

Supplementary materials

Analytical and Preparative Separation of Softwood Lignans by Supercritical Fluid Chromatography

Nikolay V. Ul'yanovskii^{1,*}, Aleksandra A. Onuchina¹, Denis V. Ovchinnikov¹, Anna V. Faleva¹, Natalia S. Gorbova² and Dmitry S. Kosyakov^{1,*}

¹ Laboratory of Natural Product Chemistry and Bioanalytics, Core Facility Center "Arktika", Northern (Arctic) Federal University, Arkhangelsk 163002, Russia

² Federal Center for Integrated Arctic Research, Arkhangelsk 163000, Russia

*Correspondence: n.ulyanovsky@narfu.ru (N.V.U.); d.kosyakov@narfu.ru (D.S.K.)

Table S1. Low-purity preparative SFC fractions of coniferous knotwood extractives.

Tree Species	Fraction No.*	Purity, %	<i>m/z</i> [M – H] [–] , (Δ, ppm)	Elemental Composition	Assumed Compound
Pine	3	70	357.1338 (-1.3)	C ₂₀ H ₂₂ O ₆	Matairesinol
		30	255.1028 (0.5)	C ₁₆ H ₁₆ O ₃	Pterostilbene
Spruce	6	30	373.1295 (0.6)	C ₂₀ H ₂₂ O ₇	5-Hydroxymatairesinol
	7	-	-	-	Many compounds
	8	-	-	-	Many compounds
Fir	4	-	-	-	Many compounds
	5	-	-	-	Many compounds
	6	78	505.1886 (3.6)	C ₂₉ H ₃₀ O ₈	Sesquilignan
	8	14	557.2404 (2.1)	C ₃₀ H ₃₈ O ₁₀	Sesquilignan
		86	539.2286 (-0.1)	C ₃₀ H ₃₆ O ₉	Sesquilignan
Larch	1	66	373.1290 (-0.7)	C ₂₀ H ₂₂ O ₇	Nortrachelogenin
	3	-	-	-	Many compounds
	4	-	-	-	Many compounds
	5	-	-	-	Many compounds
	6	-	-	-	Many compounds
	7	-	-	-	Many compounds
	9	70	716.3066 (-1.0)	C ₄₀ H ₄₈ O ₁₂	Dilignan
	11	-	-	-	Many compounds

* - corresponds to the number of the fraction collection zone depicted in Figure 3.

Table S2. ¹H NMR spectra of the isolated pure compounds.

Fraction	δ ppm	Identified Compound
Pine (<i>Pinus Sylvestris</i>)		
1	3.74 (s, 3H) 6.28 (s, 1H) 6.60 (s, 1H) 6.65 (s, 1H) 7.14 (m, 2H) 7.26 (br t, J=7.18 Hz, 1H) 7.37 (br t, J=7.70 Hz, 2H) 7.59 (br d, J=7.70 Hz, 2H) 9.47 (br s, 1H)	Pinosylvin methyl ether
2	3.76 (s, 6H) 6.37 (s, 1H) 6.71 (s, 2H) 6.76 (br d, J=8.25 Hz, 2H) 6.94 (br d, J=16.51 Hz, 1H) 7.15 (br d, J=15.96 Hz, 1H) 7.41 (br d, J=8.80 Hz, 2H) 9.59 (br s, 1H)	Pterostilbene
4	2.25 - 2.31 (m, 1H) 2.35 (s, 1H) 2.59 (br dd, J=12.65, 3.30 Hz, 1H) 2.80 (br d, J=13.76 Hz, 1H) 2.97 (br d, J=13.75 Hz, 1H) 3.69 (s, 1H) 3.72 (s, 1H) 3.89 - 3.97 (m, 1H) 6.18 (s, 1H) 6.48 - 6.53 (m, 1H) 6.55 - 6.60 (m, 1H) 6.64 (br d, J=1.65 Hz, 1H) 6.64 - 6.68 (m, 1H) 6.71 - 6.74 (m, 1H) 8.72 (s, 1H) 8.81 (s, 1H)	Nortrachelogenin
5	6.16 (s, 1H) 6.44 (s, 1H) 7.04 (d, J=3.30 Hz, 2H) 7.25 (t, J=7.36 Hz, 1H) 7.36 (br t, J=7.43 Hz, 1H) 7.57 (br d, J=7.70 Hz, 1H) 9.24 (s, 1H)	Pinosylvin
Spruce (<i>Picea abies</i>)		
1	2.39 - 2.44 (m, 1H) 2.64 - 2.70 (m, 1H) 2.70 - 2.84 (m, 1H) 3.71 (br s, 1H) 3.71 (br s, 1H) 3.82 - 3.91 (m, 1H) 4.00 - 4.10 (m, 1H) 6.47 (br d, J=7.70 Hz, 1H) 6.56 - 6.59 (m, 1H) 6.60 (s, 1H) 6.65 (br d, J=7.70 Hz, 1H) 6.67 (br d, J=8.25 Hz, 1H) 6.73 (s, 1H) 8.75 (br s, 1H) 8.79 (br s, 1H);	Matairesinol
2	2.78 (dd, J=13.94, 9.17 Hz, 1H) 3.10 (br dd, J=13.94, 4.77 Hz, 1H) 3.41 - 3.44 (m, 1H) 3.65 (s, 1H) 3.86 (s, 3H) 4.10 (t, J=8.44 Hz, 1H) 4.30 (q, J=8.07 Hz, 1H) 4.52 (t, J=8.44 Hz, 1H) 6.55 - 6.60 (m, overlapped, 1H) 6.65 (s, 1H) 6.77 (br d, J=8.80 Hz, 1H) 7.26 (overlapped, 1H) 7.28 (s, 1H)	Oxomatairesinol
3	2.58 - 2.66 (m, 1H) 2.66 - 2.76 (m, 1H) 2.92 (t, J=13.90 Hz, 1H) 3.13 (br dd, J=15.96, 4.95 Hz, 2H) 3.78 (s, 3H) 3.83 (s, 3H) 3.92 (br d, J=11.00 Hz, 1H) 4.11 (t, J=9.26 Hz, 1H) 4.17 (t, J=9.26 Hz, 1H) 6.26 (s, 1H) 6.66 (br d, J=8.25 Hz, 1H) 6.70 (br s, 1H) 6.72 - 6.82 (m, overlapped 3H)	α -Conidendrin
4	2.63 (m, 1H) 2.69 - 2.79 (m, overlapped 1H) 2.84 - 2.95 (m, overlapped 2H) 3.76 (s, 3H) 3.80 (s, 3H) 4.05 (t, J=8.80 Hz, 1H) 4.07 - 4.15 (m, 1H) 4.64 (d, J=5.14 Hz, 1H) 6.48 (dd, J=8.07, 1.83 Hz, 1H) 6.56 (d, J=1.83 Hz, 1H) 6.64 (d, J=8.07 Hz, 1H) 6.73 (overlapped, 1H) 6.74 (overlapped, 1H) 6.77 (d, J=1.47 Hz, 1H)	Iso-HMR
5	2.52 - 2.63 (m, overlapped, 2H) 2.66 - 2.79 (m, overlapped, 2H) 3.76 (s, 3H) 3.78 (s, 3H) 4.16 (dd, J=9.17, 8.07 Hz, 1H) 4.29 - 4.39 (m, overlapped, 2H) 6.48 (dd, J=8.07, 1.83 Hz, 1H) 6.53 (d, J=1.83 Hz, 1H) 6.65 - 6.68 (m, overlapped, 1H) 6.66 - 6.69 (m, overlapped 1H) 6.71 (d, J=1.83 Hz, 1H) 6.74 (d, J=8.07 Hz, 1H)	5-HMR
Fir (<i>Abies sibirica</i>)		
1	2.63 (m, 1H) 2.69 - 2.79 (m, overlapped 1H) 2.84 - 2.95 (m, overlapped 2H) 3.76 (s, 3H) 3.80 (s, 3H) 4.05 (t, J=8.80 Hz, 1H) 4.07 - 4.15 (m, 1H) 4.64 (d, J=5.14 Hz, 1H) 6.48 (dd,	Iso-HMR

