

A New Bio-Assay for Aqueous Pollution

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Patent position: PCT/AU01/00099 “Biomolecular toxicity assay”

Research Funding:

- **Hermon Slade Foundation:** \$120,000 (3 years)
- **Hazard Screen Pty Ltd:** \$120,000 (3 years)
- **NSW State & Regional Development**

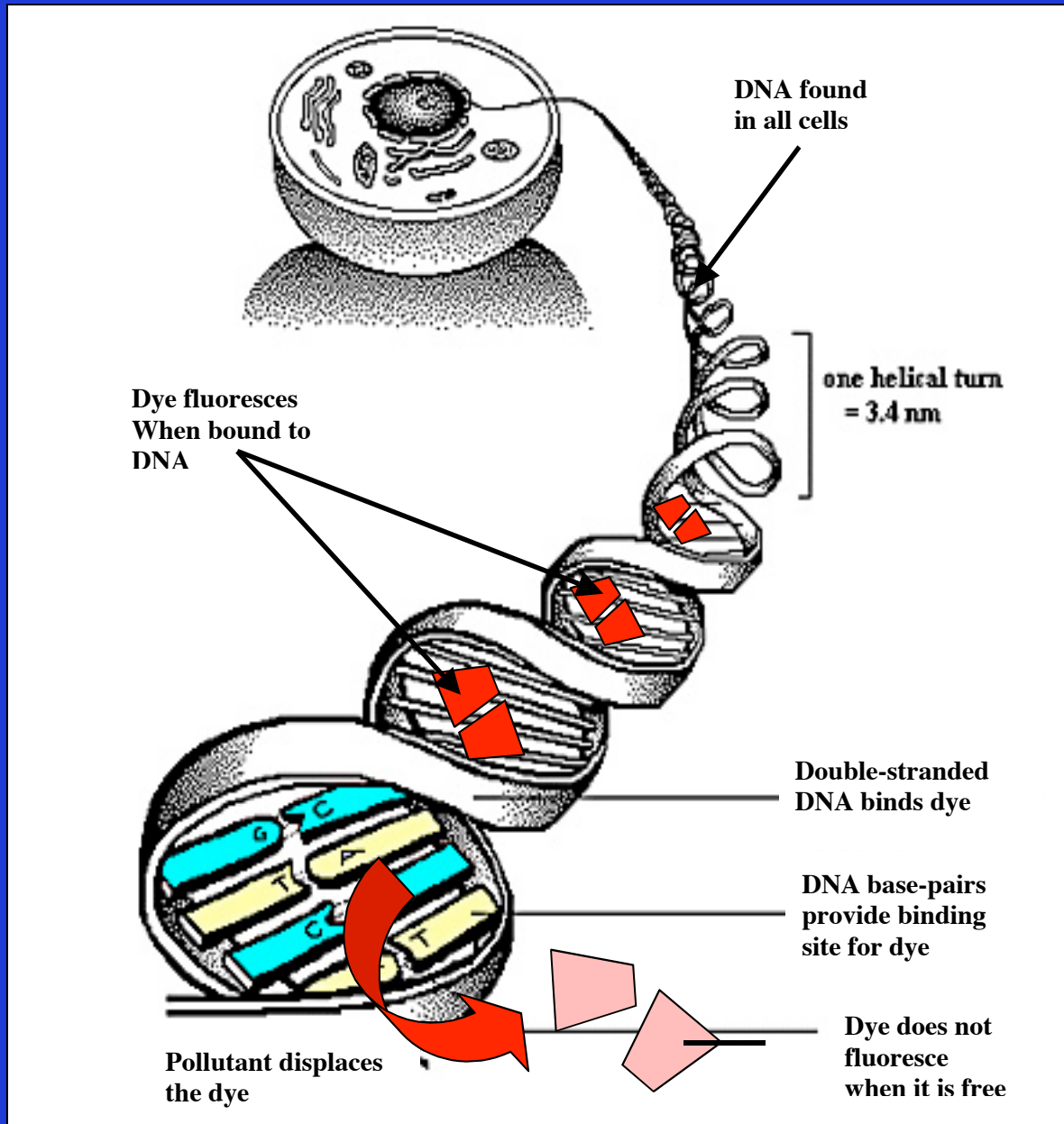
Current Bio-Assays

- **Whole Animals:** Fresh water (fish, copepods); marine (fish, Echinoderms)
- **Whole Cells** - *Microtox* luminescent bacteria,
- **Sub-cellular Particles:** Mitochondrial and sub-mitochondrial assays
- **A new Molecular Assay Based on the Fluorescence of a Dye Bound to DNA**

