

Foliar Essential Oil of White Fir (*Abies concolor* (Gordon) Lindl. ex Hildebr.): Chemical Composition and Enantiomeric Distribution

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Abstract

Members of the *Abies* genus (Pinaceae) have been used in traditional medicines in their natural ranges, and essential oils derived from *Abies* species have found use in aromatic therapy. In this work, the foliar (leaves and twigs) essential oil of *Abies concolor* have been obtained from mature trees growing in southwestern Idaho and analyzed using gas chromatographic techniques. The essential oils were rich in α -pinene (15.2–20.5%), camphene (7.5–10.2%), β -pinene (24.2–25.6%), δ -3-carene (5.5–6.5%), limonene (5.4–6.9%), and bornyl acetate (14.6–22.1%). The dominant enantiomers for the major terpenoids were (+)- α -pinene, (–)-camphene, (–)- β -pinene, (+)- δ -3-carene, (–)-limonene, and (–)-bornyl acetate. The essential oil compositions are qualitatively similar to other *Abies* species, but the enantiomeric distributions for α -pinene, β -pinene, and camphene did reveal notable variation between *Abies* essential oils.

Keywords

essential oil, terpenoids, Pinaceae, gas chromatography, chiral, enantiomer, pinene, bornyl acetate

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Introduction

Several species of *Abies* (Pinaceae) have been used in traditional medicine. For example, a paste from leaves and cones of *Abies spectabilis* (D. Don) Mirb. is used in Nepali traditional medicine to treat bone fractures,¹ leaves of *Abies pindrow* (Royle ex D. Don) Royle are employed in Ayurvedic medicine for fever, respiratory, and inflammatory conditions,² *Abies sibirica* Ledeb. leaves are used in Siberia to treat vomiting, diarrhea, epilepsy, and as a diuretic,³ preparations of *Abies webbiana* (Wall. Ex D. Don) Lindl. are used in Siddha and Ayurveda medical systems to treat a variety of ailments,⁴ and northern tribes of Native Americans used the resin of *Abies balsamea* (L.) Mill. externally as an antiseptic salve and internally to treat colds, consumption, menstrual irregularities, and as a laxative.⁵ *Abies concolor* (Gordon) Lindl. ex Hildebr. has been used by several western North American tribes. The Keres used an infusion of the foliage as a bath for rheumatism, the Paiute and Shoshoni used the resin to treat cuts, wounds, and sores, and a decoction of the resin was taken for respiratory troubles.⁶ Essential oils derived from *Abies* species have found utility in aromatherapy, including *Abies alba* Mill., *A. balsamea*, *A. concolor*, *Abies koreana* E. H. Wilson, *Abies lasiocarpa* (Hook.) Nutt.,

Abies nordmanniana (Steven) Spach, *Abies procera* Rehder, *Abies sachalinensis* (F. Schmidt) Mast., *A. sibirica*, and *A. spectabilis*.⁷

Abies concolor is native to western North America. There are two recognized subspecies, *A. concolor* subsp. *lowiana* (Gordon) A. E. Murray, the California white fir, ranges from the southern Cascade Range in Oregon, south through the western slopes of the Sierra Nevada in California, and into Baja California. *Abies concolor* subsp. *concolor*, the Rocky Mountain white fir, is found naturally in the Wasatch Range of Utah and southeastern Idaho, the Mogollon Rim of Arizona, the southern Rocky Mountains of Colorado and New Mexico, and the San Francisco Mountains of western New Mexico.^{8,9} In southwestern Idaho, *A. concolor* subsp. *concolor* is planted as an

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Table 1. Chemical Composition of *Abies concolor* Foliar Essential Oil.

