Fig.3. Zerumbone alleviated D-gal-induced neuronal apoptosis and oxidative damage in vitro.

Representative Western blotting images of Bcl-2 and Bax protein in D-gal-induced SH-SY5Ycells.

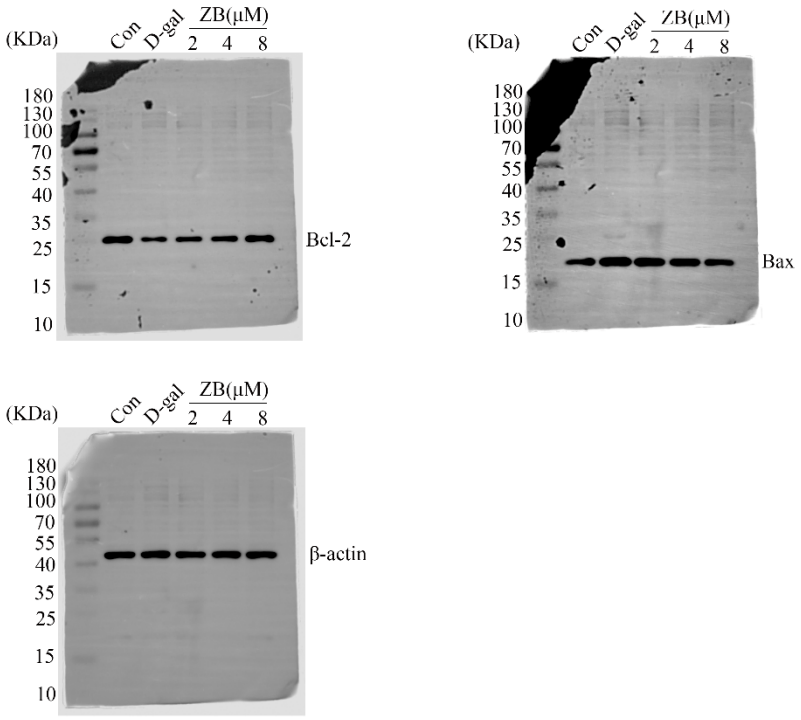
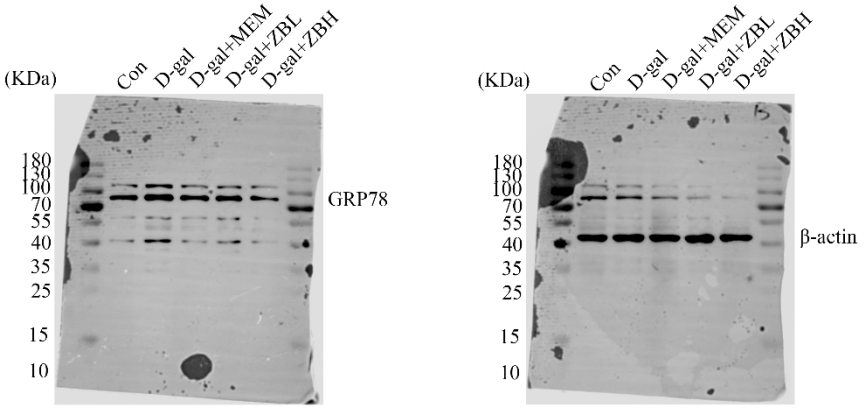
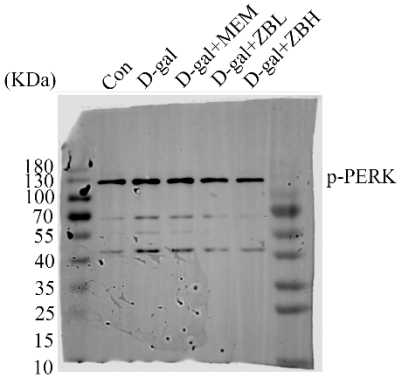
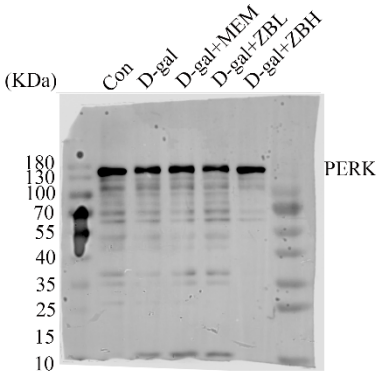


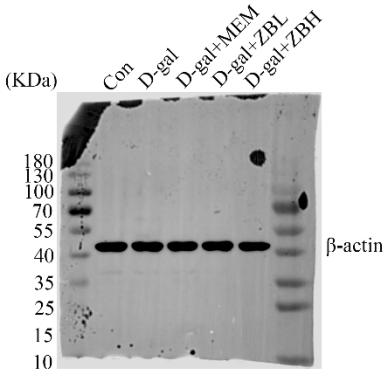
Fig.4. Zerumbone inhibited D-gal-induced activation of PERK/CHOP pathway in vivo and in vitro

