

# Supplementary material

## Metabolomics as a Potential Chemotaxonomical Tool: Application on the Selected *Euphorbia* Species Growing Wild in Serbia

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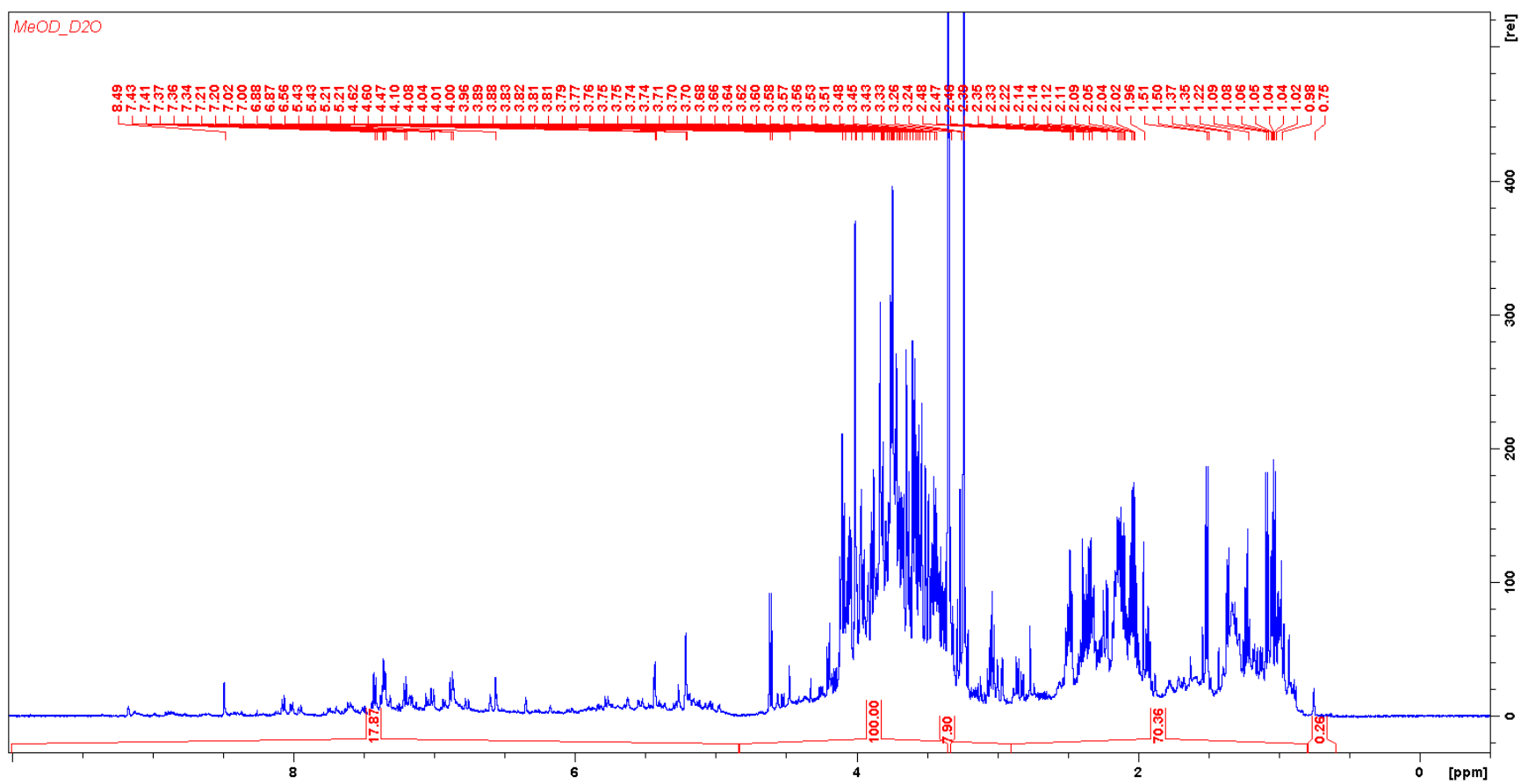


Figure S1:  $^1\text{H}$  NMR spectrum of *E. salicifolia* extract of 1:1 mixture of deuterated methanol and potassium phosphate buffer in deuterated water.

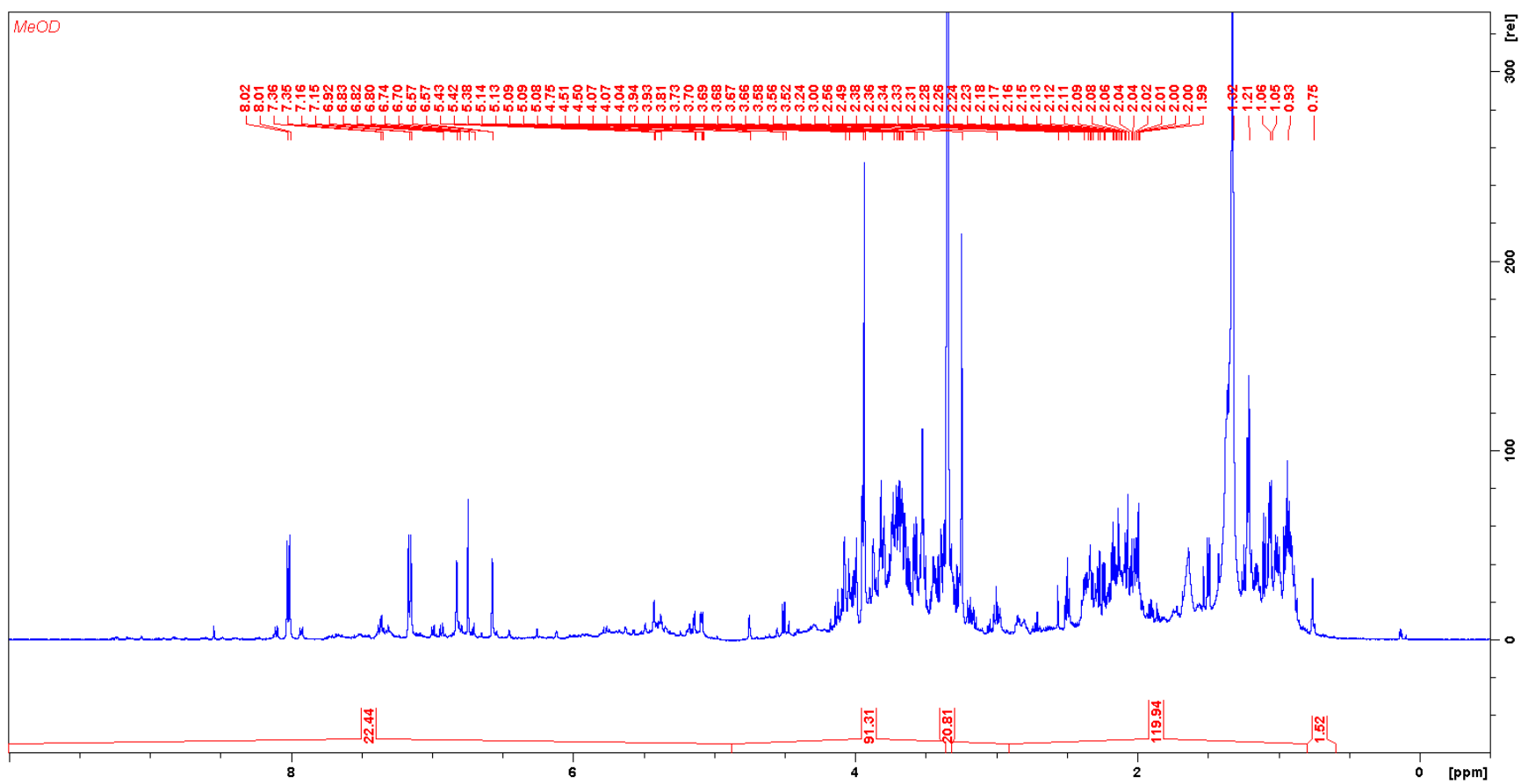


Figure S2:  $^1\text{H}$  NMR spectrum of *E. salicifolia* extract of deuterated methanol.

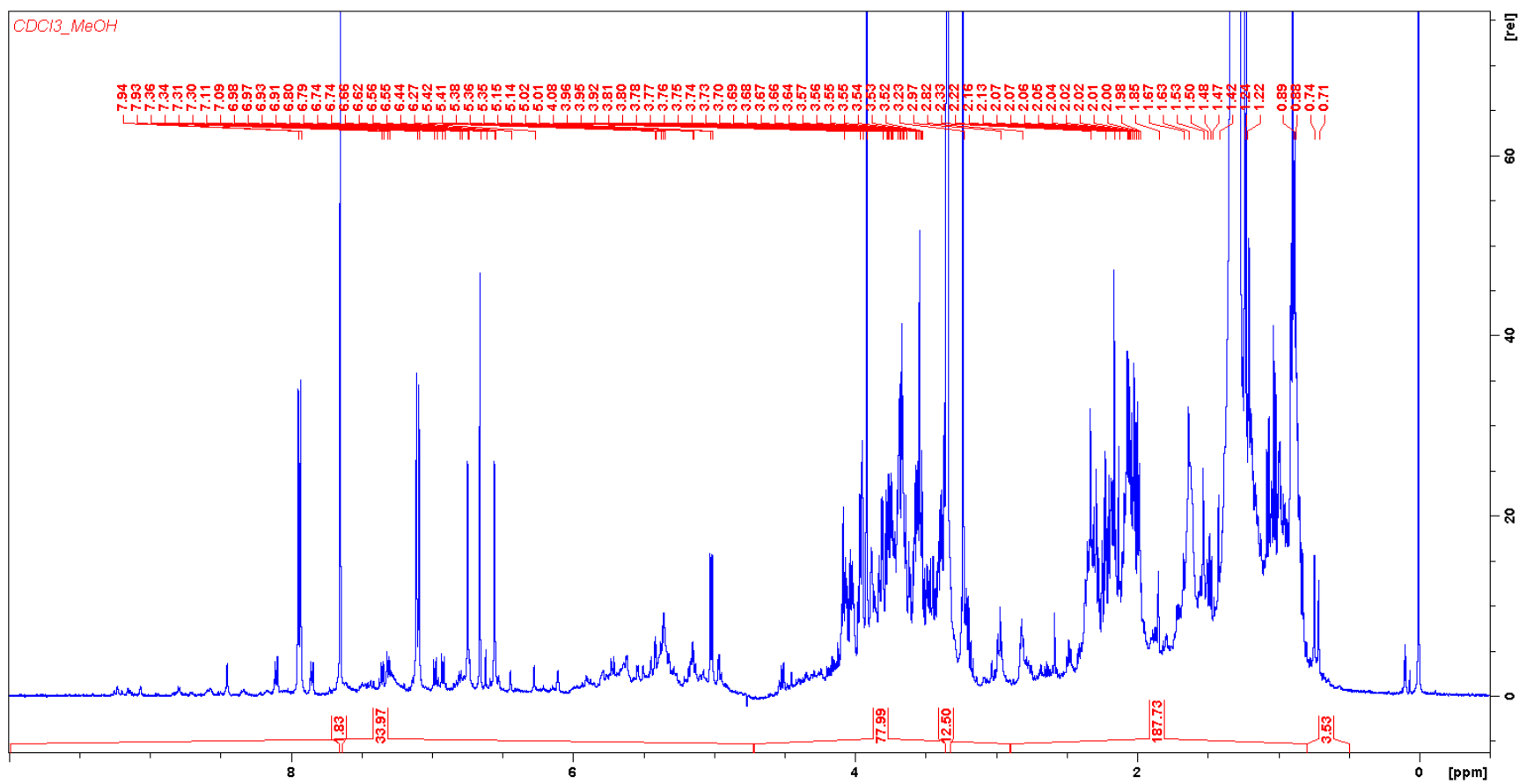


Figure S3: <sup>1</sup>H NMR spectrum of *E. salicifolia* extract of 1:1 mixture of deuterated methanol and deuterated chloroform.

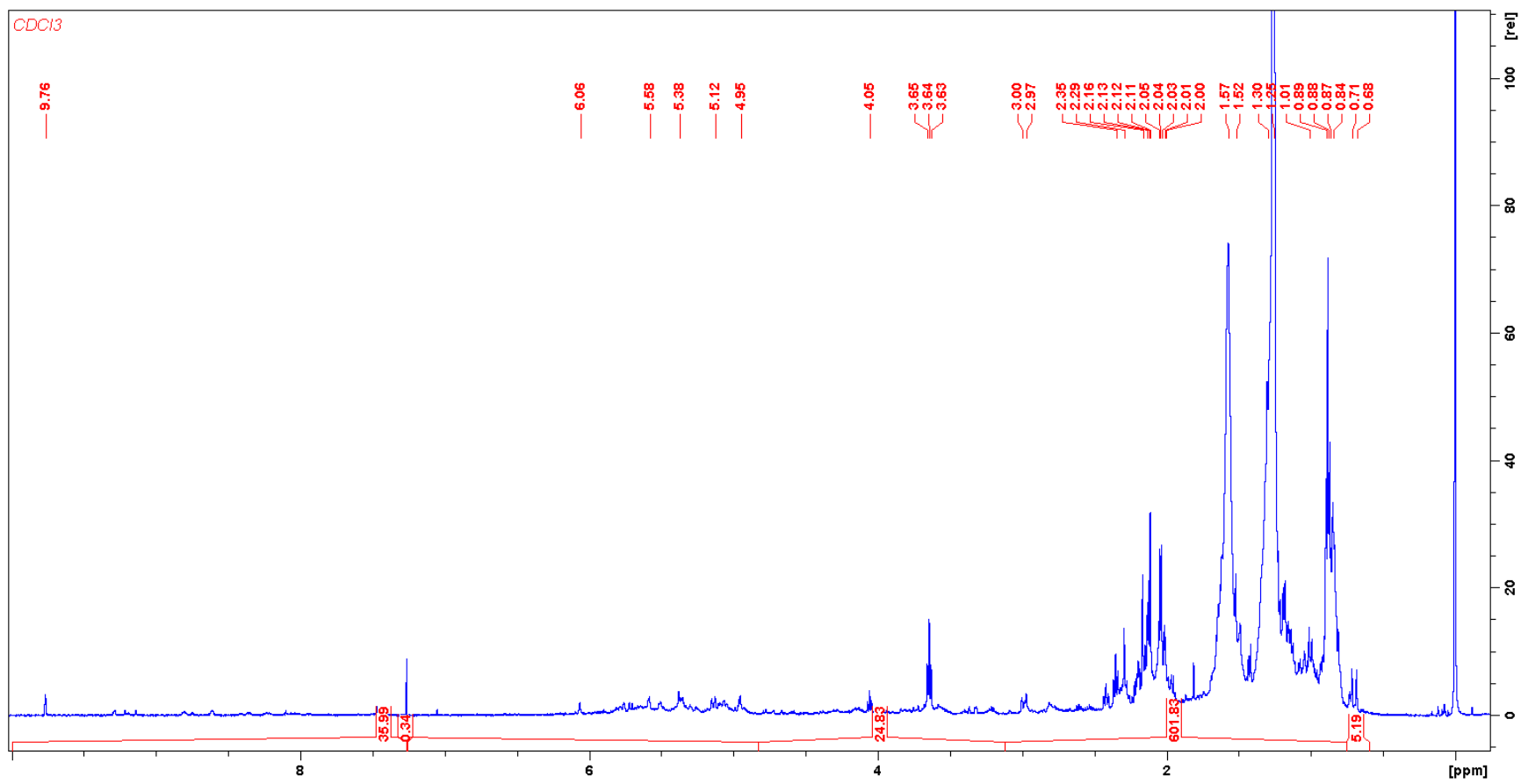


Figure S4: <sup>1</sup>H NMR spectrum of *E. salicifolia* extract of deuterated chloroform.

