Supporting Information

EPR Detection of Nitrogen-Containing Radicals in Dimethylamine Oxidation: Insights into NDMA Formation

Sifan Qiu[1], Chunxiao Xu[1], Yu Fu[1], Lingli Wang[1], Pu Wang[1], Jinhui Cao[1], Yanna Xue[1], Zhaohui Wang[1,2,3*]

[1] Shanghai Key Lab for Urban Ecological Processes and Eco-Restoration, School of Ecological and Environmental Sciences, East China Normal University, Shanghai 200241, China;

[2] Technology Innovation Center for Land Spatial Eco-restoration in Metropolitan Area, Ministry of Natural Resources, 3663 N. Zhongshan Road, Shanghai 200062, China;

[3] Shanghai Engineering Research Center of Biotransformation of Organic Solid Waste, Shanghai 200241, China.

*Corresponding author:
Email: zhwang@des.ecnu.edu.cn (Z. Wang)

Summary: 2 Text, 11 Figures, 3 Schemes.

Text S1

Dimethylamine (C_2H_7N, DMA, 40 wt.% solution in water, RG), dimethylamine (C_2H_7N, DMA (THF), 2 M solution in tetrahydrofuran, safeseal), N,N-dimethylhydroxylamine hydrochloride (C_2H_8ClNO, DIMHA·HCl, purity 99%, RG), phenyl-4,4,5,5-tetramethylimidazoline-1-oxyl 3-oxide (PTIO, purity 98%, RG), and potassium peroxymonosulfate ($KHSO_5$, PMS, purity > 98%) were supplied by Adamas-Beta Co. Ltd (Shanghai, China). Reagents including dimethylamine (C_2H_7N, DMA, 40 wt.% solution in water, standard for GC) and dimethylamine (C_2H_7N, DMA, 40 wt.% solution in water, AR) were supplied by Aladdin Chemical Reagent Co. Ltd (Shanghai, China). H_3PO_4 (85 wt.% solution in water, GR) and K_3PO_4 (AR) were supplied by General-Reagent Co. Ltd (Shanghai, China). KOH (AR) and $KMnO_4$ (AR) were supplied by Shanghai Lingfeng Chemical Reagent Co. Ltd (Shanghai, China). NaClO (Available chlorine ≤ 40%) was supplied by TCI (Shanghai) Development Co., Ltd. Hydrogen peroxide (H_2O_2, 30% aqueous solution, AR) was supplied by Sinopharm Chemical Reagent Co. Ltd (Shanghai, China). Oxygen-free pure water was obtained by Xiamen Haibiao Technology Co. Ltd (Xiamen, China).

Text S2

All reagent solutions were prepared with ultrapure water (18.25 MΩ·cm). The DMA aqueous solutions used in this study were prepared from 40% DMA aqueous solution, and the DMA (THF) was prepared from a 2 M tetrahydrofuran (THF) DMA solution. Moreover, the DMA solutions used in this study were analyzed by mass spectrometry, and no DMA oxidation products were detected, indicating that the solution does not affect the accuracy of the subsequent experiments.

Sample Preparations. In a sequential procedure, 45 μL of 0.4 M K_3PO_4, 135 μL of 1 M DMA, and 45 μL of 200 mM NaClO were successively introduced into a centrifuge tube. The resulting mixture was then transferred to an EPR instrument using capillary tubes for subsequent detection. Likewise, for investigating the impact of oxidizing agents on EPR signals, NaClO was replaced with PMS and $KMnO_4$, and the procedure was reiterated. To further investigate the presence of nitric oxide radical ($NO^•$) in the solution, an additional 10 μL of 0.1 M PTIO was introduced to the mixture to effectively capture $NO^•$. DMA solutions were titrated with H_3PO_4 or KOH solutions to adjust the pH, using a PHSJ-3F laboratory digital pH-meter.

Analytical Methods. EPR spectra were recorded on an EPR spectrometer (Bruker ESR 5000; Germany). Before testing, the EPR instrument underwent a 10-minute preheating phase. Approximately 70 μL of the mixed sample was then drawn into a 1.0 mm inner diameter glass capillary. Followed by sample loading, one end of the capillary was sealed using a wax sealing plate. Subsequently, the sealed capillary was positioned within the guide tube of the EPR instrument and firmly placed inside the EPR resonance cavity. The instrument conditions were as follows: frequency, 100 kHz; width, 0.2 mT; magnetic field, 330-345 mT; sweep time, 60 s; microwave power, 10 mW; microwave frequency, 9.46 GHz. The detected EPR signals were identified by WINSIM software simulation according to the fitted hyperfine splitting constants.[32]

