A Sucrose-derived Scaffold for Multimerization of Bioactive Peptides

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SUPPLEMENTARY DATA

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Figure S1. MALDI-TOF of **13**.

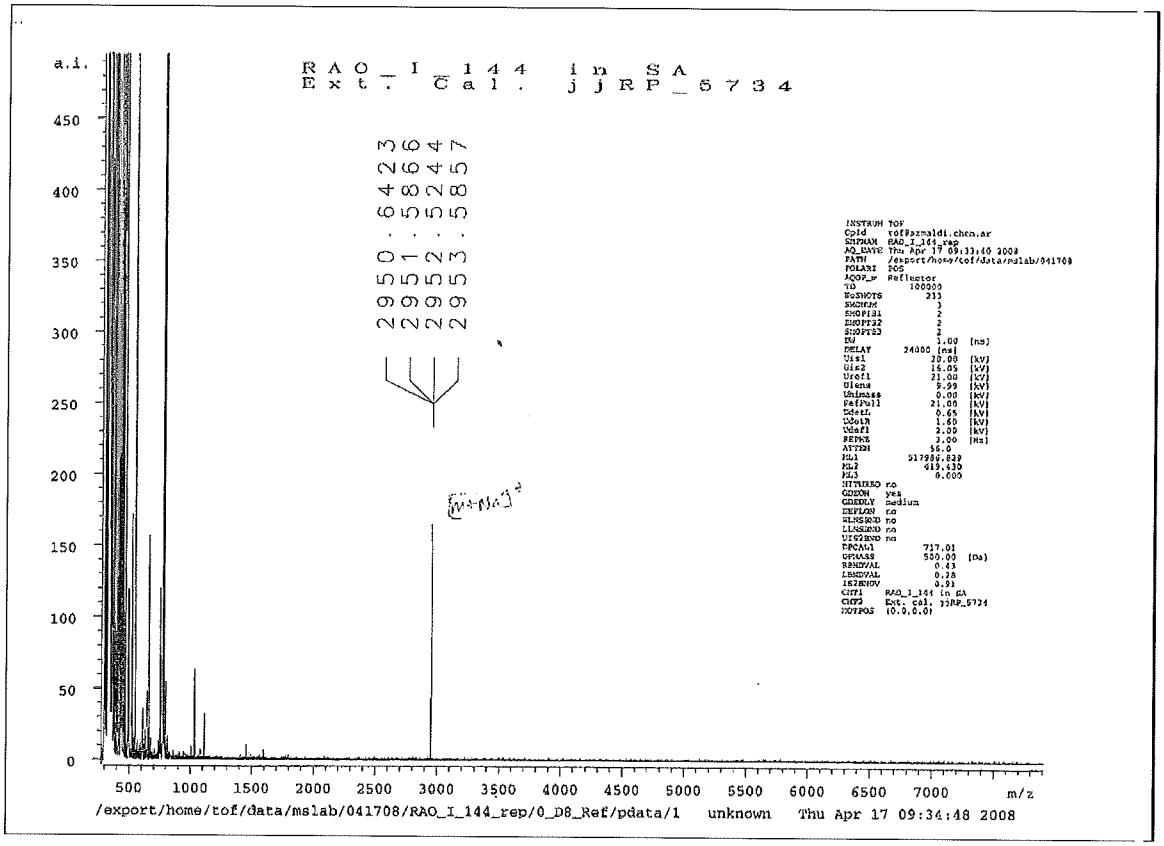
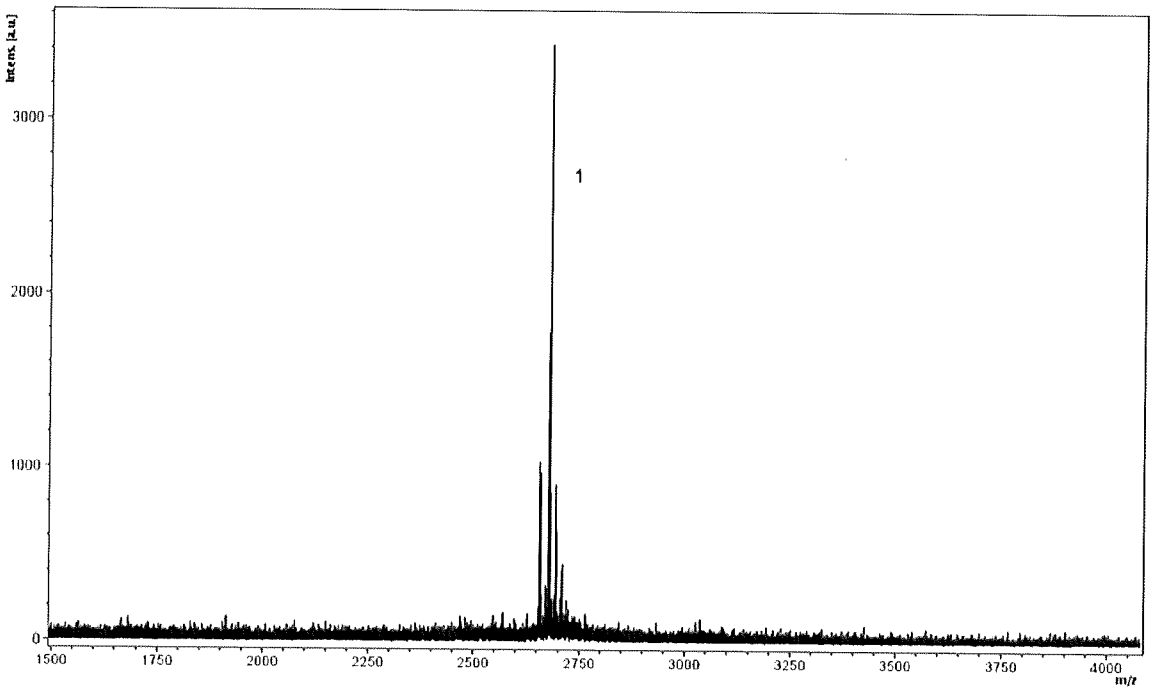