Table S1. Validation parameters of CV-ANOVA test from OPLS-DA model of *E. segueriana* vs. remaining *Euphorbia* species.

M11	SS	D F	MS	F	p	SD
Total corr.	59	59	1			1
Regression	58.26919937	8	7.283649921	508.3110046	0	2.698820114
Residual	0.730785012	51	0.0143291			0.119704001

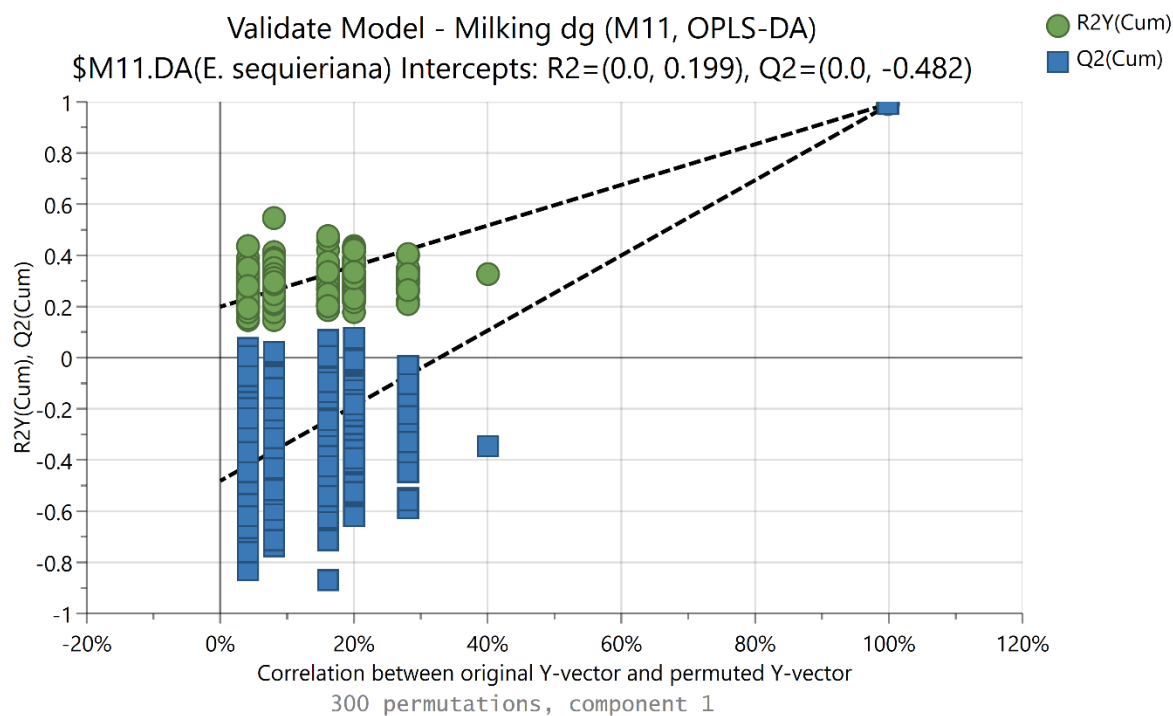


Figure S5: Permutation test of OPLS-DA model of *E. segueriana* vs. remaining *Euphorbia* species.

Table S2. Validation parameters of CV-ANOVA test from OPLS-DA model of *E. salicifolia* vs. remaining *Euphorbia* species.

M12	SS	D F	MS	F	p	SD
Total corr.	59	59	1			1
Regression	58.50939941	8	7.313670158	760.2769775	0	2.704380035
Residual	0.490606993	51	0.00961975			0.0980803

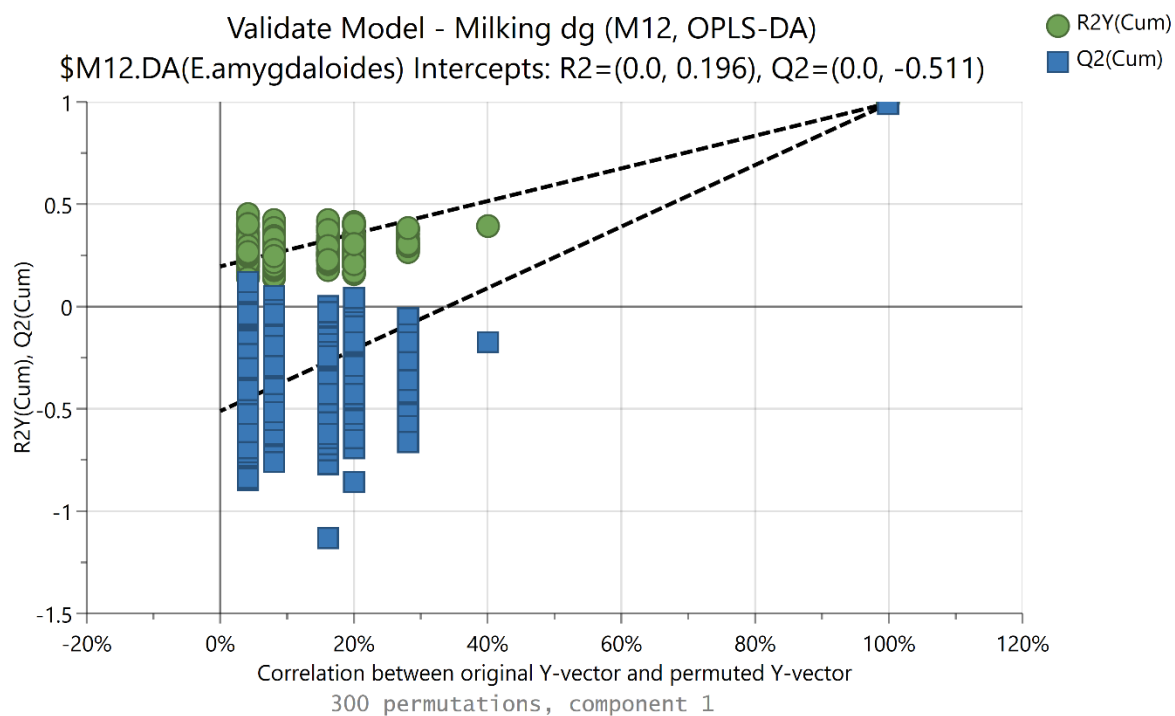


Figure S6: Permutation test of OPLS-DA model of *E. salicifolia* vs. remaining *Euphorbia* species.

Table S3. Validation parameters of CV-ANOVA test from OPLS-DA model of *E. amygdaloides* vs. remaining *Euphorbia* species.

M13	SS	DF	MS	F	p	SD
Total corr.	59	59	1			1
Regression	57.87189865	8	7.233990192	327.0490112	0	2.689610004
Residual	1.128069997	51	0.022119001			0.148725003

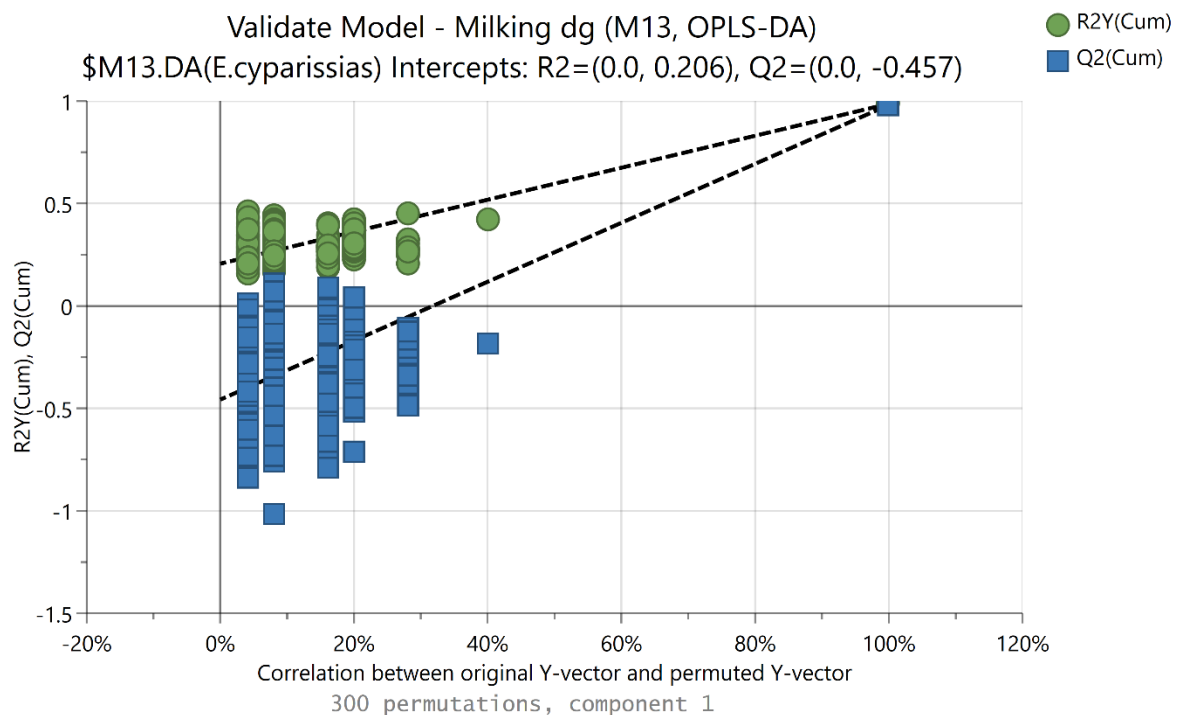


Figure S7: Permutation test of OPLS-DA model of *E. amygdaloides* vs. remaining *Euphorbia* species.



Table S4. Validation parameters of CV-ANOVA test from OPLS-DA model of *E. panonnica* vs. remaining *Euphorbia* species.

M14	SS	D F	MS	F	p	SD
Total corr.	59	59	1			1
Regression	57.73199844	8	7.216499805	290.2539978	0	2.686350107
Residual	1.268000007	51	0.024862699			0.157679006

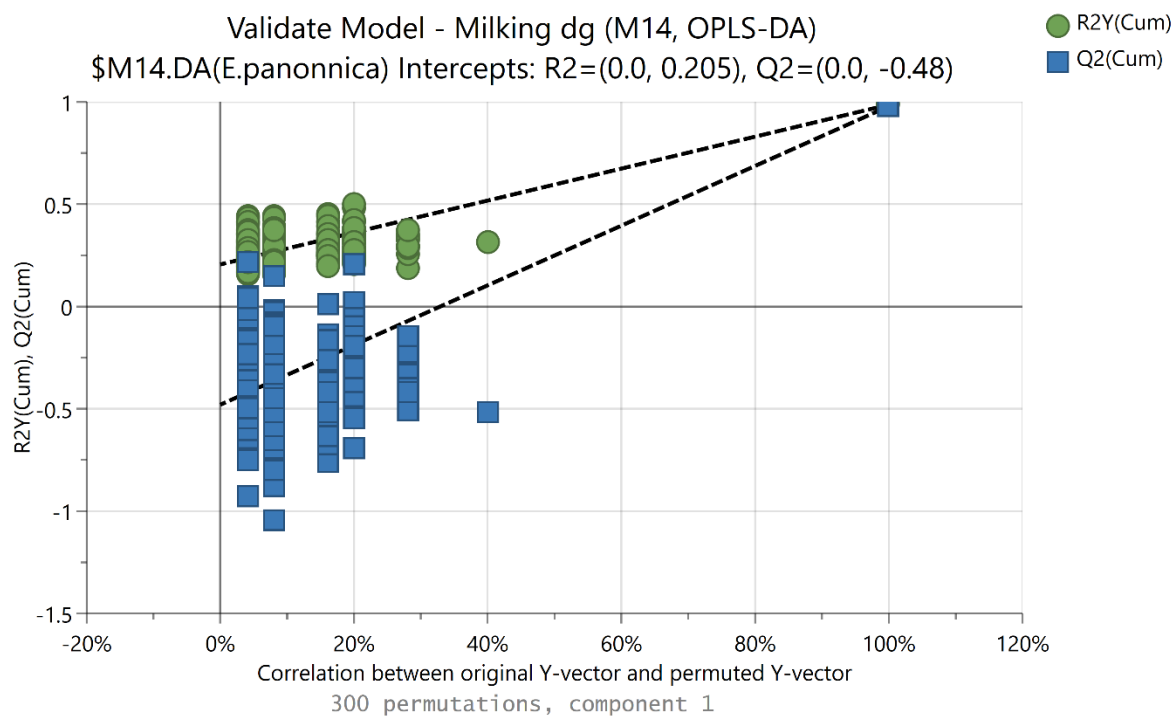


Figure S8: Permutation test of OPLS-DA model of *E. panonnica* vs. remaining *Euphorbia* species.

Table S5. Validation parameters of CV-ANOVA test from OPLS-DA model of *E. cyparissias* vs. remaining *Euphorbia* species.

