A DESIGN STRATEGY FOR COMPREHENSIVE QUALITY CONTROL OF BOTANICALS: A CASE STUDY WITH ESSENTIAL OILS

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ABSTRACT

Essential oils have been valued by many cultures for centuries. These volatile and complex hydrophobic compounds have been used for a wide variety of cosmetic and therapeutic purposes including traditional medicine, biocides, fragrances and food additives. The benefits of essential oils are centered on their functions in nature, though there are natural variations in composition and quality due to growth conditions, climate, harvest conditions, and plant parts selected. Extraction methods and storage conditions, such as exposure to oxygen, temperature and light, can also affect the integrity of essential oils. There are established standards based on analytical monographs for some essential oils. For other oils without this standard, it is vital to understand and establish identity, purity, and potency to establish reliable quality and prevent adulteration or misidentifying the oils. In order to assess and maintain reliable quality of essential oils, the quality standards were developed utilizing different evaluation methods. One of the most utilized analytical methods for assessing the quality of essential oils is GC-MS (gas chromatography-mass spectrometry), which is used to identify different components present within samples. From the chromatographic profiles, key components were selectively analyzed further to identify compounds of interest. For example, in some samples of jasmine oil, cinnamaldehyde eluted at the same retention time as methyl jasmonate. The presence of this compound in addition to the lack of other compounds demonstrated that those sample oils were of a lower quality. Methyl jasmonate is a key indicator found in high quality jasmine oils. In other examples, the essential oil was found to contain undesirable components, such as pesticides, or phototoxins. In addition to chemical analysis methods, additional safety and organoleptic assessments are fundamental to developing quality products. There are 6 key steps to developing quality products: selection, sourcing, structure, standardization, safety and substantiation. This stepwise process is great in enabling us to make our product safe and effective.

INTRODUCTION

Botanical extracts have been used for various cosmetic and therapeutic purposes. For example, ginger extracts are used to treat stomach upset, or nausea as well as soothing and calming skin irritation. Even though there are many examples of use throughout the history, the effects of plants on human physiology are poorly understood. Botanical extracts are complex mixture of different constituents with natural variations. The composition of botanical extracts could vary based on the growth conditions, climate and harvest conditions. In addition, extraction methods and storage conditions could alter the composition. It is important to set a standard for botanical extracts to maintain the quality and safety of the product.

Essential oils are special extracts from plant parts. They are mixtures of volatile compounds that produce distinct and characteristic smells. Traditionally, essential oils were used by indigenous people to improve their mood, alertness, or as medicine. There are many varieties of essential oils as well as different chemical constituents with various plant species. Most essential oils are produced by steam distillation and cold pressing, although there are some exceptions. In the case of jasmine, solvent extraction methods are used instead, due to damage caused by high-temperature steam distillation, which would be detrimental to the delicate jasmine flowers and oils. Selecting the right ingredients from trusted sources will yield quality and consistent raw materials (1).

Essential oils can be obtained from different parts of aromatic plants. For example, bark, stems, seeds, roots, flowers, as well as leaves, can be used to collect fragrant volatile oils. The major constituents of essential oils are divided into two groups: terpenes/terpenoids and aromatic/aliphatic compounds, such as citronellol and eugenol (1). Within each oil, the amounts and specific constituents change based on climate conditions, soil types, or even harvest conditions. Seasonal changes will affect the yield of essential oils. These volatile compounds are produced by the plants to function as signaling molecules, defensive mechanisms, or responses to environmental cues.

There are standard methods to evaluate the quality of raw materials in dietary supplements (5). With different botanical extracts, we could follow similar guidelines to standardize botanical extracts used in cosmetic products as well. Safe and effective products begin with quality raw materials. Appropriate evaluation of raw materials on their quality will improve the efficacy and safety of finished products, in this case, essential oils.

METHODS

Gas Chromatography-Mass Spectrometry (GC-MS)

Gas chromatography is an analytical instrument to separate different chemical substances. Separate compounds are ionized further in the mass spectrometer to identify specific components of essential oils matching the structural information to library spectra. Various essential oils were obtained from suppliers, and diluted 1:20 in chloroform. GC-MS analysis were performed on Agilent 7890B gas chromatograph (Santa Clara, CA), equipped with a split/splitless inlet in combination with an Agilent 7000C GC/MS triple quad. Compounds are fragmented by electron ionization (EI) to generate spectra, which is compared to library spectra using MassHunter Workstation (Agilent).

