



## Supporting Information

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### **Thiamine Assays—Advances, Challenges, and Caveats**

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## Supporting Information

Table S1. Properties of products formed from colorimetric reactions to detect thiamine

	<b>E</b> <b>(L*<math>\text{mol}^{-1}</math>*<math>\text{cm}^{-1}</math>)</b>	<b><math>\lambda_{\text{max}}</math></b> <b>(nm)</b>	<b>Limit of</b> <b>detection/Range</b>	<b>Interferences</b>	<b>Source</b>
Thiamine (unmodified)	3.07x10 <sup>2</sup> 8.55x10 <sup>3</sup> (TDP)	235/267	N/A	N/A	[1]
Thiochrome	2.06x10 <sup>4</sup> (TDP)	367	N/A	N/A	[1]
Bromothymol blue	N/A	420	4-10 $\mu\text{g/mL}$	Nicotinamide, ethyl alcohol	[2]
Barium sulfate precipitation	6.73x10 <sup>3</sup>	420	0.41 $\mu\text{g/mL}$ (2-32 $\mu\text{g/mL}$ )	N/A	[3]
Sodium 1,2-napthoquine-4-sulphonate	N/A	487	1.71 $\mu\text{g/mL}$ (10-40 $\mu\text{g/mL}$ )	eliminated interferences by excipients by red-shifting max wavelength of thiamine-NQS derivative	[4]
Diazotized sulfanilic acid	7.74x10 <sup>3</sup>	490	2-35 $\mu\text{g/mL}$	tested glucose, lactose, sucrose, starch, and magnesium stearate – none of which interfered with the recovery of thiamin hydrochloride	[5]
Diazotized p-aminoacetophenone	N/A	516	N/A	Uric acid, metals	[6]
Leucocrystal/iodine	1.12x10 <sup>5</sup>	589	0.19 $\mu\text{g/mL}$ (0.4-2.4 $\mu\text{g/mL}$ )	Vitamin B <sub>2</sub>	[7]
Triphenylmethane acid	(0.82-1.65)x10 <sup>5</sup> (formation of complex)	420-450	42.5 ng/mL	Vitamin B <sub>2</sub> , Vitamin B <sub>6</sub> , Vitamin C Cupric chloride Theobromine	[8]
	(1.26-3.92)x10 <sup>5</sup> (loss of dye)	550-620	22.4 ng/mL		

