Systems biology network-based discovery of a small molecule activator BL-AD008 targeting AMPK/ZIPK and inducing apoptosis in cervical cancer

Supplementary Material

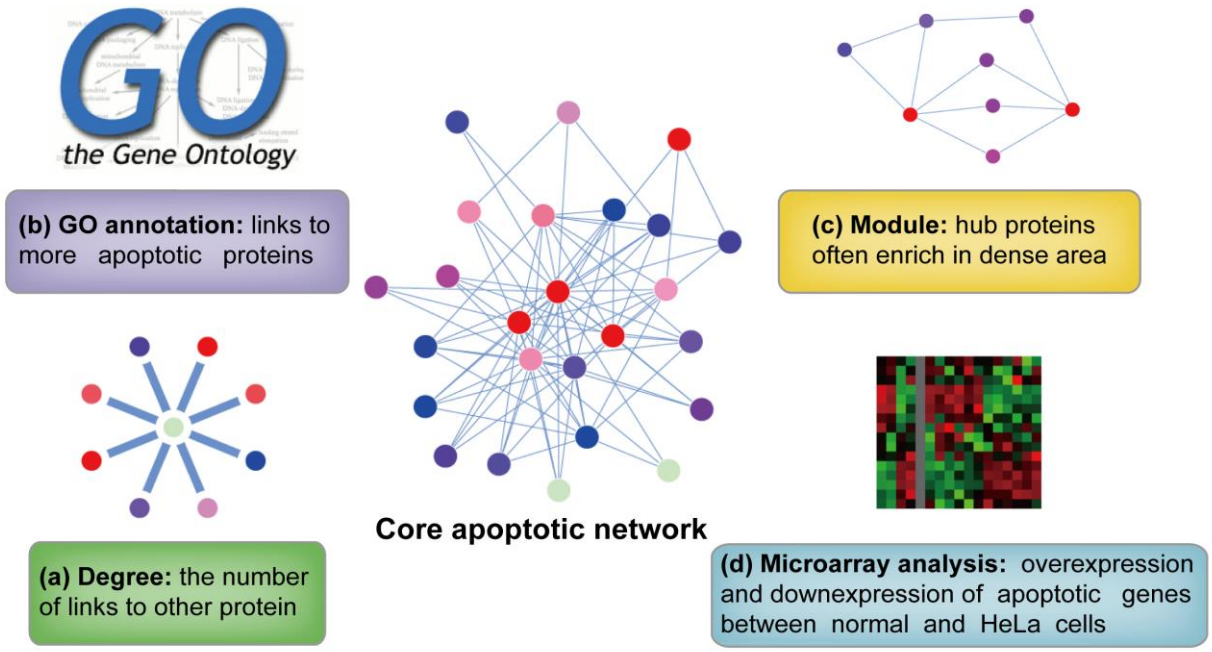


Figure S1: Multiple analyses of the apoptotic hub proteins/targets in cancer.

