Design, synthesis and *in vitro* studies of 3-amidocoumarins as novel antibiofilm agents

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1. Inhibitor Design

(i) Chemical structures of amidocoumarins **S1-S6**

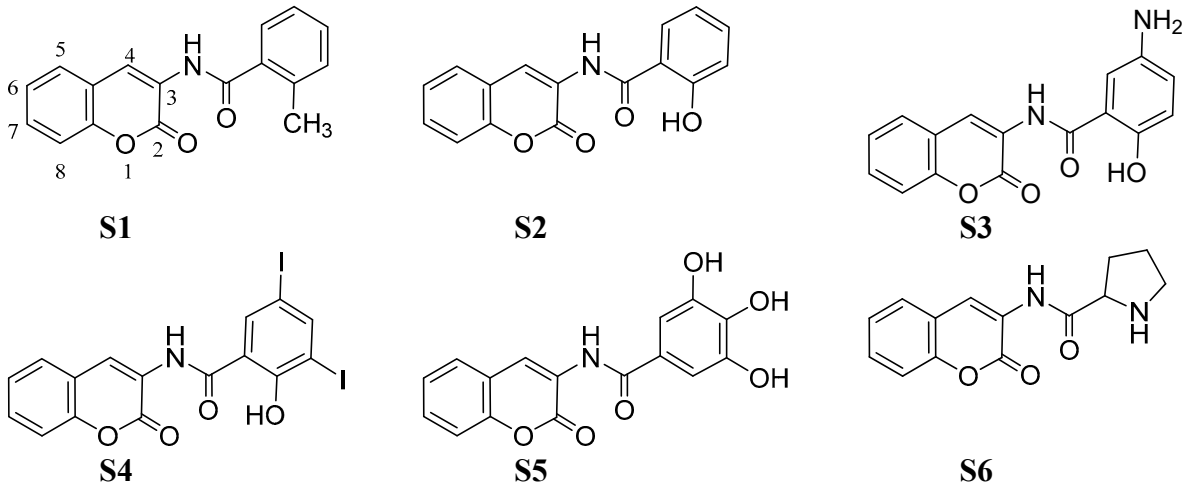
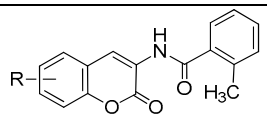


Figure S1. Chemical structures of active compounds

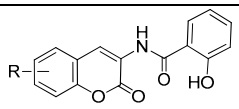
(ii) Taking **S1-S6** as parent molecule, library of 48 compounds was constructed by introducing 6,8-dichloro, 6-bromo, 6-chloro, 6-nitro, 8-methoxy, 6-methyl and 7-hydroxy group into the aromatic ring of coumarin moiety. The parent compounds and designed compounds were docked on the crystal structure of LasR (PDB code: 2uvo). The binding energies (in descending order) are depicted in **Tables S1-S6.**

Table S1. Binding energies (kcal/mol) of designed ligands S1a-h



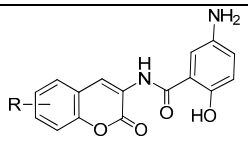
ligand	-R	Binding Energy	Run
S1	H	-10.46	2
S1b	**6-bromo**	**-11.25**	**10**
S1a	6,8-dichloro	-11.13	3
S1c	6-chloro	-11.00	4
S1f	6-methyl	-10.92	6
S1h	6-methoxy	-10.92	5
S1d	6-nitro	-10.72	6
S1g	7-hydroxy	-10.45	4
S1e	8-methoxy	-10.43	6

Table S2. Binding energies (kcal/mol) of designed ligands S2a-h



ligand	-R	Binding Energy	Run
S2	H	-10.89	10
S2b	**6-bromo**	**-11.66**	**7**
S2h	6-methoxy	-11.65	8
S2a	6,8-dichloro	-11.47	4
S2f	6-methyl	-11.23	3
S2c	6-chloro	-11.19	2
S2d	6-nitro	-10.86	10
S2e	8-methoxy	-10.72	10
S2g	7-hydroxy	-10.37	10

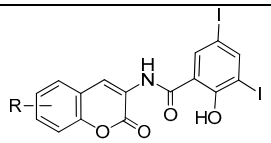
Table S3. Binding energies (kcal/mol) of designed ligands S3a-h



ligand	-R	Binding Energy	Run
S3	H	-10.48	1

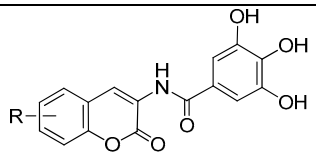
S3b	**6-bromo**	**-11.31**	**7**
S3a	6,8-dichloro	-11.08	2
S3c	6-chloro	-10.99	7
S3f	6-methyl	-10.88	8
S3h	6-methoxy	-10.86	1
S3d	6-nitro	-10.69	3
S3e	8-methoxy	-10.21	8
S3g	7-hydroxy	-10.16	2

Table S4. Binding energies (kcal/mol) of designed ligands S4a-h.