Figure S1. Historic methods utilizing colorimetric detection

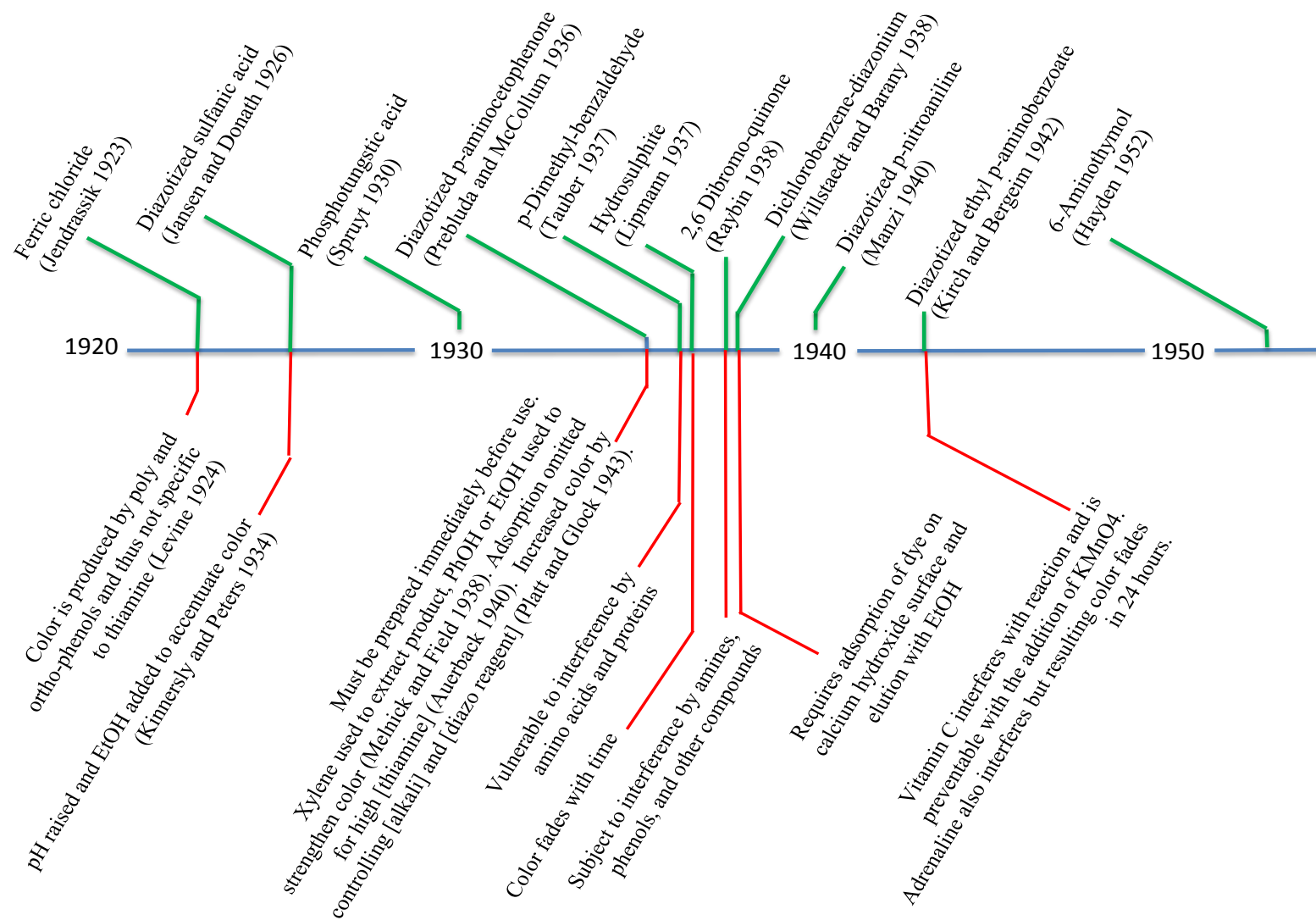


Figure S2. Historic methods using fluorometric detection

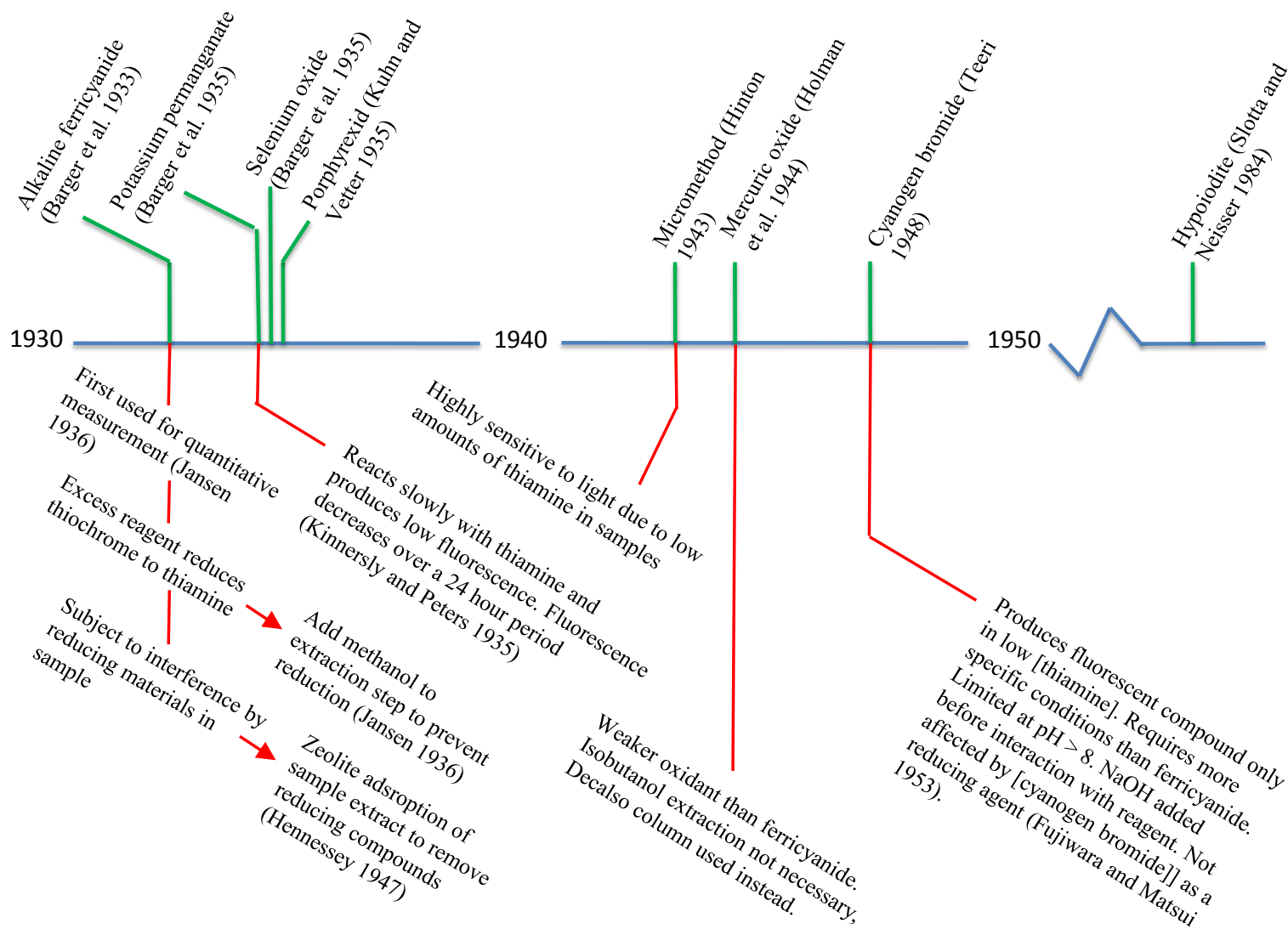


Table S2. Methods based on chemiluminescence (CL), electrochemical techniques, or electrochemiluminescence (ECL) for thiamine detection

Method basis	Platform	Range/Limit of detection	Sample matrix	Interferences	Source
<i>Chemiluminescence</i>					
CL during potassium ferricyanide oxidation	Flow-injection analysis	6.75-169 µg/mL (0.01 µg/mL)	Vitamin B1 tablets	Ascorbic acid	[34]
Enhancement of luminol/H <sub>2</sub> O <sub>2</sub> CL	Flow-injection analysis	0.05-8 µg/mL (0.01 µg/mL)	Vitamin B1 tablets and injections	Ascorbic acid	[35]
Suppression of luminol/KIO <sub>4</sub> CL	Flow-injection analysis	3.3 nM-6.7 µM (1.0 nM)	Vitamin B1 tablets and human urine	Uric acid, Cu <sup>2+</sup>	[36]
<i>Electrochemical methods</i>					
Amperometry	Anion exchange HPLC	20-250 ng	Multivitamin tablets	Vitamin C (but resolved chromatographically)	[37]
