Supporting information

Effective combination treatment of lung cancer cells by single vehicular delivery of siRNA and different anticancer drugs

Jinming Li,[‡] Yuanyuan Wang,[‡] Shanshan Xue, Jinghua Sun, Wei Zhang, Ping Hu, Liangnian Ji, Zongwan Mao[*]

MOE Key Laboratory of Bioinorganic and Synthetic Chemistry, School of Chemistry and Chemical Engineering, Sun Yat-sen University, Guangzhou 510275, China

Correspondence: Zongwan Mao

MOE Key Laboratory of Bioinorganic and Synthetic Chemistry, School of Chemistry and Chemical Engineering, Sun Yat-sen University, Guangzhou 510275, China

Tel +86 20 8411 3788

Fax +86 20 8411 2245

Email: cesmzw@mail.sysu.edu.cn

[‡]Contributed equally to this paper.

Keywords: combination therapy, lung cancer cells, Bcl-2 siRNA, anticancer drugs, quantum dot nanocarriers

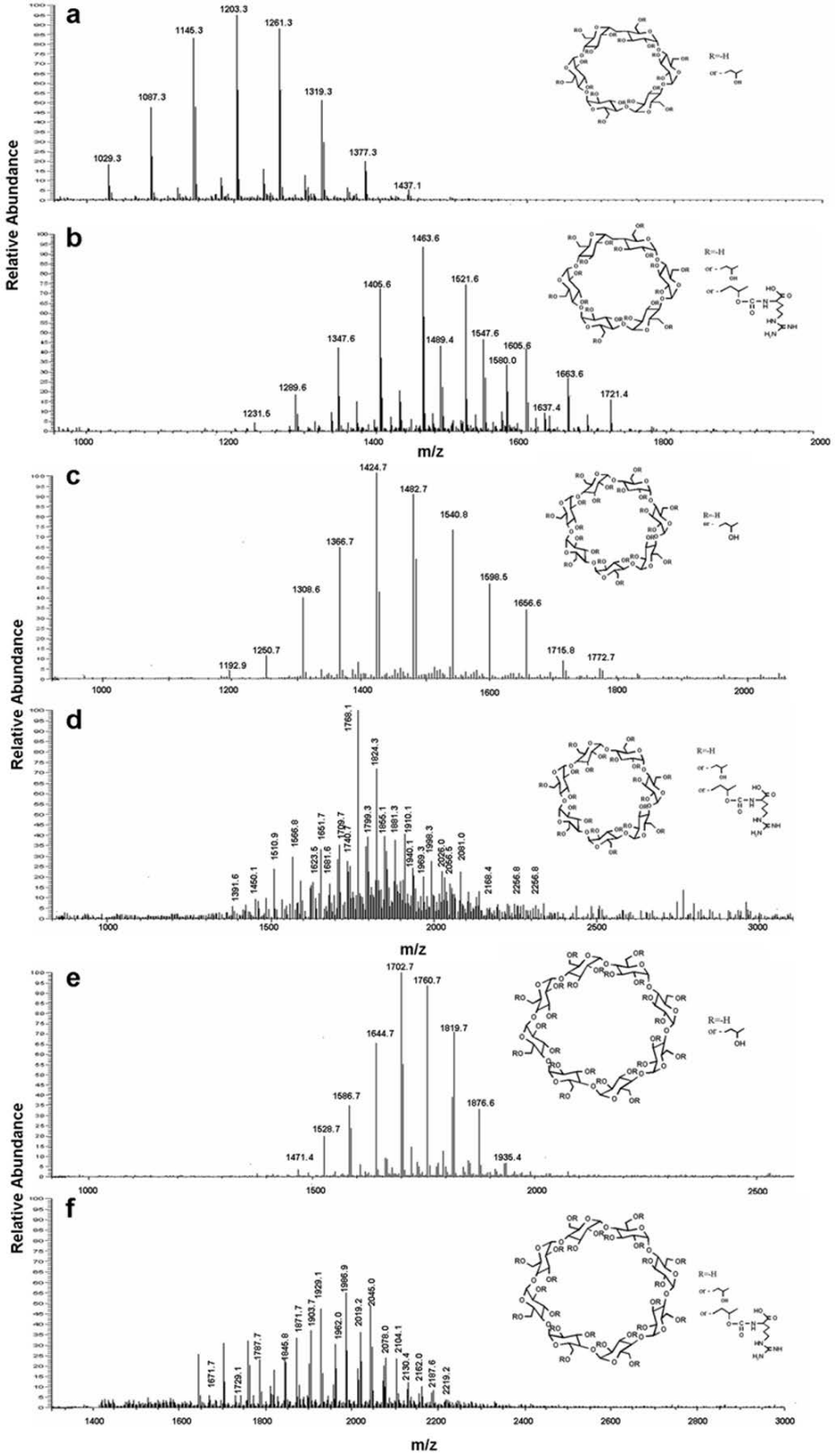

Fig. S1. Mass spectra and chemical structure of a) HP-α-CD, b) HP-α-CD-(L-Arg)$_2$, c) HP-β-CD, d) HP-β-CD-(L-Arg)$_2$, e) HP-γ-CD and f) HP-γ-CD-(L-Arg)$_2$.