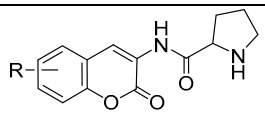
ligand	R	Binding Energy	Run
S4	H	-11.95	1
S4g	7-hydroxy	-10.94	6
S4f	6-methyl	-10.72	2
S4h	6-methoxy	-10.25	2
S4e	8-methoxy	-10.11	8
S4c	6-chloro	-10.05	6
S4d	6-nitro	-9.89	1
S4b	**6-bromo**	**-9.44**	**7**
S4a	6,8-dichloro	-8.25	2

Table S5. Table S5: Binding energies (kcal/mol) of designed ligands S5a-h.



ligand	R	Binding Energy	Run
S5	H	-10.89	6
S5b	**6-bromo**	**-11.69**	**7**
S5a	6,8-dichloro	-11.58	5
S5c	6-chloro	-11.56	1
S5h	6-methoxy	-11.43	9
S5f	6-methyl	-11.34	10
S5d	6-nitro	-11.05	1
S5e	8-methoxy	-10.80	4
S5g	7-hydroxy	-10.37	5

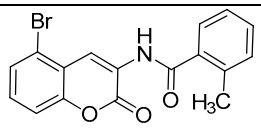
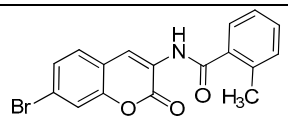
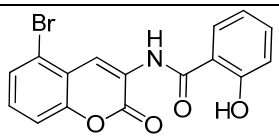
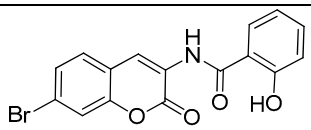
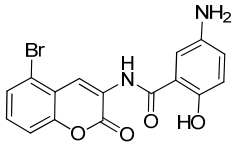
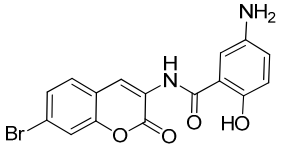
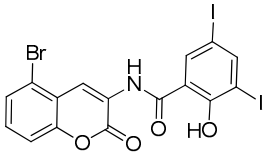
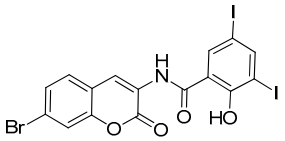
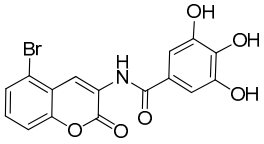
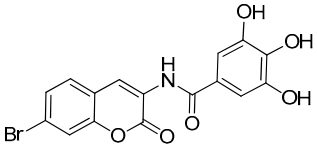
Table S6. Binding energies (kcal/mol) of designed ligands S6a-h.

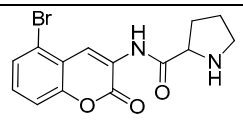
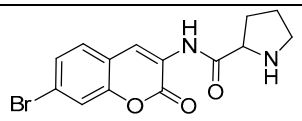


ligand	-R	Binding Energy	Run
S6	H	-10.12	3
S6b	**6-bromo**	**-10.67**	**3**
S6c	6-chloro	-10.38	10
S6f	6-methyl	-10.26	10
S6a	6,8-dichloro	-10.11	6
S6h	6-methoxy	-10.00	10
S6d	6-nitro	-9.76	3
S6g	7-hydroxy	-9.62	4
S6e	8-methoxy	-9.08	1

(iii) As ligands containing 6-bromo substituent had shown minimum binding energy in the all the sets except S2a-h. We further constructed 5-bromo and 7-bromo analogs of compounds S1-S6. All the compounds were docked into the same active site of receptor. The binding energy values are shown in **Table S7**.

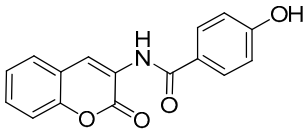
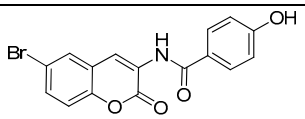
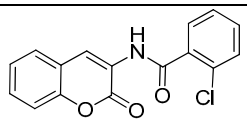
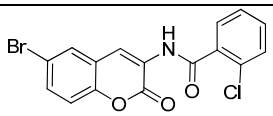
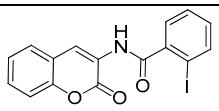
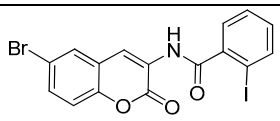
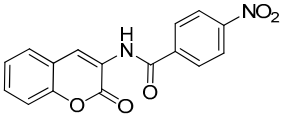
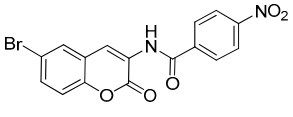
Table S7. Binding energies (kcal/mol) of designed ligands S1i,h to S6i,j.