RI _{calc}	RI _{db}	Compound	% Composition	
			#1	#2
923	923	Tricyclene	0.7	0.9
926	927	α -Thujene	0.1	0.1
934	933	α -Pinene	20.5	15.2
951	953	Camphene	7.5	10.2
973	972	Sabinene	0.2	0.1
979	978	β -Pinene	25.6	24.2
990	991	Myrcene	1.9	1.1
1007	1007	α -Phellandrene	tr	tr
1011	1009	δ -3-Carene	6.5	5.5
1017	1018	α -Terpinene	tr	tr
1025	1025	<i>p</i> -Cymene	0.2	0.2
1031	1030	Limonene	6.9	5.4
1032	1031	β -Phellandrene	2.8	2.8
1035	1034	(<i>Z</i>)- β -Ocimene	0.4	0.1
1046	1045	(<i>E</i>)- β -Ocimene	0.2	tr
1057	1057	γ -Terpinene	0.1	0.1
1085	1086	Terpinolene	0.6	0.5
1090	1090	2-Nonanone	0.1	0.2
1096	1099	6-Camphenone	0.1	0.1
1100	1101	Linalool	0.7	0.1
1101	1100	Undecane	tr	0.3
1119	1119	<i>endo</i> -Fenchol	0.1	0.1
1124	1124	<i>cis-p</i> -Menth-2-en-1-ol	tr	tr
1126	1126	α -Campholenal	0.1	0.1
1127	1127	<i>allo</i> -Ocimene	tr	tr
1140	1141	<i>trans</i> -Pinocarveol	tr	tr
1142	1142	<i>trans-p</i> -Menth-2-en-1-ol	0.1	0.1
1146	1145	Camphor	tr	0.1
1148	1148	<i>neo</i> -Isopulegol	tr	tr
1152	1152	Citronellal	0.1	0.1
1155	1156	Camphene hydrate	0.6	0.7
1158	1160	<i>iso</i> -Isopulegol	tr	tr
1161	1164	Pinocarvone	tr	tr
1163	1165	<i>iso</i> -Borneol	tr	tr
1171	1173	Borneol	0.2	0.4
1180	1180	Terpinen-4-ol	0.3	0.2
1186	1187	Cryptone	0.1	0.1
1196	1198	α -Terpineol	1.7	1.3
1202	1203	<i>p</i> -Cumenol	tr	tr
1206	1206	Decanal	tr	tr
1217	1217	<i>endo</i> -Fenchyl acetate	tr	tr
1227	1227	Citronellol	0.4	0.4
1285	1285	Bornyl acetate	14.6	22.1
1287	1287	<i>iso</i> -Bornyl acetate	tr	tr
1292	1294	2-Undecanone	---	0.1
1301	1300	Tridecane	0.1	0.3
1349	1350	Citronellyl acetate	0.5	0.6
1377	1378	Geranyl acetate	0.3	0.1
1388	1390	<i>trans</i> - β -Elemene	0.1	tr
1409	1409	Decanal	---	0.1
1418	1417	(<i>E</i>)- β -Caryophyllene	0.2	0.4
1428	1430	γ -Elemene	0.1	tr
1440	1442	Guaia-6,9-diene	0.1	tr
1447	1447	<i>iso</i> -Germacrene D	tr	tr
1454	1454	α -Humulene	0.1	0.1

(Continued)

Table 1. Continued

RI _{calc}	RI _{db}	Compound	% Composition	
			#1	#2
1473	1475	γ -Muuroolene	tr	0.1
1477	1479	α -Amorphene	0.1	tr
1479	1480	Germacrene D	tr	0.1
1480	1476	Selina-4,11-diene	0.5	0.4
1485	1488	δ -Selinene	0.1	0.1
1487	1487	β -Selinene	0.1	0.1
1490	1490	γ -Amorphene	tr	tr
1491	1492	Valencene	tr	tr
1494	1497	α -Selinene	0.3	0.3
1496	1497	α -Muuroolene	tr	tr
1511	1512	γ -Cadinene	tr	0.1
1516	1518	δ -Cadinene	0.2	0.2
1546	1546	α -Elemol	tr	tr
1556	1557	Germacrene B	0.1	tr
1559	1560	(<i>E</i>)-Nerolidol	tr	0.1
1579	1582	Caryophyllene oxide	tr	tr
1621	1624	Selina-6-en-4 β -ol	0.2	0.9
1640	1640	τ -Cadinol	tr	tr
1642	1644	τ -Muurolol	tr	0.1
1652	1655	α -Cadinol	0.1	0.1
1662	1660	Intermedeol	3.3	3.1
1695	1696	Eudesm-7(11)-en-4-ol	0.1	tr
1988	1989	Manoyl oxide	tr	tr
		Monoterpene hydrocarbons	74.2	66.4
		Oxygenated monoterpenoids	19.9	26.3
		Sesquiterpene hydrocarbons	1.8	2.0
		Oxygenated sesquiterpenoids	3.7	4.3
		Diterpenoids	tr	tr
		Others	0.2	1.0
		Total identified	99.9	99.9

