Supplementary Materials

Chemical ingredients in personal care products and their hormetic effects on freshwater photobacteria

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Q67 medium

The Q67 used in this study was sourced from Beijing Hamamatsu Corp., Ltd., Beijing, China. For culture medium preparation, the following compounds were weighed and dissolved in a 250 mL brown volumetric flask: 0.34 g KH_2PO_4, 0.895 g $Na_2HPO_4 \cdot 12H_2O$, 6.25 g $MgSO_4 \cdot 7H_2O$, 15.25 g $MgCl_2 \cdot 6H_2O$, 0.85 g $CaCl_2$, and 38.5 g NaCl. Separately, 67 g of $NaHCO_3$ was dissolved in a 1000 mL volumetric flask. For unsterilized double liquid culture medium, 10 mL each of KH_2PO_4, $Na_2HPO_4 \cdot 12H_2O$, $MgSO_4 \cdot 7H_2O$, $MgCl_2 \cdot 6H_2O$, $CaCl_2$, and NaCl solutions, along with 20 mL of $NaHCO_3$, were used. A mixture of 5 g yeast extract, 5 g tryptone, and 3 g glycerol was added to a beaker, diluted to 1000 mL with ultrapure water (conductivity 0.055 μS/cm), and the pH was adjusted to 8.5 with 2 mol/L NaOH. For a double liquid medium (unsterilized), 20 mL of each of the above solutions and 40 mL of $NaHCO_3$ were used, combined with 6.63 g of yeast extract, 8.78 g of pancreatic peptone, and 6.9 g of glycerol, then diluted to 1000 mL and pH adjusted to 8.5. The solid medium was prepared by adding 4.5–6 g of agar powder to 300 mL of the unsterilized double liquid medium.

All culture media were dispensed into conical flasks and sterilized using high-pressure steam at 121 ºC for 20 min. After cooling, the solid medium was poured into 6 cm diameter plates at a depth of 2-3 cm and stored at 4 ºC after UV sterilization. Sterilized pipettes, tips, and solid culture medium were placed on an ultra-clean table, UV-sterilized for 20 min, and then the Q67 strain stored at -20 ºC was thawed. A 1 mL pipette was used to add 0.8% NaCl solution to the strain centrifuge tube, shaking for 10 minutes to fully dissolve the freeze-dried powder. The solution was then spread onto 3-4 agar plates and incubated at 22 ± 1 ºC for 24 h in a constant temperature oscillation incubator. The grown bacteria were subcultured using sterilized inoculation rings, and the resulting strains were stored at 8 ºC, with transfers to fresh medium every three days.

Supplementary Table 1. Packaging list ingredients and detected chemical ingredients among six PCPs

