

# Colorimetric Determination of Nitrate after Reduction to Nitrite in a Paper-Based Dip Strip <sup>†</sup>

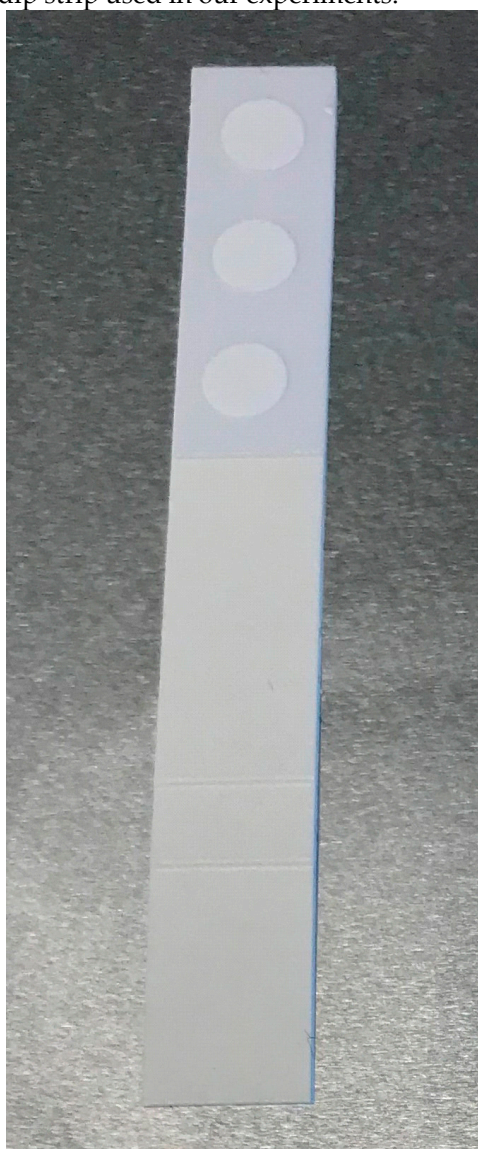
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Figure S1 shows a dip strip used in our experiments.



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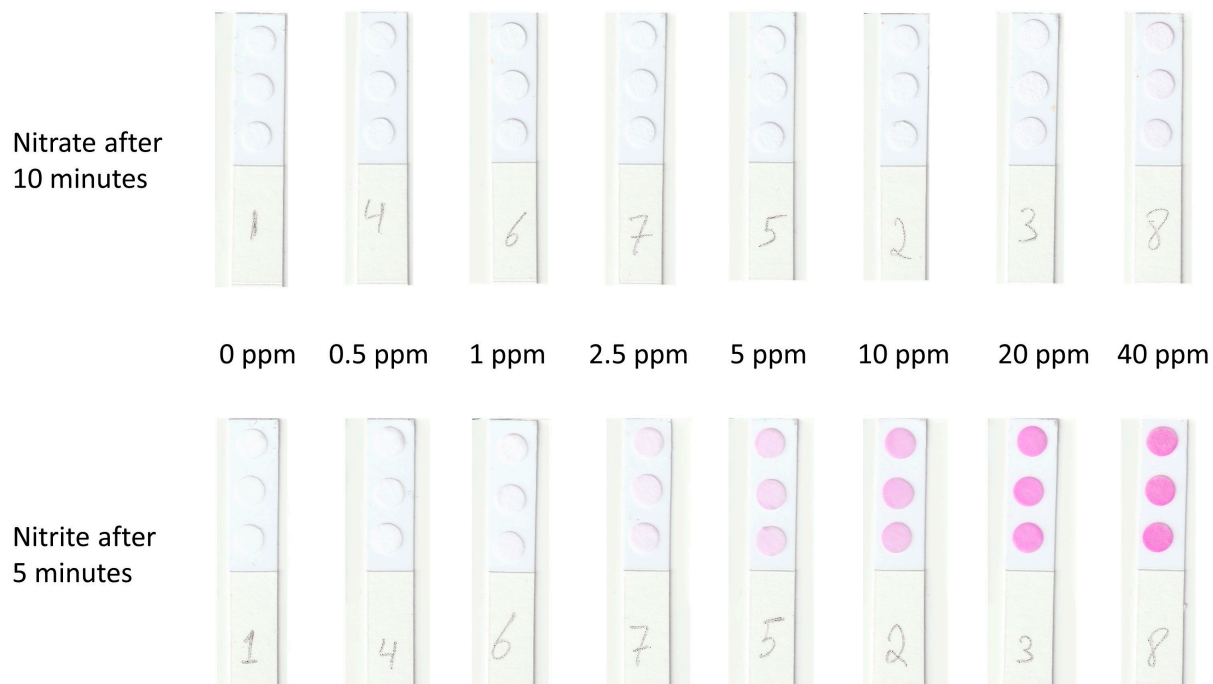
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**Figure S1.** Dip strip used in experiments.