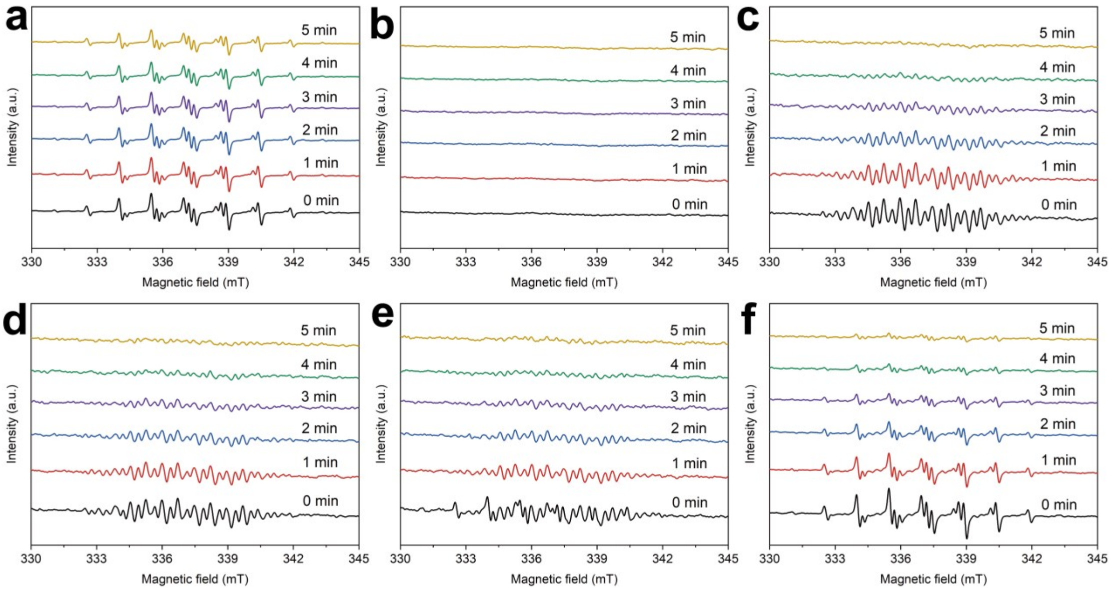


Figure S1. Time-dependent EPR spectra for (a) DMA, (b) NaClO, and the DMA/NaClO system with NaClO concentrations of (c) 40 mM, (d) 20 mM, (e) 12 mM, and (f) 1 mM. Conditions: [DMA] = 200 mM; [K_3PO_4] = 80 mM; pH = 12.5.

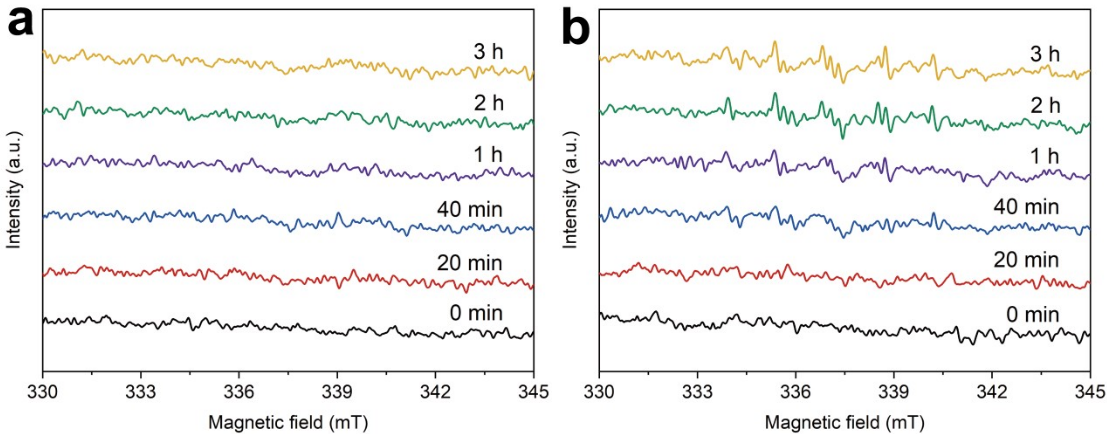


Figure S2. The temporal evolution of EPR spectra for (a) DMA (THF) and (b) DMA (THF) in oxic water. Conditions: [DMA (THF)] = 1 M; pH = 7.0. As illustrated in Figure S2, sealed DMA (THF) exhibits no signals within 3 hours, whereas DMA (THF) diluted with oxic water under sealed conditions gradually manifested DMNO$^{\bullet}$ between 1 and 3 hours. This observation confirms that dissolved oxygen in water oxidizes DMA, resulting in the generation of DMNO$^{\bullet}$.

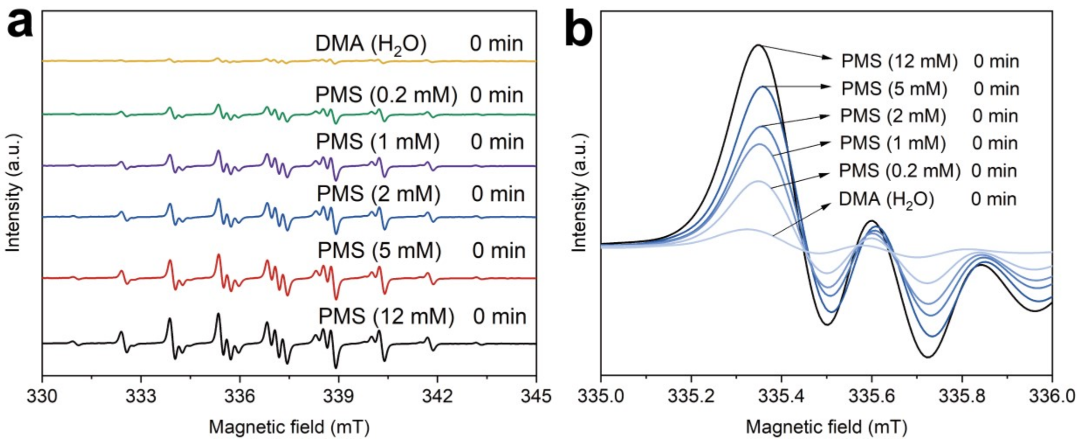


Figure S3. (a) EPR spectra during the instantaneous reaction of different concentrations of PMS and DMA, and (b) spectral comparison in the magnetic field range of 335 - 336 mT. Conditions: [DMA] = 200 mM; [K_3PO_4] = 80 mM; pH = 5.0.

