Supporting Information for

Formation of EGCG oxidation self-assembled nanoparticles and its antioxidant activity *in vitro* and hepatic REDOX regulation activity *in vivo*

Ximing Wu[1,2,3,#], Yijun Wang[2,#], Dongxu Wang[4], Ziqi Wang[3], Mingchuan Yang[2], Lumin Yang[2], Fuming Wang[2], Wei Wang[1,*] and Xiangchun Zhang[3,*]

[1]Anhui Engineering Laboratory for Medicinal and Food Homologous Natural Resources Exploration, school of Biological and Food Engineering, Hefei Normal University, Hefei, 230601, China
[2] Laboratory of Redox Biology, State Key Laboratory of Tea Plant Biology and Utilization, School of Tea & Food Science, Anhui Agricultural University, Hefei, 230036, China
[3] Tea Research Institute, Chinese Academy of Agricultural Sciences, Hangzhou, 310008, China
[4] School of Grain Science and Technology, Jiangsu University of Science and Technology, Zhenjiang, 212100, China

[#] Contributed equally to this work.

***Corresponding Authors:**

E-mail: zhangxc@tricaas.com; weiw@hfnu.edu.cn

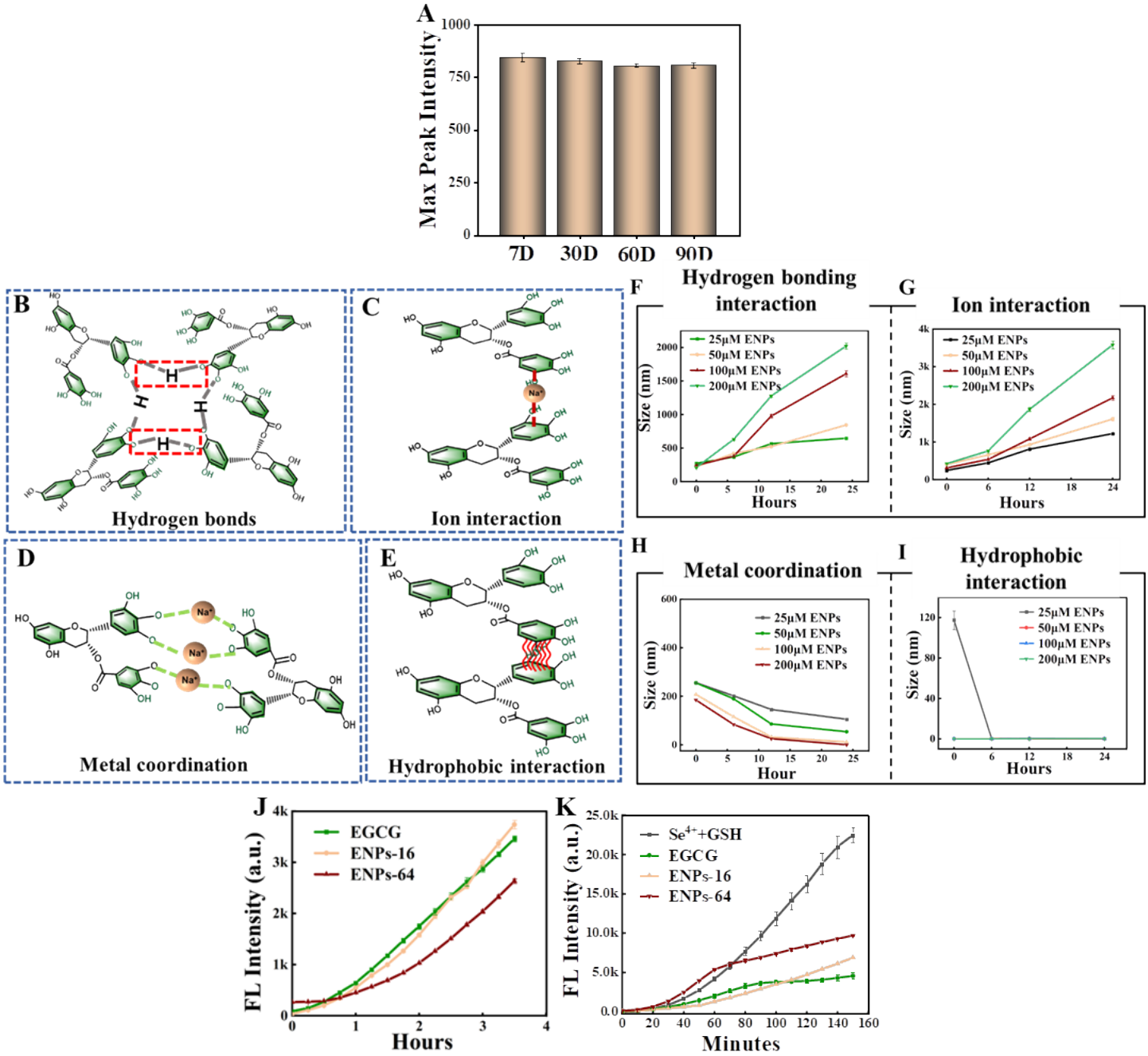

Figure S1. The characteristics of ENPs and potential intermolecular forces driving EGCG self-assembly. (A) Stability of ENPs-64 observed at 4 °C for 7, 30, 60 and 90 days. (B) The potential role of hydrogen bonds in ENPs. (C) The potential role of sodium ion interaction in ENPs. (D) The potential coordination role of sodium metal in ENPs. (E) The potential hydrophobic action of benzene ring in ENPs. (F) The size changes of ENPs after urea treatment with different time and dose at 37°C. (G) The size changes of ENPs after NaCl treatment with different time and dose at 37°C. (H) The size changes of ENPs after EDTA treatment with different time and dose at 37°C. (I) The size changes of ENPs after Tween-20 treatment with different time and dose at 37°C. (J) ROS production at 0.4 mg/mL concentration detected by 50 μM DCFH-DA. (K) Scavenging ROS in selenite/glutathione system at 0.2 mg/mL concentration. Experiments were carried out at in 200 mM PBS (1 mM $EDTANa_2$, pH 8.0) at 37 °C in the presence or absence of 50 μM DCFH-DA. Data are presented as the mean ± SEM (n= 2 or 3).