Electrochemical oxidation/fluorescence detection	Flow-injection analysis	1 ng	Multivitamin tablets, capsules, and syrup	None observed	[38]
Square-wave voltammetry	Electrochemical workstation	11 nM-2.2 µM (5.5 nM)	Vitamin B1 tablets	None observed (100-fold greater ratio of other vitamins/ions tested)	[39]
Square-wave voltammetry using Cd <sup>2+</sup> as an indicator ion	Electrochemical workstation	1 µM-4 mM (0.1 µM)	Vitamin B1 tablets	None observed (Limit substances tests at a 10-fold greater ratio)	[40]
Cyclic voltammetry using pencil lead	Electrochemical workstation	10 µM-1 mM (5.34 µM)	Vitamin B1 tablets	None observed (Testing limited to VB6)	[41]
Fast Fourier transform square-wave voltammetry	Flow-injection analysis	500 pM-400 nM (560 pM)	Vitamin B1 tablets	Not assessed	[42]
Differential pulse voltammetry using ds-DNA	Flow-injection analysis	1.0-80 µg/mL (0.44 µg/mL)	Serum, plasma, and urine samples	None reported	[43]
Adsorptive stripping differential pulse voltammetry using ds-DNA	Flow-injection analysis	2.5-800 ng/mL (1.1 µg/mL)	Serum, plasma, and urine samples	None reported	[43]