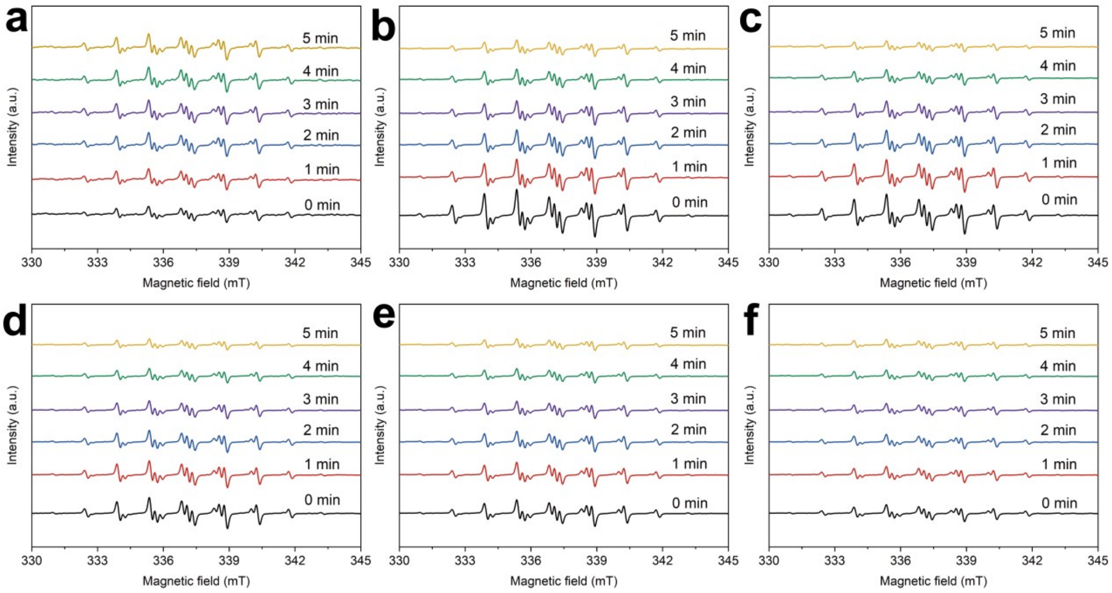


Figure S4. EPR spectra of (a) DMA and its respective reactions with PMS at concentrations of (b) 12 mM, (c) 5 mM, (d) 2 mM, (e) 1 mM, and (f) 0.2 mM. Conditions: [DMA] = 200 mM; [K_3PO_4] = 80 mM; pH = 5.0.

