Supporting Information (SI)

Azouracil and its Cu(II) catalyzed cyclization to anticancer active triazole derivative: symmetrical and asymmetrical reductive cleavage, DNA interaction and molecular docking studies

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1. General method of UV-Vis and fluorescence titration

Path length of the cells used for absorption and emission studies is 1 cm. Fluorescence measurements are performed using 5 nm x 5 nm slit width.

Genomic DNA isolation of *E. coli*

Genomic DNA of *E. coli* is isolated by the method of Chen and Kuo (1993). *E.coli* strain is grown in nutrient broth medium for 24 h at 32ºC. Two ml of culture broth is taken into micro centrifuge tube and centrifuge for 10 min at 12,000 rpm. The cell pellet is resuspended and lysed in 200 µl of lysis buffer (40 mM Tris-acetate pH 7.8, 20 mM sodium-acetate, 1 mM EDTA, 1%

SDS) using vigorous pipetting. After that, 66µl of 5M NaCl solution is added and centrifuged for 10 min at 12,000 rpm at 4ºC to remove protein and cell debris. The clear supernatant is transferred to new vial and an equal volume of chloroform is added and the vial was gently inverted at least 50 times when a milky solution was formed. Following centrifugating again in 12,000 rpm for 3 min., the extracted supernatant is transferred to another vial and the DNA precipitated with addition of 100% EtOH, washed twice with 70% EtOH, dried and redissolve in 50µl 1× TE buffer. RNA is removed by adding RNase in the lysis step for 30 min at 37°C.

Table S1 Crystallographic details and refinement data for L11, L11P and A3

Parameters	**Crystal data for L11**	**Crystal data for L11P**	**Crystal data for A3**
Empirical Formula	C_{13} H_{14} N_6 O_8	C_{13} H_{11} N_5 O_4	C_6 N_5 O_3
Moiety Formula	C_{13} H_{14} N_5 O_4, N O_3, O	C_{13} H_{11} N_5 O_4	C_6 N_5 O_2, O
Molecular weight	382.30	301.27	190.11
Crystal color	orange	yellow	Red
Crystal description	prism	plate	Block
Crystal system	orthorhombic	Monoclinic	orthorhombic
Cell parameter a α b β c γ	 21.1979(16) 90 11.9622(8) 90 6.5494(5) 90	 7.2383(6) 90 26.140(2) 98.316(5) 6.8190(6) 90	 14.061 90 6.429 90 9.700 90
Density	1.529	1.567	1.440
Volume	1660.8(2)	1276.65(18)	876.8
Space group	P n a 21	P 21/c	'P n m a'
Hall group	P 2c -2n	-P 2ybc	P 2ac 2n
Temperature	100(2)	100(2)	296(2)
Z	4	4	4
Absorption coefficient(mm-1)	0.129	0.120	0.121
F000	792	624	380
F000′	-	624.30	-
h, k, l max	28,15,8	9,34,9	16,7,11
N ref	4144	3164	I > 2\s(I)
Experimental absorbance correction T min	0.928	0.899	-

Experimental absorbance correction T max	0.982	1.000	-
Reflections threshold Expression	I > 2\s(I)	I > 2\s(I)	-
Wavelength	0.71073	0.71073	0.71073
Theta (min)	1.921	2.844	2.551
Theta (max)	28.332	28.281	24.761
Theta (full)	25.242	25.242	24.761

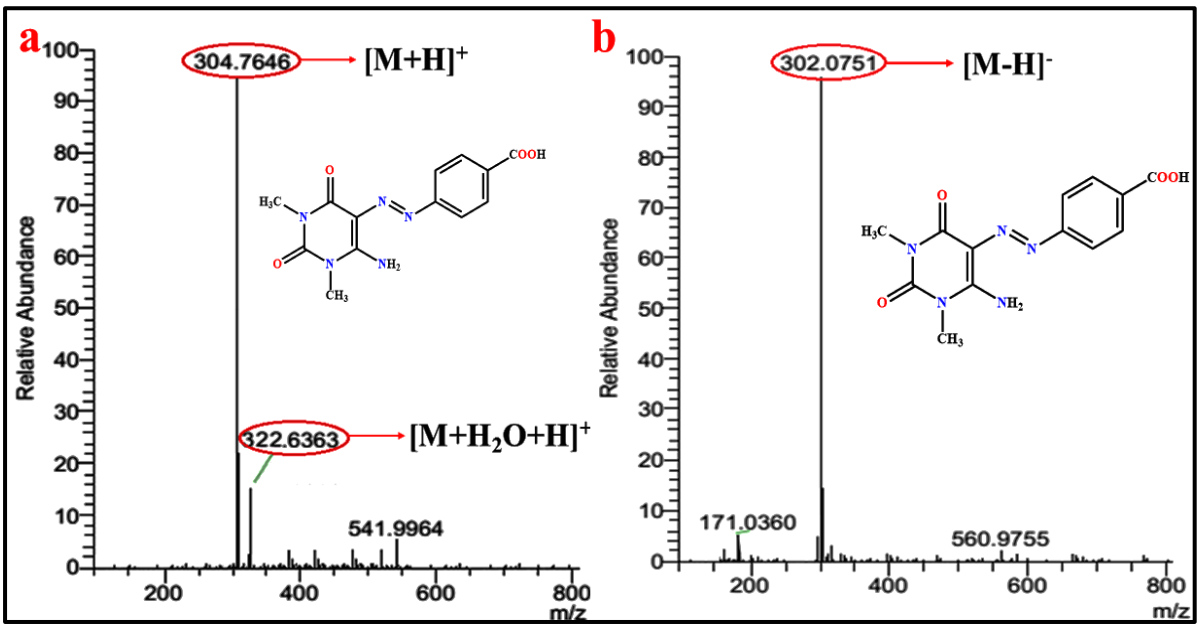

Fig.S1 QTOF-MS ES(+) spectrum of L11 in MeOH(a) and QTOF-MS ES(-) spectrum of L11 in MeOH (b)

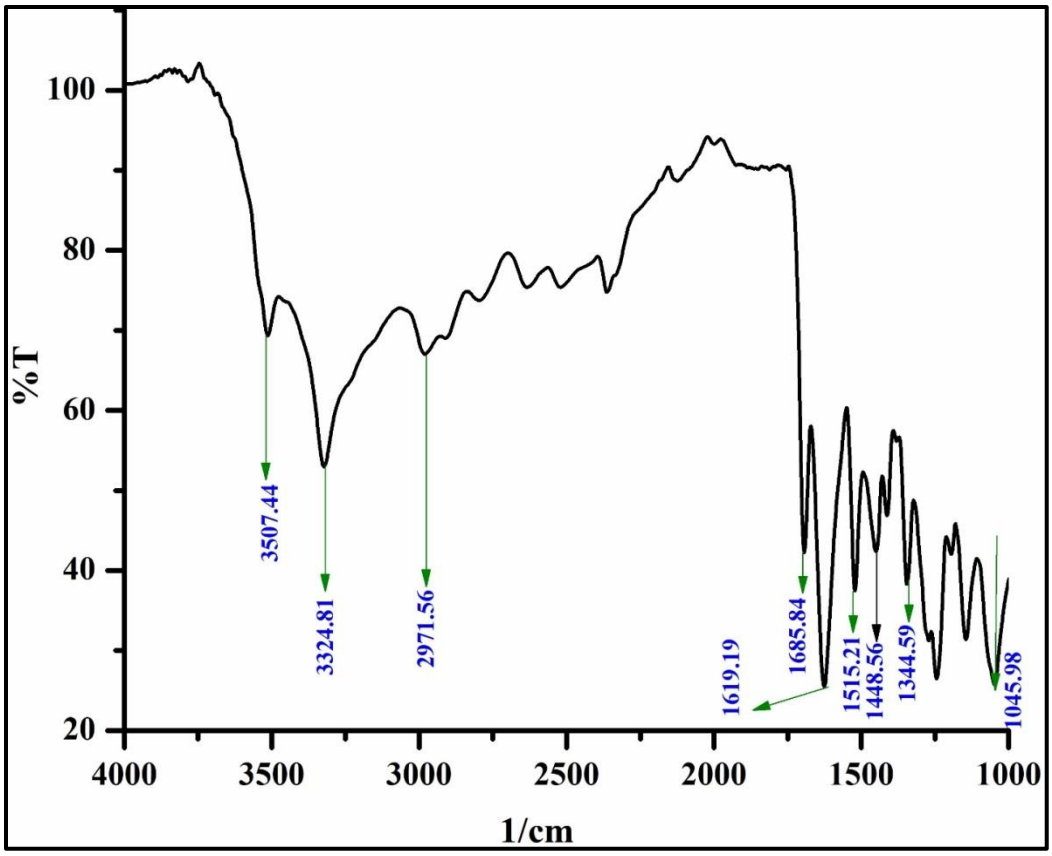

Fig.S2 FTIR spectrum of L11

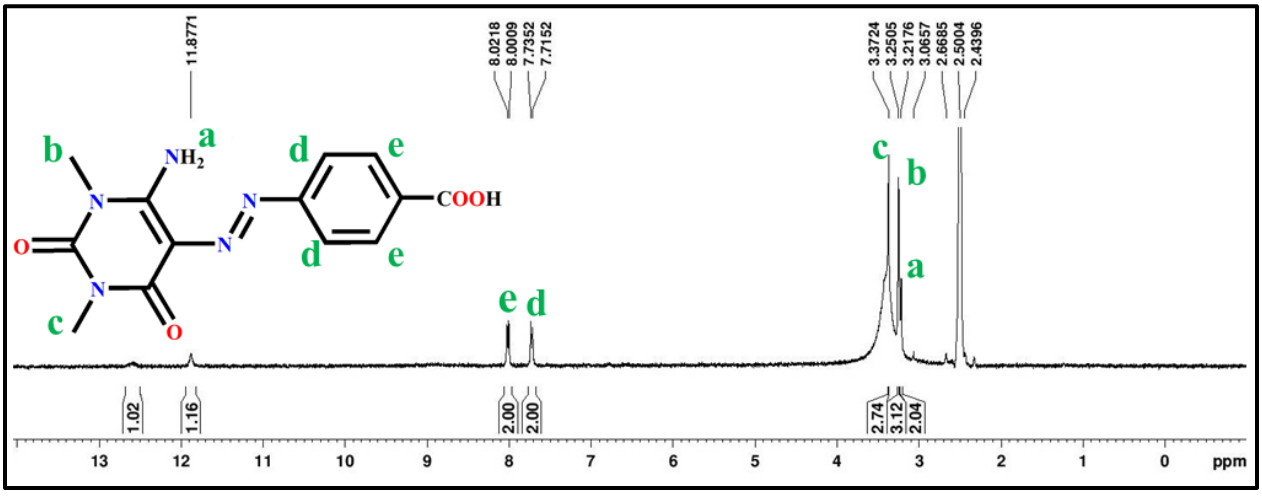

Fig.S3 ^{1}HNMR spectrum of L11 in DMSO -d_6.