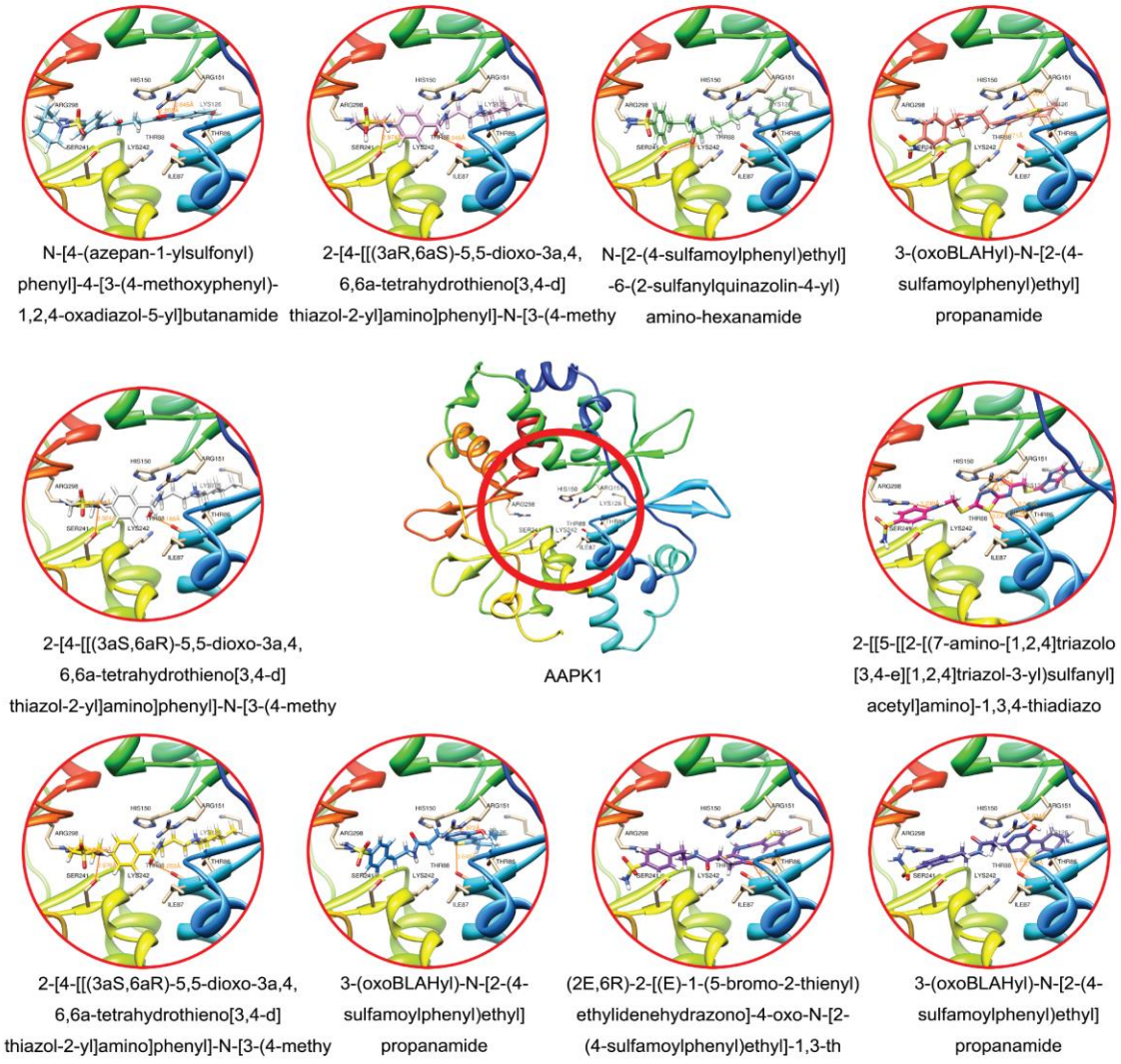


Figure S2: The consensus results of the top ten candidate compounds targeting AMPK from Drugbank and ZINC.