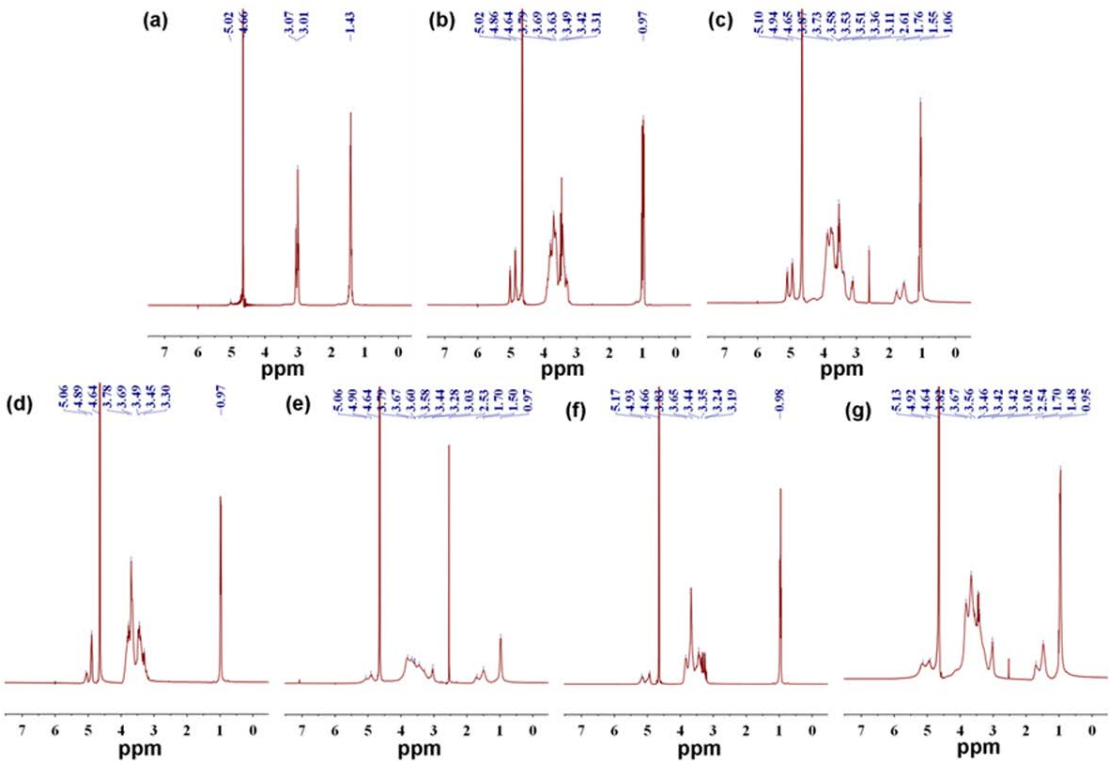

Fig. S2 ^{1}H NMR spectra (300 MHz, room temperature) in D_2O of a) $(L\text{-}Arg)_2$, b) HP-α-CD, c) HP-α-CD-$(L\text{-}Arg)_2$, d) HP-β-CD, e) HP-β-CD-$(L\text{-}Arg)_2$, f) HP-γ-CD, g) HP-γ-CD-$(L\text{-}Arg)_2$.