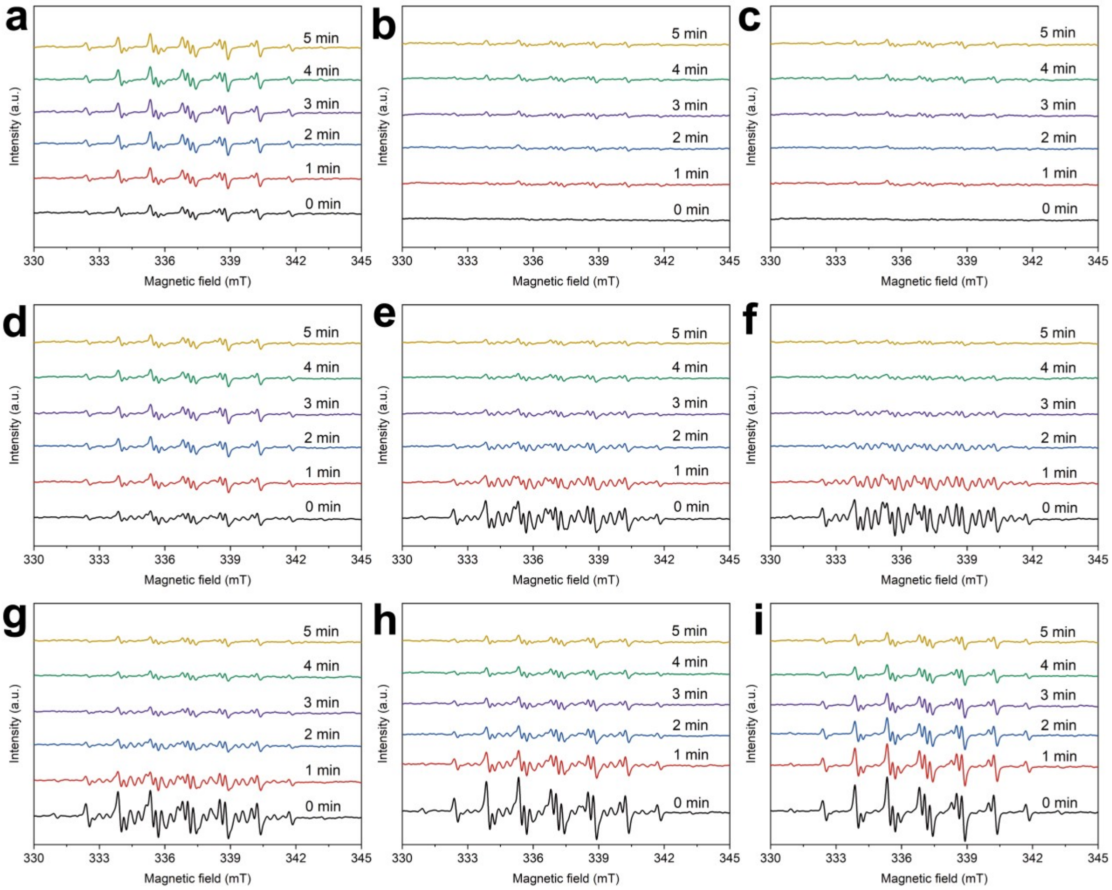


Figure S5. EPR spectra of (a) DMA and its respective reactions with $KMnO_4$ at concentrations of (b) 12 mM, (c) 5 mM, (d) 2 mM, (e) 1 mM, (f) 0.5 mM, (g) 0.2 mM, (h) 0.1 mM, and (i) 20 μM. Conditions: [DMA] = 200 mM; [K_3PO_4] = 80 mM; pH = 6.0.

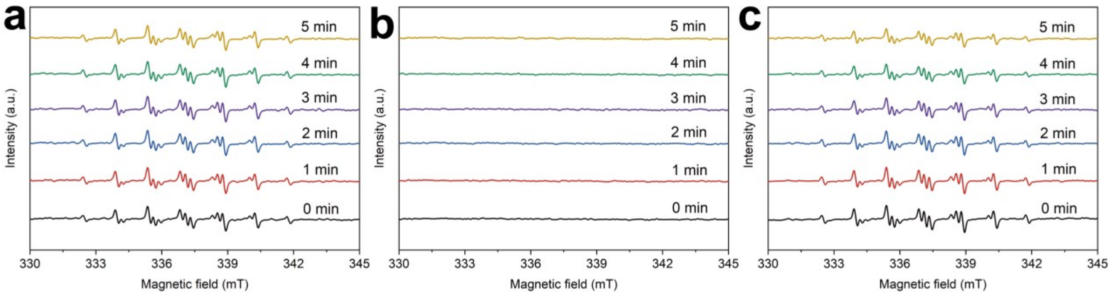


Figure S6. The EPR spectra of (a) DMA under neutral conditions, (b) at pH 3.0 with the addition of H_3PO_4, and (c) upon subsequent pH adjustment to 12.3 with KOH. Condition: [DMA] = 200 mM.

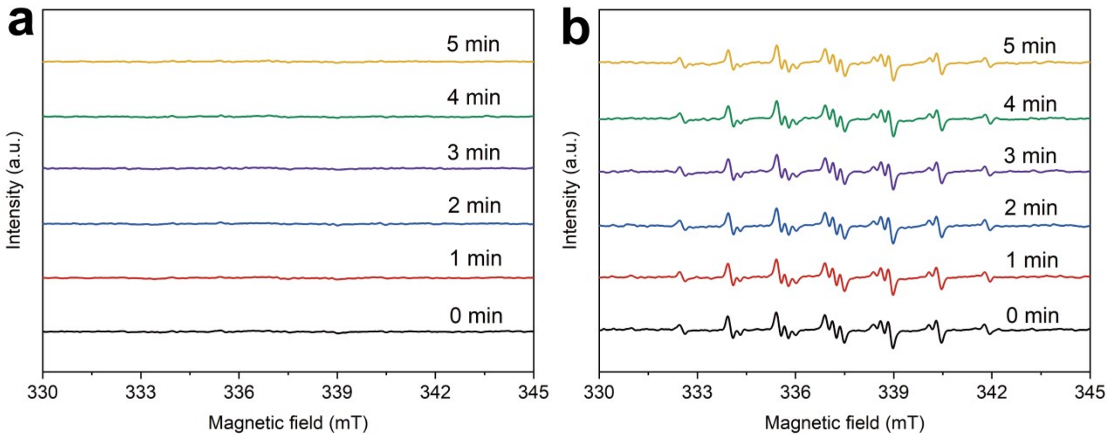


Figure S7. EPR spectra of (a) DIMHA·HCl under neutral conditions and (b) at pH 12.5 with the addition of KOH. Condition: [DIMHA·HCl] = 200 mM.