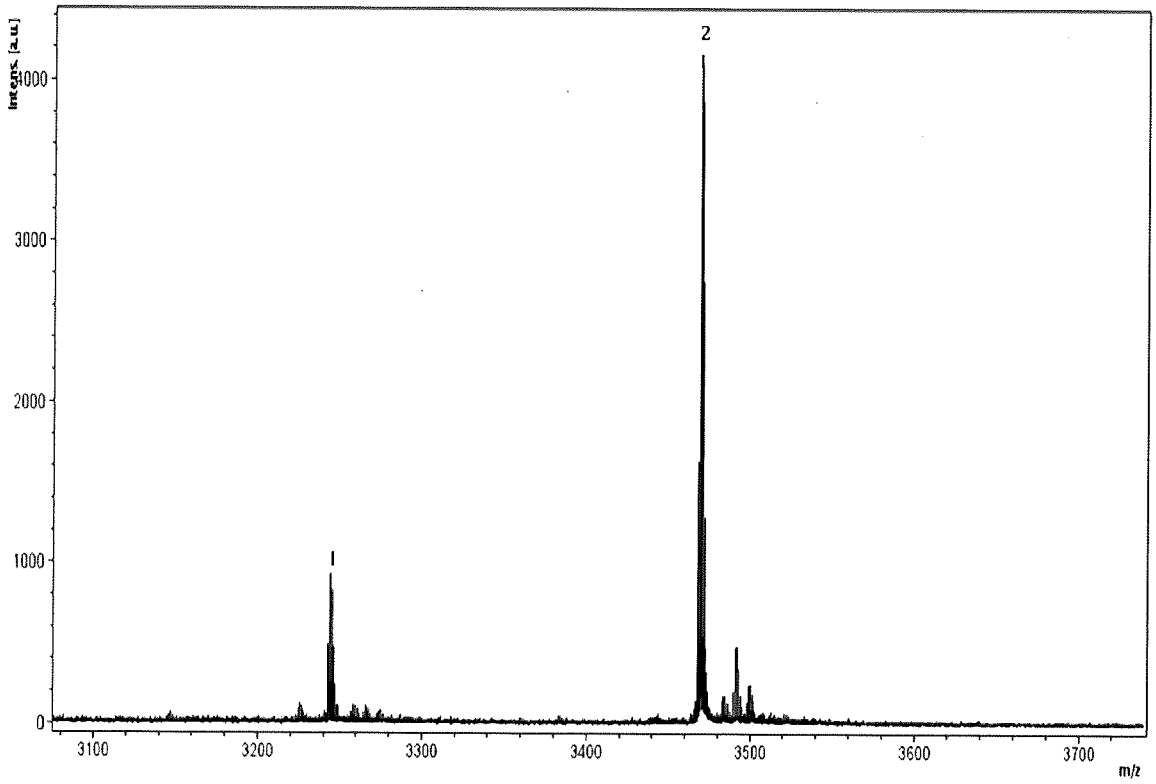
Figure S2. MALDI-TOF of **14a**.



Peak number	Number of side chains	Number of triazole rings	Number of MSH4 ligands	Number of serinamide residues*
1	8	8	1	0

