**Supplementary Materials**

Degradation of 1H-benzotriazole by UV/H2O2 and UV/TiO2:

Kinetics, mechanisms, products and toxicology

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# Text S1. Chemical reagents.

Crystal ascorbic acid, Tris-HCl, thiourea, dithiothreitol (DTT), 3-[(3-Cholamidopropyl) dimethylammonio] propanesulfonate (CHAPS), phenylmethylsulfonyl fluoride (PMSF) and nuclease were purchased from Sigma-Aldrich (USA). IPG buffer and trypsin were obtained from GE Company and Promega (V5280, USA), respectively. HPLC grade acetonitrile, formic acid, tert-butyl alcohol and ethyl alcohol were purchased from Fisher (USA). All reagents were stored at 4 °C or -20 °C as demand, and were used as received.

# Text S2. Qualitative and quantitative analysis of 1H-BTA and its intermediates.

**1. HPLC separation**

The samples were injected into a LC-30AD liquid chromatograph system (Shimadzu, Japan) with a Waters Symmetry C-18 column (2.1 × 150 mm, 3.5 µm) prior to the MS analysis. The injection volume was 10 µL, and the mobile phase was a gradient elution of 0.1% formic acid water solution (mobile phase A) and acetonitrile (mobile phase B). The gradient elution was programmed as follows: 0-1.0 min, 10% B; 1.0-2.0 min, 10%-90% B; 2.0-4.0 min, 90% B; 4.0-4.1 min, 90%-10% B; 4.1-6.0 min, 10% B (40°C, 0.3 mL min-1).

**2. Transformation products analysis**

The identification of intermediates was performed using a TripleTOF 5600+ high-resolution tandem mass spectrum (HRMS) (Applied Biosystems SCIEX, USA) equipped with a Turbo V ESI ion source and a triple quadrupole time-of-flight (TOF) device. The instrumentation conditions are listed in Table S1. Nitrogen served both as the turbo and the collision gas. Mass calibrations and resolution adjustments on the quadrupoles and TOF were performed automatically using a 10-5 M solution of polypropylene glycol introduced via a model II Harvard infusion pump. The scan range was set at *m/z* 50-600. The data were analyzed using PeakView and MasterView (Applied Biosystems SCIEX, USA).

**3. Quantitative analyses of 1H-BTA and its products**

The concentrations of 1H-BTA and its degradation products were verified using a TripleQuad 5500 tandem mass spectrum (Applied Biosystems SCIEX, USA). Most of the instrumentation conditions were similar to those of the 5600+ system, but the Collision-Induced Dissociation (CID) energy was optimized for different intermediates. The scan mode was multi-reaction monitoring, and the monitoring ion pairs are listed in Table S2.

# Text S3. Actual water matrix sampling and analysis

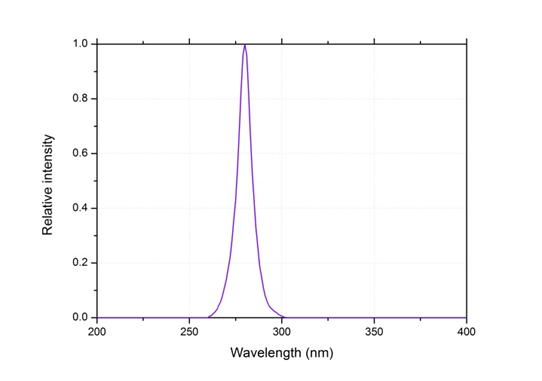
The turbidity was measured using a 1900c turbidity meter (Hach, USA). In order to investigate the impact of dissolved materials in actual water matrix on the reactions, the particles and microorganisms were removed by filtration using 0.7 μm glass fiber filter (Whatman, USA) followed by 0.22 μm polyether sulfone filter (Anpel, China). And then the basic parameters, including total organic carbon (TOC), total nitrogen (TN) and UV254, were measured. The TOC and TN were measured using a TOC-L analyzer (Shimadzu, Japan), while the UV254 was obtained by a GENESYS 10S spectrophotometer (ThermoFisher, USA). The determination of anions was performed using an ICS-2500 analyzer (Dionex, USA) with an ED50A detector. A DIONEX IonPac® AS15 column was used with 30.0 mM NaOH solution as the mobile phase.