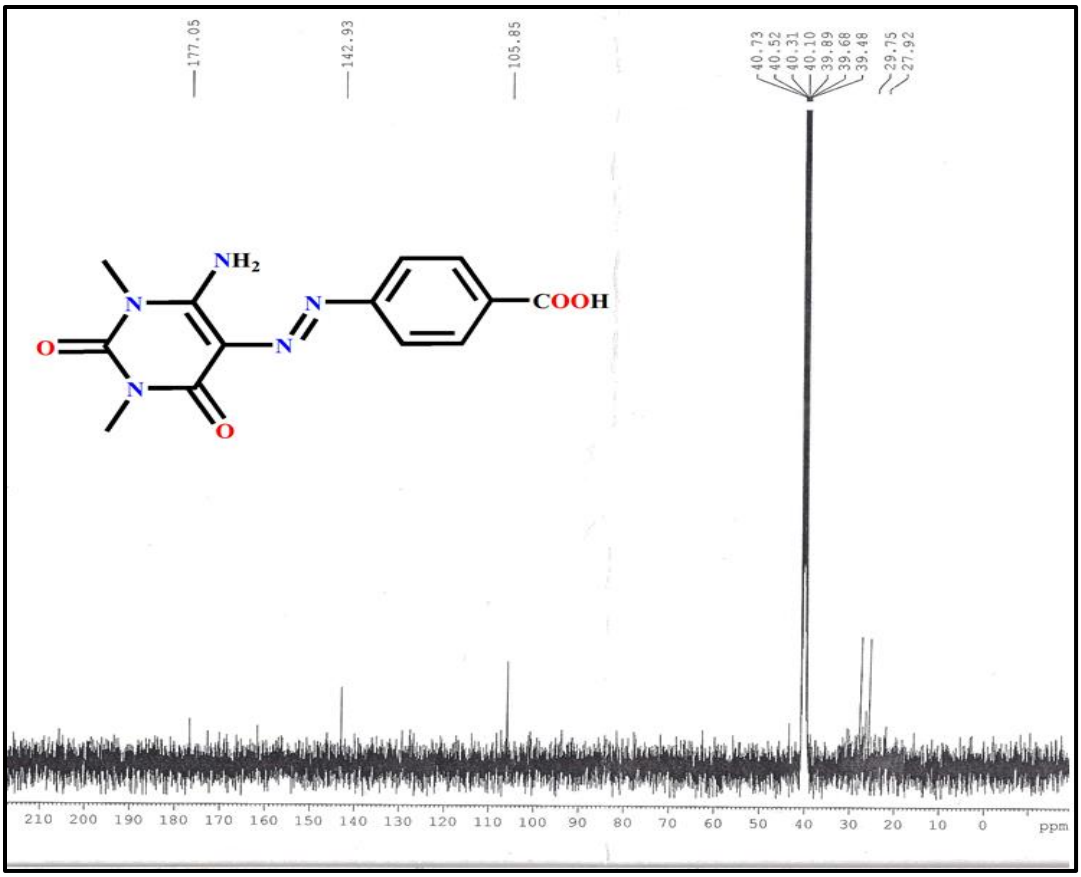

Fig.S4 ^{13} CNMR spectrum of L11 in DMSO -d_6.