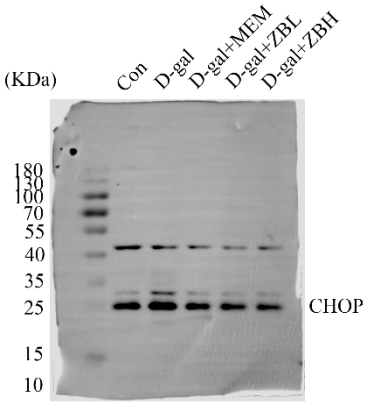
Representative Western blotting images of GRP78, p-PERK/PERK, and CHOP in D-gal-induced mice.

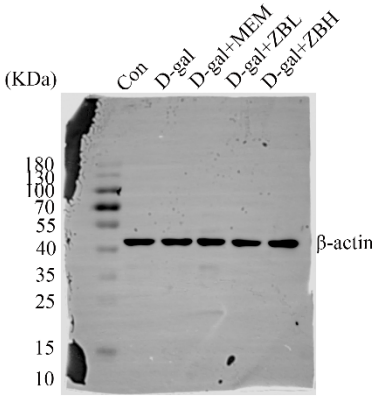



(KDa)
Con
D-gal
D-gal+MEM
D-gal+ZBL
D-gal+ZBH
180
130
100
70
55
40
35
25
15
10
p-PERK

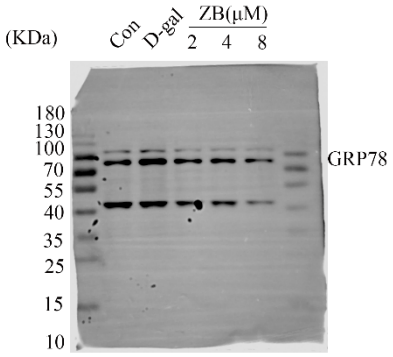

(KDa)
Con
D-gal
D-gal+MEM
D-gal+ZBL
D-gal+ZBH
180
130
100
70
55
40
35
25
15
10
PERK

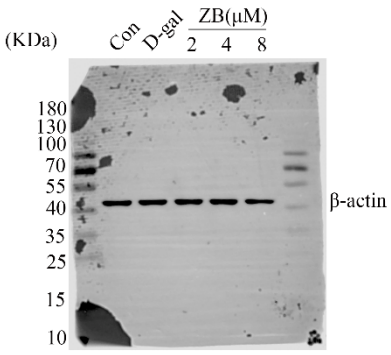

(KDa)
Con
D-gal
D-gal+MEM
D-gal+ZBL
D-gal+ZBH
180
130
100
70
55
40
35
25
15
10
β-actin

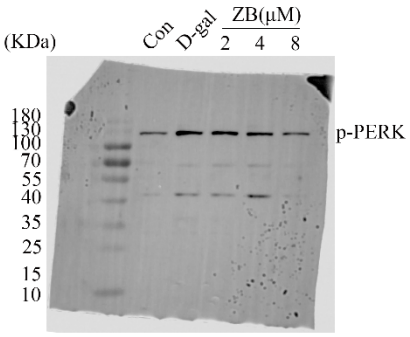

(KDa)
Con
D-gal
D-gal+MEM
D-gal+ZBL
D-gal+ZBH
180
130
100
70
55
40
35
25
15
10
CHOP

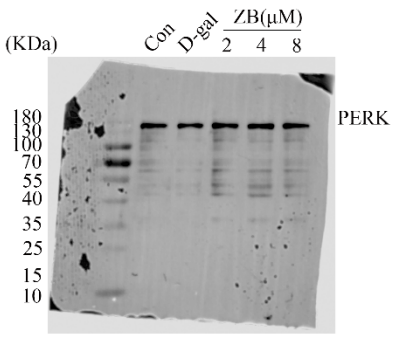

(KDa)
Con
D-gal
D-gal+MEM
D-gal+ZBL
D-gal+ZBH
180
130
100
70
55
40
35
25
15
10
β-actin

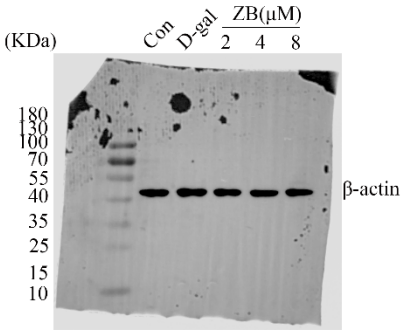
Representative Western blotting images of GRP78, p-PERK/PERK, and CHOP in D-gal-induced SH-SY5Y cells