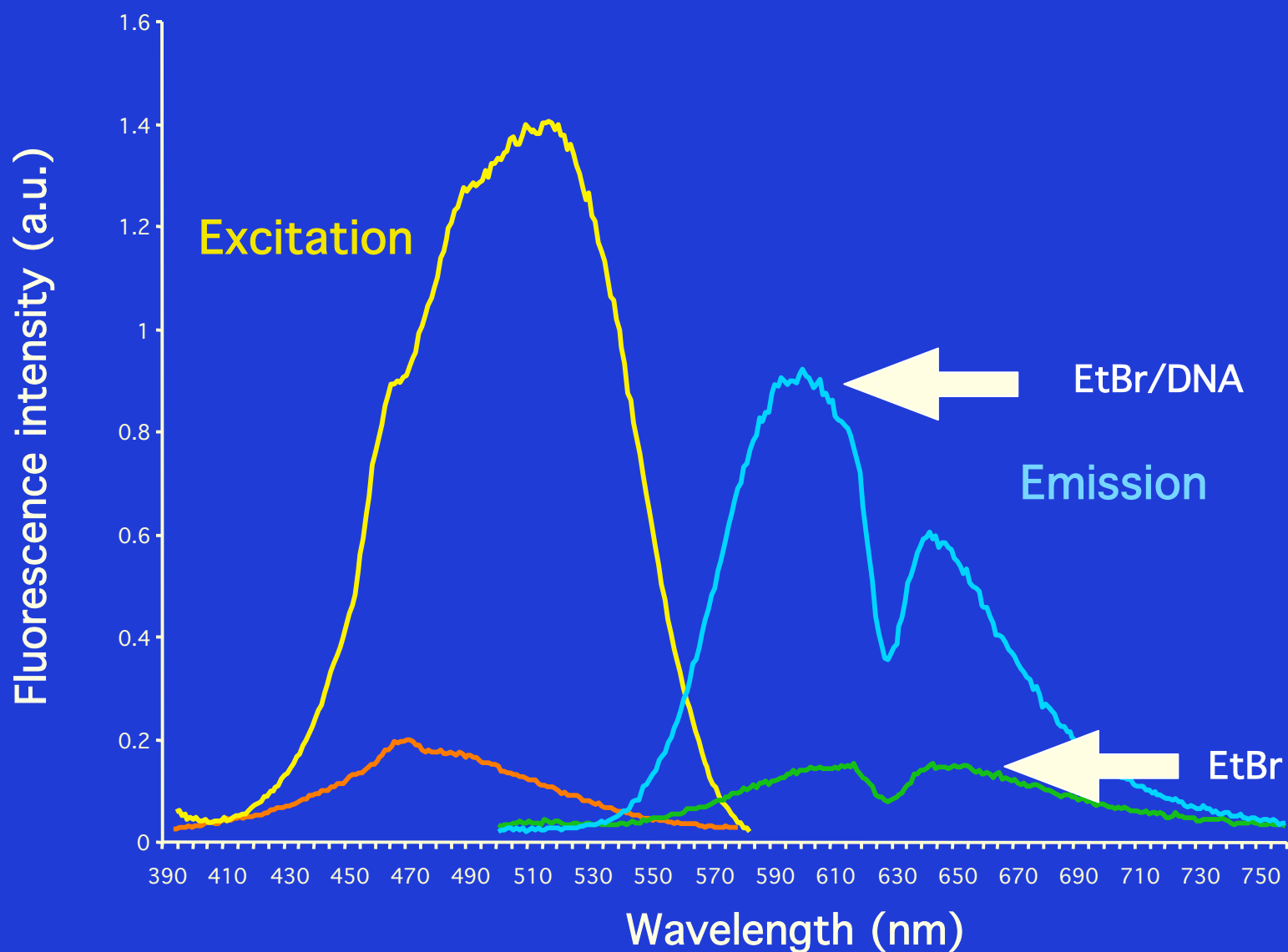
Criteria for a molecular environmental assay?