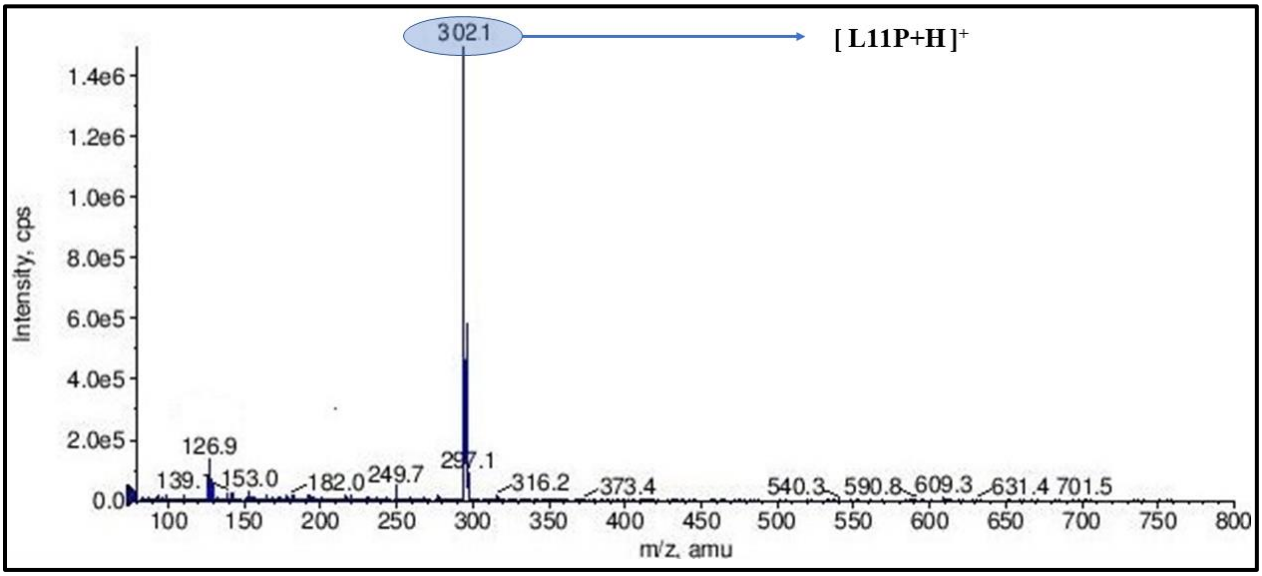

Fig.S5 QTOF-MS spectrum of L11P in MeOH

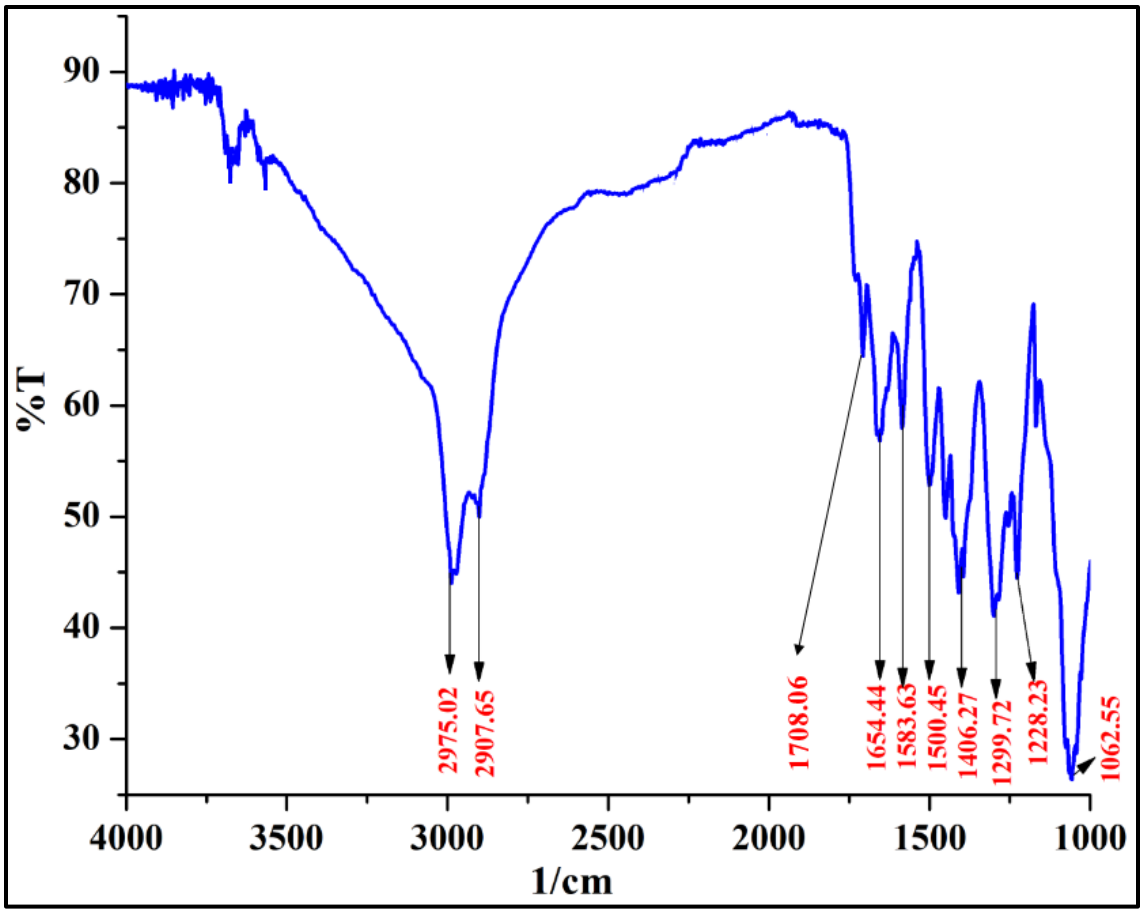

Fig.S6 FTIR spectrum of L11P

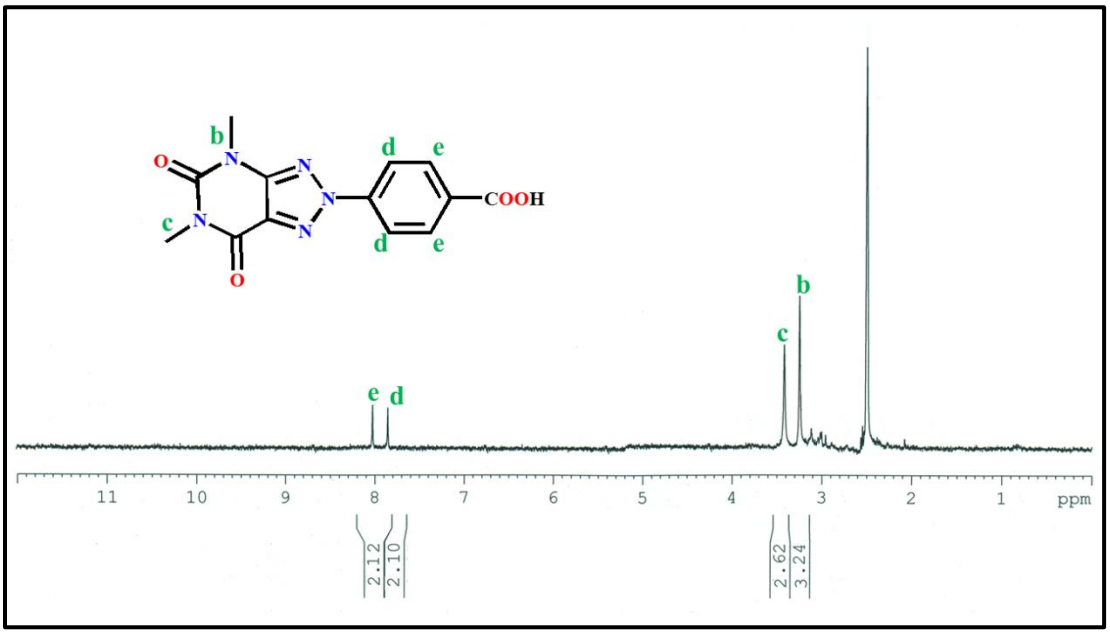

Fig.S7 ^{1}H-NMR spectrum of L11P in DMSO -d_6.

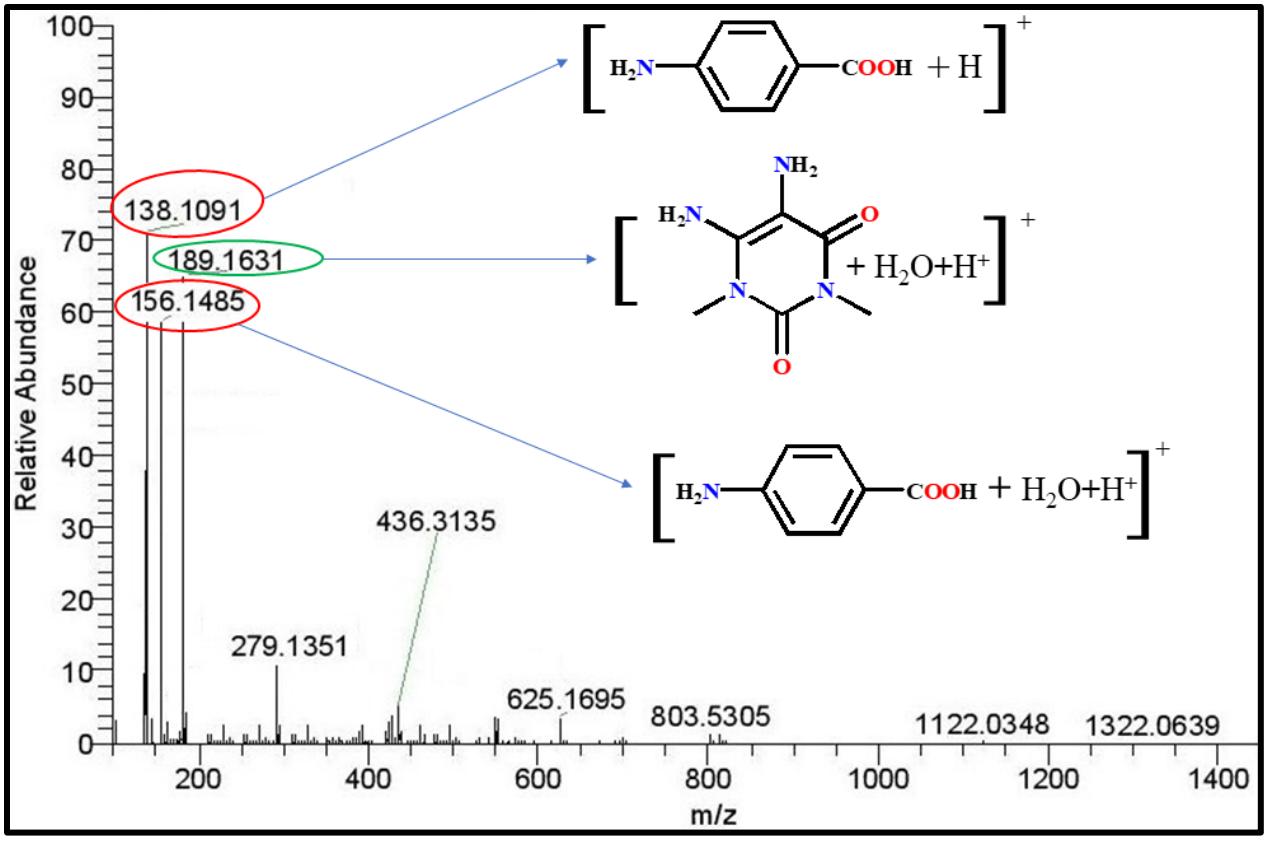

Fig.S8 QTOF-MS spectrum of A1 and A2

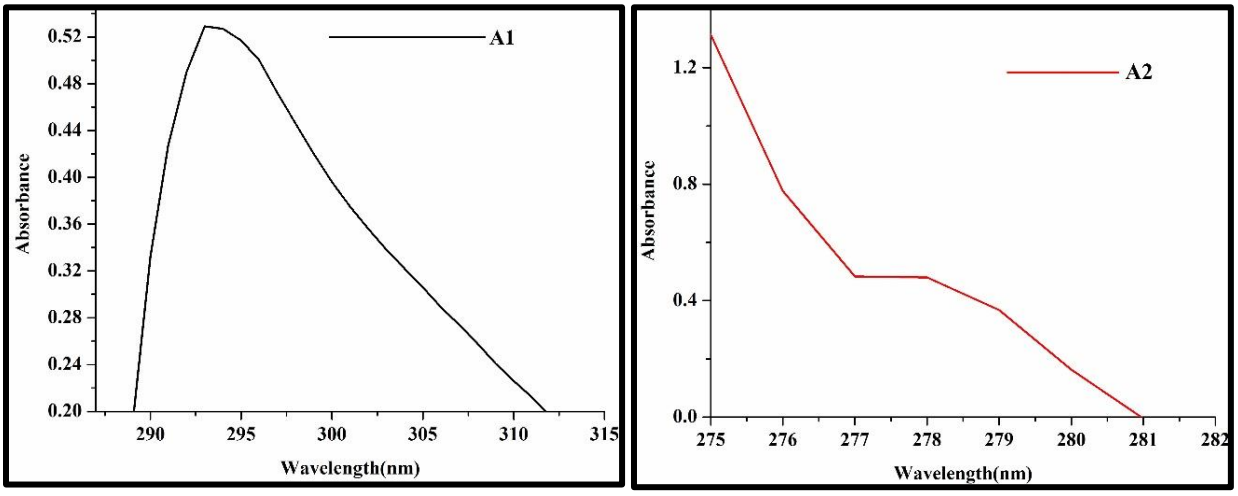

Fig.S9 UV-Vis spectrum of A1 (left) and A2 (right)

