SUPPORTING INFORMATION

Stability of ZIF-8 nanoparticles in most common cell culture media

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Cell culture media composition

Table S1. Cell culture media composition. The concentration units of all compounds are 10^{-5} M. Color code for media names corresponds to the colors in Figure 3.

	MEM	199	RPMI	alpha-MEM	DMEM	IMDM	L15
L-Alanine	0	28	0	28	0	28	250
L-Arginine	60	33	110	60	40	40	290
L-Asparagine	0	30	170	33	0	19	7,6
Aspartic acid	0	23	15	23	0	23	0
L-Cysteine	0	0,056	0	57	0	0	99
L-Cystine	10	9,9	21	10	20	29	0
L-Glutamate	0	45	14	51	0	51	0
L-Glutamine	200	68	210	200	400	400	210
L-Glycine	0	67	13	67	40	40	27
L-Histidine	20	10	9,7	20	20	20	160
Hydroxyproline	0	7,6	15	0	0	0	0
L-Isoleucine	40	15	38	40	80	80	95
L-Leucine	40	46	38	40	80	80	95
L-Lysine HCl	40	38	22	40	80	80	51
L-Methionine	10	10	10	10	20	20	50
L-Phenylalanine	20	15	9,1	19	40	40	76
L-Proline	0	35	17	35	0	35	0
L-Serine	0	24	29	24	40	40	190
L-Threonine	40	25	17	40	80	80	250
L-Tryptophan	4,9	4,9	2,5	4,9	7,8	8	9,8
L-Tyrosine	20	22	11	23	40	39	170
L-Valine	40	21	17	39	80	80	85
Total aminoacids concentration	**545**	**577**	**788**	**864**	**1068**	**1232**	**2115**

	MEM	199	RPMI	alpha-MEM	DMEM	IMDM	L15
Phenol Red•Na	2,7	4,5	1,3	2,9	4	4,239871	2,7
CaCl2	180	130		180	180	149	130
KCl	530	530	530	53	530	443	530
KH2PO4		44					44
MgCl2				2000			
MgSO4	81	81	40	81	81	81	160
NaCl	12000	14000	10000		11000	7709	14000
NaHCO3	2600		420	2600	4400	3600	
NaH2PO4	100				91	91	
Na2HPO4		40	560				0
D-Glucose	560	560	1100	560	2500	2498	
D-Galactose							5000

	MEM	199	RPMI	alpha-MEM	DMEM	IMDM	L15
Pyruvic Acid•Na				100	100	99	500
Linoleic acid					0,03		
Lipoic acid				0,097			
Para-aminobenzoic acid		0,036	0,73				
L-ascorbic acid		0,028		25			
Biotin		0,0041	0,082	0,041		0,005	
Choline Chloride	0,71	0,36	2,1	0,71	2,9	2,86	0,71
Folic Acid	0,23	0,0023	0,23	0,23	0,91	0,91	0,23
Myo-inositol	1,1	0,028	19	1,1	4	4,00	1,1
Nicotinamide	0,82	0,02	0,82	0,82	3,3	3,28	0,82
Nicotinic acid		0,02					
DL-Pantothenic Acid (hemicalcium)	0,42	0,0042	0,11	0,42	1,7	1,67	0,42
Pyridoxine•HCl		0,012	0,49				
Pyridoxal •HCl	0,49	0,012		0,49	2	1,96	
Riboflavin	0,027	0,0027	0,053	0,027	0,11	0,11	0,0019
Thiamine	0,3	0,003	0,3	0,3	1,2	1,33	0,24
α-Tocopherol		0,0023					
B12			0,00037	0,1		0,001	
Hypoxanthin		0,22					
Fe(NO3)3 9H2O					0,025		
FeSO4 7H2O							
ZnSO4 7H2O							
CuSO4 5H2O							
Thymidine				4,1			
Adenine SO4		5,4					
Adenosine				3,7			
AMP		0,058					
ATP		1,8					
Cytidine				4,1			
D-adenosine				4			
D-cytidine				4,2			
D-guanosine				3,7			
2-deoxyribose		0,37					
Guanine		0,16					
Guanosine				3,5			
D-ribose		0,33					
Thymine		0,24					
Uracil		0,27					
Uridine				4,1			
Xanthine		0,2					
Tween-80		1,8					
Putrescine							

	MEM	199	RPMI	alpha-MEM	DMEM	IMDM	L15
Calciferol		0,025					
P-retinol acetate		0,035					
HEPES						2500	
Sodium acetate		0,45					
Na2SeO3						0,01	
KNO3						0,08	

Results of simulation of experimental spectra.

