Supporting Information

A novel approach for anticancer compounds screening analysis from traditional Chinese medicine by G-quadruplex functionalized magnetic system

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The standard curve. Gradient volume, 2 μl～10 μl, of Berberine reference solution (21.67 μM) was analyzed by HPLC. The equation of the standard curve is $y = 9477 + 853.1x$, (r^2=0.9979). Here x denotes the sample amount (pmol) and y is the chromatographic peak areas.

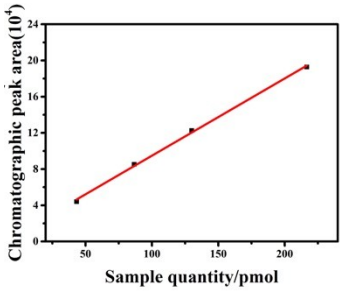

Fig. S1 The standard curve

Calculation of binding constants. As shown in the fluorescence spectrum (Fig. S2a, A), there are no fluorescence emission signal assigned for berberine (1 μM) in the 450-650 nm region. When ***c-myc*** of gradient concentration were added into the pure solution in PBSK, there were new fluorescence signals produced dramatically at 542 nm. The same thing happened with the other alkaloids, no fluorescence emission signal for pure alkaloids but it produced dramatically when ***c-myc*** G-quadruplex (G4) of gradient concentration were added. Their maximum emission wavelength is jatrorrhizine at 533 nm (Fig. S2b, A), palmatine at 540 nm (Fig. S2c, A), coptisine at 559 nm (Fig. S2d, A), columbamine at 535 nm (Fig. S2e, A). The change of spectrum after adding ***c-myc*** in PBSK indicates that all above alkaloids are able to interact with G4.

From the fluorescence titration results, the binding constants (***K_a***) of G4 and berberine (Fig. S2a, B) jatrorrhizine (Fig. S2b, B), palmatine (Fig. S2c, B), coptisine (Fig. S2d, B), columbamine (Fig S2e, B) were calculated via the Job' plots formula are 9.78×10^5, 4.27×10^5, 6.69×10^5, 8.81×10^5, 4.34×10^5, respectively.

$$(F_\infty - F_0)/(F_X - F_0) = 1 + 1/K_a[P] \qquad [1]$$

Where F_∞ is the maximum fluorescence intensity of the alkaloids induced by ***c-myc*** G4; F_0 mean the fluorescence intensity of the G4 binding to alkaloids whose concentration ratio is zero; F_x mean the measured fluorescence intensity; K_a represented binding constant; P represented the concentration of G4.

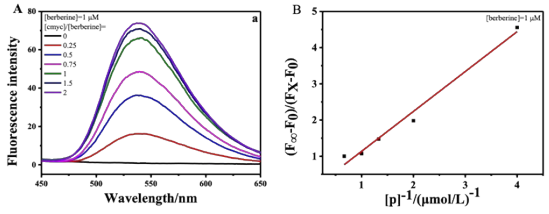

Fig.S2a (A) Fluorescence spectra of *berberine* with titration of ***c-myc*** solution. (B) Binding affinities (***Ka***) of *berbeirne* with G4.

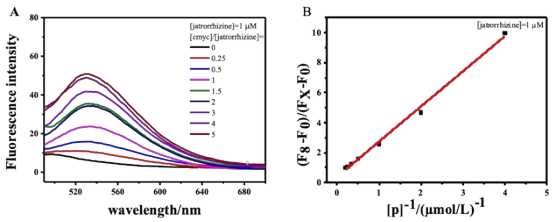

Fig.S2b (A) Fluorescence spectra of *jatrorrhizine* with titration of ***c-myc*** solution. (B) Binding affinities (***Ka***) of *jatrorrhizine* with G4.