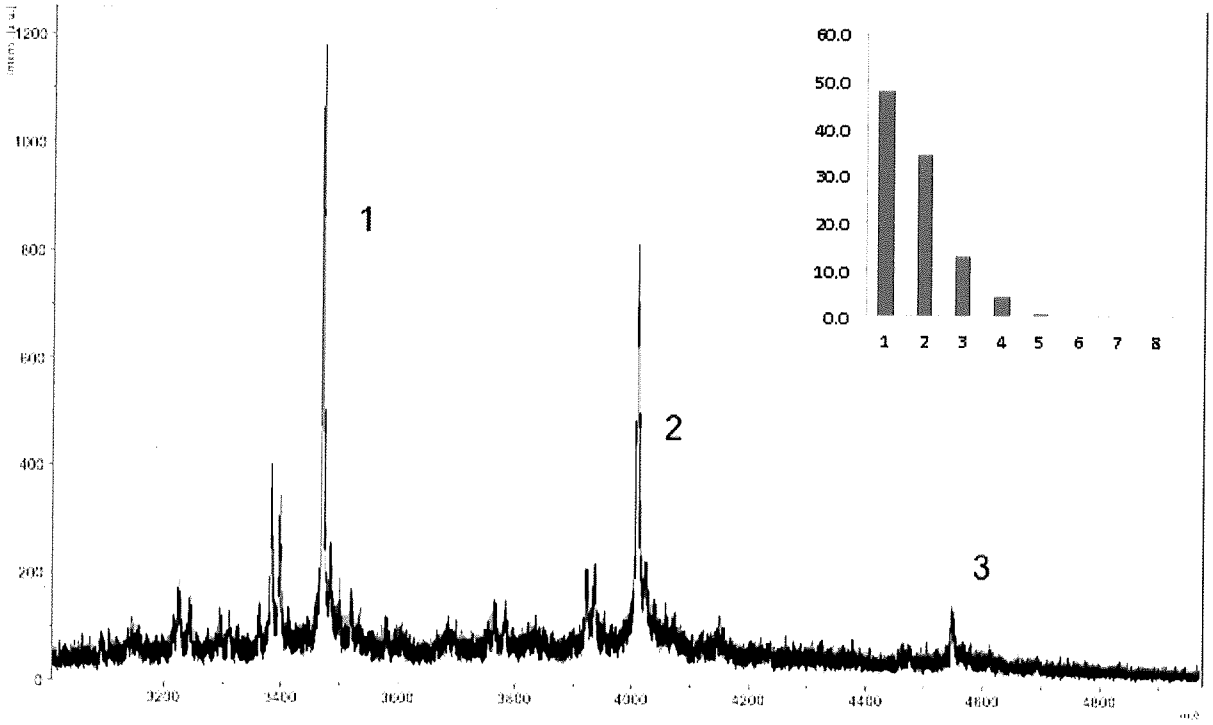
*Alkynes capped with 1-azidohexane.

Figure S3. MALDI-TOF of **14b**.



Peak number	Number of side chains	Number of triazole rings	Number of MSH4 ligands	Number of serinamide residues
1	8	7	1	6
2	8	8	1	7

Figure S4. MALDI-TOF of **14c**.



Peak number	Number of side chains	Number of triazole rings	Number of MSH4 ligands	Number of serinamide residues
1	8	8	1	7
2	8	8	2	6
3	8	8	3	5

INSERT: The statistical product distribution assuming attachment of 1.3 ligands per scaffold and equal reactivity of all alkynes throughout the reaction.

14c [2 MSH4 + 6 Serinamide]

Ligand concentration and distribution were calculated based on the theoretical molecular weight, UV absorbance, and areas under the mass spectrometric peaks.

Total MSH4 concentration by UV = 9.2 mM

Total MSH4 concentration by the theoretical molecular weight = 13.8 mM

Calculated number of MSH4 per scaffold by UV = 2 × (9.2/13.8) = 1.3

Calculated by areas under the mass spectrometric peaks:

Weighted average molecular weight Mw = 3772 (determined by area under the relevant peaks, see below)

Calculated molecular weight M = 4005 (based on 2 MSH4 residues per scaffold)

MSH4 molecular weight = 782

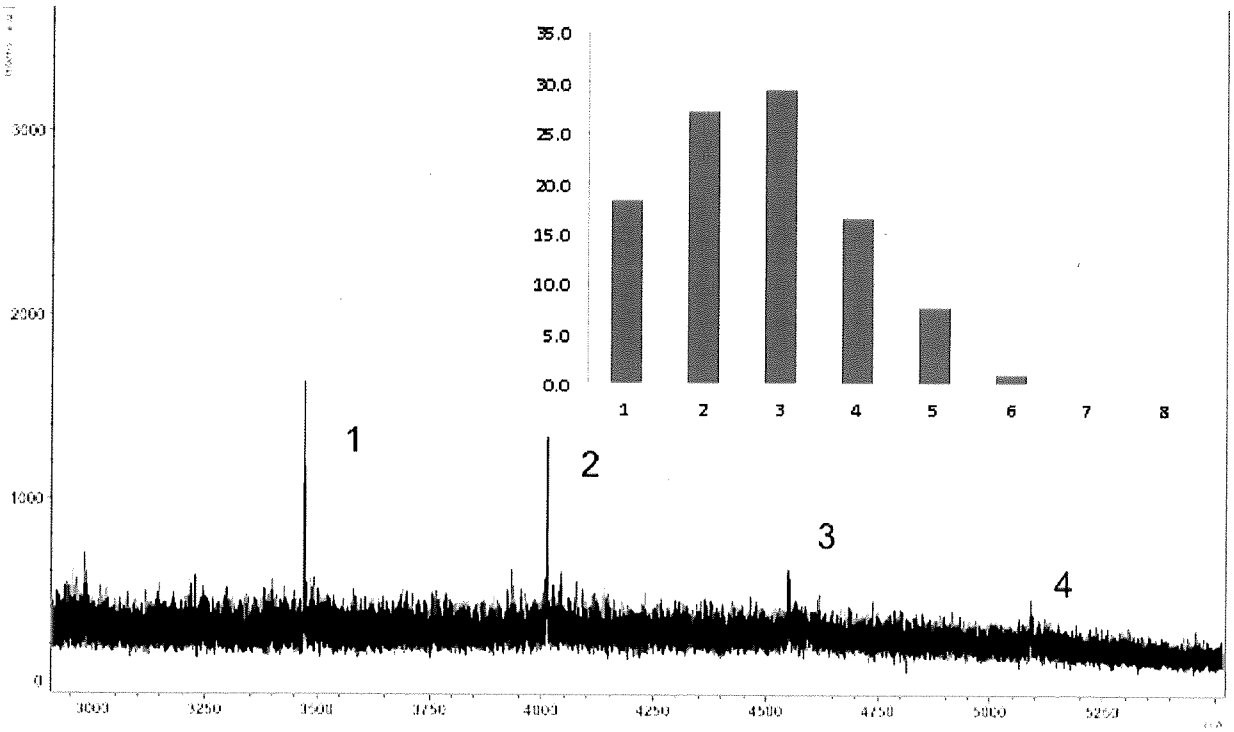
Total MSH4 weight in the scaffold = 2 × 782 –(4005-3772) = 1331 which is equal to 1.7 MSH4 per scaffold.