RI_{calc} = Retention index calculated using a homologous series of *n*-alkanes on a ZB-5 ms column. RI_{db} = Retention index obtained from the databases.¹³⁻¹⁶ tr = trace (< 0.05%).

ornamental shade tree.¹⁰ In this work, the essential oil from the leaves and twigs of *A. concolor*, collected from two individuals growing in suburban Kuna, Idaho, were obtained and analyzed using gas chromatographic techniques.

Results and Discussion

Essential Oil Composition

The foliar essential oils from two different *A. concolor* trees were obtained as colorless liquids in 0.737% and 0.895% yield. The essential oil compositions of two different samples of *A. concolor* are presented in Table 1. The major components were α -pinene (20.5%, 15.2%), camphene (7.5%, 10.2%), β -pinene (25.6%, 24.2%), δ -3-carene (6.5%, 5.5%), limonene (6.9%, 5.4%), and bornyl acetate (14.6%, 22.1%). The essential oil compositions of the Kuna *A. concolor* samples are in qualitative agreement with several essential oils growing in urban locations in Belarus, which showed α -pinene (7.3-13.2%), camphene

(12.8-17.6%), β -pinene (28.0-35.9%), δ -3-carene (0.2-1.3%), limonene (5.4-7.2%), and bornyl acetate (17.5-25.4%).¹¹ Likewise, *A. concolor* foliar essential oil from the Kaibab Plateau, Arizona, showed α -pinene (21.0-25.8%), camphene (0.9-5.8%), β -pinene (45.6-57.4%), δ -3-carene (0.0-0.2%), and limonene (10.7-13.5%); neither β -phellandrene nor bornyl acetate were reported, however.¹²

Based on leaf morphology¹⁷ the collected plants in this study are *A. concolor* subsp. *concolor*. However, based on geographical location,¹⁸ they may actually be a hybrid of *A. concolor* and *Abies grandis* (Douglas ex D. Don) Lindl. A comparison of the concentrations of the major components of *A. concolor* and *A. grandis* is summarized in Table 2. The α -pinene concentrations in *A. concolor* from this study are higher than those found in *A. concolor* subsp. *lowiana* from California or *A. concolor* subsp. *concolor* from Utah,¹⁹ but comparable to those reported for *A. concolor* from Arizona.¹² α -Pinene concentration was also low in *A. grandis* from Idaho.²⁰ Conversely, β -pinene concentrations were lower in *A. concolor* in this study compared to *A. concolor* from Arizona,¹² from California or from Utah,¹⁹ but comparable to *A. concolor* from Belarus¹¹ or *A. grandis* from Idaho.²⁰ Importantly, reported limonene concentrations are greater than β -phellandrene concentrations in *A. concolor*¹⁹ whereas β -phellandrene concentrations are greater than limonene concentrations for *A. grandis*.²⁰ In this present study, limonene concentrations were slightly greater than β -phellandrene and suggests an intermediate ratio. Likewise, the concentrations of bornyl acetate in this study are intermediate between those of *A. concolor* (from Arizona, California, or Utah) and that from *A. grandis* from Idaho. The volatile phytochemistry of *A. concolor* from southwestern Idaho in this study, then, may suggest these two samples are actually hybrids of *A. concolor* and *A. grandis*.

As a comparison, the major components of the leaf essential oils of commercial samples of *A. balsamea* from Canada and *A. sibirica* from Russia (unpublished data from samples from the collection of the Aromatic Plant Research Center, Lehi, Utah) are also summarized in Table 2. Based on these data, the essential oil compositions of *A. concolor* from southwestern Idaho are more similar to those of *A. balsamea* than to *A. sibirica* oils.