	J=8.07, 1.83 Hz, 1H) 6.56 (d, J=1.83 Hz, 1H) 6.64 (d, J=8.07 Hz, 1H) 6.73 (overlapped, 1H) 6.74 (overlapped, 1H) 6.77 (d, J=1.47 Hz, 1H)	
2	2.18 (t, J=6.88 Hz, 1H) 2.41 (dd, J=13.48, 11.28 Hz, 1H) 2.54 - 2.60 (m, 1H) 2.81 (dd, J=13.48, 4.68 Hz, 1H) 3.45 (br s, 1H) 3.55 (dd, J=7.98, 6.88 Hz, 1H) 3.62 - 3.67 (m, 1H) 3.74 (s, 3H) 3.74 (s, 3H) 3.84 - 3.88 (m, 1H) 4.58 - 4.71 (m, 2H) 6.57 (dd, J=8.25, 1.65 Hz, 1H) 6.65 - 6.71 (m, overlapped 3H) 6.74 (d, J=1.65 Hz, 1H) 6.82 (d, J=1.65 Hz, 1H) 8.68 (s, 1H) 8.81 (s, 1H)	Lariciresinol
3	(600 MHz, DMSO-d6) δ ppm 1.82 (br t, J=5.78 Hz, 2H) 2.45 - 2.61 (m, overlapped 4H) 3.28 - 3.43 (m, 4H) 3.68 (s, 6H) 4.51 (br t, J=4.68 Hz, 2H) 6.49 (dd, J=7.70, 1.65 Hz, 2H) 6.62 (br s, 2H) 6.63 (overlapped, 2H) 8.61 (br s, 2H);	Secoisolariciresinol
7	1.83 (br t, J=5.78 Hz, 2H) 2.50 (m, overlapped 4H) 3.23 (m, overlapped, 2H) 3.28 - 3.43 (m, overlapped 4H) 3.53 - 3.60 (m, overlapped 2H) 3.65 - 3.76 (s, overlapped 9H) 4.17 (m, overlapped 1H) 4.71 (m, overlapped 1H) 6.49 (br d, J=7.70 Hz, 1H) 6.55 - 6.61 (m, 1H) 6.61 - 6.64 (m, 2H) 6.68 (br d, J=9.35 Hz, 2H) 6.76 (br d, J=7.70 Hz, 1H) 6.89 (br d, J=8.25 Hz, 1H) 6.98 (br s, 1H)	Secoisolariciresinol-sesquilignan
9	1.88 (br t, J=5.78 Hz, 4H) 2.53 - 2.63 (m, 4H) 3.26 - 3.46 (overlapped 8H) 3.67 (s, 6H) 3.74 (s, 6H) 4.52 (br t, J=4.68 Hz, 4H) 6.53 (br d, J=7.70 Hz, 2H) 6.54 - 6.57 (overlapped 2H) 6.62 (overlapped 4H) 6.66 (br s, 2H)	Dilignan, disecoisolariciresinol, 5-5 bond
Larch (<i>Larix sibirica</i>)		
2	1.82 (br t, J=5.78 Hz, 2H) 2.45 - 2.61 (m, overlapped 4H) 3.28 - 3.43 (m, 4H) 3.68 (s, 6H) 4.51 (br t, J=4.68 Hz, 2H) 6.49 (dd, J=7.70, 1.65 Hz, 2H) 6.62 (br s, 2H) 6.63 (overlapped, 2H) 8.61 (br s, 2H)	Secoisolariciresinol
8	1.83 (br t, J=5.78 Hz, 2H) 2.50 (m, overlapped 4H) 3.23 (m, overlapped, 2H) 3.28 - 3.43 (m, overlapped 4H) 3.53 - 3.60 (m, overlapped 2H) 3.65 - 3.76 (s, overlapped 9H) 4.17 (m, overlapped 1H) 4.71 (m, overlapped 1H) 6.49 (br d, J=7.70 Hz, 1H) 6.55 - 6.61 (m, 1H) 6.61 - 6.64 (m, 2H) 6.68 (br d, J=9.35 Hz, 2H) 6.76 (br d, J=7.70 Hz, 1H) 6.89 (br d, J=8.25 Hz, 1H) 6.98 (br s, 1H)	Secoisolariciresinol-sesquilignan
10	4.48 (br dd, J=11.00, 5.50 Hz, 1H) 4.96 (br d, J=11.00 Hz, 1H) 5.72 (br d, J=5.50 Hz, 1H) 5.83 (s, 1H) 5.87 (s, 1H) 6.74 (m, overlapped 2H) 6.87 (s, 1H)	Taxifolin

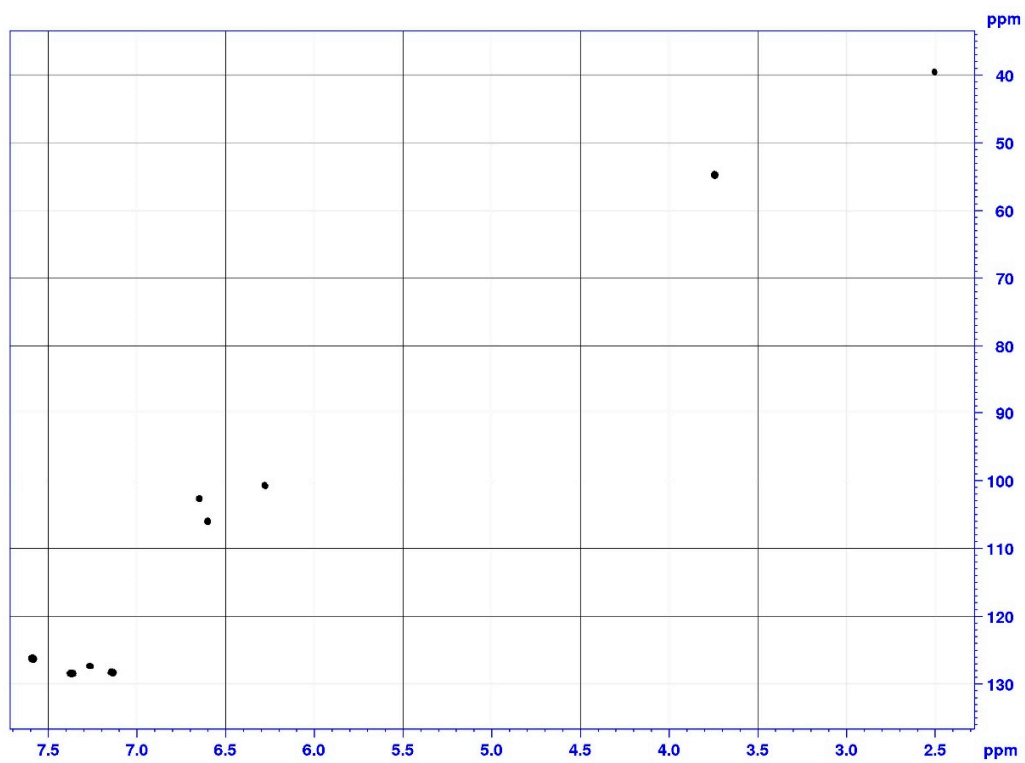


Figure S1. ^1H - ^{13}C HSQC NMR spectrum of Fraction 1 isolated from pine (*Pinus Sylvestris*) extract.