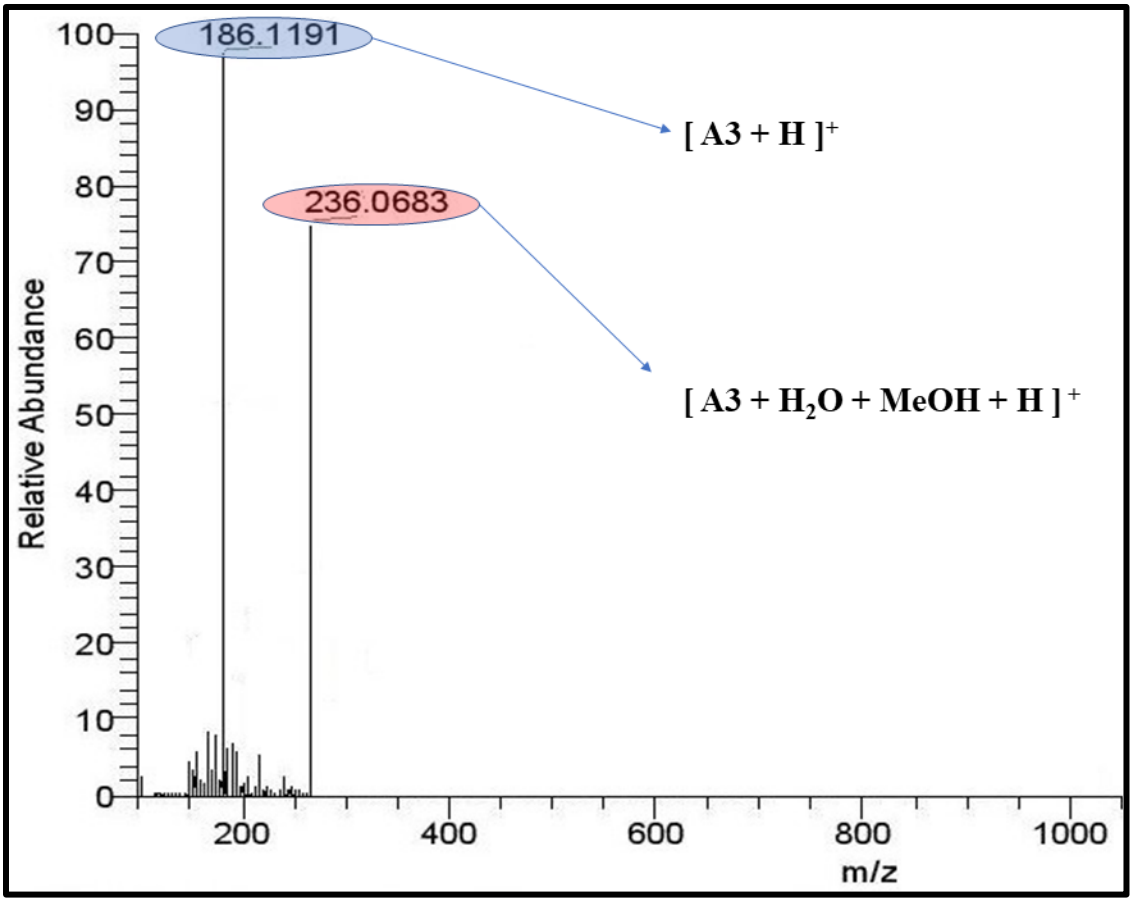

Fig.S10 QTOF-MS spectrum of A3 in MeOH

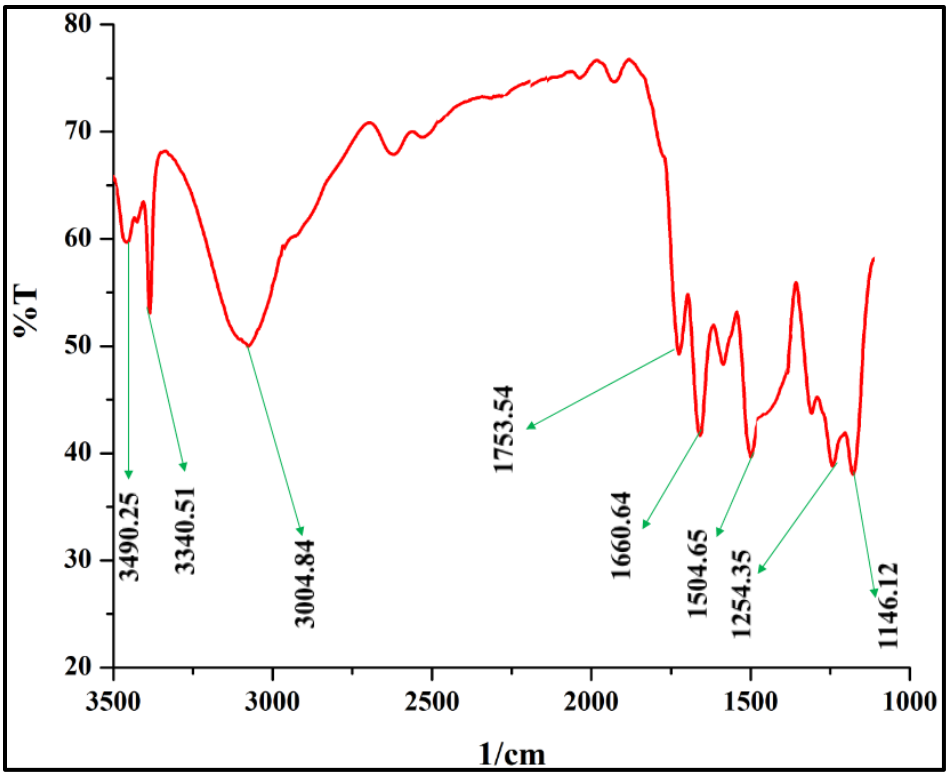

Fig.S11 FTIR spectrum of A3

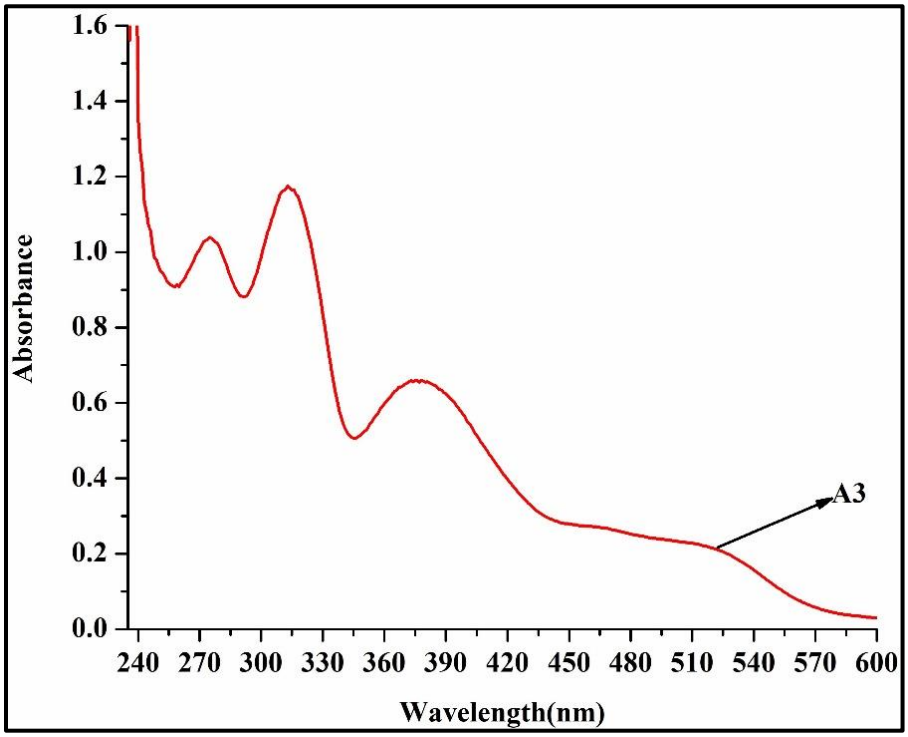

Fig.S12 UV-Vis spectrum of A3

Table S2 Selected bond lengths and bond angles of L11

ATOMS	ANGLE	ATOMS	LENGTH
N12A_a C1 C6	113.7(4)	C1 N12A_a	1.317(5)
N12A_a C1 C2	126.5(4)	C1 C6	1.431(6)
C6 C1 C2	119.8(3)	C1 C2	1.464(6)
C6 C1 N12B_b	138.6(10)	C1 N12B_b	1.48(3)
C2 C1 N12B_b	101.4(10)	C2 O7	1.219(5)
O7 C2 N3	120.2(4)	C2 N3	1.389(5)
O7 C2 C1	123.2(4)	N3 C4	1.378(5)
N3 C2 C1	116.6(3)	N3 C8	1.471(5)
C4 N3 C2	124.2(3)	C4 O9	1.207(5)
C4 N3 C8	119.3(3)	C4 N5	1.393(6)
C2 N3 C8	116.5(3)	N5 C6	1.369(5)
O9 C4 N3	122.2(4)	N5 C10	1.473(6)
O9 C4 N5	120.5(4)	C6 N11	1.299(6)
N3 C4 N5	117.3(3)	C8 H8A	0.9800
C6 N5 C4	124.2(4)	C8 H8B	0.9800
C6 N5 C10	119.0(4)	C8 H8C	0.9800
C4 N5 C10	116.7(4)	C10 H10A	0.9800
N11 C6 N5	120.1(4)	C10 H10B	0.9800
N11 C6 C1	122.0(4)	C10 H10C	0.9800
N5 C6 C1	117.9(4)	N11 H11A	0.8800
N3 C8 H8A	109.5	N11 H11B	0.8800
N3 C8 H8B	109.5	N12A_a N13A_a	1.299(6)
H8A C8 H8B	109.5	N13A_a C14A_a	1.421(6)
N3 C8 H8C	109.5	N13A_a H13A_a	0.8800
H8A C8 H8C	109.5	C14A_a C19A_a	1.395(6)
H8B C8 H8C	109.5	C14A_a C15A_a	1.396(7)
N5 C10 H10A	109.5	C15A_a C16A_a	1.381(7)
N5 C10 H10B	109.5	C15A_a H15A_a	0.9500
H10A C10 H10B	109.5	C16A_a C17A_a	1.398(7)
N5 C10 H10C	109.5	C16A_a H16A_a	0.9500
H10A C10 H10C	109.5	C17A_a C18A_a	1.395(8)
H10B C10 H10C	109.5	C17A_a C20	1.505(7)
C6 N11 H11A	120.0	C18A_a C19A_a	1.389(7)
C6 N11 H11B	120.0	C18A_a H18A_a	0.9500
H11A N11 H11B	120.0	C19A_a H19A_a	0.9500
N13A_a N12A_a C1	117.2(4)	N12B_b N13B_b	1.27(2)