# Text S4. Scavenging studies

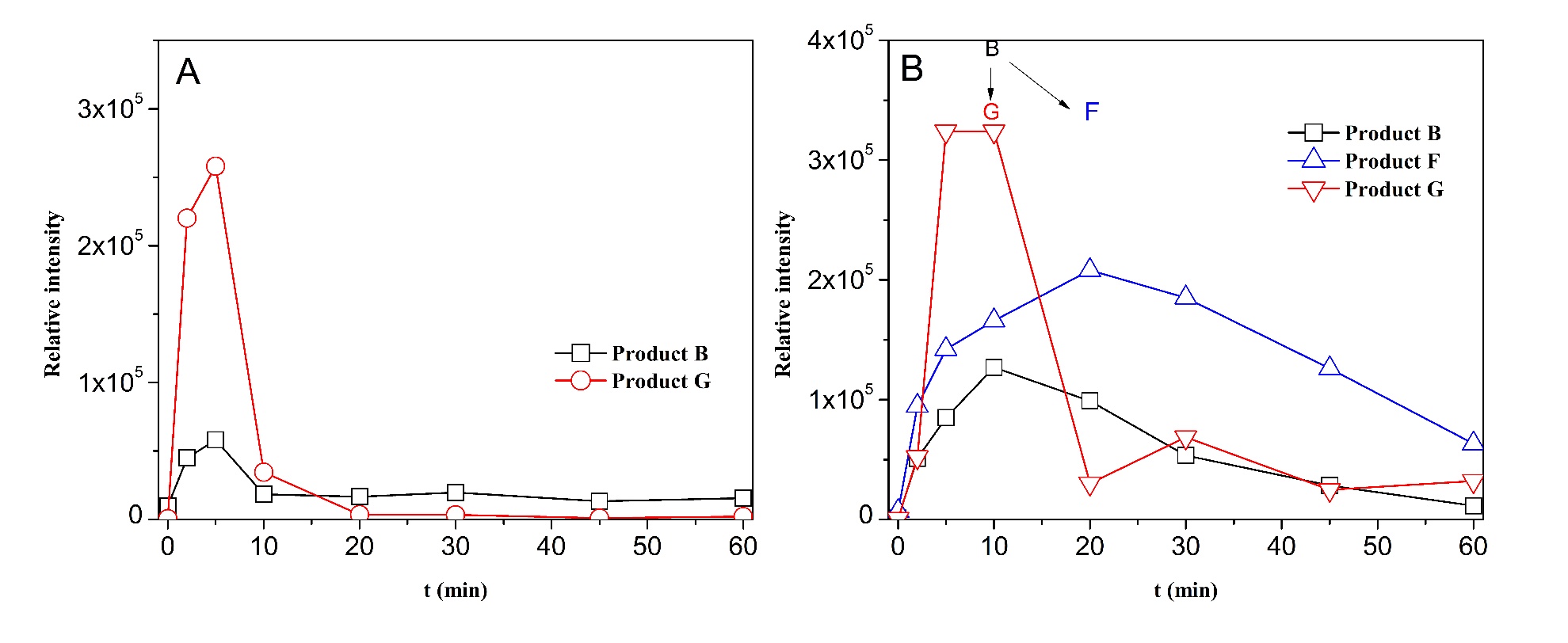
To reveal the mechanisms within this promotion, additions of typical radical scavengers were performed. Removal efficiency was severely inhibited in the existence of 10 mM ascorbic acid, and *k*app decreased from 1.63 × 10-3 s-1 to 0.11 × 10-3 s-1 (UV/H2O2), and from 1.87 × 10-3 s-1 to 0.03 × 10-3 s-1 (UV/TiO2) (Figs. 1b and 1c). Ascorbic acid is a strong reductant and can scavenge most of the oxidants, suggesting that the degradation of 1H-BT was dominated by oxidation. Still, ascorbic acid has a high *ε* (3259 ± 80 M-1 cm-1) at280 nm (Table S3). To further identify the reactive species in UV/H2O2 and UV/TiO2 system, EtOH and TBA, with reaction rate constants to •OH(*k*·OH) at 1.9 × 109 M-1 s-1 (eq. 2) and 6.0 × 108 M-1 s-1 (eq. 3) [21](#_ENREF_21), respectively, were spiked. Furthermore, *ε* of EtOH and TBA at 280 nm are 0.67 ± 0.07 M-1 cm-1 and 1.40 ± 0.10 M-1 cm-1, indicating their weak UV screening effects. The *k*app decreased from 1.63 × 10-3 s-1 to 0.64 × 10-3 s-1 (EtOH) and 0.69 × 10-3 s-1 (TBA) (Fig. 1b), indicating an inhibition of •OH oxidation in UV/H2O2 system. After EtOH and TBA were added to UV/TiO2 system, the *k*app decreased from 1.87 × 10-3 s-1 to 0.15 × 10-3 s-1 and 0.18 × 10-3 s-1, respectively (Fig. 1c). Accordingly, •OH oxidation was dominating for the 1H-BT degradation during UV/H2O2 and UV/TiO2 treatment.