- **Biologically relevant**
- **Simple**
- **Reproducible**
- **Robust and portable**
- **Does not involve vertebrate animals**
- **EPA approved**

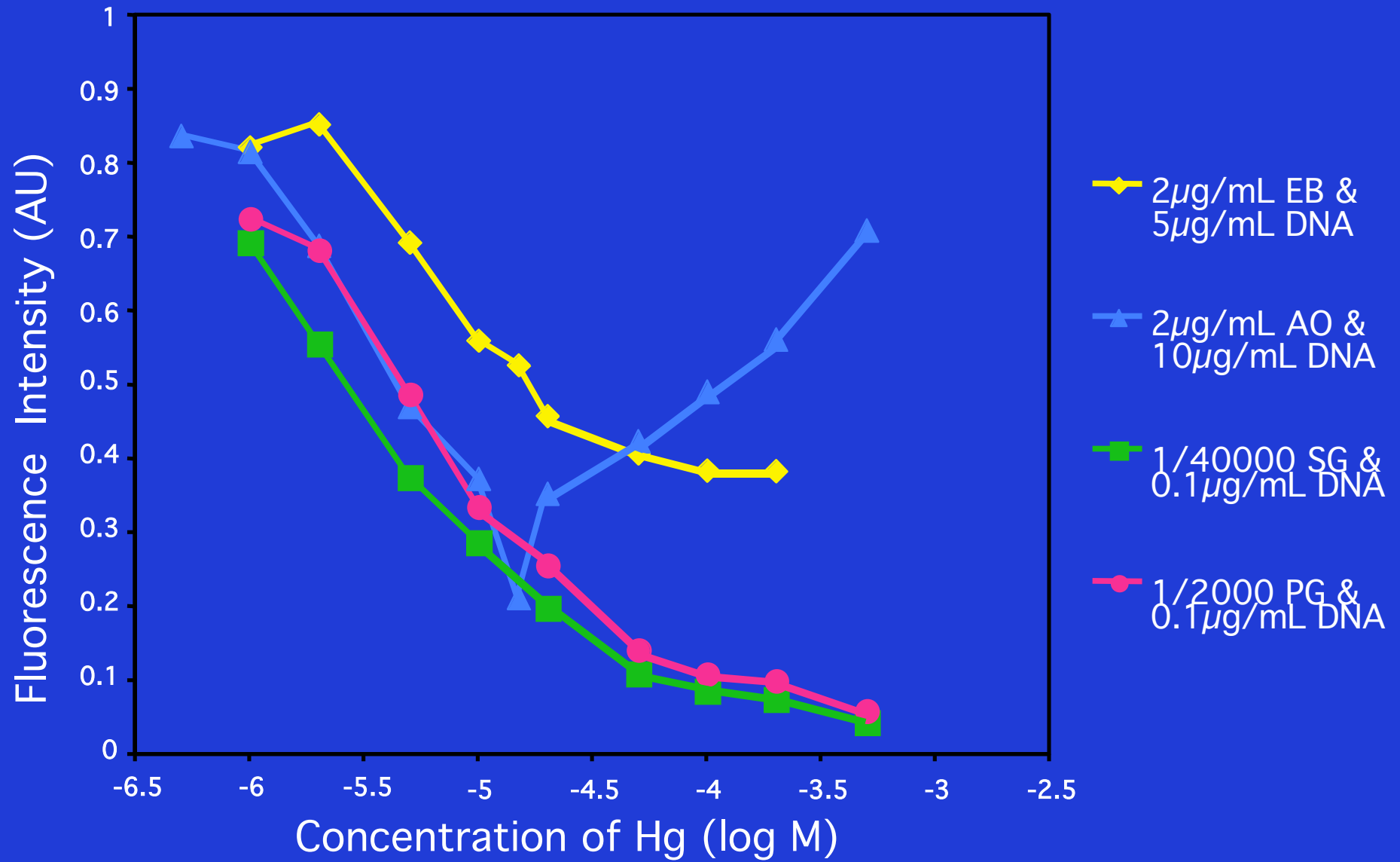
How Does It Work?