NO.	PCPs	Packaging list ingredients	Detected chemical ingredients	DD%
M3	Make-up water 3	24K gold foil, collagen, narrow leaf blueberry fruit extract, trehalose, blueberry extract, sparse ETA palm fruit extract, phenoxyethanol, essence, and glycerin (9)	**Glycerin**, 1,3-butanediol, **phenoxyethanol**, polysorbate 80, Tween 80, Carbomer, triethanolamine, EDTA disodium, **trehalose**, adenosine, **gold**, and **essence** (12)	44%
M2	Make-up water 2	Propylene glycol, PEG-7 glycerol coconut ester, PEG-60 hydrogenated castor oil, coconut-based glucoside, glycerin, butanediol, C12-15 alcohol benzoate, dioctyl carbonate, decyl glucoside, oleanol polyether-3, oleanol polyether-5, phenoxyethanol, essence, citric acid, hydroxyphenyl methyl ester, hydroxyphenyl ethyl ester, EDTA disodium, benzyl salicylate, linalool, benzyl alcohol, α - iso methyl ionone, and extract of Lamia grandiflora (22)	**Propylene glycol**, **1,3-butylene glycol**, **glycerin**, **sunflower glucoside**, **coconut glucoside**, **PEG-7 glycerol coconut ester**, **PEG-60 hydrogenated castor oil**, **C12-C15 alcohol benzoate**, **dioctyl carbonate**, **oleanol polyether**, **phenoxyethanol**, **essence**, **citric acid**, **disodium EDTA**, **methyl hydroxybenzene**, **ethyl hydroxybenzene**, **iso methyl ionone**, **benzyl salicylate**, and **linalool** (19)	14%
S2	Skin water 2	Sorbitol (sugar), butanediol, carbomer, white flower Chunhuang chrysanthemum extract, glycerin, European basswood extract, sunflower seed oil, tocopherol acetate, butanol toluene, sodium citrate, and essence (11)	**Sorbitol, glycerin**, panthenol, phenoxyethanol, **1,3-butanediol**, isothiazolinone, EDTA disodium, **sodium citrate**, lactic acid, oleanol polyether-20, polyethylene glycol -8, vitamin E acetate, and **essence** (13)	54%
S5	Skin water 5	PEG-8, glycerin, betaine, octyl glycol, essence, saccharin, PPG-26-butanol polyeth-26, PEG-40, hydrogenated castor oil, oxothiazolidine carboxylic acid, caprylic acid, salicylic acid, bark extract of Casuarina falciformis, ascorbic acid glucoside, phenoxyethanol, chlorophenylene glycol, propionic acid (ester)/C10-30, alkanol acrylate cross-linked polymer, and tocopherol (vitamin E) (18)	**Glycerin**, **betaine, octyl glycol**, **polyethylene glycol -8**, **phenoxyethanol**, **chlorophenylene glycol ether**, **octyl salicylic acid**, alancysteine, ascorbic acid, **essence**, **PEG-40 hydrogenated castor oil**, and dextrin (12)	50%
S6	Skin water 6	PEG-7 glycerol coconut oil ester, phenoxyethanol, slamonium chloride water pyroxene, hydrogenated starch hydrolysate, EDTA trisodium, essence (daily use) (6)	Silicon dioxide, magnesium silicate, **phenoxyethanol**, polyethylene glycol-8 (4)	83%
T5	Toner 5	Butanediol, glycerol, aloe vera leaf juice, phenoxyethanol, hydroxyproline, willow bark extract, serine, urea sacs, disodium ascorbate sulfate, tiger tail fruit extract, hydrolyzed yeast, olive leaf extract, (animal) placental protein, acetyl tyrosine, peony root extract, and wild kudzu root extract (16)	**Glycerol**, **1,3-butanediol**, **phenoxyethanol**, **hydroxyproline**, serine, salicylic acid, allantoin, ascorbate glucoside (8)	75%

The detected chemical ingredient is shown in bold black font if it matches the ingredient listed on the packaging.

Supplementary Table 2. Twenty-three PCP chemical ingredients were fitted with S-shaped CRC parameters (E_m, E_{max}, EC_{up}, and H_{up}) and statistical measures (R^2 and *RMSE*), EC_{50}, and their 95%OCI for Q67 at 0.25 h[a]