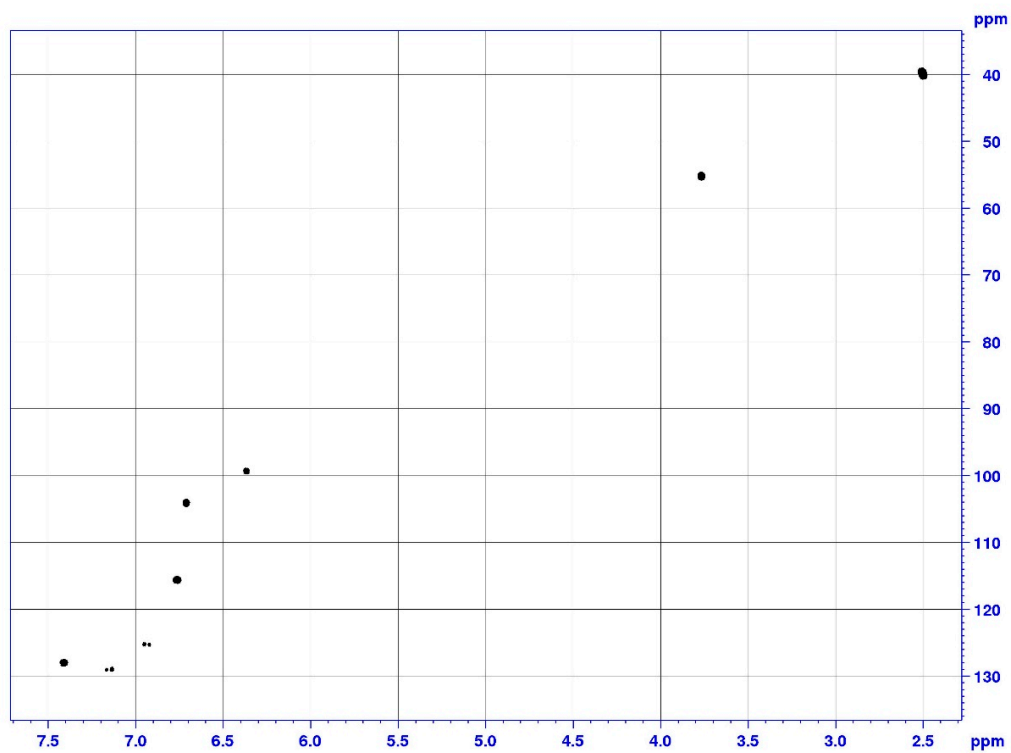


Figure S2. ^1H - ^{13}C HSQC NMR spectrum of Fraction 2 isolated from pine (*Pinus Sylvestris*) extract.

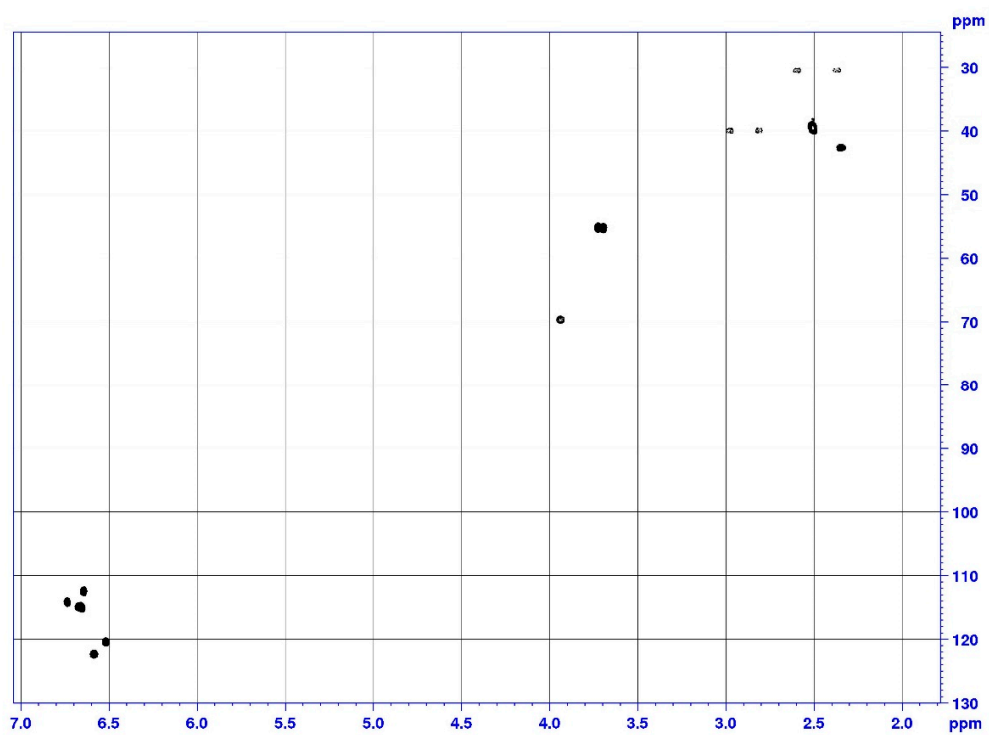


Figure S3. ^1H - ^{13}C HSQC NMR spectrum of Fraction 4 isolated from pine (*Pinus Sylvestris*) extract.

