AMICBASE – Essential Oils

Supplementary Information

Alexander Pauli

ReviewScience, 90513 Zirndorf, Fürther Str. 13, Germany
December 2006

Essential oils are commonly a mixture of lipophilic compounds belonging to the class of terpenes, aliphatic and aromatic compounds mainly. Analyses have been shown that many of them contain more than 100 individual compounds, and therefore, it is difficult to attribute specific properties of the entire oils to the presence or absence of a single compound.

Although essential oils are prominent for their use in phyto- and aromatherapy as beneficial agents to treat human disorders, their main use is their industrial utilization for food flavouring and the processing of raw materials for the chemical industry. Due to the ability of essential oils to volatize at room temperature they are used as fragrants especially in perfumes or are taken as a source for valuable fragrant compounds.

Medicinal Uses of Essential Oils

In pharmacopoeias of European countries essential oils are accepted and recommended, e.g. for treatment of catarrhal diseases in children. In 1991 in every Indian state, about one-third of the governmental medical posts are occupied by physicians who belong to the traditional systems. The Indian physician Sushruta reported 600 BC the medicinal use of essential oil containing plants for treatment of infections of the mouth, jaw and teeth. Traditional practitioners, medicinal plants and herbal medicines are accepted in large sections of the population in developing countries and public interest had also increased in industrialized countries during the last decades. In 1998 the World Health Organization summarized the regulatory situation on plants used in traditional medicine of 52 countries (1).

Essential Oils vs. Drugs

In laboratory testing the antimicrobial activity of essential oils is much lower than of pharmaceutical drugs that are designed to treat severe human infectious diseases. Especially, the minimal inhibitory concentration (MIC) of essential oils, which is the most precise measure of antimicrobial activity, is about 1.000 to 1.000.000 fold lower than obtained with

antibacterial and antifungal drugs. By these facts it is hard to explain why some essential oils are successful in treating and healing infections. One explanation can be that essential oils reach locally high concentrations, e.g. when given onto skin in pure form, or adsorb and accumulate in tissue that contains lipophilic material, e.g. in the lungs after inhalation. Another, frequently given explanation is the occurrence of synergistic effects. Their existence has been already demonstrated in laboratory experiments, e.g. the antispasmodic action of pure eugenol (ca. 80% in clove oil) is lower than of clove oil, which contains the eugenol-synergist acetyl eugenol. One can suggest that mechanism-based processes are responsible for such synergistic effects, however, it seems to be impossible to examine all possible interactions in only one essential oil that contains over 100 different compounds.

(1) A. Pauli: Anticandidal Low Molecular Compounds from Higher Plants with Special Reference to Compounds from Essential Oils, Med. Res. Rev. 26, 223-68 (2006)

Database AMICBASE – Essential Oils

The database AMICBASE – Essential Oils schedules growth inhibitory properties of essential oils towards microorganisms. It is running in the MA Windows environment and is available as MS Excel table plus as MS Access database (all trademarks of Microsoft Corp.). The database field description is given next:

Plants

Plant Material Tested: Essential oil, powder or extract
Common Names: Common plant names
Scientific Name: Name officially used
Main Components: Data on chemical composition

Microorganism

Bacteria: yes/no field
Molds: yes/no field
Yeast: yes/no field

Test Method

SDT - yes/no field: Serial dilution test: microorganisms were cultivated in liquid growth medium; resulting inhibitory data: MIC, MMC, NIC.
ADlT - yes/no field: Agar dilution test: microorganisms were cultivated on agar surface; resulting inhibitory data: MIC.
ADT - yes/no field: Agar diffusion test: growth inhibitory compounds cause a zone with no growth; resulting inhibitory data: inhibition zone (mm).
Other - yes/no field: Other methods not listed above
Annotation: Annotations on test results

Inhibitory Data

MIC: Minimal Inhibitory Concentration in mg/l (ppm): the concentration causing no growth of microorganisms - occurrence of growth is usually determined with the naked eye.
MMC: Minimal Microbicidal Concentration in mg/l: the concentration causing no growth of microorganisms - growth was controlled in subcultures.

PC: Phenol coefficient: this is an index used in older literature, that describes the activity relative to phenol (PC of phenol = 1)
NIC: Non-Inhibitory Concentration in mg/l: the concentration at which a microbial strain survived - given among 'Annotation'.
PC: Phenol coefficient: this calculates the inhibitory activity of a test compound relative to phenol.

Evaluation

The evaluations being done in this field represent relative numbers only. Comparison of results obtained with different testing methods cannot be compared without errors. Evaluated data obtained with the same testing method yield most reliable data.