Peak #	M+S*	Mi (Mass)	Ni (Area)	MiNi	Mi^2Ni
1	1+7	3469	6178	21431482	74345811058
2	2+6	4009	4690	18802210	75378059890
3	3+5	4548	579	2633292	11976212016
			11447	42866984	1.617E+11

Mn =	$\sum_i MiNi/\sum_i Ni$	3745
Mw=	$\sum iMi^2Ni/\sum iMiNi$	3772
polydispersity	Mw/Mn	1.01

*M= MSH4, S = Serinamide

Figure S5. MALDI-TOF of **14d**.



Peak number	Number of side chains	Number of triazole rings	Number of MSH4 ligands	Number of serinamide residues
1	8	8	1	7
2	8	8	2	6
3	8	8	3	5
4	8	8	4	4

INSERT: The statistical product distribution assuming attachment of 2.6 ligands per scaffold and equal reactivity of all alkynes throughout the reaction.

14d [3 MSH4 + 5 Serinamide]

Ligand concentration and distribution were calculated based on the theoretical molecular weight, UV absorbance, and areas under the mass spectrometric peaks.

Total MSH4 concentration by UV = 7.7 mM

Total MSH4 concentration by the theoretical molecular weight = 8.9 mM

Calculated number of MSH4 per scaffold by UV = 3 × (7.7/8.9) = 2.6

Calculated by areas under the mass spectrometric peaks:

Weighted average molecular weight Mw = 3947 (determined by area under the relevant peaks, see below)

Calculated molecular weight M = 4545 (based on 3 MSH4 residues per scaffold)

MSH4 molecular weight = 782

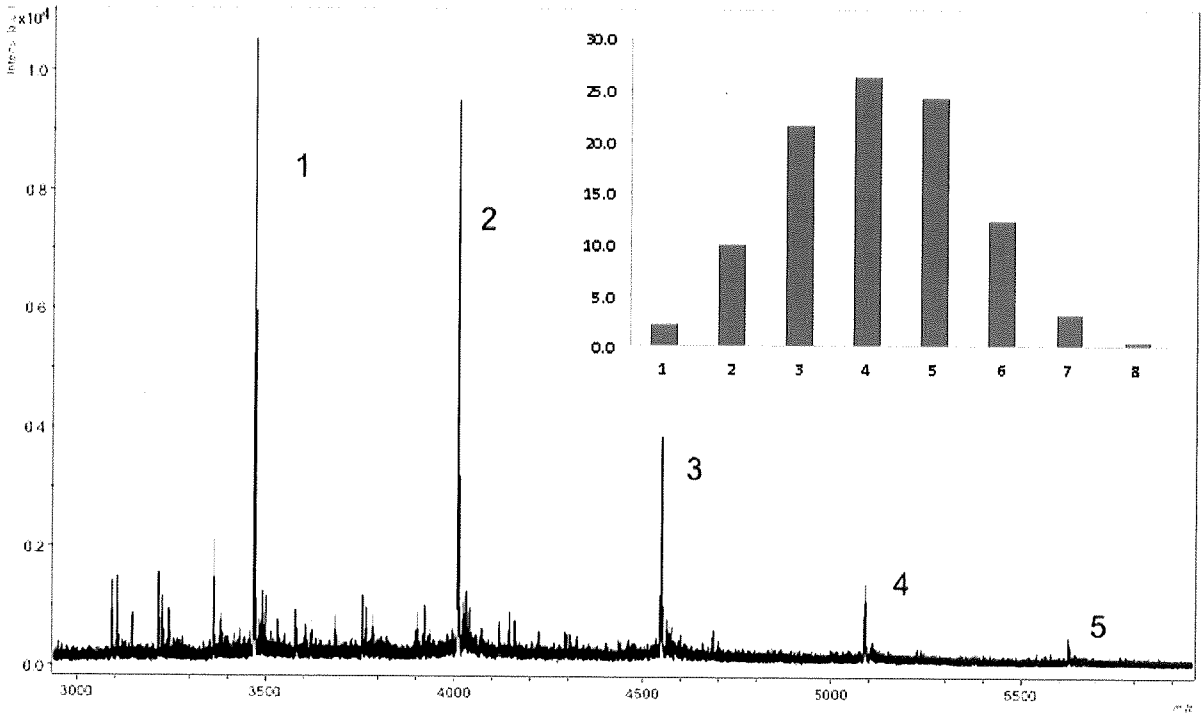
Total MSH4 weight in the scaffold = 3 × 782 –(4545-3947) = 1748 which is equal to 2.2 MSH4 per scaffold.