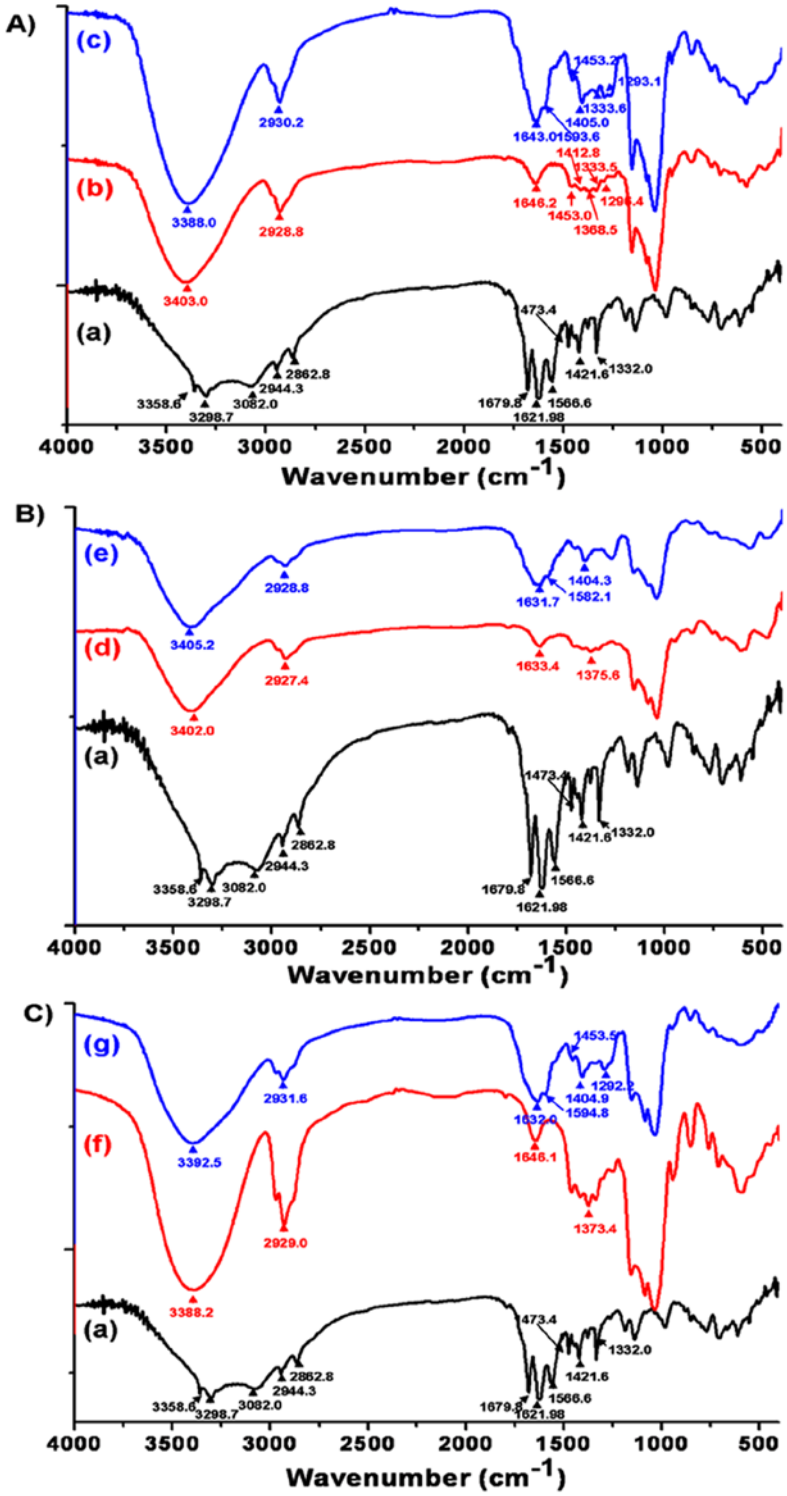

Fig. S3 Infrared spectrum of a) L-Arg, b) HP-α-CD, c) HP-α-CD-(L-Arg)$_2$, d) HP-β-CD, e) HP-β-CD-(L-Arg)$_2$, f) HP-γ-CD, g) HP-γ-CD-(L-Arg)$_2$. The obtained products were dispersed in KBr and the mixtures were compressed to form disks for FTIR spectrum.