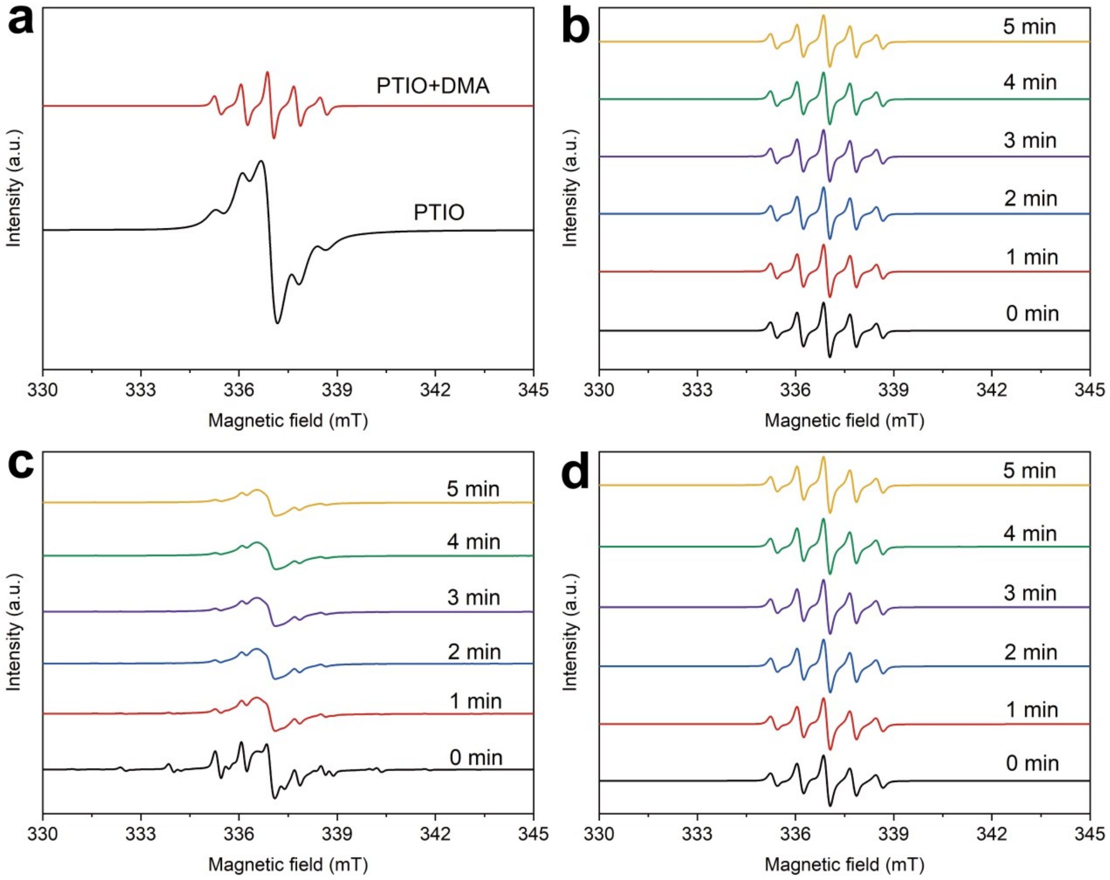


Figure S8. EPR spectra of (a) PTIO alone and PTIO added to the DMA, (b) PTIO added to the DMA/NaClO system, (c) PTIO added to the DMA/PMS system, and (d) PTIO added to the DMA/$KMnO_4$ system. Conditions: [DMA] = 200 mM; [K_3PO_4] = 80 mM; [NaClO] = 200 mM; [$KMnO_4$] = 0.5 mM; [PMS] = 12 mM. As shown in Figure S8a, free radical signals were detected in both PTIO and DMA/PTIO mixtures. Simulation of these signals indicated that they correspond to PTIO (α_N = 8.2 G (×2)). Upon adding PTIO to the DMA/NaClO system, the simulation of the signals in Figure S8b still indicates PTIO, with no evidence of imino nitroxide 2-phenyl-4,4,5,5-tetramethylimidazoline 1-oxyl (PTI) signals.

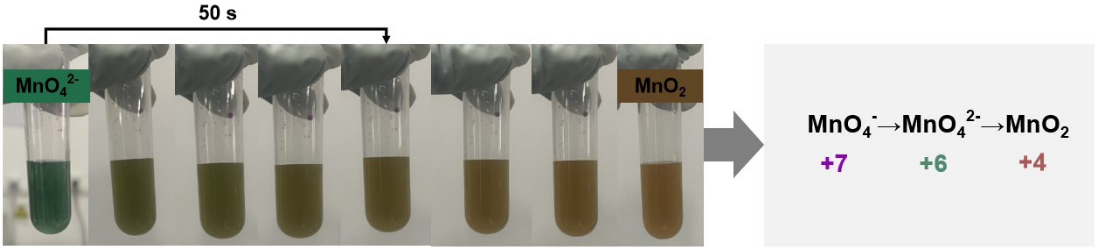


Figure S9. Color change of solution when $KMnO_4$ is immediately added to DMA.