M15	SS	D F	MS	F	p	SD
Total corr.	59	59	1			1
Regression	58.48870087	6	9.748109818	1010.400024	0	3.122200012
Residual	0.511334002	53	0.00964781			0.098223299

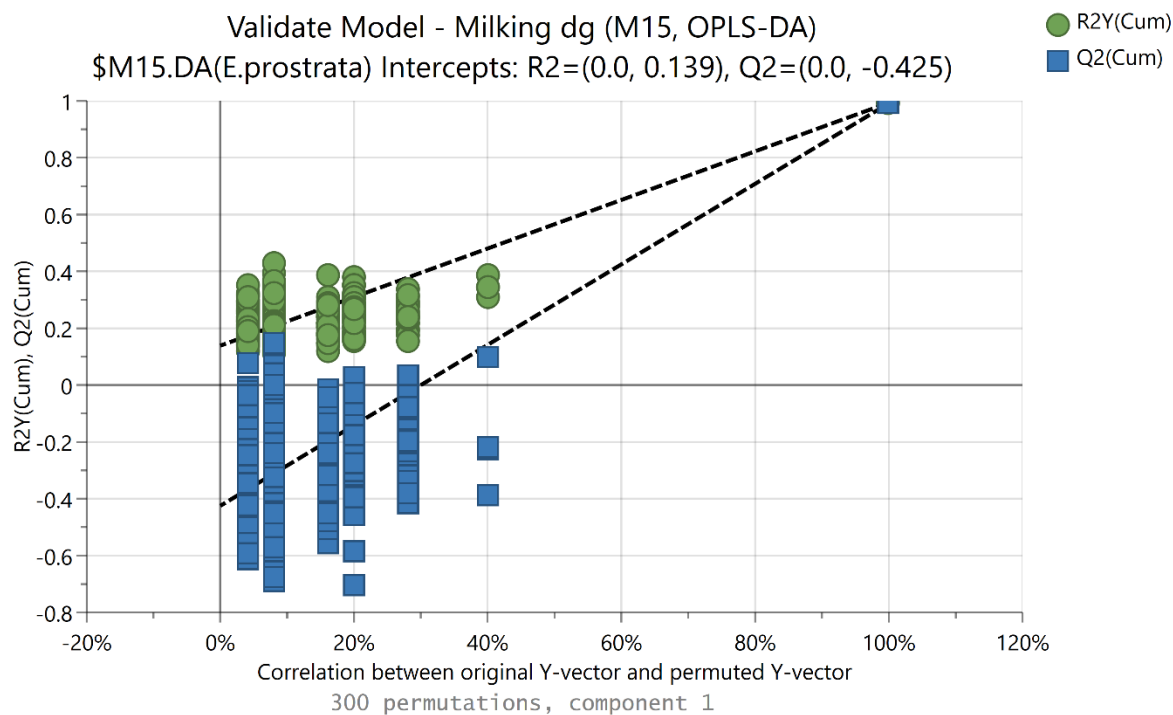


Figure S9: Permutation test of OPLS-DA model of *E. cyparissias* vs. remaining *Euphorbia* species.

Table S6. Validation parameters of CV-ANOVA test from OPLS-DA model of *E. maculata* vs. remaining *Euphorbia* species.

M16	SS	D F	MS	F	p	SD
Total corr.	59	59	1			1
Regression	58.3791008	4	14.5948	1292.719971	0	3.820310116
Residual	0.62094599	55	0.0112899			0.106253996

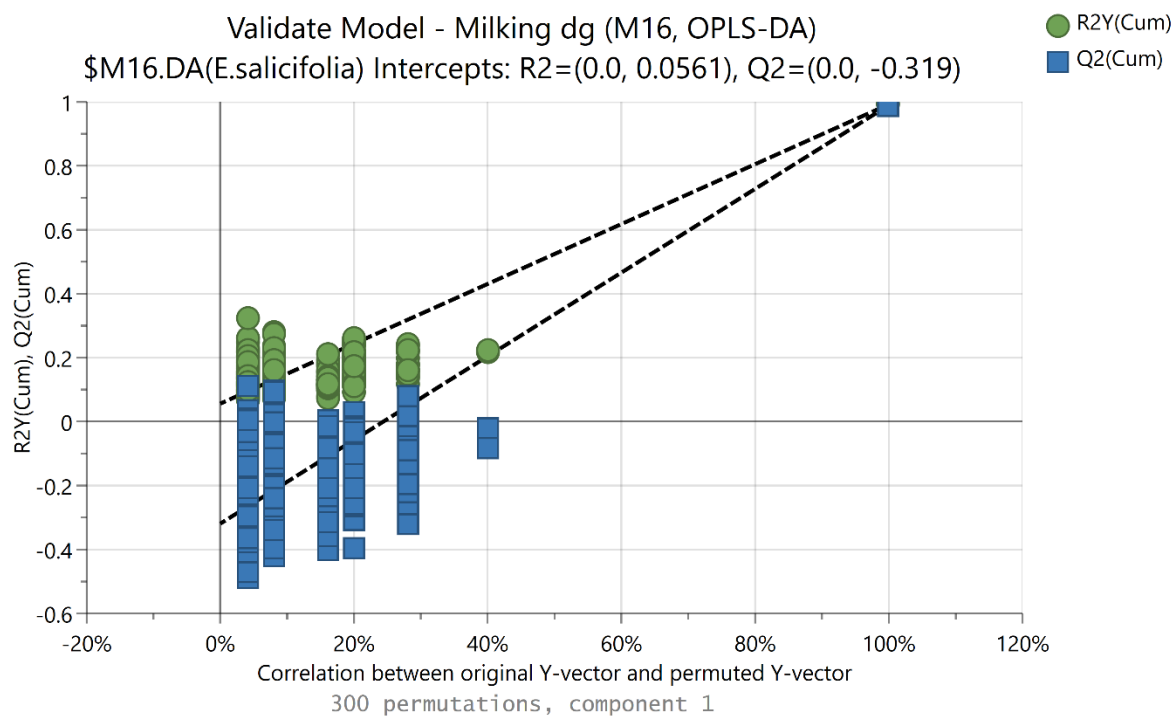


Figure S10: Permutation test of OPLS-DA model of *E. cyparissias* vs. remaining *Euphorbia* species.

Table S7. PCA and OPLS-DA model parameters of tested *Euphorbia* species.

Models									
Number	Model	Type	A	N	R2X(cum)	R2Y(cum)	Q2(cum)	Date	Title
2	M2	PCA-X	17	60	0.994		0.975	10/21/2022	
11	M11	OPLS-DA	1+3+0	60	0.609	0.993	0.988	10/23/2022	<i>E. segueriana</i> vs. remaining <i>Euphorbia</i>
12	M12	OPLS-DA	1+3+0	60	0.635	0.994	0.992	10/23/2022	<i>E. salicifolia</i> vs. remaining <i>Euphorbia</i>
13	M13	OPLS-DA	1+3+0	60	0.671	0.986	0.981	10/23/2022	<i>E. amygdaloides</i> vs. remaining <i>Euphorbia</i>
15	M15	OPLS-DA	1+2+0	60	0.595	0.993	0.991	10/23/2022	<i>E. panonnica</i> vs. remaining <i>Euphorbia</i>
14	M14	OPLS-DA	1+3+0	60	0.675	0.986	0.979	10/23/2022	<i>E. cyparissias</i> vs. remaining <i>Euphorbia</i>
16	M16	OPLS-DA	1+1+0	60	0.442	0.992	0.989	11/13/2022	<i>E. maculata</i> vs. remaining <i>Euphorbia</i>

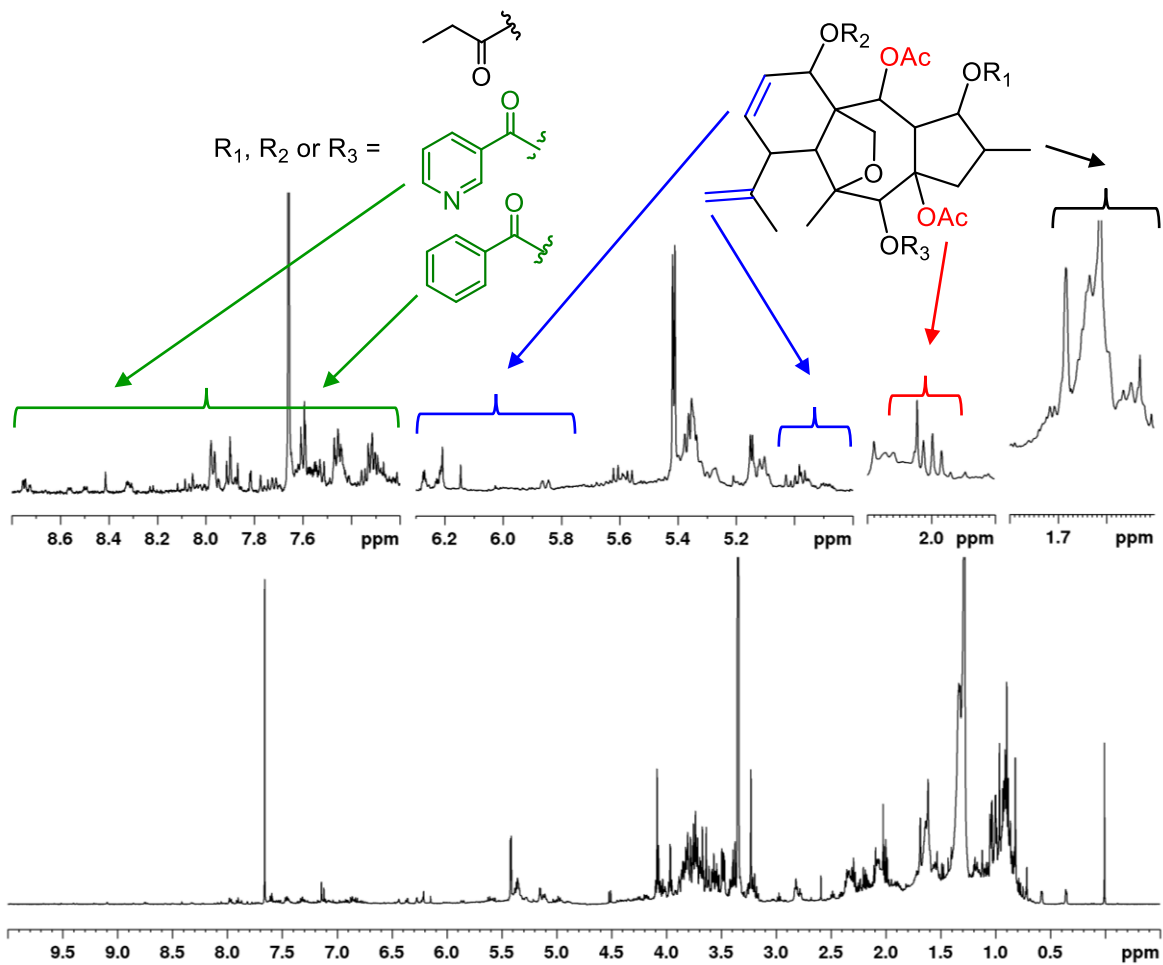


Figure S11: The  $^1\text{H}$  NMR spectrum of *E. segueriana* extract obtained with MeOD:CDCl<sub>3</sub>(1:1) with assignment of characteristic resonances

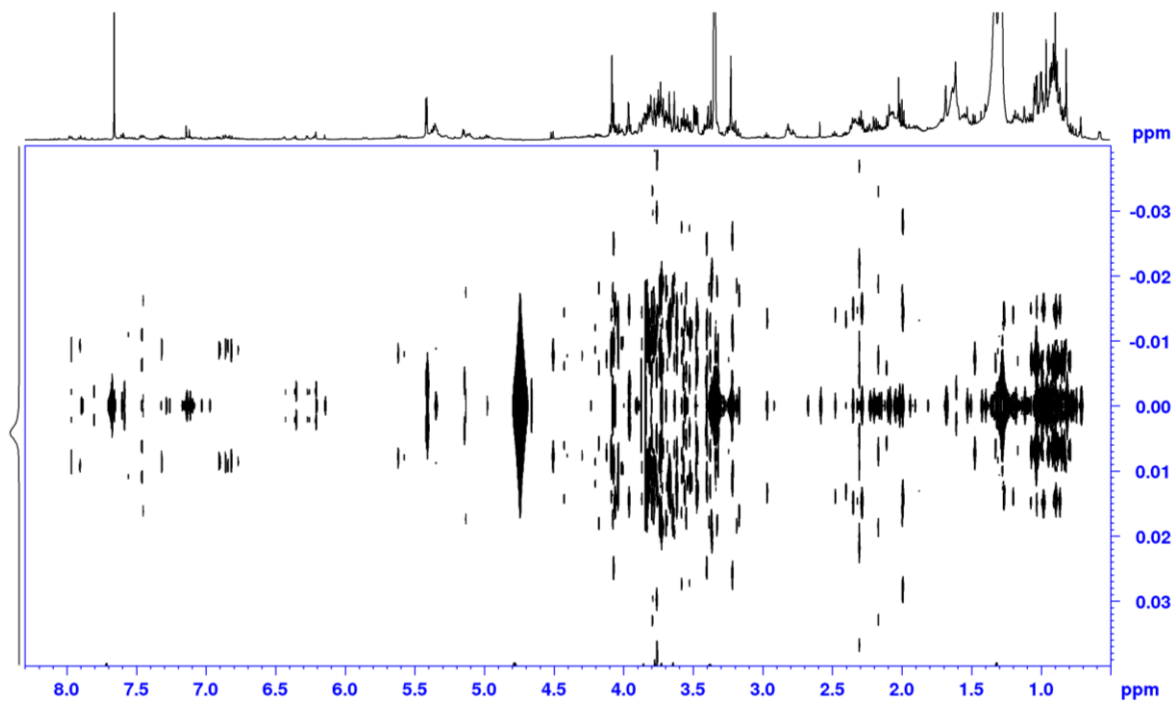


Figure S12: The H,H-J-resolved NMR spectrum of *E. segueriana* extract obtained with MeOD:CDCl<sub>3</sub>(1:1)

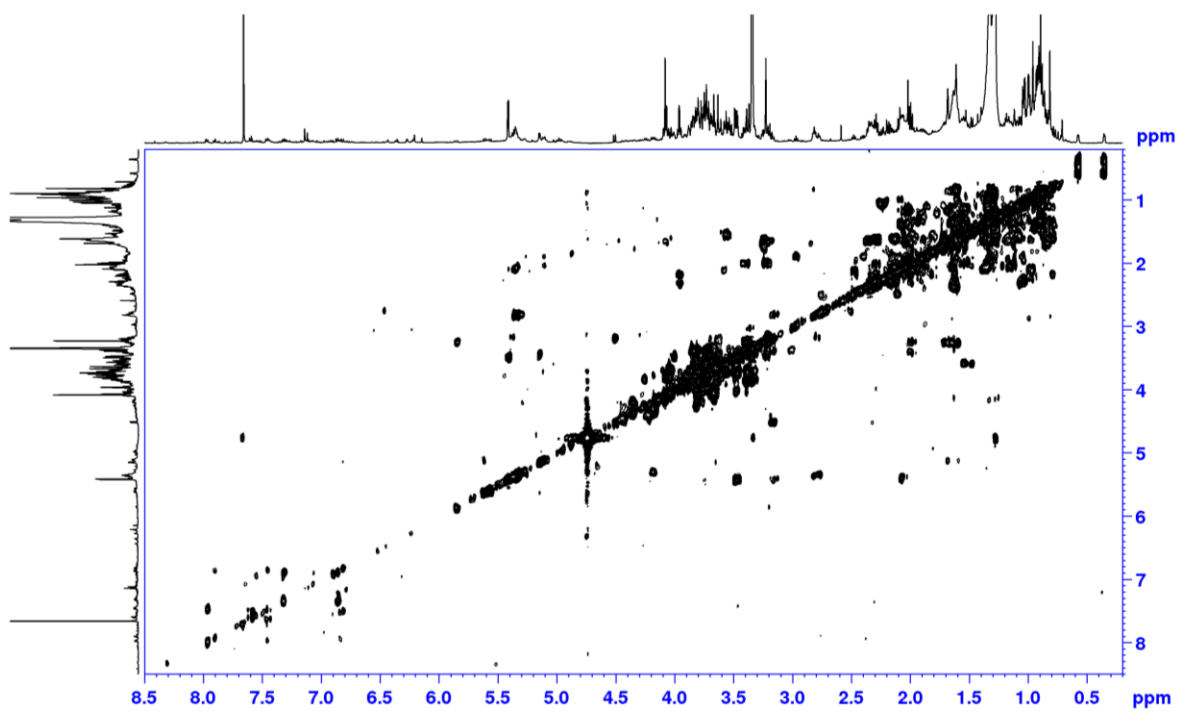


Figure S13: The COSY NMR spectrum of *E. segetiana* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