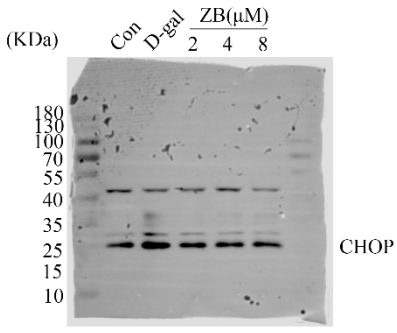












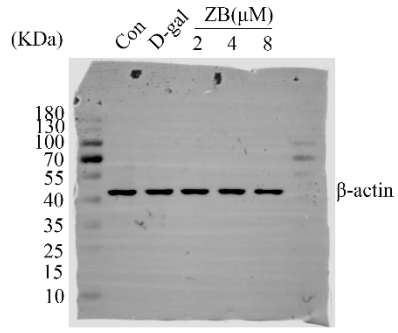
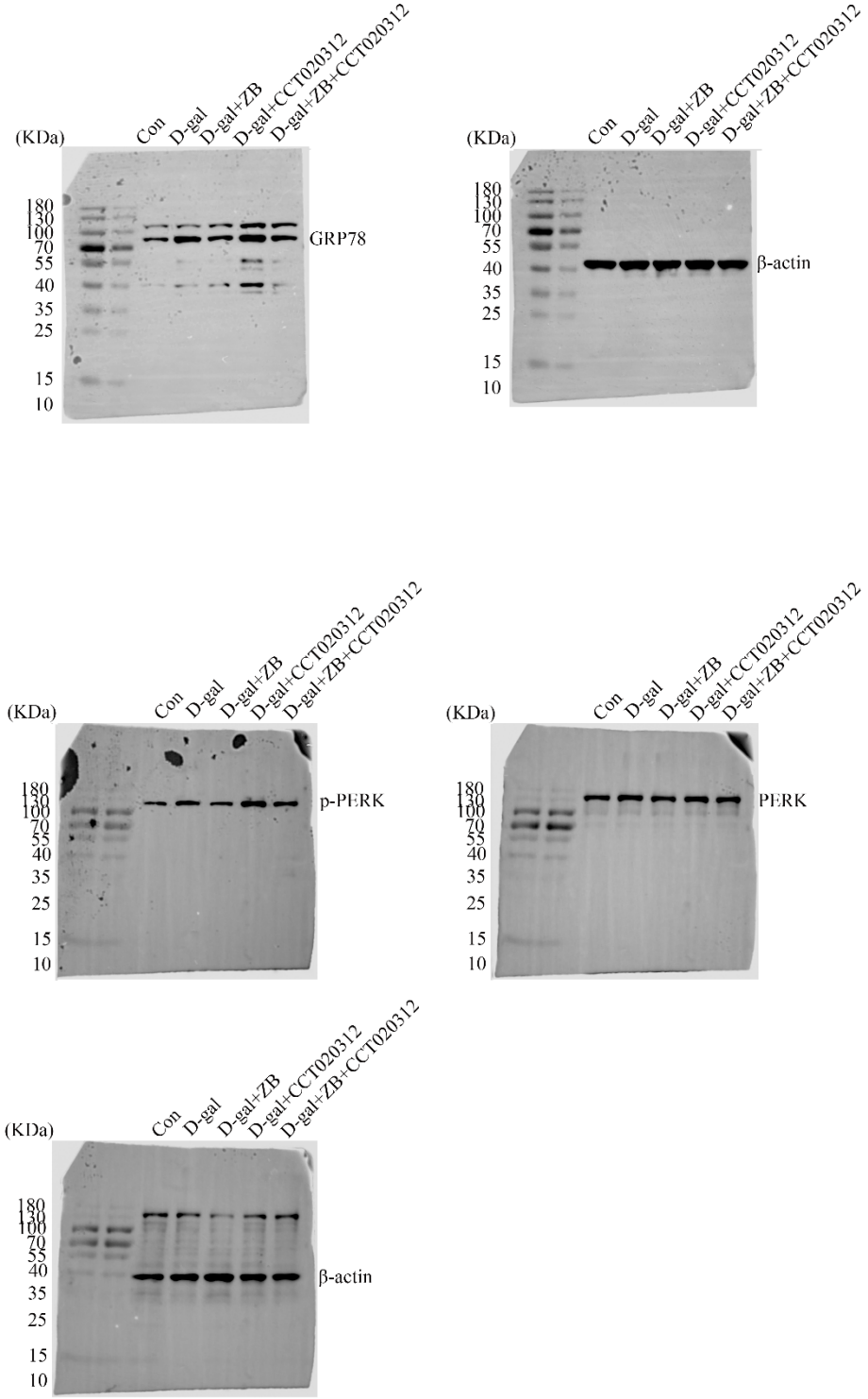
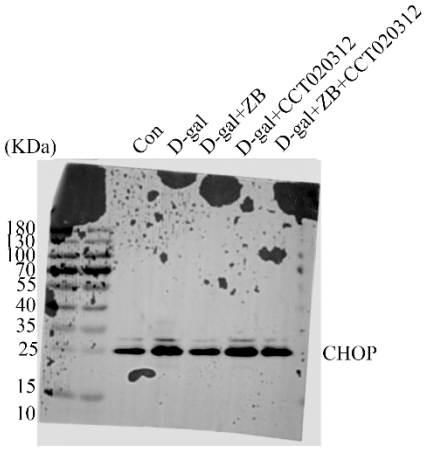
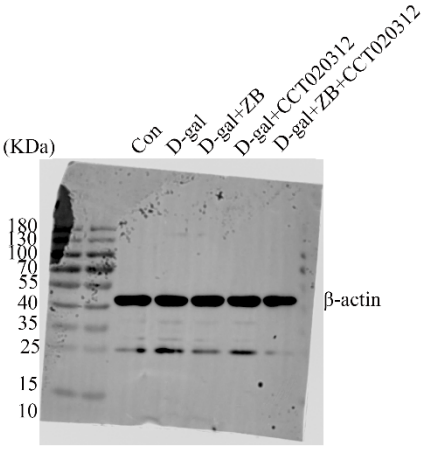
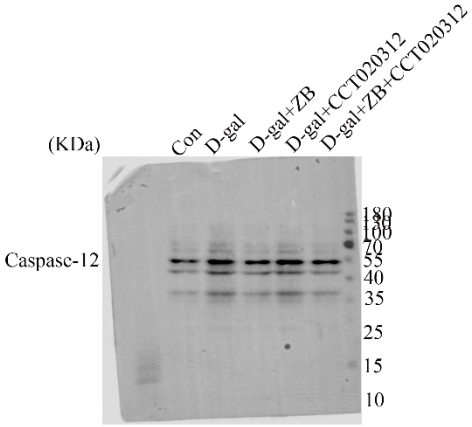


