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## ABSTRACT

There are approximately 200 species of jasmine around the world, and they are cultivated for flowers and fragrance. Due to the sweet and romantic nature of the jasmine fragrance, over 80 percent of women's fragrances contain jasmine. Jasmine essential oils (absolutes) contain multiple volatile compounds that contribute to the distinct smell. Some compounds present in jasmine essential oils include benzyl acetate, benzyl alcohol, citronellol, and methyl jasmonate. Jasmine essential oils are widely used for their calming and sedative properties. Despite their widespread use in the cosmetic industry, the effects of jasmine essential oils on the skin are not well studied. Jasmine essential oils are thought to produce hydrating effects in the skin, although there is a lack of scientific evidence to support this benefit. This work was conducted in order to study the effects of jasmine essential oils in the skin. Prior to assessing the biological activities of jasmine essential oils, the components of jasmine essential oils were analyzed by GC-MS (gas chromatography-mass spectrometry). Biological effects were analyzed using full thickness in vitro skin equivalents and Affymetrix microarrays. Bioinformatics were performed using iPathwayGuide from Advaita Bio. Results showed different patterns of gene expression for each of the jasmine essential oils. Multiple genes and biological processes involved in epidermal functions, including keratinocyte differentiation, were regulated, suggesting that jasmine essential oils may improve the epidermal skin barrier.

## INTRODUCTION

Essential oils are mixtures of volatile compounds that produce distinct, characteristic smells. Traditionally, essential oils were used by indigenous people to improve their mood, alertness, or as medicine. Most essential oils are produced by steam distillation and cold pressing, though 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. Therefore, jasmine oils should be called jasmine absolutes, however jasmine oils are also called jasmine essential oils for simplicity.

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. 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 (2). It is known as a stress hormone because it is produced by plants after biotic and abiotic stress (3).

*Jasminum officinale* is widespread in central Asia and jasmine essential oils are used in different skin care products for calming, hydrating, and for fragrance. In fact, jasmine is widely used in women's fragrance, which could be problematic, since jasmine essential oils are one of the more common contact allergens (4). In a European study conducted between 2000 and 2008, 1.6 percent of patients tested positive for contact allergy to jasmine absolute (4). Therefore, it is important to evaluate the allergen present within jasmine essential oils, 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 pesticides or synthetic molecules. Despite the broad use of jasmine essential oils in skin care products, the biological function in the skin is not well understood. In this study, biological activities of jasmine essential oils were evaluated using human skin equivalents.



## OBJECTIVE

To characterize jasmine essential oils by analyzing the quality and the activity through chemical and biological methods.

## METHODS

### 1. 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. Several jasmine essential oils were obtained from suppliers, and GC-MS analyses 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. Different essential oil samples were diluted in chloroform by 1:20. Compounds are fragmented by electron ionization (EI) to generate spectra, which is compared to library spectra using MassHunter Workstation (Agilent).

### 2. AFFYMETRIX AND QPCR OPENARRAY

In order to evaluate different jasmine essential oils on biological activities, full-thickness human skin equivalents were obtained from MatTek (Ashland, MA). These human skin equivalents were made from normal human fibroblasts and normal human keratinocytes. After equilibrating the skin equivalents according to the manufacturer's protocol, 15µL of essential oil was applied to the surface of each skin culture and distributed evenly using a sterile glass spreader. Six replicates were included in each treatment group. After a 24-hour incubation period, the skin equivalents were harvested. Maxwell 16 LEV Simply RNA Tissue Kits (Promega, Madison, WI) were used to isolate RNA, followed by determination of RNA concentration and quality using a NanoDrop 2000 spectrophotometer (Thermo Scientific, Waltham, MA).

For microarray analysis, samples were labelled using Affymetrix 3' IVT Plus Kits (Affymetrix, Santa Clara, CA) and hybridized onto Affymetrix Human Genome U133 Plus 2.0 GeneChips (HU133 Plus 2.0 Array). GeneChips were scanned with an Affymetrix GCS3000 GeneChip Scanner and bioinformatics analysis was conducted using BioConductor R software and iPathwayGuide (Advaita Bio, Plymouth, MI).

For qPCR analysis, cDNA was synthesized using High Capacity DNA Synthesis Kits (Life Technologies, Grand Island, NY). 380 validated Taqman gene expression assays for target genes and 4 endogenous control genes were assembled into a custom OpenArray (Life Technologies) and analyzed using a QuantStudio 12K Flex instrument (Life Technologies). A statistical data analysis was performed using RealTime StatMiner software v 4.2.