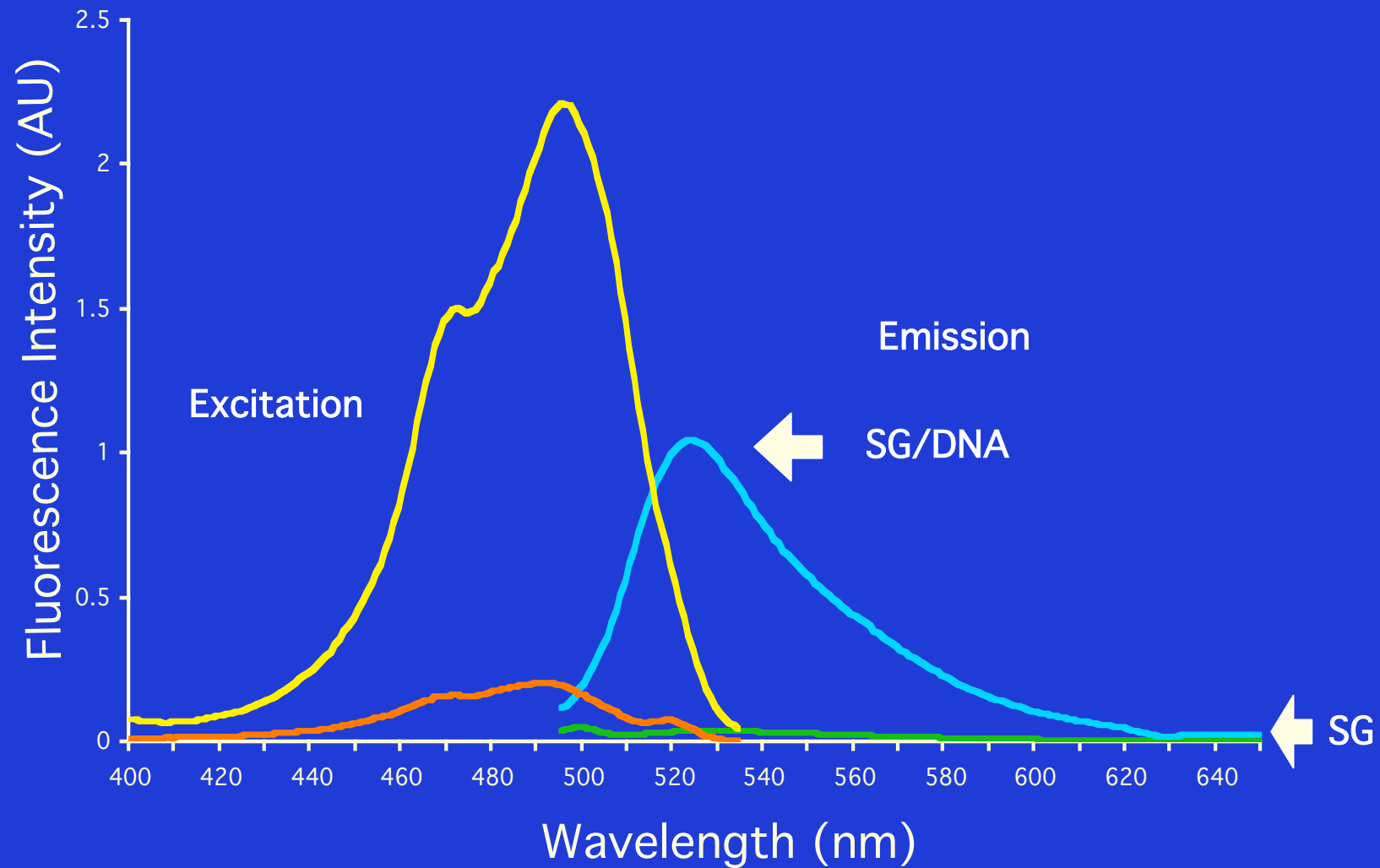
Fluorescence of EtBr Bound to DNA



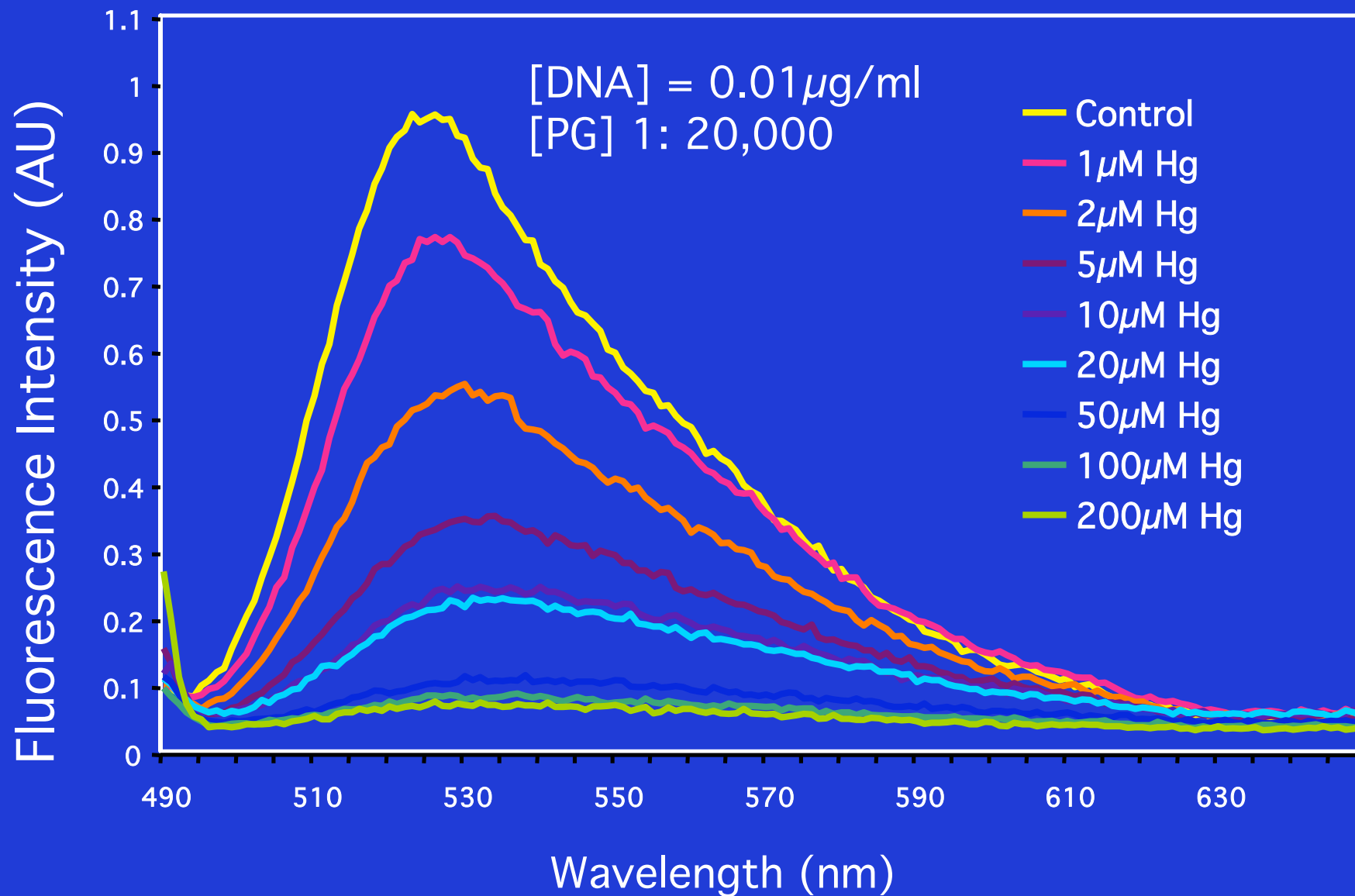
Comparing Different DNA Dyes



Fluorescence of Sybr Green-DNA

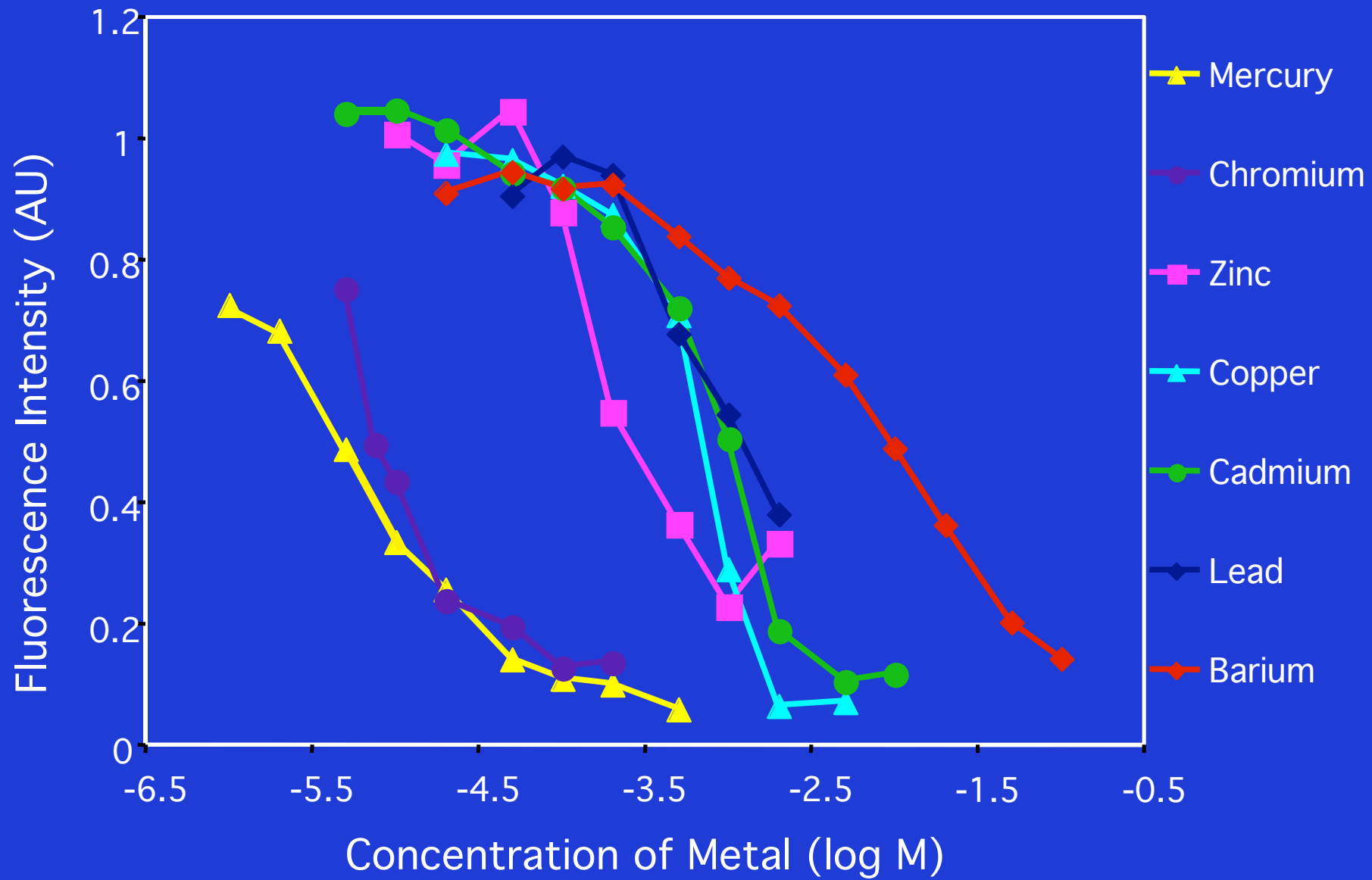


DNA-Pico Green