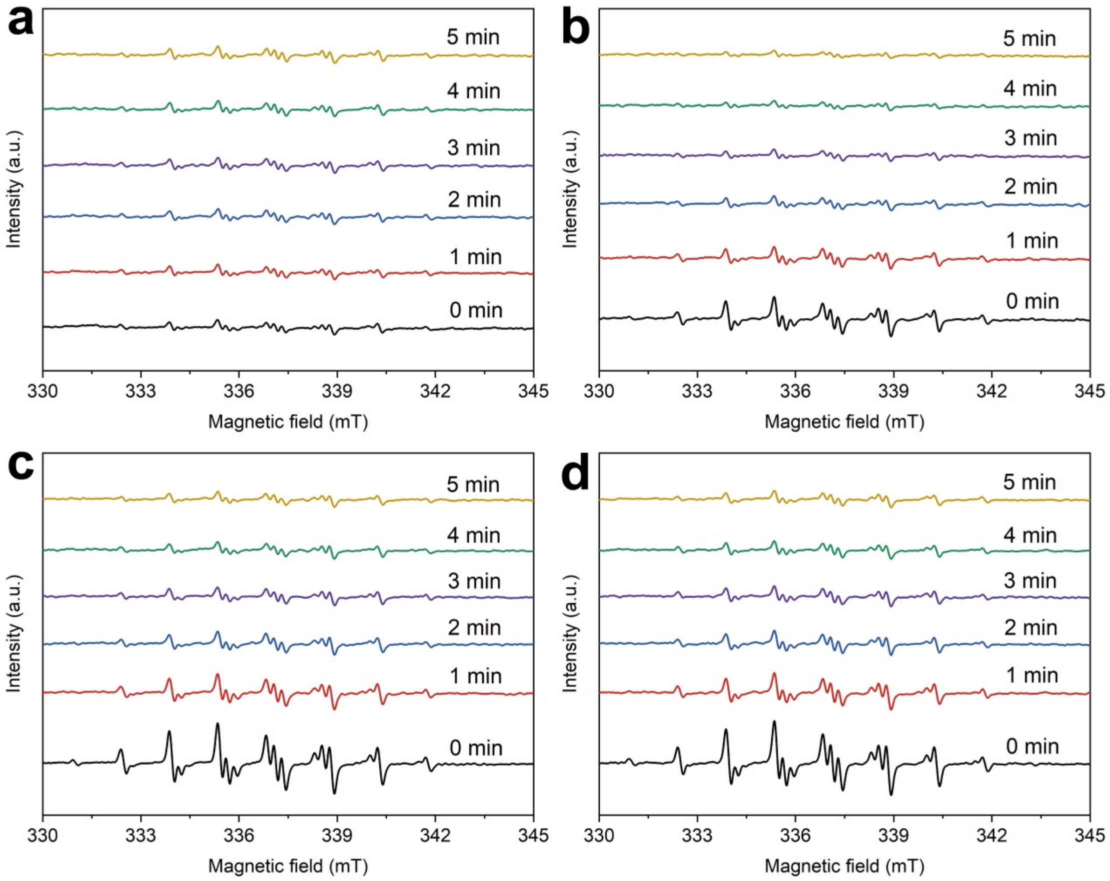


Figure S10. EPR spectra of (a) DMA and the reaction between $KMnO_4$ and DMA are shown at pH 7.0 with initial concentrations of (d) 1 mM, (c) 2.5 mM, and (b) 5 mM. Conditions: [DMA] = 200 mM; $[K_3PO_4]$ = 80 mM.