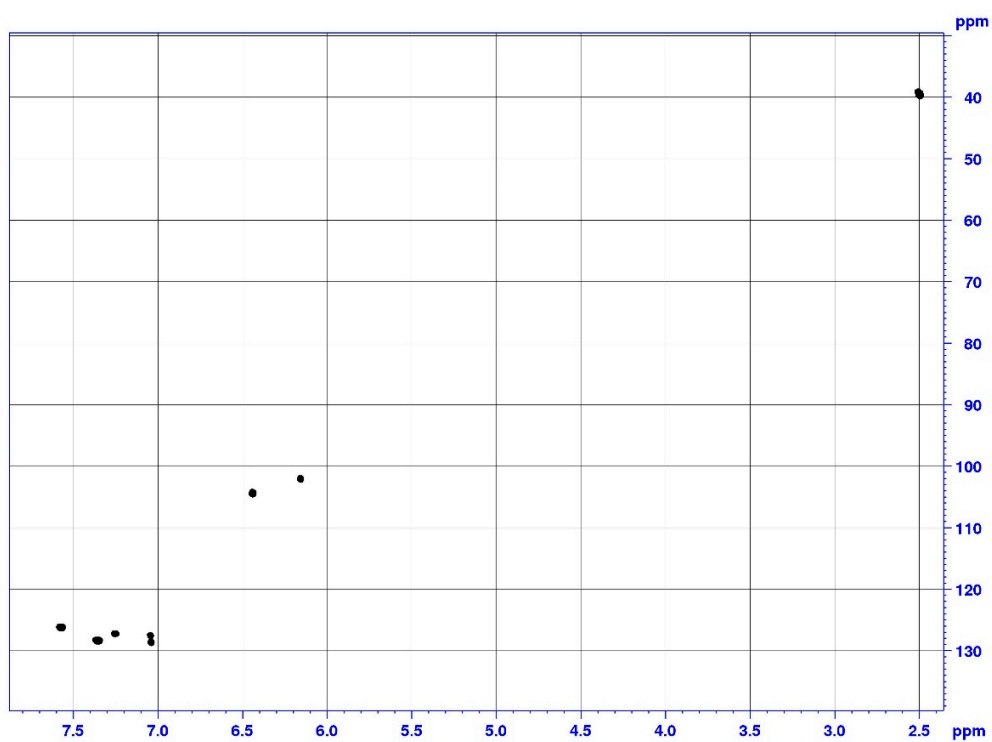


Figure S4. ^1H - ^{13}C HSQC NMR spectrum of Fraction 5 isolated from pine (*Pinus Sylvestris*) extract.

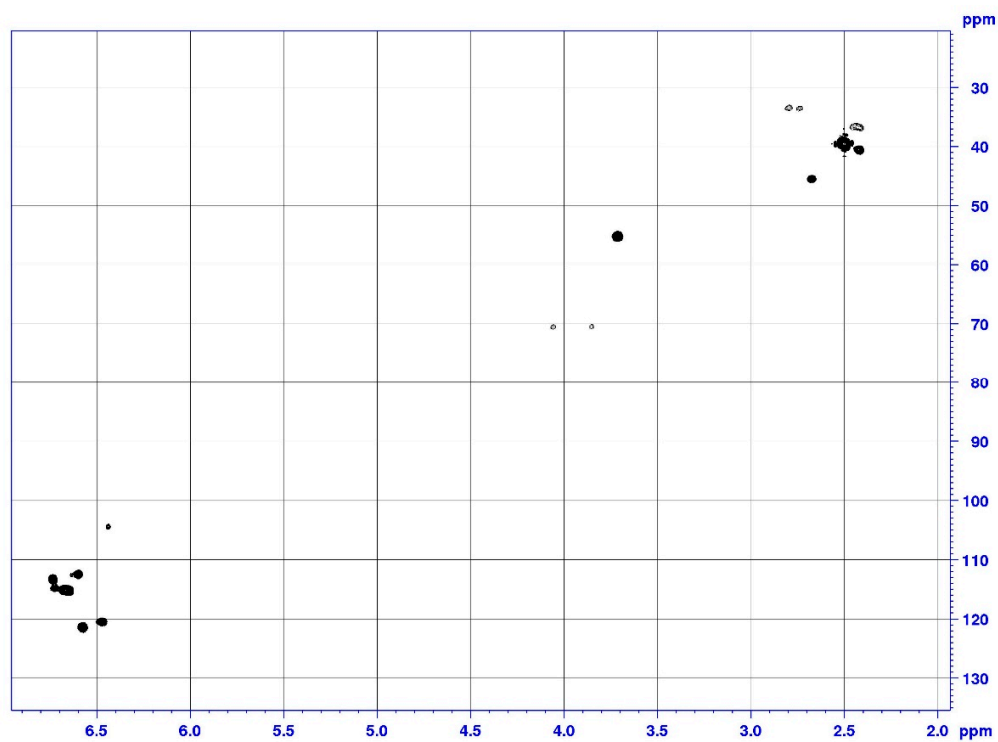


Figure S5. ^1H - ^{13}C HSQC NMR spectrum of Fraction 1 isolated from spruce (*Picea abies*) extract.

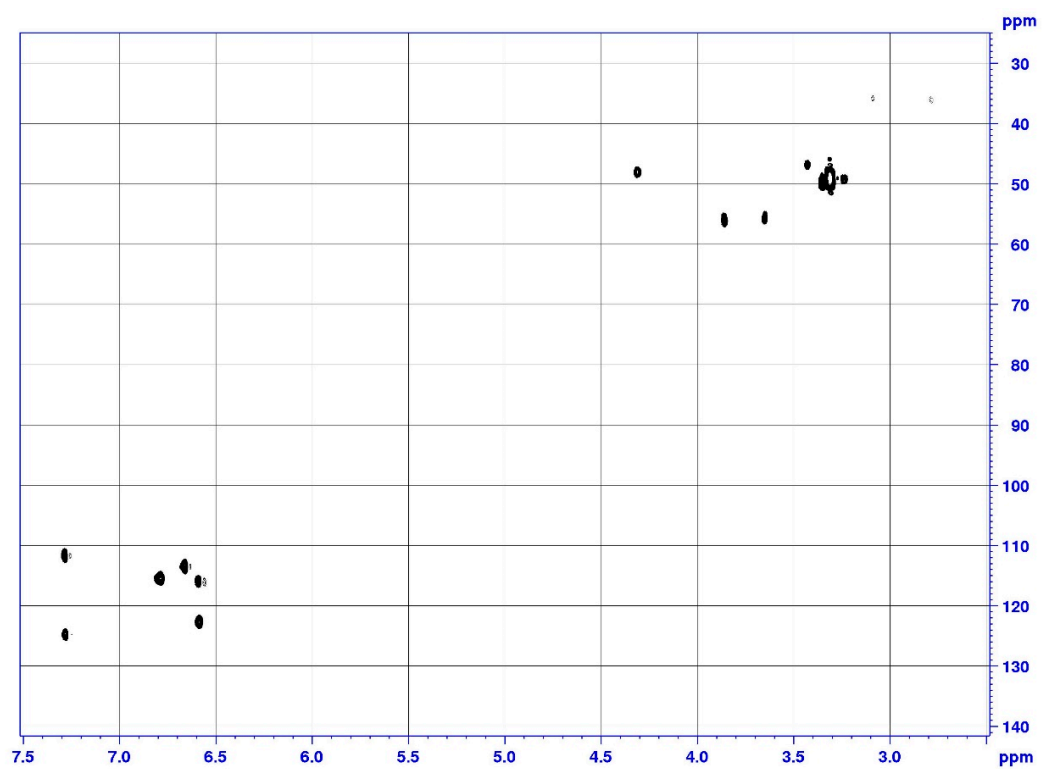


Figure S6. ^1H - ^{13}C HSQC NMR spectrum of Fraction 2 isolated from spruce (*Picea abies*) extract.