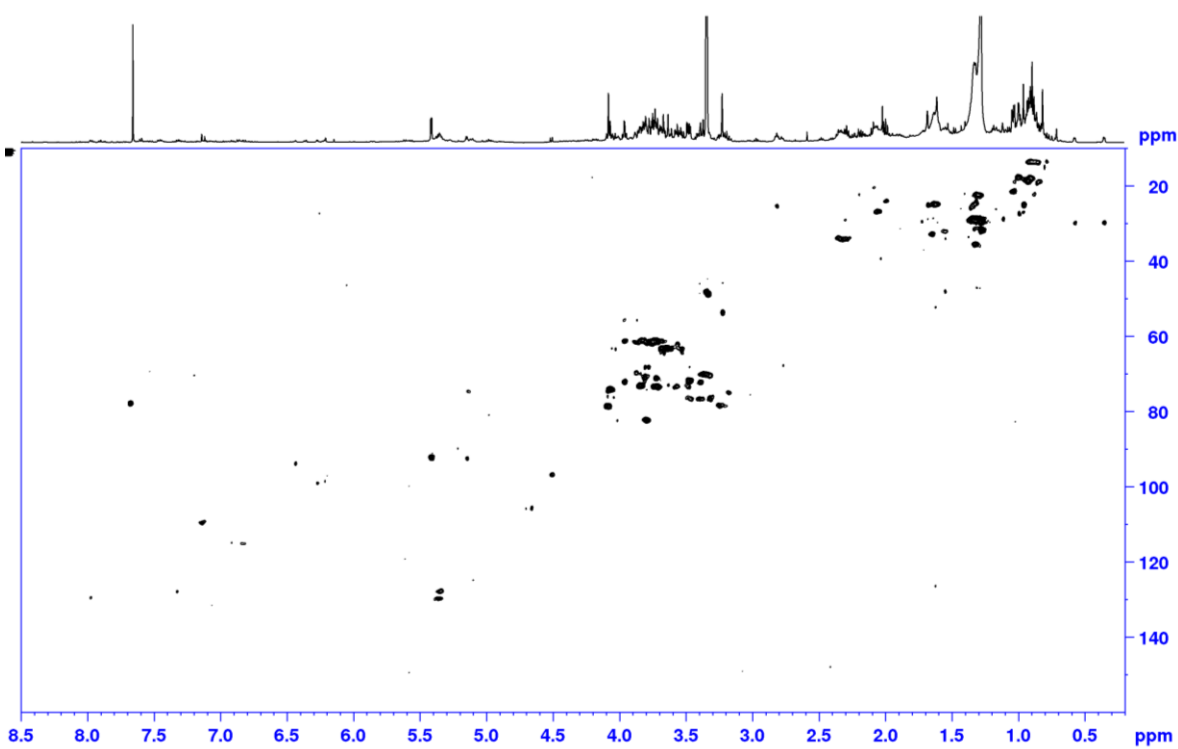


Figure S14: The HSQC NMR spectrum of *E. segetiana* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

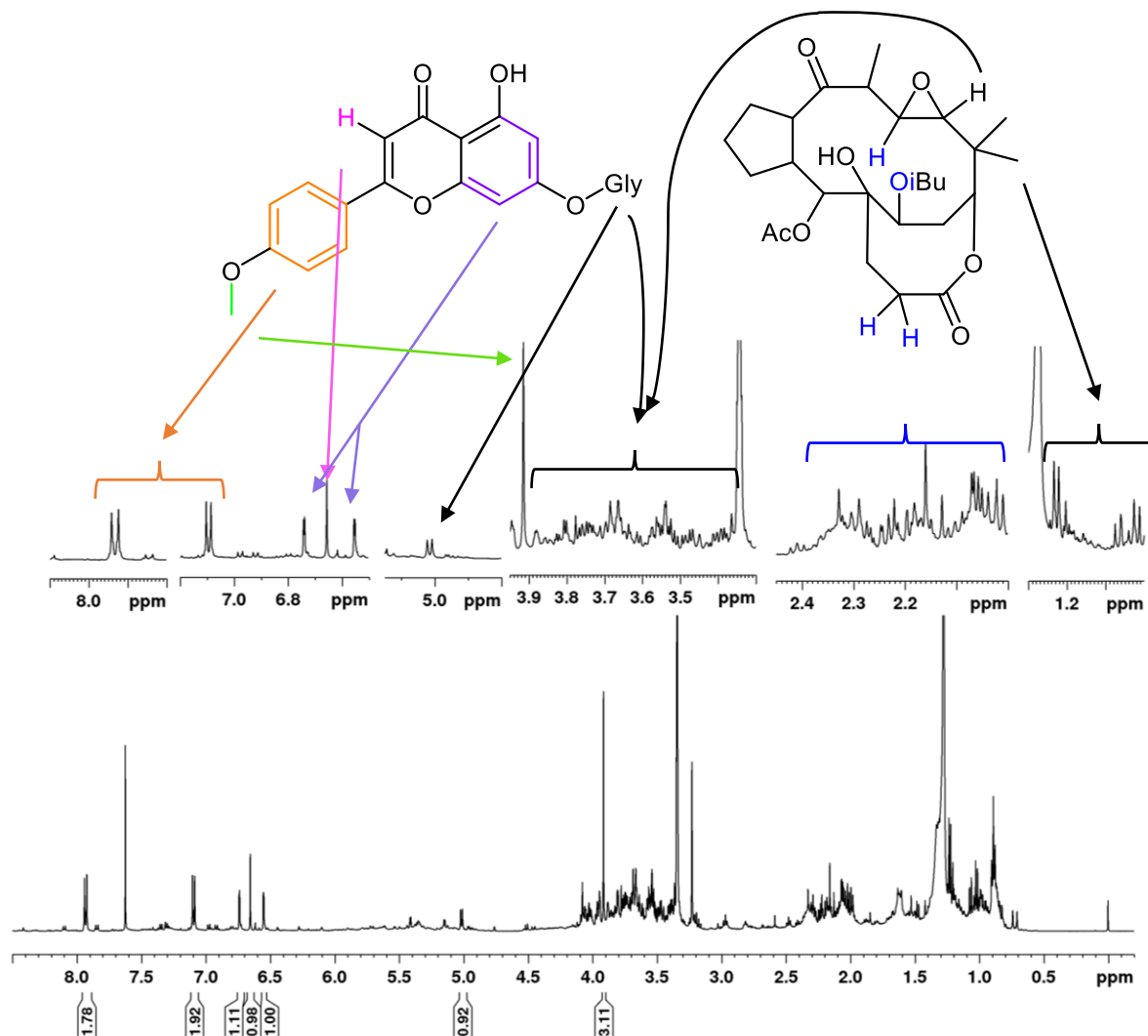


Figure S15: The  $^1\text{H}$  NMR spectrum of *E. salicifolia* extract obtained with MeOD:CDCl<sub>3</sub> (1:1) with assignment of characteristic resonances

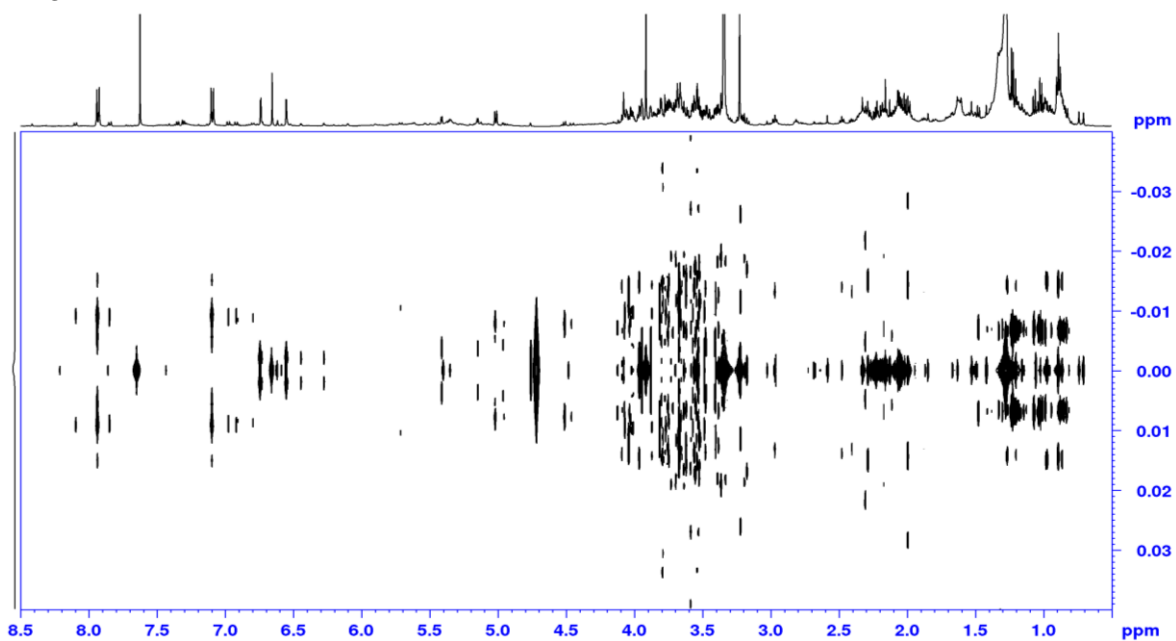


Figure S16: The  $^1\text{H}$ ,  $^1\text{H}$ -J-resolved NMR spectrum of *E. salicifolia* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

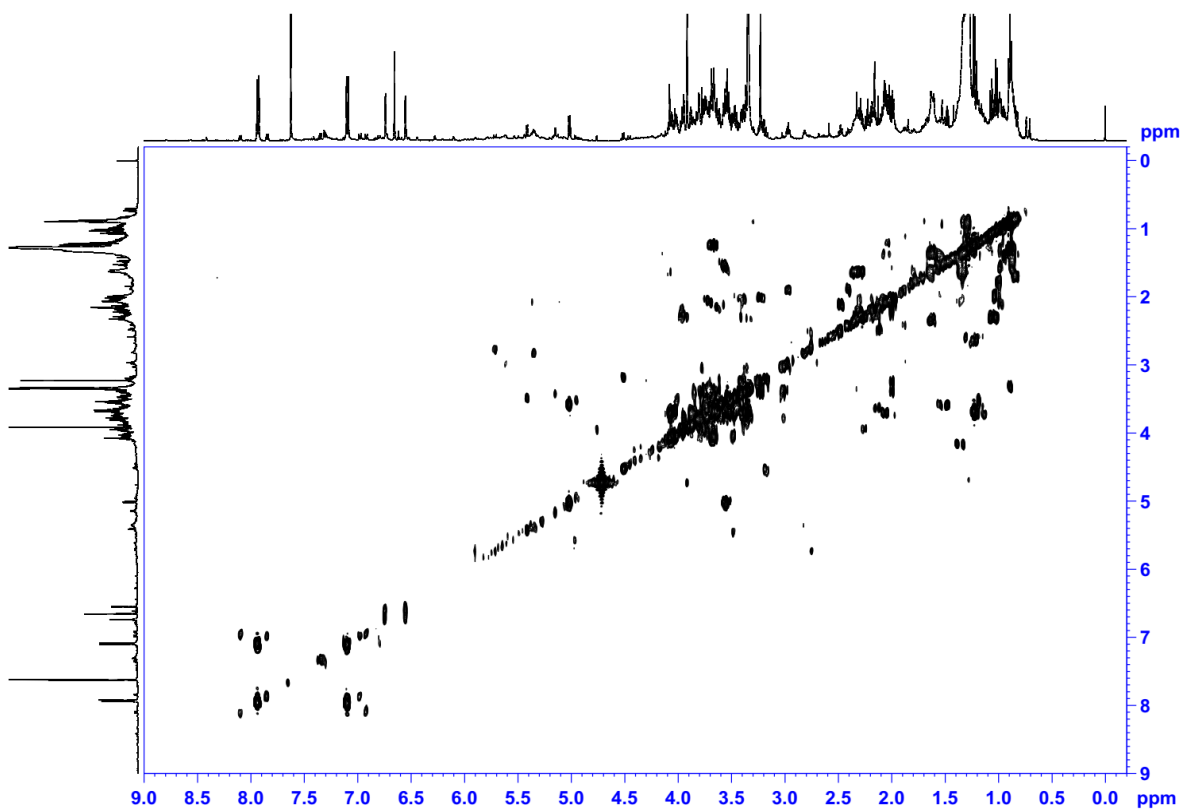


Figure S17: The COSY NMR spectrum of *E. salicifolia* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