Chemical ingredient	E_m	E_{max}	EC_{up}	H_{up}	R^2	RMSE	EC_{50} (g/mL)	OCI_{left} (g/mL)	OCI_{right} (g/mL)
AAG	0.084	1.033	3.510E–3	5.261	0.9868	0.05589	3.349E–3	2.716E–3	4.030E–3
ASA	0.123	0.996	1.740E–3	9.941	0.9979	0.02060	1.692E–3	1.622E–3	1.759E–3
BEN	0.059	0.748	4.772E–2	1.752	0.9904	0.02993	6.634E–2	4.574E–2	9.988E–2
BET	–0.027	1.042	1.062E–1	2.323	0.9922	0.0445	1.049E–1	7.813E–2	1.415E–1
BUT	–0.016	0.982	5.399E–2	1.217	0.9974	0.00937	6.827E–2	4.968E–2	–
CIA	0.029	1.016	6.700E–4	5.644	0.9952	0.03705	6.565E–4	5.788E–4	7.404E–4
COG	–0.068	1.001	4.100E–4	1.533	0.994	0.03167	4.409E–4	3.291E–4	5.814E–4
DEG	–0.167	1.111	2.900E–4	1.369	0.9973	0.02602	3.071E–4	2.461E–4	3.795E–4
DIC	0.083	0.981	4.679E–1	1.137	0.9913	0.01679	4.121E–1	3.177E–1	–
GLO	–0.109	1.251	1.480E–1	0.794	0.9966	0.02157	1.135E–1	8.400E–2	1.537E–1
IPO	0.035	1.300	5.757E–2	2.159	0.9921	0.03187	4.480E–2	3.590E–2	5.570E–2
ITZ	0.081	1.038	1.100E–4	6.804	0.9914	0.04242	1.050E–4	9.298E–5	1.173E–4
LAA	0.132	1.347	8.000E–5	5.154	0.9908	0.0354	6.943E–5	6.198E–5	7.631E–5
LIO	–0.034	1.000	4.900E–4	1.571	0.9968	0.02603	5.102E–4	3.992E–4	6.521E–4
MET	0.101	1.132	1.174E–2	0.954	0.9918	0.02986	7.243E–3	4.366E–3	1.131E–2
ODO	0.024	1.007	2.700E–4	1.433	0.9917	0.0352	2.590E–4	1.762E–4	3.843E–4
PDO	–0.035	0.752	2.320E–1	1.143	0.9938	0.01623	4.482E–1	3.351E–1	–
PEG400	0.043	1.005	3.341E–2	1.651	0.9949	0.02622	3.144E–2	2.433E–2	4.091E–2
POE	–0.017	1.146	9.200E–4	1.137	0.9974	0.02231	7.597E–4	5.875E–4	9.795E–4
PRO	0.050	1.296	1.278E–1	1.709	0.993	0.0205	9.156E–2	7.613E–2	–
SBO	–0.024	2401.927	7.051E+03	0.948	0.9887	0.02407	1.032E+00	7.967E–1	1.186E+00
SOC	–0.595	8.445	9.557E+01	0.247	0.9945	0.02869	3.089E–2	1.966E–2	4.762E–2
TEA	–0.214	1.161	2.076E–2	0.967	0.9978	0.01973	2.245E–2	1.812E–2	2.790E–2

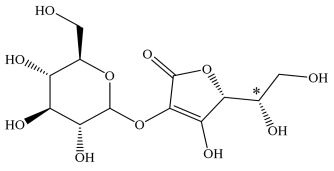
–: none available

a: Data for ASA, BET, GLO, ODO, PEG400, and POE were sourced from our previous study [1].

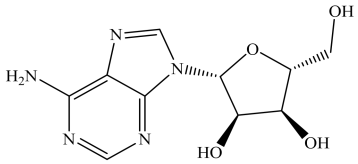
Supplementary Table 3. The J-shaped CRC fitting parameters (E_m, EC_{down}, H_{down}, E_{max}, EC_{up}, H_{up}, and E_0) and statistical parameters (R^2 and $RMSE$) of 18 chemical ingredients for Q67[a]

