Composition and potential anticancer activities of essential oils obtained from myrrh and frankincense

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Abstract. The present study aimed to investigate the composition and potential anticancer activities of essential oils obtained from two species, myrrh and frankincense, by hydrodistillation. Using gas chromatography-mass spectrometry (GC-MS), 76 and 99 components were identified in the myrrh and frankincense essential oils, respectively, with the most abundant components, 2-Cyclohexen-1-one, 4-ethyl-4-hydroxy-3,5,5-trimethyl- and n-Octylacetate, accounting for 12.01 and 34.66%, respectively. The effects of the two essential oils, independently and as a mixture, on five tumor cell lines, MCF-7, HS-1, HepG2, HeLa and A549, were investigated using the MTT assay. The results indicated that the MCF-7 and HS-1 cell lines showed increased sensitivity to the myrrh and frankincense essential oils compared with the remaining cell lines. In addition, the anticancer effects of myrrh were markedly increased compared with those of frankincense, however, no significant synergistic effects were identified. The flow cytometry results indicated that apoptosis may be a major contributor to the biological efficacy of MCF-7 cells.

Introduction

Commiphora myrrha has a yellow oleo-gum resin that exists in its stem and is used worldwide for the production of myrrh, particularly in China and Egypt. The constituents of myrrh include volatile oil (2-8%), resin (23-40%), gum (40-60%) and bitter principles (10-25%). Previous studies have shown that myrrh exhibits cytotoxic, analgesic, anti-inflammatory, anticancer, antiparasitic and hypolipidemic activities (1-4).

Frankincense is an aromatic resin obtained from trees of the genus Boswellia and has been hypothesized to exhibit a number of health supporting properties, including the treatment of rheumatoid arthritis and anti-inflammatory, antibacterial, antifungal and anticancer activities (5-8). Frankincense oil is prepared by the steam distillation of frankincense gum resin and is frequently used in aromatherapy practices. According to previous studies, the constituents of frankincense oil vary according to the climate, harvest conditions and geographical sources of the frankincense resin (9).

Notably, these two resinous drugs are always prescribed simultaneously in traditional Chinese medicine and are primarily administered for the treatment of blood stagnation and inflammation diseases, as well as for the relief of swelling and pain (10). A previous study identified that the combination of frankincense and myrrh oils exhibited synergistic effects on Cryptococcus neoformans and Pseudomonas aeruginosa (11).

The present study investigated the chemical composition of hydrodistilled frankincense and myrrh oils from Ethiopia. In addition, the anticancer activities of the prepared essential oils against the MCF-7, HepG2, HeLa and A549 cell lines were investigated to determine whether synergistic effects were observable in vitro. The results illustrated that certain cells (MCF-7 and HS-1 cells) demonstrate increased sensitivity to the two essential oils, and the anticancer effects of myrrh is superior to frankincense. No synergistic effect was observed.

Materials and methods

Materials. Dry sap samples were obtained in Ethiopia from the stem bark of Boswellia carterii and Commiphora pyranthoides Engler in August 2009. The plant materials were identified by a botanist at Harbin Medicine University-Daqing (Daqing, China) and a voucher specimen was stored at the Department of Pharmacology (School of Pharmacy, Harbin Medicine University-Daqing).
Extraction of essential oils. Subsequent to being frozen for 24 h, 30 g of the air-dried frankincense and myrrh samples were crushed into a powder. The essential oils from each sample were obtained through hydrodistillation for 3 h, according to the AB method described previously (12). Subsequently, the essential oils were diluted with 1% Tween 80 for a bioactivity analysis. The solution was prepared by mixing the myrrh and frankincense essential oils in a 1:1 ratio.

GC-MS analysis. Analyses of the constituents of the essential oils were performed using gas chromatography mass spectrometry (GC-MS; Agilent Technologies, Santa Clara, CA, USA) and the GCMS-QP2010S mass spectrometer (Shimadzu Corp., Kyoto, Japan) with Rtx®-50 elastic quartz capillary column (30x0.25 mm, 0.25 µm) and helium carrier gas (Beijing BAIF Gases Industry Co., Ltd., Beijing, China). The injector temperature was 230°C and the interface and ion-source heating temperatures were 300°C and 230°C, respectively. The temperature program consisted of 60°C for 1 min and 220°C for 15 min, with a heating rate of 5°C/min. The column head pressure was 70 kPa, the EI-mode was 70 eV and the scan-range was 20-500 amu with a time cycle of 0.65 sec. Mass spectral correlations were performed using NIST05.

