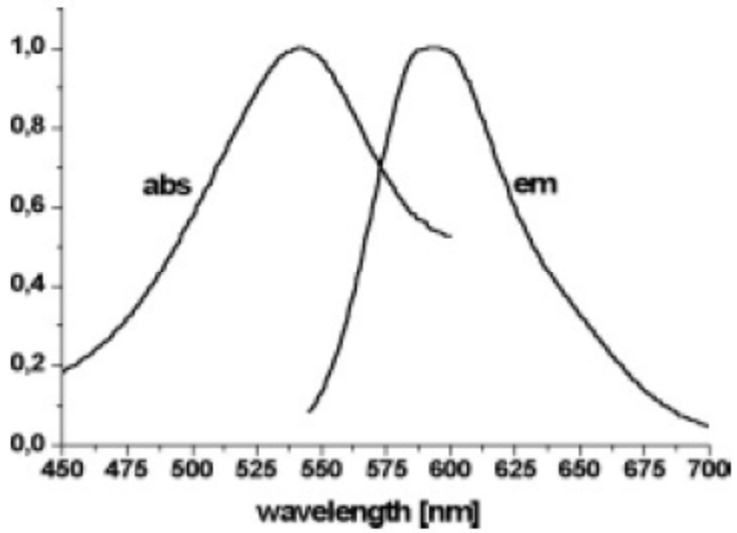
72701 Chromeo™ P540

Chemical Properties:

CONTENTS: Supplied as a 1 mg lyophilized blue solid.
Soluble in DMF, methanol and acetonitrile.
MW: 393.29; m.p. >300 °C

Absorption and emission spectra of Chromeo P540 protein-conjugate



Fluorescent Properties:
Chromeo P540 detects proteins and peptides by exhibiting a color change from blue to red upon binding to primary amines. On conjugation to the primary amino groups, the label undergoes a shortwave spectral shift of more than 100 nm.
Chromeo P540 displays a weak fluorescence with a quantum yield below 1 % in solution. On conjugation to the amine, the quantum yield rises to 40 %. This property allows a distinct detection of primary amines, proteins and other bio-molecules.

Absorption:	590 nm (free), 542 nm (conjugated)
Emission:	627 nm (free), 594 nm (conjugated)
e **L/(mol·cm):**	54 000 (free) 31 000 (conjugated) both in methanol, 25 000 (conjugated) in aqueous solution
Quantum Yield:	< 1 % (free), ~ 40 % (conjugated)

Quality Control:
The Dye has been quality tested by conjugation to BSA and spectro-photometrical evaluation.

Storage and Guarantee:
To ensure stability, the lyophilized dye should be stored at 4°C in the dark. This product is guaranteed for 6 months from the date of arrival.

Protocol for labeling proteins with Chromeo P540 Preparation of the working solution
Dissolve 1 mg of Chromeo P540 in 100 µl of dimethylformamide (DMF). Do not use amine-containing solutions or buffers as solvent. The stock solution can be stored in the dark at 4°C for 6 months.

Labeling reaction
Dissolve 2 mg of HSA (or another protein) in 0.5 ml of bicarbonate buffer (0.1 M, preferably of pH 9.0) and add 5-10 µl of the working solution drop-wise to the protein solution. Gently stir the reaction mixture at room temperature for 30 minutes. The reactive dye in solution is blue. The blue colour disappears and becomes yellow, when the dye is stored in basic solution.

Bicarbonate buffer, pH 9.0
2.1 g of NaHCO3 are dissolved in 500 ml doubly distilled water. The buffer is adjusted to pH 9.0 with 1 N NaOH.

Purification of the conjugated protein.
For some applications the purification of the dye conjugated protein may be necessary. The labeled protein is purified by size-exclusion chromatography using Sephadex G25 as stationary phase and phosphate buffer of pH 7.2 (22 mM) as the eluent. The red band indicates the labeled protein.

Phosphate buffer, (22 mM) pH 7.2
5.67 g Na2HPO4 x 12 H2O and 0.96 g NaH2PO4 x 2H2O are dissolved in 1 L of ddH2O. The buffer is adjusted with 1 N HCl to pH 7.2.

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.