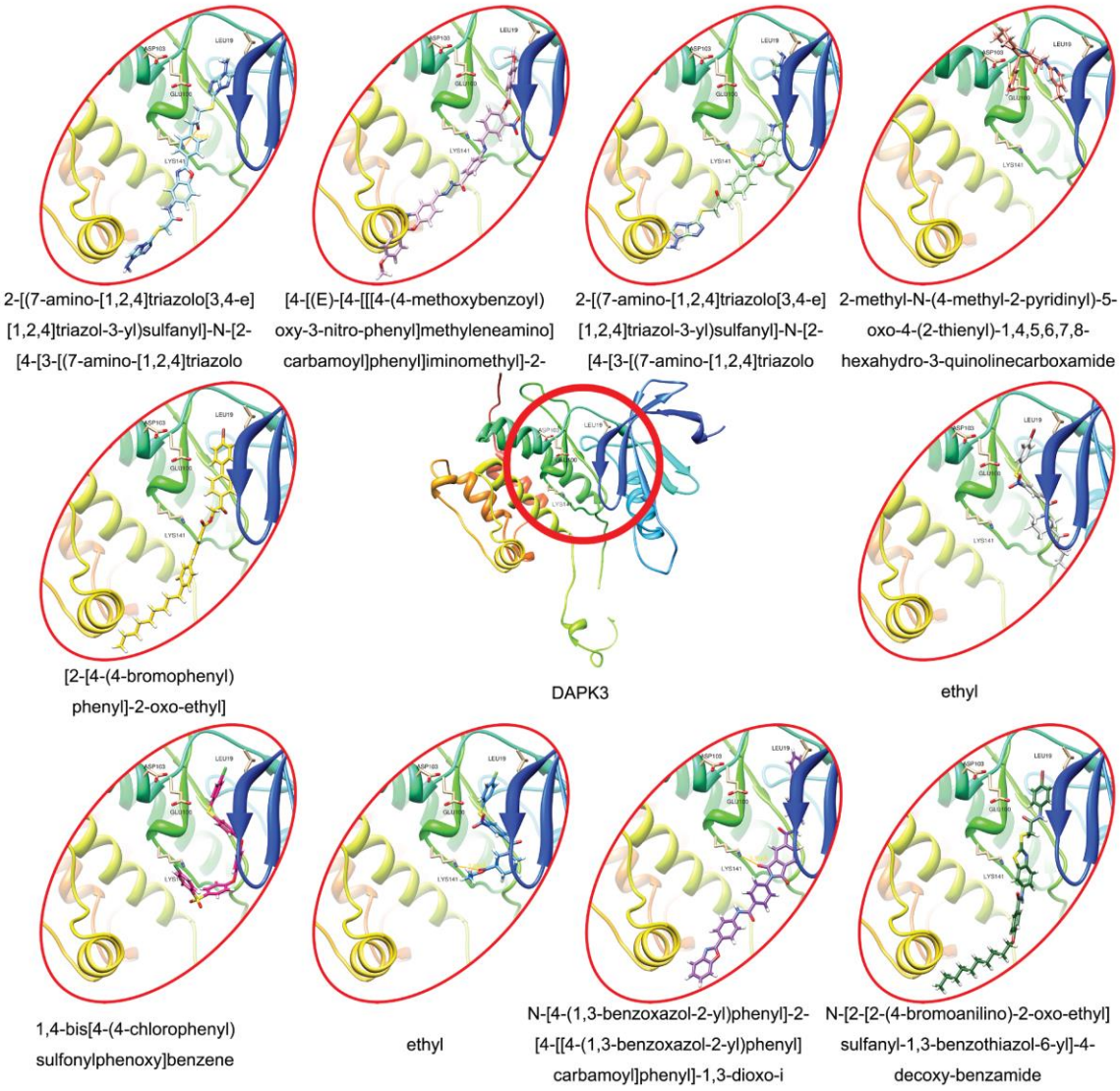


Figure S3: The consensus results of the top ten candidate compounds targeting ZIPK from Drugbank and ZINC.