Cell culture. Human cell lines (American Type Culture Collection, Rockville, MD, USA) obtained from breast (MCF-7) and hepatocellular (HepG2) carcinomas and cervical (HeLa), skin (HS-1) and small cell lung (A549) cancers, were maintained in monolayer tissue culture Petri dishes prior to examination. RPMI-1640 medium was supplemented with 10% fetal bovine serum (both Sigma-Aldrich, St. Louis, MO, USA), 100 IU/ml penicillin, 100 µg/ml streptomycin and 2 mM/l glutamine and cultures were maintained in a humidified atmosphere at 37°C in 5% CO₂.

MTT antiproliferative assay. 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) method was used to determine the effects of frankincense and/or myrrh essential oils on cell proliferation in the MCF-7, HepG2, HeLa, HS-1 and A549 cell lines. Briefly, 5x10³ cells/well were evenly distributed and incubated on 96-well plates (Iwaki, Tokyo, Japan) overnight. The cells were then treated with frankincense, myrrh and a mixture of the essential oils at concentrations of 0, 5, 10, 20, 40, 60, 80, 160, 180, 320 and 640 µg/ml, and incubated for 24 and 48 h. Subsequently, the medium in each well was replaced with 20 µl MTT (5 mg/ml in PBS) and incubated at 37°C for 4 h. The purple-blue formazan precipitate was dissolved in 100 µl dimethyl sulfoxide and the optical density was measured at a wavelength of 570 nm on a 96 -well plate reader (Thermo Labsystems, Franklin, MA USA). The IC₅₀ was calculated as the concentration of compounds that achieved a 50% inhibition of cell viability. Data were analyzed using a SlideWrite program (Advanced Graphics Software, Inc., Rancho Santa Fe, CA, USA) to determine the IC₅₀ of each drug independently.

Synergistic effect analysis. Isobologram curves were derived as described previously (13): IC₅₀ A and B = Dₓ / ICₓ,A + Dᵧ / ICₓ,B; where IC₅₀ A and B indicates the combination concentration of drugs A and B at 50% inhibition, ICₓ,A and ICₓ,B indicates the concentration of the drugs that result in 50% inhibition independently and Dₓ and Dᵧ indicates the concentrations of the two drugs as a mixture to achieve 50% inhibition. The isobologram curve was generated by plotting doses of drugs A vs. B predicted to simultaneously achieve 50% cell growth inhibition. A standard line of Loewe additivity was included to indicate a lack of interaction, and points below and above the line indicated synergy and antagonism, respectively.

Cell apoptosis assay. Flow cytometry was used for the quantitative measurement of apoptosis. Briefly, 1x10⁶ MCF-7 cells were treated with 0, 10, 20 and 40 µg/ml frankincense and/or myrrh essential oils for 24 and 48 h, respectively. The cells were then collected by trypsinization and washed once with cold PBS. BD tubes were used and 100 µl suspension was added to each labeled tube followed by 10 µl Annexin V-FITC and 10 µl PI (20 µg/ml). Following incubation for ≥20 min at room temperature in the dark, 400 µl PBS binding buffer was added to each tube without washing. Within 30 min, the mixtures were analyzed using flow cytometry (BD FACSaria; BD Biosciences, San Jose, USA).

Results

GC-MS analysis. The content of the extracted oil of myrrh and frankincense was ~0.41 ml (2.05%, ml/g) and 0.62 ml (2.06%, ml/g), respectively, and the total ion figures of the constituents were obtained by GC-MS analysis. The area normalization method was adopted to integrate the total ion peaks and the minimum area of the comparatively small peaks was set. Using a standard mass spectrum, 76 components were identified that accounted for 87.54% of the total myrrh essential oil (Table I). In addition, 99 components were identified that accounted for 91.26% of the total frankincense essential oil (Table II).

