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Classification of Chemicals in Black Locust *Robinia pseudoacacia* Wood and Bark

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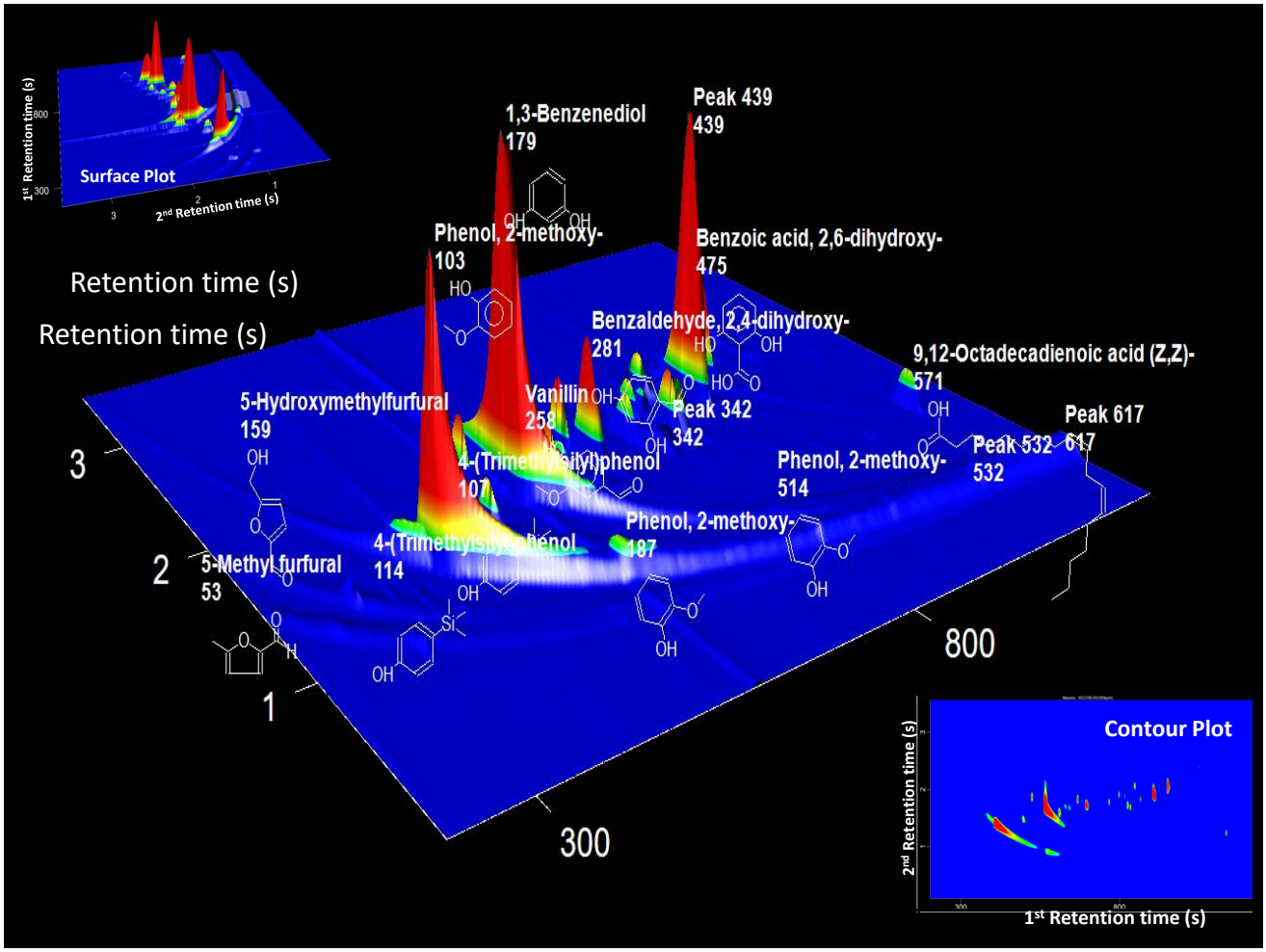

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Abstract

Black locust (*Robinia pseudoacacia*) is a native tree in southeastern United States. The wood from this tree is known for resistance to decay and excellent longevity even in soil contact. It is a durable species used for fence post material. It is also known for the stilbene chemicals resveratrol and piceatannol. These chemicals exhibit biological activities, mainly antioxidant properties. Identifying the chemicals that contribute to the durability of black locust could lead to the development of relevant medicinal alternatives and other products. In this study, black locust wood and bark were extracted using acetone–water solutions. Using the extractions, we analyzed mixtures from the bark and wood. Two-dimensional gas chromatography-time of flight mass spectrometry (GC×GC-TOFMS) Pegasus BT 4D was used to characterize and identify the chemical composition of the extracts. The chemicals were categorized based on their functionality. The chemicals identified were separated into two categories, those shared by both wood and bark and those not shared. The extractions had some of the same chemicals as identified in the NIST and Wiley libraries. After the chemicals were categorized, we were able to determine which chemicals were specific to wood and bark.