## REFERENCES

- Food and Chemical Toxicology 46: 446-475, 2008
- Helvetica Chimica Acta 45: 675-685, 1962
- Essential Oil Safety: A Guide for Health Care Professionals, Second Edition, 2014
- Contact Dermatitis 63: 277-283, 2010

## RESULTS

### 1. Analytical Chemistry

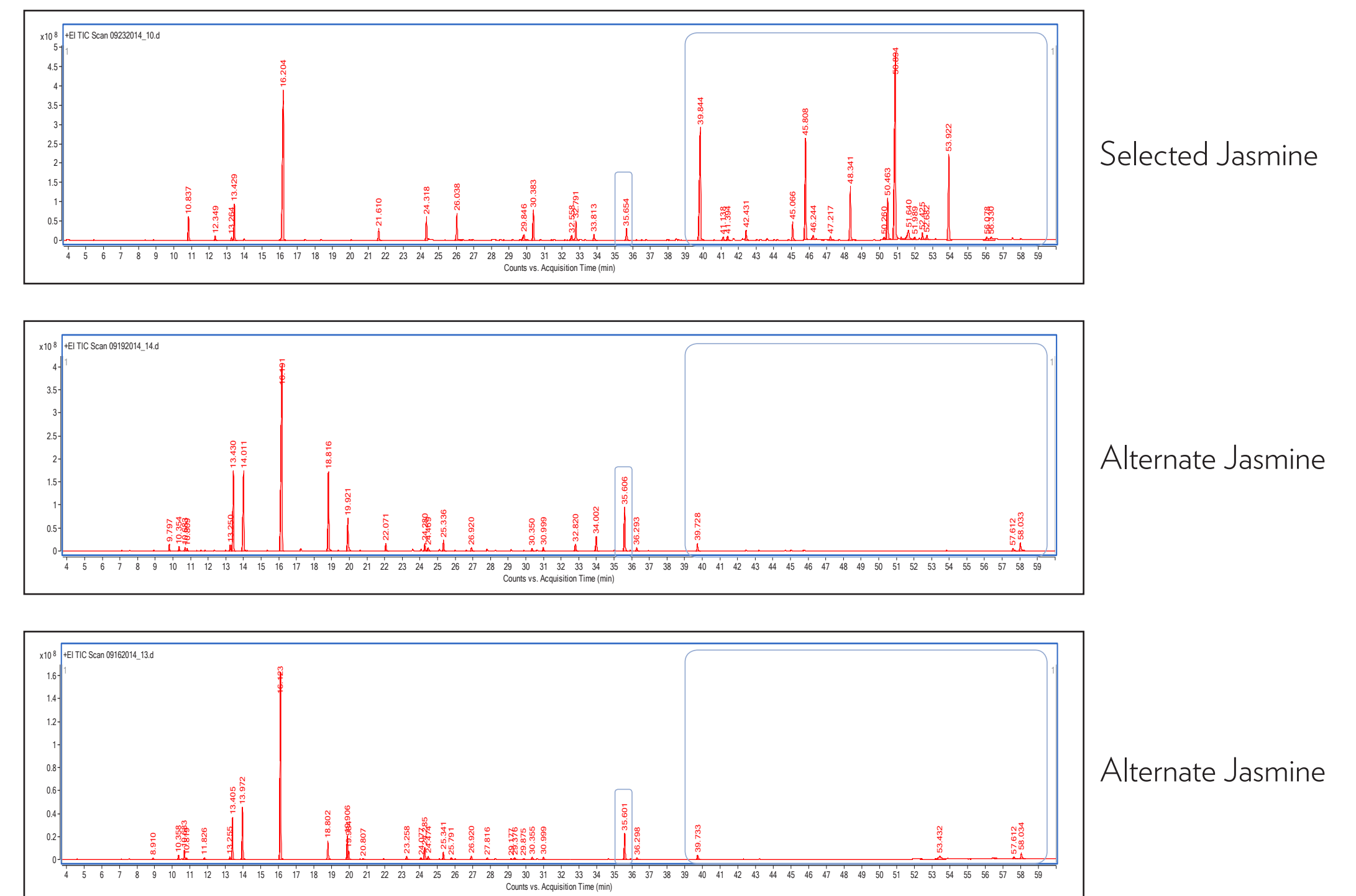


FIGURE 1: Chromatograms from Different Jasmine Essential Oils

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 1. Different jasmine essential oils exhibited altered chromatograms. The top chromatogram shows the closest fingerprint of the chromatogram of unadulterated *Jasminum officinale* essential oils, and was chosen for further studies. The middle and the bottom chromatograms are different from the top one, but similar to each other. The peaks after 40 minutes on retention time are noticeably missing from the middle and the bottom chromatograms. Further fragmentation on mass spectrophotometry revealed that the peaks near 35.6 minutes retention time were identified to be different molecules (Figure 2). 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  $\alpha$ -pentyl-cinnamaldehyde, also known as amyl cinnamaldehyde. As mentioned previously, methyl jasmonates are first identified from jasmine essential oils and are a well-known constituent. On the other hand, amyl cinnamaldehydes are not typically present in jasmine essential oils. In addition, amyl cinnamaldehyde is an allergen, listed on the 26 fragrance allergens by the European Union. These results suggest that the middle and bottom chromatograms are altered jasmine essential oils.