EtOH + •OH → products (*k* = 1.9 × 109 M-1 s-1) (S1)

TBA + •OH → products (*k* = 6.0 × 108 M-1 s-1) (S2)

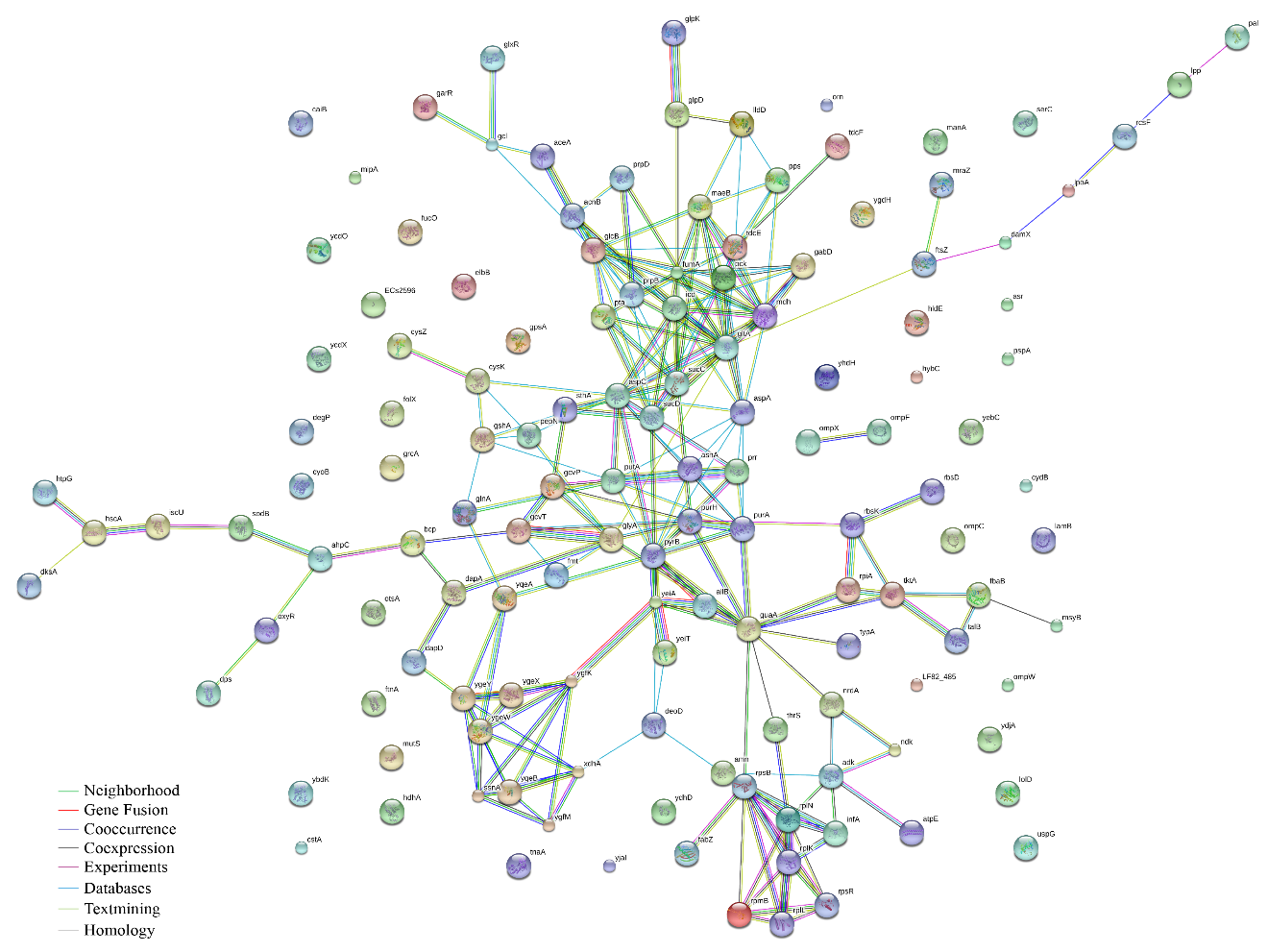