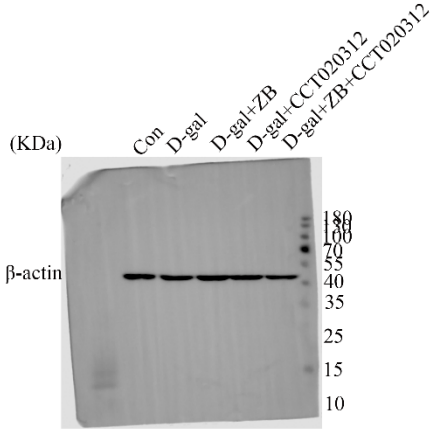
Fig.5. Zerumbone inhibited D-gal-induced ER stress and apoptosis by PERK-dependent pathway in vitro. When CCT020312 (5 μM) and ZB (8 μM) were applied to treat cells together, representative Western blotting images of GRP78, p-PERK/PERK, CHOP, Caspase-12 and Bcl-2/Bax in D-gal-induced SH-SY5Y cells.

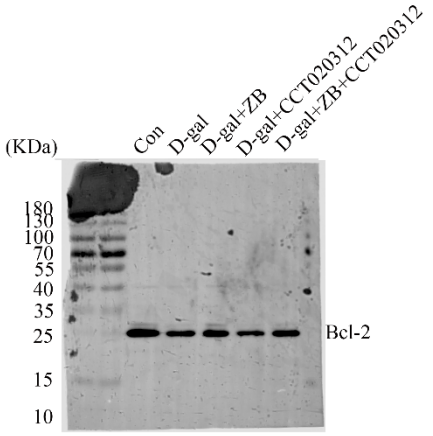


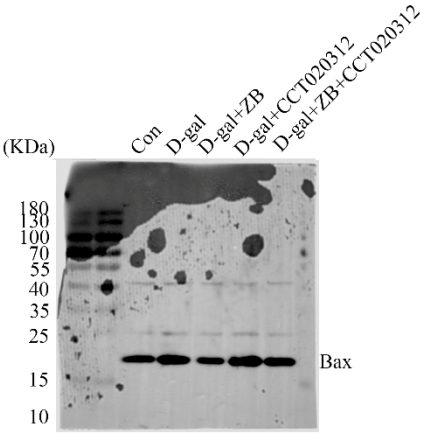

(KDa)
Con
D-gal
D-gal+ZB
D-gal+CCT020312
D-gal+ZB+CCT020312
180
130
100
70
55
40
35
25
15
10
CHOP

