SUPPLEMENTARY INFORMATION

Manuscript title: Fe-TAML/Hydrogen Peroxide Degradation of Concentrated Solutions of the Commercial Azo Dye Tartrazine

Authors: Evan S. Beach, Ryan T. Malecky, Roberto R. Gil, Colin P. Horwitz, and Terrence J. Collins

11 pages (8 tables, 3 Figures)

Table S1: Tartrazine color reduction at 400 nm (3.09×10^{-2} M dye, 15 min.)

		[Fe-TAML] / 10^{-6} M				
		3.75	9.375	18.75	37.5	75
$[H_2O_2]$ / 10^{-6} M	330000	21%	34%	48%	52%	44%
	500000	20%	37%	50%	67%	70%
	660000	21%	38%	52%	66%	79%
	1250000	21%	39%	55%	65%	75%
	2000000	21%	36%	51%	62%	74%

Reaction conditions: room temperature, 0.1 M pH 10 buffer.

Table S2: Tartrazine color reduction at 400 nm (3.09×10^{-2} M dye, 60 min.)

		[Fe-TAML] / 10^{-6} M				
		3.75	9.375	18.75	37.5	75
$[H_2O_2]$ / 10^{-6} M	330000	21%	39%	48%	50%	43%
	500000	30%	44%	58%	71%	82%
	660000	24%	50%	59%	70%	79%
	1250000	30%	46%	61%	71%	81%
	2000000	20%	42%	58%	67%	81%

Reaction conditions: room temperature, 0.1 M pH 10 buffer.

Table S3: Tartrazine color reduction at 400 nm (3.09×10^{-5} M dye, 15 min.)

		[Fe-TAML] / 10^{-6} M				
		0.375	0.9375	1.875	3.75	7.5
$[H_2O_2]$ / 10^{-6} M	312.5	11%	17%	18%	15%	12%
	625	18%	30%	32%	29%	21%
	1250	29%	48%	54%	50%	37%
	1875	38%	61%	68%	65%	50%
	3750	54%	75%	82%	86%	75%

Reaction conditions: room temperature, 0.1 M pH 10 buffer.

Table S4: Tartrazine color reduction at 400 nm (3.09×10^{-5} M dye, 60 min.)

		[Fe-TAML] / 10^{-6} M				
		0.375	0.9375	1.875	3.75	7.5
$[H_2O_2]$ / 10^{-6} M	312.5	25%	24%	21%	17%	13%
	625	43%	45%	39%	32%	22%
	1250	63%	74%	65%	53%	39%
	1875	72%	85%	83%	70%	52%
	3750	79%	90%	92%	92%	78%

Reaction conditions: room temperature, 0.1 M pH 10 buffer.

Table S5: FD&C Red No. 40 color reduction at 496 nm ($3.75x10^{-2}$ M dye)

		[I] / 10^{-6} M				
		0.375	0.9375	1.875	3.75	7.5
$[H_2O_2]$ / 10^{-6} M	37500	n/d	n/d	n/d	n/d	12%
	75000	n/d	n/d	n/d	n/d	19%
	150000	n/d	n/d	n/d	n/d	32%
	230000	n/d	n/d	n/d	n/d	44%
	330000	3%	13%	19%	31%	50%
	500000	1%	8%	17%	28%	42%
	660000	0%	8%	18%	28%	39%
	1250000	0%	4%	12%	23%	33%
	2000000	0%	2%	7%	19%	32%

Table S6: FD&C Red No. 40 color reduction at 496 nm ($3.75x10^{-5}$ M dye)

		[I] / 10^{-6} M				
		0.375	0.9375	1.875	3.75	7.5
$[H_2O_2]$ / 10^{-6} M	312.5	97%	97%	97%	95%	92%
	625	97%	98%	98%	97%	95%
	1250	99%	99%	99%	98%	97%
	1875	99%	99%	99%	98%	97%
	3750	100%	100%	99%	99%	97%

