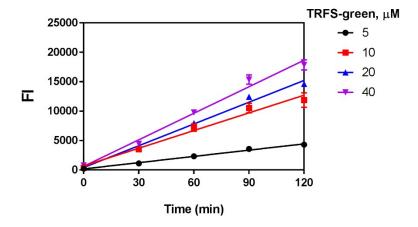
## Effects of TRFS-green concentration on the reduction of TRFS-green by *E. coli* Trx system

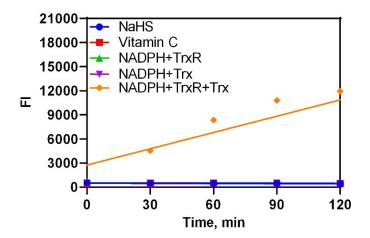
Various concentration of TRFS-green probe was added in the solution containing 200  $\mu$ M NADPH, 50 nM *E. coli* TrxR, 1  $\mu$ M *E. coli* Trx in TE buffer (50 mM Tris-HCl, 1 mM EDTA, pH 7.5). The fluorescent intensity of the solution were measured every 30 minutes for 2 h using the VERSA microplate reader at 37 °C, with the excitation at 438 nm and the emission at 538 nm.



**Fig. S1** Effects of TRFS-green concentration on the reduction of TRFS-green by E. coli Trx system. Data were obtained from three independent experiments and shown as mean  $\pm$  SD.

## Comparison of the reduction of TRFS-green by *E. coli* Trx system with some small reductants

TRFS-Green (10  $\mu$ M) were mixed with 1 mM NaHS (), 1 mM vitamin C, 100 nm *E. coli* TrxR /1  $\mu$ M Trx1 in the presence of 200  $\mu$ M NADPH in TE buffer, separattively. Then the fluorescence intensity of the solution was determined at 37 °C every 30 min for 2 h using the VERSA microplate reader, with the excitation at 438 nm and the emission at 538 nm.



**Fig. S2** Comparison of the reduction of TRFS-green by *E. coli* Trx system with some small reductants including vitamin C, NaHS. Data were obtained from three independent experiments and shown as mean  $\pm$  SD.