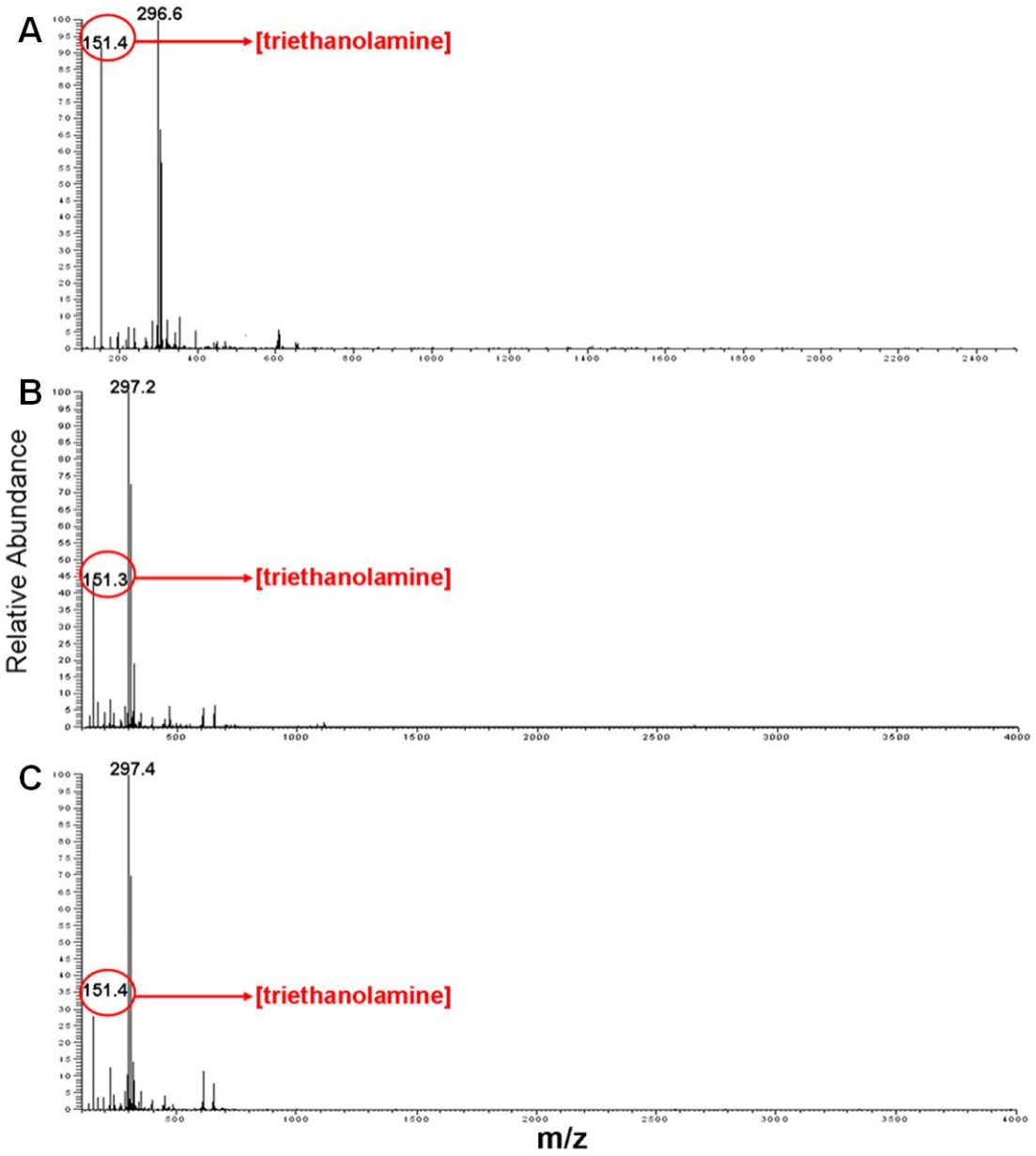

Fig. S4. MS of filter liquor obtained from the ligand exchange reaction for synthesis of A) HP-α/β-CD-(L-Arg)$_2$-QDs, B) HP-α/γ-CD-(L-Arg)$_2$-QDs and C) HP-β/γ-CD-(L-Arg)$_2$-QDs by ultrafiltration (Mw=10 K).