Keywords: GC×GC, extractives, *Robinia pseudoacacia*, wood, bark

Contents

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Classification of Chemicals in Black Locust *Robinia pseudoacacia* Wood and Bark

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Introduction

Wood chemical research has been around since the early 1900s. Years ago, it was called "naval stores" research. Separation and isolation of complex mixtures were difficult to accomplish because of similarity in the chemical formula, functional groups, and volatility of the compounds. Naval stores research was important, but analytical techniques for resolving the complex mixture were limited. In the twenty-first century, instrumental analysis is more advanced and is well-equipped to handle the resolution needed to identify and characterize trace components. GC×GC is well-suited to investigate trace chemical composition in black locust. Sarikurkcu and others (2015) investigated antioxidant and enzyme inhibitory activity in various solvents. Sergent and others (2014) identified the stilbene chemicals resveratrol and piceatannol between leaves during different seasons. Kirker and others (2013) isolated and monitored fatty acids contribution to the overall wood durability of wood species. Moore and others (2015a, 2015b, 2017a, 2017b, 2017c) identified chemicals using gas chromatography-mass analyzer and found that the concentration plays a role in the number of chemicals identified from the chemical fractionations. In this study, GC×GC was used to compare chemicals identified in the wood and bark of black locust. This technique is important for identifying chemicals with low concentrations. Chemical mapping is facilitated by the easy visualization of 3D chromatographic results. The same solvent solutions were used for extracting wood and bark. GC×GC was used to identify chemical differences and similarities of bark and wood. The chemicals were categorized and classified on the chromatograms.

Experimental

Materials

Solvents were purchased from Thermo Fisher Scientific (Waltham, Massachusetts, USA). Tetracosane 99% was used for the calibration standard and was purchased from Sigma-Aldrich (St. Louis, Missouri, USA).

Sample Preparation

Black locust wood chips and bark were collected and Wiley-milled to 40 mesh. A soxhlet extraction with 9:1 acetone/water was run for 10 hours on the wood and bark samples separately. The solvent extracts were dried by a rotary evaporator and placed in the freezer.

Instrumentation Operations

GC×GC TOFMS was used to analyze the black locust extractives. Table 1 is the list of the oven and mass spectrometer parameters. The methodology was the same for each run. The injection temperation and transfer lines were the same for each run. The split ratio and modulation time changed according to concentration and chemicals. The splits were dependent on the concentration of the samples. The modulation time changed according to the chemicals retained on the second dimension column.

Table 1. Gas chromatography (GC) and two-dimensional gas chromatography-time of flight mass spectroscopy (GC×GC-TOFMS) (Pegasus BT 4D) instrument parameters

Gas chromatograph agilent 7890B with LECO dual stage quad jet modulator and LPAL 3 autosampler	
Injection	1 uL, split 20–150:1 at 250 °C
Carrier gas	He at 1.4 mL/min
Primary column	Rxi-5 ms, 30-m by 0.25-mm i.d. by 0.25-µm coating (Restek, Bellefonte, PA, USA)
Secondary column	Rxi-17SilMS, 0.60-m by 0.25-mm by 0.25-µm coating (Restek, Bellefonte, PA, USA)
Oven programming	Primary column starting temperature 50 °C, ramped 10 °C/min to 320 °C, held 5 min. Secondary column temperature parallel ramp offset by +5 °C (total run time 32 min).
Modulation	3–5 s with temperature maintained +15 °C relative to second oven.
Transfer line	280 °C
Mass spectrometer LECO Pegasus BT 4D	
Ion source temperature	250 °C
Mass range	50–650 m/z
Acquisition rate	150 spectra/s (2D)

Results and Discussion

A mixture of acetone and water (9:1) was used to remove extracts from the black locust wood (blw) and bark (blb). GC×GC-TOFMS was calibrated with known concentrations of tetracosane. Tetracosane was also tested as an unknown standard. Both extracts were injected in replicates. There were 1,343 and 1,868 chemicals from the acetone extracts of blw and blb, respectively. The NIST and Wiley libraries were used to identify these chemicals. Table 2 shows the number of chemicals detected at various similarity matches. There were 566 and 857 unknown chemicals detected for blw and blb extracts, respectively.

Because high concentration produced tailing, some chemicals were duplicated. All chemicals identified more than once were removed from the list of chemicals. Any chemical with isomers was not included in the final list. From the list of chemicals detected, 106 and 180 were left with greater than 800 similarity for blw and blb extracts, respectively. This is not shown in Table 2, but 83 chemicals were shared between extracts. They were not included in the total for each extract.

The list of chemicals in Tables 3 and 4 are from the bark and wood solvent extractions, respectively. There are no duplicates, and the chemicals are specific to their solvent extractions. The chemicals listed have spectral similarity value greater than 900. The percentage peak area was calculated using the sum of all chemicals with matches greater than 800 similarity. The blw extracts did not yield as many spectral matches with 900 similarity as did the blb extracts.

Chemicals in Table 3 with significant peak area were lactic acid 5.2% and dihydro-3-methylene-5-methyl- 2-furanone 3.67%. There were other significant peak area percentages not shown in the table because they did not have spectral matches greater than 900, including b-d-glucopyranose 3.80%; stigmast-4-en- 3-one 10.44%; 3-hydroxypropane-1,2-diyl diacetate 6.38%; and l-sorbose 8.46%. B-d-glucopyranose is a synthetic simple monosaccharide used as an energy source. L-sorbose is the sugar known as monosaccharide, which has the configuration of a natural sugar. Stigmast-4-en-3-one is a sito-sterol used to decrease cholesterol levels. Of the chemicals shown in Table 4, heptadecane had the most significant peak area at almost 5.60%. Chemicals that were under the 900 similarity were 2,4-dihydroxy-benzaldehyde 5.13%; 3-methoxymethylhept-1-yn-3-ol 4.26%; and 4-methoxy-2-pentanone 30.56%.