MTT antiproliferative assay. Myrrh and frankincense essential oils exhibited an inhibitory effect on the cell lines and a dose-dependent inhibition effect was noted. Among the five
Table I. Chemical composition of myrrh essential oil.

| No. | Compound                                                                 | RI<sup>a</sup> | %<sup>b</sup> |
|-----|---------------------------------------------------------------------------|----------------|-------------|
| 1   | Bicyclo[2.2.1]heptane, 2-(1-methylethenyl)-                              | 969            | 0.03        |
| 2   | Azulene                                                                   | 1069           | 0.05        |
| 3   | (+)-Cycloisosativene                                                      | 1125           | 0.27        |
| 4   | Acetic acid, octyl ester                                                 | 1183           | 0.10        |
| 5   | Ylangene                                                                  | 1221           | 0.10        |
| 6   | Copaene                                                                   | 1221           | 5.50        |
| 7   | 5H-inden-5-one, 1,2,3,6,7,7a-hexahydro-7a-methyl-                          | 1237           | 1.73        |
| 8   | Seychellene                                                               | 1275           | 0.57        |
| 9   | Cyclohexane, 1,2-dienethenyl-4-(1-methylethylidene)-, cis-               | 1281           | 0.36        |
| 10  | Biurea                                                                    | 1328           | 0.02        |
| 11  | β-bourbonene                                                              | 1339           | 2.06        |
| 12  | (+)-Sativien                                                              | 1339           | 0.11        |
| 13  | Isosativene                                                               | 1339           | 0.02        |
| 14  | α-cubebene                                                                | 1344           | 0.39        |
| 15  | δ-elemene                                                                 | 1377           | 2.51        |
| 16  | 7-Tetracyclo[6.2.1.0(3.8)0(3.9)]undecanol, 4,4,11,11-tetramethyl-         | 1385           | 0.03        |
| 17  | Aromadendrene                                                             | 1386           | 0.63        |
| 18  | Tricyclo[6.3.0.0(2,4)]undec-8-ene, 3,3,7,11-tetramethyl-                  | 1391           | 0.23        |
| 19  | Aromadendrene, dehydro-                                                  | 1396           | 4.62        |
| 20  | β-elemene                                                                 | 1398           | 8.57        |
| 21  | α-longipinene                                                             | 1403           | 0.07        |
| 22  | 1,4-Disopropyl-2,5-dimethylbenzene                                        | 1403           | 0.65        |
| 23  | 2-Cyclohexen-1-one, 4-ethyl-4-hydroxy-3,5,5-trimethyl-                     | 1406           | 12.00       |
| 24  | 1,1,4,7-Tetramethyl-1a,2,3,4,4a,5,6,7b-octahydro-1H-cyclopenta[ez]azulene   | 1419           | 0.47        |
| 25  | α-bergamotene                                                             | 1430           | 0.70        |
| 26  | trans-α-bergamotene                                                       | 1430           | 0.93        |
| 27  | Cyperene                                                                  | 1432           | 0.25        |
| 28  | γ-muurolene                                                               | 1435           | 0.78        |
| 29  | Aminourea                                                                 | 1437           | 0.03        |
| 30  | α-amorphene                                                               | 1440           | 1.96        |
| 31  | Naphthalene, 1,2,3,4,4a,7-hexahydro-1,6-dimethyl-4-(1-methylethyl)-        | 1440           | 0.13        |
| 32  | β-panasinesene                                                            | 1441           | 0.41        |
| 33  | 7-Oxabicyclo[4.1.0]heptane, 2,2,6-trimethyl-1-(3-methyl-1,3-butanediyl)-5-methylen- | 1452           | 0.54        |
| 34  | Bicyclo[5.3.0]decane, 2-methylene-5-(1-methylvinyl)-8-methyl-              | 1456           | 4.37        |
| 35  | Aromadendrene oxide-(2)                                                  | 1462           | 0.18        |
| 36  | γ-elemene                                                                 | 1465           | 4.52        |
| 37  | Nitrogen                                                                  | 1468           | 0.03        |
| 38  | β-cadinene                                                                | 1469           | 2.74        |
| 39  | 1-Cycloheptene, 1,4-dimethyl-3-(2-methyl-1-propene-1-yl)-4-vinyl-          | 1480           | 0.42        |
| 40  | α-guaiene                                                                 | 1490           | 0.20        |
| 41  | α-bulnesene                                                               | 1490           | 1.17        |
| 42  | 4,11,11-Trimethyl-8-methylenebicyclo[7.2.0]undec-4-ene                     | 1494           | 1.68        |
| 43  | Humulen-(v1)                                                              | 1494           | 0.36        |
| 44  | 2,4-Dimethyl-3-nitrobicyclo[3.2.1]octan-8-one                             | 1498           | 0.20        |
| 45  | Germacrene                                                                | 1515           | 0.52        |
| 46  | Germacrene D                                                              | 1515           | 3.81        |
| 47  | Cyclopenta[c,d]pentane-1,3-dione, hexahydro-4-(2-methyl-2-propenyl)-2,2,4-trimethyl- | 1518           | 0.35        |
| 48  | Elemol                                                                    | 1522           | 3.96        |
| 49  | 4-(1-Methylethylidene)-1,2-divinylcyclohexane                             | 1530           | 0.57        |
| 50  | Epiglobulol                                                               | 1530           | 0.27        |
| 51  | Cyclononasiloxane, octadecamethyl-                                        | 1535           | 0.43        |
| 52  | Ent-spathulenol                                                           | 1536           | 3.34        |
| 53  | (-)-Spathulenol                                                           | 1536           | 0.32        |
| 54  | 3,7-Cyclodecadien-1-one, 10-(1-methylethenyl)-, (E,E)-                    | 1562           | 2.00        |
Table I. Continued.

