SI methods

Detection of DHE through blood brain barrier

The six male adult SD rats (200-220 g) were intraperitoneally injected with DHE for 1 hour, and then the rats were anesthetized with 4% chloral hydrate. The cerebrospinal fluid was collected from the cerebellomedullary cistern by puncturing the foramen magnum and the cerebrospinal fluid volume between 50-100 μl. Next, the cerebrospinal fluid by adding 2 times the volume of acetonitrile-grade chromatography was centrifuged 20 min at 20,000 g centrifugal force after vortex, and the supernatant was taken out in 1.5 ml EP tube in the nitrogen blowing instrument on the dry, then the dried sample was reconstituted by mobile phase with a volume of 50 μl. The reconstitution ratio was acetonitrile: pure water = 4:1. Finally, the reconstituted sample and a certain concentration of DHE standard solution were analyzed by LC-MS/MS. Analytical method: The Agilent 1200 high-performance liquid chromatography (HPLC) system consisted of a quaternary delivery system, a degasser, an auto-sampler and an UV detector. The chromatograph was equipped with an Elite Hypersil ODS (2.1×150 mm, 5 μm) analytical column. The mobile phase consisted of acetonitrile and 0.1% formicacid in aqueous solution at a flow rate of 0.5 ml/min. An Applied Biosystems MDSSciex API3200 Triple Quadrupole Mass Spectrometer (MS/MS) equipped with an electrospray ionization (ESI) source was used to analyze potential metabolites, and the system was operated in negative mode for DHE (Q1-Q3: 231.4-185.2).

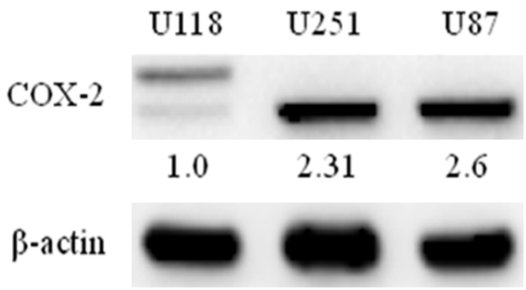

Figure S1. The expression of COX-2 protein in the glioblastoma cells. The expression of COX-2 protein in the U118, U251 and U87 cells were evaluated by western blot. One representative immunoblot of three independent experiments is shown. The β-actin served as the protein loading control. Densitometric analysis of COX-2 protein are quantified by Image J software.

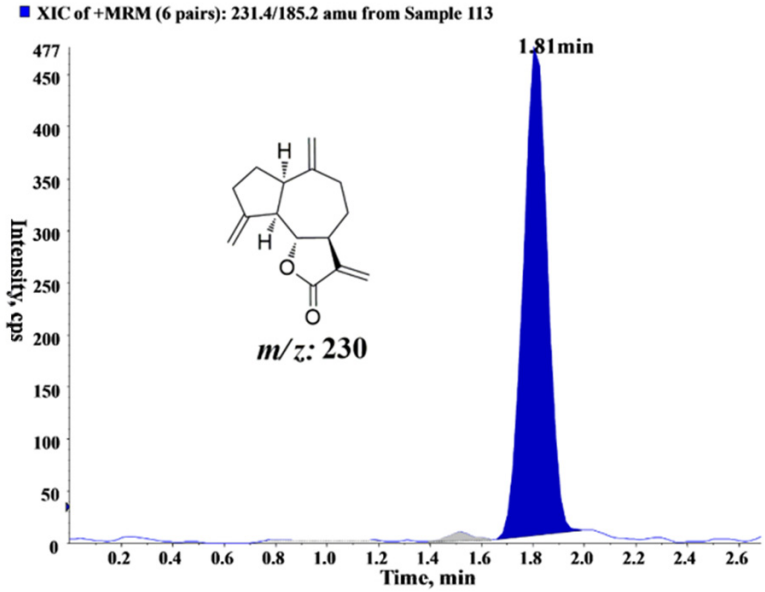

Figure S2. DHE can penetrate the blood brain barrier. LC-MS spectrum of cerebrospinal fluid sample on SD rat model after pretreatment with DHE (1 h). The cerebrospinal fluid samples were prepared as described in "Materials and Methods" section.