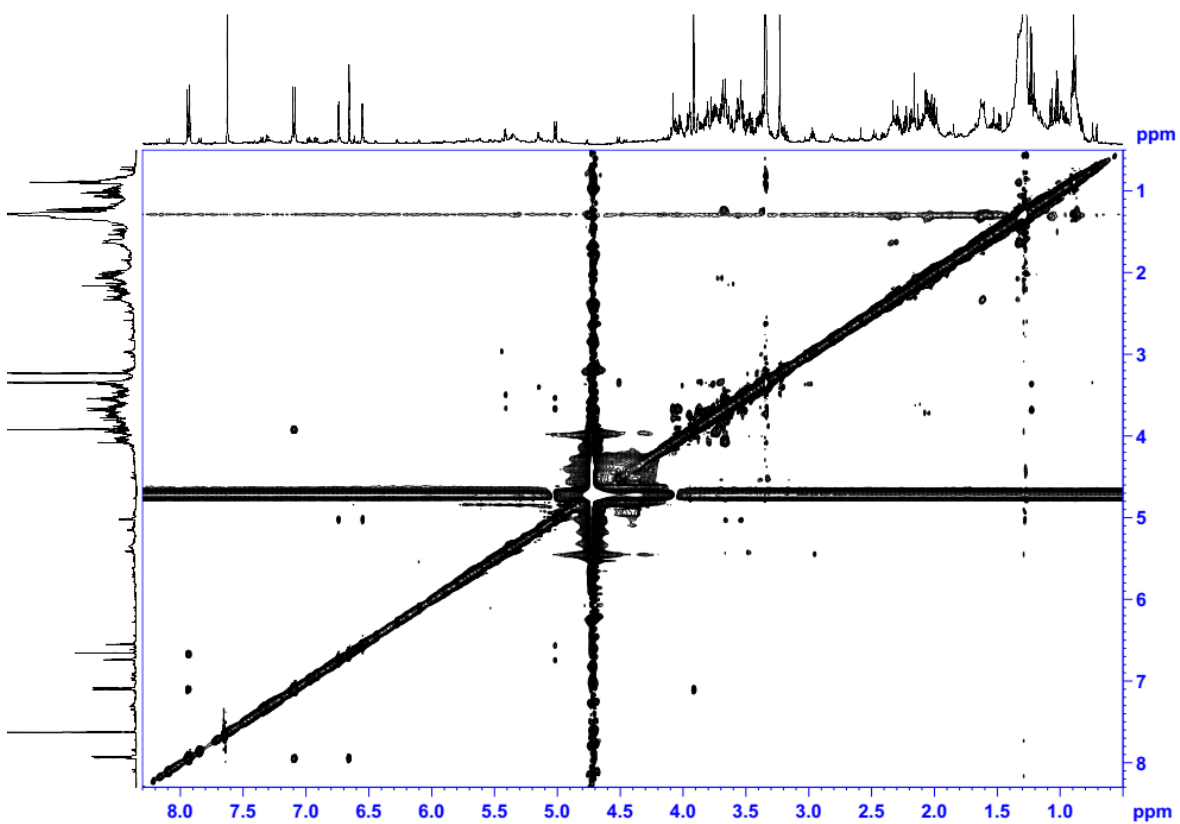


Figure S18: The NOESY NMR spectrum of *E. salicifolia* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)



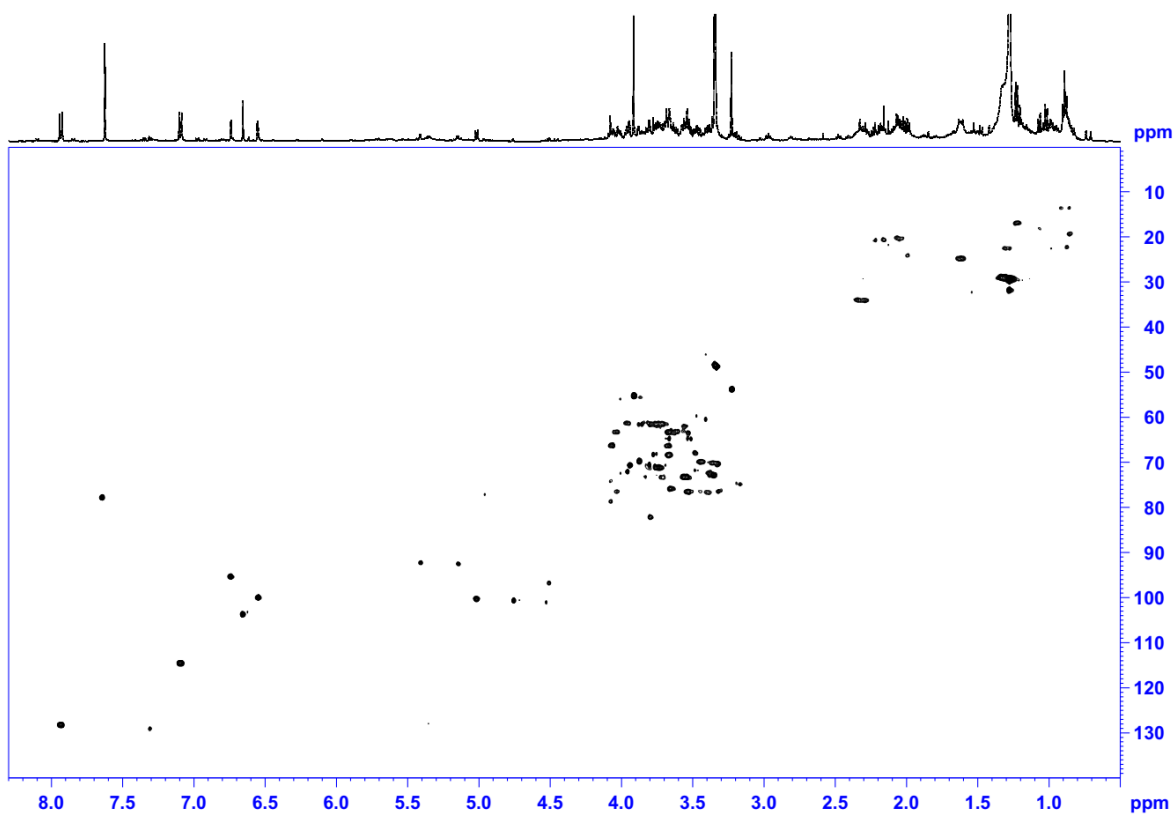


Figure S19: The HSQC NMR spectrum of *E. salicifolia* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

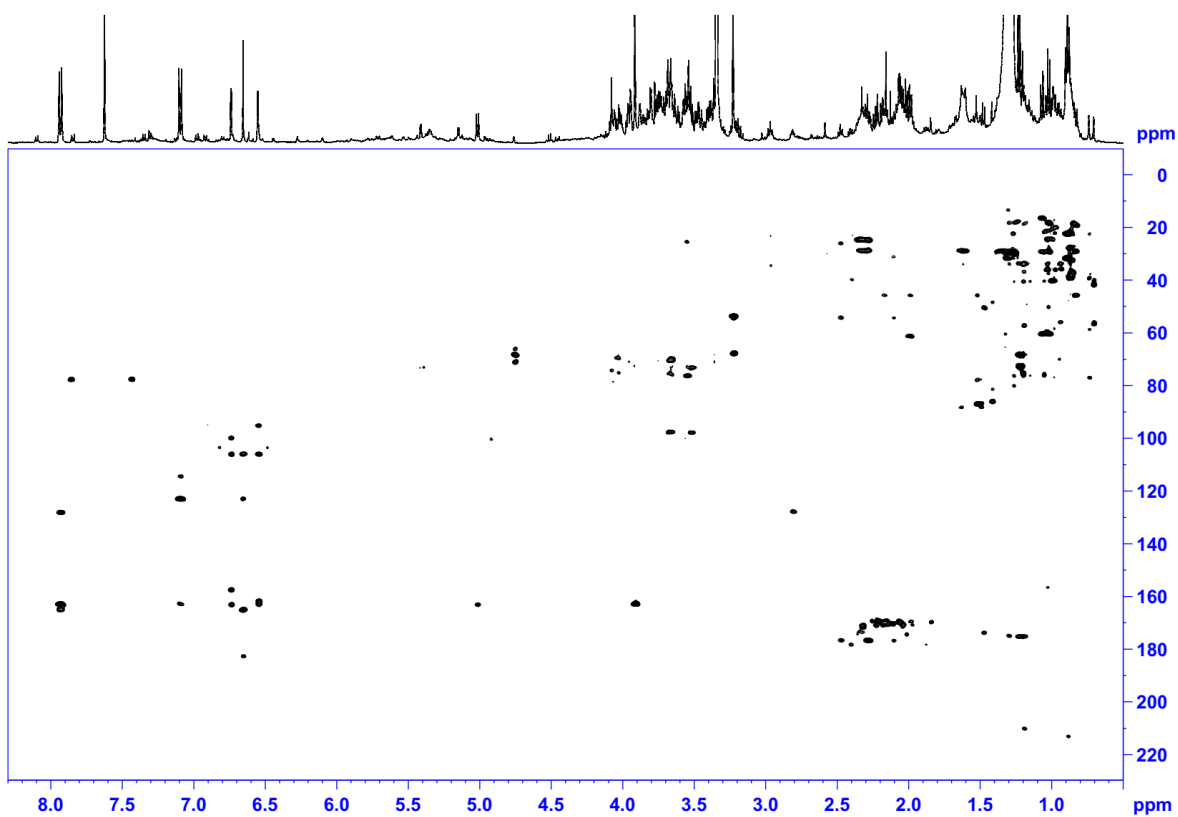


Figure S20: The HMBC NMR spectrum of *E. salicifolia* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

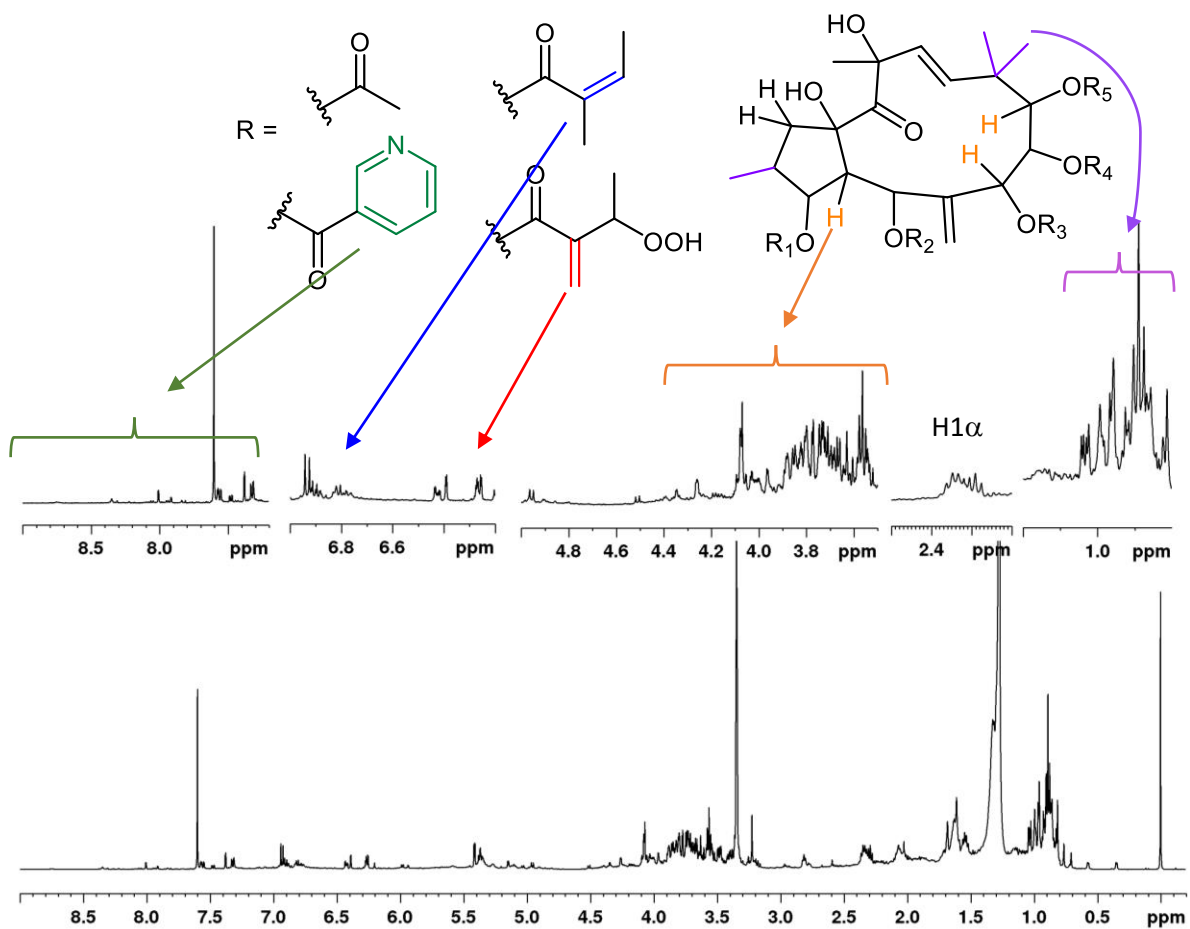


Figure S21: The  $^1\text{H}$  NMR spectrum of *E. amigdaloides* extract obtained with MeOD:CDCl<sub>3</sub> (1:1) with assignment of characteristic resonances

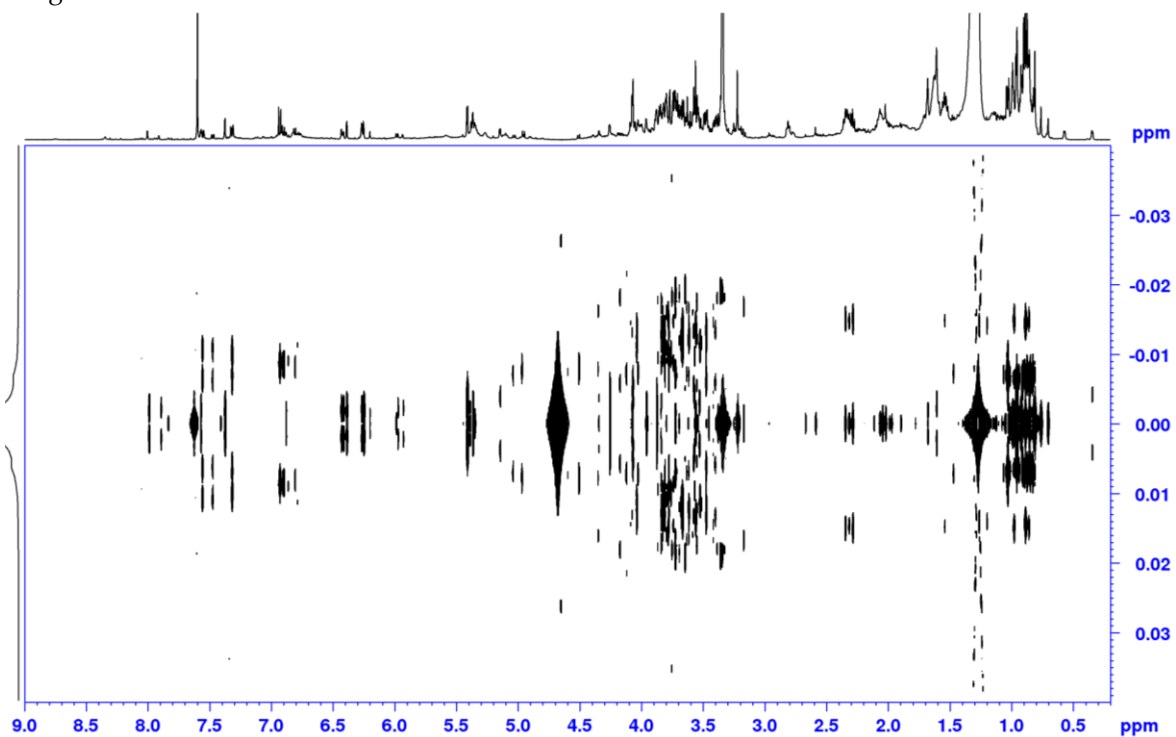


Figure S22: The  $^1\text{H}$ ,  $^1\text{H}$ -J-resolved NMR spectrum of *E. amigdaloides* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