Because they are part of the same tree structure, bark and wood are expected to have similar chemicals. There were 83 shared chemicals in both extraction systems. Wood chemicals are known to migrate throughout the life of the tree. Tables 5 and 6 list chemicals from bark and wood, respectively, with spectral matches greater than 900 similarity. 2-propanone 5.67%; (pyro)-catechol 4.10%; glycerin 27.1%; mome inositol 21.6%; and 1,3- benzenediol 5.50% had the highest peak area among the 900 similarity matches. Benzene 10.57% also had a significant peak area even though it was below the 900 similarity criteria.

Table 2. Number of chemicals detected by two-dimensional gas chromatography-time of flight mass spectrometry (GC×GC-TOFMS) and identified by NIST (blw) and Wiley (blb) libraries

Extract	Total >700	Peaks unknown	Peaks known	>800 known	>800 known[a]
blw	1,343	566	777	455	106
blb	1,868	857	1,011	573	180

[a]Number of known chemicals after removing duplicates, shared, and isomers.

In Table 6, pyro-catechol 4.85% was significant. Other chemicals with percentages higher than pyro-catechol were 1,2,3 benzenetriol 5.30%; resorcinol 64.4%; and 4-(3-hydrooxyprop-1-en-1yl)-2-methoxyphenol 7.77%. These were not shown in Table 6 because they fell below the similarity cut off. Resorcinol is extremely concentrated and is part of the benzenediol group. Benzenediols are known for having antimicrobial activity. Resorcinol is used in antiseptic and disinfectant topical pharmaceutical products for treatment of skin disorders and infections such as acne, dermatitis, eczema, calluses, and warts.

When injected into the GC×GC TOFMS, complex mixtures display various classes of chemicals in different regions on the chromatogram. We call this molecular chemical mapping (MCM). This is similar to the periodic table. GC×GC chromatograms can be used to create a fingerprint for various complex mixtures. This is one of the best ways to visually and chemically identify unknown chemicals in complex mixtures.

Figures 1, 2, and 3 have similar profiles. Figures 1 and 2 are the first 20 min, and Figure 3 started at 11 min into the run of the extractives. Figure 3 is the rest of the full analysis of black locust wood and bark. The bark chemicals were more concentrated than the wood chemicals. Most of the chemicals observed in the bark were also observed in the wood but at lower concentrations. These chromatograms follow the same profile as the other complex extract mixtures. The labeling of the regions is similar. On the bottom right are long chain hydrocarbons eluting according to the volatility. On the top right are the sterol chemicals. And in the bottom middle are fatty acids. On the top in the middle between 1.8 and 2.8 seconds in the second dimension, there are several identified pyranose and sugar chemicals. The majority of the chemicals between second dimension 1.5 and 3 seconds are aromatic chemicals with alcohols, acids, and methoxy groups as the ring substituents.

Table 3. Two-dimensional gas chromatography-time of flight mass spectrometry (GC×GC TOFMS) list of compounds specifically identified in black locust bark acetone extracts >900 similarity