| No. | Compound                          | RI  | %   |
|-----|-----------------------------------|-----|-----|
| 55  | Nerolidol                         | 1564| 0.04|
| 56  | Humulene                          | 1579| 0.80|
| 57  | t-cadinol                         | 1580| 1.90|
| 58  | β-cadinol                         | 1580| 0.41|
| 59  | Longipinocarveol, trans-          | 1599| 0.51|
| 60  | Azulen-2-ol, 1,4-dimethyl-7-(1-methylethyl)- | 1602| 0.78|
| 61  | Nickel tetracarbonyl              | 1623| 0.02|
| 62  | 6-Isopropenyl-4,8a-dimethyl-1,2,3,5,6,7,8,8a-octahydro-naphthalen-2-ol | 1690| 0.13|
| 63  | Cadalene                          | 1706| 0.16|
| 64  | 2-(4a,8-Dimethyl-1,2,3,4,4a,5,6,7-octahydro-naphthalen-2-yl)-prop-2-en-1-ol | 1745| 0.25|
| 65  | 3a,9b-Dimethyl-1,2,3a,4,5,9b-hexahydrocyclopenta[a]naphthalen-3-one | 1747| 0.06|
| 66  | Benzofuran, 2,3-dihydro-2-methyl-5-phenyl- | 1763| 0.07|
| 67  | Bicyclo[4.1.0]heptan-2-ol, 1β-(3-methyl-1,3-butadienyl)-2α,6β-dimethyl-3β-acetoxy- | 1801| 0.02|
| 68  | Nerolidol isobutyrate            | 1889| 0.05|
| 69  | Dihexyl phthalate                 | 1908| 0.04|
| 70  | 2(3H)-Naphthalenone, 4,4a,5,6,7,8-hexahydro-4-phenyl- | 1918| 1.89|
| 71  | N-(Trifluoroacetyl)-N,O,O',O''-tetrakis(trimethylsilyl) norepinephrine | 2151| 0.89|
| 72  | Dinordesoxo-9-methyl-1, 8-diacetyleseroline | 2152| 0.22|
| 73  | 4-Butylbenzoic acid, 2,7-dimethyloct-7-en-5-yn-4-yl ester | 2223| 0.22|
| 74  | Retinol acetate                   | 2362| 0.21|
| 75  | (4α,5α,17β)-3,17-dihydroxy-4,5-epoxyandrost-2-ene-2-carbonitrile | 2427| 0.24|
| 76  | (+)-Epi-bicyclosesquiphellandrene | 2682| 0.56|

*Retention index; †relative percentage obtained from peak area.

Table II. Chemical composition of frankincense essential oil.