Peak #	M+S*	Mi (Mass)	Ni (Area)	MiNi	Mi^2Ni
1	1+7	3469	13655	47369195	1.64324E+11
2	2+6	4010	11192	44879920	1.79968E+11
3	3+5	4549	4556	20725244	94279134956
4	4+4	5087	1267	6445229	32786879923
			30670	119419588	4.71358E+11

Mn =	$\sum_i MiNi/\sum_i Ni$	3894
Mw=	$\sum iMi^2Ni/\sum iMiNi$	3947
polydispersity	Mw/Mn	1.01

*M= MSH4, S = Serinamide

Figure S6. MALDI-TOF of **14e**.



Peak number	Number of side chains	Number of triazole rings	Number of MSH4 ligands	Number of serinamide residues
1	8	8	1	7
2	8	8	2	6
3	8	8	3	5
4	8	8	4	4
5	8	8	5	3

INSERT: The statistical product distribution assuming attachment of 4.1 ligands per scaffold and equal reactivity of all alkynes throughout the reaction.

14e [5 MSH4 + 3 Serinamide]

Ligand concentration and distribution were calculated based on the theoretical molecular weight, UV absorbance, and areas under the mass spectrometric peaks.

Total MSH4 concentration by UV = 14.5 mM

Total MSH4 concentration by the theoretical molecular weight = 17.3 mM

Calculated number of MSH4 per scaffold by UV = 5 × (14.5/17.3) = 4.1

Calculated by areas under the mass spectrometric peaks:

Weighted average molecular weight Mw = 4043 (determined by area under the relevant peaks, see below)

Calculated molecular weight M = 5622 (based on 5 MSH4 residues per scaffold)

MSH4 molecular weight = 782

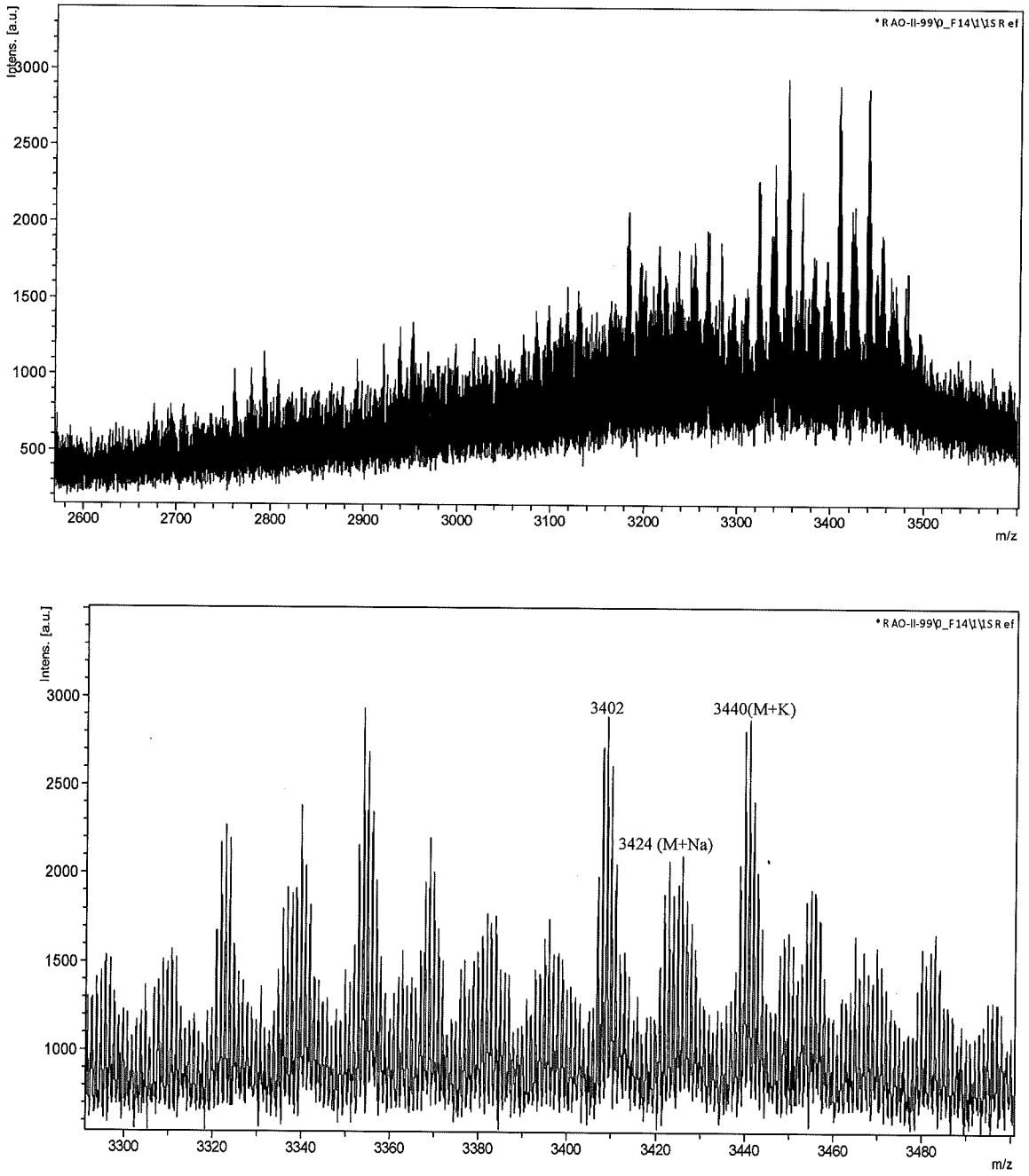
Total MSH4 weight in the scaffold = 5 × 782 –(5622-4043) = 2331 which is equal to 3.0 MSH4 per scaffold.

