

Supplementary Information for

Circulating heparin oligosaccharides rapidly target the hippocampus in sepsis potentially impacting cognitive functions

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Exact mass: 2652.8997

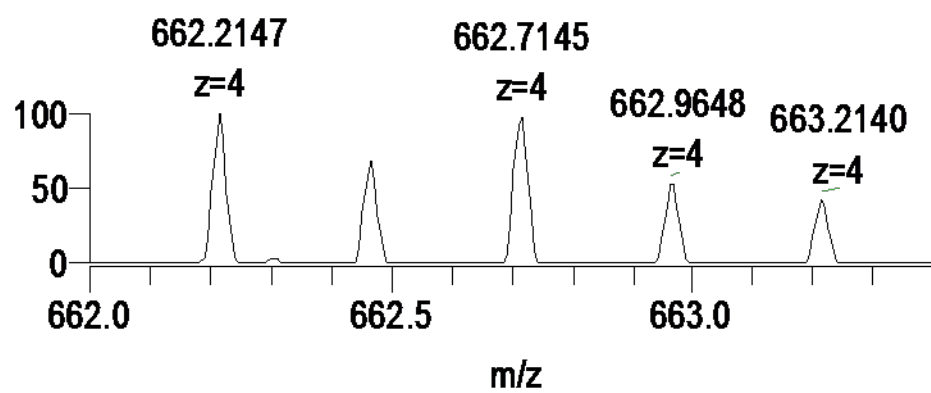


Fig. S1. HRMS (negative model) of compound 11.

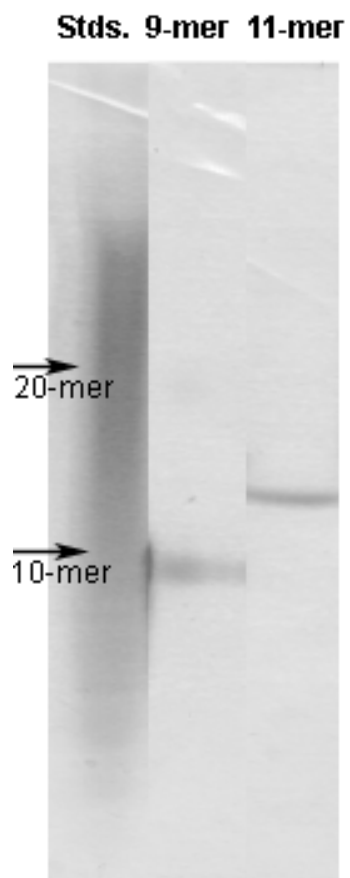


Fig. S2. PAGE analysis of compound **11** (9-mer) and **9** (11-mer). Heparin oligosaccharide standards prepared from partial heparin lyase 1 depolymerization of bovine lung heparin are shown on left by counting bands from disaccharide up five bands to 10-mer and 10 bands to 20-mer molecular size can be determined.