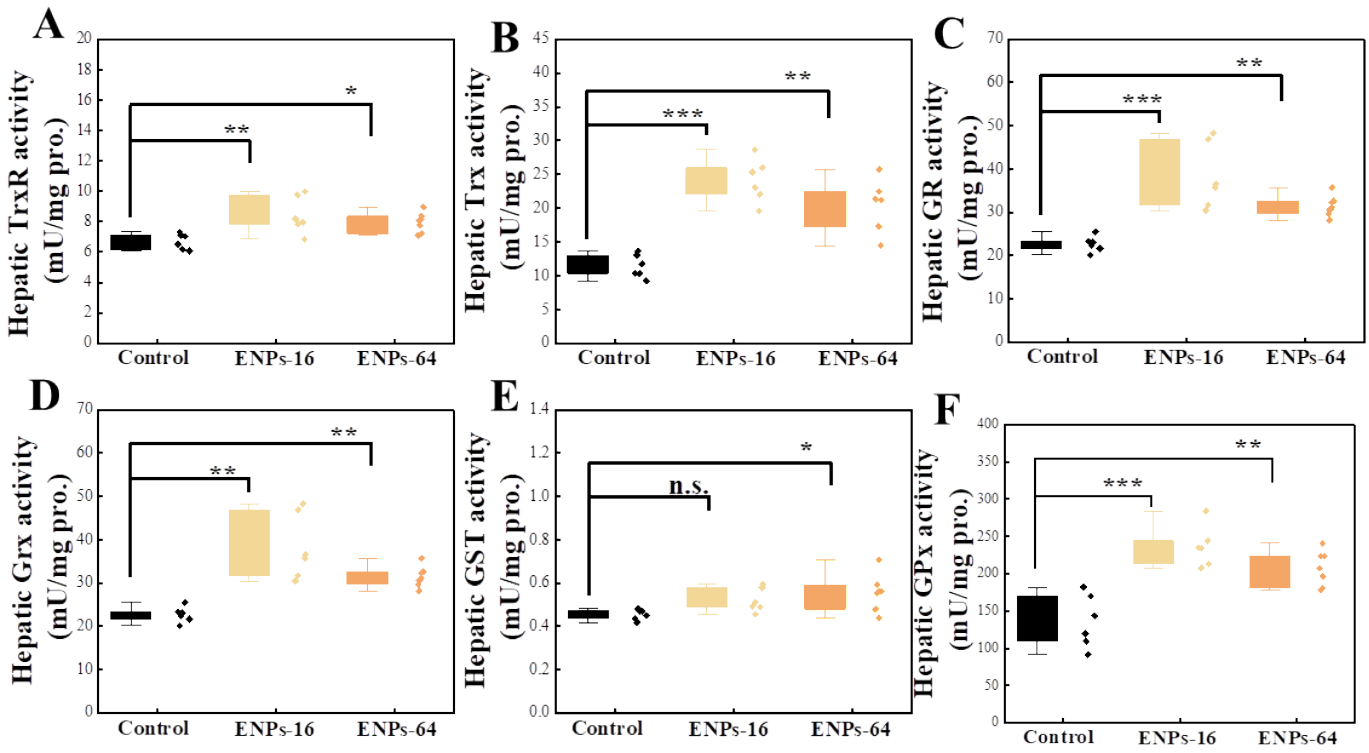

Figure S2. Influence of ENPs-16 and ENPs-64 on hepatic antioxidant enzymes activities. Kunming mice (n = 6/group) were i.p. administered with PBS as control or ENPs-16 and ENPs-64 respectively, at a dose of 80 mg/kg daily for 3 days. (A) Hepatic TrxR activity. (B) Hepatic Trx activity. (C) Hepatic GR activity. (D) Hepatic Grx activity. (E) Hepatic GST activity. (F) Hepatic GPx activity. Data are presented as the mean ± SEM. * P<0.05, ** P<0.01, and *** P<0.001, compared to the control group.

