BCH-499

Analysis of lipid peroxidation in tissues

Background:

Lipid peroxidation is a free radical-induced process leading to oxidative deterioration of polyunsaturated lipids resulting in cellular damage. The reaction of malondialdehyde (MDA) with thiobarbituric acid (TBA) to form a colored complex that can be quantified spectrophotometrically is the basis of commonest method for the assessment of lipid peroxidation in biological materials.

Reagents:

Potassium chloride

Trichloroacetic acid

Thiobarbituric acid

Tetraethoxypropane

Method:

Tissues are weighed and homogenized (10% w/v) in 0.15 M potassium chloride in a homogenizer using a motor-driven Teflon pestle. Tissue homogenate (1 mL) is incubated at 37°C in a shaker for 1 h. One milliliter of 10% (w/v) trichloroacetic acid is mixed with homogenate followed by centrifugation at 3000 rpm for 10 min. Aliquots (1 mL) of the clear supernatant are mixed with 1 mL of 0.67% (w/v) 2-thiobarbituric acid and placed in a boiling water bath for 10 min, cooled and diluted with 1mL distilled water. The absorbance of solution is recorded at 535 nm, and the concentration of MDA is calculated using tetraethoxypropane as an external standard.