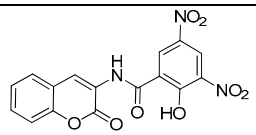
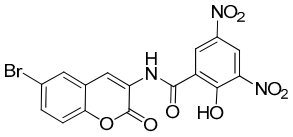
Ligand	Structure	Binding Energy	Run
S1i		-10.94	9
S1j		-11.02	1
S2i		-10.99	2
S2j		-11.59	5
S3i		-10.90	7
S3j		-11.21	5
S4i		-10.12	5
S4j		-10.5	8
S5i		-11.05	10
S5j		-11.75	6

S6i		-10.13	3
S6j		-10.25	9

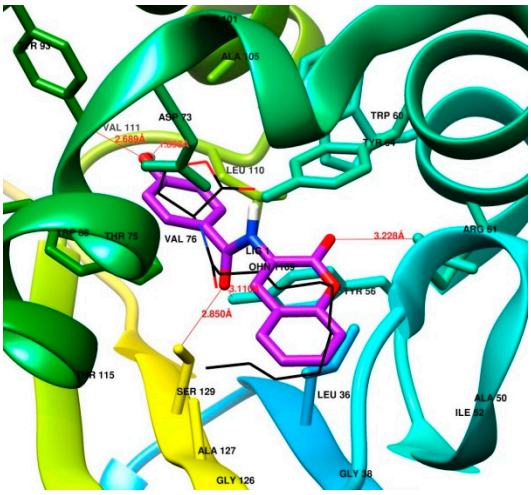
(iv) Binding energies of newly constructed compounds **S7-S11** and their 6-Br analogs **S7a-S11a** (Table S8).

Table S8: Binding energies (kcal/mol) of designed ligands S7-S11 and S7a-S11a

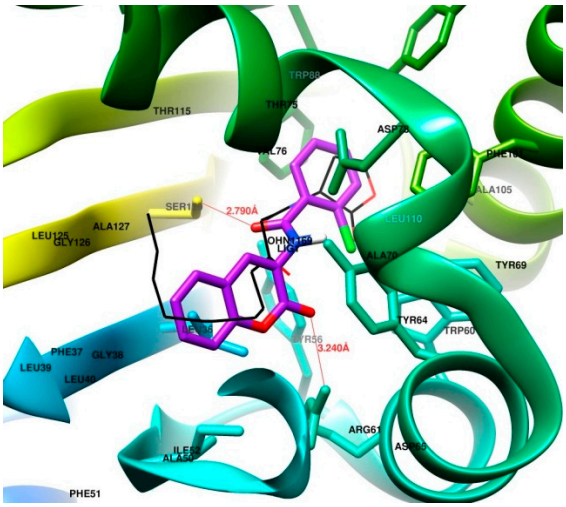
ligand	Structure	Binding Energy	Run
S7		-11.36	8
S7a		-11.47	4
S8		-10.51	2
S8a		-11.45	5
S9		-10.32	6
S9a		-11.65	9
S10		-9.84	2
S10a		-10.67	3

S11		-10.46	3
S11a		-10.31	5

2. Molecular docked complex of 3-benzamidocoumarin **4a-4o** with LasR (2uv0).



4a



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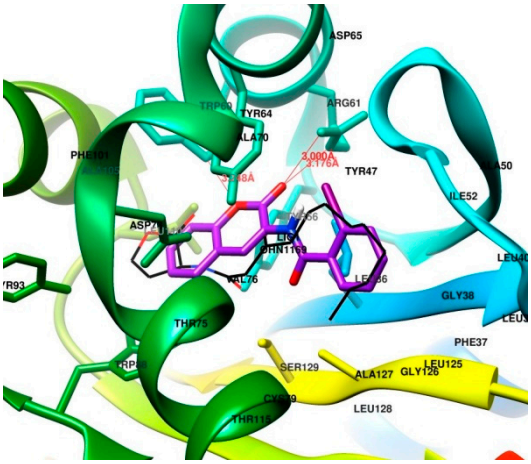