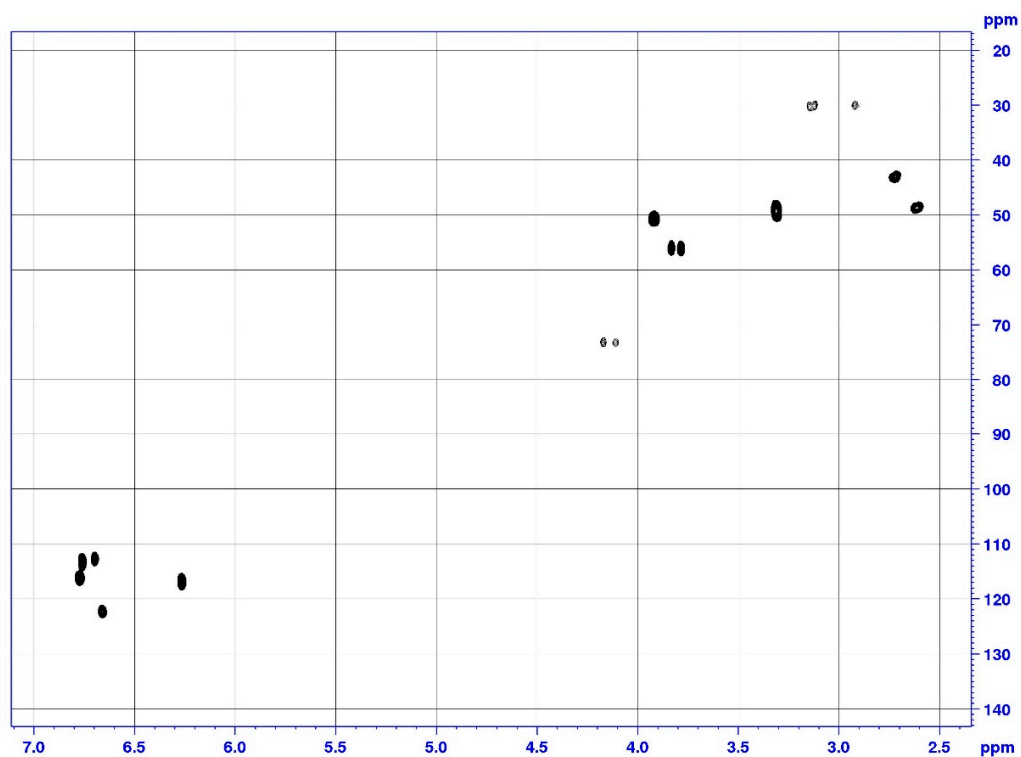


Figure S7. ^1H - ^{13}C HSQC NMR spectrum of Fraction 3 isolated from spruce (*Picea abies*) extract.

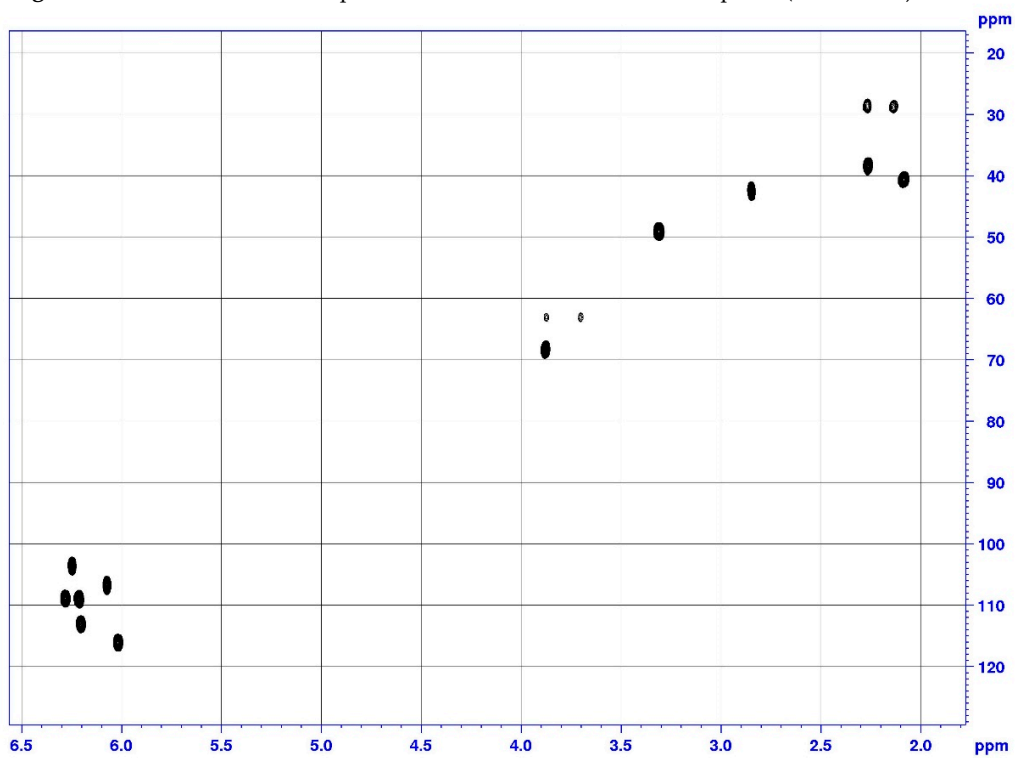


Figure S8. ^1H - ^{13}C HSQC NMR spectrum of Fraction 4 isolated from spruce (*Picea abies*) extract.

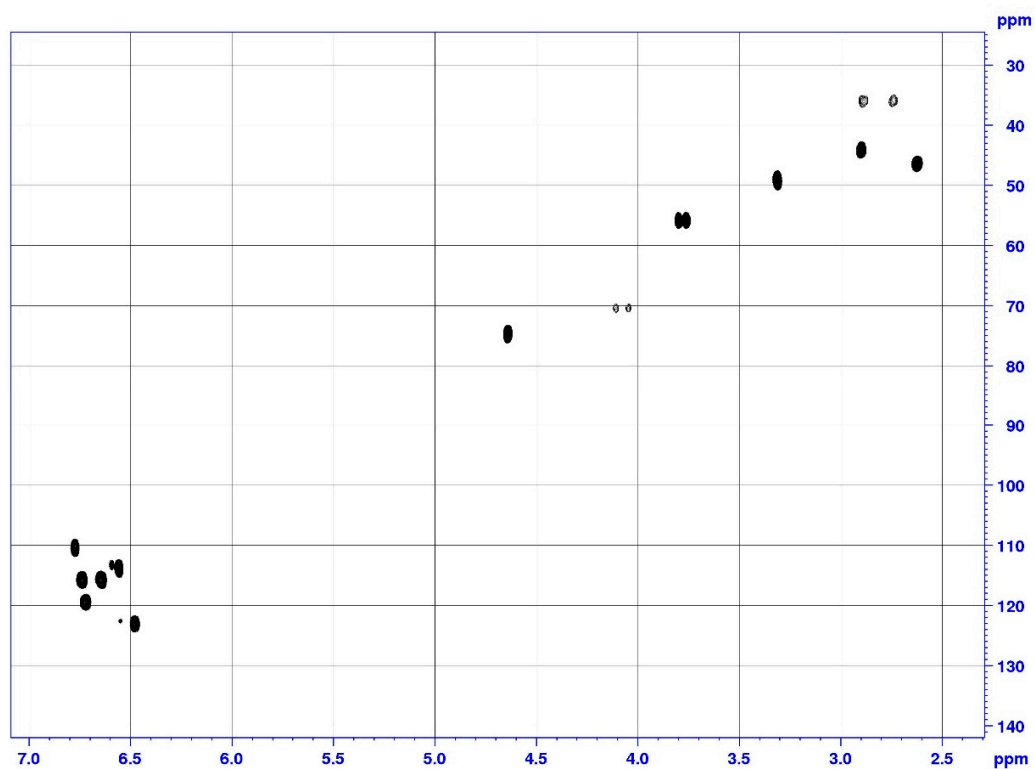


Figure S9. ^1H - ^{13}C HSQC NMR spectrum of Fraction 5 isolated from spruce (*Picea abies*) extract.