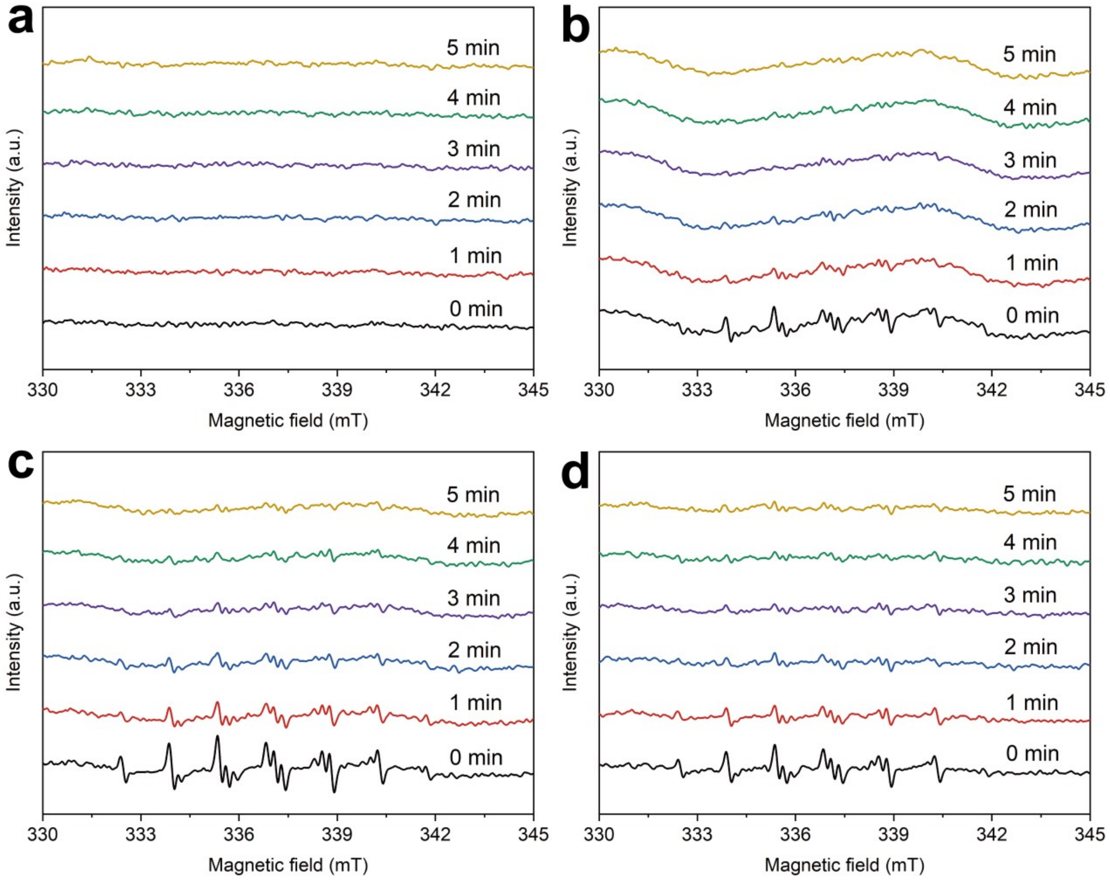
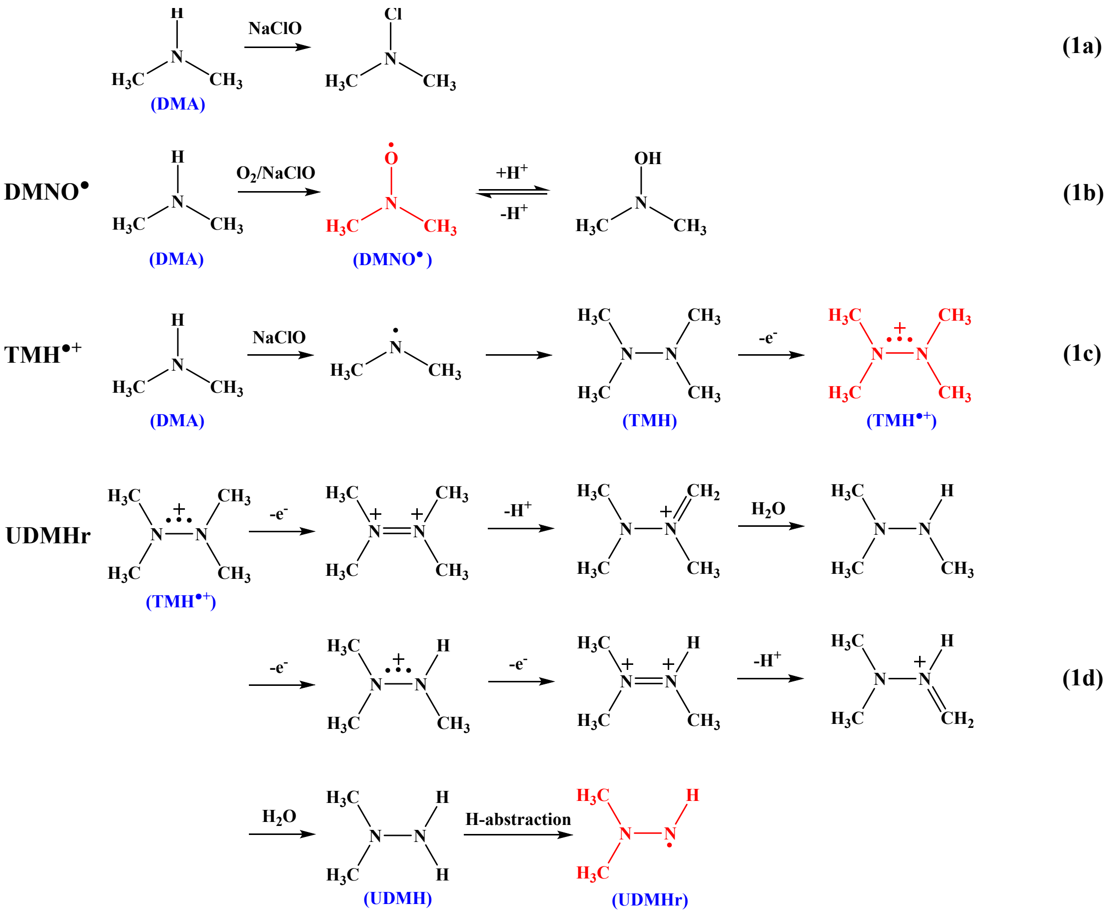
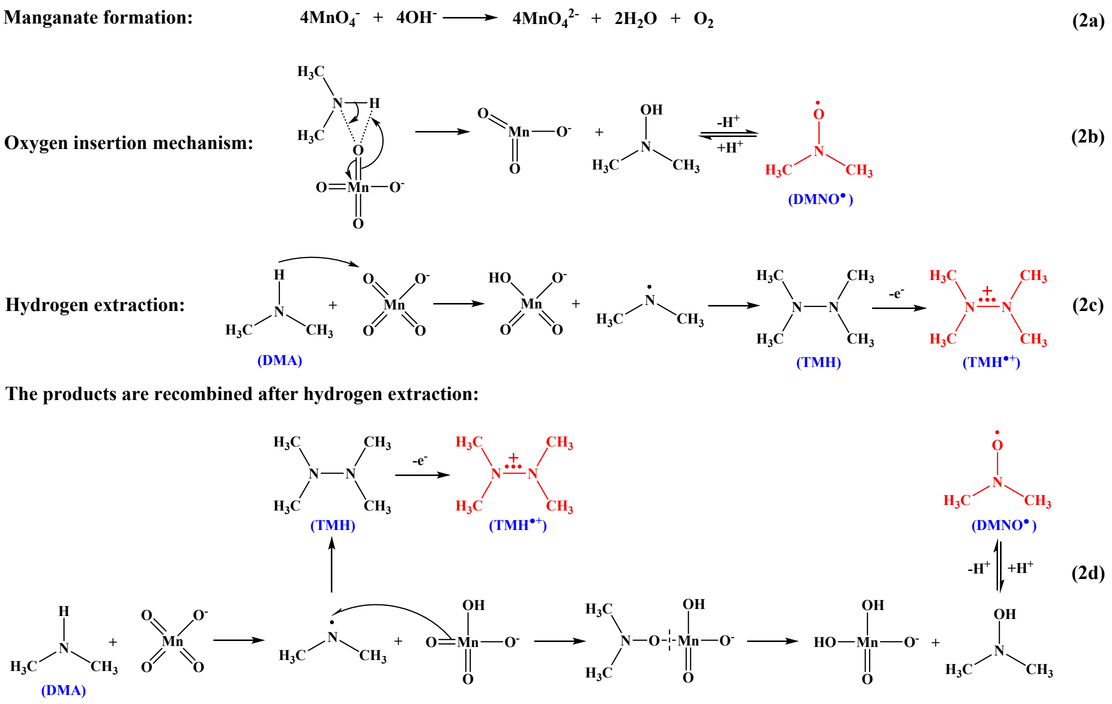