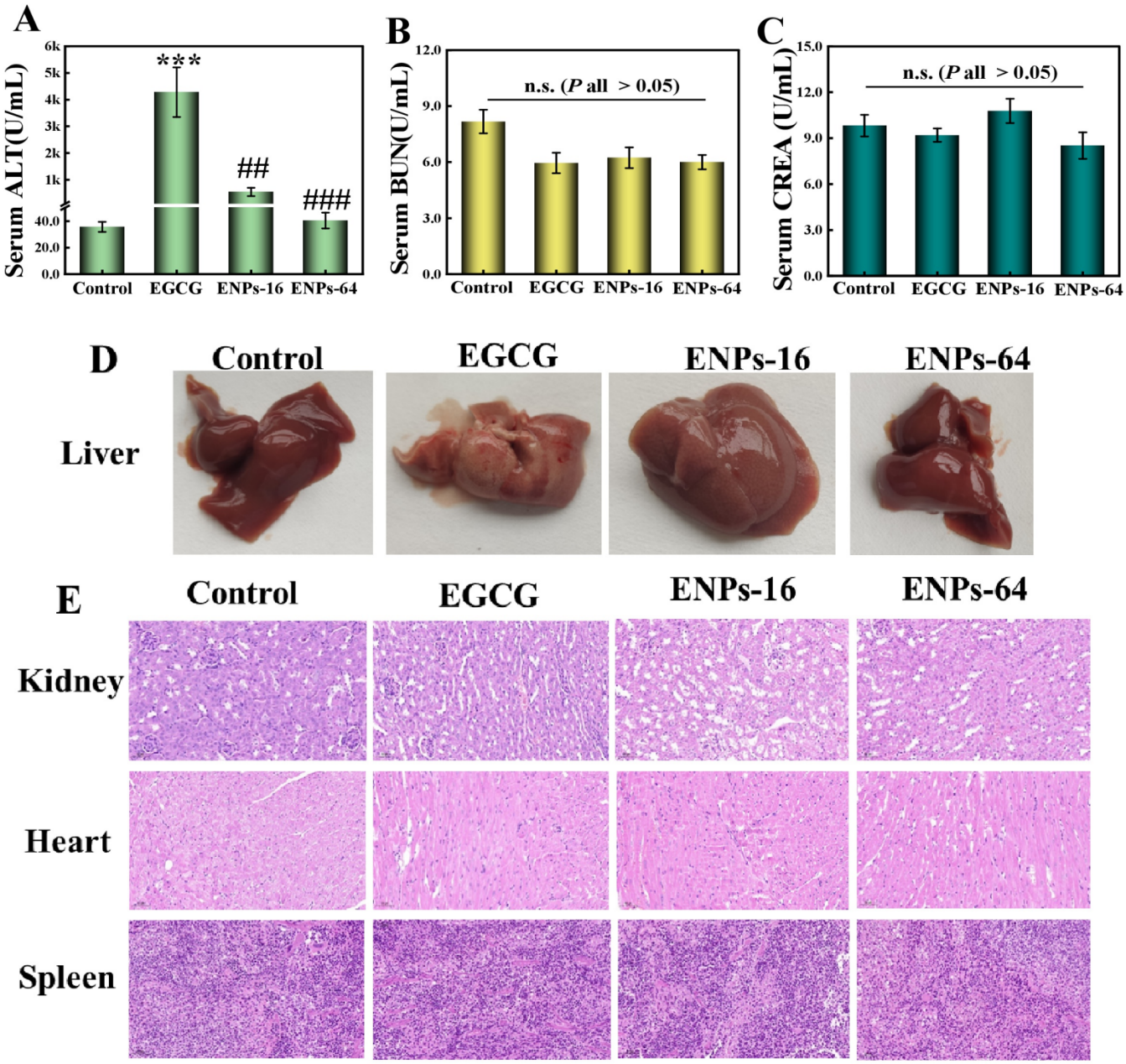

Figure S3. The blood biochemical index and pathologies evaluation of ENPs in mice. Kunming mice were i.p. administered with PBS as control or EGCG, ENPs-16 and ENPs-64 respectively, at a dose of 100 mg/kg daily for 4 days. (A) Serum AST activity. (B) Serum BUN activity. (C) Serum CREA activity. Data are presented as the mean ± SEM. *** $P<0.001$, compared to control group. ## $P<0.01$ and ### $P<0.001$, compared to EGCG group. (D) Liver images in control, EGCG, ENPs-16 or ENPs-64 treated group mice. (E) Histopathological images of kidney, heart and spleen in control, EGCG, ENPs-16 or ENPs-64 treated group mice. Samples were collected at the end of the experiment (H&E stained, × 200 magnification) and H&E staining was performed to investigate the histological changes in all experimental groups.

Table S1. Gene-specific primers used.

Genes	Primers	Sequences
Bax	Sense	TGGAGATGAACTGGACAGCAATAT
	Antisense	GCAAAGTAGAAGAGGGCAACCAC
NRF2	Sense	GACGGGACTATTGAAGGCTGTGA
	Antisense	TCGGCTGGGACTCGTGTTCA
Gclc	Sense	GCCTGGAGCCTCTGAAGAACAA
	Antisense	CGTGCTGTGCCAGAAGATGAT
HO1	Sense	TCAGAAGGGTCAGGTGTCCAGA
	Antisense	GCATAGACTGGGTTCTGCTTGTT
NQO1	Sense	GGCGAGAAGAGCCCTGATTG