0 = no growth inhibitory activity
1 = weak activity against few microorganisms
2 = slight growth inhibitory activity
3 = moderate growth inhibitory activity
4 = strong growth inhibitory activity

Literature

Author, Title, Journal: Citation of the data source
Reference: Number of reference

Types of Inhibitory Data

Antimicrobial inhibitory data alone are not the complete information in the judging of the therapeutic value of an essential oil. Further questions have to be answered, first of all aspects related to toxicity and allergenic potential. Antimicrobial inhibitory data are useful in answering questions in technical subject areas: Which dose is required to stabilize a product against microbial decay ? To what extent contributes an added amount of essential oil to the stability of a given product ?

Agar diffusion test (ADT)

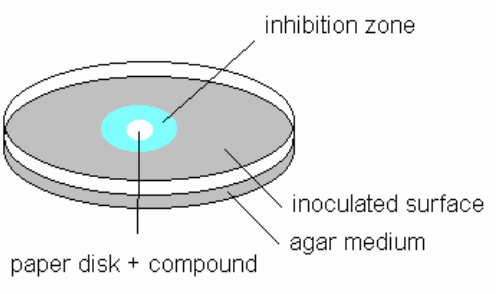

Figure I. Model of the agar diffusion test

In the agar diffusion test a sample (essential oil, solved or pure compound) is given to a reservoir (hole or paper disk), while microorganisms were cultivated on top of the agar surface. After a defined time an inhibition zone may be formed, if first a given compound fulfils two preconditions, which are first the ability to diffuse through the agar medium and second the compound possesses growth inhibitory properties towards the cultivated microbial species, respectively. In other words: compounds that do not diffuse through the agar medium do not show any antimicrobial properties in this test method. This is the case with e.g. farnesol, which forms no inhibition zone but strongly inhibits growth of e.g. *Bacillus subtilis* in other test systems.

Results obtained by this method are inhibition zones in millimetres. In the evaluation field of AMICBASE – Essential Oil database results of the most active oils were characterized with the number 4. Oils causing smaller inhibition zones with the numbers 3 and 2 and such oils which inhibit to a small extent or do not show any inhibitory activity were characterized with

1 and 0. The evaluation has been done for every publication separately, and therefore, a comparison of the data is critical.

Agar dilution test (ADlT)

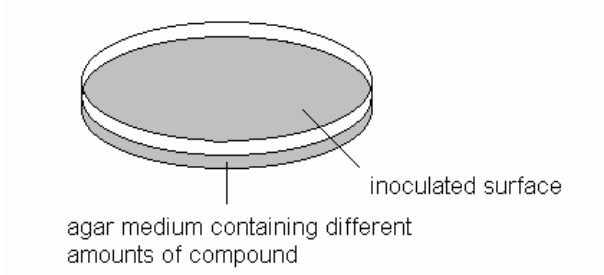

Figure II. Model of the agar dilution test

In the agar dilution test a sample is incorporated in the agar medium and microorganism are grown onto the agar surface. The minimum concentration causing absence of growth on the agar surface after defined time characterizes the minimal inhibitory concentration (MIC in mg/l).

Serial dilution test (SDT)

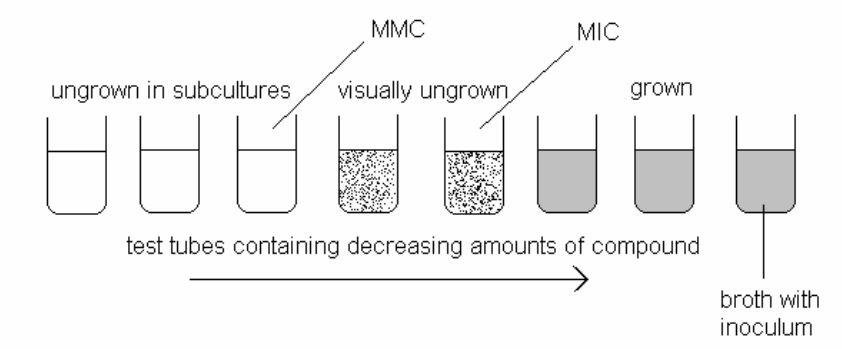

Figure III. Model of the serial dilution test

In the serial dilution test the essential oil sample (requires addition alcohol or detergents) and microbial cells are added to the liquid culture medium. The minimum concentration causing absence of growth in the culture medium after defined time characterizes the minimal inhibitory concentration (MIC in mg/l). Absence of growth is determined with the naked eye.

Additionally, this method allows the determination of the minimum microbicidal concentration (MMC in mg/ml), which characterizes the concentration causing the complete absence of surviving microbial cells. It is determined in subcultures taken from visually clear tubes.