Table S2. Fraction of the probe inside ZIF-8 calculated from the CW EPR spectra.

Sample	Fraction of loaded guest molecules, %		Sample	Fraction of loaded guest molecules, %
HEPES (pH=7.4)	91		**MEM**	56
HEPES (pH=7.8)	94		**MEM + 10 mM MIM**	61
PBS (pH=7.4)	76		**MEM + 30 mM MIM**	61
PBS (pH=7.8)	88		**OPTIMEM**	27
10% FBS	94		**OPTIMEM + 10 mM MIM**	41
10% FBS + 10 mM MIM	93		**RPMI**	8
199	63		**RPMI + 10 mM MIM**	13
199 + 10 mM MIM	70		**α-MEM**	52
DMEM	30		**α-MEM + 10 mM MIM**	53
DMEM + 10 mM MIM	46			
DMEM + 30 mM MIM	58			
IMDM	15			
IMDM + 10 mM MIM	20			
L-15	11			
L-15 + 10 mM MIM	22			
L-15 + 30 mM MIM	36			
Sample	**Fraction of loaded guest molecules, %**		**Sample**	**Fraction of loaded guest molecules, %**
Ala	78		**Lys**	80
Arg	80		**Lys 1 mM**	87
Asn	59		**Lys 10 mM**	66
Asn + 10 mM MIM	77		**Met**	66
Asp 2.5 mM	65		**Met + 10 mM MIM**	76
Cys	17		**Phe**	47
Cys + 10 mM MIM	10		**Phe + 10 mM MIM**	63
Cys 1 mM	71		**Pro**	84
Cys 10 mM	1		**Ser**	61
Cys 2.5 mM	52		**Ser + 10 mM MIM**	85
Glu	73		**Ser 1 mM**	86

Sample	Fraction of loaded guest molecules, %		Sample	Fraction of loaded guest molecules, %
Gly	72		**Ser 10 mM**	51
Gly + 10 mM MIM	90		**Thr**	62
His	41		**Thr + 10 mM MIM**	73
His + 10 mM MIM	40		**Trp**	63
His 1 mM	86		**Trp + 10 mM MIM**	66
His 10 mM	4		**Tyr 1mM**	74
His 2.5 mM	66		**Val**	72
Ile	75		**Leu**	67

Parameters β, Ka, and fraction of spin probes escaped from ZIF-8 for studied amino acids.

Amino acid	Log(β)	pKa	Log(β)-2*pKa	Fraction of escaped guest molecules, %
Pro	9,69	10,34	-10,99	16,0
Arg	7,56	8,82	-10,08	20,4
Ala	8,56	9,5	-10,44	22,2
Ile	8,08	9,36	-10,64	25,3
Glu	8,54	9,39	-10,24	27,2
Gln	7,94	8,83	-9,72	27,8
Val	8,24	9,32	-10,4	27,9
Gly	9,01	9,38	-9,75	28,4
Met	6,93	8,91	-10,89	33,7
Trp	8,76	9,085	-9,41	36,7
Thr	8,14	8,71	-9,28	37,9
Ser	8,31	8,84	-9,37	39,1
His	11,68	8,92	-6,16	59,1
Cys	17,98	10,23	-2,48	83,5

We analyzed two processes: Zn^{2+}+2AA ↔ $Zn(AA)_2$ with constant β and HAA ↔ H^+ + AA^- with constant K_a, where AA is an amino acid and HAA indicates its protonated form. Equilibrium constants can be written as follows:

$$\beta = \frac{[ZnAA_2]}{[Zn^{2+}][AA^-]^2};\ K_a = \frac{[H^+][AA^-]}{[HAA]};$$