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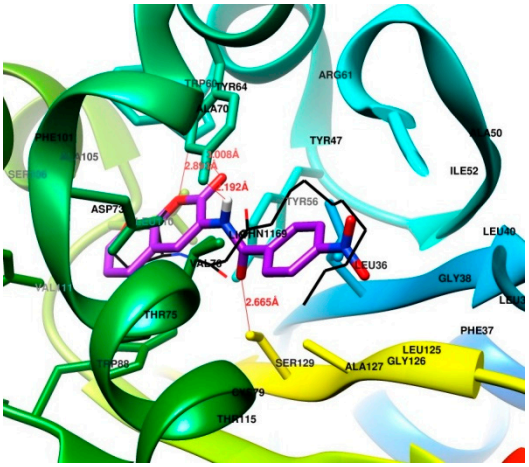

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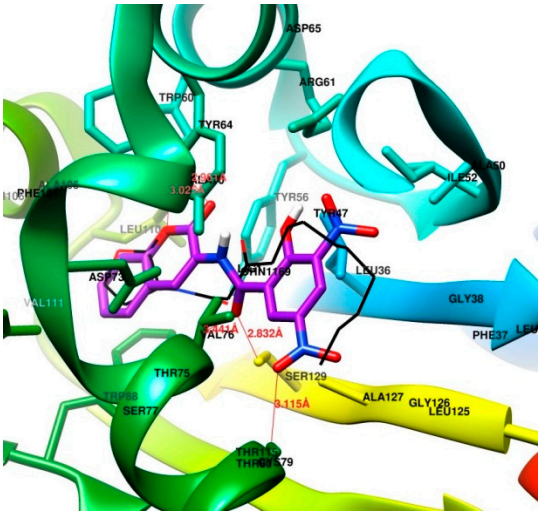

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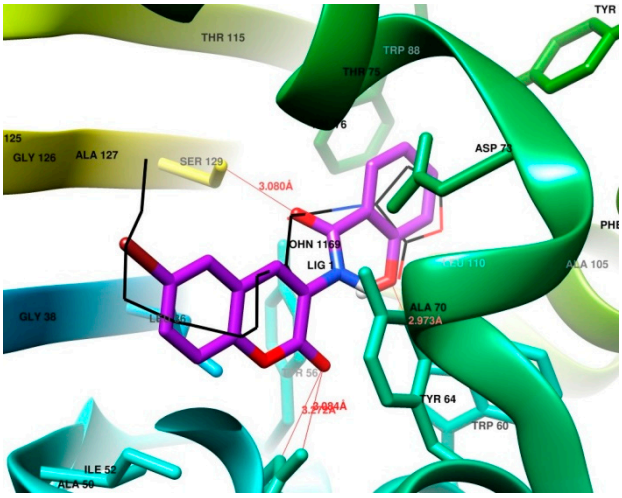

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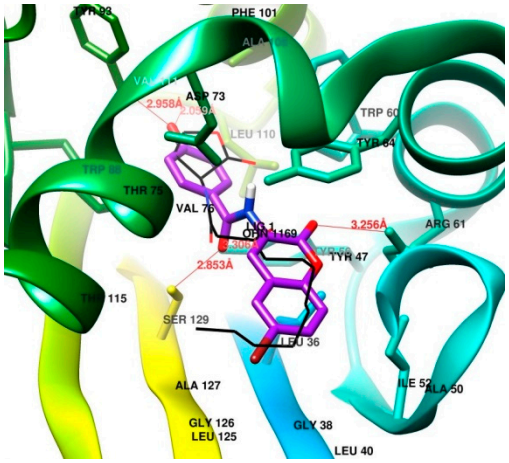

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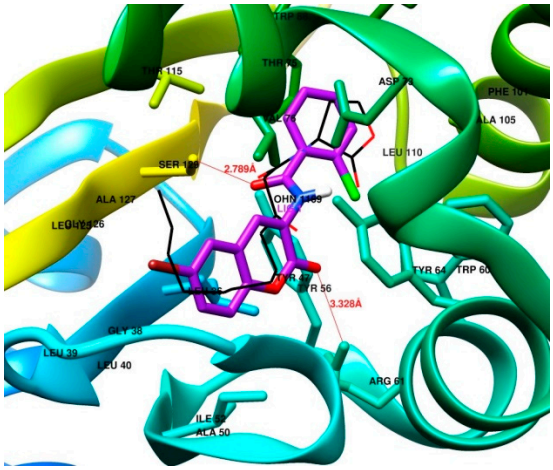
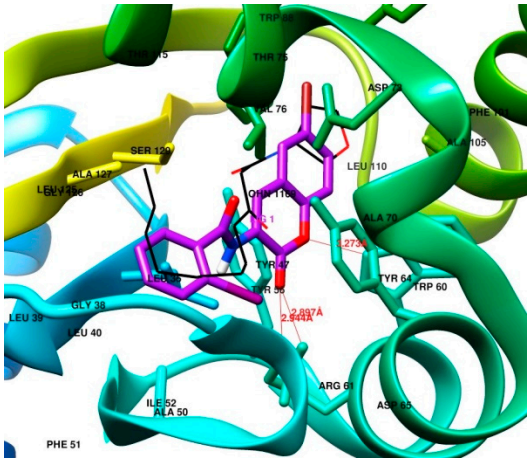