Number	Chemical identified[a]	Formula	Peak area (%)	Retention time (s)	Similarity
1	2-Pentanone, 4-hydroxy-4-methyl-	$C_6H_{12}O_2$	0.256	200 3.429	933
2	2,3-Dimethylfuran	C_6H_8O	0.178	204.998 3.221	927
3	2-Furanmethanol	$C_5H_6O_2$	0.318	214.993 1.064	932
4	2,5-Furandione	$C_4H_2O_3$	0.170	214.993 1.429	960
5	(E)-4-(1',1'-Bis(ethoxycarbonyl)ethyl)-4-	$C_{20}H_{34}O_5$	0.163	219.991 0.607	944
6	L-Lactic acid	$C_3H_6O_3$	5.151	219.991 0.955	942
7	2-Hydroxy-gamma-butyrolactone	$C_4H_6O_3$	0.121	319.943 1.811	914
8	4-Hydroxy-5,6-di-hydro-(2H)-pyran-2-one	$C_5H_6O_3$	0.019	324.941 1.522	948
9	Cyclohexene, 1-methyl-4-(1-methylethenyl)-,	$C_{10}H_{16}$	0.550	354.927 0.954	907
10	1,2-Propadiene-1,3-dione	C_3O_2	0.042	354.927 1.927	961
11	Benzene, methyl-	C_7H_8	0.073	379.915 1.048	912
12	2H-Pyran-2-one, tetrahydro-	$C_5H_8O_2$	0.048	379.915 1.908	904
13	Dihydro-3-methylene-5-methyl-2-furanone	$C_6H_8O_2$	3.669	394.908 1.798	920
14	1-(2-Furanyl)-2-hydroxyethanone	$C_6H_6O_3$	0.174	399.905 1.563	931
15	p-(1-Propenyl)-toluene	$C_{10}H_{12}$	0.314	409.901 1.143	923
16	1,2,3-Propanetriol, 1-acetate	$C_5H_{10}O_4$	0.365	409.901 1.512	930
17	Benzeneethanol	$C_8H_{10}O$	0.038	429.891 1.453	901
18	Benzoic acid	$C_7H_6O_2$	0.437	464.875 1.354	904
19	1-Propanone, 1-phenyl-	$C_9H_{10}O$	0.079	474.87 1.445	957
20	(S)-(+)-2',3'-Dideoxyribonolactone	$C_5H_8O_3$	0.134	489.863 2.152	947
21	5-(1-Hydroxyethyl)-2-oxolanone	$C_6H_{10}O_3$	0.213	499.858 1.971	911
22	(5R)-5-(Hydroxymethyl)-2-oxolanone	$C_5H_8O_3$	0.666	509.853 1.800	928
23	Erythritol	$C_4H_{10}O_4$	0.684	529.844 1.837	919
24	Benzeneacetic acid	$C_8H_8O_2$	0.491	539.839 1.446	936
25	(5E)-6,10-Dimethyl-5,9-undecadien-2-one	$C_{13}H_{22}O$	0.135	704.761 1.174	921
26	DL-Arabinitol	$C_5H_{12}O_5$	1.933	739.745 1.962	903
27	Vanillic acid	$C_8H_8O_4$	1.332	784.723 1.756	908
28	Pentadecanal-	$C_{15}H_{30}O$	0.047	889.674 1.067	905
29	1,2,3,4,5,6-Hexanehexol	$C_6H_{14}O_6$	1.141	939.65 2.198	932
30	(9E)-9-Hexadecen-1-ol	$C_{16}H_{32}O$	0.177	989.626 1.074	927
31	11-Hexadecen-1-ol, (Z)-	$C_{16}H_{32}O$	1.179	1,104.57 1.136	944
32	Octadecanal	$C_{18}H_{36}O$	0.165	1,239.51 1.117	908
33	Eicosanoic acid	$C_{20}H_{40}O_2$	0.342	1,254.5 1.167	908
34	Docosanal	$C_{22}H_{44}O$	0.256	1,284.49 1.152	917
35	2,6,10,14,18,22-Tetracosahexaene, 2,6,10,15,19,23-hexamethyl-	$C_{30}H_{50}$	1.148	1,464.4 1.381	935
36	7-exo-Acetylmethylbicyclo[4.1.0]hept-2-ene	$C_{11}H_{18}O$	0.044	1,479.39 4.356	928
37	7-Hydroxy-7-phenyl-3,9-diisopropyl-2,10-dioxadispiro[3.3.3.1]dodecan-1,11-dione	$C_{22}H_{28}O_5$	0.402	1,779.25 0.978	969

[a]Name from Wiley library.

Identifying regions for classes of chemicals (RCC) located on the GC×GC chromatograms is important when characterizing complex mixtures. GC×GC is being used to create a standard way to establish a fingerprint for complex mixtures. Because of the resolving power and the visual view of MCM on a GC×GC chromatogram, we can better identify chemicals in extracts. This development will change the way we investigate complex mixtures.

Table 4. Two-dimensional gas chromatography-time of flight mass spectroscopy (GC×GC TOFMS) list of compounds specifically identified in black locust wood acetone extracts >900 similarity

Number	Chemical identified[a]	Formula	Peak area (%)	Retention time (s)	Similarity
1	2-Furancarboxaldehyde	$C_5H_4O_2$	0.159	204.998 1.155	901
2	4-Hydroxy-4-methylpentane-2-one	$C_6H_{12}O_2$	0.905	209.995 0.987	938
3	2,4-Dihydroxy-2,5-dimethyl-3(2H)-furan-3-one	$C_6H_8O_4$	0.161	309.948 1.120	912
4	Nonanal	$C_9H_{18}O$	0.349	414.898 1.069	922
5	1,2,4,5-Tetroxane 3,3,6,6-tetramethyl-	$C_6H_{12}O_4$	0.441	479.868 0.549	939
6	1,3-Isobenzofurandione	$C_8H_4O_3$	0.039	604.808 1.922	910
7	Heptadecane	$C_{17}H_{36}$	5.594	644.789 0.864	910
8	Vanillin	$C_8H_8O_3$	0.673	669.778 1.809	928
9	Benzaldehyde 2,4-dihydroxy-	$C_7H_6O_3$	5.138	699.763 1.697	946
10	p-Hydroxyphenylacetone	$C_9H_{10}O_2$	0.179	699.763 1.829	902
11	6-Hydroxybenzofuran-3-one	$C_8H_6O_3$	0.804	824.704 2.033	907
12	(2R,3S,5α,22R,23R,24S)-25-Methoxy-6,6-(ethylenedioxy)-2,3;22,23-bis(isopropylidenedioxy)ergostane-22,23-diol	$C_{37}H_{62}O_7$	0.474	1,174.54 0.511	925
13	Benzene 1-[[(1-ethenyl-5-ethoxy-2-methoxy-4-pentynyl)oxy]methyl]-4-methoxy- [S-(R*,S*)]-	$C_{18}H_{24}O_4$	0.032	1,329.47 2.786	944
14	Squalene	$C_{30}H_{50}$	1.208	1,464.4 1.383	935
15	2-Ethyl-3-(n-butyl)penta-3,4-dienoic acid	$C_{11}H_{18}O_2$	0.318	1,524.37 4.564	900
16	2-(5-Hydroxy-1-benzofuran-3-yl) benzene-1,4-diol	$C_{14}H_{10}O_4$	0.019	1,554.36 3.882	901
17	Hentriacontane	$C_{31}H_{64}$	1.598	1,569.35 1.306	940
18	9,19-Cyclolanost-24-en-3-ol (3β)-	$C_{30}H_{50}O$	0.673	1,709.29 3.802	911
19	1H-Indene 3-(4-methoxyphenyl)-2-phenyl-	$C_{22}H_{18}O$	0.013	1,819.23 2.832	919

[a]Name from NIST library.