And concentration of deprotonated amino acids is

$$[AA^-] = \frac{K_a[HAA]}{[H^+]}.$$

Using this concentration in equation with β provides combination of β and K_a:

$$\beta = \frac{[ZnAA_2][H^+]^2}{[Zn^{2+}]K_a{}^2[HAA]^2};$$

$$\frac{\beta K_a{}^2[HAA]^2}{[H^+]^2} = \frac{[ZnAA_2]}{[Zn^{2+}]}.$$

Since the whole process of ZIF-8 dissolution and zinc interaction with amino acids is complicated, we analyzed correlation between βK_a^2 and the amount of escaped radical.

Particle size distribution

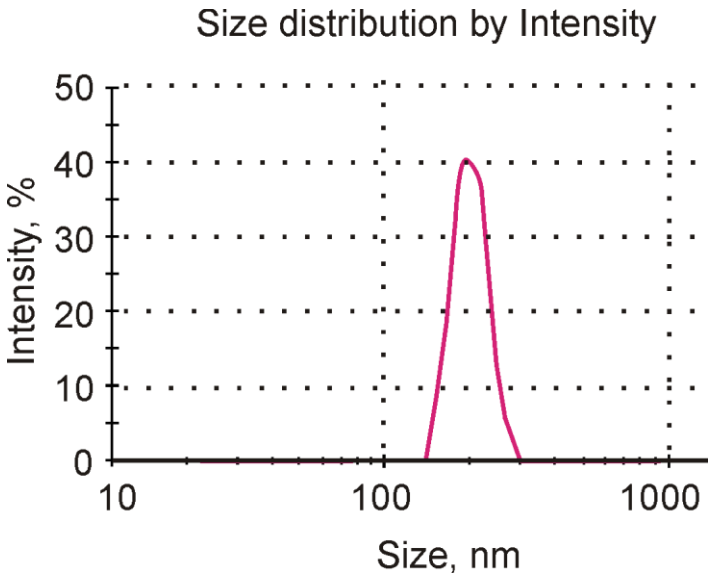

Figure S1. Particle size distribution curves of ZIF-8 nanoparticles in water measured by dynamic light scattering.

CW EPR spectra of R@ZIF-8 at pH=4.0 and free radical R in H_2O

Spectrum of fully dissociated R@ZIF-8 at pH=4 (citrate buffer) showed no differences compared to the spectrum of free radical in water.

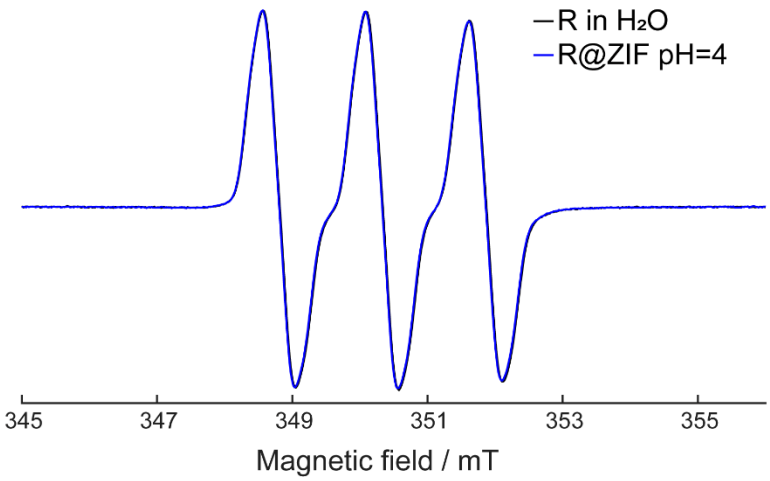

Figure S2. X-band room temperature CW EPR spectra of R@ZIF-8 at pH=4 and free radical R in water. Both spectra were normalized by double integral.