4g



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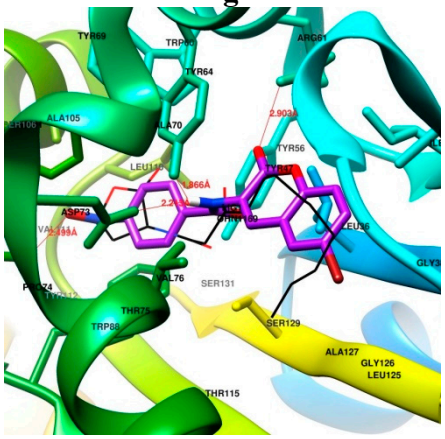

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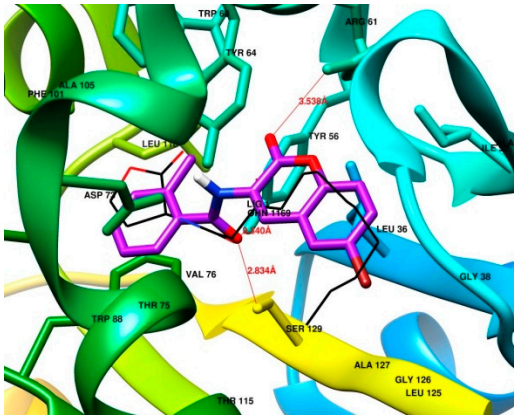

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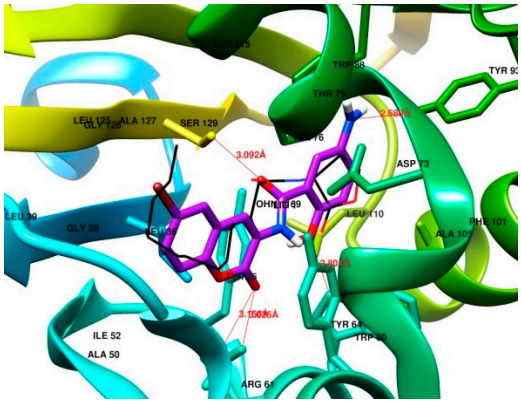

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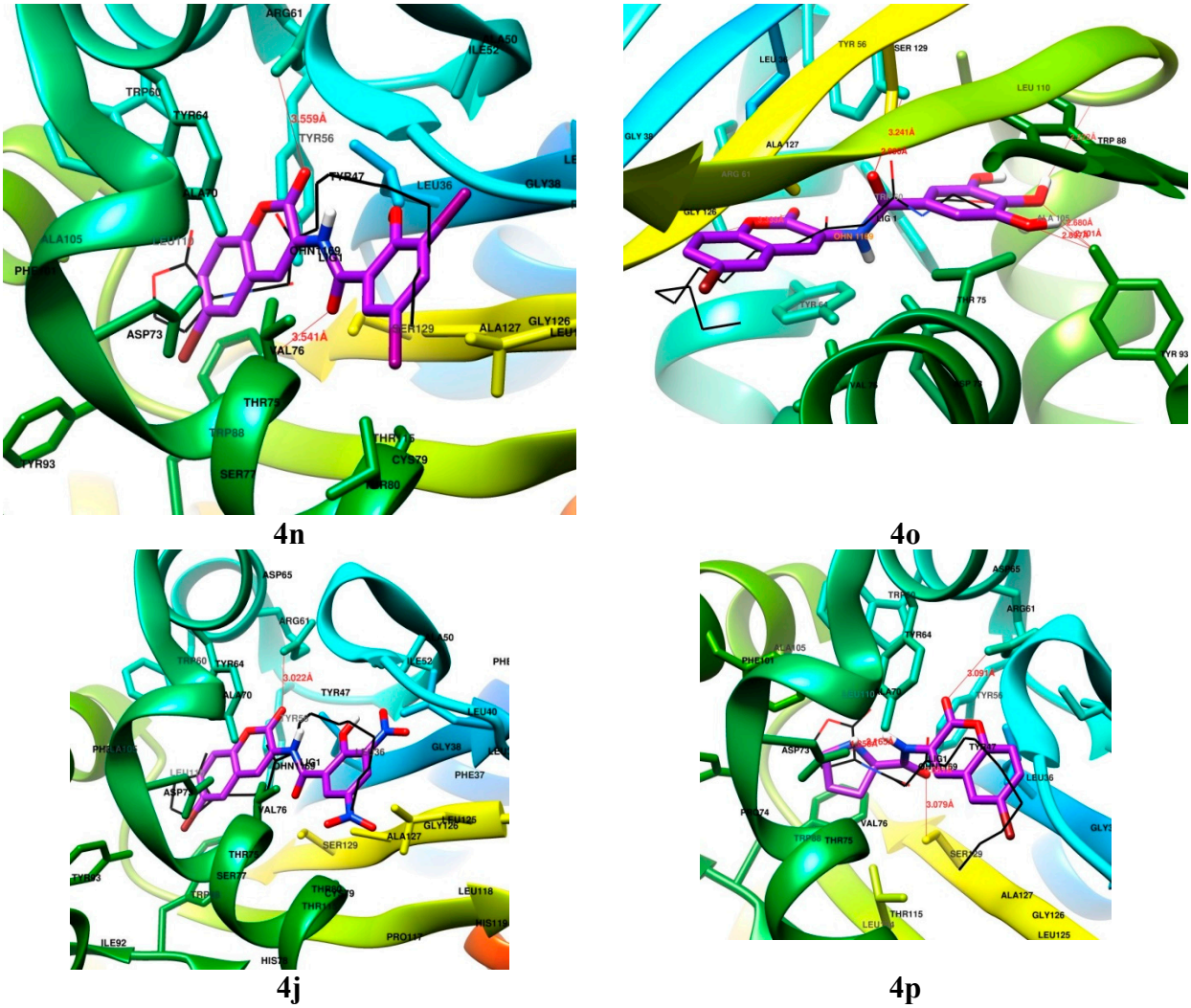