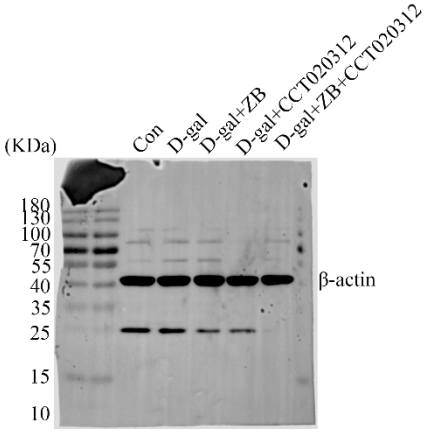

(KDa)
Con
D-gal
D-gal+ZB
D-gal+CCT020312
D-gal+ZB+CCT020312
180
130
100
70
55
40
35
25
15
10
β-actin


(KDa)
Con
D-gal
D-gal+ZB
D-gal+CCT020312
D-gal+ZB+CCT020312
Caspase-12
180
130
100
70
55
40
35
25
15
10

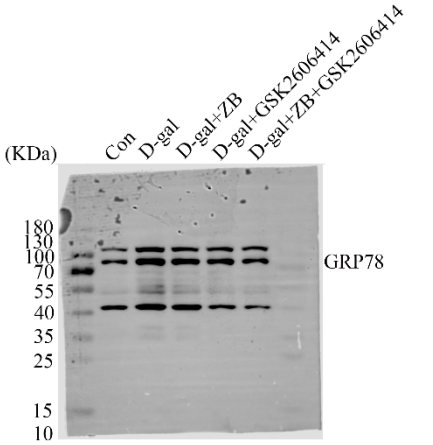

(KDa)
Con
D-gal
D-gal+ZB
D-gal+CCT020312
D-gal+ZB+CCT020312
β-actin
180
130
100
70
55
40
35
25
15
10

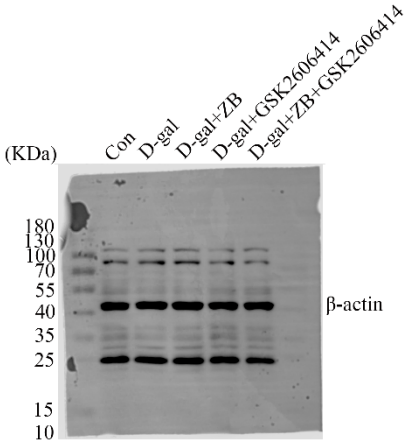


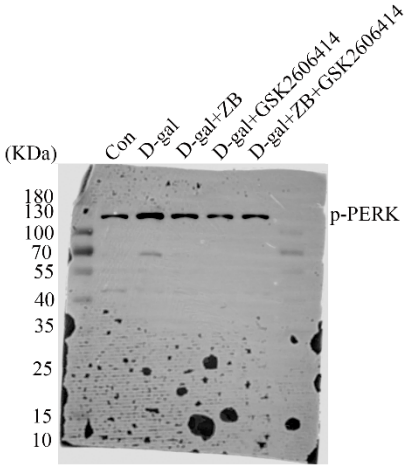


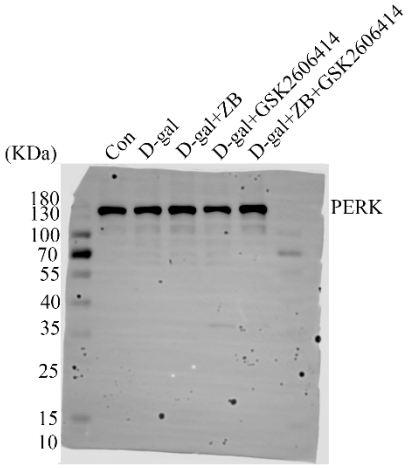


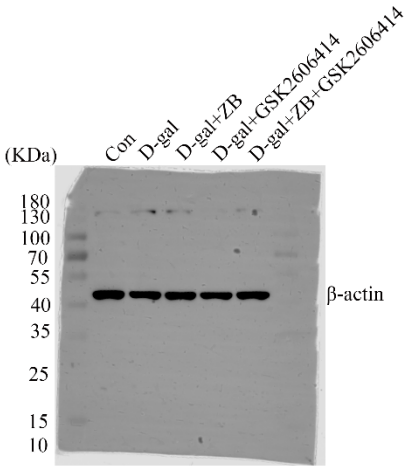
When GSK2606414 (0.25 μM) and ZB (8 μM) were applied to treat cells together, representative Western blotting images of GRP78, p-PERK/PERK, CHOP, Caspase-12 and Bcl-2/Bax in D-gal-induced SH-SY5Y cells.