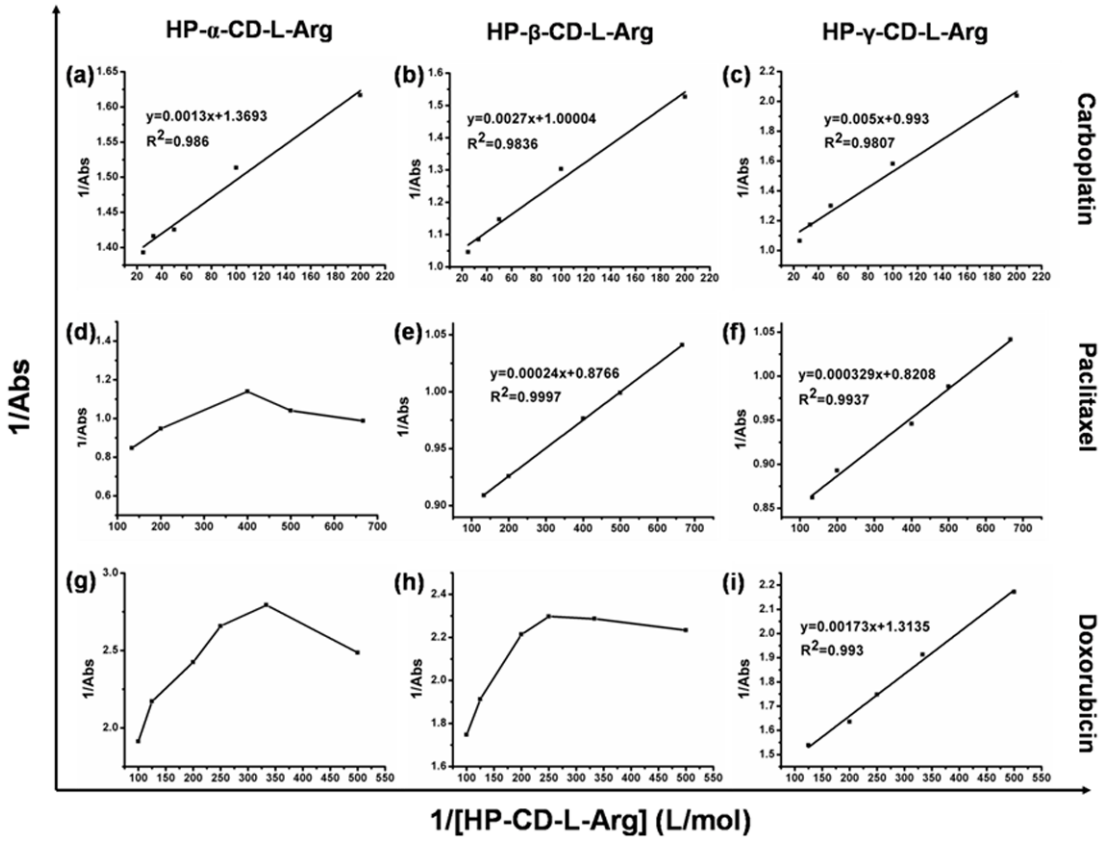

Fig. S5. The absorbance change of a) Carbo/HP-α-CD-(L-Arg)$_2$, b) Carbo/HP-β-CD-(L-Arg)$_2$, c) Carbo/ HP-γ-CD-(L-Arg)$_2$, d) Tax/ HP-α-CD-(L-Arg)$_2$, e) Tax/HP-β-CD-(L-Arg)$_2$, f) Tax/HP-γ-CD-(L-Arg)$_2$, g) Dox/ HP-α-CD-(L-Arg)$_2$, h) Dox/HP-β-CD-(L-Arg)$_2$ and i) Dox/HP-γ-CD-(L-Arg)$_2$ complexes as the concentration of HP-CD-(L-Arg)$_2$ increased, which was determined by UV-vis spectroscopy. The existence of linear relationship between above two parameters indicated formation of 1:1 inclusion complexes of drugs with HP-CD-(L-Arg)$_2$.