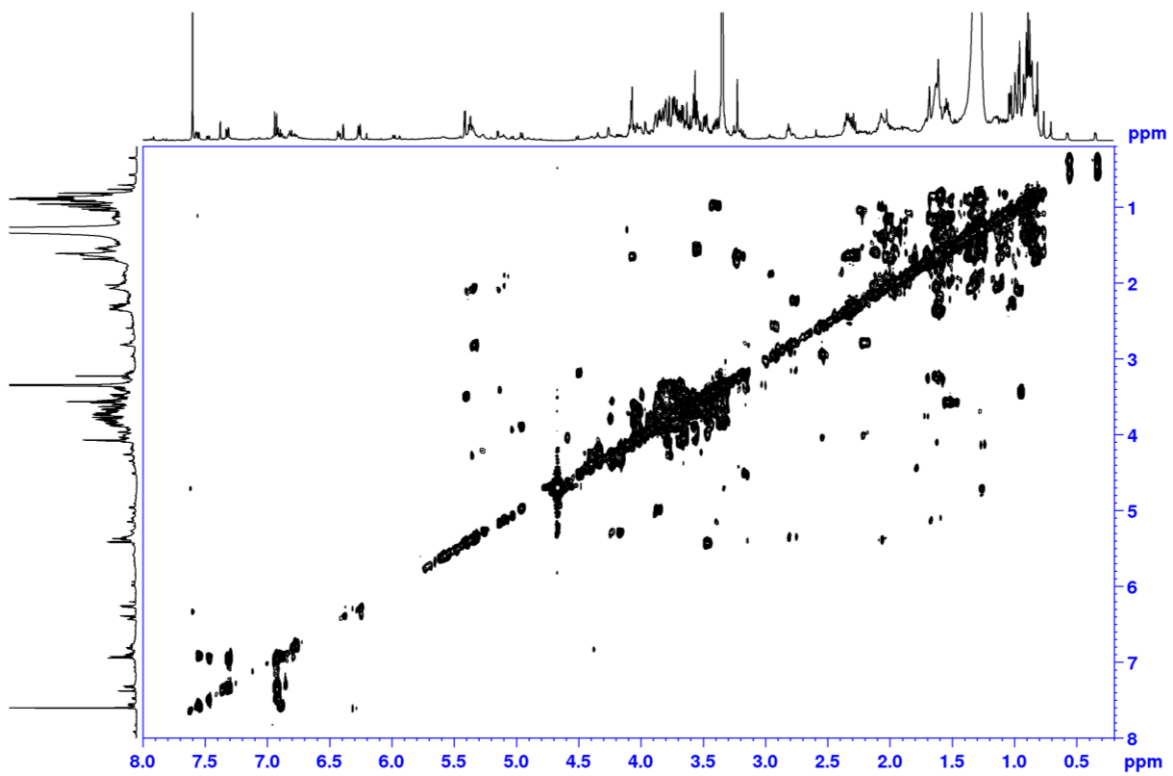


Figure S23: The COSY NMR spectrum of *E. amigdaloides* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

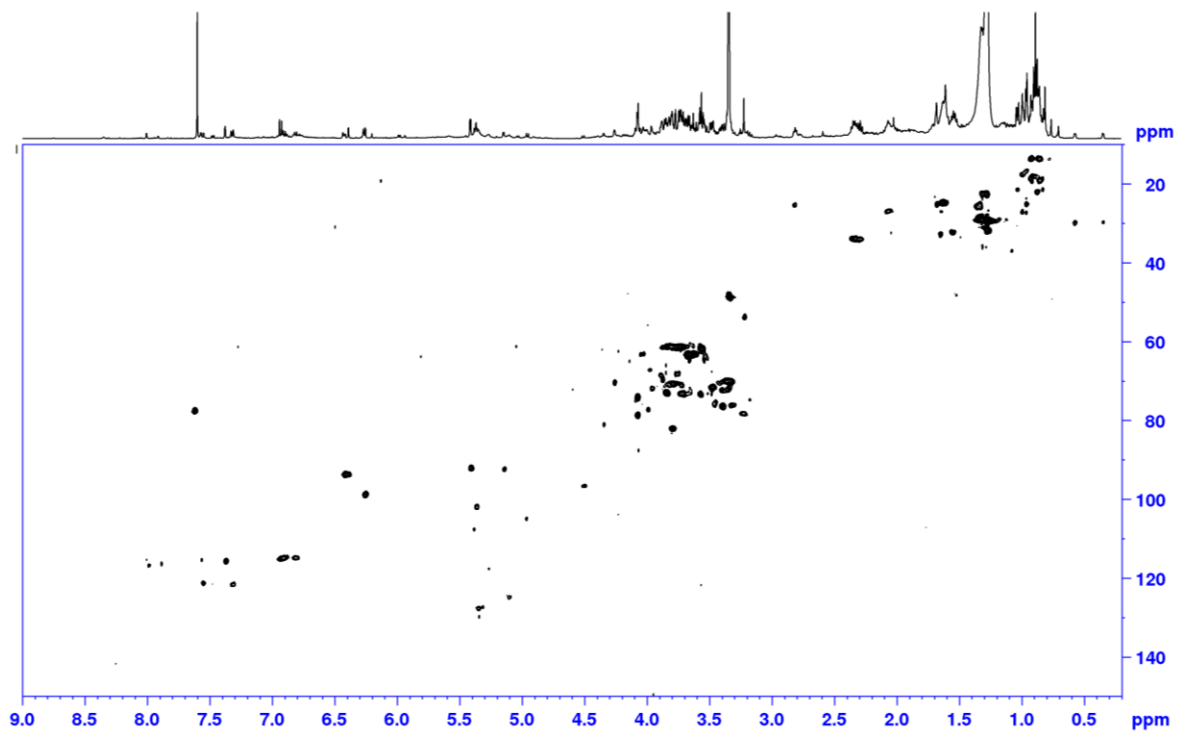


Figure S24: The HSQC NMR spectrum of *E. amigdaloides* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

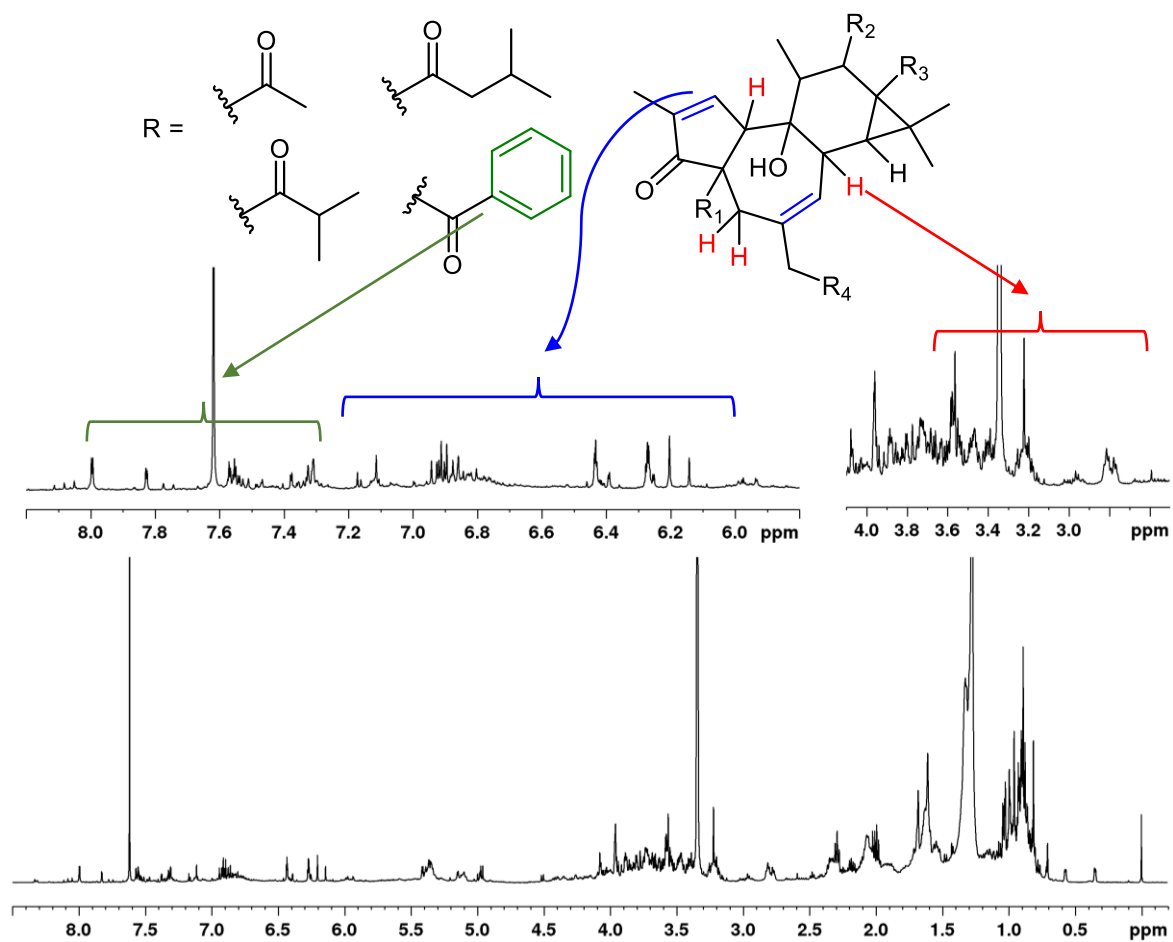


Figure S25: The  $^1\text{H}$  NMR spectrum of *E. panonnica* extract obtained with MeOD:CDCl<sub>3</sub> (1:1) with assignment of characteristic resonances

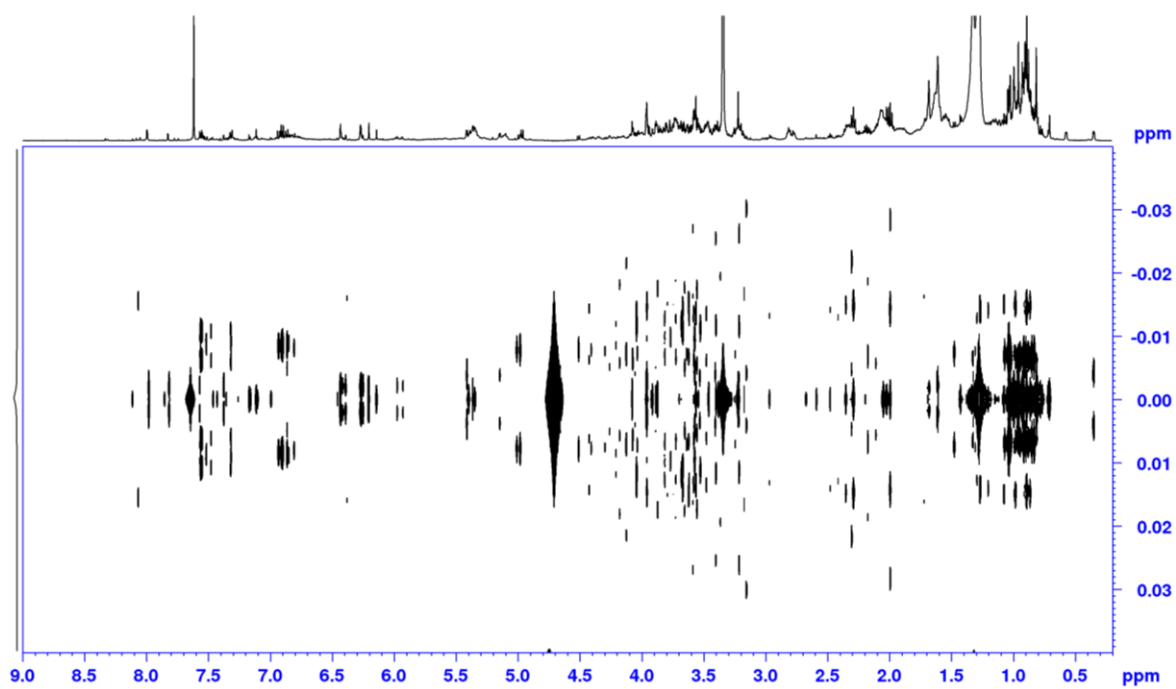


Figure S26: The  $^1\text{H}$ ,  $^1\text{H}$ -J-resolved NMR spectrum of *E. panonnica* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

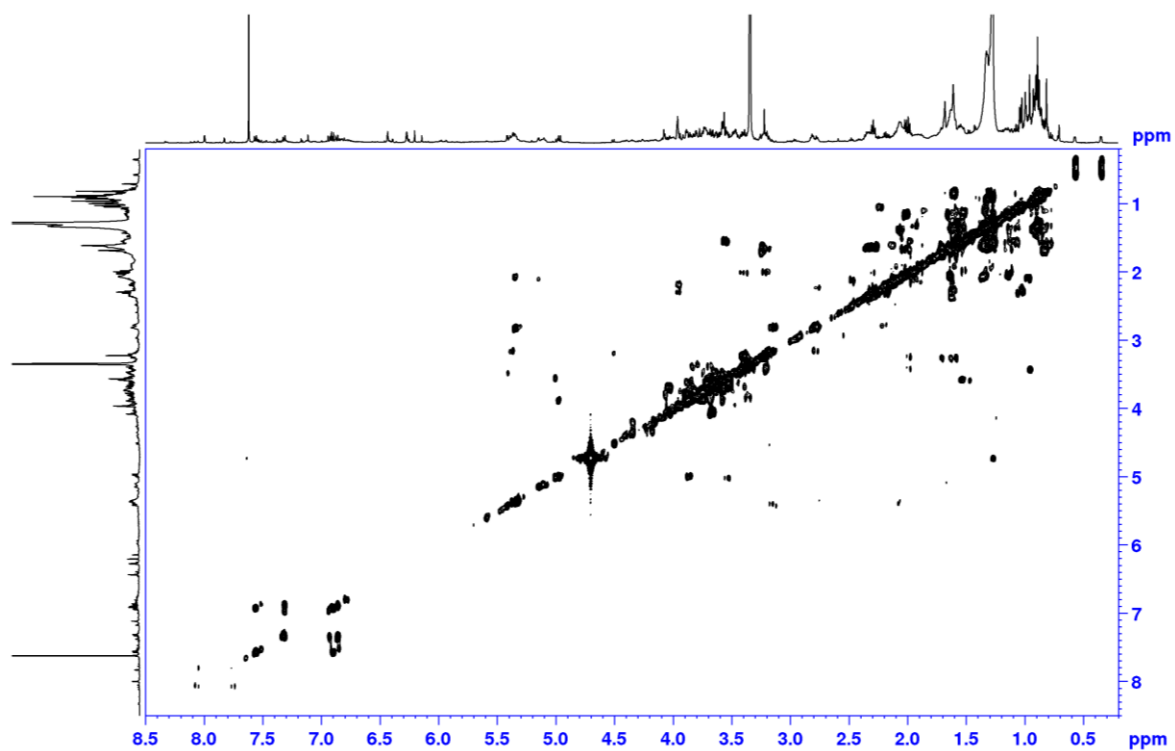


Figure S27: The COSY NMR spectrum of *E. panonnica* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