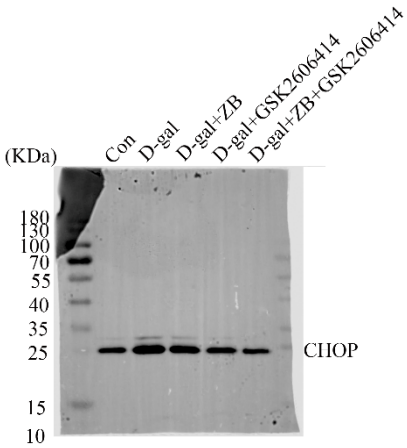


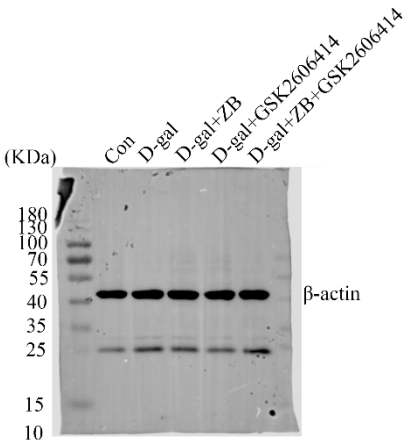


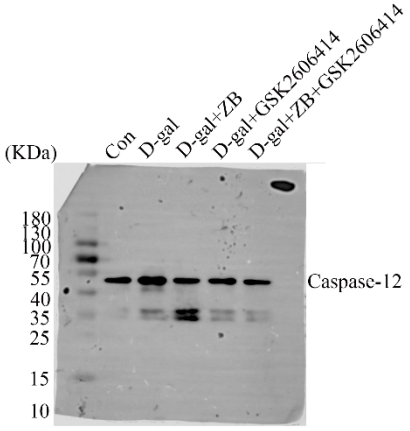

(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
p-PERK

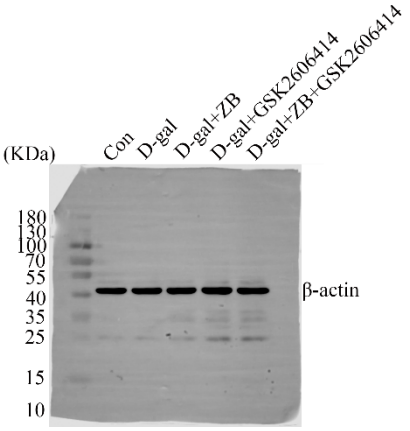

(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
PERK

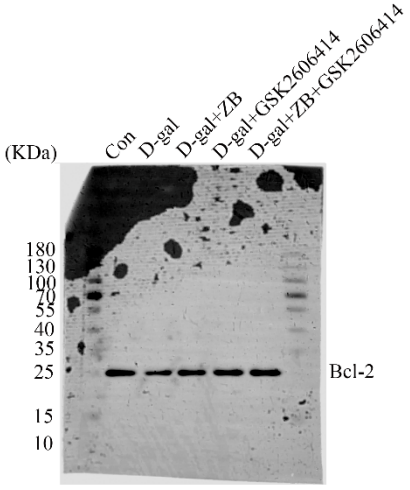

(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
β-actin

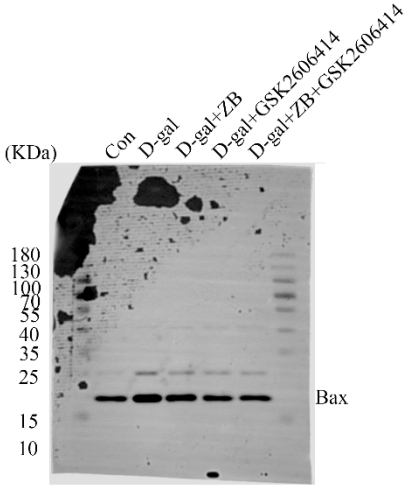

(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
CHOP


(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
β-actin


(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
Caspase-12


(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
β-actin


(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
Bcl-2


(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
Bax

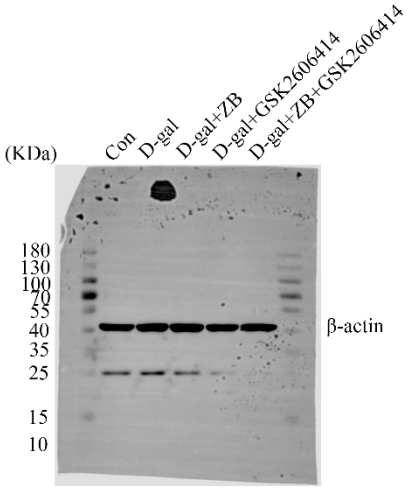

(KDa)
Con
D-gal
D-gal+ZB
D-gal+GSK2606414
D-gal+ZB+GSK2606414
180
130
100
70
55
40
35
25
15
10
β-actin

Fig.6 Calculation of medium efficiency equations and molecular docking

Representative Western blotting images of p-PERK/PERK in D-gal-induced SH-SY5Y cells.

