Supporting Information

Inactivation of Murine Norovirus and Fecal Coliforms by Ferrate(VI) in Secondary Effluent Wastewater

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Total Pages: 11
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31 **Text S1**

32 **Chemicals.** Phosphate buffer (PB) solutions (0.01 M) were prepared by diluting amounts of

33 Na_2HPO_4 and $NaH_2PO_4 \cdot H_2O$ in DI water to achieve the desired pH. The secondary effluent

34 wastewater (SEW) at pH 8.2 was grab sampled (prior to disinfection step) at the Pottersburg

35 Wastewater Treatment Plant (London, Ontario, Canada), an activated sludge-based treatment

36 plant. Sodium phosphate dibasic (Na_2HPO_4; ≥99% purity) and sodium thiosulfate ($Na_2S_2O_3$;

37 99% purity) were purchased from Sigma-Aldrich, Canada. Sodium phosphate monobasic

38 monohydrate ($NaH_2PO_4 \cdot H_2O$; ≥98% purity) was purchased from Caledon Laboratory Chemicals

39 (Georgetown, Ontario, Canada). Polypropylene (0.45 μm) and regenerated cellulose (0.2 μm)

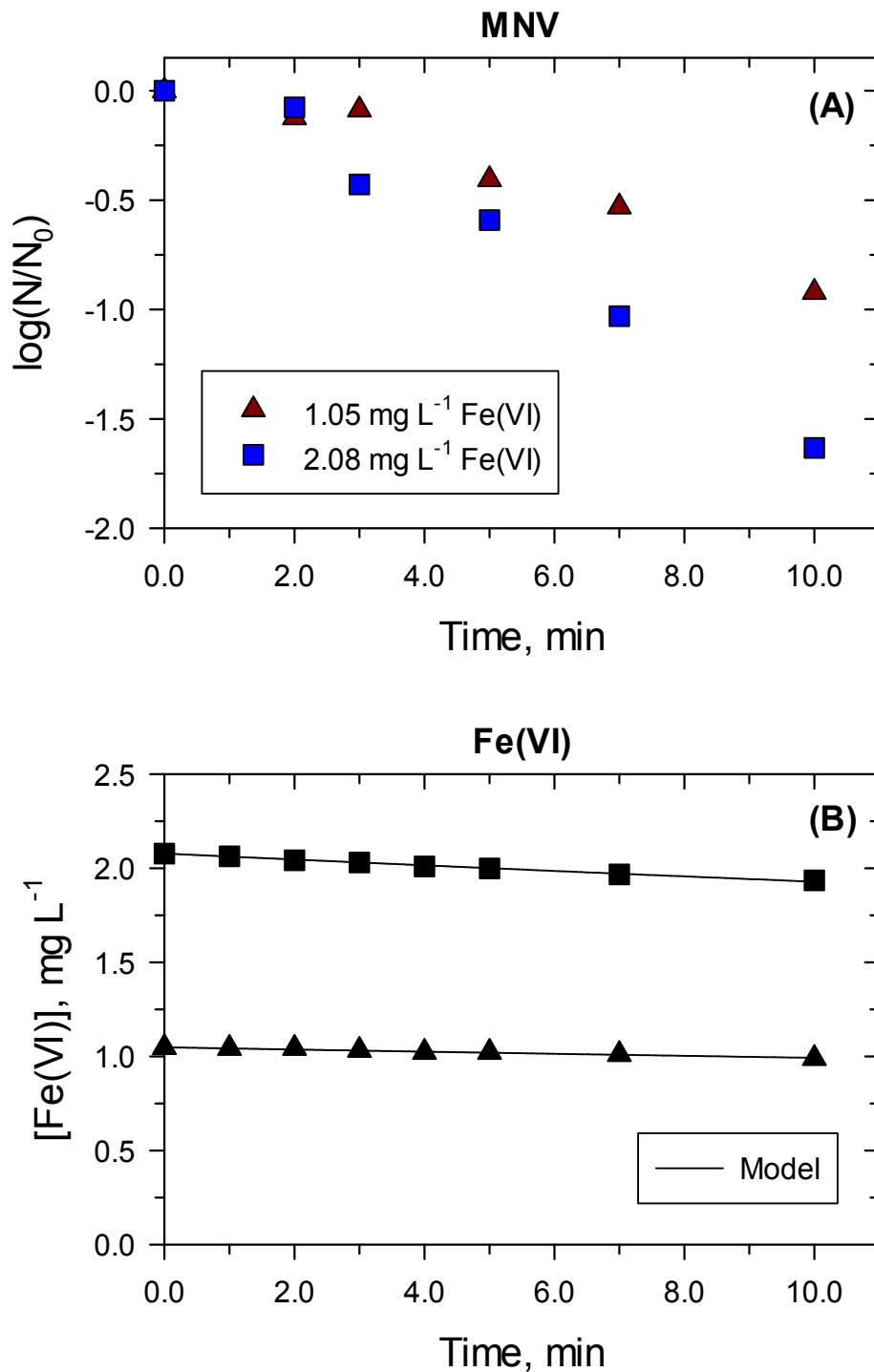40 syringe filters were acquired from VWR International (Mississauga, Ontario, Canada).