# Figure S1. UVLED module and emission spectra for 280 nm UV-LED



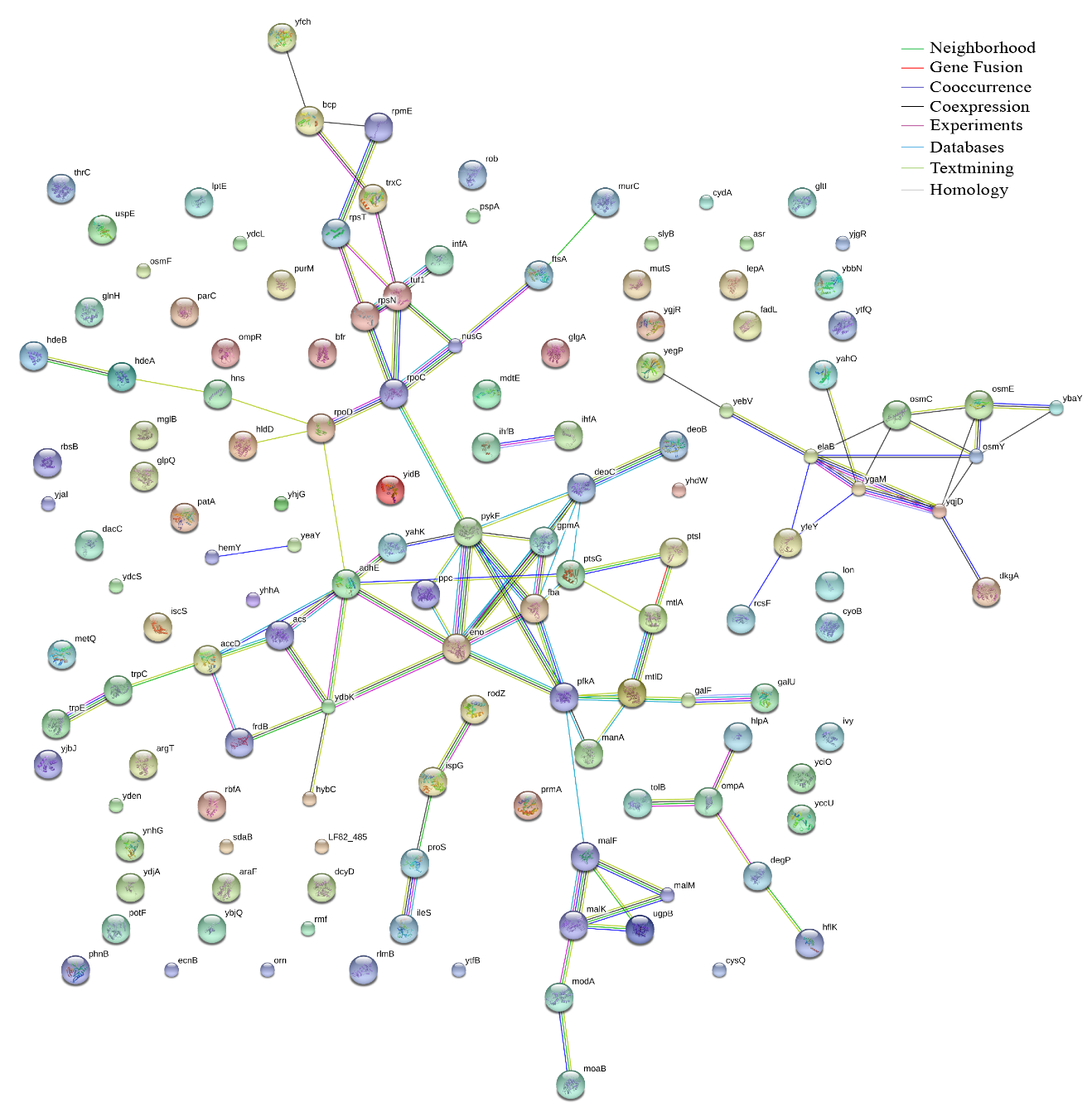
# Figure S2. Evolution tendency of several products in UV/H2O2 (a) and UV/TiO2 (b) systems.