Enantiomeric Distribution

Chiral gas chromatographic analysis was carried out to determine the enantiomeric distributions of the terpenoid constituents (Table 3). The (+)-enantiomer was predominant for α -pinene, in contrast to that found in the seed and cone essential oils of *A. concolor* from Łódź, Poland.²¹ (-)- α -Pinene also predominated in the leaf essential oil of *A. alba* from Poland,²² the seed essential oil of *A. koreana* from Poland,²³ and the leaf and shoot essential oils of *Abies veitchii* Lindl., *Abies mariesii* Mast., *A. sachalinensis* from Japan,²⁴ and *A. balsamea* from Canada (Table 3). On the other hand, (+)- α -pinene was the dominant enantiomer in the leaf essential oil of *A. sibirica* from Austria,²² *A. sibirica* from Russia (Table 3), *Abies firma* Siebold & Zucc. leaf essential oil from Japan,²⁴ and the leaf

essential oil of *Abies spectabilis* subsp. *langtangensis* (Silba) Silba from Nepal.²⁵ Thus, the enantiomeric distribution of α -pinene seems to vary between and within *Abies* species. (-)- β -Pinene was the dominant enantiomer in *A. concolor* essential oil from Kuna, as well as essential oils of *A. alba*, *A. sibirica*,²² *A. spectabilis*,²⁵ *A. koreana*,²⁶ *A. sachalinensis*,²⁷ *A. balsamea* (Table 3), and *A. procera*.²⁸ Interestingly, (+)- β -pinene was more abundant than the (-)-enantiomer in the seed and cone essential oils of *A. concolor* from Poland.²¹

The (-)-enantiomer of limonene and camphene predominated in *A. concolor* foliar essential oil in this study as well as in *A. concolor* seed and cone essential oils,²¹ *A. balsamea* (Table 3), *A. alba* and *A. sibirica* leaf essential oils,²² *A. koreana* seed essential oil,²³ and *A. firma*, *A. veitchii*, *A. mariesii*, and *A. schalinensis* leaf essential oils.²⁴ On the other hand, (+)-camphene was the major enantiomer in *A. spectabilis*,²⁵ *A. procera*,²⁸ and Russian *A. sibirica* (Table 3) essential oils. The only enantiomer of δ -3-carene in *A. concolor* foliar essential oil was (+)- δ -3-carene, consistent with that observed for *A. firma*, *A. mariesii*,²⁴ *A. spectabilis* subsp. *langtangensis*,²⁵ *A. procera*,²⁸ *A. balsamea* and *A. sibirica* (Table 3) essential oils. Both (-)-borneol and (-)-bornyl acetate were the exclusive enantiomers in *A. concolor* essential oil, which was also observed in *A. procera* essential oil²⁸ as well as *A. balsamea* and *A. sibirica* commercial essential oils (Table 3).

Conclusions

The essential oil compositions and enantiomeric distributions of terpenoids for *Abies concolor*, growing in a suburban environment in southwestern Idaho have been presented. The compositions are qualitatively similar to other *Abies* essential oils. However, a quantitative comparison of the major components with other *Abies concolor* and *Abies grandis* may indicate the southwestern Idaho samples to be hybrids of *A. concolor* and *A. grandis*. The enantiomeric distributions for α -pinene, β -pinene, and camphene did reveal variation between *Abies* species. The similarities between *A. concolor* and *A. balsamea* essential oils are notable and account for the similar traditional uses of these two species.

Materials and Methods

Plant Material

Branch tips (leaves and stems) of *A. concolor* were obtained from two different mature trees growing in Kuna, Idaho (43° 30'11"N, 116°25'36"W, 823 m elevation) on 26 July, 2021. The trees were identified by Kathy Swor and William Setzer in the field and verified by Daniel Murphy, Collections Curator, Idaho Botanical Garden, based on leaf morphology.¹⁷ Geographical location, however, suggests that this may be an *A. concolor* - *A. grandis* hybrid.¹⁸ A voucher specimen (WNS22143658) has been deposited in the herbarium of the University of Alabama in Huntsville (HALA). The fresh foliar materials (148.8 and 214.0 g, respectively) were hydrodistilled

Table 2. Comparison of Concentration Ranges (%) of Major Components of *Abies* Essential Oils.

