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Supplementary Material

For

A dual-fluorescent whole-well imaging approach for screening active compounds against doxorubicin-induced cardiotoxicity from natural products

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S1. Preparing the fractions from ZQFZ.

S2. Cardiopretective effects of fractions from ZQFZ in primary screening.

S3. Dose-response curve of rutin by dual staining

S4.The total ion chromatogram and dose-dependent effects of five active components

1. Preparing fractions from ZQFZ

ZQFZ granules (90g) were dissolved in water (400mL) and centrifuged to obtain aqueous extracts of ZQFZ. The supernatant was loaded onto a glass column (4.6×60 cm) packed with the D101 macro porous resin for chromatographic separation. After the aqueous extracts of ZQFZ was absorbed for 0.5 hour, the column was eluted with H_2O, ethanol-H_2O (40:60), ethanol-H_2O (95:5) at the speed of $2L \cdot h^{-1}$ to obtain crude fractions A02 (12.5g), A03 (5.2g), and A04 (0.7g), respectively. The crude fractions were subjected to preparative HPLC to prepare subfractions.

Preparation of subfractions was performed on a 1200 series LC system (Agilent, Palo Alto, CA, USA) supplemented with a G-1361 preparative dual pump, a G1365Dmultichanneldetector, a Buchi self-motion fraction collector, a UV detector recorded 210, 230, 254 and 280nm, and a Zorbax SB-C_{18} column (21.2 × 250 mm, 7 μm) .The mobile phase consisted of H_2O (phase A) and acetonitrile (phase B) and the flow rate was $12mL \cdot min^{-1}$.

2.0gfraction A03 was dissolved in 6mL 50% ethanol and centrifuged at 10000 rpm. The supernatant was injected to preparative LC. Subfractions were collected every three minutes from 3.5 minute. The elute gradient was as follows: 0-65 min, 5%-20% B; 65-85 min, 20%-30% B; 85-91 min, 30%-95% B. The obtained subfractions were named as fraction **B01-B30**.

0.5g fraction A04 was dissolved in 3mL 70% ethanol and centrifuged at 10,000 rpm. The supernatant was injected to preparative LC. Subfractions were collected every three minutes from 3.5 minute. The elute gradient was as follows: 0-40 min, 20%-35% B; 40-60 min, 35%-95% B; 60-63 min, 95% B. The obtained subfractions were named as fraction **C01-C22**. All subfractions were concentrated to less than 4 mL under reduced pressure, subsequently freeze-dried and stored in -20 ºC until use.