Peak #	M+S*	Mi (Mass)	Ni (Area)	MiNi	Mi^2Ni
1	1+7	3468	16244	56334192	1.95367E+11
2	2+6	4007	18993	76104951	3.04953E+11
3	3+5	4547	8374	38076578	1.73134E+11
4	4+4	5087	2140	10886180	55377997660
5	5+3	5628	521	2932188	16502354064
			46272	184334089	7.45334E+11

Mn =	$\sum_i MiNi/\sum_i Ni$	3984
Mw=	$\sum iMi^2Ni/\sum iMiNi$	4043
polydispersity	Mw/Mn	1.01

*M= MSH4, S = Serinamide

Figure S7. MALDI-TOF of **15a**.



Peak mass $[M+H]^+$	Number of side chains	Number of triazole rings	Number of CCK4 ligands	Number of serinamide residues
3402	8	8	1	7

15a [1 CCK4 + 7 serinamides]
Ligand/multimer concentration was calculated based on the UV absorbance.
Multimer concentration = CCK4 concentration = 2.1 mM by UV

15b [2 CCK4 + 6 serinamides]
Ligand concentration and distribution were calculated based on the theoretical molecular weight, UV absorbance.
Total CCK4 concentration by UV = 5.0 mM
Total CCK4 concentration by the theoretical molecular weight = 7.8 mM
Calculated number of CCK4 ligands per scaffold = 2 X (5.0/7.8) = 1.3

15c [3 CCK4 + 5 serinamides]
Ligand concentration and distribution were calculated based on the theoretical molecular weight, UV absorbance.
Total CCK4 concentration by UV = 5.5 mM
Total CCK4 concentration by the theoretical molecular weight = 7.6 mM
Calculated number of CCK 4 ligands per scaffold = 3 X (5.5/7.6) = 2.2

15d [5 CCK4 + 3 serinamides]
Ligand concentration and distribution were calculated based on the theoretical molecular weight, UV absorbance.
Total CCK4 concentration by UV = 5.1 mM
Total CCK4 concentration by the theoretical molecular weight = 7.1 mM
Calculated number of CCK 4 ligands per scaffold = 5 X (5.1/7.1) = 3.6

Figure S8. ^{1}H-NMR of Compound **9**.