Experimental conditions: UVLED intensity (280 nm) = 0.023 mW cm-2, solution temperature 25 ± 2°C, pH 6.5-7.2, [1H-BTA]0 = 8.39 μM, [H2O2]0 = 4.41 mM, [TiO2]0 = 2.50 mM.



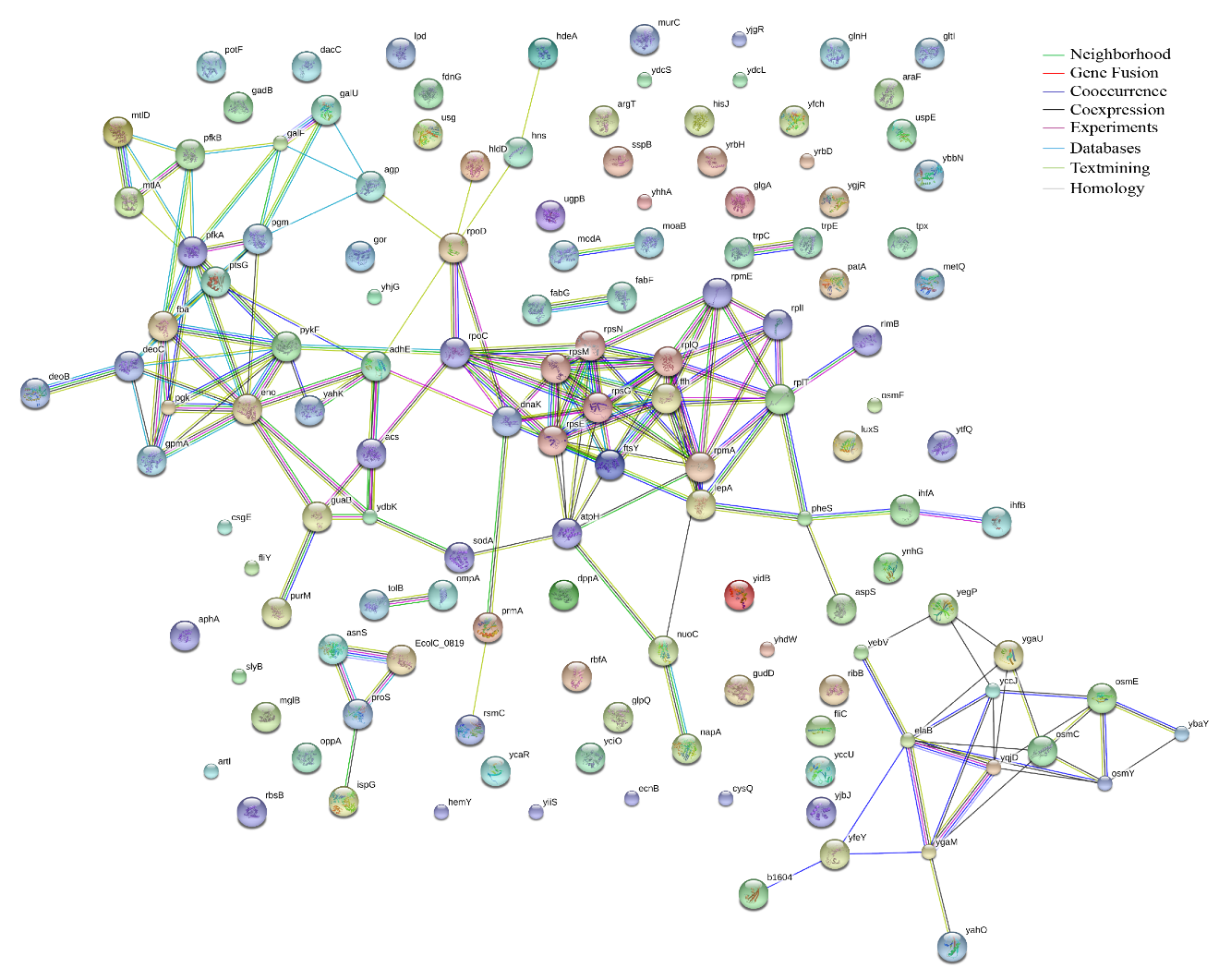
# Figure S3. Biological networks of down-regulated expressed proteins in *E. coli* after the exposure to the intermediates in UV/H2O2 treatment compared to untreated 1H-BTA.

Experimental conditions: UVLED intensity (280 nm) = 0.023 mW cm-2, solution temperature 25 ± 2°C, pH 6.5-7.2, [1H-BTA]0 = 8.39 μM, [H2O2]0 = 4.41 mM, reaction time = 30 min.