Figure S11. EPR spectra of (a) DMA and the reaction between $KMnO_4$ and DMA are shown at pH 2.5 with initial concentrations of (d) 1 mM, (c) 2.5 mM, and (b) 5 mM. Conditions: [DMA] = 200 mM; $[K_3PO_4]$ = 80 mM.

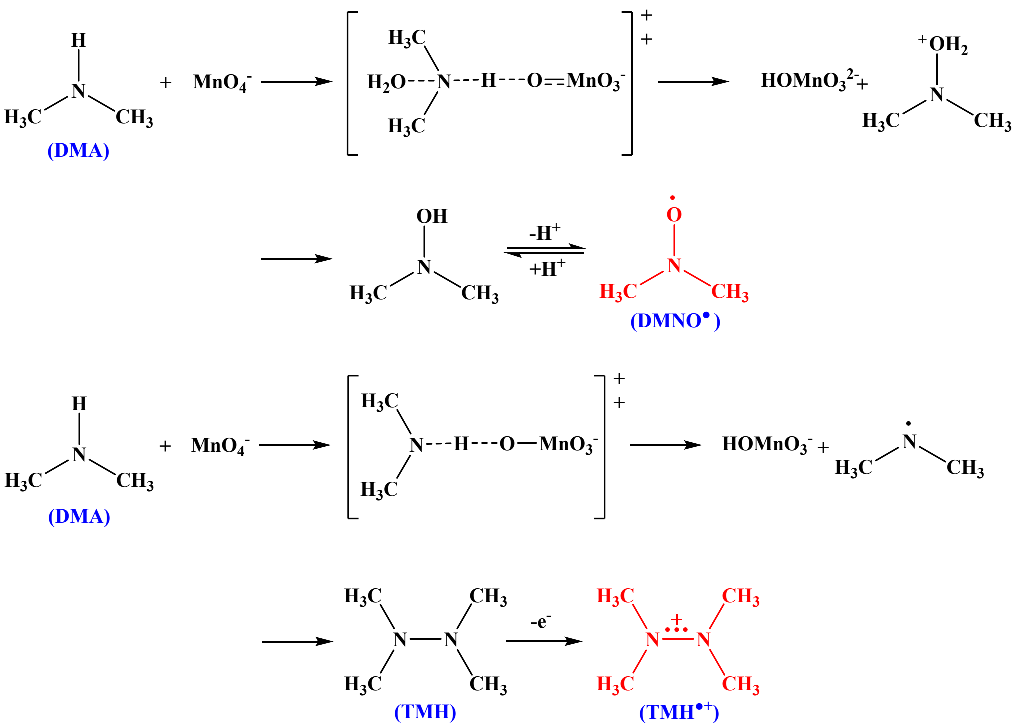


Scheme S1. Formation mechanisms of DMNO$^{•}$, TMH$^{•+}$, and UDMHr in DMA/NaClO system.



$4MnO_4^- + 4OH^- \longrightarrow 4MnO_4^{2-} + 2H_2O + O_2$ (2a)

Scheme S2. The first possible formation mechanism of $DMNO^{\bullet}$ and $TMH^{\bullet+}$ in DMA/$KMnO_4$ system.



(3a)

(3b)

Scheme S3. The second possible formation mechanism of DMNO$^\bullet$ and TMH$^{\bullet+}$ in DMA/KMnO$_4$ system.