Dosage optimization for decolorization of FD&C Red No. 40 and H_2O_2. For FD&C Red No. 40, dye concentrations of $3.75x10^{-2}$ M and $3.75x10^{-5}$ M were used. The "extent of decolorization" was defined as the percent reduction in absorbance at the FD&C Red No. 40 λ_{max} of 496 nm. To examine whether the tartrazine findings could be extended to other dyes, a second azo dye, FD&C Red No. 40 (Red 40), was subjected to a single-addition dosage optimization study. Red 40 is also widely used, but is naphthol-based rather than a pyrazolone dye (Fig. S1). The pK_a of the naphthol in Red 40 is about 11.4, nearly two units higher than the pyrazolone in tartrazine.[1] Therefore, much less of the reactive common anion form should be available as the pH drops from 10 to 8 in a high dye concentration decolorization process. Red 40 requires 47 molar equivalents of H_2O_2 for mineralization compared to 45 for tartrazine.

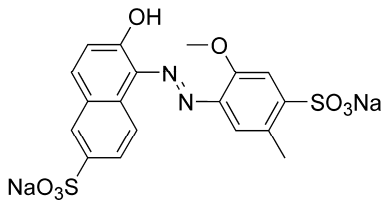

Fig. S1. Structure of FD&C Red No. 40.

Decolorization of low concentration solutions of Red 40 is faster than for comparable concentrations of tartrazine. By UV/Visible spectroscopy, there is no difference in the Red 40 reaction mixtures at 15 or 60 minutes, whereas tartrazine is more degraded at 60 minutes. However, at a high dye concentration, Red 40 decolorization was less extensive than for a comparable concentration of tartrazine: the maximum decolorization was 50%, as opposed to 79%, for single doses of Fe-TAML and H_2O_2. As with tartrazine, pH

adjustment with single additions of $7.5x10^{-5}$ M Fe-TAML and H_2O_2 did not improve performance for a low H_2O_2 dose (0.33 M), but when the H_2O_2 dose was increased to 2.0 M, decolorization increased to 72%. Total decolorization with the $7.5x10^{-5}$ M Fe-TAML dose could only be achieved by reducing the dye concentration slightly to $2.4x10^{-2}$ M and maintaining the high H_2O_2 concentration. It is possible that the Red 40 degradation products inhibit or poison the catalyst. Aggregation properties of Red 40 may also be different than tartrazine. While only a small fraction of the degradation products of Red 40 were identified by NMR spectroscopy, some were common to those from tartrazine degradation, but not surprisingly there were also significant differences. Thus inhibitors or poisons for the Fe-TAML catalysts may be substrate-specific.

The relationship between decolorization, catalyst concentration, and H_2O_2 concentration for treatment of a high dye concentration is shown in Fig. S2. At high catalyst dose (shown in bold), there is a sharp optimum at 0.33 M H_2O_2, but at lower Fe-TAML doses, the optimum is not as pronounced. Tartrazine is less sensitive to hydrogen peroxide concentration. The different behavior observed for Red 40 and tartrazine show that the experimental dosage ranges must be thoroughly explored to maximize the catalytic performance. Optimal degradation of a high concentration of Red 40 is seen at the catalyst:dye:H_2O_2 ratio of 1:500:4400, but only 50% of the dye is removed. At low dye concentration, every dosing combination tested resulted in >90% decolorization. Almost 100% decolorization occurred at ratios close to 1:100:10000. Thus, as found for tartrazine, the Fe-TAML/H_2O_2 system is more efficient at the higher concentration of Red 40.