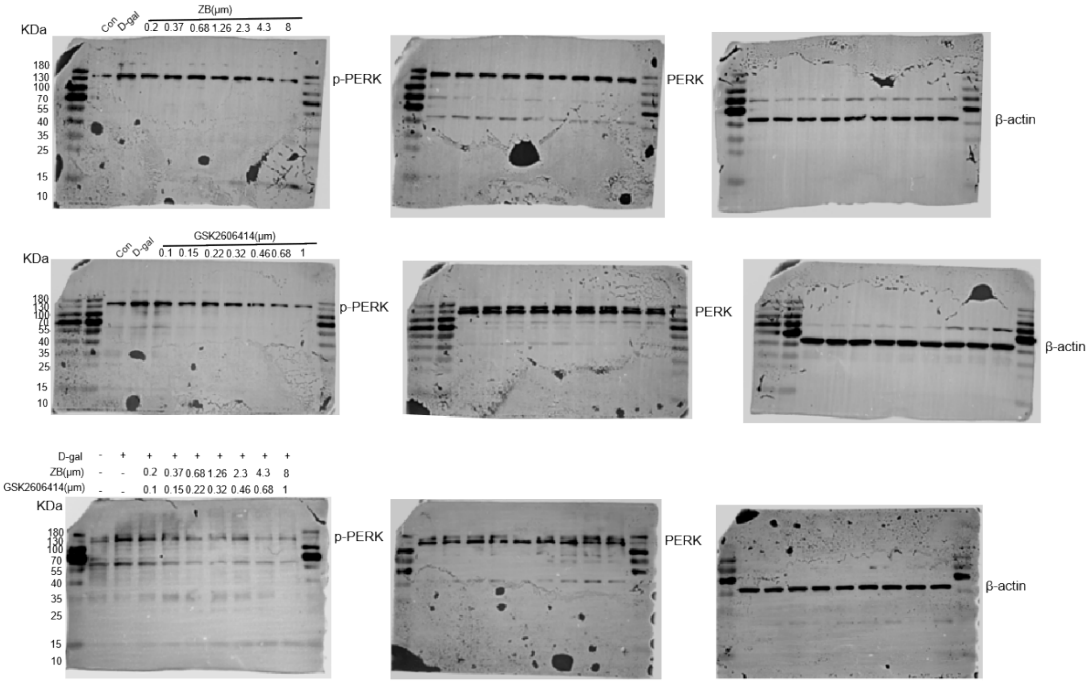
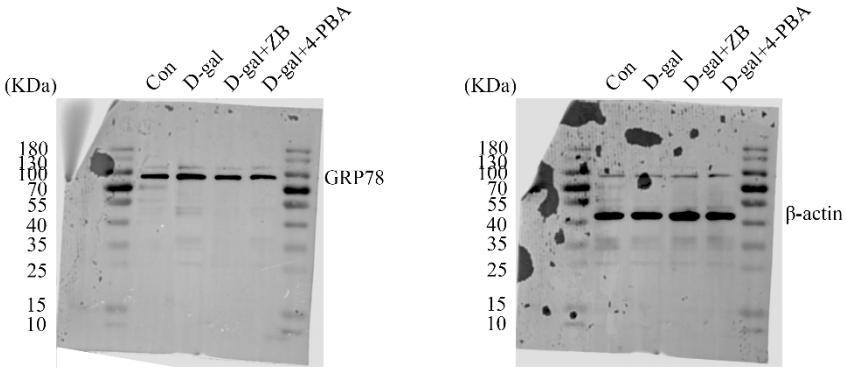
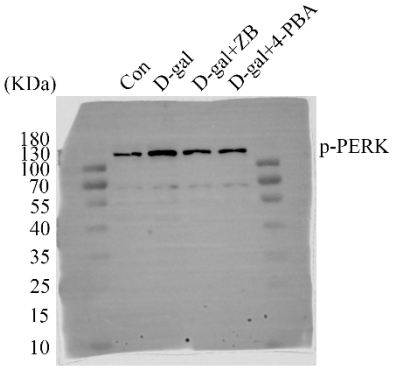
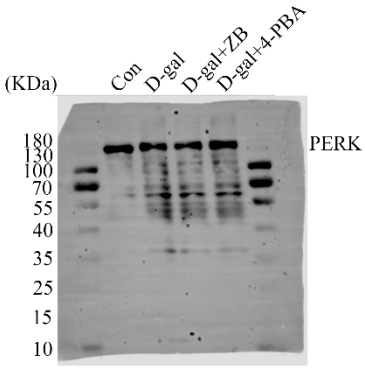