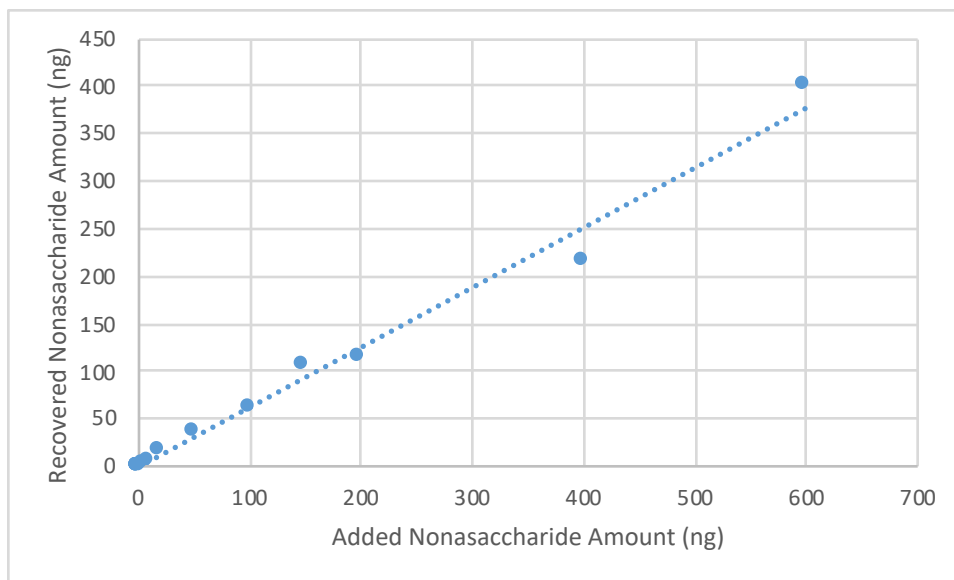
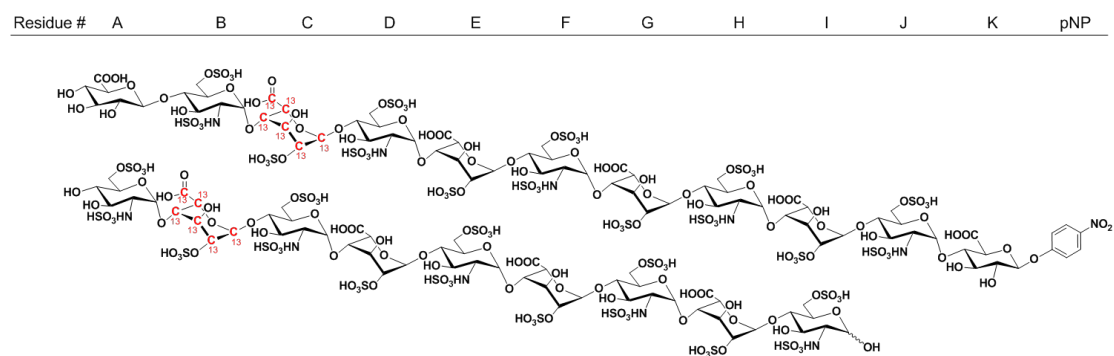


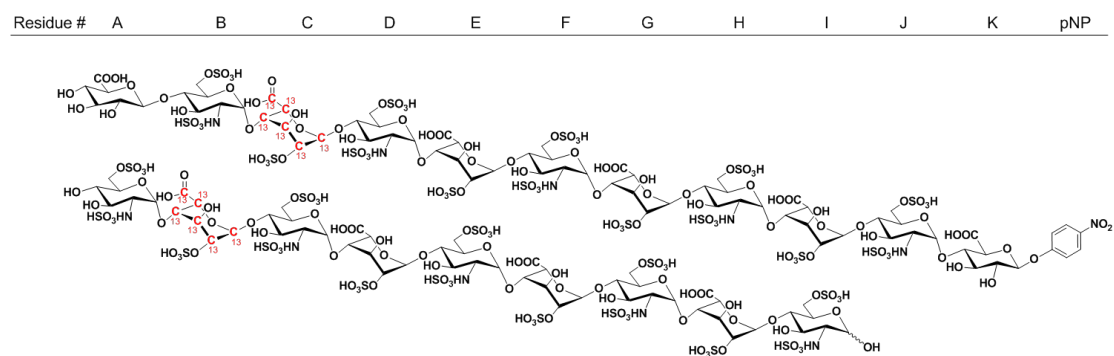
Fig. S3. Nonsaccharide recovery was linear over the range of 0.01-600 ng in 5 μ L of plasma sample. ($y = 0.6302x + 0.0152$, $R^2 = 0.9877$)

Table S1. ^1H NMR chemical shift assignments (in ppm) of compound 9



	1	2	3	4	5	6a	6b
A	4.53	3.28	3.47	3.41	3.69	—	—
B	5.34	3.20	3.59	3.68	3.97	4.13	4.27
C	5.15	4.25	4.12	4.02	4.74	—	—
D	5.34	3.20	3.59	3.68	3.97	4.13	4.27
E	5.15	4.25	4.12	4.02	4.74	—	—
F	5.34	3.20	3.59	3.68	3.97	4.13	4.27
G	5.15	4.25	4.12	4.02	4.74	—	—
H	5.34	3.20	3.59	3.68	3.97	4.13	4.27
I	5.15	4.25	4.12	4.02	4.74	—	—
J	5.53	3.23	3.60	3.75	3.89	4.15	4.33
K	5.22	3.65	3.89	3.78	3.92	—	—