Chemical ingredient	Time (h)	E_m	EC_{down}	H_{down}	E_{max}	EC_{up}	H_{up}	E_0	R^2	RMSE
AAG	12	–9.303	2.970E–4	1.266	–6.674	4.200E–3	14.176	7.574	0.9968	0.07702
ADE	12	–7.100	4.310E–4	2.139	20.013	1.186E+00	0.253	0.392	0.975	0.23473
BEN	12	–16.580	2.091E–2	5.907	1.382	3.000E–4	0.506	–0.025	0.9956	0.04279
BET	12	–14.621	1.868E–2	2.188	1.299	4.600E–2	2.527	–0.192	0.9993	0.12414
CEG	12	–257.943	9.900E–5	42.847	0.894	0.000E+00	2.302	0.026	0.9914	0.06668
DEG	12	–28.432	1.210E–4	43.985	1.314	0.000E+00	0.714	0.025	0.9901	0.06444
E4C	12	-0.145	1.400E–5	27.021	0.952	2.810E–4	1.818	-0.191	0.9979	0.03044
IPO	12	–0.430	2.430E–4	28.516	1.265	5.800E–3	1.653	–0.220	0.9935	0.07975
ITZ	12	–4.387	1.120E–4	3.227	1.112	1.000E–4	11.151	0.058	0.9954	0.05535
LAA	12	–0.635	4.700E–5	17.198	1.442	1.000E–4	7.726	–0.328	0.9903	0.08447
LIO	12	–3.080	4.360E–4	0.639	0.38	1.200E–3	4.481	0.666	0.9978	0.0521
ODO	12	–0.594	1.316E–2	2.391	1.368	1.019E–1	1.108	–0.071	0.9959	0.04414
PGC	0.25	–1.099	1.700E–5	154.498	2.458	5.400E–3	0.229	–0.039	0.992	0.03968
PGC	12	–1.888	1.800E–5	26.591	0.888	0.000E+00	0.892	0.063	0.9926	0.05322
POE	12	–0.384	1.280E–4	2.624	1.438	6.000E–4	1.459	–0.360	0.9986	0.0329
SBO	12	–49.053	7.298E–1	1.63	1.518	2.437E–1	4.258	–0.079	0.9998	0.03181
SER	12	–3.370	1.807E–1	1.609	1.012	1.814E–1	8.888	–0.026	0.9984	0.03787
TW80	0.25	–5.024	3.200E–5	0.298	81.03	3.393E+00	1.348	3.566	0.9902	0.04249

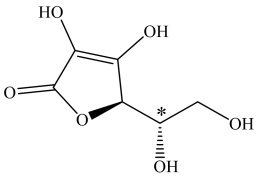
a: Data for BET, E4C, ODO, PEG400, and POE were sourced from our previous study [1].



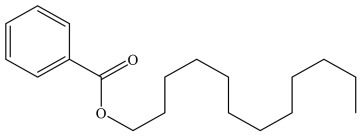
Ascorbic acid 2-glucoside



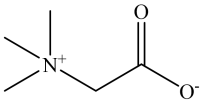
Adenosine



Ascorbic acid



Benzoic acid C12-15-alkyl esters



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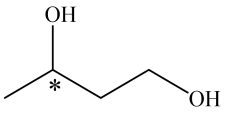
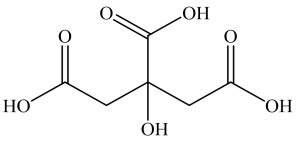


1,3-Butanediol



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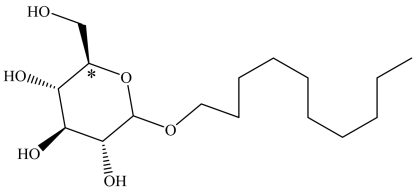

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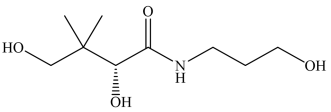

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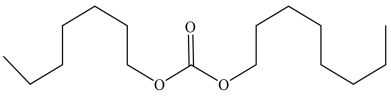
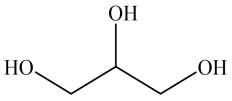


Dicaprylyl carbonate



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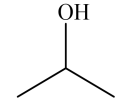

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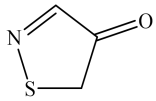

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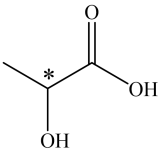