GC-MS Conditions:

Column:	HP-5MS UI (0.25mm x 30m x 0.25µm)
	Carrier Gas type: Helium
	Mode: Constant flow at 1 mL/min
Injection:	Mode: Split
	Initial Temp: 250 °C
	Split Ratio: 50:1
	Injection volume: 1.0 µl
Detection:	Transfer line: 300 °C
	MS in scan mode (40-400amu)
	Solvent delay: 3.75 min
Oven:	Initial temp: 60 °C
	Ramp: 3°C/min to 240 °C
	Total run time: 60 min

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4. Contact Dermatitis 63: 277-283, 2010

RESULTS

Raw material selection plays a critical role in the quality control process. Lavender essential oils are obtained from *Lavandula angustifolia* flower spikes. However, another lavender hybrid, lavandin (*Lavandula x intermedia*) produces more oils from bigger flowers. There are some differences between lavender and lavandin. Lavandin essential oils have higher amounts of camphor (7-12%), whereas lavender oils usually have less than 1%. Camphor is used for anti-itch or other treatments, but FDA limits the concentration of camphor to be less than 11% in the consumer products. Lavandin oil is less expensive, so some lavender raw materials are mixed with lavandin. Figure 1 shows the chromatograms from selected lavender oils and alternate raw materials which contains higher amounts of camphor.

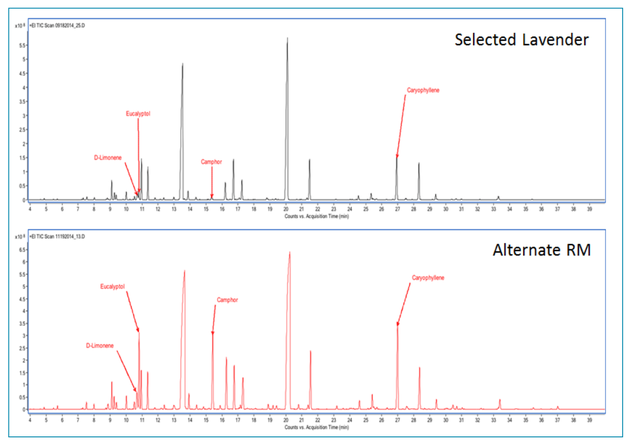
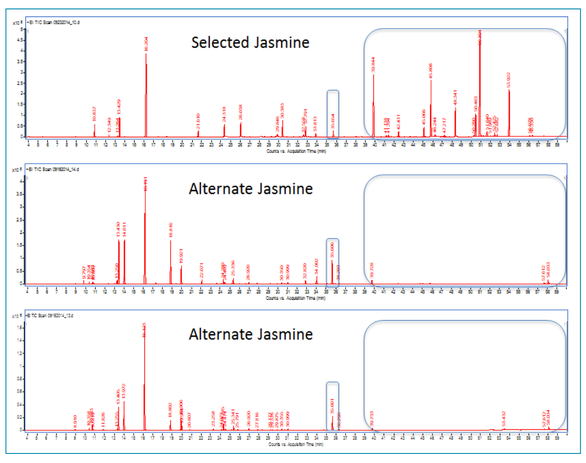
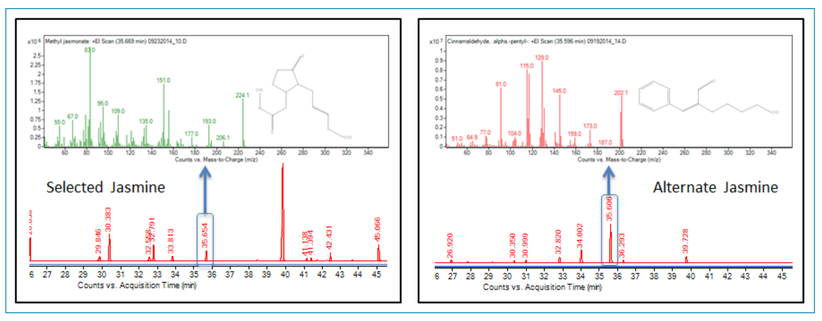


Figure 1: Lavender raw material selection. Gas chromatograms show the different lavender raw materials. The retention time for camphor is around 15.4 min. The selected lavender raw material has a barely detectable peak for camphor, whereas an alternate raw material shows a noticeable peak for camphor. Differential constituents are pointed out to demonstrate the discrepancies.