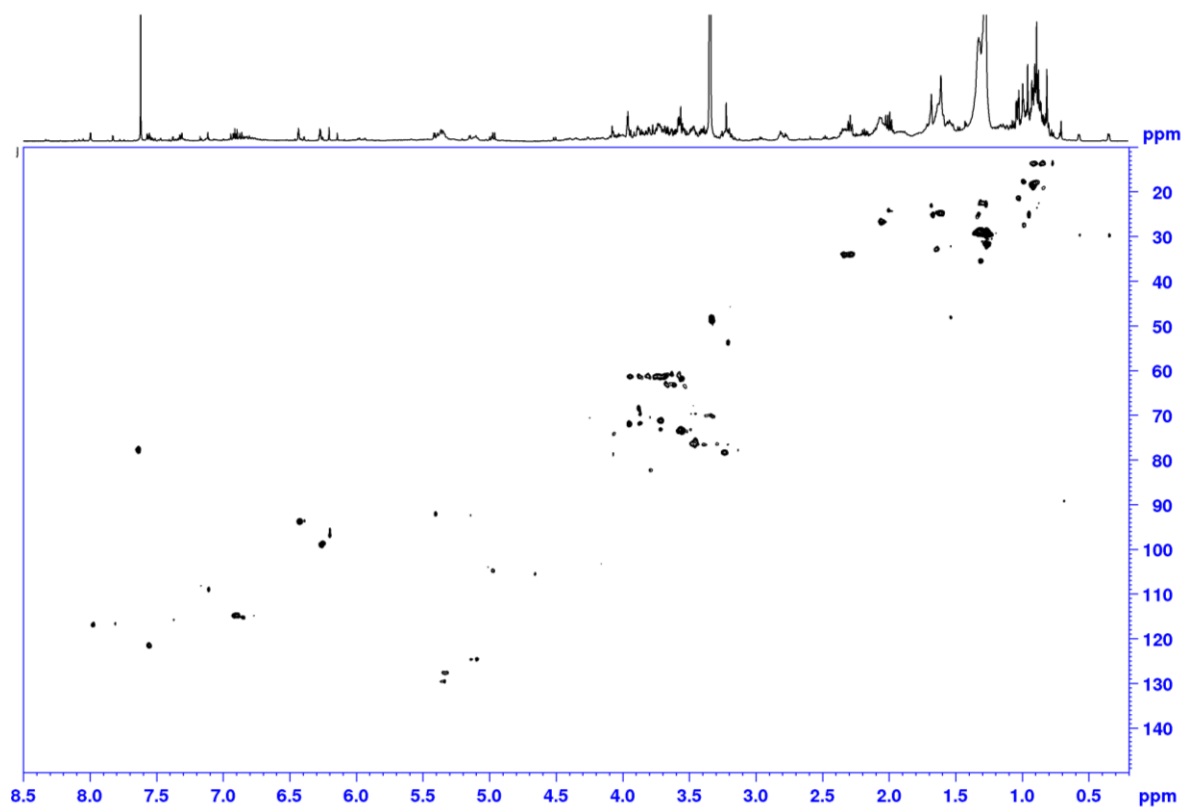


Figure S28: The HSQC NMR spectrum of *E. panonnica* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

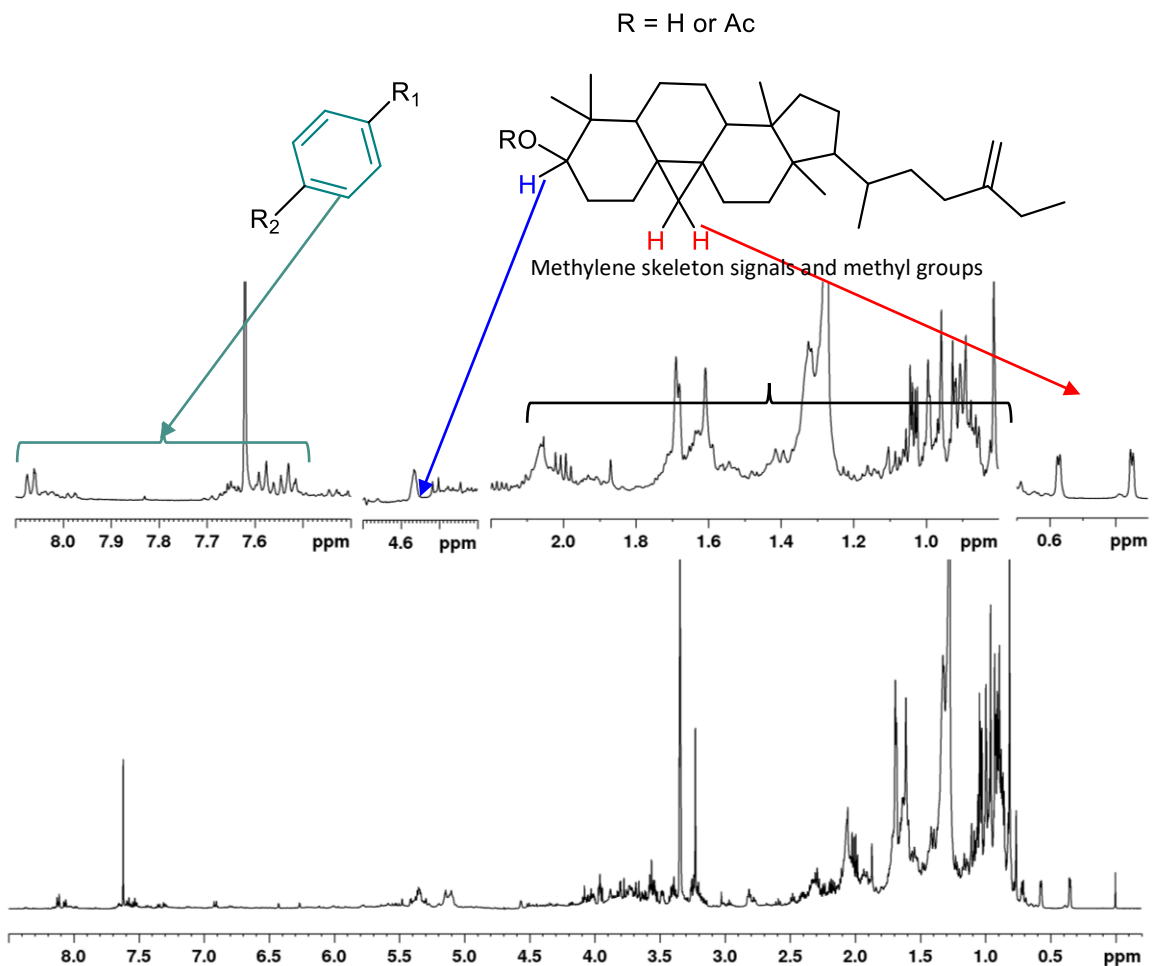


Figure S29: The  $^1\text{H}$  NMR spectrum of *E. cyparisisis* extract extract obtained with MeOD:CDCl<sub>3</sub> (1:1) with assignment of characteristic resonances

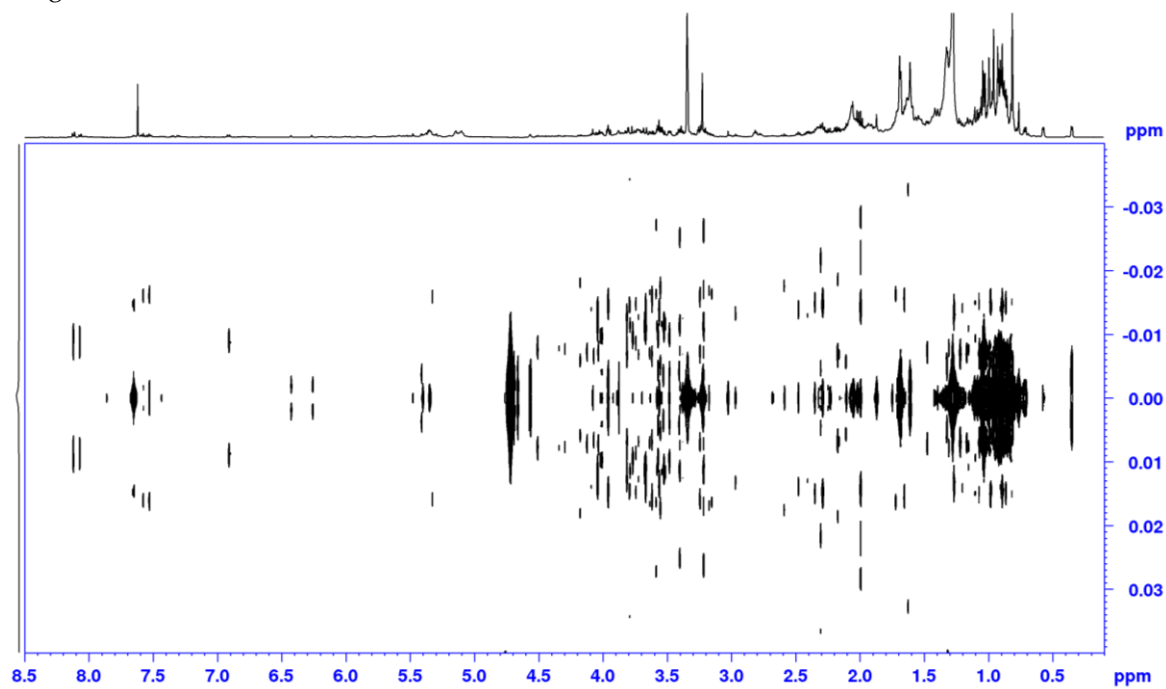


Figure S30: The  $^1\text{H}$ , $^1\text{H}$ -J-resolved NMR spectrum of *E. cyparisisis* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

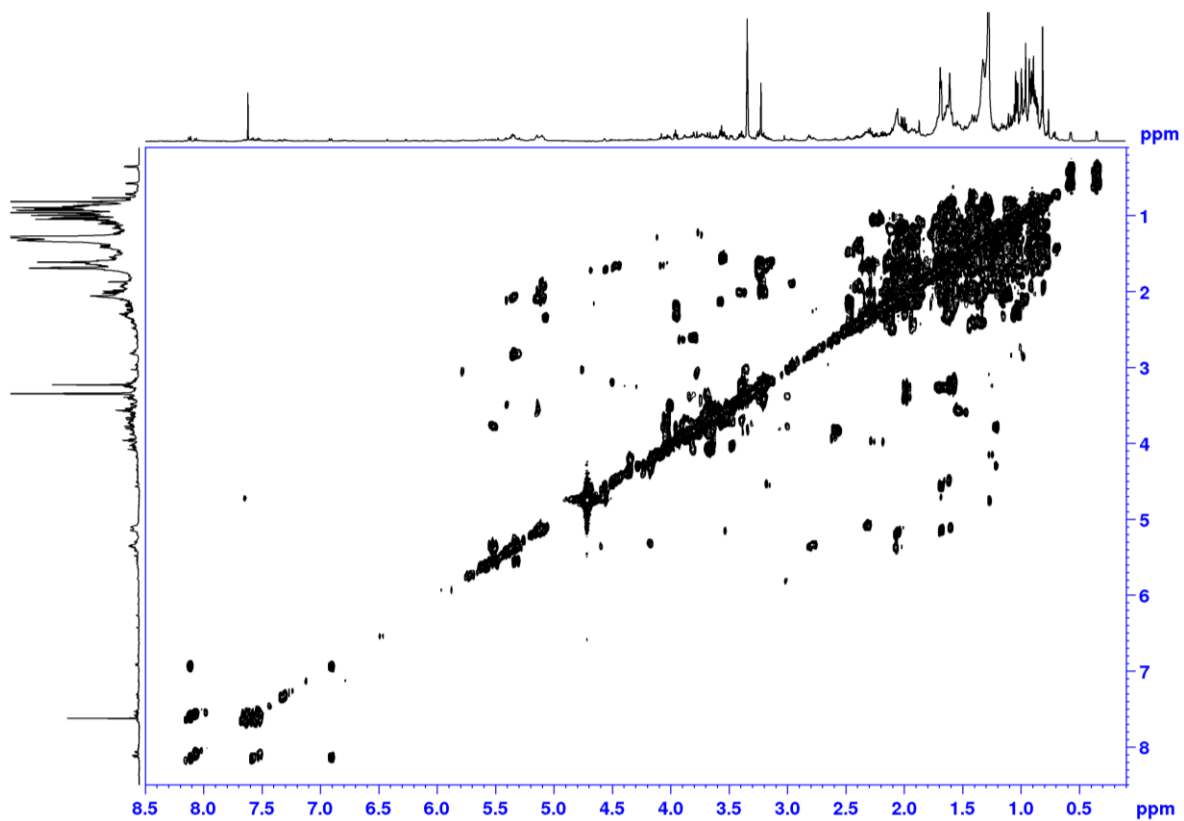


Figure S31: The COSY NMR spectrum of *E. cyparisisas* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