Figure S2: Molecular docked complex of compounds 4a-4o with LasR (2uv0).

3. Spectral data of compound **4a**

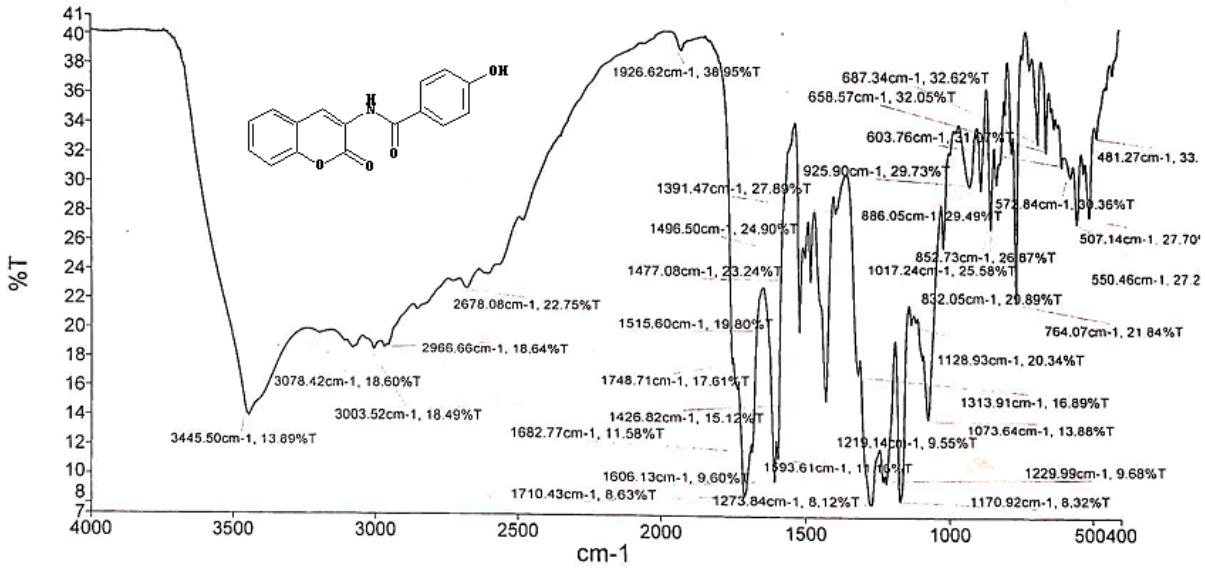

Figure S3: IR spectrum of 4a.