41 **Fe(VI) Preparation and Quantification.** The wet chemical method was used to synthesize solid

42 potassium ferrate(VI) (K_2FeO_4; 98% purity).[1] Fe(VI) solutions were prepared prior to each

43 experiment by diluting the desired amount (4-8 mg) of solid $K_2Fe^{VI}O_4$ to DI water (12-15 mL)

44 followed by centrifugation at 3400 rpm and 23±1 °C. The concentration of Fe(VI) in the solution

45 was determined spectrophotometrically by measuring the absorbance at 510 nm using molar

46 absorptivity ($\varepsilon_{pH\ 6.2}$ = 476 M^{-1} cm^{-1}, $\varepsilon_{pH\ 7.2}$ = 663 M^{-1} cm^{-1}, $\varepsilon_{pH\ 7.7}$ = 908 M^{-1} cm^{-1}, $\varepsilon_{pH\ 8.2}$ = 1050

47 M^{-1} cm^{-1} and $\varepsilon_{pH\ 9}$ = 1150 M^{-1} cm^{-1}).[2,3]

48 **Analytical Methods.** The standard membrane filtration method (9222D) was applied to measure

49 the concentration (colony-forming units (CFU) 100 mL^{-1}) of fecal coliforms (FC) in secondary

50 effluent wastewater (SEW).[4] Levels of chemical oxygen demand (dichromate method;

51 DOC316.53.01099) and nitrogen (s-TKN method; DOC316.53.01258) in SEW (Table S1) were

52 measured by performing the USEPA-approved Hach methods and testing kits (Hach Odyssey

DR/2500; Hach, Loveland, Colorado, USA). Solids-related parameters (Table S1) of the SEW were measured according to the standard methods.[4] The UV transmittance (UVT) at 254 nm was determined using a REALUVT meter (REALTECH, Whitby, Ontario, Canada), and the absorbance at 400 nm was measured using the Hach spectrophotometer. Turbidity was measured by a Hach 2100AN Turbidimeter (Table S1). Fisher Scientific accumet™ Portable pH (AP62) and conductivity (AP65) Meters were used to measure the pH and conductivity, respectively.

REFERENCES

(1)   Sharma, V. K.; Zboril, R.; Varma, R. S. Ferrates: greener oxidants with multimodal action in water treatment technologies. *Acc. Chem. Res.* **2015**, *48* (2), 182–191.

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(3)   Luo, Z.; Strouse, M.; Jiang, J.-Q.; Sharma, V. K. Methodologies for the analytical determination of ferrate(VI): a review. *J. Environ. Sci. Health. A. Tox. Hazard. Subst. Environ. Eng.* **2011**, *46* (5), 453–460.

(4)   APHA. Standard methods for the examination of water and wastewater. In; American Public Health Association: Washington D.C., 1998.