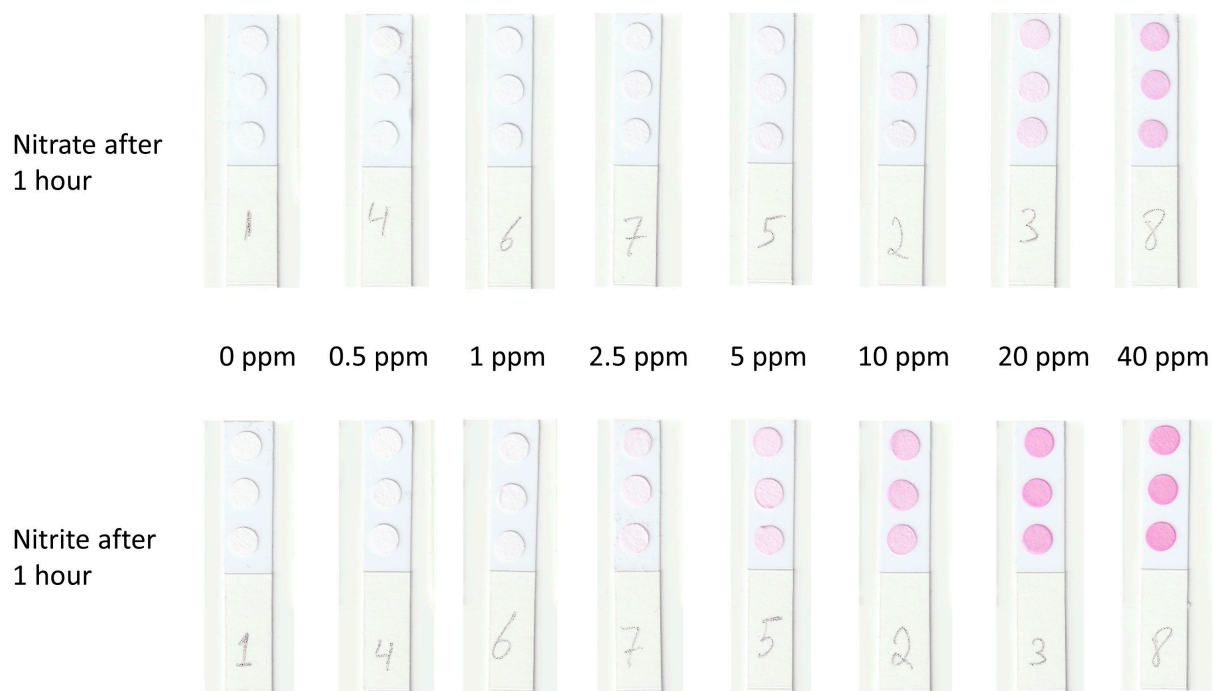
Figure S2 shows the dip strips after the solutions are dried on the detection zones. Solutions “A” and “B” are prepared following the procedure outlined by Thongkam et al. [1].