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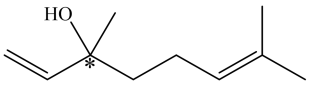

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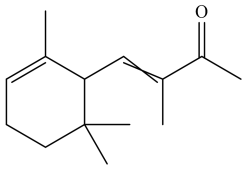
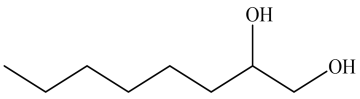
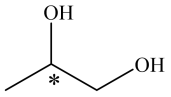


α-iso-Methylionone



1,2-Octanediol



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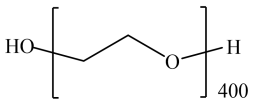

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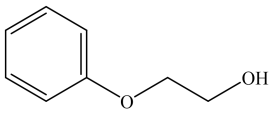

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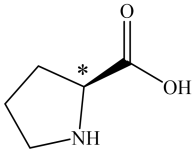

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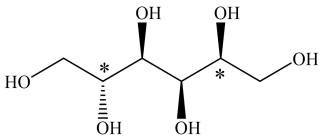

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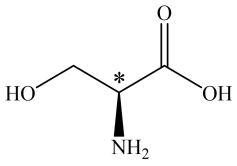

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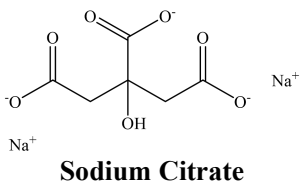

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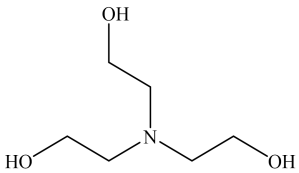
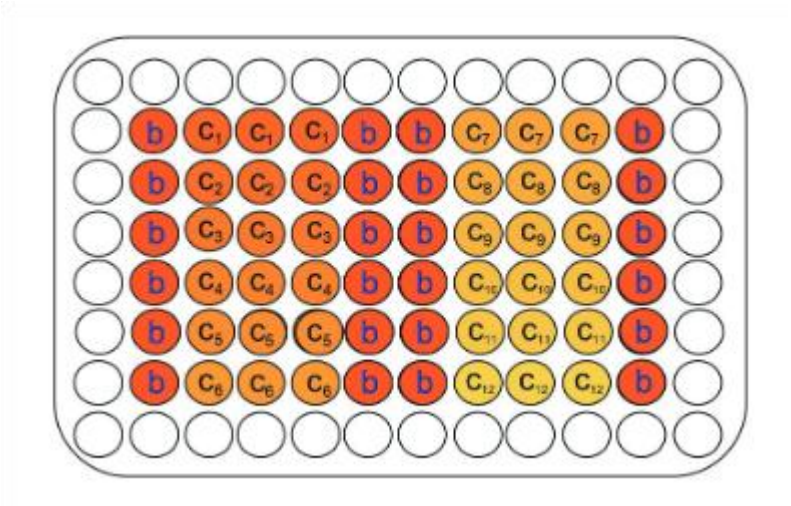


Triethanolamine

Supplementary Figure 1. Structures of chemical ingredients.



Supplementary Figure 2. Microplate design in the long-term microplate toxicity analysis (L-MTA), where b refers to blank and c_i to the i^{th} concentration gradient [2].

References

1. Xu, Y.-Q.; Li, K.; Wang, Z.-J.; Huang, P.; Liu, S.-S. Transfer pattern of hormesis into personal care product mixtures from typical hormesis-inducing compounds. *Science of the Total Environment* **2023**, *855*, 158981, doi:10.1016/j.scitotenv.2022.158981.
2. Liu, S.S.; Zhang, J.; Zhang, Y.H.; Qin, L.T. APTox: Assessment and Prediction on Toxicity of Chemical Mixtures. *Acta Chimica Sinica* **2012**, *70*, 1511-1517, doi:10.6023/a12050175.