	Antisense	GTTCATAGCATAGAGGTCAGATTCG
Gss	Sense	ATGCCCAGTCAGTATAATTCACAGA
	Antisense	GACCCACCCTGCTCAGTTCC
TrxR	Sense	ACCTGGGCATCCCTGGAGAC
	Antisense	GCACCATTACAGTGACGTCTAAGC
Trx	Sense	CCTTCTTCCATTCCCTCTGTGAC
	Antisense	TTTCCTTGTTAGCACCGGAGAAC
Thbs	Sense	CCCCTACAACCACAACCCTGAC
	Antisense	ACTGATCTCCAACCCCATCCAT
MDM2	Sense	AGGCAGAAGAAGGCTTGGATGT
	Antisense	TGGAAGTCGATGGTTGGGAATA
P53	Sense	ATCTACAAGAAGTCACAGCACATGA
	Antisense	TCTTCCAGATACTCGGGATACAAAT
β-actin	Sense	GCTGAGAGGGAAATCGTGCGT
	Antisense	ACCGCTCGTTGCCAATAGTGA

Table S2. Blood routine examination of EGCG, ENPs-16 and ENPs-64.

	Control	EGCG	ENPs-16	ENPs-64
WBC (10^9/L)	2.00±0.36	1.97±0.93$^{n.s.}$	2.32±0.68$^{n.s.}$	2.12±0.48$^{n.s.}$
Lymph (10^9/L)	1.23±0.17	1.07±0.74$^{n.s.}$	1.40±0.53$^{n.s.}$	1.30±0.63$^{n.s.}$
Mon (10^9/L)	0.10±0.00	0.13±0.06$^{n.s.}$	0.13±0.13$^{n.s.}$	0.14±0.08$^{n.s.}$
RBC (10^{12}/L)	8.34±1.74	8.59±0.43$^{n.s.}$	7.87±0.35$^{n.s.}$	7.99±0.32$^{n.s.}$

Notes: $P < 0.05$ in all groups in each tested item and n.s.were labeled in each tested item. Kunming mice (n = 7/group) were i.p. administered with PBS as control or EGCG, ENPs-16 and ENPs-64 respectively, at a dose of 100 mg/kg daily for 4 days. Data are presented as the mean ± SEM.