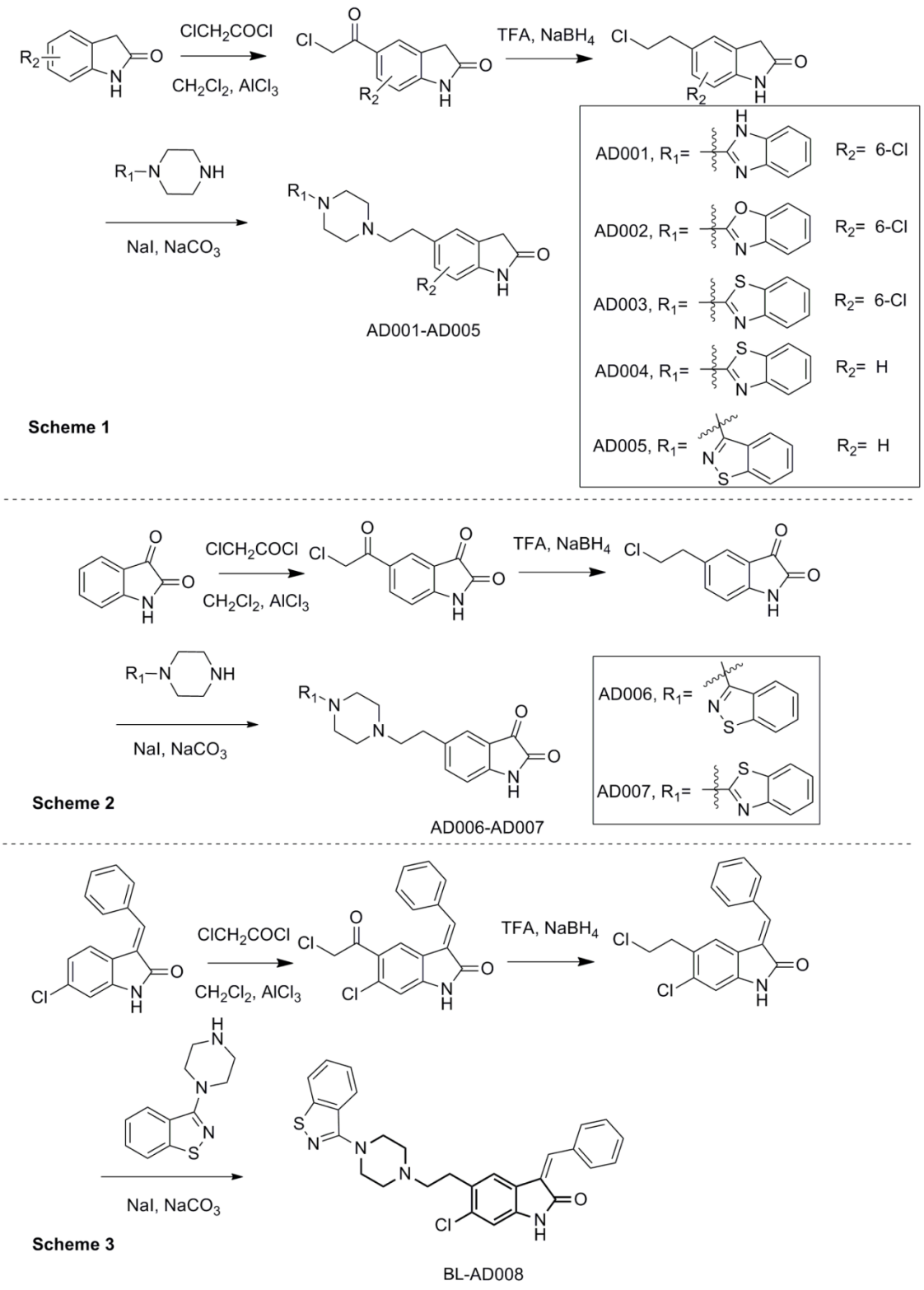


Figure S4: The synthesis of BL-AD008 as a novel dual-target activator.

Table S1: Integration of data sources for predicting human protein-protein interactions.

Prediction type	Dataset name	Time stamping	Element number
Co-expression	Pairwise co-expression set	2014/10/14	8, 030, 028
Enriched domain pair	Domain-domain interaction set	2014/11/17	148, 938
Ortholog interactions	Interolog mapping PPI set	2014/5/10	7, 090
Shared biological process	Shared biological process enrichment set	2014/10/04	31, 318
Gold standard	**Data set**	**Time stamping**	**Element number**
Positive(GSP)	Good standard positive set	2014/10/05	85, 083
	Testing set	2014/5/10	25, 620
Negative(GSN)	Good standard negative set	2014/5/14	23, 169, 177
Raw pair	Raw protein-protein interaction set	2014/5/14	204, 919, 890

Table S2: Protein-protein interactors from core apoptotic subnetwork.