**Figure S2.** Dip strip used in experiments after the solutions are dried on the detection zones.



**Figure S3.** Color formed in the detection zone vs. nitrate or nitrite concentrations after several minutes.



**Figure S4.** Color formed in the detection zone vs. nitrate or nitrite concentrations after 1 hour

**Table S1.** ImageJ analysis of nitrate detection zones after 10 minutes. Test order was randomized.

Standard	Nitrate Concentration (ppm)	Run Order	Unweighted Intensity Value (a.u.)	Weighted Intensity Value (a.u.)	RGB Intensity Value (a.u.)	Red Intensity Value (a.u.)	Green Intensity Value (a.u.)	Blue Intensity Value (a.u.)
1	0	1	251.065	250.971	250	247.881	252.005	253.42
2	0	1	250.3	250.078	249.075	246.752	251.142	253.048
3	0	1	249.826	249.729	248.423	245.995	251.015	252.622
4	0.5	4	250.304	250.113	248.996	247.024	251.116	252.811
5	0.5	4	250.768	250.663	249.449	247.326	251.816	253.267
6	0.5	4	249.793	249.693	248.323	246.101	250.948	252.464
7	1	6	250.82	250.806	249.156	248.226	251.697	252.702
8	1	6	250.534	250.469	249.381	247.83	251.358	252.553
9	1	6	249.875	249.682	248.274	246.758	250.636	252.298
10	2.5	7	250.956	250.839	249.492	248.2	251.688	253.09
11	2.5	7	250.103	249.868	249.003	246.913	250.825	252.594
12	2.5	7	249.781	249.456	248.357	246.777	250.249	252.33
13	5	5	251.465	251.335	250.03	249.109	252.03	253.442
14	5	5	250.478	250.259	249.098	247.619	251.08	252.805
15	5	5	249.514	249.166	247.936	246.408	249.998	252.082
16	10	2	250.71	250.4	249.574	248.004	251.087	253.071
17	10	2	250.191	249.802	248.598	247.099	250.592	252.805
18	10	2	249.397	249.066	248.045	246.134	249.974	252.016
19	20	3	251.108	250.714	249.908	249.185	251.02	253.28
20	20	3	249.749	249.249	248.456	247.59	249.575	252.159
21	20	3	249.372	248.773	248.188	247.035	249.093	252.012
22	40	8	247.819	246.824	246.109	246.922	246	250.803
23	40	8	247.738	246.659	246.487	246.627	245.811	251.176
24	40	8	246.629	245.46	245.661	245.274	244.634	250.453

**Table S2.** ImageJ analysis of nitrate detection zones after 1 hour. Test order was randomized.

Standard	Nitrate Concentration (ppm)	Run Order	Unweighted Intensity Value (a.u.)	Weighted Intensity Value (a.u.)	RGB Intensity Value (a.u.)	Red Intensity Value (a.u.)	Green Intensity Value (a.u.)	Blue Intensity Value (a.u.)
1	0	1	250.708	250.871	249.273	248.809	251.696	251.837
2	0	1	251.095	251.222	249.545	249.177	252.009	252.341
3	0	1	250.669	251.05	248.913	248.353	252.192	251.781
4	0.5	4	249.77	250.163	247.717	248.449	251.024	250.11
5	0.5	4	251.419	251.592	249.831	249.609	252.385	252.494
6	0.5	4	251.026	251.232	249.387	249.104	252.11	252.075
7	1	6	251.13	251.163	249.643	249.383	251.796	252.458
8	1	6	250.696	250.796	249.562	248.94	251.518	251.857
9	1	6	250.147	250.235	248.482	248.329	250.954	251.392
10	2.5	7	250.441	250.207	248.822	249.208	250.414	251.915
11	2.5	7	249.429	248.967	248.173	248.297	248.926	251.159
12	2.5	7	249.72	249.53	248.227	248.385	249.817	251.141
13	5	5	250.291	249.589	248.821	249.437	249.246	252.091
14	5	5	249.306	248.575	247.909	248.395	248.166	251.39
15	5	5	248.256	247.477	246.676	247.455	246.979	250.296
16	10	2	247.854	246.366	246.621	248.435	244.532	250.735
17	10	2	247.351	245.736	246.125	248.07	243.705	250.429
18	10	2	247.209	246.249	246.127	246.836	245.284	248.664
19	20	3	241.457	237.454	240.588	245.128	231.5	247.874
20	20	3	242.773	239.433	242.29	245.69	234.549	248.121
21	20	3	240.116	235.907	239.801	244.07	229.614	246.821
22	40	8	227.251	219.413	227.482	236.837	206.934	237.933