N12A_a N13A_a C14A_a	**120.8(4)**	**N13B_b C14B_b**	**1.45(2)**
N12A_a N13A_a H13A_a	**119.6**	**N13B_b H13B_b**	**0.8800**
C14A_a N13A_a H13A_a	**119.6**	**C14B_b C15B_b**	**1.37(2)**
C19A_a C14A_a C15A_a	**121.9(4)**	**C14B_b C19B_b**	**1.41(2)**
C19A_a C14A_a N13A_a	**115.3(4)**	**C15B_b C16B_b**	**1.40(2)**
C15A_a C14A_a N13A_a	**122.8(4)**	**C15B_b H15B_b**	**0.9500**
C16A_a C15A_a C14A_a	**118.5(4)**	**C16B_b C17B_b**	**1.39(2)**
C16A_a C15A_a H15A_a	**120.8**	**C16B_b H16B_b**	**0.9500**
C14A_a C15A_a H15A_a	**120.8**	**C17B_b C18B_b**	**1.38(3)**
C15A_a C16A_a C17A_a	**121.0(5)**	**C17B_b C20**	**1.43(3)**
C15A_a C16A_a H16A_a	**119.5**	**C18B_b C19B_b**	**1.36(2)**
C17A_a C16A_a H16A_a	**119.5**	**C18B_b H18B_b**	**0.9500**
C18A_a C17A_a C16A_a	**119.4(5)**	**C19B_b H19B_b**	**0.9500**
C18A_a C17A_a C20	**119.5(5)**	**C20 O21**	**1.210(5)**
C16A_a C17A_a C20	**121.0(5)**	**C20 O22**	**1.310(5)**
C19A_a C18A_a C17A_a	**120.6(5)**	**O22 H22**	**0.8400**
C19A_a C18A_a H18A_a	**119.7**	**N1_1a O2_1a**	**1.272(15)**
C17A_a C18A_a H18A_a	**119.7**	**N1_1a O3_1a**	**1.275(17)**
C18A_a C19A_a C14A_a	**118.5(4)**	**N1_1a O1_1a**	**1.292(18)**
C18A_a C19A_a H19A_a	**120.8**	**N1_2b O1_2b**	**1.272(16)**
C14A_a C19A_a H19A_a	**120.8**	**N1_2b O2_2b**	**1.297(15)**
N13B_b N12B_b C1	**114(2)**	**N1_2b O3_2b**	**1.386(16)**
N12B_b N13B_b C14B_b	**113(2)**		
N12B_b N13B_b H13B_b	**123.6**		
C14B_b N13B_b H13B_b	**123.6**		
C15B_b C14B_b C19B_b	**121.9(18)**		
C15B_b C14B_b N13B_b	**125(2)**		
C19B_b C14B_b N13B_b	**114(2)**		
C14B_b C15B_b C16B_b	**117(2)**		
C14B_b C15B_b H15B_b	**121.3**		
C16B_b C15B_b H15B_b	**121.3**		
C17B_b C16B_b C15B_b	**121(2)**		
C17B_b C16B_b H16B_b	**119.4**		
C15B_b C16B_b H16B_b	**119.4**		
C18B_b C17B_b C16B_b	**119(2)**		
C18B_b C17B_b C20	**116(2)**		
C16B_b C17B_b C20	**125(2)**		
C19B_b C18B_b C17B_b	**121(2)**		

C19B_b C18B_b H18B_b	119.7		
C17B_b C18B_b H18B_b	119.7		
C18B_b C19B_b C14B_b	119(2)		
C18B_b C19B_b H19B_b	120.7		
C14B_b C19B_b H19B_b	120.7		
O21 C20 O22	124.9(4)		
O21 C20 C17B_b	115.3(13)		
O22 C20 C17B_b	119.7(13)		
O21 C20 C17A_a	123.2(4)		
O22 C20 C17A_a	111.9(4)		
C20 O22 H22	109.5		
O2_1a N1_1a O3_1a	114.4(19)		
O2_1a N1_1a O1_1a	121.6(19)		
O3_1a N1_1a O1_1a	124.0(18)		
O1_2b N1_2b O2_2b	112.4(16)		
O1_2b N1_2b O3_2b	123.1(15)		
O2_2b N1_2b O3_2b	121.6(17)		

Table S3 Selected bond lengths and bond angles of L11P

ATOMS	ANGLE	ATOMS	LENGTH
N3 C1 N4	127.7(2)	C1 N3	1.328(3)
N3 C1 C2	110.4(2)	C1 N4	1.372(3)
N4 C1 C2	121.8(2)	C1 C2	1.385(4)
N2 N1 C2	102.5(2)	N1 N2	1.333(3)
N1 C2 C1	108.9(2)	N1 C2	1.342(4)
N1 C2 C4	128.5(2)	C2 C4	1.449(4)
C1 C2 C4	122.6(2)	N2 N3	1.354(3)
N1 N2 N3	116.5(2)	N2 C5	1.429(3)
N1 N2 C5	121.5(2)	N4 C3	1.372(3)
N3 N2 C5	122.0(2)	N4 C4A	1.471(3)
C1 N3 N2	101.7(2)	C4A H4A1	0.9800
C3 N4 C1	119.1(2)	C4A H4A2	0.9800
C3 N4 C4A	120.3(2)	C4A H4A3	0.9800
C1 N4 C4A	120.6(2)	C3 O1	1.225(3)
N4 C4A H4A1	109.5	C3 N5	1.402(3)
N4 C4A H4A2	109.5	N5 C4	1.407(3)
H4A1 C4A H4A2	109.5	N5 C5A	1.472(3)
N4 C4A H4A3	109.5	C5A H5A1	0.9800
H4A1 C4A H4A3	109.5	C5A H5A2	0.9800

H4A2 C4A H4A3	**109.5**	**C5A H5A3**	**0.9800**
O1 C3 N4	**120.6(2)**	**C4 O2**	**1.207(3)**
O1 C3 N5	**121.1(2)**	**C5 C6**	**1.386(4)**
N4 C3 N5	**118.4(2)**	**C5 C10**	**1.388(3)**
C3 N5 C4	**126.4(2)**	**C6 C7**	**1.389(4)**
C3 N5 C5A	**116.2(2)**	**C6 H6**	**0.9500**
C4 N5 C5A	**117.4(2)**	**C7 C8**	**1.393(4)**
N5 C5A H5A1	**109.5**	**C7 H7**	**0.9500**
N5 C5A H5A2	**109.5**	**C8 C9**	**1.385(4)**
H5A1 C5A H5A2	**109.5**	**C8 C11**	**1.496(4)**
N5 C5A H5A3	**109.5**	**C9 C10**	**1.384(4)**
H5A1 C5A H5A3	**109.5**	**C9 H9**	**0.9500**
H5A2 C5A H5A3	**109.5**	**C10 H10**	**0.9500**
O2 C4 N5	**122.0(2)**	**C11 O4**	**1.203(3)**
O2 C4 C2	**126.9(3)**	**C11 O3**	**1.336(3)**
N5 C4 C2	**111.1(2)**	**O3 H3A**	**0.8412**
C6 C5 C10	**122.2(2)**		
C6 C5 N2	**118.7(2)**		
C10 C5 N2	**119.1(2)**		
C5 C6 C7	**118.6(2)**		
C5 C6 H6	**120.7**		
C7 C6 H6	**120.7**		
C6 C7 C8	**120.1(2)**		
C6 C7 H7	**119.9**		
C8 C7 H7	**119.9**		
C9 C8 C7	**119.9(2)**		
C9 C8 C11	**118.9(2)**		
C7 C8 C11	**121.2(2)**		
C10 C9 C8	**120.9(2)**		
C10 C9 H9	**119.5**		
C8 C9 H9	**119.5**		
C9 C10 C5	**118.2(2)**		
C9 C10 H10	**120.9**		
C5 C10 H10	**120.9**		
O4 C11 O3	**123.8(2)**		
O4 C11 C8	**123.4(2)**		
O3 C11 C8	**112.8(2)**		
C11 O3 H3A	**111.0**		