| No. | Compound                          | RI  | %   |
|-----|-----------------------------------|-----|-----|
| 1   | α-pinene                          | 948 | 0.07|
| 2   | Sabinene                          | 897 | 0.02|
| 3   | Nopinene                          | 943 | 0.02|
| 4   | β-myrcene                         | 958 | 0.03|
| 5   | Octanal                           | 1005| 0.03|
| 6   | Hexyl acetate                     | 984 | 0.10|
| 7   | o-Cymene                          | 1024| 0.03|
| 8   | D-Limonene                        | 1018| 0.30|
| 9   | Eucalyptol                        | 1059| 0.09|
| 10  | β-trans-ocimene                   | 976 | 0.04|
| 11  | β-cis-ocimene                     | 976 | 0.13|
| 12  | Tricyclene                        | 998 | 0.01|
| 13  | n-Octanol                         | 1059| 3.27|
| 14  | β-linalool                        | 1082| 0.38|
| 15  | Nonanal                           | 1104| 0.02|
| 16  | 1,3-Dimethylcyclohexene           | 852 | 0.58|
| 17  | L-pinocarveol                     | 973 | 0.02|
| 18  | Isoborneol                        | 1138| 0.03|
| 19  | 4-Terpineol                       | 1137| 0.07|
| 20  | Naphthalene                       | 1231| 0.09|
| 21  | 3-Cyclohexene-1-methanol          | 1137| 0.09|
| 22  | n-Octyl acetate                   | 1183| 34.66|
| 23  | cis-Geraniol                      | 1128| 0.03|
| 24  | n-Decanol                         | 1158| 0.09|
Table II. Continued.

| No. | Compound                                                                 | RI$^a$ | %b |
|-----|--------------------------------------------------------------------------|--------|-----|
| 25  | 1,7,7-Trimethylbicyclo[2.2.1]hept-2-yl acetate                         | 1277   | 1.08 |
| 26  | 2-Dodecanone                                                            | 1151   | 0.02 |
| 27  | Octane                                                                  | 1042   | 0.03 |
| 28  | n-Nonyl acetate                                                         | 1282   | 0.03 |
| 29  | Benzyl butyl ether                                                      | 1264   | 0.02 |
| 30  | (-)-Myrtenyl acetate                                                   | 1314   | 0.04 |
| 31  | Bornylene                                                               | 1243   | 0.03 |
| 32  | δ-elemene                                                               | 1377   | 0.67 |
| 33  | Citronellol acetate                                                    | 1302   | 0.38 |
| 34  | 1,10-Decanediol                                                        | 1356   | 0.04 |
| 35  | Longicyclene                                                            | 1184   | 0.07 |
| 36  | Cubebene                                                                | 1344   | 0.08 |
| 37  | Nerol acetate                                                          | 1352   | 0.82 |
| 38  | Cyclobuta[1,2:3,4]dicyclopentene, decahydro-3a-methyl-6-methylene-1-(1-methylethyl)-, [1S-(1α,3α,3β,6αβ,6βα)]- | 1339   | 0.16 |
| 39  | Decyl acetate                                                           | 1381   | 0.72 |
| 40  | 1,4-Methanoazulene, decahydro-4,8,8-trimethyl-9-methylene-, (1S,3aR,4S,8aS)- | 1398   | 0.40 |
| 41  | Cyclopentane, 1-acetoxyethyl-3-isopropenyl-2-methyl-                  | 1315   | 0.07 |
| 42  | Caryophyllene oxide                                                    | 1494   | 0.13 |
| 43  | Bergamotol, Z-α-trans-                                                  | 1673   | 0.05 |
| 44  | Isoamyl caprylate                                                      | 1364   | 0.03 |
| 45  | (+)-Sativen                                                             | 1339   | 0.05 |
| 46  | Longicyclene                                                            | 1184   | 0.09 |
| 47  | Dodecanoic acid, 4-penten-1-yl ester                                   | 1281   | 0.04 |
| 48  | α-humulene                                                              | 1579   | 0.07 |
| 49  | Hexahydrobenzylacetone                                                 | 1440   | 0.03 |
| 50  | α-Amorphene                                                             | 1429   | 0.18 |
| 51  | Germacrere                                                              | 1515   | 0.76 |
| 52  | (Z)-11-Tetradecen-1-ol acetate                                         | 1787   | 0.28 |
| 53  | α-muurolone                                                             | 1440   | 0.09 |
| 54  | α-dodecene                                                              | 1235   | 0.02 |
| 55  | β-bisabolene                                                            | 1500   | 0.06 |
| 56  | γ-muurolone                                                             | 1435   | 0.05 |
| 57  | Methyl dodecanoate                                                     | 1457   | 0.03 |
| 58  | γ-cadinene                                                              | 1469   | 0.12 |
| 59  | Isophytol                                                               | 1899   | 0.03 |
| 60  | 2-(4-Ethyl-4-methyl-3-(isopropenyl)cyclohexyl)propan-2-ol               | 1300   | 0.08 |
| 61  | γ-elemene                                                               | 1465   | 0.19 |
| 62  | 1,10-Decanediol                                                        | 1501   | 0.11 |
| 63  | Hexyl octanoate                                                        | 1580   | 0.64 |
| 64  | 4-Camphenylbutan-2-one                                                 | 1451   | 0.11 |
| 65  | (-)-Spathulenol                                                         | 1536   | 0.23 |
| 66  | (-)-δ-cadinol                                                           | 1420   | 0.09 |
| 67  | (2E,6E,10E)-12-Hydroxy-3,7,11-trimethyl-2,6,10-dodecatrienyl acetate    | 2076   | 0.14 |
| 68  | 1-Pentadecanol                                                         | 1543   | 0.05 |
| 69  | Octyl heptanoate                                                       | 1602   | 0.04 |
| 70  | (Z)-11-Tetradecenyl acetate                                            | 1787   | 0.19 |
| 71  | 10-Isopropenyl-3,7-cyclodecadien-1-one                                  | 1745   | 0.06 |
| 72  | Octanoic acid, phenylmethyl ester                                      | 1756   | 0.05 |
| 73  | Octanoic acid, octyl ester                                             | 1779   | 0.32 |
| 74  | 2,4a,5,6,7,8,9a-octahydro-3,5,5-trimethyl-9-methylene-1H-benzocycloheptene | 1826   | 0.04 |
| 75  | Farnesyl acetate                                                       | 1834   | 0.06 |
| 76  | Lanceol, cis                                                           | 1737   | 0.07 |
| 77  | Cycloheptane, 4-methylene-1-methyl-2-(2-methyl-1-propen-1-yl)-1-vinyl-  | 1541   | 0.82 |
cell lines, MCF-7 and HS-1 were sensitive to the myrrh and frankincense essential oils (Table III).