# Figure S4. Biological networks of up-regulated expressed proteins in *E. coli* after the exposure to the intermediates in UV/TiO2 treatment compared to untreated 1H-BTA.

Experimental conditions: UVLED intensity (280 nm) = 0.023 mW cm-2, solution temperature 25 ± 2°C, pH 6.5-7.2, [1H-BTA]0 = 8.39 μM, [TiO2]0 = 2.50 mM, reaction time = 30 min.



# Figure S5. Biological networks of down-regulated expressed proteins in *E. coli* after the exposure to the intermediates in UV/TiO2 treatment compared to untreated 1H-BTA.

Experimental conditions: UVLED intensity (280 nm) = 0.023 mW cm-2, solution temperature 25 ± 2°C, pH 6.5-7.2, [1H-BTA]0 = 8.39 μM, [TiO2]0 = 2.50 mM, reaction time = 30 min.

# Table S1. Operational parameters of mass spectrum.

|  |  |  |
| --- | --- | --- |
| Parameter (unit) | TripleTOF 5600+ | TripleQuad 5500 |
| Ion source mode | Positive | Positive |
| Scan mode | Full scan | Multi reaction monitoring |
| ESI needle voltage (V) | 4500 | 4500 |
| Turbo-gas temperature (°C) | 450 | 450 |
| Curtain gas pressure (psi) | 35 | 35 |
| Nebulizer gas pressure (psi) | 30 | 30 |
| Auxiliary gas pressure (psi) | 30 | 30 |
| Declustering potential (V) | 50 | 50 |
| CID energy (eV) | 30 ± 15 | See in Table S2 |

# Table S2. CID energy and monitoring ion pairs for Triple Quad 5500 MS/MS.

|  |  |  |  |
| --- | --- | --- | --- |
| Name | Molecular Formula | CID Energy (eV) | Ion Pairs |
| 1H-BTA | C6H5N3 | 45 | 120.1/64.9 |
| Product A | C6H5N3O | 25 | 136.1/80.1 |
| Product B | C6H5N3O2 | 30 | 152.0/68.1 |
| Product C | C6H5N3O3 | 35 | 168.0/66.0 |
| Product D | C5H5N3O4 | 40 | 172.0/52.0 |
| Product E | C4H3N3O4 | 20 | 158.0/140.0 |
| Product F | C6H3N3O2 | 30 | 150.0/66.0 |
| Product G | C4H3N3O2 | 30 | 126.0/80.0 |
| Product H | C4H3N3O3 | 35 | 142.0/69.0 |

# Table S3. Molar absorption coefficients of different solutions.

|  |  |
| --- | --- |
| Contents | Molar absorption coefficient |
| 280 nm (M-1 cm-1) |
| 1H-BTA | 3806 ± 216 |
| EtOH | 0.67 ± 0.07 |
| TBA | 1.40 ± 0.10 |
| ascorbic acid | 3259 ± 80 |
| OH- | 22 ± 3 |
| Cl- | 252 ± 28 |
| NO3- | 139 ± 41 |

# Table S4. Quantifiable identified proteins in *Escherichia coli* after exposure to 1H-BTA or its products

See in Supplementary Materials (Part II).

# Table S5. KEGG enrichment of significantly up- and down-regulated proteins after exposure to the intermediate mixtures compared to 1H-BTA