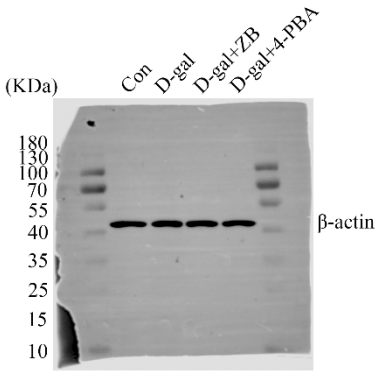
Fig.7. Zerumbone reversed tunicamycin-induced the activation of PERK/CHOP pathway and apoptosis in vitro.

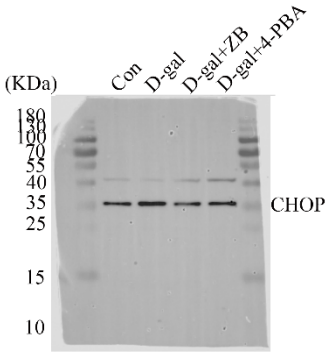
Representative Western blotting images of GRP78, p-PERK/PERK, CHOP, Caspase-12 and Bcl-2/Bax in tunicamycin-induced SH-SY5Y cells.

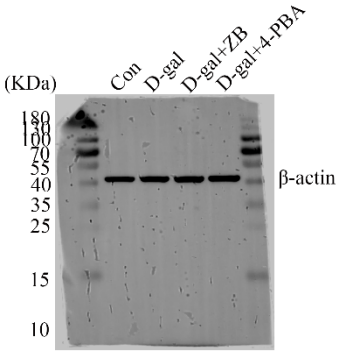


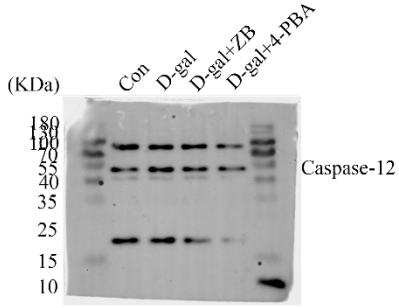

(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
p-PERK

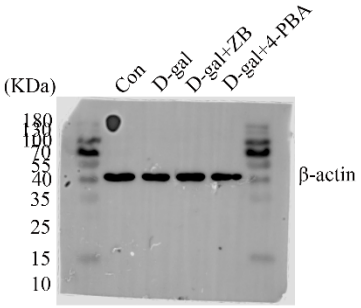

(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
PERK

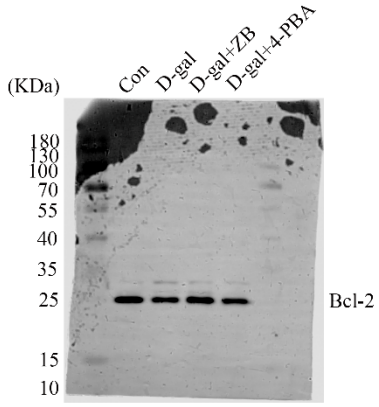

(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
β-actin

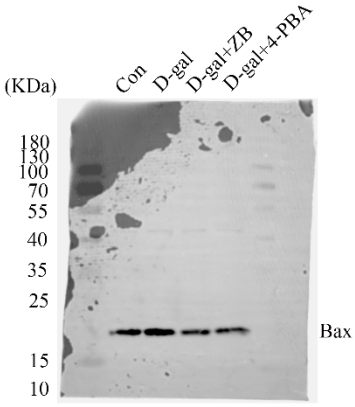

(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
CHOP

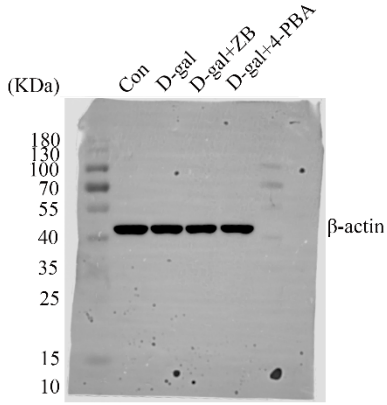

(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
β-actin


(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
Caspase-12


(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
β-actin


(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
Bcl-2


(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
Bax


(KDa)
Con
D-gal
D-gal+ZB
D-gal+4-PBA
180
130
100
70
55
40
35
25
15
10
β-actin