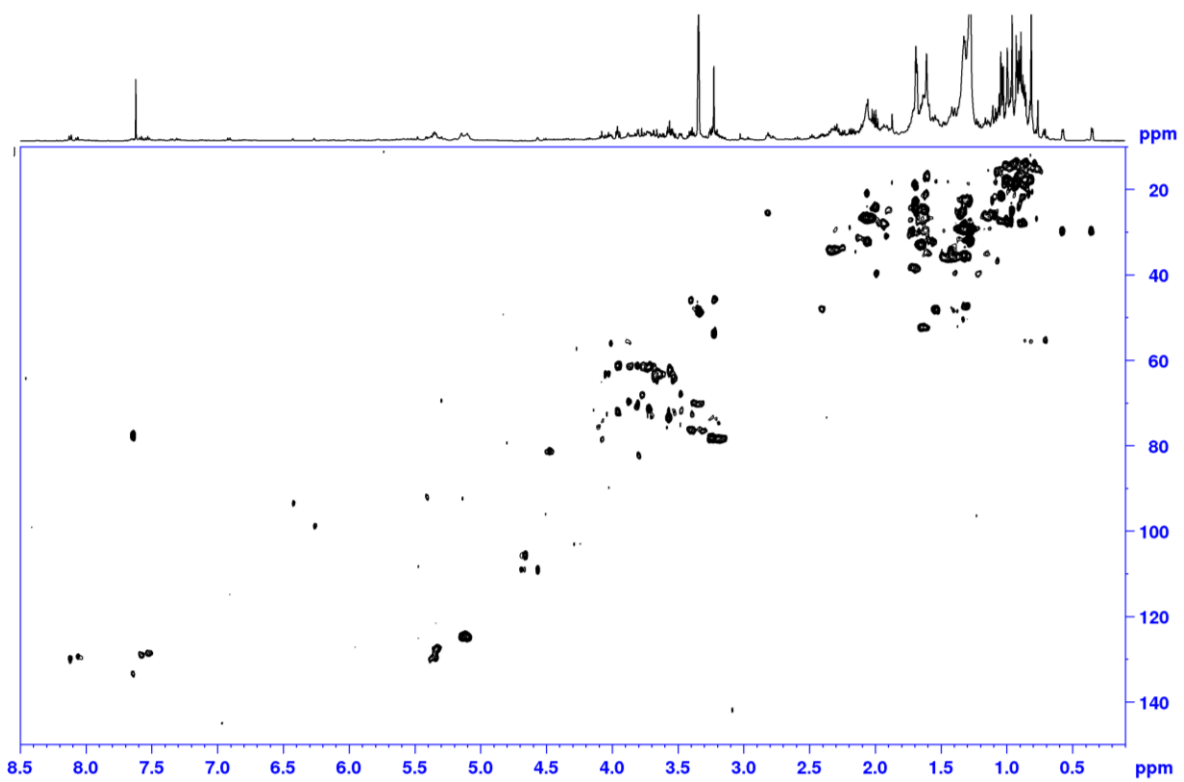


Figure S32: The HSQC NMR spectrum of *E. cyparisisas* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

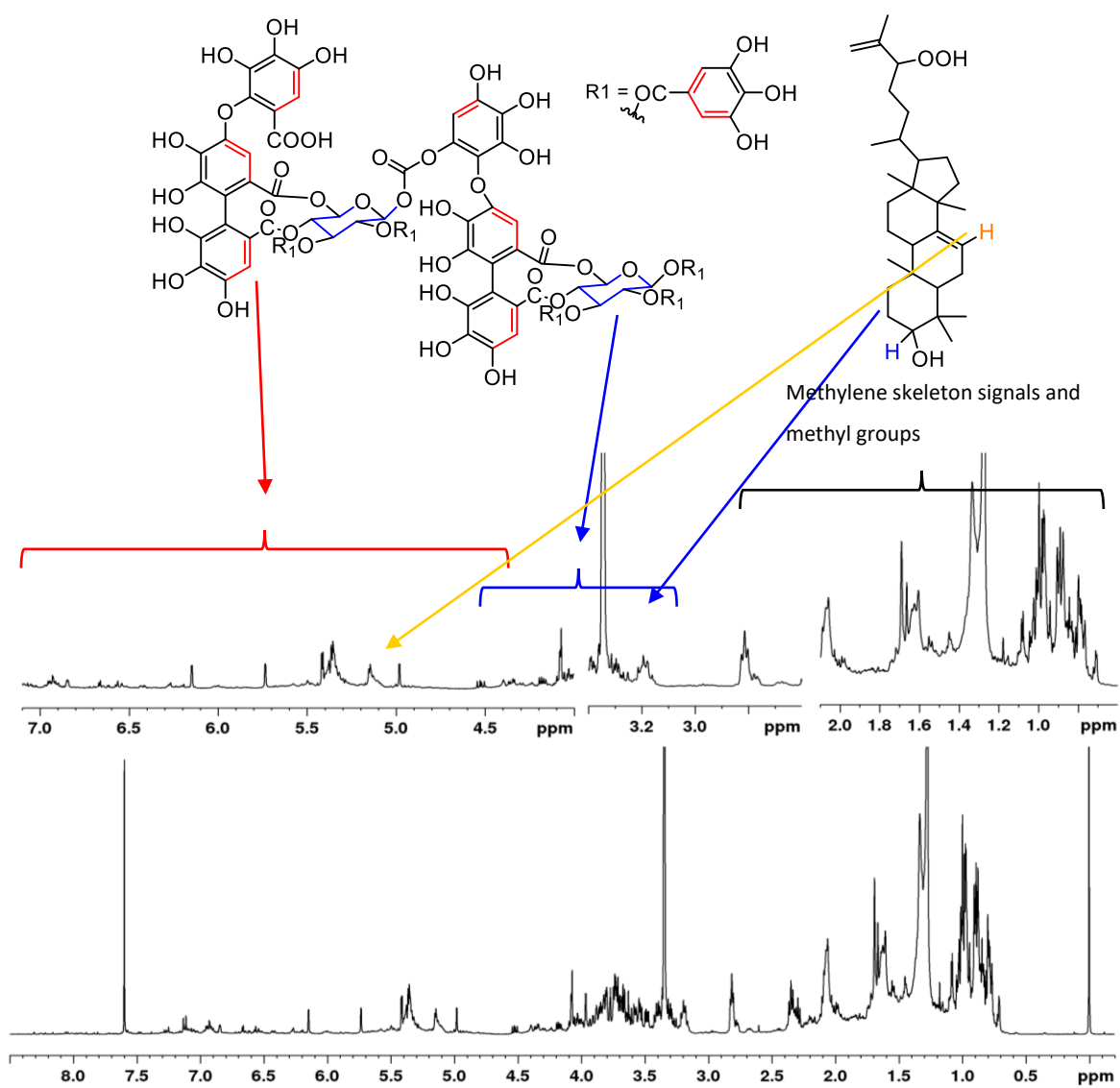


Figure S33: The  $^1\text{H}$  NMR spectrum of *E. maculata* extract extract obtained with MeOD: $\text{CDCl}_3$  (1:1) with assignment of characteristic resonances

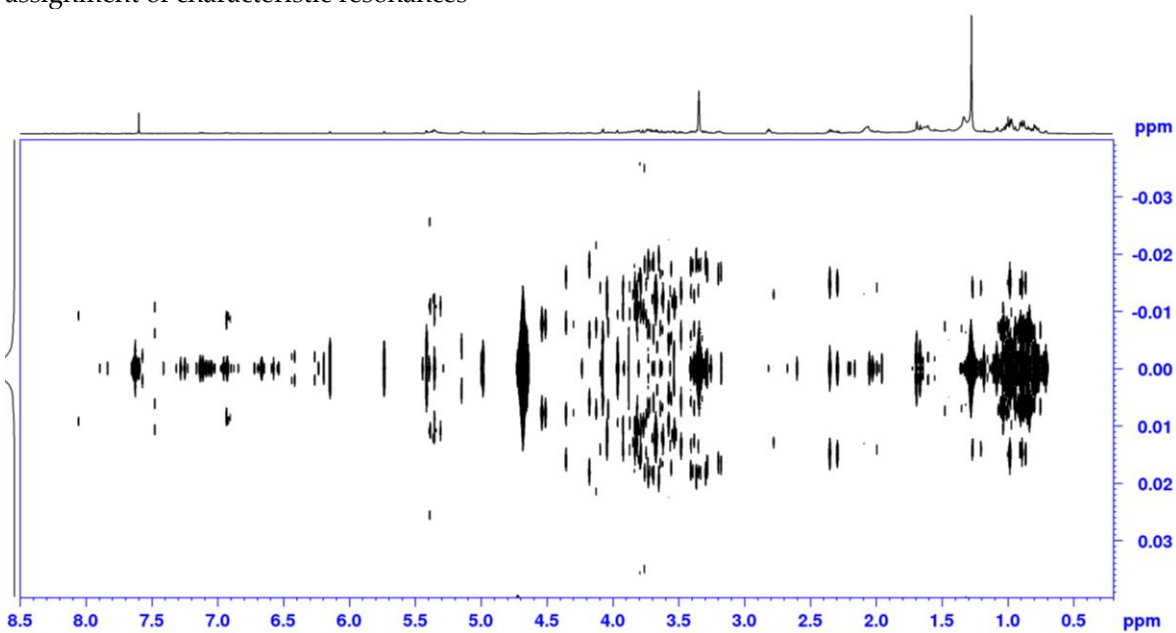


Figure S34: The  $^1\text{H}$ ,  $^1\text{H}$ -J-resolved NMR spectrum of *E. maculata* extract obtained with MeOD: $\text{CDCl}_3$  (1:1)



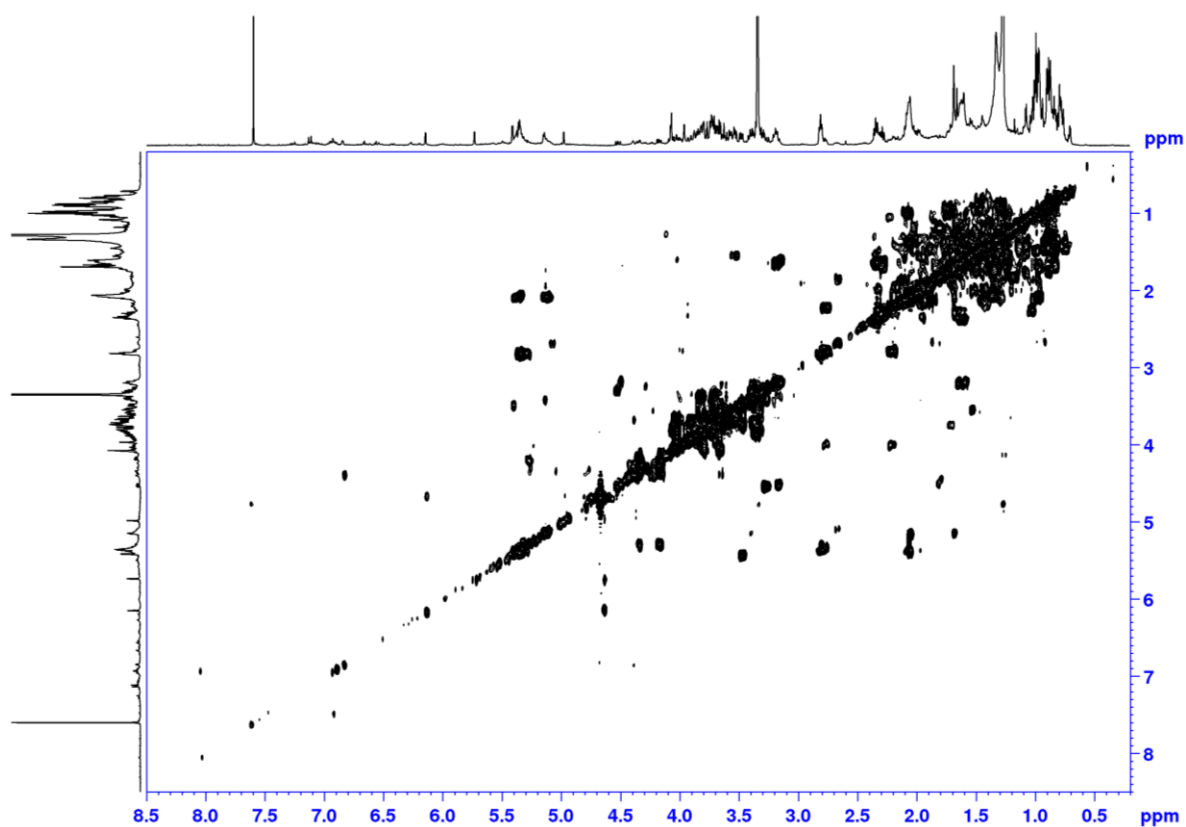


Figure S35: The COSY NMR spectrum of *E. maculata* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)

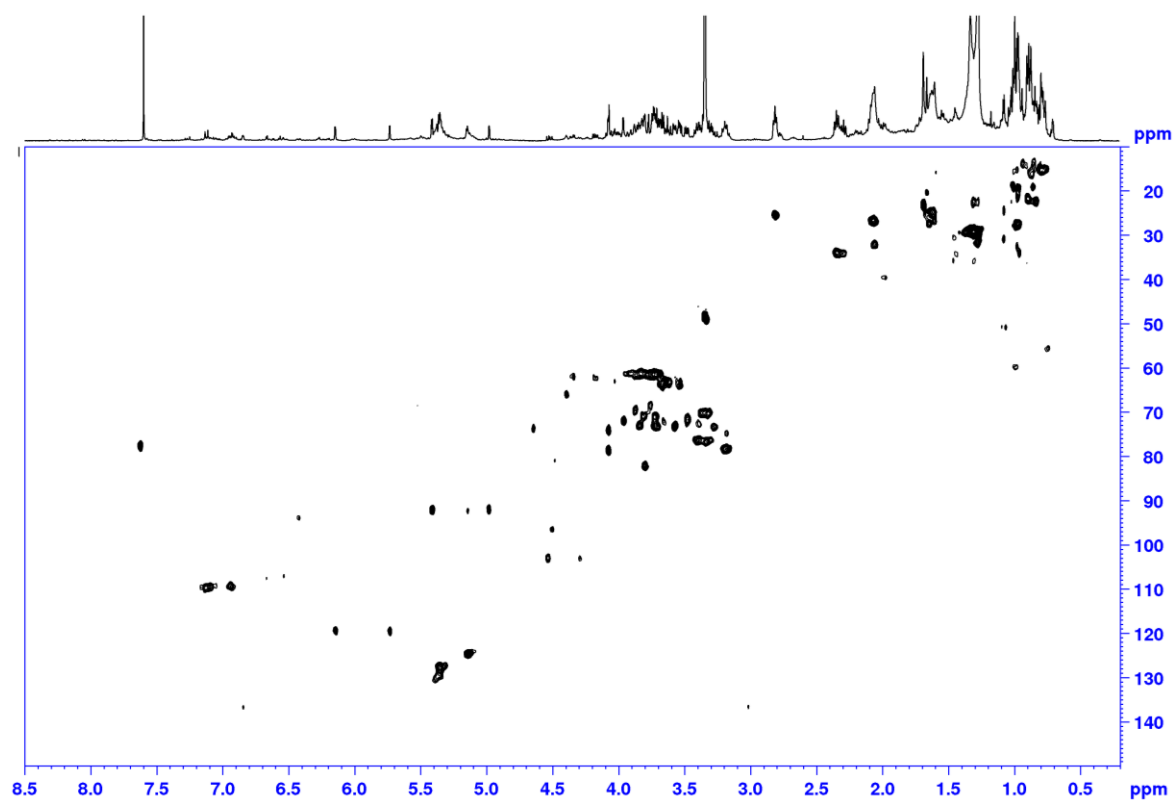


Figure S36: The HSQC NMR spectrum of *E. maculata* extract obtained with MeOD:CDCl<sub>3</sub> (1:1)