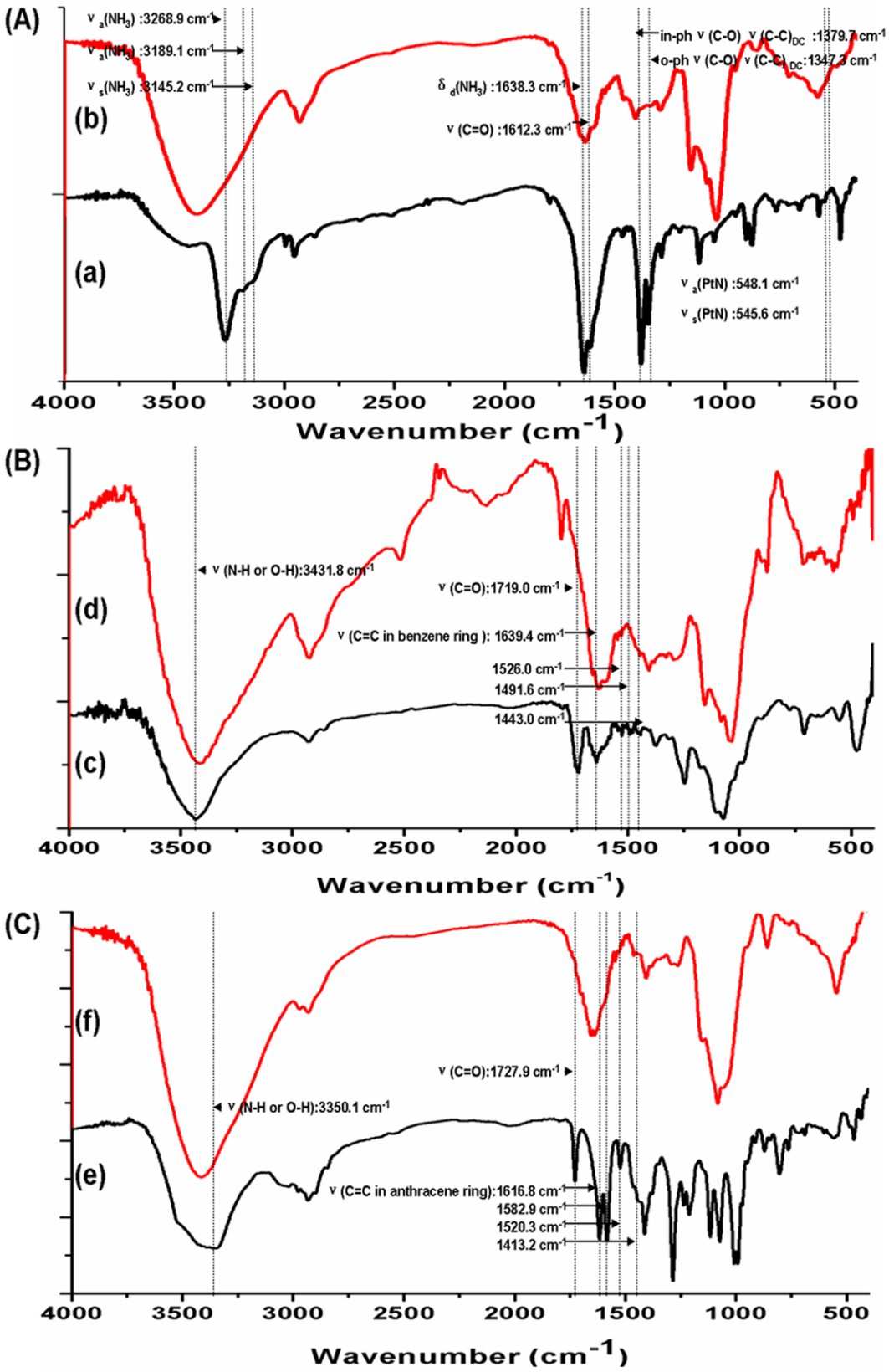

Fig. S6. The FTIR spectra of a) Carbo, b) Carbo/HP-α-CD-(L-Arg)$_2$, c) Tax, d) Tax /HP-β-CD-(L-Arg)$_2$, e) Dox, f) Dox/HP-γ-CD-(L-Arg)$_2$ complexes.