Table S3: The interactors with AMPK in apoptosis.

Table S4: The interactors with ZIPK in apoptosis.

Table S5: Modified candidate small molecule compounds from A1 to A10.

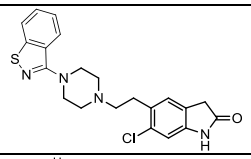
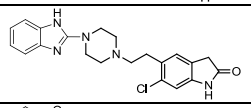
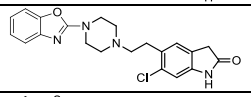
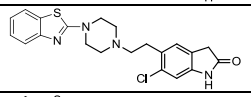
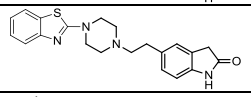
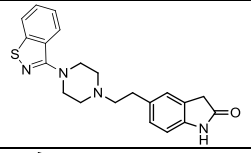
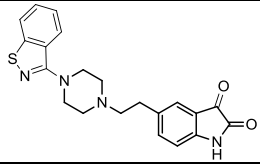
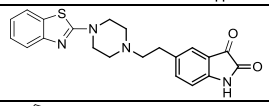
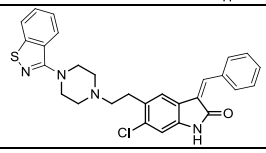
Compound	**IUPAC Name**	**Chemical Structures**
A-1	5-(2-(4-(benzo[d]isothiazol-3-yl)piperazin-1-yl)ethyl)-6-chloroindolin-2-one	
AD-001	5-(2-(4-(1H-benzo[d]imidazol-2-yl)piperazin-1-yl)ethyl)-6-chloroindolin-2-one	
AD-002	5-(2-(4-(benzo[d]oxazol-2-yl)piperazin-1-yl)ethyl)-6-chloroindolin-2-one	
AD-003	5-(2-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)ethyl)-6-chloroindolin-2-one	
AD-004	5-(2-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)ethyl)indolin-2-one	
AD-005	5-(2-(4-(benzo[d]isothiazol-3-yl)piperazin-1-yl)ethyl)indolin-2-one	
AD-006	5-(2-(4-(benzo[d]isothiazol-3-yl)piperazin-1-yl)ethyl)indoline-2,3-dione	
AD-007	5-(2-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)ethyl)indoline-2,3-dione	
BL-AD-008	5-(2-(4-(benzo[d]isothiazol-3-yl)piperazin-1-yl)ethyl)-3-benzylidene-6-chloroindolin-2-one	

Table S6: Binding free energies and individual energy terms of BL-AD008 in complex with both AMPK and ZIPK (kcal/mol).

Contribution	AD008-AMPK	AD008-ZIPK
ΔE_{int}^{ele}	-7.11(0.23)	-0.91(0.04)
ΔE_{int}^{vdw}	-38.04 (0.39)	-31.52(0.40)
ΔG_{sol}^{nopol}	-4.2(0.04)	-3.66(0.05)
ΔG_{sol}^{ele}	21.73(0.27)	12.82(0.56)
ΔG_{sol} [a]	17.53(0.26)	9.16(0.52)
ΔG_{ele} [b]	14.62(0.38)	11.91(0.59)
-TΔS	-12.7	-10.9
ΔG_{bind}	-40.12(0.41)	-34.18(0.34)

[a] The polar/nonpolar ($\Delta G_{sol}^{ele} + \Delta G_{sol}^{nopol}$) contributions.

[b] The electrostatic ($\Delta E_{int}^{ele} + \Delta G_{sol}^{ele}$) contributions. All energies are averaged over 300 snapshots and are given in kcal/mol. Calculation of ΔG_{bind} does not explicitly consider entropy contributions. The values in parentheses represent the standard error of the mean.