Jasminum officinale is widespread in central Asia and jasmine essential oils are used in different skin care products for calming, hydrating, and for fragrance. The typical volatile components of jasmine essential oils include benzyl acetate, benzyl alcohol, citronellol and methyl jasmonate. Methyl jasmonate was first isolated from jasmine essential oils (3). There are several jasmine essential oils with different qualities available from various suppliers. After initial organoleptic evaluation, a few were analyzed further using a GC-MS. Examples of gas chromatograms are shown in Figure 2. Different jasmine essential oils exhibited altered chromatograms. Further fragmentation on mass spectrophotometry revealed that the peaks near 35.6 min retention time were identified to be different molecules (Figure 3). While one from the top was identified to be methyl jasmonates, a critical component of jasmine essential oils, ones from the bottom two were identified as α-pentyl-cinnamaldehyde, also known as amyl cinnamaldehyde. Amyl cinnamaldehydes are not typically present in jasmine essential oils. In addition, amyl cinnamaldehyde is an allergen. These results suggest that the middle and bottom chromatograms are altered jasmine essential oils.



Figures 2 & 3: Jasmine raw material selection. Jasmine oils from different suppliers show different chromatogram patterns. While multiple, distinctive peaks are present in the selected jasmine, alternate jasmines show missing constituents after 40 minutes of retention time. Further analysis with mass spectrophotometry identified a peak near 35.6 min from selected jasmine is methyl jasmonate. Similar peaks from alternate jasmines were identified as α-pentyl-cinnamaldehyde.



Jasmine essential oils are one of the more common contact allergens among essential oils (4). In a study conducted between 2000 and 2008, 1.6 % of patients tested positive for contact allergy to jasmine absolute (4). Therefore, it is important to evaluate the known allergens present within jasmine essential oils as well as others, since different species, extraction methods, and culture conditions will all vary the amount of allergen present within each lot. In addition to allergens, essential oils are often examined for chemical adulterants, such as heavy metals, pesticides, phototoxins or synthetic molecules. Five heavy metals, arsenic, lead, cadmium, mercury and antimony, were measured for the safety of different essential oils. Among essential oils selected and sourced based on the 6S quality process, none had any heavy metals above 0.016 ppm, which is well below the unavoidable impurities allowed based on the global regulatory bodies. In addition to heavy metals, pesticides were also measured. Some essential oils had only trace levels present, if any were detectable. Lemon essential oils are known to contain phototoxic substances when cold-pressed. Additional distillation was performed to remove the phototoxin from the lemon oil.

Chemical analysis is not the only safety evaluations done for essential oils. An example of safety assessments is irritection (InVitro International, Placentia, CA, USA), which will evaluate potential dermal irritation *in vitro*. Other critical safety evaluations are sting tests and human repeat insult patch tests (HRIPT). Sting tests will evaluate irritation or sensitivity to essential oils, when applied to the skin. HRIPT is an *in vivo* test to evaluate allergic contact sensitization or reaction on the skin of volunteers. HRIPT is supervised by a clinical dermatologist, who will assess the potential safety concerns.

There are several different ways to substantiate the quality of essential oils. There are clinical studies to evaluate claims or simpler *in vitro* assays to evaluate specific target validations. For example, rose essential oils evaluated on skin equivalents demonstrated improved skin structures (data not shown). In addition to above mentioned evaluations, there are other assays performed to evaluate the quality of these essential oils. One example is an organoleptic test, which is performed throughout the quality control steps. Organoleptic evaluation utilizes the sense organs, and the smell is extremely important for essential oils. If peppermint essential oil passes all the quality control steps, but doesn't smell like peppermint, would anyone think it is peppermint oil?

CONCLUSIONS

Selection:
select the best possible ingredients

Sourcing:
quality ingredients for the highest efficacy

Specification:
identify constituents

Standardization:
set for consistent efficacy and quality

Safety:
extensive analysis for safety

Substantiation:
evaluation of efficacy