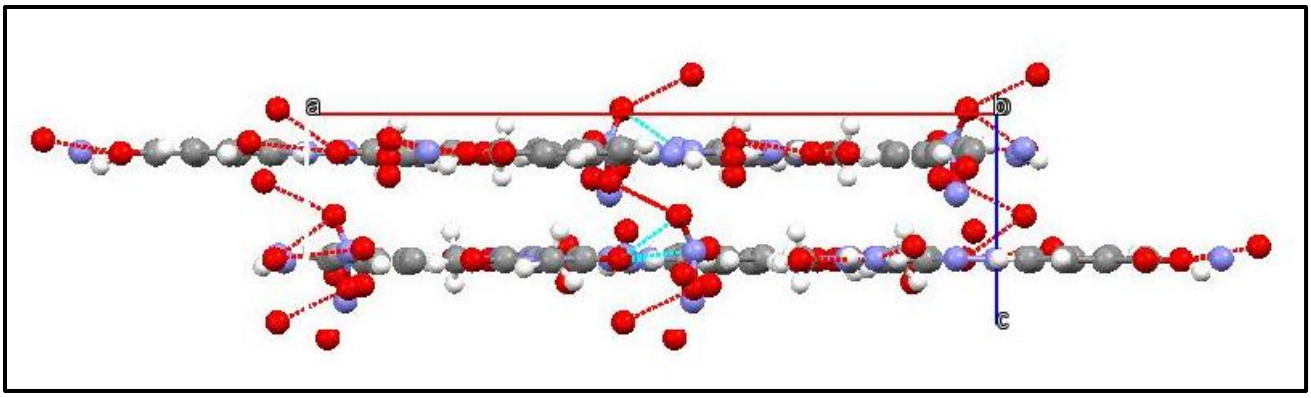

Fig. S13 Packing diagram of L11

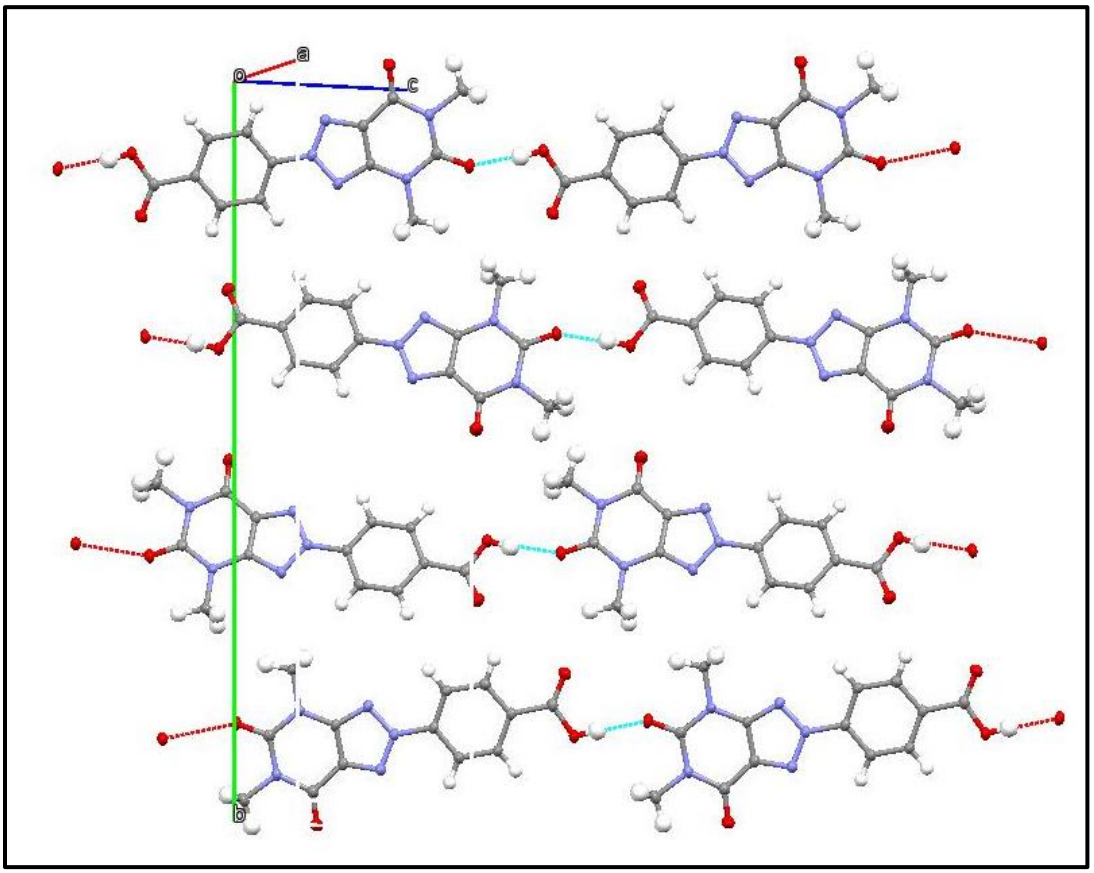

Fig. S14 Packing diagram of L11P

Table S4 Selected bond lengths and bond angles of A3

ATOMS	ANGLE	ATOMS	LENGTH
N2 N1 C009	118.6(6)	O2 C00B	1.329(8)
C00B N4 C00C	122.9(5)	O002 C00C	1.224(8)
C00B N4 C00D	120.0(6)	N1 N2	1.305(8)
C00C N4 C00D	117.2(6)	N1 C009	1.347(9)
C00C N005 C00A	124.8(6)	N3 C00A	1.215(8)
C00C N005 C00E	116.3(6)	N4 C00B	1.379(8)
C00A N005 C00E	118.9(6)	N4 C00C	1.400(9)
N3 C00A N005	120.4(6)	N4 C00D	1.491(9)
N3 C00A C009	124.6(6)	N005 C00C	1.385(9)
N005 C00A C009	115.0(6)	N005 C00A	1.412(9)
O2 C00B N4	120.0(6)	N005 C00E	1.483(9)
O2 C00B C009	121.2(6)	C00A C009	1.463(10)
N4 C00B C009	118.7(6)	C00B C009	1.430(10)
O002 C00C N005	121.4(7)		
O002 C00C N4	120.8(6)		
N005 C00C N4	117.7(6)		
N1 C009 C00B	126.2(6)		
N1 C009 C00A	112.9(6)		
C00B C009 C00A	120.8(6)		

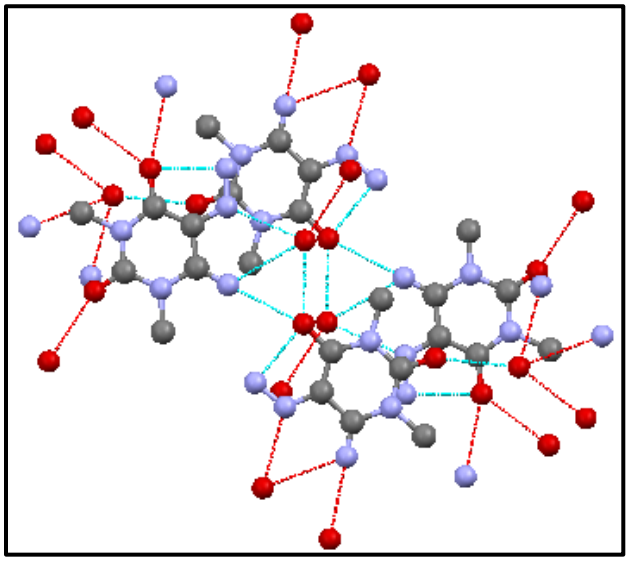

Fig. S15 Packing diagram of A3

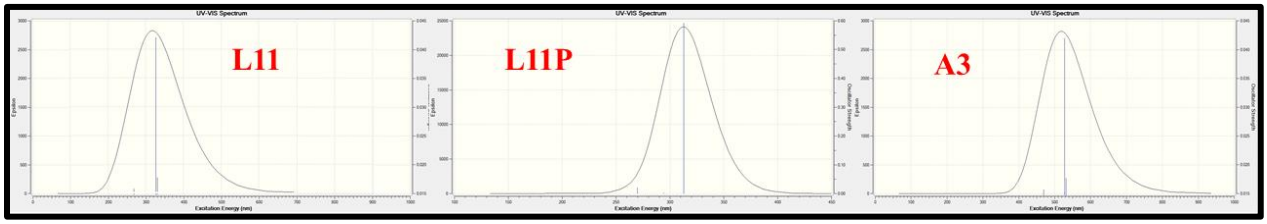

Fig.S16 Theoretical UV spectrum of L11 (left), L11P (middle) and A3 (right).

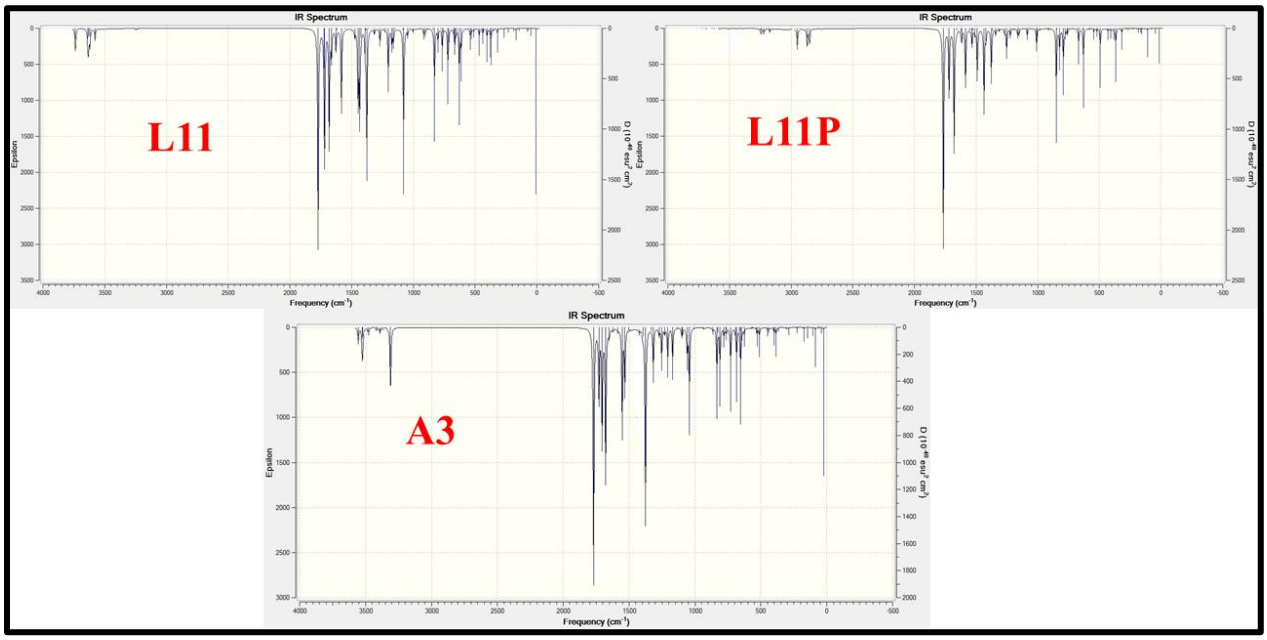

Fig.S17 Theoretical IR spectrum of L11(left), L11P(right) and A3 (lower middle).