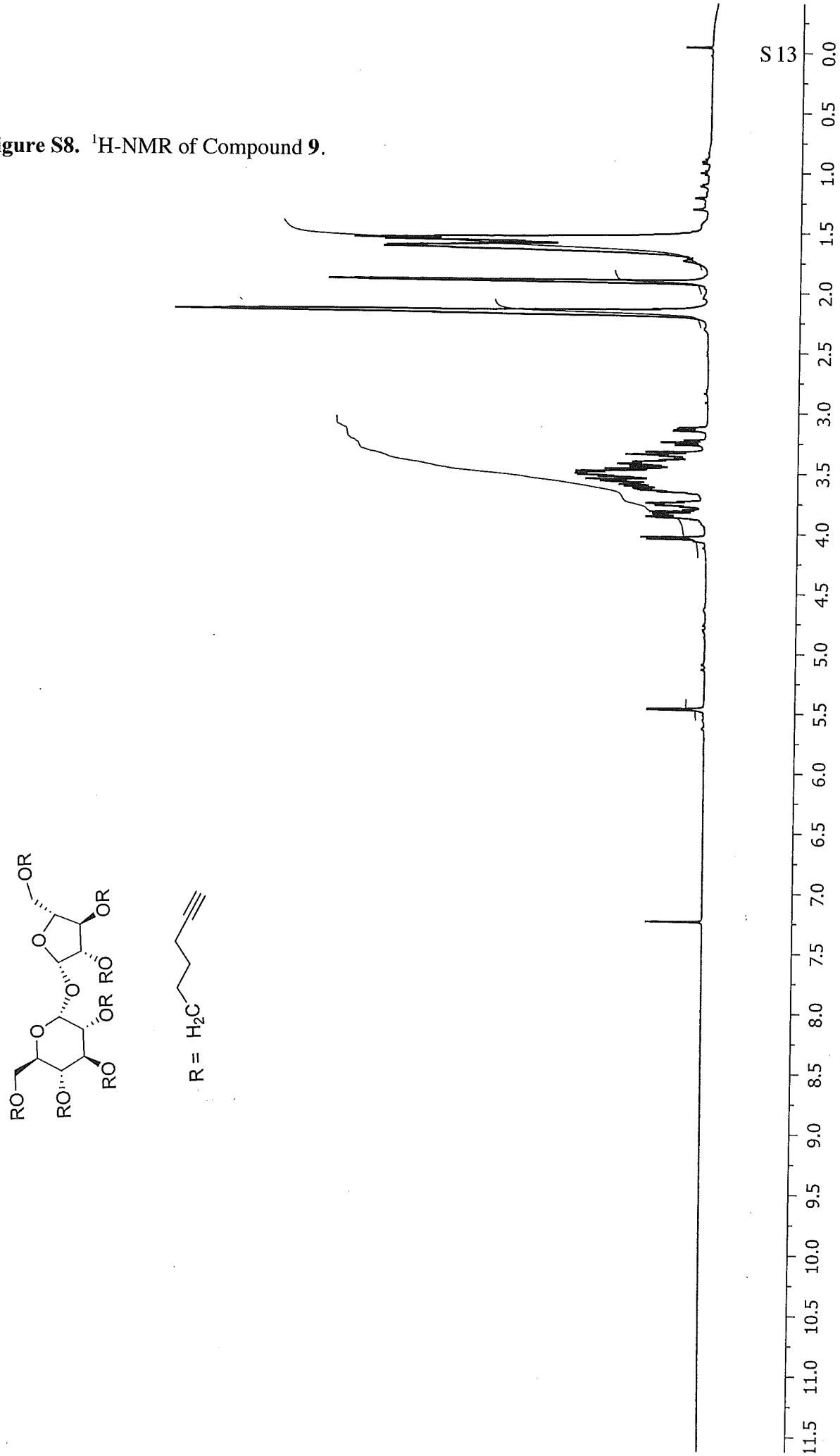


Figure S9. ^{13}C-NMR of Compound **9**.