Figure S1. (A) Effect of Fe(VI) concentration on inactivation of MNV with time, and (B) Decay of Fe(VI) with time at different concentrations of Fe(VI) (data fitted by eq 4). (Experimental conditions: $[MNV]_{1.05 \text{ mg L}^{-1}} = 917$ PFU mL^{-1}, $[MNV]_{2.08 \text{ mg L}^{-1}} = 429$ PFU mL^{-1}, 0.01 M phosphate buffer (pH = 8.2±0.1), T = 23±1 °C).

MNV

legend:
- 1.05 mg L^{-1} Fe(VI)
- 2.08 mg L^{-1} Fe(VI)
- Model

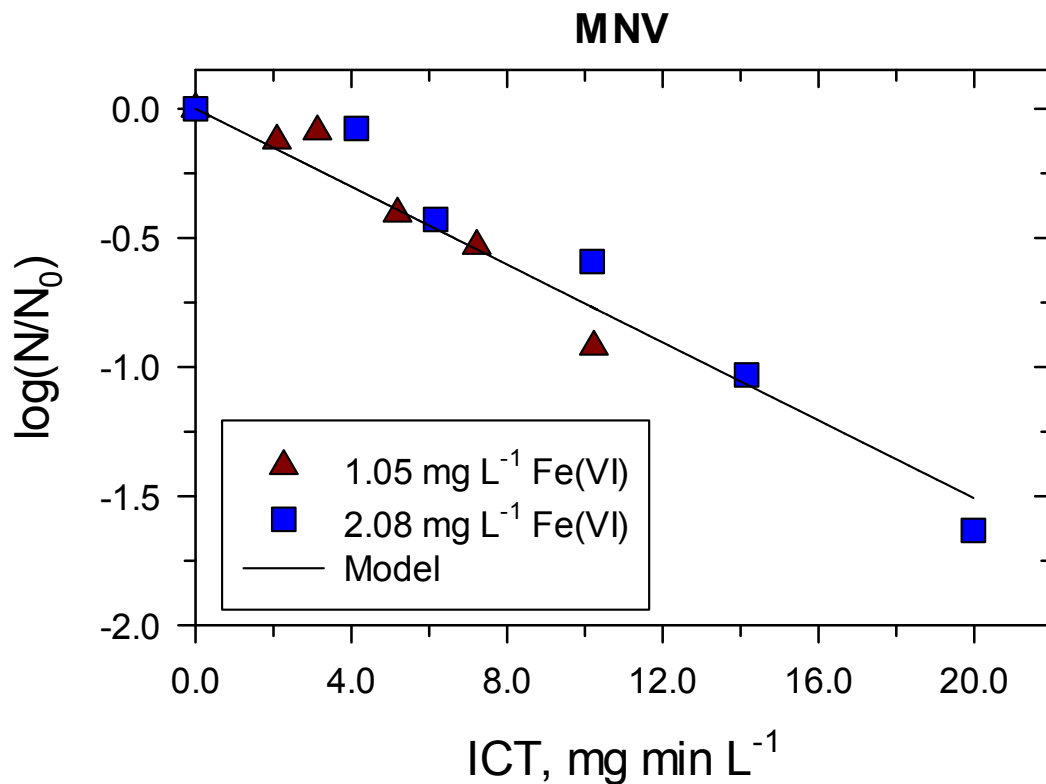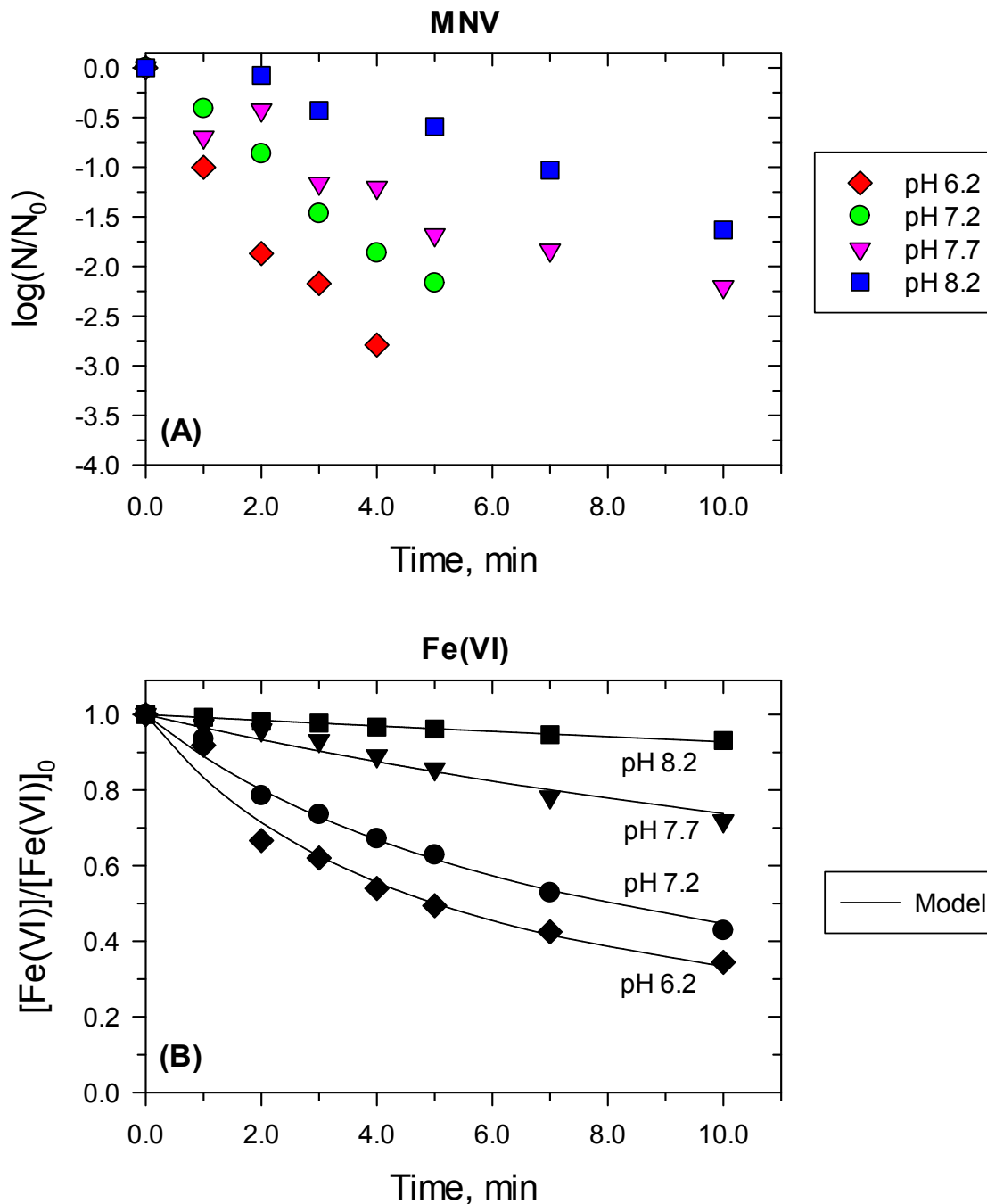y-axis: log(N/N$_0$)

x-axis: ICT, mg min L^{-1}

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Figure S2. ICT dose response curve for the inactivation of MNV by Fe(VI) (Fitting data of Figure S1(A) using Chick-Watson model (eq 1) (ICT calculated using eq 5). (Experimental conditions: $[MNV]_{1.05\ mg\ L^{-1}} = 917$ PFU mL^{-1}, $[MNV]_{2.08\ mg\ L^{-1}} = 429$ PFU mL^{-1}, 0.01 M phosphate buffer (pH = 8.2±0.1), T = 23±1 °C).