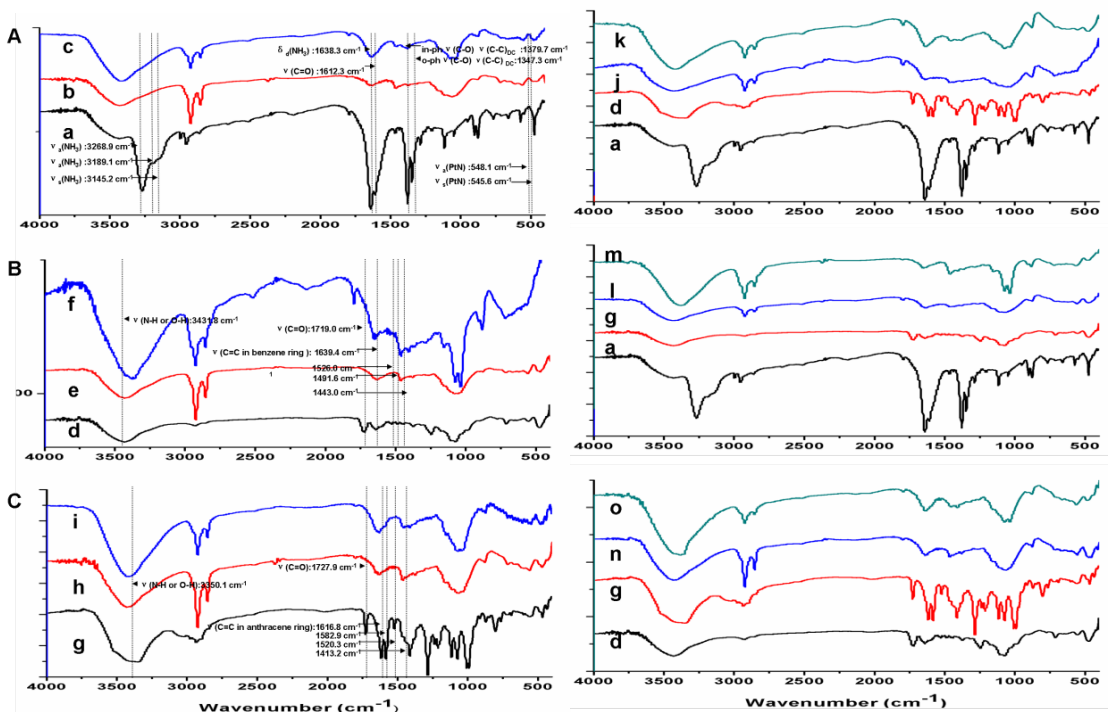

Fig. S7. The FTIR spectra of a) Carbo, b) HP-α-CD-(L-Arg)$_2$-QDs, c) Carbo/ HP-α-CD-(L-Arg)$_2$-QDs, d) Tax, e) HP-β-CD-(L-Arg)$_2$-QDs, f) Tax/HP-β-CD-(L-Arg)$_2$-QDs, g) Dox, h) HP-γ-CD-(L-Arg)$_2$-QDs, i) Dox/ HP-γ-CD-(L-Arg)$_2$-QDs, j) HP-α/β-CD-(L-Arg)$_2$-QDs, k) Carbo/Tax/ HP-α/β-CD-(L-Arg)$_2$-QDs, l) Carbo/Dox/HP-α/γ-CD-(L-Arg)$_2$-QDs, n) HP-β/γ-CD-(L-Arg)$_2$-QDs and o) Tax/Dox/HP-β/γ-CD-(L-Arg)$_2$-QDs.

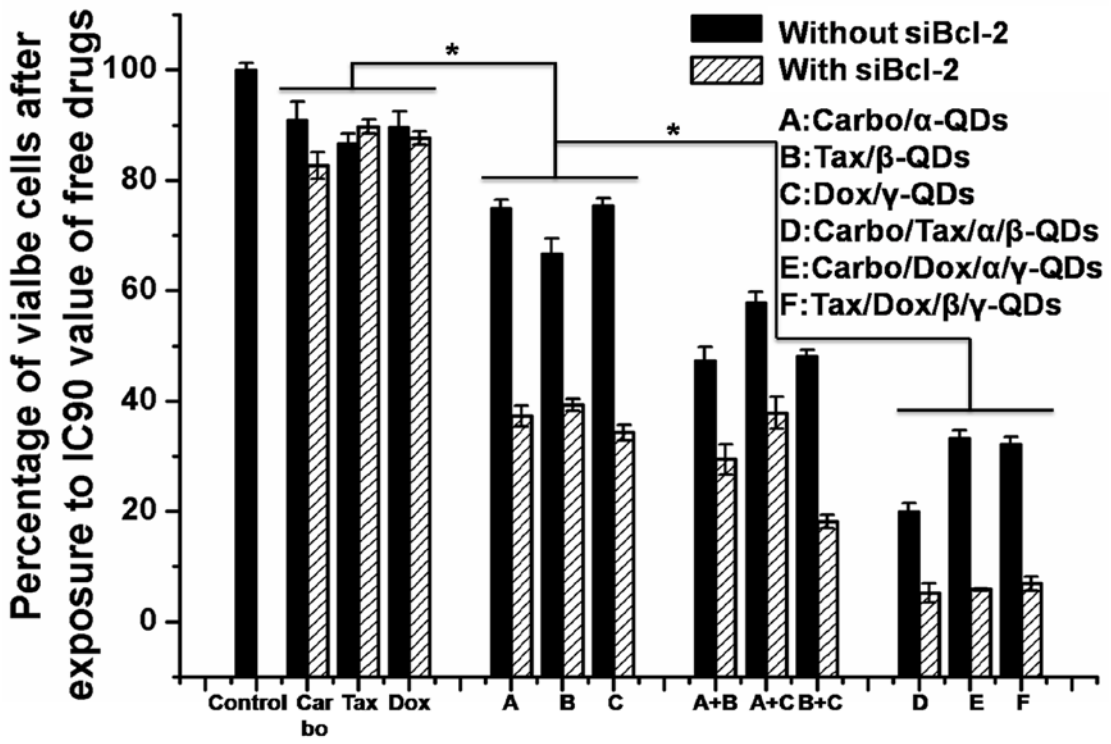

Fig. S8. Cell viability in A549 cells after exposure to free drugs, drug loaded QDs in the presence or absence of siBcl-2 or their cocktail for 72 h at IC90 concentration of free drugs was detected by MTT assay. The concentration of siRNA was 50 nM. The incubation time was 72 h.

Tabel. S1 Theoretical values of molecular weight and data detected by MS for A) HP-α-CD and HP-α-CD-(L-Arg)$_2$, B) HP-β-CD and HP-β-CD-(L-Arg)$_2$, C) HP-γ-CD and HP-γ-CD-(L-Arg)$_2$ at different degree of hydroxypropylation and with different number of (L-Arg)$_2$ coupled to HP-CDs.