Table S5 Comparison of experimental value of IR with theoretical value

Compounds	Assignment	Experimental value	Theoretical value
L11	**ν(-NH_2)**	**3507.44**	**3577.90**
	ν (Aromatic C-H)	**3324.81**	**3254.97**
	ν(MeN-C=O)	**1685.84**	**1682.41**
	ν(C=C)	**1619.19**	**1618.38**
	ν(-N=N-)	**1448.56**	**1448.31**
	ν(O=C-N)	**1344.59**	**1367.52**

L11P	**ν(Sp3C-H)**	**2975.02**	**2990.54**
	ν(MeN-C=O)	**1708.06**	**1704.50**
	ν(C=N)	**1583.63**	**1647.43**
	ν(O=C-N)	**1299.72**	**1367.34**
A3	**ν(-NH_2)**	**3490.25, 3340.51**	**3504.72, 3320.75**
	ν(Sp3C-H)	**3004.84**	**2874.25**
	ν(MeN-C=O)	**1753.54**	**1770.19**
	ν(C=C)	**1660.64**	**1651.70**
	ν(O=C-N)	**1254.35**	**1316.80**

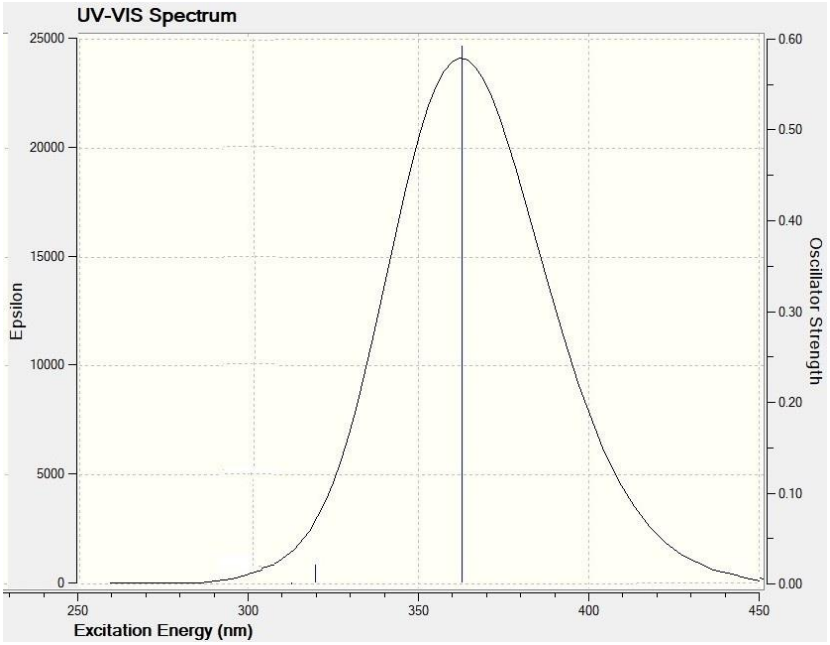

Fig.S18 Theoretical UV spectrum of six membered intermediate.

Table S6 TDDFT calculation in methanol

Compound	Electronic Transitions	Energy[a](eV)	Wavelength (nm)	f[b]	Transitions involved
L11	$S_0 \rightarrow S_1$	1.6867 eV	504.20 nm	0.0023	HOMO-1→LUMO HOMO-1→LUMO+1 HOMO-1→LUMO+4 HOMO-1→LUMO+2
	$S_0 \rightarrow S_2$	2.3477 eV	328.11 nm	0.0963	HOMO-2→LUMO HOMO-1→LUMO
	$S_0 \rightarrow S_3$	2.4262 eV	311.03 nm	0.0005	HOMO-1→LUMO

Table S7 TDDFT calculation in gas phase

Compound	Electronic transitions	Energy[a] (eV)	Wavelength (nm)	f[b]	Transitions involved
L11P	$S_0 \rightarrow S_1$	3.9656 eV	312.65 nm	0.5927	HOMO→LUMO
	$S_0 \rightarrow S_2$	4.2110 eV	294.43 nm	0.0019	HOMO-6→LUMO HOMO-1→LUMO HOMO-1→LUMO+1
	$S_0 \rightarrow S_3$	4.5961 eV	269.76 nm	0.0207	HOMO-4→LUMO+2 HOMO-3→LUMO HOMO-2→LUMO HOMO→LUMO+2

Table S8 TDDFT calculation in gas phase

Compound	**Electronic transitions**	**Energy[a] (eV)**	**Wavelength (nm)**	**f[b]**	**Transitions involved**
Intermediate six membered ring compound	$S_0 \rightarrow S_1$	2.0971 eV	591.22 nm	0.0034	HOMO-1 →LUMO
	$S_0 \rightarrow S_2$	2.8929 eV	428.58 nm	0.0024	HOMO-2→LUMO+1 HOMO →LUMO+1
	$S_0 \rightarrow S_3$	3.6204 eV	342.46 nm	0.0016	HOMO-2→LUMO+1 HOMO→LUMO+1 HOMO→LUMO +3

Table S9 TDDFT calculation in gas phase

Compound	**Electronic transitions**	**Energy[a](eV)**	**Wavelength (nm)**	**f[b]**	**Transitions involved**
A3	$S_0 \rightarrow S_1$	1.9339 eV	641.12 nm	0.0056	HOMO-1 →LUMO
	$S_0 \rightarrow S_2$	3.0388 eV	408.34 nm	0.0027	HOMO-2→LUMO HOMO →LUMO+1
	$S_0 \rightarrow S_3$	3.3542 eV	369.64 nm	0.0020	HOMO-2→LUMO+1 HOMO→LUMO+1 HOMO→LUMO +3

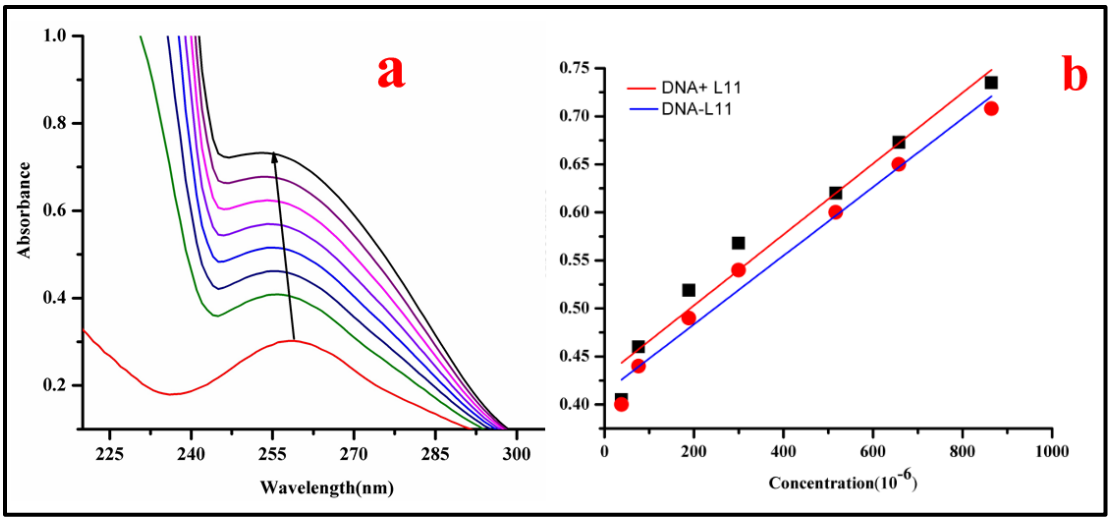

Fig.S19 (a) Change of absorbance of DNA (38.09 µM) upon gradual addition of L11 (µM) at 23^{O}C: (1) 0.0, (2) 22.21, (3) 75.98, (4) 188.56, (5) 299.47, (6) 516.49, (7) 657.70 and (8) 805.54; (b) comparison of absorbance at 260 nm between [DNA- L11] adduct *vs.* summation of free L11 and free DNA.