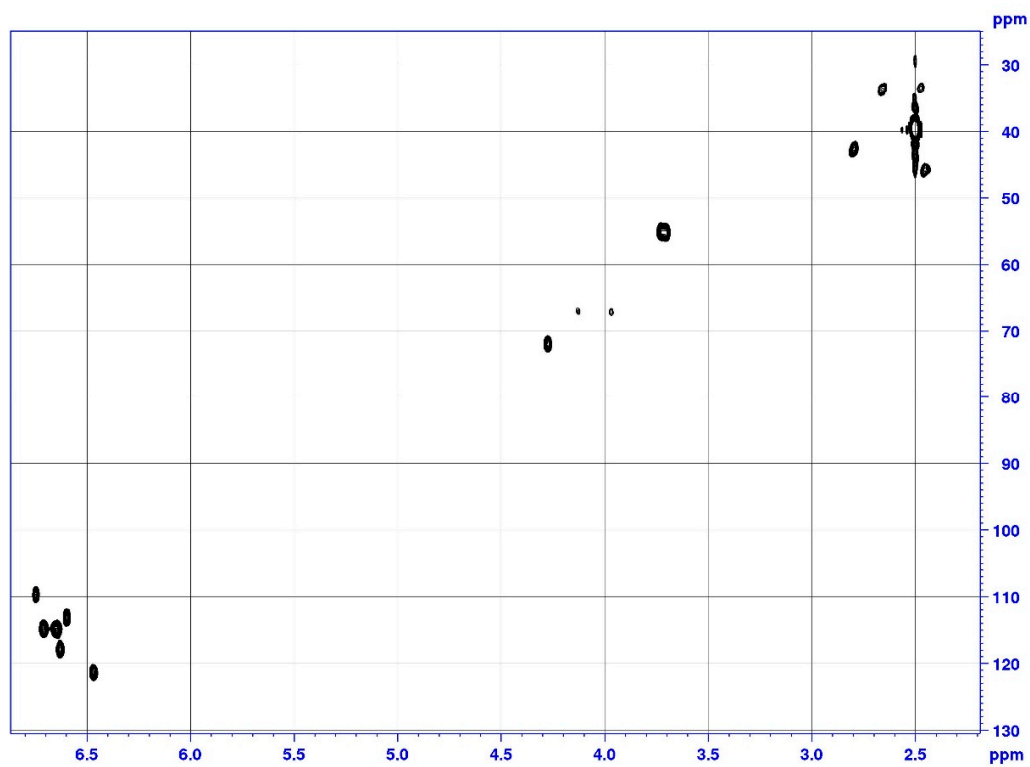


Figure S10. ^1H - ^{13}C HSQC NMR spectrum of Fraction 1 isolated from fir (*Abies sibirica*) extract.

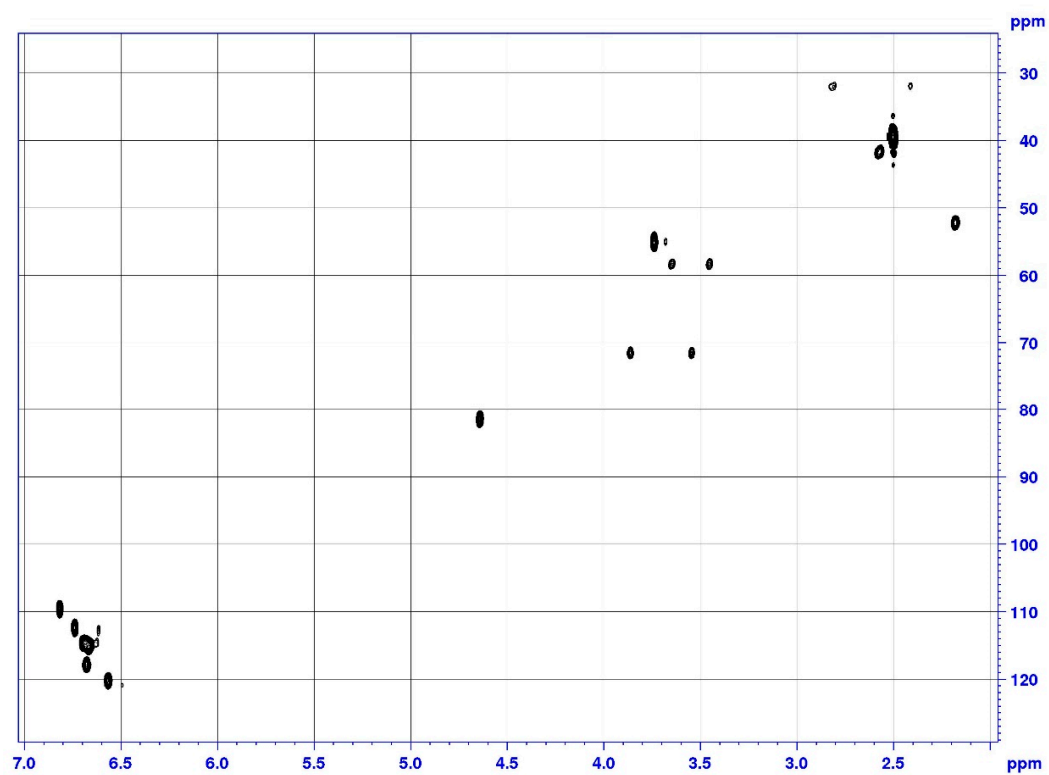


Figure S11. ^1H - ^{13}C HSQC NMR spectrum of Fraction 2 isolated from fir (*Abies sibirica*) extract.

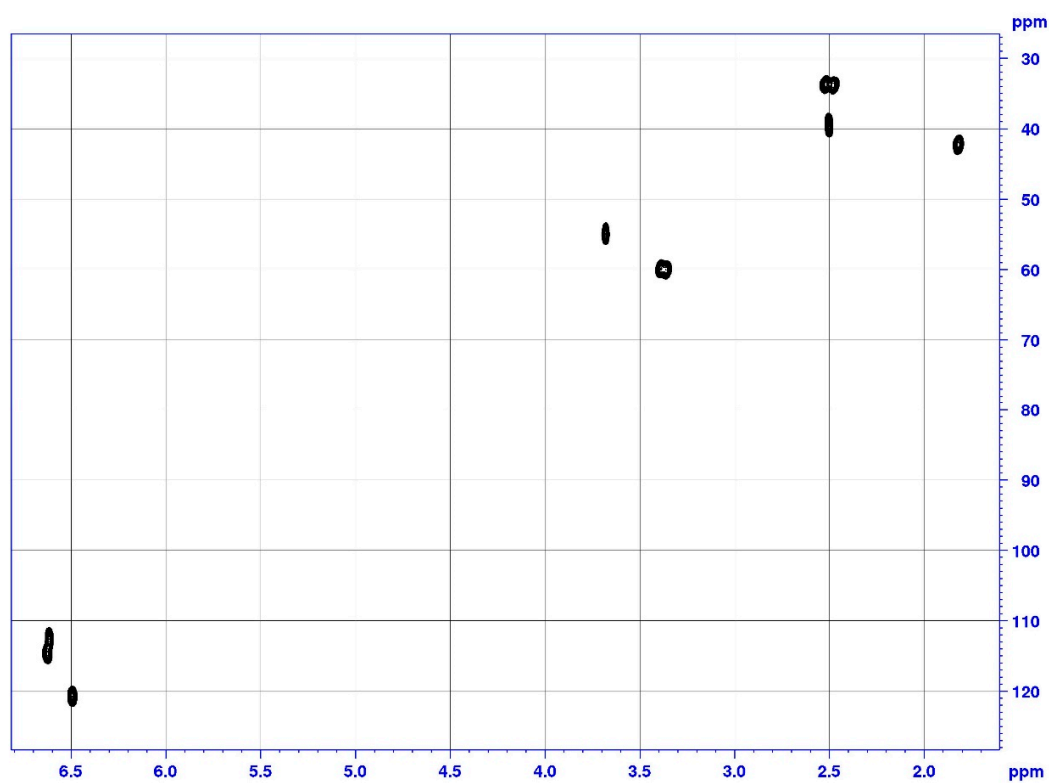


Figure S12. ^1H - ^{13}C HSQC NMR spectrum of Fraction 3 isolated from fir (*Abies sibirica*) extract.