Table 5. Two-dimensional gas chromatography-time of flight mass spectrometry (GC×GC TOFMS) list of shared compounds identified in black locust bark acetone extracts >900 similarity

Number	Chemical identified[a]	Formula	Peak area (%)	Retention time (s)	Similarity
1	Butyrolactone	$C_4H_6O_2$	0.110	259.972 1.656	933
2	Benzene, (1-methylethyl)-	C_9H_{12}	0.233	264.969 0.979	904
3	2-Hydroxy-2-cyclopenten-1-one	$C_5H_6O_2$	0.156	264.969 1.313	902
4	1,3-Dioxolane-4-methanol, 2,2-dimethyl-	$C_6H_{12}O_3$	0.482	279.962 1.094	913
5	Glycerin	$C_3H_8O_3$	27.057	294.955 1.590	934
6	Phenol	C_6H_6O	0.441	309.948 1.234	931
7	Furan, 2-pentyl-	$C_9H_{14}O$	0.618	319.943 0.958	955
8	2-Propanone	C_3H_6O	5.674	324.941 0.569	960
9	Ethanol, 2-(2-ethoxyethoxy)-	$C_6H_{14}O_3$	0.193	329.938 1.204	924
10	p-Cymene	$C_{10}H_{14}$	0.577	349.929 1.033	934
11	Cyclohexene, 1-methyl-4-(1-methylethenyl)-, (S)-	$C_{10}H_{16}$	0.550	354.927 0.954	907
12	2(3H)-Furanone, dihydro-3-hydroxy-4,4-dimethyl-, (±)-	$C_6H_{10}O_3$	2.319	359.924 1.548	937
13	Phenol, 2-methoxy-	$C_7H_8O_2$	0.592	409.901 1.401	903
14	Undecane	$C_{11}H_{24}$	0.079	414.898 0.810	916
15	4H-Pyran-4-one, 3-hydroxy-2-methyl-	$C_6H_6O_3$	0.105	429.891 1.570	902
16	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	$C_6H_8O_4$	2.080	454.879 1.489	929
17	Butanedioic acid	$C_4H_6O_4$	0.202	459.877 1.322	929
18	Octanoic acid	$C_8H_{16}O_2$	0.033	469.872 1.056	927
19	(Pyro)-catechol	$C_6H_6O_2$	4.135	499.858 1.462	935
20	Nonanoic acid	$C_9H_{18}O_2$	0.027	549.834 1.071	913
21	1,3-Benzenediol	$C_6H_6O_2$	5.506	564.827 1.685	908
22	Phenol, 2,6-dimethoxy-	$C_8H_{10}O_3$	0.448	629.797 1.692	931
23	2-Propenal, 3-(2-furanyl)-	$C_7H_6O_2$	0.067	699.763 0.520	954
24	β-D-Glucopyranose, 1,6-anhydro-	$C_6H_{10}O_5$	2.199	724.752 2.200	932
25	Benzoic acid, 4-hydroxy-	$C_7H_6O_3$	0.177	739.745 1.731	901
26	Vanillic acid	$C_8H_8O_4$	1.332	784.723 1.756	908
27	Mome inositol	$C_7H_{14}O_6$	21.607	859.688 1.999	920
28	Myristic acid	$C_{14}H_{28}O_2$	0.036	914.662 1.091	924
29	Benzoic acid, 4-hydroxy-3,5-dimethoxy-	$C_9H_{10}O_5$	0.613	954.643 1.968	901
30	Hexadecanoic acid	$C_{16}H_{32}O_2$	1.488	1,039.6 1.108	923
31	Hexadecanal	$C_{16}H_{32}O$	0.120	1,334.46 1.175	919
32	1-Docosene	$C_{22}H_{44}$	1.047	1,409.43 1.239	935
33	1,15-Pentadecanediol	$C_{15}H_{32}O_2$	0.174	1,449.41 1.585	905
34	2,6,10,14,18,22-Tetracosahexaene, 2,6,10,15,19,23-hexamethyl-	$C_{30}H_{50}$	1.148	1,464.4 1.381	935
35	Octacosanol	$C_{28}H_{58}O$	0.777	1,494.39 1.370	938
36	Campesterol	$C_{28}H_{48}O$	0.421	1,634.32 2.750	915
37	Stigmasterol	$C_{29}H_{48}O$	0.719	1,649.31 2.854	929
38	β-Sitosterol	$C_{29}H_{50}O$	1.981	1,674.3 3.119	933
39	Lup-20(29)-en-3-one	$C_{30}H_{48}O$	1.855	1,704.29 4.136	930
40	Lupeol	$C_{30}H_{50}O$	0.511	1,714.28 4.226	943
41	Furan, 2,3-dihydro-	C_4H_6O	0.137	1,849.22 0.394	999

[a]Name from Wiley library.