Table S2. ^1H NMR chemical shift assignments (in ppm) of compound 11



	1	2	3	4	5	6a	6b
A	5.33	3.20	3.58	3.50	3.90	4.19	4.33
B	5.16	4.26	4.15	4.04	4.83	—	—
C	5.33	3.20	3.58	3.70	3.90	4.19	4.33
D	5.16	4.26	4.15	4.04	4.83	—	—
E	5.33	3.20	3.58	3.70	3.90	4.19	4.33
F	5.16	4.26	4.15	4.04	4.83	—	—
G	5.33	3.20	3.58	3.70	3.90	4.19	4.33
H	5.16	4.26	4.15	4.04	4.83	—	—
I	5.33	3.20	3.58	3.70	3.90	4.19	4.33

Table S3. Nonasaccharide recovery percentage in plasma.

Nonasaccharide amount added to plasma (ng)	Plasma (μ L)	Nonasaccharide recovered Amount (ng)	Recovery Percentage
0.01	5	0.0052	52%
0.05	5	0.0435	87%
0.1	5	0.071	71%
0.5	5	0.395	79%
1	5	0.74	74%
2	5	1.28	64%
5	5	3.85	77%
10	5	6.9	69%
20	5	16.6	83%
50	5	36.5	73%
100	5	63	63%
150	5	106.5	71%
200	5	116	58%
400	5	216	54%
600	5	402	67%

Table S4. Weight and volume information for sampled organs. Range, mean, and standard deviation for the defatted-dry weight (milligrams) of brain, muscle, liver, spleen, fat, heart, kidney, lung and hippocampus. Range, mean, and standard deviation for the volumes of collected plasma and urine.

Organ	Brain	Muscle	Liver	Spleen	Fat	Heart	Kidney	Lung	Hippocampus	Plasma (µL)	Urine (µL)
Range (mg)	21.9-45.5	42.8-109.6	23.7-241.9	23.1-123.7	9.4-55.1	33.1-86.6	36.3-86.3	13.1-25.8	1.7-5.5	20-340	40-200
Mean (mg)	35.0	60.8	113.5	36.2	17.4	43.6	50.1	18.0	2.8	151.7	156.2
Std. Dev.	8.5	14.7	40.7	21.0	9.0	12.0	12.2	3.2	0.9	59.1	88.5

Table S5. The average and standard deviation for the amounts of nonasaccharide quantified in organs (ng/mg) and biological fluids (ng/mL). The LOQ for the LC-MS method is 70 pg, and LOD is 20 pg. If between LOD and LOQ but shows both ¹³C and ¹²C peak with identical retention time then presence is recorded. If between LOD and LOQ and both ¹³C and ¹²C peaks are not detected then no presence is recorded.

Organ	Brain	Muscle	Liver	Spleen	Fat	Heart	Kidney	Lung	Hippocampus	Urine (ng/mL)	Plasma (ng/mL)
Control Mean (ng/mg)	0	0	0	0	0	0	0	0	0	0	0
Sham 30 min mean (ng/mg)	0	0	0	0.001	0	0	0	0	0	12791	1100
Sham 30 min standard deviation	0	0	0	0.001	0	0	0	0	0	722	135
Sham 120 min mean (ng/mg)	0	0	0	0.005	0.003	0.003	0.002	0	0	9438	529
Sham 120 min standard deviation	0	0	0	0.005	0.005	0.005	0.003	0	0	6749	58
Sham 240 min mean (ng/mg)	0	0	0.002	0.001	0.005	0.005	0.003	0	0	8813	0
Sham 240 min standard deviation	0	0	0.003	0.002	0.005	0.001	0.003	0	0	3212	0
CLP 30 min mean (ng/mg)	0	0	0	0.002	0	0	0	0	0.285	5739	14450
CLP 30 min standard deviation	0	0	0	0.005	0	0.001	0.001	0	0.045	0	2030
CLP 120 min mean (ng/mg)	0.002	0	0	0.006	0	0	0	0.005	0.319	7723	0
CLP 120 min standard deviation	0.002	0	0	0.008	0	0	0	0.006	0.088	4629	0
CLP 240 min mean (ng/mg)	0	0	0.003	0.004	0.009	0	0	0	0.361	795	0
CLP 240 min standard deviation	0	0	0.00296	0.004	0.008	0	0	0	0.040	924	0