

## Product Information

### Arachidonic acid

Product Number **A 9673**  
Storage Temperature -0 °C

#### Product Description

Molecular Formula: C<sub>20</sub>H<sub>32</sub>O<sub>2</sub>  
Molecular Weight: 304.5  
CAS Number: 506-32-1  
Density: 0.92 g/ml  
Molarity: 3.02 M (based on a density of 0.92 g/ml and a molecular weight of 304.5)  
Melting point: -49 °C<sup>1</sup>  
Boiling point: 163 °C<sup>1</sup>

Many of the lipids that act as second messengers in cell signaling pathways are derived from arachidonic acid. Arachidonic acid is an ω<sub>6</sub>-unsaturated fatty acid that is a constituent of cellular membranes. It is esterified to the *sn*-2 position of glycerophospholipids. Arachidonic acid and its metabolites play important roles in a variety of biological processes, including signal transduction, contraction, chemotaxis, cell proliferation and differentiation, and apoptosis. Under normal conditions, the concentration of free, non-esterified arachidonic acid is virtually undetectable and its release is under tight metabolic and physiological regulation. Activation of phospholipases such as phospholipase A<sub>2</sub> releases arachidonic acid from membrane phospholipids making it available for oxidative metabolism by several enzyme systems including cyclooxygenases, lipoxygenases, and cytochrome P450 enzymes. Free arachidonic acid can regulate specific cellular processes, including modulating calcium flux and activating certain kinases and lipases. Arachidonic acid is the precursor for the biosynthesis of all the eicosanoid messengers, such as prostaglandins, thromboxanes, leukotrienes, lipoxins, and hydroxyeicosatetraenoic acids (HETEs).

Polyunsaturated fatty acids (such as arachidonic acid) autooxidize by three competing pathways.<sup>2</sup> After formation of a peroxy radical, the following can occur:

1. abstraction of hydrogen atoms to give hydroperoxide products,
2. beta-scission of the carbon-oxygen bond to give back carbon radicals, including isomerized carbon radicals,
3. cyclizing to give a cyclic peroxy radical.

A procedure for determination of the amount of oxidation in a lipid using the oxidation index has been published.<sup>3</sup> The oxidation index is the ratio of the absorbance at 233 nm to the absorbance at 215 nm. The latter wavelength was chosen since there is little contribution of the fatty acid carbonyl to the absorbance at this wavelength, thus allowing Beer's Law to be followed.

Other useful references describe lipid oxidation<sup>4</sup> and analytical methods to monitor oxidation.<sup>5</sup>

#### Precautions and Disclaimer

For Laboratory Use Only. Not for drug, household or other uses.

#### Preparation Instructions

Arachidonic acid is soluble in either ethanol, DMSO, or dimethyl formamide at 100 mg/ml and in chloroform or methanol at 50 mg/ml. Arachidonic acid is sparingly soluble in neutral buffers. Arachidonic acid is soluble at 10 mg/ml in 98% ethanol.<sup>7</sup> Solubility of 109 mg of arachidonic acid in 1 ml absolute ethanol gave a clear, colorless solution, but further dilution with 9 volumes of phosphate buffered saline gave a white, opaque suspension.

#### Storage/Stability

Arachidonic acid, diluted in either ethanol, DMSO, or dimethyl formamide, is stable for up to 6 months when stored at -20 °C when purged and stored with an inert gas such as argon or nitrogen. Aqueous solutions of arachidonic acid should be used within 12 hours. Although aqueous solutions may be stable for more than 12 hours, it is recommended to make fresh preparations each day. Purging solutions with an inert gas will prolong stability.

### Procedure

Arachidonic acid can be transesterified to the methyl ester for subsequent analysis by capillary gas chromatography.<sup>6</sup> The prepared sample is promptly injected into a capillary GC, usually approximately 20 µg in a 1 µl volume. Typical capillary column conditions are: Capillary column: 30 meters, 0.25 mm ID, SP2330 phase. Temperature: injector, 225 °C; detector, 250 °C. Column temperature program: 192 °C to 220 °C at 4 °C per minute. Initial temperature held zero minutes, final temperature held 4 minutes.

### References

1. The Lipid Handbook, 2nd edition, Gunstone, F. D., et al., Chapman and Hall (London, UK), dictionary section p 106.
2. Fox, J., Fatty acids' spontaneous oxidation clarified. Chemical and Engineering News, pages 18-19 (1981).
3. Klein, R. A., The detection of oxidation in liposome preparations. Biochim. Biophys. Acta., **210(3)**, 486-489 (1970).
4. Frankel, E. N., Lipid oxidation: mechanisms, products and biological significance. J. Am. Oil Chem. Soc., **61(12)**, 1908-1917 (1984).
5. Kim, R. S., and LaBella, F. S., Comparison of analytical methods for monitoring autooxidation profiles of authentic lipids. J. Lipid Res., **28(9)**, 1110-1117 (1987).
6. Metcalfe, L. D., and Wang, C. N., Rapid preparation of fatty acid methyl esters using organic base-catalyzed transesterification. J. Chromatogr. Sci., **19**, 530-535 (1981).
7. Prostaglandins and Related Substances; A Practical Approach, Benedetto, C., et al., IRL Press (Washington, DC), p 158.

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