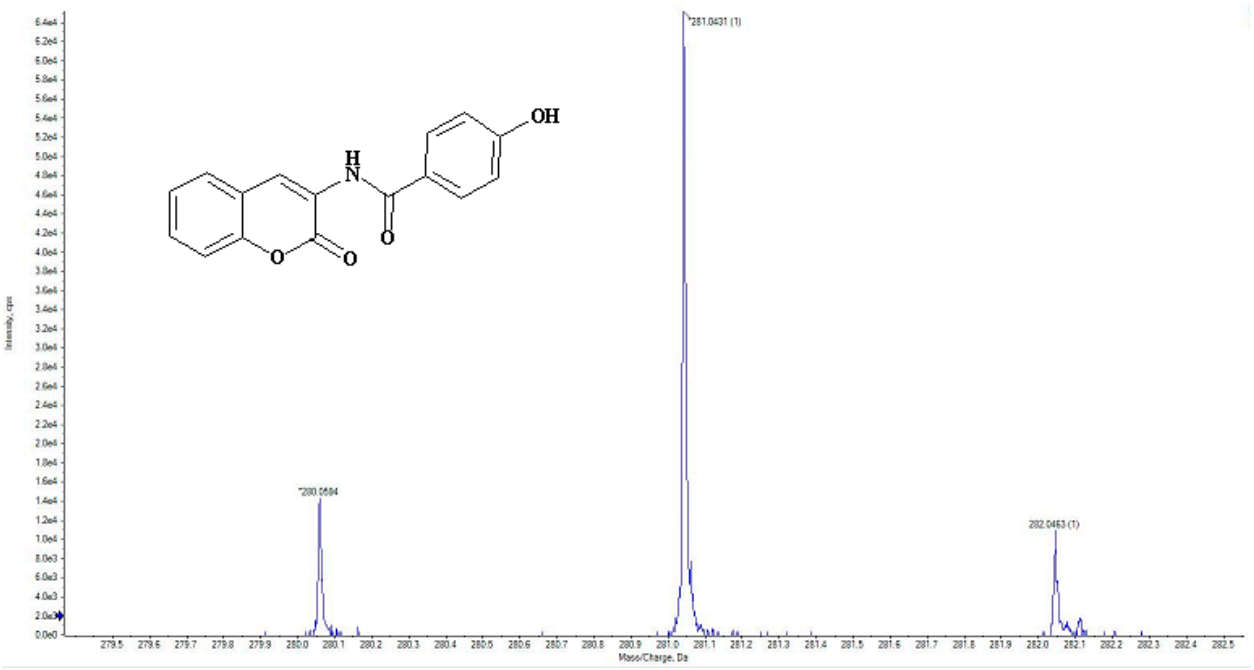

Figure S4: HRMS spectra of 4a.