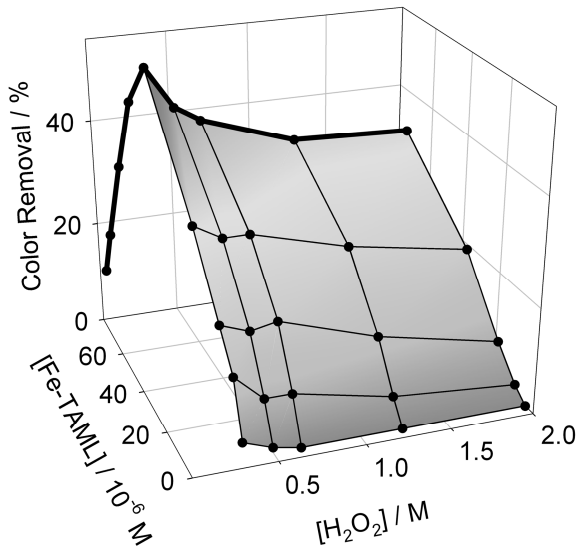

Fig. S2. FD&C Red No. 40 color reduction at 496 nm as functions of Fe-TAML and H_2O_2 doses, $3.75x10^{-2}$ M dye.

Ion chromatography. Testing for inorganic ions was completed by West Coast Analytical Service, Inc. of Santa Fe Springs, CA. The proprietary method SOP 4020 Rev. 6, which is based on EPA method 300.0, was used. The column was Dionex AS9-SC/AG9-SC. The eluent was 2 mM Na_2CO_3 and 0.75 mM $NaHCO_3$. The flow was 2.0 mL/min. $300x10^{-6}$ L of sample was injected. A suppressed conductivity detector was used. Testing for oxalic acid was carried out at Carnegie Mellon University using a Dionex AS9-SC/AG9-SC column, 9 mM Na_2CO_3 eluent at 1.0 mL/min flow rate, and $100x10^{-6}$ L sample injection.

Table S7: Ion chromatography results

	Nitrite	Nitrate	Sulfate
Detected concentration	ND	0.81 mM	4.02 mM
Percentage of theoretical amount assuming 100% mineralization	ND	0.66%	6.5%

ND = not detected.

Chemical Oxygen Demand. Samples were treated with Thermo Orion High-Range COD Test Reagent (CODHP0) in a Thermo Orion COD Thermoreactor at 150 ^{o}C for 2 hours. The COD values were measured with an Orion AQUAfast II AQ2040 colorimeter. For each sample, 5 COD tests were run using 100, 80, 60, 40, and 20 microliters of sample mixed with HPLC water to make 200 microliters total. Samples containing carbonate buffer were treated by adding an equal volume of concentrated sulfuric acid (Fisher scientific, technical grade) to the sample (exothermic reaction), and then 200, 160, 120, 80, and 40 microliters of the cooled acid mixture was mixed with HPLC water to make 200 microliters total. The measured COD values from the 5 tests were plotted vs. volume of sample used, and fit to a straight line using Igor Pro software version 5.0.4.8 (Wavemetrics, Inc.). The lines were extrapolated to 200 microliters and the COD values at that point were taken as the "actual" COD, per Test Reagent instructions.

Table S8. COD results.

Sample no.	Sample composition	COD (g/L)*
1	tartrazine	17.8
2	tartrazine + buffer	18.1
3	tartrazine + buffer + H_2O_2, catalase quench	18.7
4	tartrazine + catalase	19.2
5	tartrazine + buffer + Fe-TAML/H_2O_2, catalase quench	16.5

Samples were prepared using 3.09×10^{-2} M tartrazine, 0.1 M pH 10 buffer (carbonate), 0.58 M H_2O_2, 3.75×10^{-5} M Fe-TAML, and 0.59 g/L catalase. * Theoretical tartrazine COD is 22.3 g/L.

Comparison of COD samples 1 and 2 shows that the carbonate buffer does not significantly affect the COD measurements. Similarly, comparison of samples 3 and 4 shows that catalase is effective in removing H_2O_2 from the sample (H_2O_2 is a known to interfere with the COD test[2, 3]). However, the catalase itself does make a small contribution to the COD of approximately 1 g/L (compare samples 3 and 4 with samples 1 and 2). The observed COD value for tartrazine alone was only 80% of the theoretical amount. Dye molecules often contain functional groups that are resistant to the COD test reagent.[4, 5] Thus, the test carries uncertainties, but it can provide a lower limit or worst-case scenario for the efficiency of the use of H_2O_2.