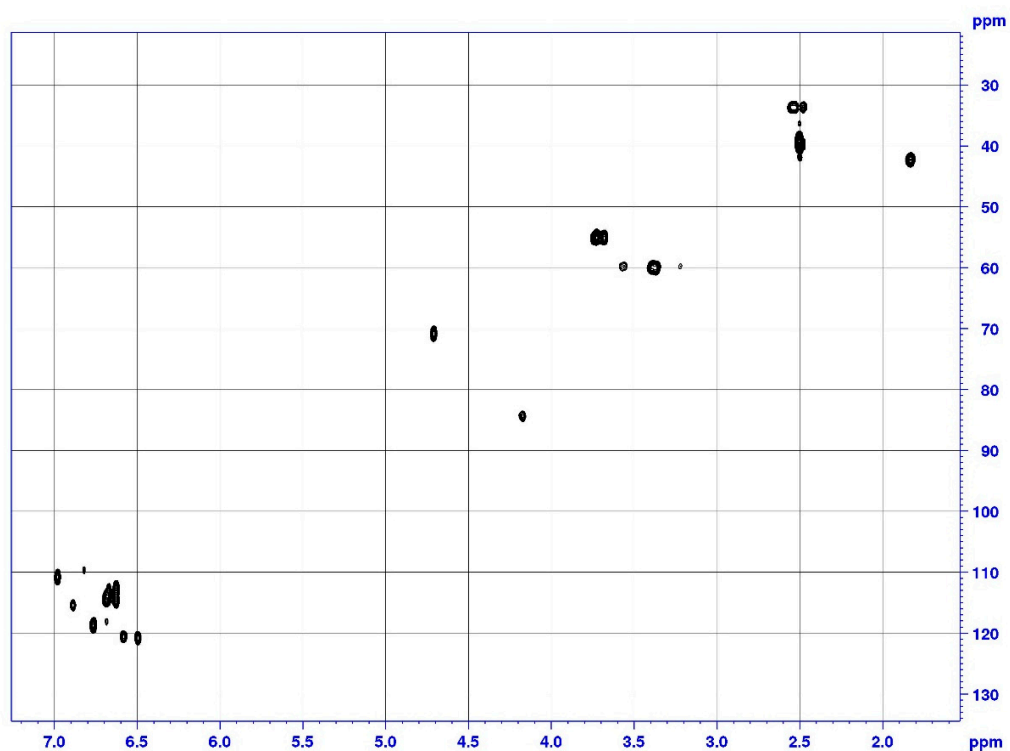


Figure S13. ^1H - ^{13}C HSQC NMR spectrum of Fraction 7 isolated from fir (*Abies sibirica*) extract.

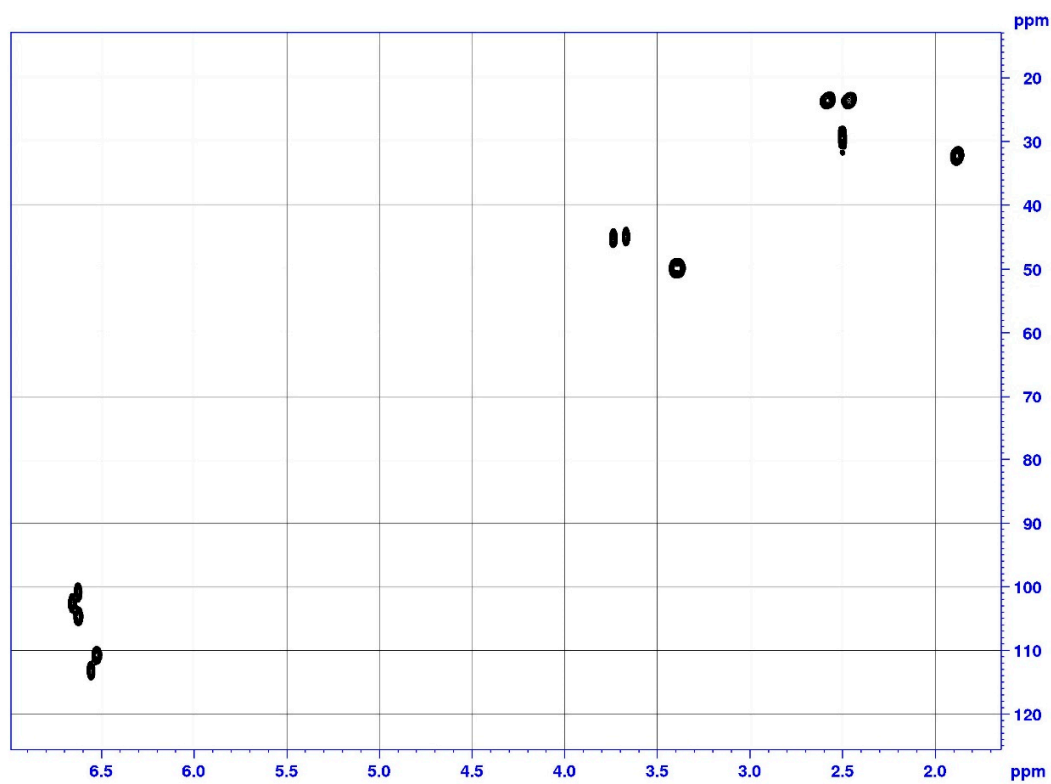


Figure S14. ^1H - ^{13}C HSQC NMR spectrum of Fraction 9 isolated from fir (*Abies sibirica*) extract.