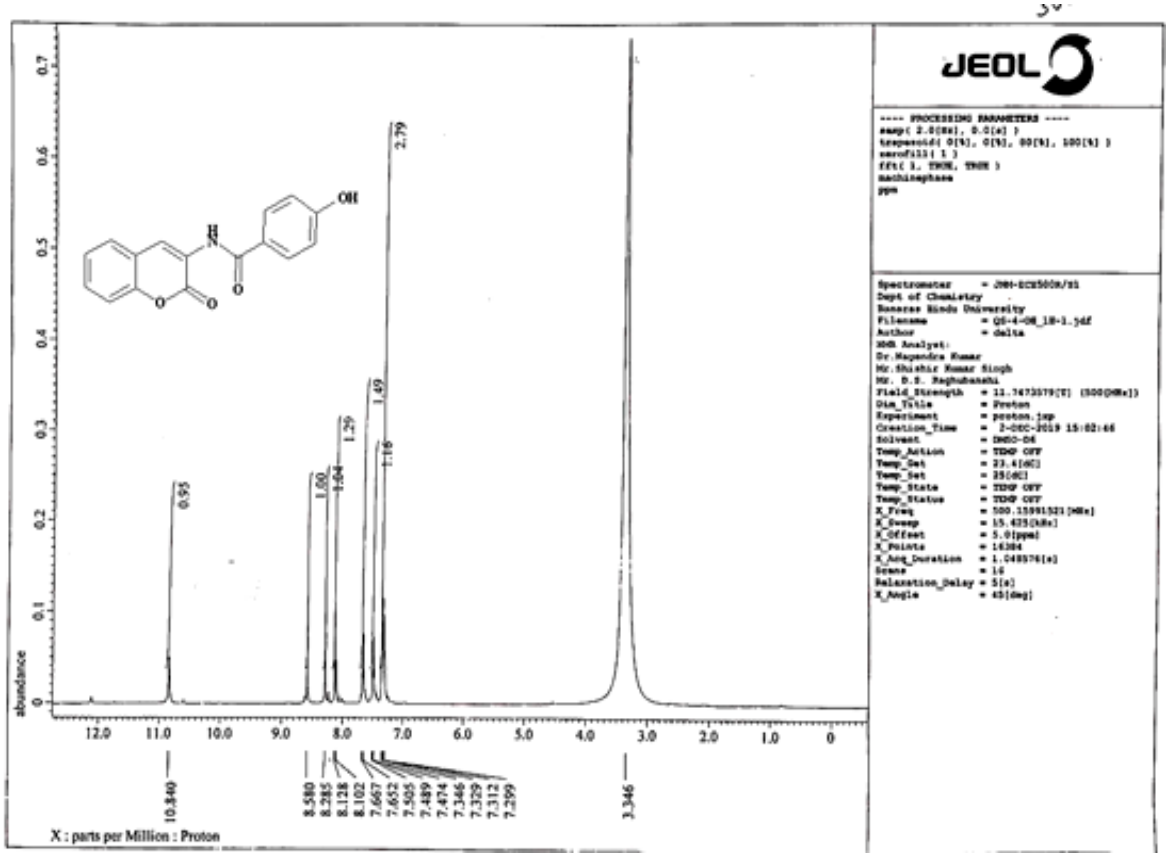

Figure S5: ^{1}H NMR of 4a.

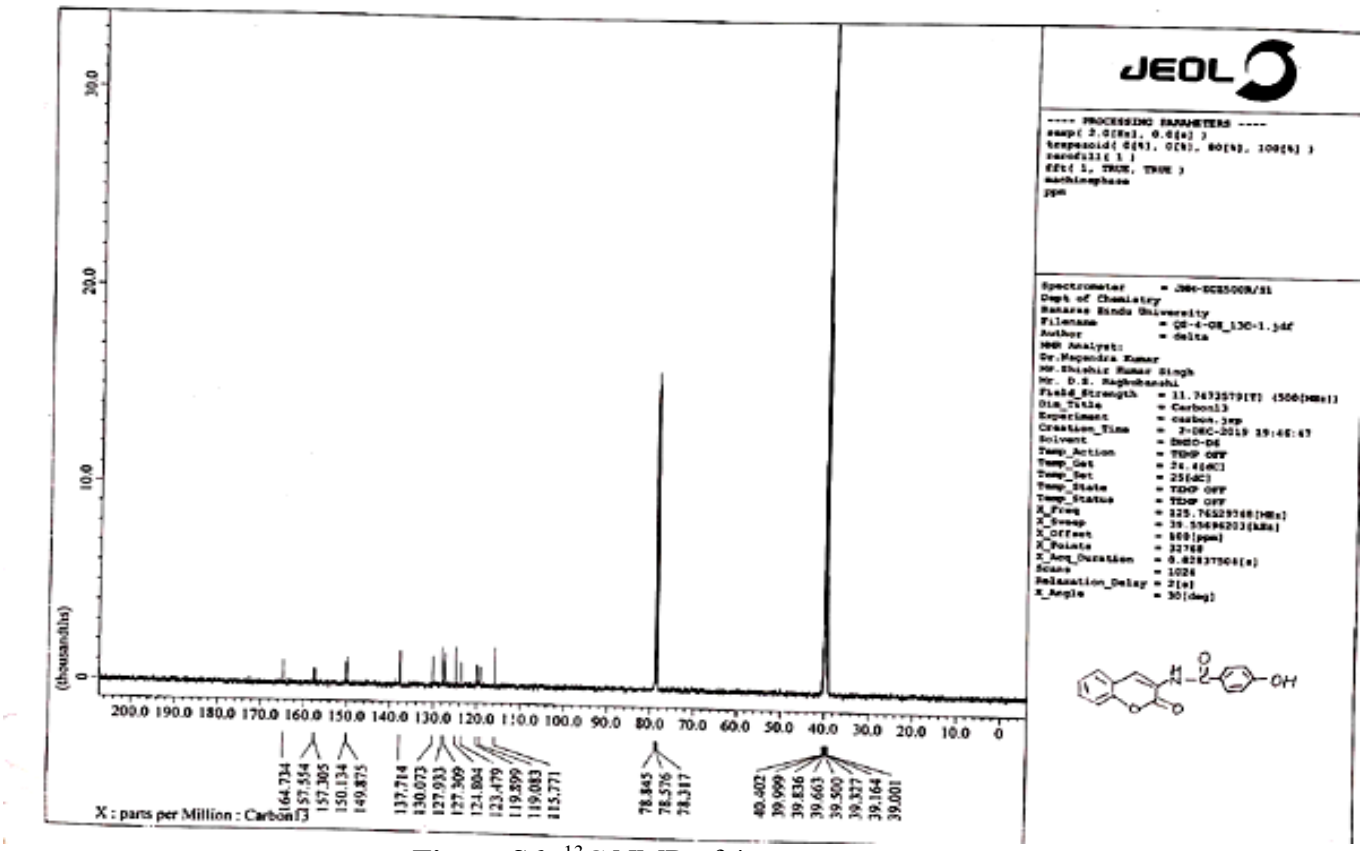

Figure S6: ^{13}C NMR of 4a.