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| KEGG pathways | Up-regulated proteins  UV/H2O2 : 1H-BTA | Down-regulated proteins  UV/H2O2 : 1H-BTA | Up-regulated proteins  UV/TiO2 : 1H-BTA | Down-regulated proteins  UV/TiO2 : 1H-BTA |
| Metabolic pathways | aceA, acnB, allB, asnA, aspA, aspC, cysK, dapA, dapD, deoD, fabG, fabZ, fbaB, fbp, fumA, gabD, garR, gcl, gcvP, gcvT, ghrB, glcB, glgP, glmM, glnA, glnS, glpK, gltA, glxR, glyA, gshA, gshB, hemC, hldE, icd, lldD, maeB, mdh, menB, ndk, nrdA, pck, pps, psd, pta, purA, purH, putA, pyrB, pyrI, ribC, ribE, rpiA, serC, sthA, sucC, sucD, talB, tdcE, thrB, tktA, xdhA, yeiA, yeiT |  | aceA, acnB, adk, allB, asnA, aspA, aspC, atpE, cydB, cyoB, cysK, dapA, dapD, deoD, fabZ, fbaB, fumA, gabD, garR, gcl, gcvP, gcvT, glcB, glnA, glpK, gltA, glxR, glyA, gshA, guaA, hldE, icd, lldD, maeB, manA, mdh, ndk, nrdA, otsA, pck, pepN, pps, pta, purA, purH, putA, pyrB, rpiA, serC, sthA, sucC, sucD, talB, tdcE, tktA, xdhA |  |
| Carbon metabolism | aceA, acnB, cysK, fbaB, fbp, fumA, gcvP, gcvT, glcB, gltA, glyA, icd, maeA, maeB, mdh, pck, pps, pta, rpiA, serC, sucC, sucD, talB, tktA, yqeA | accD, acs, eno, fba, frdB, gpmA, pfkA, ppc, pykF, sdaB, ydbK | aceA, acnB, cysK, fbaB, fumA, gcvP, gcvT, glcB, gltA, glyA, icd, maeB, mdh, pck, pps, pta, rpiA, serC, sucC, sucD, talB, tktA, yqeA | acs, eno, fba, gpmA, lpd, pfkA, pfkB, pgk, pykF, ydbK |
| Microbial metabolism in diverse environments | aceA, acnB, allB, aspC, cysK, dapA, dapD, fbaB, fbp, fucO, fumA, gabD, ghrB, glcB, glnA, gltA, glyA, icd, maeB, mdh, pck, pps, pta, rpiA, serC, sucC, sucD, talB, thrB, tktA, xdhA, yqeA |  | aceA, acnB, allB, aspC, cysK, dapA, dapD, fbaB, fucO, fumA, gabD, glcB, glnA, gltA, glyA, hybC, icd, maeB, mdh, pck, pps, pta, rpiA, serC, sucC, sucD, talB, tktA, xdhA, yqeA | acs, adhE, agp, aphA, cysQ, eno, fba, fdnG, gpmA, lpd, napA, pfkA, pfkB, pgk, pgm, pykF, usg, ydbK |
| Ribosome | rplA, rplB, rplC, rplD, rplI, rplK, rplN, rplU, rplX, rplY, rpmB, rpsI, rpsL, rpsR |  | rplK, rplL, rplN, rpmB, rpsB, rpsR | rplI, rplQ, rplT, rpmA, rpmE, rpsE, rpsG, rpsM, rpsN |
| Glyoxylate and dicarboxylate metabolism | aceA, acnB, fucO, garR, gcl, glcB, glnA, gltA, glxR, glyA, mdh |  | aceA, acnB, fucO, garR, gcl, glcB, glnA, gltA, glxR, glyA, mdh |  |
| Citrate cycle | acnB, fumA, gltA, icd, mdh, pck, sucC, sucD |  | acnB, fumA, gltA, icd, mdh, pck, sucC, sucD |  |
| Alanine, aspartate and glutamate metabolism | asnA, aspA, aspC, gabD, glnA, purA, putA, pyrB, pyrI |  | asnA, aspA, aspC, gabD, glnA, purA, putA, pyrB |  |
| Pyruvate metabolism | fucO, fumA, glcB, lldD, maeA, maeB, mdh, pck, pps, pta, tdcE |  | fucO, fumA, glcB, lldD, maeB, mdh, pck, pps, pta, tdcE |  |
| Biosynthesis of amino acids | acnB, asnA, aspC, cysK, dapA, dapD, fbaB, glnA, gltA, glyA, icd, rpiA, serC, talB, thrB, tktA |  | acnB, asnA, aspC, cysK, dapA, dapD, fbaB, glnA, gltA, glyA, icd, rpiA, serC, talB, tktA |  |
| Methane metabolism | fbaB, fbp, glyA, mdh, pps, pta, serC | acs, eno, fba, gpmA, pfkA, ppc | fbaB, glyA, mdh, pps, pta, serC | acs, eno, fba, gpmA, pfkA, pfkB |
| Propanoate metabolism | acnB, prpB, prpD, pta, sucC, sucD, tdcE |  | acnB, prpB, prpD, pta, sucC, sucD, tdcE |  |
| Biosynthesis of secondary metabolites | acnB, asnA, aspC, dapA, deoD, fbaB, fbp, fumA, glgP, gltA, glyA, hemC, icd, mdh, menB, ndk, pck, purH, putA, rpiA, sucC, sucD, talB, tktA |  | acnB, adk, asnA, aspC, dapA, deoD, fbaB, fumA, gltA, glyA, icd, manA, mdh, ndk, pck, purH, putA, rpiA, sucC, sucD, talB, tktA | acs, adhE, eno, fba, gadB, galF, galU, glgA, gpmA, guaB, ispG, lpd, pfkA, pfkB, pgk, pgm, purM, pykF, trpC, trpE |
| Pentose phosphate pathway | fbaB, fbp, ghrB, rbsK, rpiA, talB, tktA |  | fbaB, rbsK, rpiA, talB, tktA | deoB, deoC, fba, pfkA, pfkB, pgm |
| One carbon pool by folate | fmt, gcvT, glyA, purH |  | fmt, gcvT, glyA, purH |  |
| Purine metabolism | allB, amn,deoD, ndk, nrdA, purA, purH, xdhA, yqeA |  | adk, allB, amn, deoD, guaA, ndk, nrdA, purA, purH, xdhA, yqeA |  |
| Glycolysis / Gluconeogenesis |  | acs, adhE, eno, fba, gpmA, pfkA, ptsG, pykF |  | acs, adhE, agp, eno, fba, gpmA, lpd, pfkA, pfkB, pgk, pgm, ptsG, pykF |
| ABC transporters |  | araF, argT, glnH, gltI, malF, malK, metQ, mglB, modA, osmF, potF, rbsB, ugpB, yhdW |  | araF, argT, artI, dppA, fliY, glnH, gltI, hisJ, metQ, mglB, modA, oppA, osmF, potF, rbsB, ugpB, yhdW |
| Aminoacyl-tRNA biosynthesis |  |  |  | EcolC\_0819, asnS, aspS, pheS, proS |
| Arginine and proline metabolism |  |  | aspC, glnA, prr, putA, yqeA |  |