23	40	8	226.406	218.295	226.936	236.557	205.316	237.265
24	40	8	226.213	218.288	227.038	236.08	205.625	236.868

**Table S3.** ImageJ analysis of nitrite detection zones after 5 minutes. Test order was randomized.

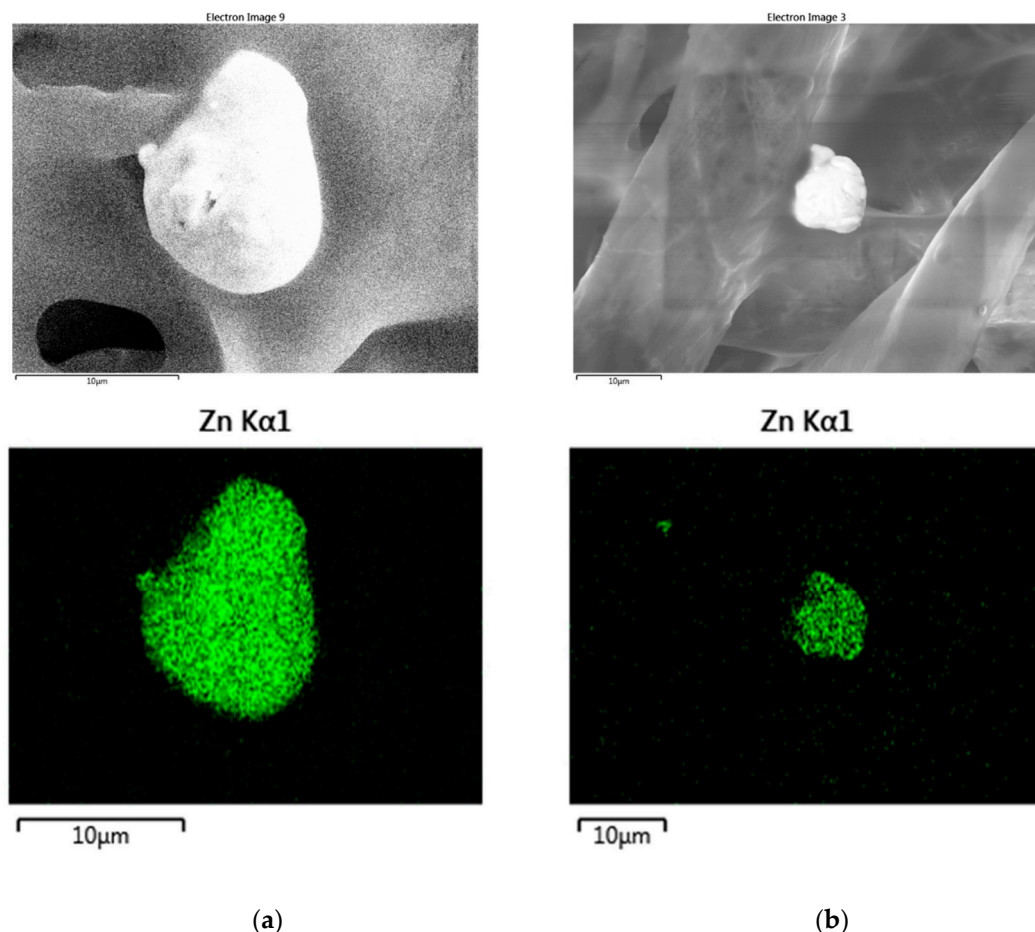
Standard	Nitrite Concentration (ppm)	Run Order	Unweighted Intensity Value (a.u.)	Weighted Intensity Value (a.u.)	RGB Intensity Value (a.u.)	Red Intensity Value (a.u.)	Green Intensity Value (a.u.)	Blue Intensity Value (a.u.)
1	0	1	251.968	251.992	250.763	249.775	252.717	253.685
2	0	1	251.915	251.921	251.017	249.224	252.844	253.869
3	0	1	251.914	251.922	251.088	249.225	252.863	253.824
4	0.5	4	250.573	250.351	249.432	248.685	250.736	252.598
5	0.5	4	250.827	250.421	249.969	248.271	251	253.222
6	0.5	4	250.833	250.53	249.698	248.514	251.057	253.077
7	1	6	251.263	250.69	250.133	250.075	250.58	253.265
8	1	6	250.23	249.522	249.352	248.713	249.393	252.785
9	1	6	250.141	249.434	249.385	248.292	249.478	252.789
10	2.5	7	244.944	242.643	244.496	246.011	239.397	249.934
11	2.5	7	246.589	244.712	246.11	247.005	242.292	250.884
12	2.5	7	245.889	243.943	245.226	246.302	241.452	250.324
13	5	5	237.605	231.712	237.139	242.977	223.035	246.796
14	5	5	237.572	231.989	237.177	242.493	223.784	246.594
15	5	5	237.035	231.612	237.142	241.717	223.603	246.021
16	10	2	223.378	212.67	223.991	235.964	195.814	238.215
17	10	2	223.55	212.965	223.816	235.84	196.413	238.282
18	10	2	225.718	216.553	226.289	235.858	202.565	238.437
19	20	3	208.288	191.069	210.08	232.827	162.211	230.008
20	20	3	207.778	189.631	210.133	233.183	159.454	230.876
21	20	3	208.566	190.976	210.341	232.842	161.898	231.138
22	40	8	199.566	182.199	202.669	229.865	150.953	217.881
23	40	8	199.691	181.343	202.147	230.718	148.732	219.557
24	40	8	196.835	177.116	199.941	228.792	142.568	219.166