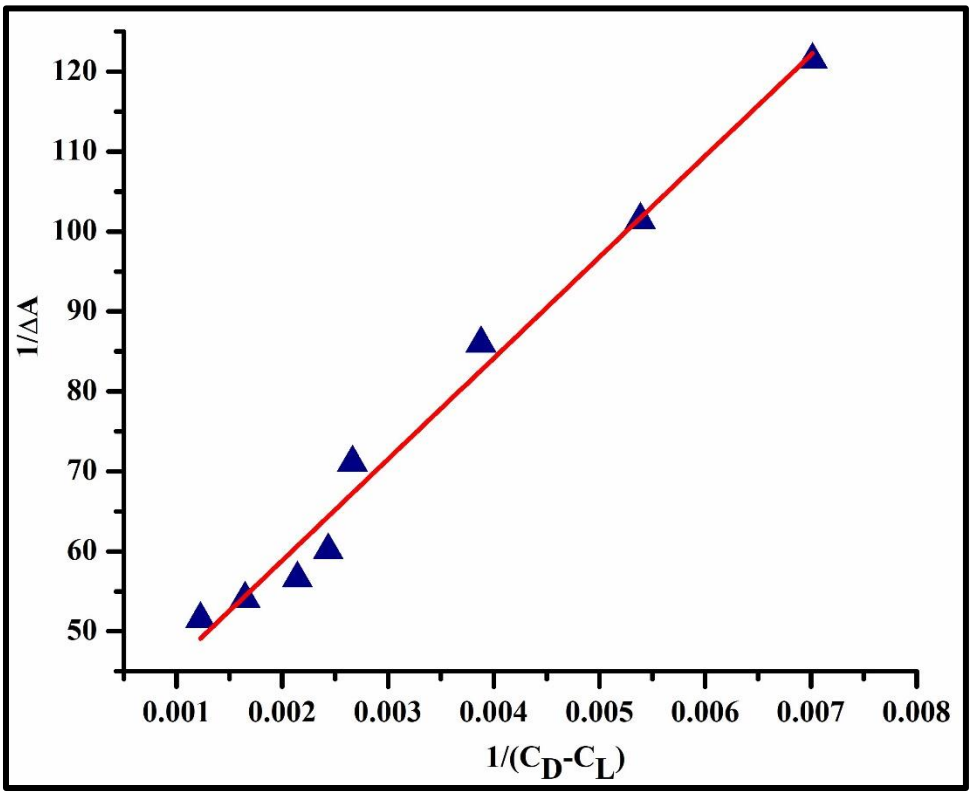

Fig.S20 Plot of the inverse of (C_D - C_L) with the inverse of ΔA

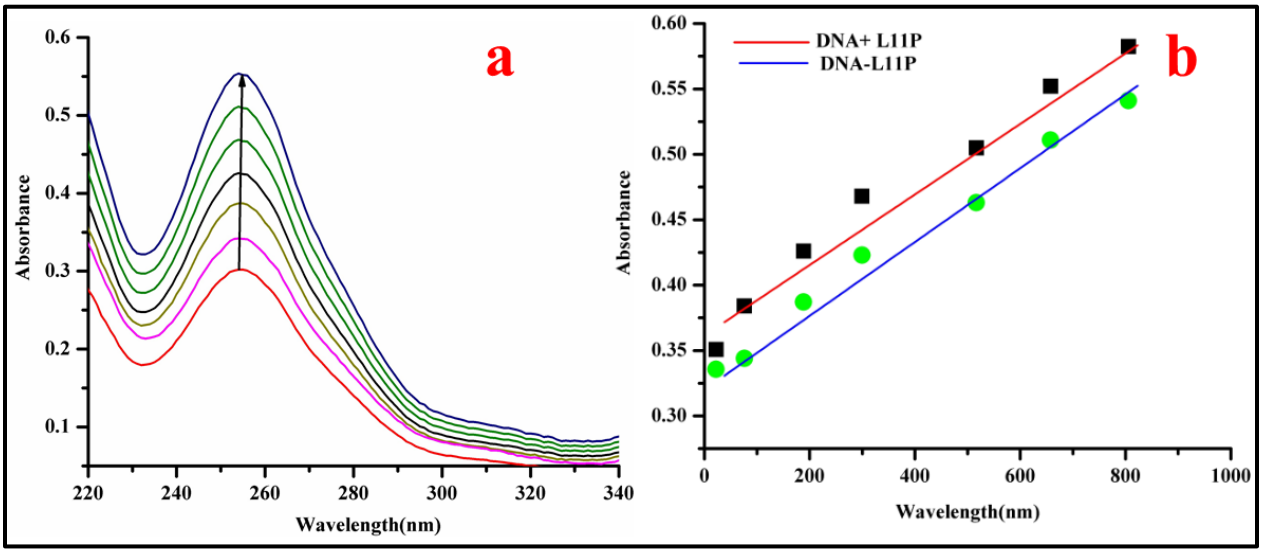

Fig.S21 (a) Change of absorbance of DNA (38.09 µM) upon gradual addition of L11P (µM) at 23^{O}C: (1) 0.0, (2) 22.21, (3) 75.98, (4) 188.56, (5) 299.47, (6) 516.49, (7) 657.70 and (8) 805.54; (b) comparison of absorbance at 260 nm between [DNA- L11P] adduct *vs.* summation of free L11P and free DNA.

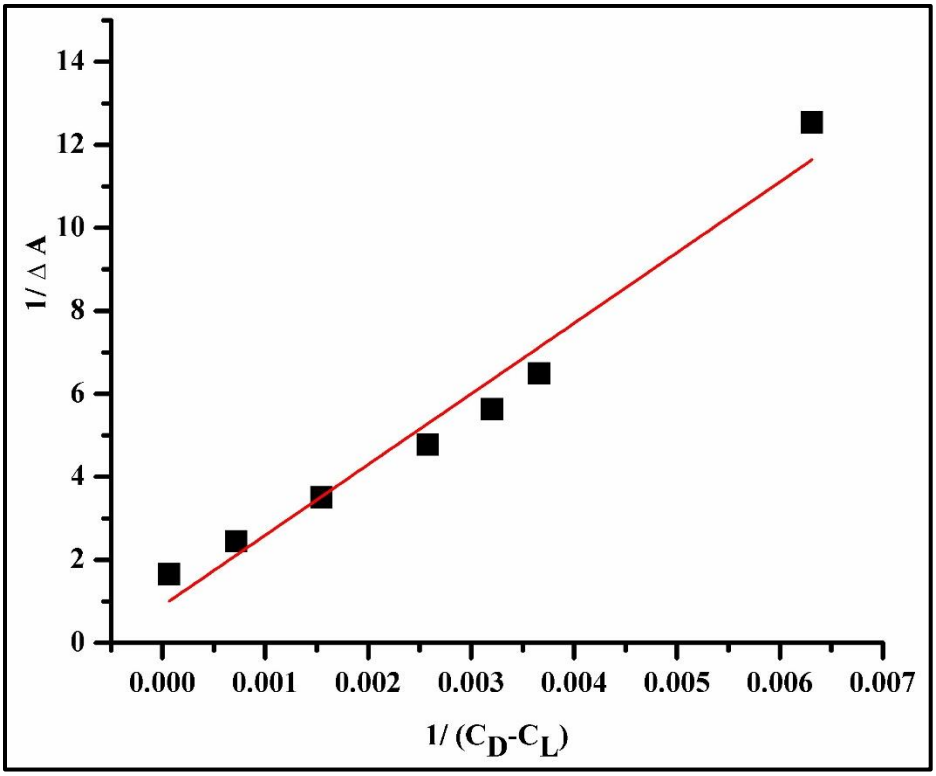

Fig.S22 Plot of the inverse of (C_D - C_L) with the inverse of ΔA

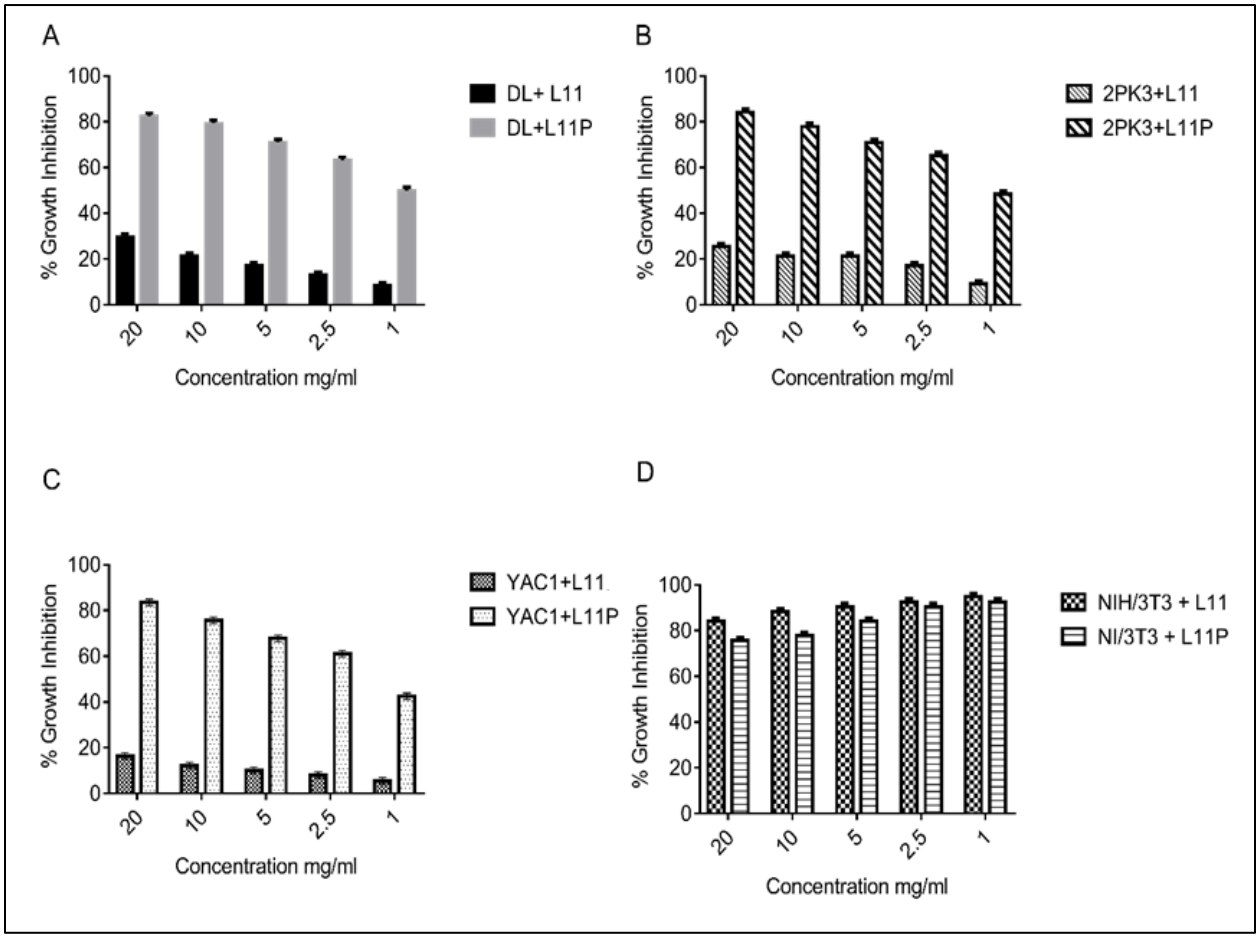

Fig.S23 The growth inhibition of DL (A), 2PK3 (B), YAC1 (C) and NIH/3T3 (D) cells treated with L11 and L11P with indicated concentrations for 48h