A

Number of hydroxy-propyl	Theoretical values	Values in MS	Number of Arg	Theoretical values	Values in MS
1	1030.92	1029.3	1	1231.12	1231.5
2	1089.00	1087.3	1/2	1289.20/1489.39	1289.6/1489.4
3	1147.08	1145.3	1/2	1347.27/1547.47	1347.6/1547.6
4	1205.16	1203.3	1/2	1405.36/1605.55	1405.6/1605.6
5	1263.24	1261.3	1/2	1463.43/1663.63	1463.6/1663.6
6	1321.32	1319.3	1/2	1521.51/1721.70	1521.6/1721.4
7	1379.40	1377.3	1/2	1579.59/1779.78	1580.0/--
8	1437.48	1437.1	1/2	1637.67/1837.86	1637.4/--

B

Number of hydroxypropyl	Theoretical values	Values in MS	Number of Arg	Theoretical values	Values in MS
1	1193.06	1192.9	1	1393.26	1391.6
2	1251.14	1250.7	1/2	1451.33/1651.53	1450.1/1651.7
3	1309.22	1308.6	1/2/3	1509.42/1709.61/1909.81	1510.9/1709.7/1910.1
4	1367.30	1366.7	1/2/3/4	1567.50/1767.69/1967.89/2168.08	1566.8/1768.1/1969.3/2168.4
5	1425.38	1424.7	1/2/3	1625.58/1825.77/2025.96	1623.5/1824.3/2026.9
6	1483.46	1482.7	1/2/3	1683.65/1883.85/2084.04	1799.3/1881.3/2081.0
7	1541.54	1540.8	1/2/3	1741.73/1941.93	1740.7/1940.1
8	1599.62	1598.5	1/2/3	1799.81/2000.01/2200.20	1801.1/1998.3/2200.5
9	1657.70	1656.6	1/2/3	1857.89/2058.09/2258.28	1855.1/2056.5/2256.8
10	1715.78	1715.8	1/2/3	1915.97/2116.2/2316.4	--/--/2316.4
11	1773.85	1772.7	1/2/3	1974.1/2174.2/2374.4	1972.2/2172.1/2372.1

C

Number of hydroxypropyl	Theoretical values	Values in MS	Number of Arg	Theoretical values	Values in MS
1	1355.2	--	1	1555.4	--
2	1413.3	--	1/2	1613.5/1813. 7	--/--
3	1471.4	1471.4	1/2/3	1671.6/1871.7/ 2071.9	1671.7/1871.7
4	1529.4	1528.7	1/2/3/4	1729.6/1929.8/ 2130.0/2330.2	1729.1/1929.1/ 2130.4/--
5	1587.5	1586.7	1/2/3	1787.7/1987.9/ 2188.1	1787.7/1986.9/ 2187.6
6	1645.6	1644.7	1/2/3	1845.8/2046.0/ 2246.2	1845.8/2045.0/ --
7	1703.7	1702.7	1/2/3	1903.9/2104.1/ 2304.2	1903.7/2104.1/ --
8	1761.8	1760.7	1/2/3	1962.0/2162.1/ 2362.3	1962.0/2162.0/ --
9	1819.8	1819.7	1/2/3	2020.0/2220.2 2420.4	2019.2/2219.2/ --
10	1877.9	1876.6	1/2/3	2078.1/2278.3/ 2478.5	2078.0/--/--
11	1936.0	1935.4	1/2/3	2136.2/2336.4/ 2536.6	--/--/--

Table S2. Zeta potentials of α-QDs, β-QDs, γ-QDs, α/β-QDs, α/γ-QDs and β/γ-QDs.

QDs	α-QD	β-QD	γ-QD	α/β-QD	α/γ-QD	β/γ-QD
Zeta potential (mV)	26.9	21.4	17.8	22.6	20.6	19.8

Table S3. Stability constants of inclusion complexes of carboplatin, paclitaxel and doxorubicin respectively with HP-α-CD-(L-Arg)$_2$, HP-β-CD-(L-Arg)$_2$ and HP-γ-CD-(L-Arg)$_2$, determined by UV spectroscopic titration. "--" indicated the nonexistence of formation of 1:1 inclusion complexes of drugs with HP-CD-(L-Arg)$_2$.

Stability constant (10^3 M^{-1})	HP-α-CD-$(L\text{-}Arg)_2$	HP-β-CD-$(L\text{-}Arg)_2$	HP-γ-CD-$(L\text{-}Arg)_2$
Carboplatin	1.05	0.37	0.20
Paclitaxel	--	3.65	2.49
Doxorubicin	--	--	0.76