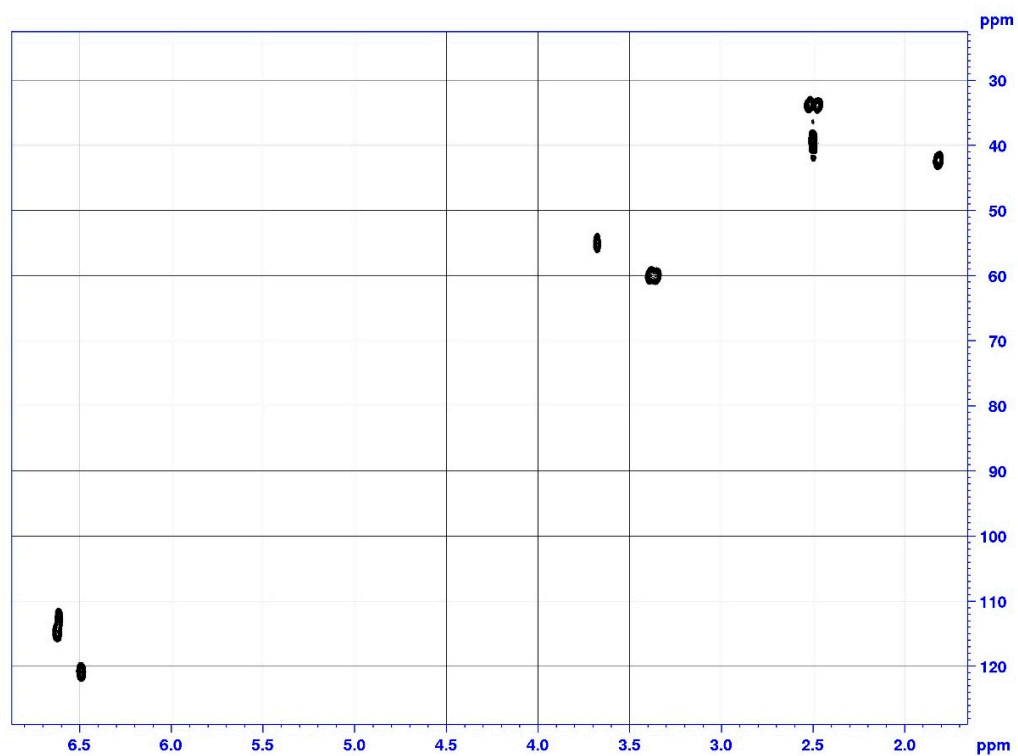


Figure S15. ^1H - ^{13}C HSQC NMR spectrum of Fraction 2 isolated from larch (*Larix sibirica*) extract.

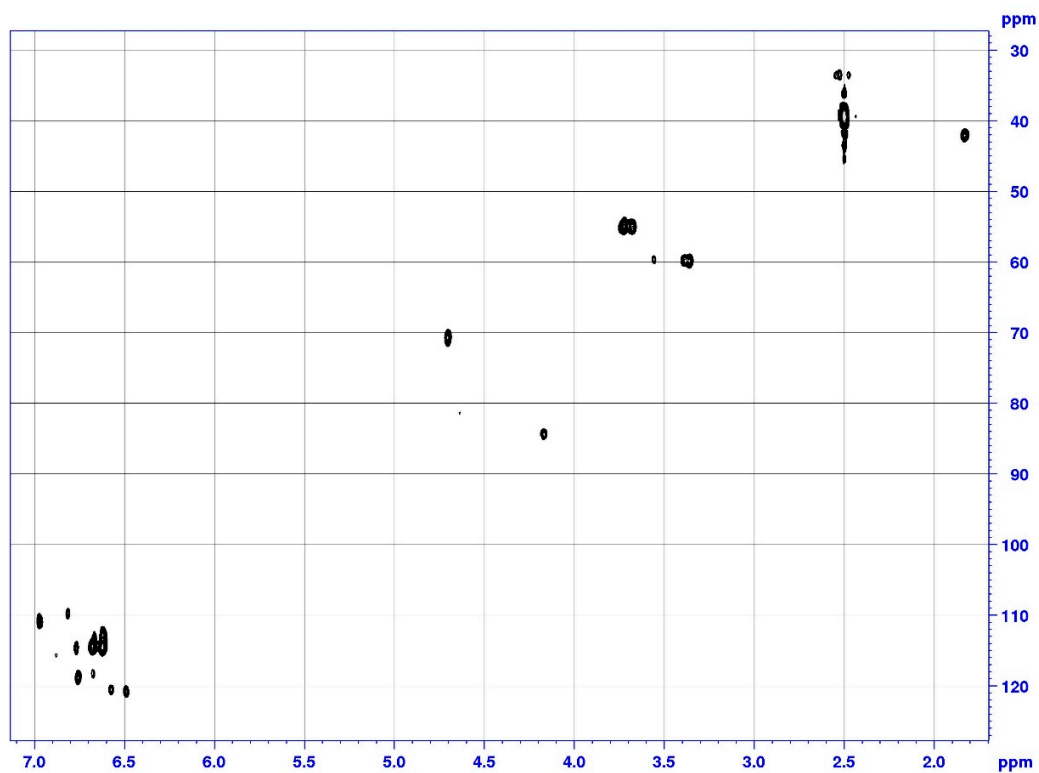


Figure S16. ^1H - ^{13}C HSQC NMR spectrum of Fraction 8 isolated from larch (*Larix sibirica*) extract.

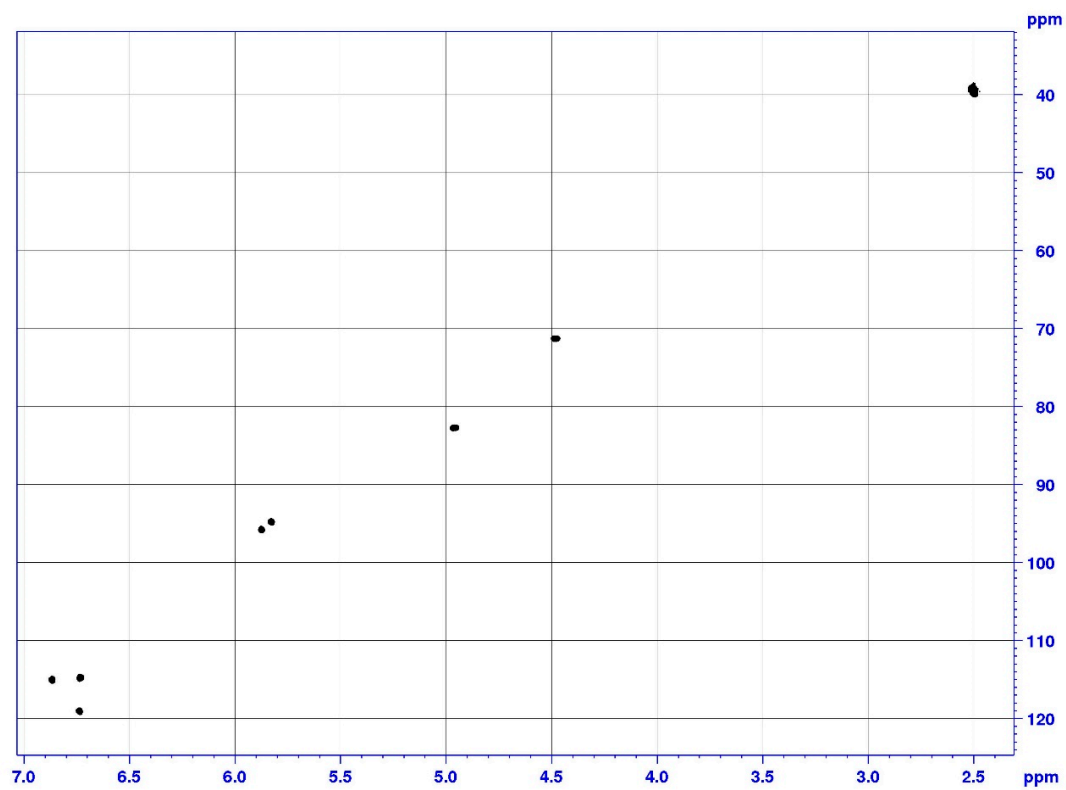


Figure S17. ^1H - ^{13}C HSQC NMR spectrum of Fraction 10 isolated from larch (*Larix sibirica*) extract.