Table S3. Methods based on changes of nanoparticle properties

Nanoparticle type	Surface modification	Signal with increasing [thiamine]	Interaction with thiamine	Limit of detection/Range	Source
Eu-doped Y <sub>2</sub> O <sub>3</sub> NPs	Captopril	Fluorescence enhancement ( $\lambda_{\text{ex}}=\text{NA}$ ; $\lambda_{\text{em}}=612$ nm)	Electrostatic	144 nM/ linear assay range to 44 $\mu\text{M}$	[44]
Core-shell silica NPs (C-dots)	Coconut water natural products	Fluorescence enhancement ( $\lambda_{\text{ex}}=367$ nm; $\lambda_{\text{em}}=430$ nm)	Displacement of Cu <sup>2+</sup> from C-dot surface	280 nM/10-50 $\mu\text{M}$	[45]
Iron phthalocyanine entrapped in silica NPs	-	Fluorescence enhancement	Catalyzed oxidation to thiochrome	2.0 nM (5.0 nM-1 $\mu\text{M}$ )	[46]
Gold NPs	4-amino-6-hydroxy-2-mercaptopyrimidine	Fluorescence enhancement ( $\lambda_{\text{ex}}=520$ nm; $\lambda_{\text{em}}=781$ nm)	Electrostatic	6.8 fM (10-120 pM)	[47]
Gold NPs	Polyphenols	Fluorescence enhancement ( $\lambda_{\text{ex}}=356$ nm; $\lambda_{\text{em}}=425$ nm)	Oxidation to thiochrome	0.5 $\mu\text{M}$	[48]
Gold NPs	-	Colorimetric signal at 590 nM	Gold interaction with thiazole sulfur	54 nM (0.15-3.5 $\mu\text{M}$ )	[49]
Gold NPs	Citrate	Resonance Raleigh scattering (368 nm)	Electrostatic/hydrophobic	0.9 ng/mL (0-2.8x10 <sup>-7</sup> M)	[50]
CdSe quantum dots	Citrate	Fluorescence quenching ( $\lambda_{\text{ex}}=380$ nm; $\lambda_{\text{em}}=591$ nm)	Adsorption	70 ng/mL (5-40 $\mu\text{g/mL}$ )	[51]
CdTe nanorods	Thioglycolic acid and cysteine	Fluorescence enhancement ( $\lambda_{\text{ex}}=530$ nm; $\lambda_{\text{em}}=665$ nm)	Coordination of thiazole sulfur with Cd <sup>2+</sup>	30 nM (0.1-3 $\mu\text{M}$ )	[52]

Table S4. Microorganisms used in thiamine bioassays

Organism	Limit of Detection (µg/L)	Length of incubation	Signal type	Applications	Disadvantages	Source
<i>Cryptococcus albid</i>	10	5 days	Optical density	Marine water	1. Thiazole moiety of thiamine could interfere with thiamine in the bioassay. 2. The growth of <i>Cryptococcus albid</i> would be affected by salinity. 3. Time consuming incubation.	[53]
<i>Escherichia coli</i>	0.012.5	19 hours	Turbidity of culture medium	Blood	1. <i>E. coli</i> also responded to cocarboxylase.	[54]
<i>Kloeckera brevis</i>	0.25	18-24 hours	Turbidity of culture medium	Foods	1. <i>K. brevis</i> also responded to cocarboxylase	[55]
<i>Lactobacillus fermenti</i>	5	16-18 hours	Turbidity of culture medium	Foods, tissues, urine and vitamin concentrates	1. <i>L. fermenti</i> also responded to phosphorylated esters of thiamine. 2. <i>L. fermenti</i> tends to behave occasionally in an inexplicably anomalous manner, giving inconsistent results upon repeated assay.	[56]
<i>Lactobacillus viridescens</i>	1.3	20 hours	Turbidity of the culture medium	Foods	3. <i>Lactobacilli</i> are often stimulated nonspecifically by substances in biologic materials.	[57]
<i>Monochrysis lutheri</i>	0.002	2-3 days	<sup>14</sup> C-CO <sub>2</sub> uptake / cell counts	Marine water and fresh water	1. Pyrimidine moiety of thiamine could interfere with thiamine in the bioassay. 2. Time consuming incubation.	[58]
<i>Ochromonus malhamensi</i>	0.3	4-6 days	Optical density of the culture	Blood, urine, cerebrospinal fluid, liver and brain	1. Time consuming incubation.	[59]
<i>Phycomyces blakesleeanus</i>	5	10 days	Weight of mycelium from the culture	Blood and plants tissues	1. Thiazole and pyrimidine moiety could interfere with thiamine in the bioassay. 2. Time consuming incubation.	[60]
<i>Saccharomyces cerevisiae</i>	0.39	15 hours	Turbidity of culture medium	-	1. Pyrimidine, thiazole and sulfite degradation products of thiamine could interfere with thiamine in the bioassay .	[61]
<i>Staphylococcus aureus</i>	2-4	36 hours	Turbidity of culture medium	Pork liver, kidney and ham	1. <i>S. aureus</i> also responds to pyrimidine and thiazole moieties of thiamine. 2. <i>S. aureus</i> is pathogenic for humans	[62]
<i>Streptococcus salivarius</i>	0.2	24 hours	Turbidity of culture medium	Foods	1. <i>S. salivarius</i> also responded to cocarboxylase and monophosphothiamine.	[63]

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