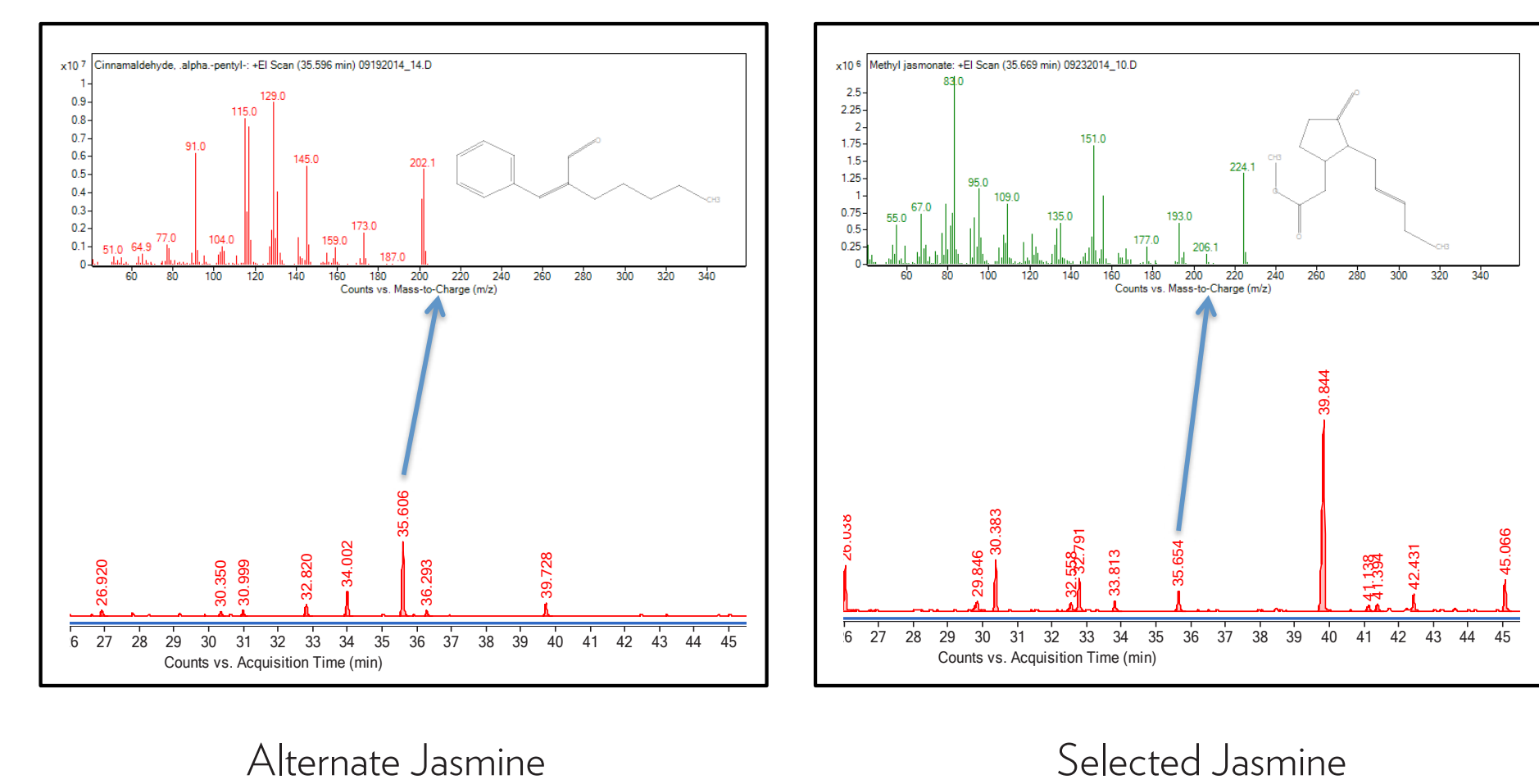


FIGURE 2: Identification of Key Constituents by Mass Spectrophotometry

### 2. Biological Activities

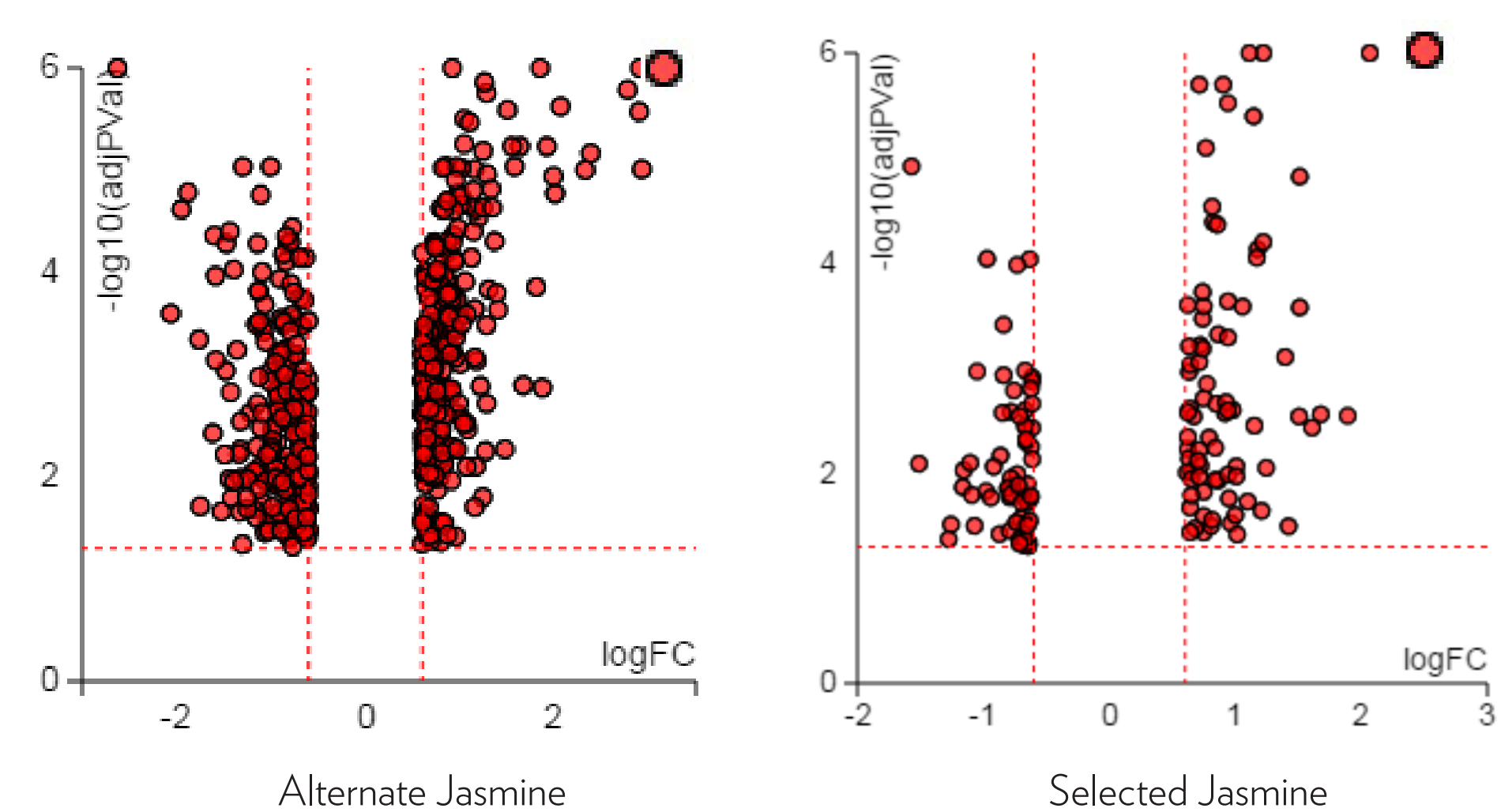


FIGURE 3: Volcano Plots from Different Jasmine Essential Oils

Following application of the essential oils on the skin equivalents, gene expression was first analyzed by microarray, followed by qPCR validation of genes that regulate skin functions. Different jasmine essential oils modulated different genes, demonstrated by volcano plots from the microarray (Figure 3). Volcano plots show profiles of statistically significant data only (P value on the y-axis with the fold change on the x-axis). Gene expression data generated from the microarrays was validated with qPCR, examining skin specific target genes (Figure 4). Among different skin genes positively stimulated, several targets were involved in the skin barrier formation, as well as antioxidant enzymes and growth factors/receptors. The data suggest that jasmine essential oils may improve the epidermal skin barrier and improve the response to stress via upregulation of antioxidant pathways.