CW EPR spectra of initial R@ZIF-8 and R@ZIF-8 5-fold diluted with water

R@ZIF-8 dilution with water did not lead to MOF degradation: 99% of the spectrum corresponds to the loaded fraction.

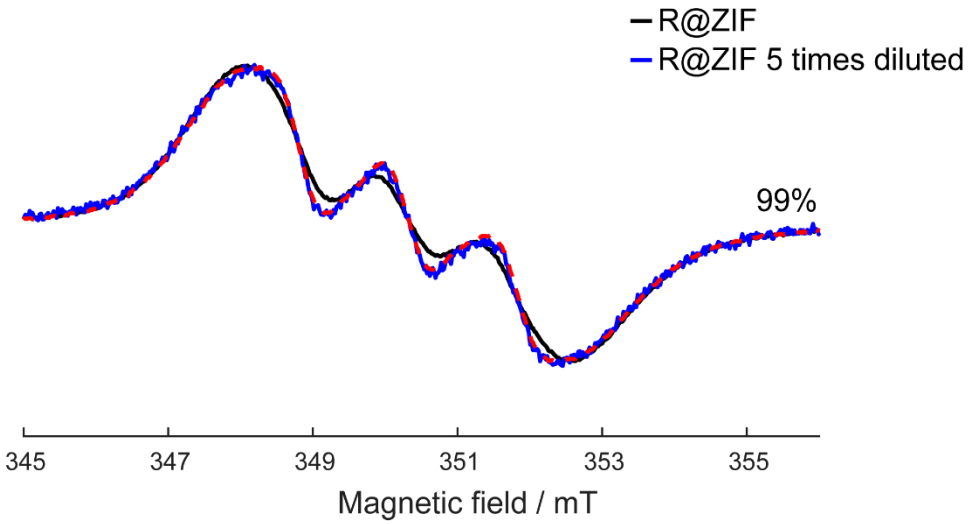

Figure S3. X-band room temperature CW EPR spectra of R@ZIF-8 and R@ZIF-8 diluted with water. Red dashed line shows spectrum simulation, the number on the right – the fraction of the probe inside ZIF-8. Spectra were normalized by double integral.

CW EPR of R@ZIF-8 in different media

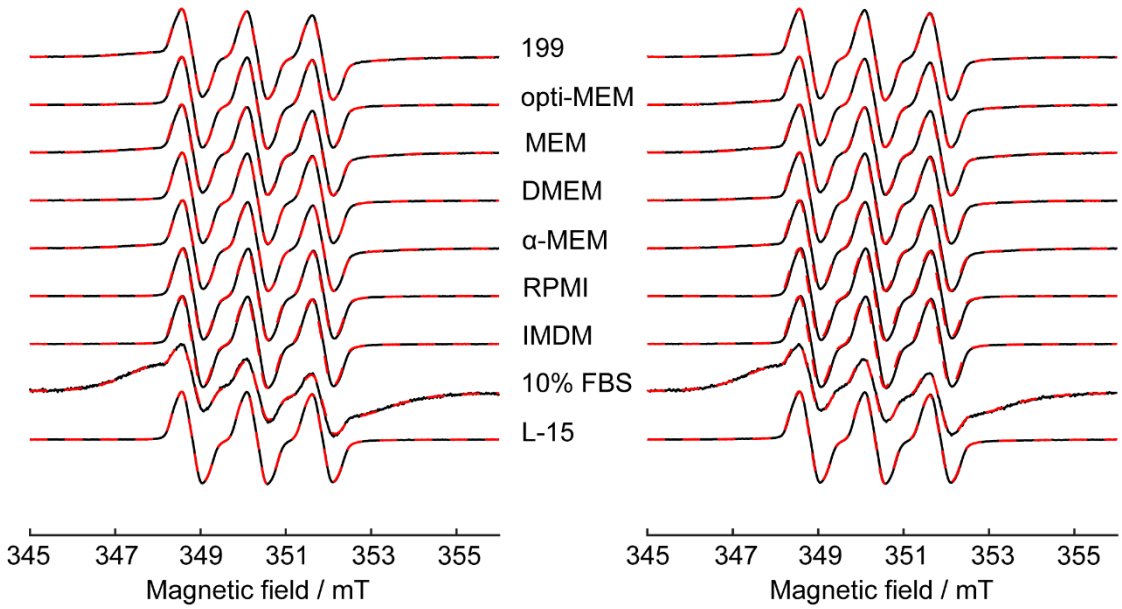

Figure S4. X-band room temperature CW EPR spectra of R@ZIF-8 after 5-fold dilution in culture media. On the left - spectra of samples without MIM, on the right - samples with

addition of 10mM MIM. Red dashed lines show simulations. All spectra were normalized by double integral.