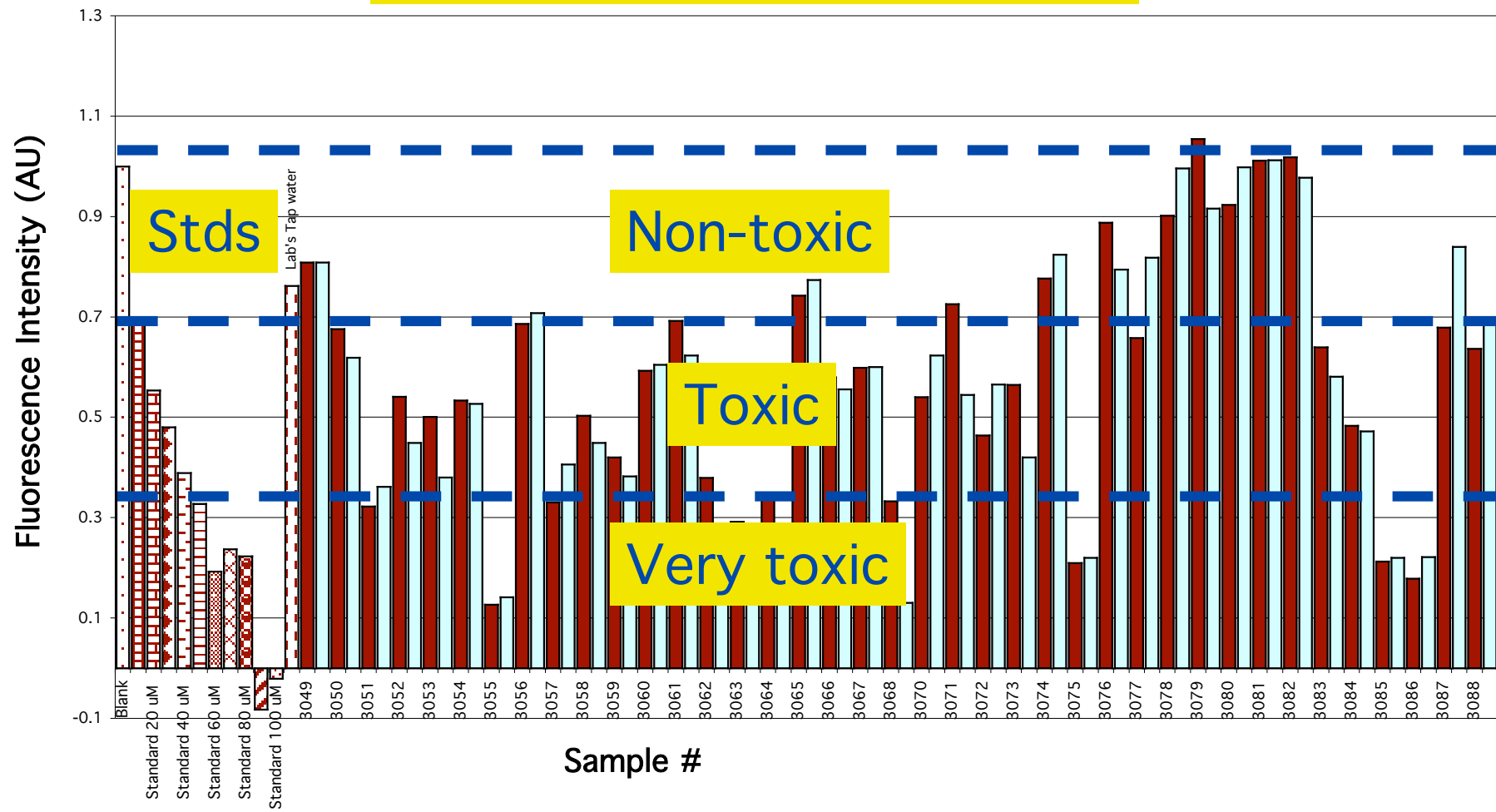


Relative Biotoxicities of Metal Cations

Heavy Metal	Hg	Cr	Zn	Cu	Pb	Cd	Ba
EC ₅₀ (μ M)	2.2	9.1	174	673	745	860	4403



EPA-supplied water samples



Blank	Standard 10 uM	Standard 20 uM	Standard 30 uM	Standard 40 uM	Standard 50 uM	Standard 60 uM	Standard 70 uM	Standard 80 uM	Standard 90 uM
Standard 100 uM	Lab's Tap water	3049	3049	3050	3051	3052	3052	3052	3052
3053	3053	3054	3054	3055	3055	3056	3056	3057	3057
3058	3058	3059	3059	3060	3060	3061	3061	3062	3062
3063	3063	3064	3064	3065	3065	3066	3066	3067	3067
3068	3068	3070	3070	3071	3071	3072	3072	3073	3073
3074	3074	3075	3075	3076	3076	3077	3077	3078	3078
3079	3079	3080	3080	3081	3081	3082	3082	3083	3083
3084	3084	3085	3085	3086	3086	3087	3087	3088	3088