Table 6. Two-dimensional gas chromatography-time of flight mass spectrometry (GC×GC TOFMS) list of shared compounds identified in black locust wood acetone extracts >900 similarity

Number	Chemical identified[a]	Formula	Peak area (%)	Retention time (s)	Similarity
1	2-Propanone	C_3H_6O	0.650	214.993 0.168	972
2	(Furan-2-yl)methanol	$C_5H_6O_2$	0.051	214.993 1.086	929
3	2-Propenal, 3-(2-furanyl)-	$C_7H_6O_2$	0.164	224.988 0.360	929
4	Butyrolactone	$C_4H_6O_2$	0.083	259.972 1.677	930
5	2(5H)-Furanone	$C_4H_4O_2$	0.041	259.972 1.720	946
6	Benzene, (1-methylethyl)-	C_9H_{12}	0.283	264.969 0.992	925
7	1,3-Dioxolane-4-methanol, 2,2-dimethyl	$C_6H_{12}O_3$	1.613	274.965 1.126	956
8	Hexanoic acid	$C_6H_{12}O_2$	0.132	299.953 0.989	935
9	2,4-Dihydroxy-2,5-dimethyl-3(2H)-furan-3-one	$C_6H_8O_4$	0.161	309.948 1.120	912
10	Phenol	C_6H_6O	0.889	309.948 1.238	937
11	Furan, 2-pentyl-	$C_9H_{14}O$	0.498	319.943 0.964	961
12	Ethanol, 2-(2-ethoxyethoxy)-	$C_6H_{14}O_3$	0.253	329.938 1.212	950
13	3-Hydroxy-4,4-dimethyldihydro-2(3H)-furanone	$C_6H_{10}O_3$	1.329	359.924 1.552	947
14	Phenol, 2-methoxy-	$C_7H_8O_2$	1.160	409.901 1.406	919
15	Undecane	$C_{11}H_{24}$	0.060	414.898 0.814	951
16	4H-Pyran-4-one, 3-hydroxy-2-methyl-	$C_6H_6O_3$	0.128	429.891 1.576	929
17	2,3-Dihydro-3,5-dihydroxy-6-methyl-4H-pyran-4-one	$C_6H_8O_4$	0.307	454.879 1.489	908
18	(Pyro)-catechol	$C_6H_6O_2$	4.852	499.858 1.462	911
19	Nonanoic acid	$C_9H_{18}O_2$	0.028	549.834 1.071	920
20	Phenol, 2,6-dimethoxy-	$C_8H_{10}O_3$	0.778	629.797 1.692	928
21	1,4-Benzenediol, 2-methoxy-	$C_7H_8O_3$	0.083	664.78 1.790	918
22	Vanillin	$C_8H_8O_3$	0.673	669.778 1.809	928
23	β-D-Glucopyranose, 1,6-anhydro-	$C_6H_{10}O_5$	2.348	724.752 2.195	929
24	Mome inositol	$C_7H_{14}O_6$	2.376	844.695 2.052	921
25	Myristic acid	$C_{14}H_{28}O_2$	0.052	914.662 1.090	919
26	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	0.491	1,039.6 1.100	933
27	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	0.359	1,134.56 1.235	936
28	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-	$C_{18}H_{30}O_2$	0.234	1,139.56 1.284	932
29	1-Docosene	$C_{22}H_{44}$	0.109	1,314.47 1.168	948
30	Hexadecanal	$C_{16}H_{32}O$	0.037	1,334.46 1.178	906
31	Squalene	$C_{30}H_{50}$	1.208	1,464.4 1.383	935
32	Heptacosane	$C_{27}H_{56}$	0.141	1,489.39 1.156	930
33	Campesterol	$C_{28}H_{48}O$	0.470	1,634.32 2.750	909
34	Stigmasterol	$C_{29}H_{48}O$	0.829	1,649.31 2.855	928
35	β-Sitosterol	$C_{29}H_{50}O$	0.975	1,674.3 3.122	928
36	Lup-20(29)-en-3-one	$C_{30}H_{48}O$	0.063	1,704.29 4.128	905
37	Octacosanol	$C_{28}H_{58}O$	0.084	1,734.27 2.771	917

[a]Name from NIST library.