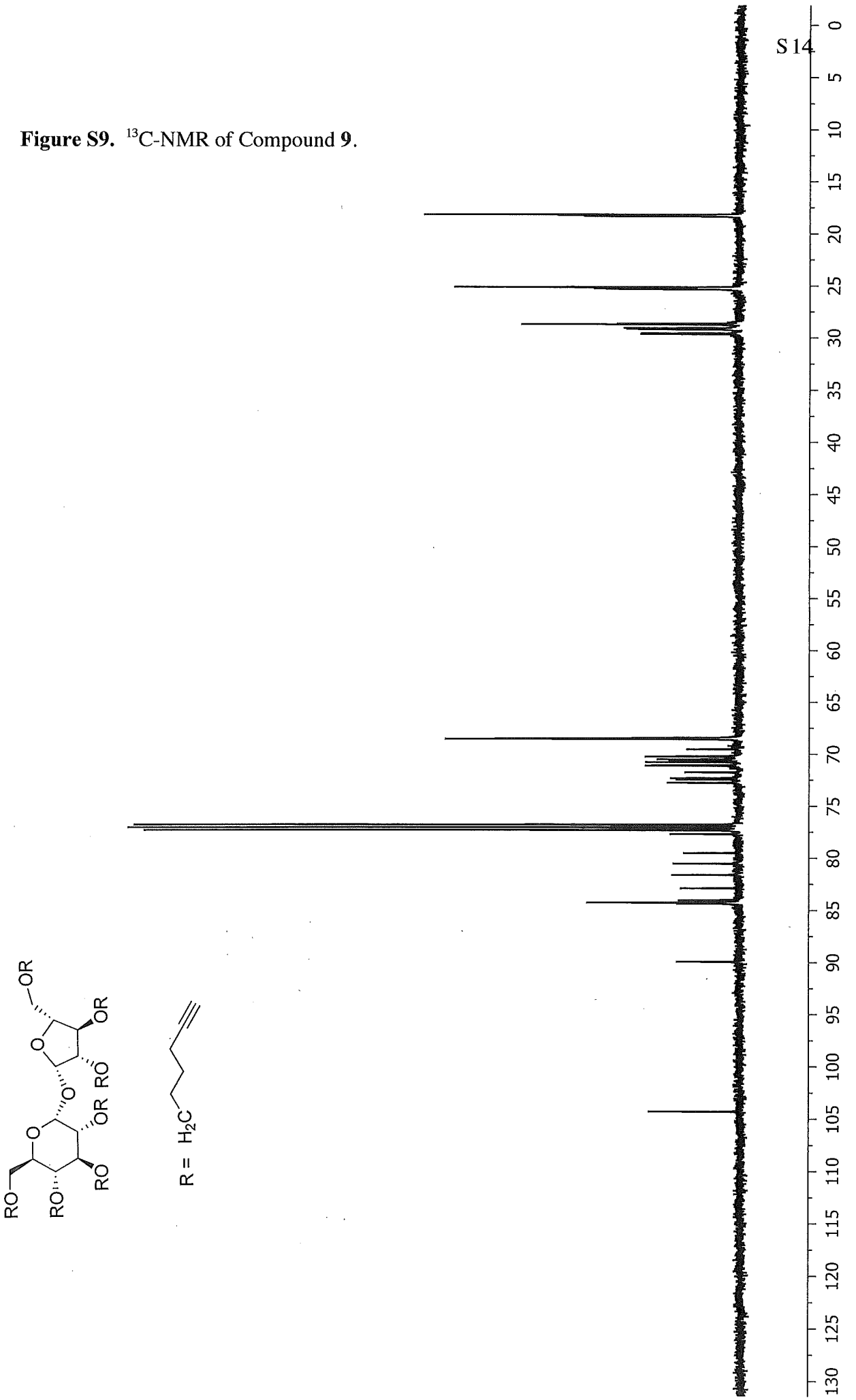


Figure S10. ^{1}H-NMR of Compound **11**.

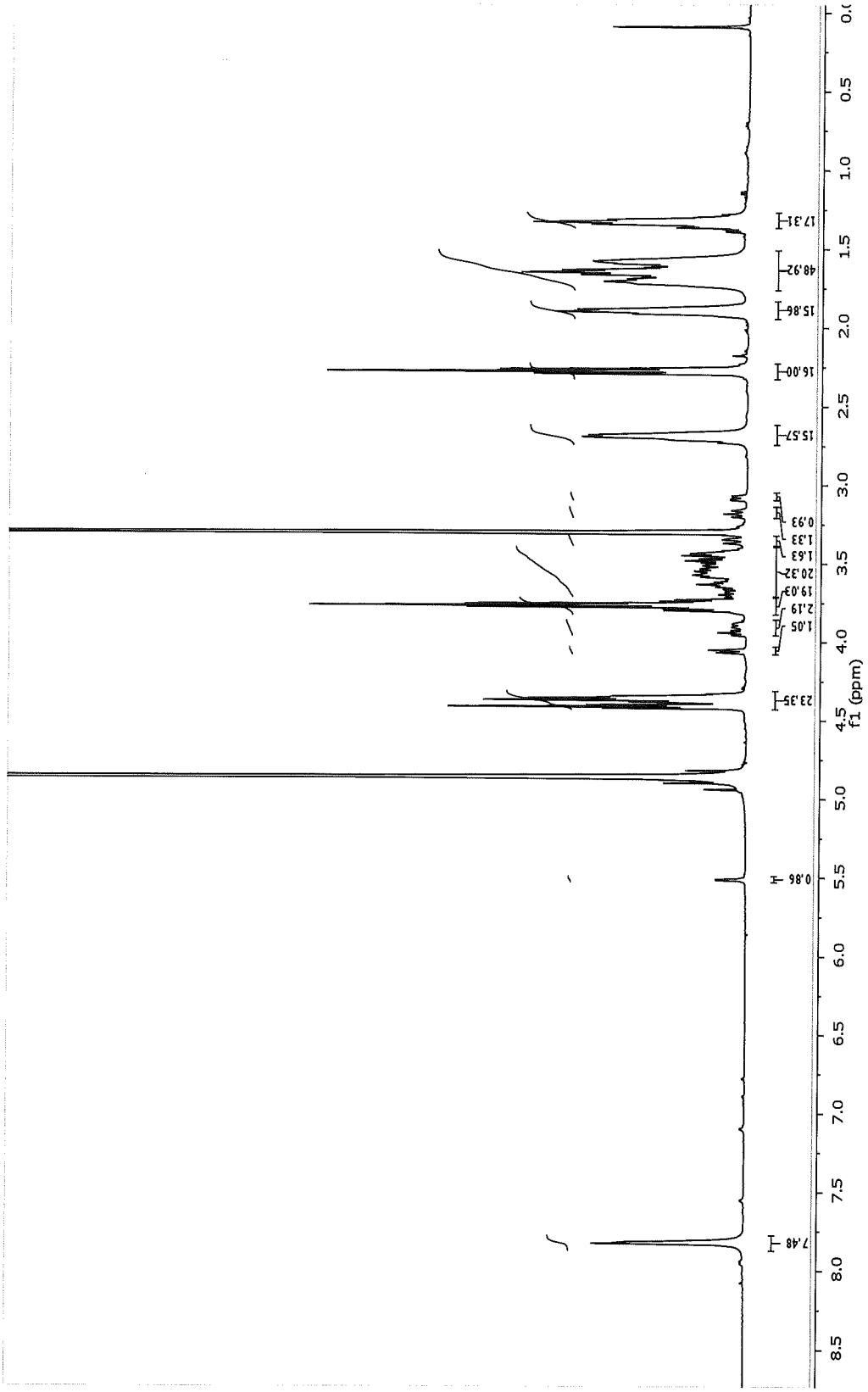


Figure S11. ^{13}C-NMR of Compound **11**.