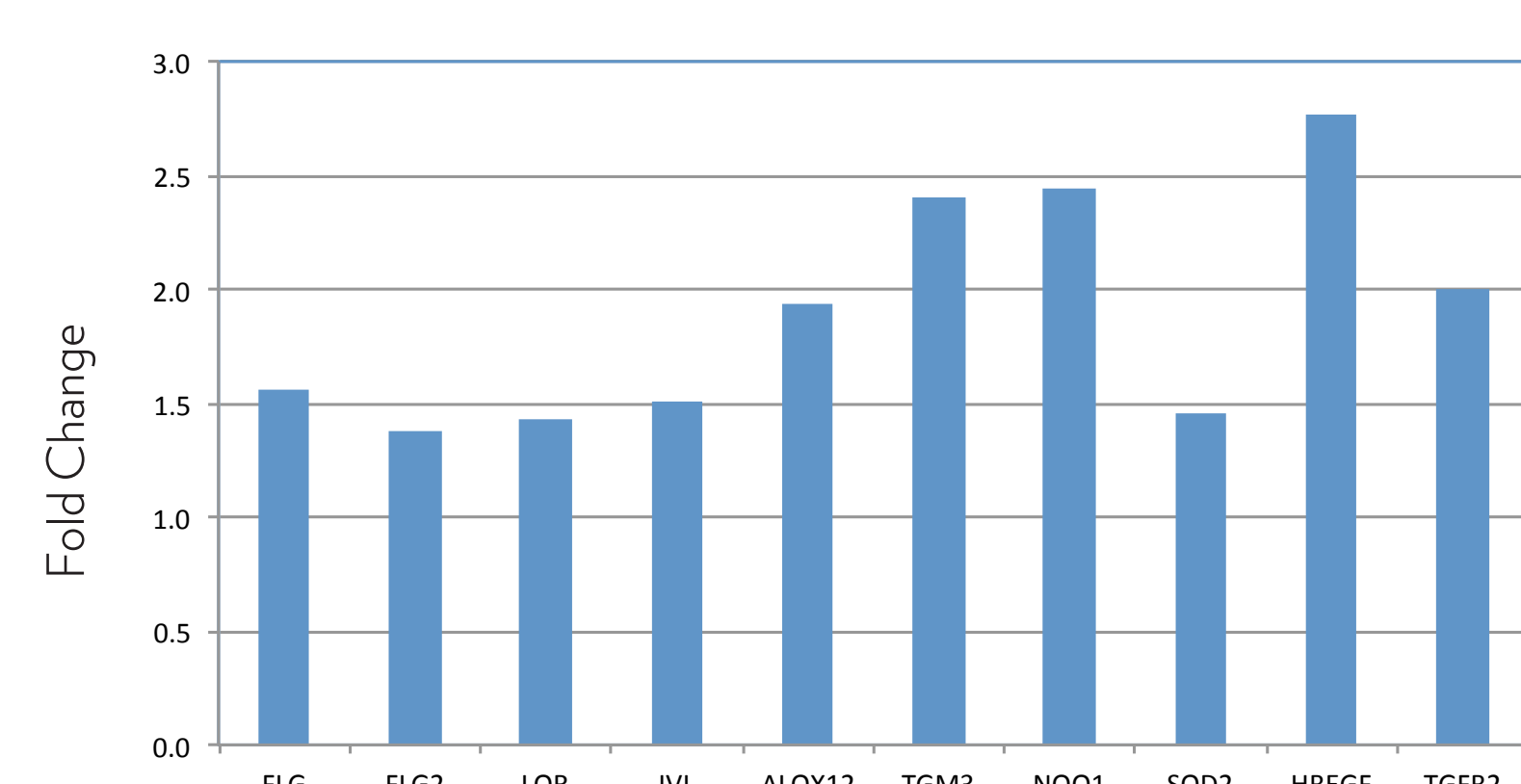


FIGURE 4: qPCR Validation of Targeted Skin Genes

## CONCLUSIONS

- Quality of jasmine essential oils varies among differentially sourced raw materials.
- Different jasmine essential oils have different biological activities.
- Jasmine essential oils regulated genes involved in terminal differentiation of keratinocytes, suggesting a role in epidermal barrier function.