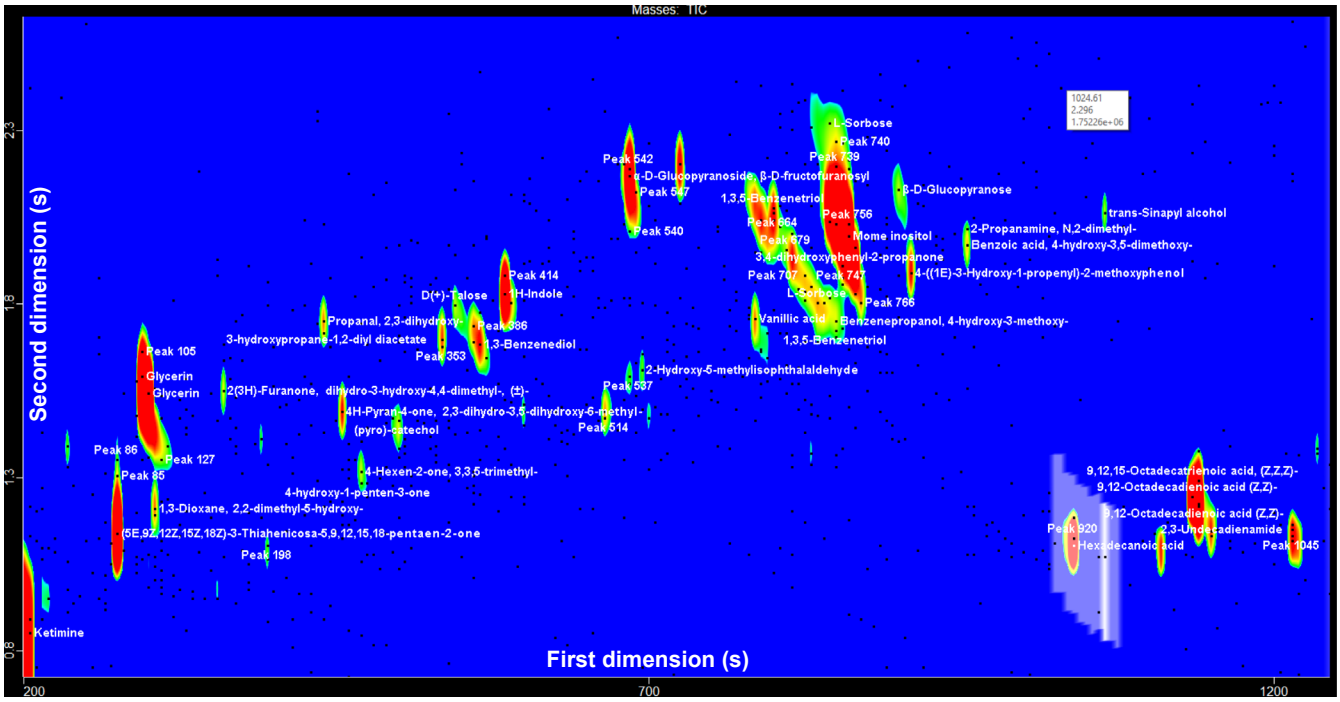

Figure 1. Two-dimensional gas chromatography (GC×GC) chromatogram contour plot of black locust bark acetone extracts.

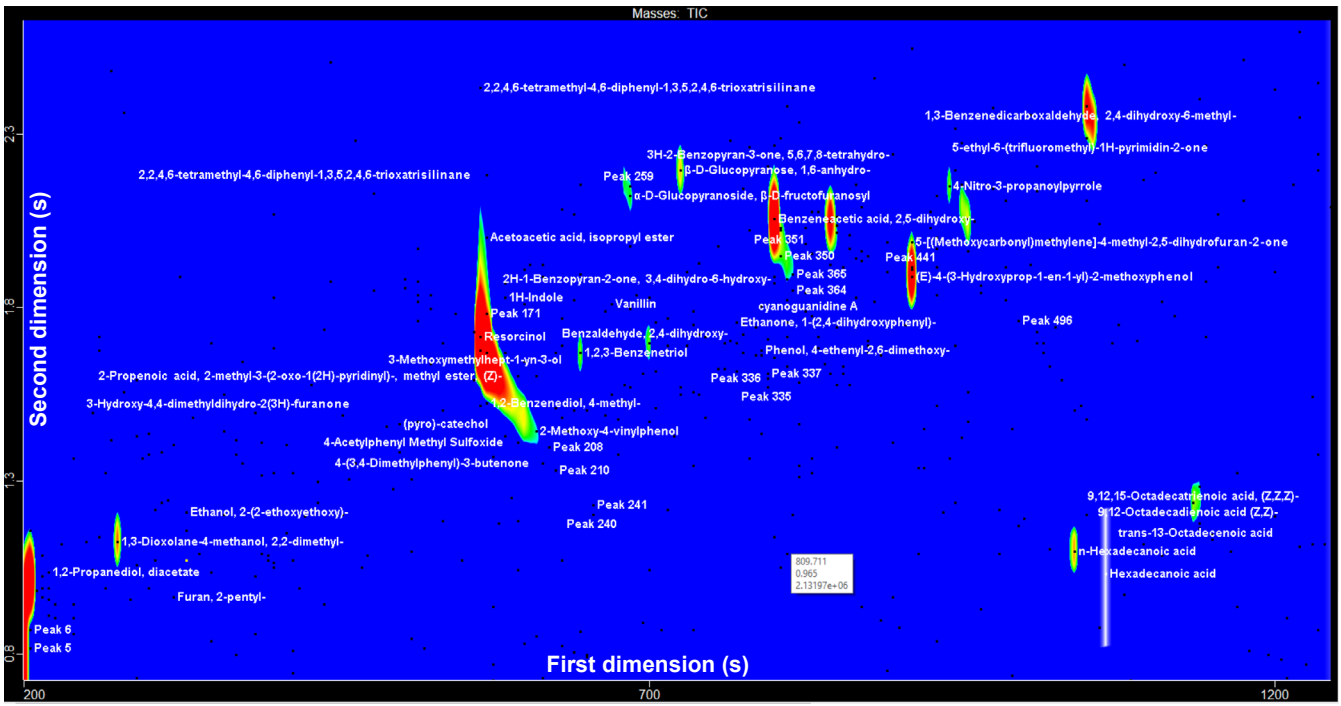

Figure 2. Two-dimensional gas chromatography (GC×GC) chromatogram contour plot for black locust wood acetone extracts.