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Figure S3. (A) Effect of pH on inactivation of MNV by Fe(VI) with time, and (B) Decay of Fe(VI) with time at different pH (data fitted by eq 4), in 0.01 M phosphate buffer (PB). (Experimental conditions: $[MNV]_{pH=6.2}$ = 37126 PFU mL^{-1}, $[MNV]_{pH=7.2}$ = 1467 PFU mL^{-1}, $[MNV]_{pH=7.7}$ = 4800 PFU mL^{-1}, $[MNV]_{pH=8.2}$ = 429 PFU mL^{-1}, $[Fe(VI)]_{pH=6.2}$ = 2.04 mg L^{-1}, $[Fe(VI)]_{pH=7.2}$ = 2.36 mg L^{-1}, $[Fe(VI)]_{pH=7.7}$ = 2.14 mg L^{-1}, $[Fe(VI)]_{pH=8.2}$ = 2.08 mg L^{-1}, and T = 23±1 °C).
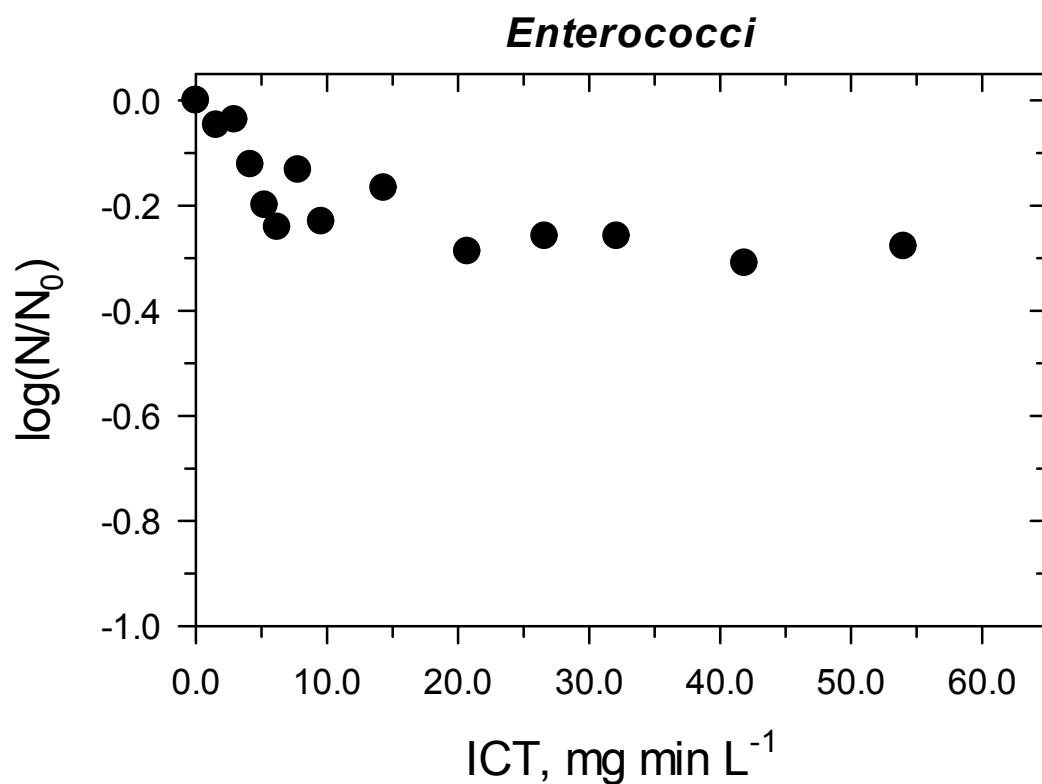
Figure S4. ICT dose response for the inactivation of enterococci by Fe(VI), in secondary effluent wastewater (SEW). (Experimental conditions: [Enterococci] = 3798 CFU 100 mL^{-1}, [Fe(VI)] = 1.16-8.29 mg L^{-1}, pH = 8.2±0.2, T = 23±1 °C).

Table S1. Water quality characteristics of the secondary effluent wastewater (SEW) used in the study.