2. Cardiopretective effects of fractions from ZQFZ in primary screening

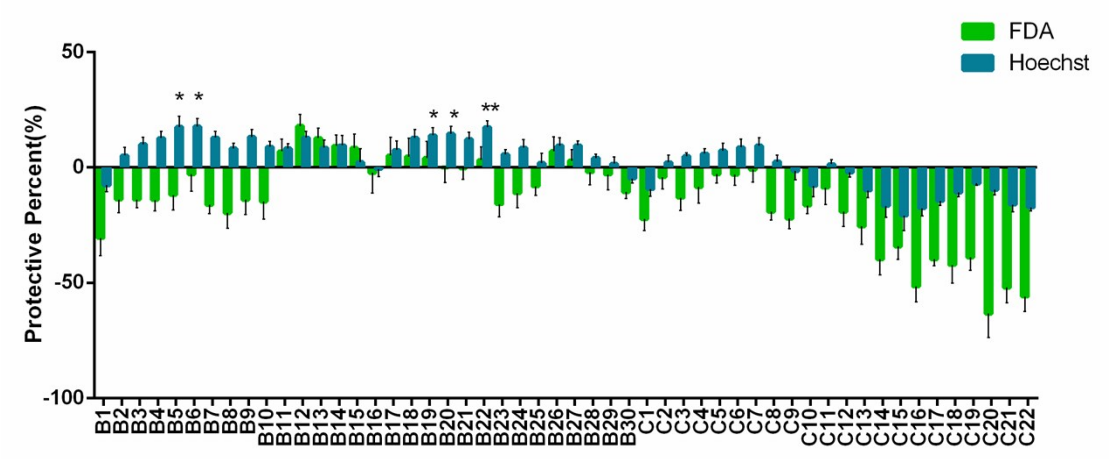

Fig.S1 The cardioprotective effects of fractions isolated from ZQFZ

3. Dose-response curve of rutin by dual staining

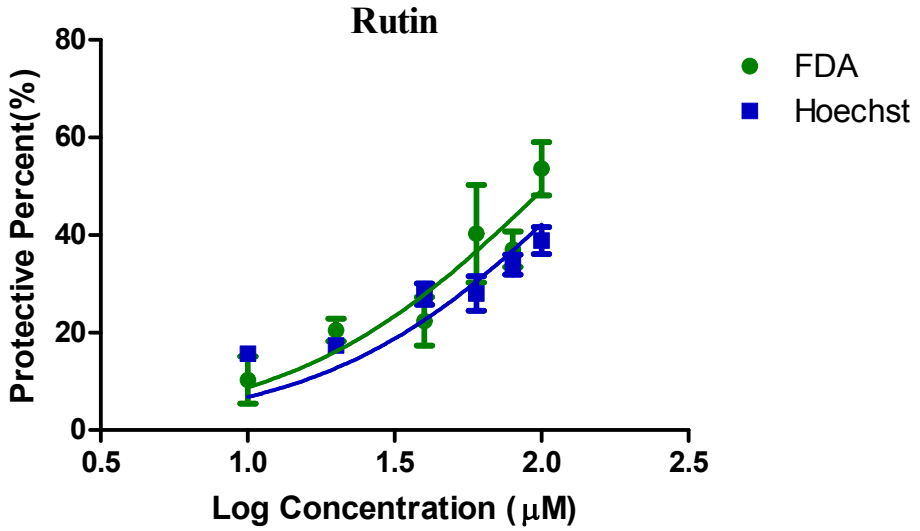

Fig. S2 Dose-response curve of rutin for its cardioprotective effects measured by dual staining of FDA(•) and Hoechst(■) .

4. The total ion chromatogram and dose-dependent effects of five active components

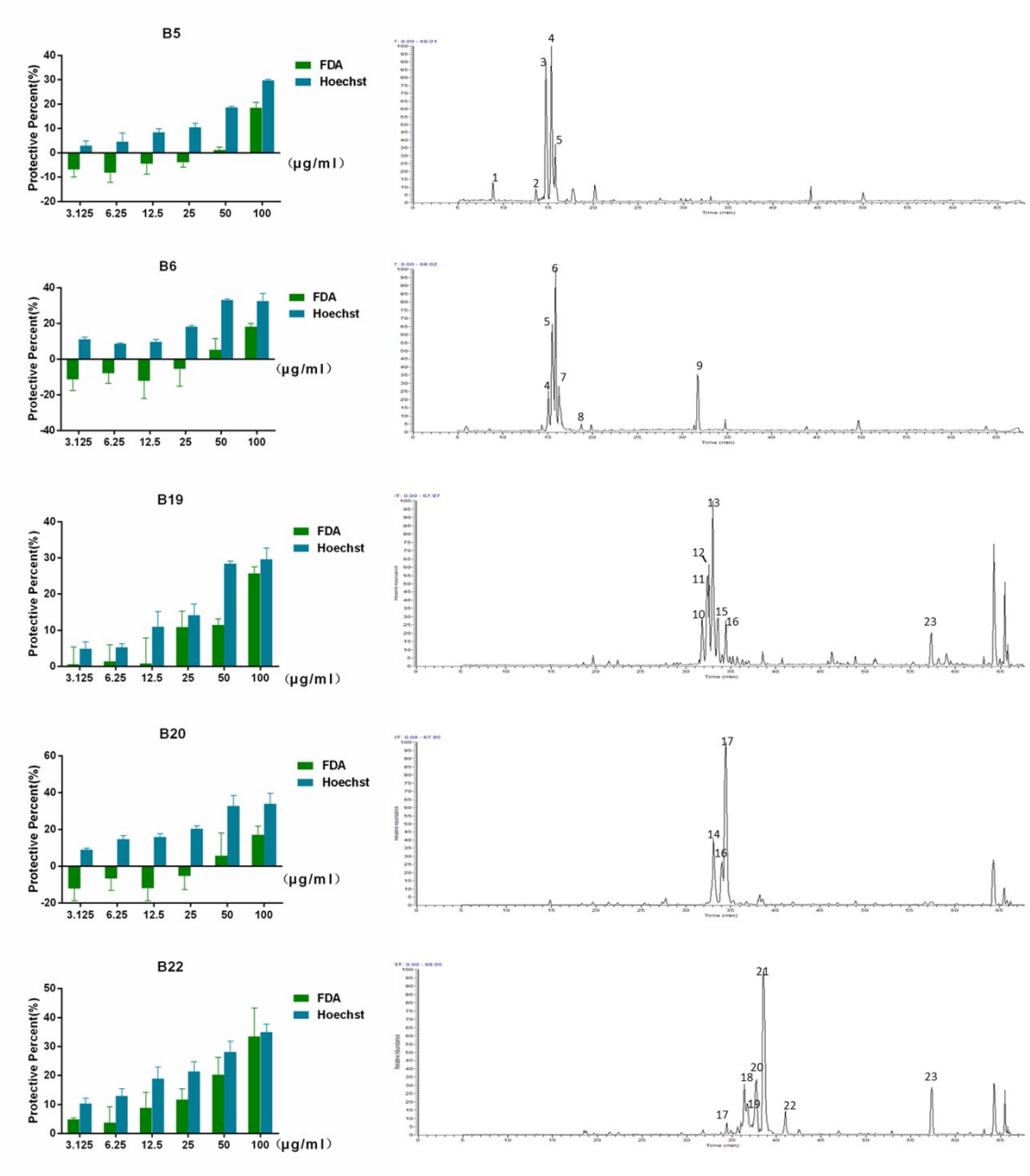

Fig. S3 The total ion chromatogram of five active components from ZQFZ using LC-MS in negative ion mode and their dose-dependent effects