**Table S4.** ImageJ analysis of nitrite detection zones after 1 hour. Test order was randomized.

Standard	Nitrite Concentration (ppm)	Run Order	Unweighted Intensity Value (a.u.)	Weighted Intensity Value (a.u.)	RGB Intensity Value (a.u.)	Red Intensity Value (a.u.)	Green Intensity Value (a.u.)	Blue Intensity Value (a.u.)
1	0	1	250.839	251.172	249.499	249.891	251.894	251.043
2	0	1	250.589	250.904	249.515	249.458	251.678	250.958
3	0	1	249.964	250.319	248.89	248.907	251.084	250.251
4	0.5	4	251.285	251.279	250.064	250.566	251.594	251.894
5	0.5	4	249.559	249.551	248.777	248.619	249.947	250.361
6	0.5	4	250.555	250.619	249.029	249.666	251.099	251.178
7	1	6	250.137	249.825	248.439	249.604	249.772	251.014
8	1	6	248.426	248.114	246.579	247.851	248.095	249.341
9	1	6	249.126	248.847	248.082	248.336	248.915	250.228
10	2.5	7	244.941	243.052	243.378	246.964	240.268	247.431
11	2.5	7	247.549	246.235	246.605	248.628	244.486	249.299
12	2.5	7	246.198	244.804	245.142	248.282	242.602	247.49
13	5	5	251.626	237.591	240.418	246.993	231.026	246.577
14	5	5	241.667	238.154	239.708	246.463	232.346	245.961
15	5	5	240.273	236.707	239.698	245.062	230.868	244.626
16	10	2	228.775	221.865	228.932	239.326	210.077	236.632
17	10	2	228.699	221.705	228.175	239.476	209.75	236.551
18	10	2	231.959	226.094	231.647	241.007	215.879	239.018
19	20	3	215.88	204.922	216.451	235.059	185.15	227.573



20	20	3	213.68	202.513	214.403	233.036	182.436	225.753
21	20	3	214.992	204.514	215.893	233.238	185.695	226.038
22	40	8	209.424	198.099	210.888	232.793	176.237	219.352
23	40	8	208.019	195.834	209.695	232.543	172.57	219.095
24	40	8	206.98	194.586	208.561	231.346	171.189	218.475



**Figure S5.** Zinc microparticles observed using an electron scanning microscope with EDS analysis in the nitrate test fields of commercial dip strips (a) Quantofix 91313 (b) Quantofix 91351.

## Reference

1. Thongkam, T., & Hemavibool, K. An environmentally friendly microfluidic paper-based analytical device for simultaneous colorimetric detection of nitrite and nitrate in food products. *Microchem. J.* **2020**, 159, doi: 10.1016/j.microc.2020.105412