

## Product Information

### Cytochrome c from equine heart

Product Number **C2867**

Storage Temperature  $-20\text{ }^{\circ}\text{C}$

CAS<sup>#</sup> 9007-43-6

Synonym: Ferricytochrome c (oxidized state)

#### Product Description

Cytochrome c is an electron-carrying mitochondrial protein. It is a heme protein containing a single polypeptide chain and a single heme group, which is covalently attached to Cys<sup>14</sup> and Cys<sup>17</sup>. The ready fluctuation of cytochrome c within the cell between the ferrous and ferric states, makes it an efficient biological electron-transporter and it plays a vital role in cellular oxidations in both plants and animals. It is generally regarded as a universal catalyst of respiration, forming an essential electron-bridge between the respirable substrates and oxygen.

This cytochrome c product is prepared from equine heart using trichloroacetic acid by a modification of a published method.<sup>1</sup> Alternatively, acetic acid may be used to prepare cytochrome c (Product No. C7752). The trichloroacetic acid method may reduce the amount of superoxide dismutase (SOD) present, but tends to cause dimerization or acid-modified structures of cytochrome c. In contrast, acetic acid preparations may have slightly higher amounts of SOD, but a lower proportion of dimeric cytochrome c.

The product is supplied as a lyophilized powder. The final step before lyophilization is extensive dialysis against 6 mM ammonium hydroxide, which is volatile under lyophilization conditions, so the final product should not contain any buffer salts. The product is mainly the oxidized form of the protein. The reduced form of cytochrome c can be prepared with either sodium dithionite or sodium ascorbate, followed by gel filtration.<sup>2</sup>

Molecular mass:<sup>3</sup> 12,384 Da This MW is confirmed by analysis of representative lots by MADLI-TOF mass spectrometry.

Isoelectric point (pI):<sup>4</sup> range of 10.0 – 10.5

Spectral properties:<sup>5</sup>

$\lambda_{\text{max}} = 550\text{ nm}$  (reduced form)

$E^{\text{mM}} = 29.5$

(reduced form, 0.1 M phosphate buffer, pH 6.8)

$E^{\text{mM}} = 8.4$

(oxidized form, 0.1 M phosphate buffer, pH 6.8)

Purity:  $\geq 99\%$  (SDS-PAGE) Purity confirmed on representative lots by HPLC-electrospray MS.

Purity:  $\geq 95\%$  (spectral assay)

#### Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

#### Preparation Instructions

Cytochrome c is soluble in water or buffered solutions at pH  $7.0 \pm 1.0$  (up to 200 mg/ml), yielding a clear, dark red solution. For general purposes reconstitution is done at 10 mg/ml.

#### Storage/Stability

Store cytochrome c at  $-20\text{ }^{\circ}\text{C}$ . The product, as supplied, is stable for 5 years.

#### Recommended storage time for aqueous solutions:

Storage at  $-20\text{ }^{\circ}\text{C}$  (freezer) – 6 months

Storage at  $2-8\text{ }^{\circ}\text{C}$  (refrigerator) – 2 weeks

Storage at  $20-25\text{ }^{\circ}\text{C}$  (ambient temperature) – 3 days

#### References

1. Hagihara, I. *et al.*, *Biochem. Prep.*, **6**, 1 (1958).
2. Dixon, H.B., and McIntosh, R., *Nature*, **213(74)**, 399-400 (1967).
3. *Handbook of Biochemistry* (CRC Press, 1968).
4. *Brain Research*, **153**, 477-493 (1978).
5. Van Gelder, B.F., and Slater, E.C., *Biochim. Biophys. Acta*, **58**, 593-595 (1962).

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