# Table S6. Water quality parameters of natural water matrix

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Source water #1 | Finished water #1 | Source water #2 | Finished water #2 |
| pH | 6.8 | 6.9 | 6.8 | 6.5 |
| Turbidity (NTU) | 14.9 | 0.4 | 5.0 | 0.1 |
| TOC (mg L-1) | 11.06 | 8.59 | 10.41 | 6.77 |
| TN (mg L-1) | 3.83 | 2.15 | 2.10 | 1.20 |
| UV254 (cm-1) | 0.0499 | 0.0137 | 0.0469 | 0.0121 |
| Cl- (mg L-1) | 14.97 | 12.74 | 7.21 | 13.10 |
| SO42- (mg L-1) | 30.69 | 20.56 | 18.90 | 20.38 |
| NO3- (mg L-1) | 14.46 | 9.30 | 10.03 | 3.53 |

# Table S7. EE/O values for UV/H2O2 and UV/TiO2 systems

|  |  |  |  |
| --- | --- | --- | --- |
| UV/H2O2 System a | EE/O b | UV/TiO2 System a | EE/O b |
| pH = 7.0 (control) | 0.0365 | pH = 7.0 (control) | 0.0319 |
| pH = 3.0 | 0.0297 | pH = 3.0 | 0.2035 |
| pH = 5.0 | 0.0358 | pH = 5.0 | 0.0324 |
| pH = 9.0 | 0.0391 | pH = 9.0 | 0.0588 |
| pH = 11.0 | 0.0436 | pH = 11.0 | 0.0914 |
| Cl- (100 mg L-1) | 0.0348 | Cl- (100 mg L-1) | 0.0331 |
| NO3- (100 mg L-1) | 0.0497 | NO3- (100 mg L-1) | 0.0412 |
| Humic acid (100 mg L-1) | 0.0641 | Humic acid (100 mg L-1) | 0.2673 |
| Source water #1 | 0.0955 | Source water #1 | 0.2341 |
| Finished water #1 | 0.0398 | Finished water #1 | 0.0650 |
| Source water #2 | 0.0757 | Source water #2 | 0.2453 |
| Finished water #2 | 0.0391 | Finished water #2 | 0.0882 |

a Experimental conditions: solution temperature 25 ± 2°C, pH 6.5-7.2, [1H-BTA]0 = 8.39 μM, [H2O2]0 = 4.41 mM, [TiO2]0 = 2.50 mM.

b The unit of EE/O is kWh m-3 order-1. The EE/O is defined as the electrical energy in kilowatt-hour (kWh) required to degrade a specific contaminant by one order of magnitude in 1 m3 contaminated water or air.