Spectra of R@ZIF-8 in MEM at different concentrations

We diluted initial R@ZIF-8 with MEM 5-, 10- and 15-times v/v. The results demonstrated that the amount of escaped guest molecules increased with the amount of MEM added. The 10 and 15 times diluted sample spectrum had no component corresponding to the R@ZIF-8.

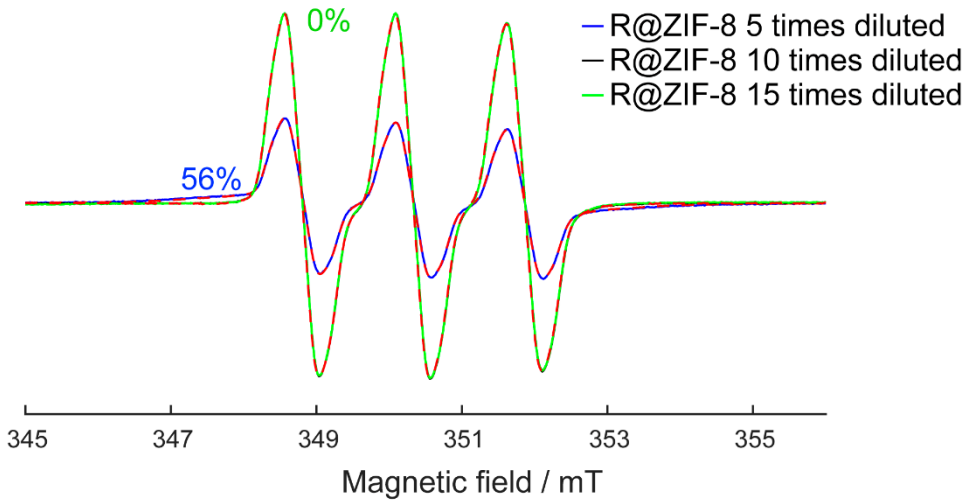

Figure S5. X-band room temperature CW EPR spectra of R@ZIF-8 and R@ZIF-8 diluted with MEM. Red dashed lines show spectra simulation, the numbers on the figure demonstrate fractions of loaded guest molecules, color code corresponds to the legend. All spectra were normalized by double integral.

EPR spectra of R@ZIF-8 with amino acids

All amino acids were dissolved in 5mM HEPES buffer, pH=7.4 at 5mM concentration except for Asp – 2.5mM Tyr – 1mM. Tyr and Asp were poorly soluble at this concentration.