Synergistic effect analyses. All points were identified above the standard line of Loewe additivity, therefore, no synergistic effects were identified in the isobologram and combination index curves (Fig. 1).

Cell apoptosis assay. The flow cytometry results showed that the myrrh, frankincense and the mixture of essential oils were capable of inducing apoptosis in the MCF-7 cells in a concentration-dependent manner (Fig. 2). A dose-dependent induction of the apoptotic cells was performed to investigate the apoptosis rate. The early- and late-stage apoptosis rates of the MCF-7 cells induced by 40 µg/ml myrrh, frankincense and the mixture of essential oils were 36.0, 77.3 and 45.8%, respectively (P<0.01).

Discussion
In the present study, the constituents of the essential oils of myrrh and frankincense were identified to include monoterpenes, sesquiterpenes, alcohols and esters.
2-Cyclohexen-1-one, 4-ethyl-4-hydroxy-3,5,5-trimethyl was demonstrated to account for the highest percentage of the components in myrrh (12.0%), followed by β-elemene, copaene and aromadendrene, dehydro (6.18, 5.50 and 4.62%, respectively). By contrast, n-Octyl acetate was the most significant component of frankincense, accounting for 34.66%, followed by nerolidolosobutrate, 3,7,11-trimethyl-1,6,10-dodecatrien-3-ylster-formic acid, δ-elemene and n-Octanol (18.29, 9.61, 5.61 and 3.24%, respectively). In contrast with the results of a previous study (14), additional components were detected in the frankincense oil, including β-elemene, α-pinene and n-Octanol (5.61, 0.07 and 3.24%, respectively).

A significant inhibitory effect was noted in the cell lines following treatment with the myrrh essential oil compared with treatment with frankincense and the mixture of essential oils. This observation indicated that apoptosis may be a major contributor to the biological efficacy of the MCF-7 cells. The apoptosis rate was higher in the myrrh essential oil group compared with that of the frankincense and mixture of essential oil groups at three concentrations (P<0.01). In addition, the results indicated that the breast cancer cell line exhibited increased sensitivity to the myrrh essential oil. To the best of our knowledge, the present study investigated the synergistic effects of the two drugs in the tumor cell lines for the first time. No synergistic effects were identified, which is in contrast to results observed using the Chinese folk formula (10).

Using cancer cell apoptosis induction trials, previous studies have identified that specific components of myrrh and frankincense essential oils are capable of inducing cancer cell apoptosis. For example, sesquiterpenes have anticancer activity of different extracts of *Commiphora myrrha*. J Pharm Pharmacol 61: 1653-1656, 2009.

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