prone to condensation. That could also be partially the reason for the unusually low content of some compounds detected.

From the **Results and Discussion** part it is obvious that the student has studied the 328 references well because he exhaustively compared them with his results. However, there are some discrepancies. As I understand, two distillations were done, probably with different plant materials, because the EO yields from the same suppliers are very different, see chapter 7.1. vs 7.3.1. If the first batch was used for the antibacterial tests and the second one for the chemical analyses, there is a risk that the composition is not the same. On the other hand, the results of different desorption media and sampling techniques are interesting, showing also how tricky the analysis of volatiles is.

In terms of language and typography, there is almost nothing to improve. The vast majority of the text is correct. I recommend using non-breaking space between numbers and units, placing figure captions below the figures, and writing the names of chemical individuals without the first capital letter.

Overall, the work is of a high quality, with a substantial degree of novelty and invention. Hopefully, the results will help to increase the use of plant-derived compounds in respiratory infection treatment.

In my opinion, the thesis meets the requirements of the Czech University of Life Science Prague, and I recommend its acceptance for defence.

2

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In Prague 08/01/2024

  
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