Total Carbon. Total carbon (TC) is the sum of Total Organic Carbon (TOC) and Total Inorganic Carbon (TIC). TC measurements were recorded with an OI Analytical Solids

TOC Analyzer. Combustion was at 900 ^{o}C for 7.5 min. Samples were tested using 200 μL aqueous reaction mixture in closed cups packed with quartz wool.

Toxicity. Samples (20 mL) initially contained 3.09×10^{-2} M dye in pH 10 buffer (0.1 M carbonate). One sample was tested without treatment. The second sample was treated with 7.5×10^{-5} M Fe-TAML and 0.625 M H_2O_2, and quenched after 15 minutes by adding 12.5 mg catalase to the reaction mixture. Microtox testing was completed by Coastal Bioanalysts, Inc. of Gloucester, VA, according to protocols published by AZUR Environmental (Carlsbad, CA). The test was performed on a model 500 analyzer. Samples were osmotically adjusted by addition of 200 mg NaCl (ACS reagent grade) per 10 mL of solution. Sample pH was adjusted by addition of 6 N HCl. Color correction was performed. *Vibrio fischeri* bacteria were stored frozen until reconstituted immediately prior to testing, and a reference toxicant test using zinc(II) sulfate heptahydrate was run with the same lot of bacteria.

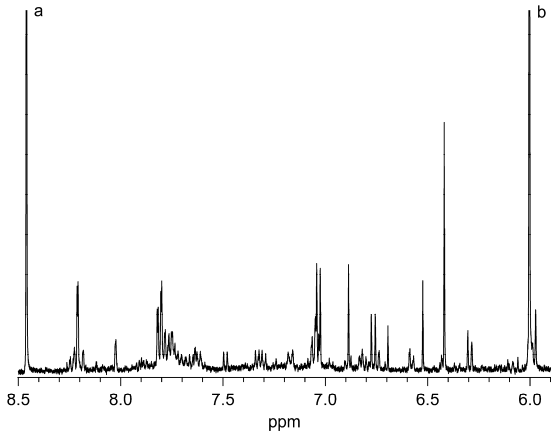

Fig. S3. ^{1}H NMR spectrum of a 4-phenolsulfonic acid degradation mixture. Reaction conditions: 4.36×10^{-2} M 4-phenolsulfonic acid sodium salt in 0.1 M pH 10 buffer, 1.2×10^{-4} M Fe-TAML, 1.1 M H_2O_2, 15 min. reaction time, catalase quench. The aromatic region is shown, between formic acid (peak a, 8.46 ppm) and maleic acid (peak b, 6.00 ppm). NOTE: peaks a and b have been cropped for clarity. The integral of the region between 8.0 and 7.5 is comparable to the integral of the formic acid peak.

References

1. Pérez-Urquiza, M.; Beltrán, J. L., *J. Chromatogr., A* **2001,** *917*, 331-336.
2. Talinli, I.; Anderson, G. K., *Water Res.* **1992,** *26*, 107-110.
3. Kang, Y. W.; Cho, M.-J.; Hwang, K.-Y., *Water Res.* **1999,** *33*, 1247-1251.
4. Clesceri, L. S.; Greenberg, A. E.; Eaton, A. D., Eds. *Standard Methods for the Examination of Water and Wastewater*. 20th ed.; United Book Press Inc.: Baltimore, Maryland, 1998.
5. The calculated COD of the pyrazolone ring (5.44 g/L O_2 for 3.09×10^{-2} M $C_4HN_2NaO_3$) is about the same as the discrepancy between theoretical and observed COD for tartrazine.