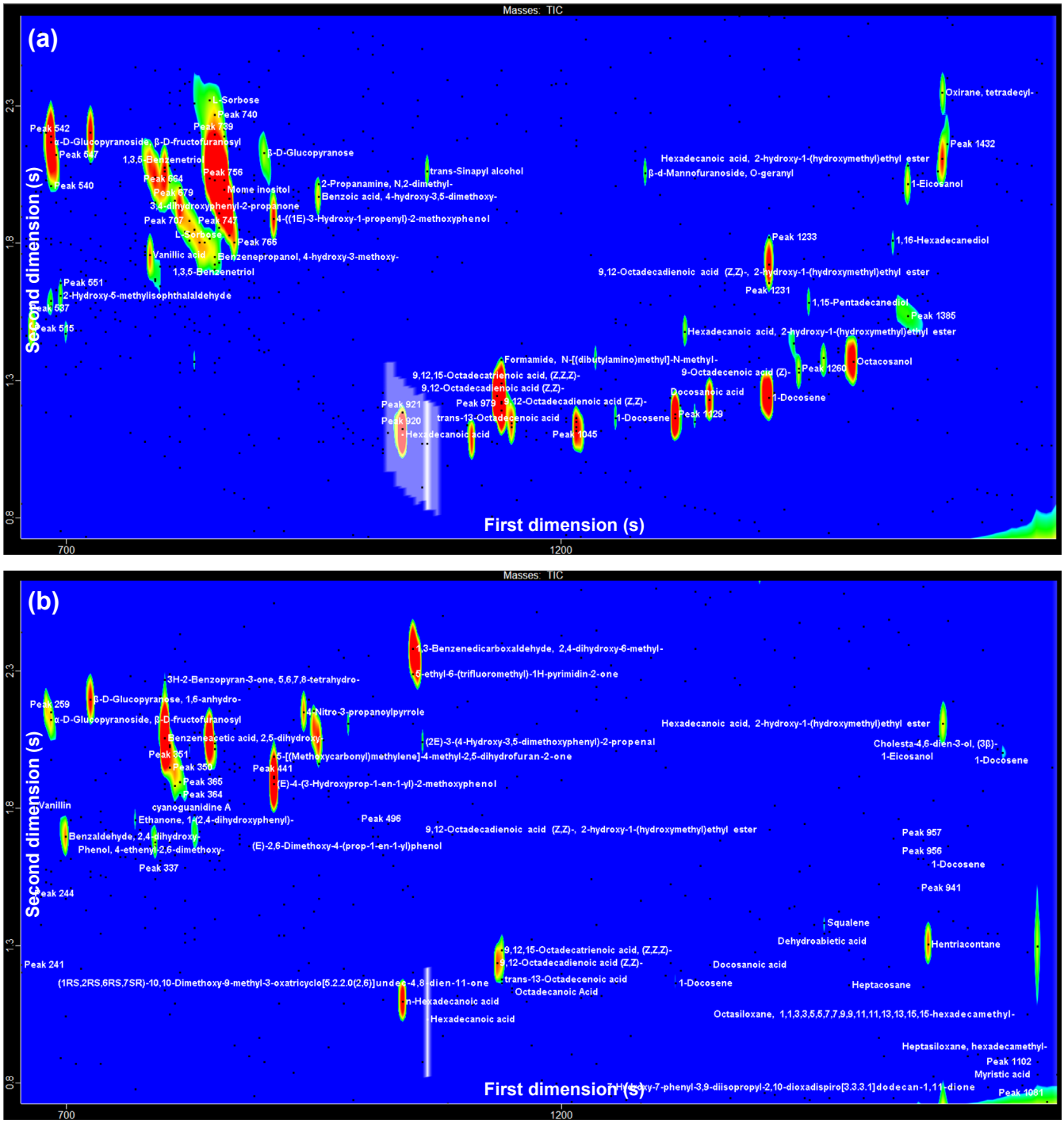

Figure 3. Two-dimensional gas chromatography (GC×GC) chromatogram contour plots for black locust (a) bark and (b) wood acetone extracts starting at 11 min into the run of the extractives.

Conclusions

Black locust is known for being a durable wood species. Identifying the chemicals that contribute to the durability can lead to other relevant research on medicinal alternatives. GC×GC technology opens the peak capacity where trace components are observed and can be detected in a complex mixture. One of the best techniques for characterizing black locust extracts is GC×GC TOFMS. There were 1,343 and 1,868 chemicals detected in blw and blb extracts, respectively. NIST and Wiley libraries were used to identify the chemicals based on the spectral matches. There were unknown chemicals detected in blw (566) and blb (857) extracts. Benzenediols were highly concentrated in the bark and wood. This chemical is one of several reasons black locust has high decay resistance when placed in soils. Various classes of chemicals were identified and labeled on the chromatogram. The molecular mapping on GC×GC will assist with labeling and classifying the unknown chemicals detected in the bark and wood extracts for future work. This will lead to developing or isolating chemicals with alternative medicinal applications.

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