Parameter	Value[a]
Fecal coliforms, FC (CFU 100 mL^{-1})	11440 ± 303
pH	8.2 ± 0.2
Conductivity, (μS cm^{-1})	1017 ± 14
UV$_{400}$, (cm^{-1})[b]	0.012 ± 0.001
UVT$_{254}$, (%)	77.1 ± 0.1
UV$_{254}$, (cm^{-1})	0.113 ± 0.001
Chemical Oxygen Demand, COD (mg L^{-1})	37 ± 3
Soluble Chemical Oxygen Demand, SCOD (mg L^{-1})	24 ± 6
Total Nitrogen, TN (mg N L^{-1})	13.3 ± 0.2
Soluble Total Nitrogen, STN (mg N L^{-1})	12.0 ± 0.4
Total Kjeldahl Nitrogen, TKN (mg N L^{-1})	2.7 ± 0.2
Nitrites and Nitrates, NO$_2^-$–N + NO$_3^-$–N (mg N L^{-1})	10.6 ± 0.1
Particulate Organic Nitrogen, (mg N L^{-1})	1.3 ± 0.4
Ammonia + Soluble Organic Nitrogen (mg N L^{-1})	1.4 ± 0.4
Total Phosphorus, TP (mg P L^{-1})[c]	0.45 ± 0.13
Turbidity, (NTU)	5.0 ± 0.1
Total Solids, TS (mg L^{-1})	653 ± 10
Total Suspended Solids, TSS (mg L^{-1})	13 ± 3
Total Dissolved Solids, TDS (mg L^{-1})	640 ± 10

[a]Average ± standard deviation of triplicate analysis of the grab sampled SEW

[b]Indication of color

[c]Measured by the wastewater treatment plant at the same day we collected the SEW

105 **Table S2.** Kinetic parameters determined in this study for the inactivation of MNV by Fe(VI)
106 (k_d) and the decomposition of Fe(VI) (k_2) in 0.01 M phosphate buffer.

pH	[MNV] (PFU mL^{-1})	[Fe(VI)] (mg L^{-1})	k_2, 10^{-1} (L mg^{-1} min^{-1})	k_d, 10^{-1} (L mg^{-1} min^{-1})
6.2	37126	2.04	0.98±0.06	4.80±0.17
7.2	935	1.16	0.49±0.02	2.23±0.06
7.2	1467	2.36	0.53±0.02	
7.7	4800	2.14	0.17±0.01	1.39±0.10
8.2	917	1.05	0.05±0.002	0.75±0.04
8.2	429	2.08	0.04±0.001	

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Table S3. Kinetic parameters determined in this study for inactivating MNV and FC by Fe(VI) in secondary effluent wastewater.

pH	[MNV] (PFU mL⁻¹)	[Fe(VI)] (mg L⁻¹)	[FC] (CFU (100 mL)⁻¹)	D (mg L⁻¹)	k_1 (min⁻¹)	MNV[a] k_d, 10⁻¹ (L mg⁻¹ min⁻¹)	FC[b] β	k_d (Lᵐ mg⁻ᵐ min⁻ᵐ)	m	k_p (L mg⁻¹ min⁻¹)
8.2	-	1.16	11372	0.21±0.04	0.37±0.04	-				
8.2	32327	2.06	11771	0.41±0.09	0.12±0.02	1.35±0.04	0.017±0.003	2.08±0.05	1.08±0.06	0.03±0.018
8.2	24123	8.29	11177	0.57±0.16	0.08±0.004					

[a]MNV: Murine norovirus; [b]FC: Fecal coliforms

Table S4. Model predicted ICT values required for 1-, 2-, 3-, and 4-$\log_{10}$ reduction of murine norovirus (MNV) and fecal coliforms (FC) by Fe(VI) in secondary effluent wastewater.

Reduction	ICT (mg min L^{-1})	
	FC	MNV
1-$\log_{10}$	1	7
2-$\log_{10}$	17	15
3-$\log_{10}$	N/A	22
4-$\log_{10}$	N/A	30