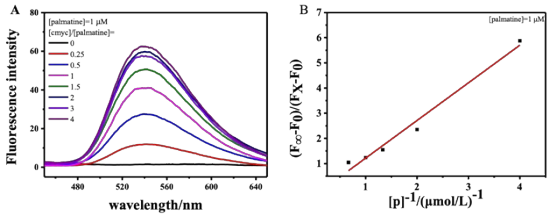

Fig.S2c (A) Fluorescence spectra of *palmatine* with titration of ***c-myc*** solution. (B) Binding affinities (***Ka***) of *palmatine* with G4.

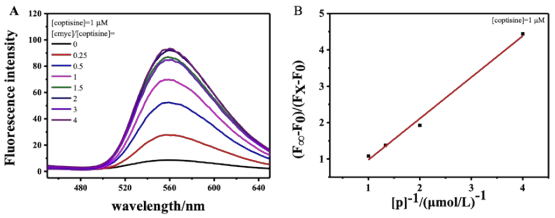

Fig.S2d (A) Fluorescence spectra of *coptisine* with titration of ***c-myc*** solution. (B) Binding affinities (***Ka***) of *coptisine* with G4.

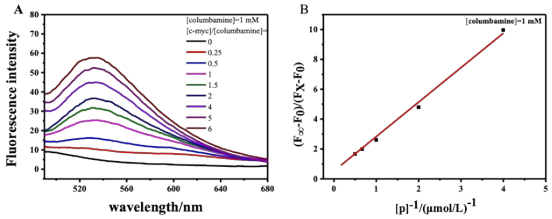

Fig.S2e (A) Fluorescence spectra of columbamine with titration of c-myc solution. (B) Binding affinities (Ka) of columbamine with G4.

Application in *Cortex Phellodendri Chinensis*. *Cortex Phellodendri Chinensis*, another commonly used TCM, was screened by the ***c-myc***-MBs (magnetic microbeads) screening system. As shown in Fig. S3A, the *Rhizoma Coptidis* extract (Fig. S3A, a) and its eluate (Fig. S3A, b) were analyzed by HPLC. There were three compounds identified as columbamine (peak 1), jatrorrhizine (peak 2), berberine (peak 3). The columbamine was too little to calculate. As shown in Fig. S3B, the ranks of ***f*(*Ka*)** and ***f*(*c*)** of the other 2 compounds behaved consistently. The results further verified that the screening efficiency of ***c-myc***-MBs system is relative to ligands' affinity with G4.

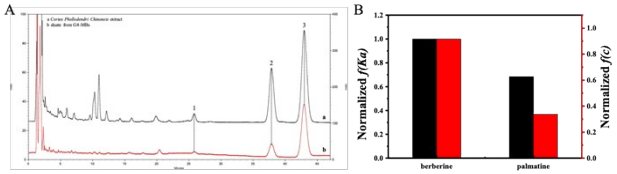

Fig.S3 (A) HPLC spectra of (a)the extract of *Cortex Phellodendri Chinensis*, and (b) the screening eluate from it by ***c-myc***-MBs system. 1 columbamine, 2 jatrorrhizine, 3 berberine. (B) Comparison of the ***f(Ka)*** of three alkaloids, berberine, palmatine, jatrorrhizine, and ***f(c)*** of them screening from the extract of *Cortex Phellodendri Chinensis* by ***c-myc***-MBs system.

The optimization of the incubation time. To optimize the incubation time of under-screen solution with ***c-myc*** G4, the G4 ligand berberine was employed. We tested the amount of the captured solution after the screening process with ***c-myc***-MBs for 5, 15, 30, 120 min, respectively. The results (Figure. S4) show that the absorption intensities of berberine interacting with ***c-myc*** G4 increased with the time extension and the signal got saturated after 30 minutes, so we chose 30 minutes as the incubation time.

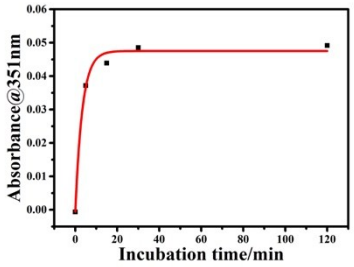

Fig.S4 The change of the absorption intensity of the captured berberine solution with increasing incubation time.