Compounds	<i>A. concolor</i> ^a	<i>A. concolor</i> ^b	<i>A. concolor</i> ^c	<i>A. concolor</i> subsp. <i>lowiana</i> ^d	<i>A. concolor</i> subsp. <i>concolor</i> ^e	<i>A. grandis</i> ^f	<i>A. balsamea</i> ^g	<i>A. sibirica</i> ^h
α-Pinene	15.2, 20.5	21.0-25.8	7.3-13.2	4.4-5.1	8.9	4.4	17.8-31.3	10.8-12.0
Camphene	7.5, 10.2	0.9-5.8	12.8-17.6	1.4-2.7	2.9	10.7	0.6-4.2	19.3-22.0
β-Pinene	24.2, 25.6	45.6-57.4	28.0-35.9	42.0-47.1	43.9	20.3	22.1-32.8	1.7-2.3
δ-3-Carene	5.5, 6.5	0.0-0.2	0.2-1.3	0.1-0.2	1.6	0.7	1.4-9.8	10.8-12.6
Limonene	5.4, 6.9	10.7-13.5	5.4-7.2	17.6-23.0	9.3	2.1	11.9-23.3	4.5-5.6
β-Phellandrene	2.8, 2.8	n.r.	n.r.	2.0-2.5	1.1	18.9	6.3-12.2	2.8-3.8
Bornyl acetate	14.6, 22.1	n.r.	17.5-25.4	1.2-6.6	6.4	26.2	0.3-6.4	30.2-34.8

^aThis work. ^b From Arizona. ¹² ^c From Belarus. ¹¹ ^d From California. ¹⁹ ^e From Utah. ¹⁹ ^f From northern Idaho. ²⁰ ^g Based on 10 commercial samples from Canada. ^h Based on 15 commercial samples from Russia. n.r. = not reported.

Table 3. Enantiomeric Distribution of Chiral Terpenoid Components in the Essential Oils of *Abies* Species.

Compounds	Content, (+) : (-)							
	<i>Abies concolor</i> ^a		<i>Abies balsamea</i> ^b				<i>Abies sibirica</i> ^c	
	Sample #1	Sample #2	Sample A	Sample B	Sample C	Sample D	Sample A	Sample B
α-Thujene	16.3 : 83.7	15.5 : 84.5	11.3 : 88.7	13.4 : 86.6	12.4 : 87.6	12.5 : 87.5	100 : 0	100 : 0
α-Pinene	75.1 : 24.9	68.9 : 31.1	30.6 : 69.4	33.5 : 66.5	36.4 : 63.6	33.6 : 66.4	55.0 : 45.0	54.9 : 45.1
Camphene	4.5 : 95.5	2.8 : 97.2	26.9 : 73.1	27.3 : 72.7	26.3 : 73.7	26.6 : 73.4	96.6 : 3.4	96.6 : 3.4
β-Pinene	1.4 : 98.6	1.3 : 98.7	1.2 : 98.8	1.5 : 98.5	1.3 : 98.7	1.5 : 98.5	4.5 : 95.5	4.0 : 96.0
δ-3-Carene	100 : 0	100 : 0	100 : 0	100 : 0	100 : 0	100 : 0	100 : 0	100 : 0
Limonene	10.2 : 89.8	12.1 : 87.9	2.5 : 97.5	3.5 : 96.5	3.4 : 96.6	3.7 : 96.3	0 : 100	0 : 100
β-Phellandrene	0.9 : 99.1	0.4 : 99.6	0.5 : 99.5	0.3 : 99.7	0.6 : 99.4	0.4 : 99.6	0.5 : 99.5	0.5 : 99.5
Linalool	71.1 : 28.9	---	---	---	---	---	---	---
Borneol	0 : 100	0 : 100	0 : 100	0 : 100	0 : 100	0 : 100	0 : 100	0 : 100
Terpinen-4-ol	37.1 : 62.9	40.5 : 59.5	28.6 : 71.4	30.3 : 69.7	28.2 : 71.8	27.9 : 72.1	---	---
α-Terpineol	14.8 : 85.2	12.9 : 87.1	7.8 : 92.2	11.5 : 88.5	10.8 : 89.2	9.2 : 90.8	10.0 : 90.0	15.4 : 84.6
Citronellol	34.6 : 65.4	34.4 : 65.6	---	---	---	---	---	---
Bornyl acetate	0 : 100	0 : 100	0 : 100	0 : 100	0 : 100	0 : 100	0 : 100	0 : 100
Citronellyl acetate	0 : 100	0 : 100	---	---	---	---	---	---
(<i>E</i>)-β-Caryophyllene	0 : 100	0 : 100	---	---	---	---	---	---
δ-Cadinene	0 : 100	0 : 100	---	---	---	---	---	---

^aThis work. ^b Commercial samples from Canada. ^c Commercial samples from Russia.

using a Likens-Nickerson apparatus with continuous extraction with dichloromethane for 3 h to give, after evaporation of the solvent, colorless essential oils (1.097 and 1.915 g, respectively).

