



## Product Information

### Bisbenzimidazole H 33258

Product Number **B2883**

Storage Temperature -0 °C

#### Product Description

Molecular Formula: C<sub>25</sub>H<sub>24</sub>N<sub>6</sub>O 3HCl

Molecular Weight: 533.9

CAS Number: 23491-45-4

Melting Point: >300 °C<sup>1</sup>

Fluorescent properties:

Free dye: Excitation maximum = 338 nm, Emission maximum = 505 nm (5 mM HEPES, 10 mM NaCl, pH 7.0)<sup>2</sup>

DNA complex: Excitation maximum = 356 nm, Emission maximum = 465 nm (5 mM HEPES, 10 mM NaCl, pH 7.0)<sup>2</sup>

Synonym: Hoechst 33258

This product is a fluorescent dye that binds to AT-rich regions of DNA, allowing it to be used to detect and quantitate DNA. Therefore, it is useful as a fluorescent stain for chromosomes<sup>2,3,4</sup>. There is a logarithmic relationship between the binding of this dye to AT base pairs and fluorescence values.<sup>5</sup> Therefore, this product can be used to quantitate DNA at the nanogram level using cellular homogenates.<sup>4</sup> However, the presence of proteins can affect the determination of the DNA content in these crude homogenates.<sup>6</sup> This product is also suitable for the determination of Q-banding patterns of chromosomes.<sup>7</sup> Overall, the dye is generally nontoxic to cells. The combination of dye plus light is also nontoxic, unless the cells have incorporated 5-bromo-2'-deoxyuridine.<sup>8</sup> Since the dye will penetrate cell membranes, it can be used either with cells or cell-free samples. In contrast, Hoechst 33342 (Product No. B 2261) is a poor fluorochrome for chromosome analysis, since it penetrates cell membranes poorly.<sup>9</sup>

This product has also been used for mammalian cell sorting based on DNA content<sup>3,9</sup> and for bacterial characterization by flow cytometry.<sup>10</sup> Other applications include high speed chromosome sorting at high salt concentrations, effects on virus replication, and spectral DNA studies<sup>11,12</sup>.

This dye is used as a small relatively nontoxic, cell-permeable molecule that can bind aptamers to regulate gene expression in mammalian cells (An aptamer is a short nucleic acid sequence that can bind a small molecule with high affinity and specificity).<sup>13</sup>

#### Precautions and Disclaimer

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

#### Preparation Instructions

This product is soluble in water (10 mg/ml), yielding a clear, yellow to brown solution. Heating may be required for complete solubilization. It has been observed that this material will precipitate from phosphate buffered solutions.

#### Storage/Stability

Stock solutions (200 µM) of Hoechst 33258 in water are reported to be stable for at least two months when kept at 2-8 °C in the dark.<sup>3</sup> SDS (0.1% w/v) has been reported to quench fluorescence within a few minutes.

#### References

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