Gas Chromatography – Mass Spectrometry

The GC-MS analysis of the foliar essential oils of *A. concolor* samples were performed using a Shimadzu GCMS-QP2010 Ultra Gas Chromatography-Mass Spectrometry instrument (Shimadzu Scientific Instruments, Columbia, MD, USA) fitted with a ZB-5 ms fused silica capillary GC column, (5% Phenyl polymethylsiloxane, 60 m × 0.25 mm i.d. × 0.25 μm film thickness) (Phenomenex, Torrance, CA, USA). Helium (99.9%) was used as the carrier gas at a flow rate of 2.0 mL/min. Mass spectra were recorded at 70 eV, the mass range was from 40 to 400 *m/z*. The ion source, interface, and injector port temperatures were 260 °C. For each sample, a 5% *w/v* solution in CH₂Cl₂ was prepared, and 0.1 μL was injected using a split ratio of 1:24. The oven temperature was initially 50 °C,

increased to 260 °C at a rate of 2 °C/min. The components of the essential oils were identified by comparison of their mass spectral fragmentation patterns and on the basis their GC retention indices (RI), calculated using a homologous series of *n*-alkanes (C₈-C₄₀) injected under the same chromatographic conditions, with those available in the MS databases.¹³⁻¹⁶ The data were handled using the LabSolutions GCMS solution software version 4.45 (Shimadzu Scientific Instruments, Columbia, MD, USA).

Gas Chromatography with Flame Ionization Detection

The GC-FID analyses were carried out using a Shimadzu GC 2010 (Shimadzu Scientific Instruments, Columbia, MD, USA) equipped with flame ionization detector, a split/splitless injector, and Shimadzu autosampler AOC-20i, with a ZB-5 capillary column (60 m × 0.25 mm i.d.; film thickness 0.25 μm) (Phenomenex, Torrance, CA, USA). The oven temperature was programmed as above for GC-MS. Injector and detector

temperatures were maintained at 260 °C, respectively. The carrier gas was helium and the flow rate was 1.0 mL/min. For each sample, a 5% w/v solution in CH₂Cl₂ was prepared, and 0.1 µL was injected using a split ratio of 1:31. The percent compositions of the essential oils were calculated from peak areas with quantification using the external standard method; calibration curves of representative compounds from each class were used for quantification.

Chiral Gas Chromatography – Mass Spectrometry

The chiral GC-MS analysis was performed using a Shimadzu GCMS-QP2010S (Shimadzu Scientific Instruments, Columbia, MD, USA) gas chromatograph equipped with a Restek B-Dex 325 chiral capillary GC column (Restek Corp., Bellefonte, PA, USA) (30 m × 0.25 mm ID × 0.25 µm film thickness). The temperature of the injection was 260 °C. The samples were diluted to 3% w/v with CH₂Cl₂ and the injection volume was 0.1 µL using a split ratio of 1:25. Helium was used as carrier gas at a constant flow rate of 1.8 mL/min. The initial oven temperature was 50 °C and then increased at a rate of 1.5 °C/min to 120 °C, then heated to 260 °C at a rate of 2 °C/min, and then held at 200 °C for an addition 5 min. The temperature of the detector was 260 °C. The peaks were identified by comparing retention times (RT) with authentic samples obtained from Sigma-Aldrich (Milwaukee, WI, USA). Relative enantiomer percentages were determined based on peak areas.

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Author Contribution Statement

W.N.S. designed the study; K.S. and W.N.S. collected the plant material; W.N.S. carried out the hydrodistillations; A.P. and A.R. obtained the GC-MS, GC-FID, and chiral GC data; P.S. and W.N.S. analyzed the GC data. All authors contributed to the writing and editing of the manuscript.

Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical Approval

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Statement of Human and Animal Rights

This article does not contain any studies with human or animal subjects.

Trial Registration

Not applicable, because this article does not contain any clinical trials.

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