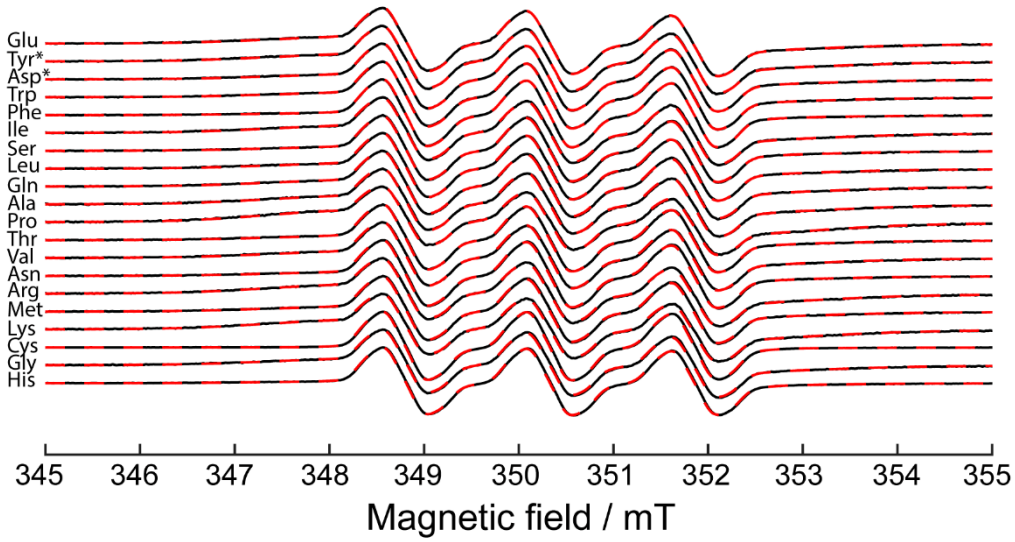

Figure S6. X-band room temperature CW EPR spectra of R@ZIF-8 after addition of amino acids with 5mM concentration (except Trp and Asp). Red dashed lines show simulations. All spectra were normalized by double integral.

EPR spectra at different concentrations of amino acids

All samples were made with a 5mM HEPES buffer at pH=7.4.

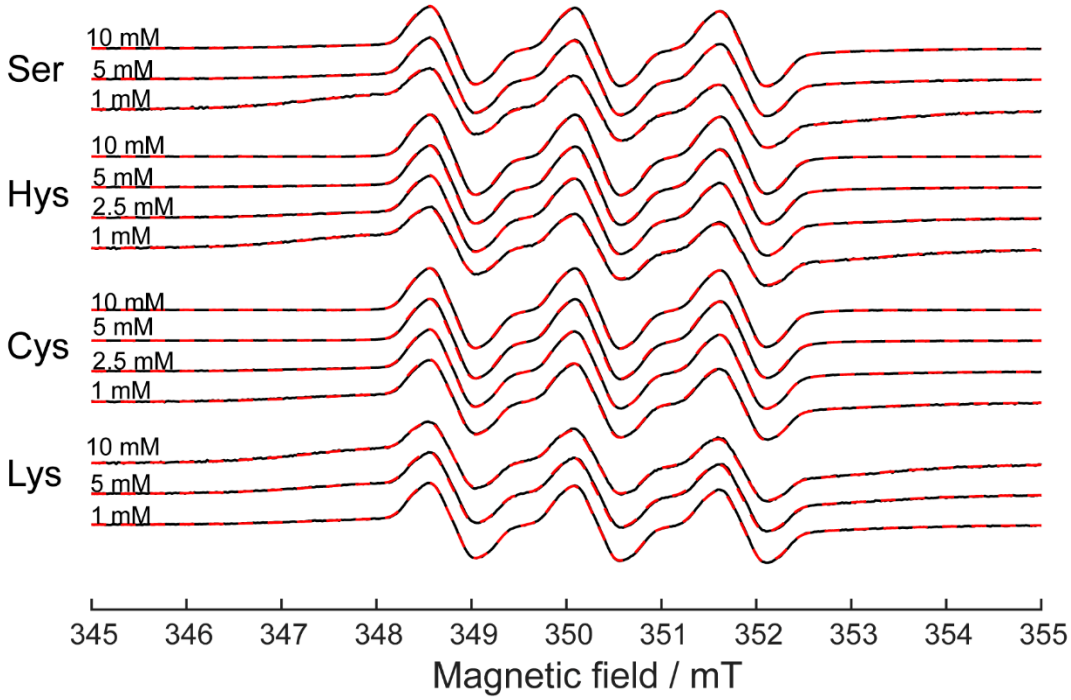

Figure S7. CW EPR spectra of R@ZIF-8 with amino acids addition at different concentrations. Red dashed lines show spectra simulation. All spectra were normalized by double integral.

EPR spectra of samples with 2-methylimidazole (MIM) addition.

We added 10mM MIM to all culture media and 9 amino acids to test whether MIM would stabilize nanoparticles. Moreover, MEM, DMEM and L-15 were tested with 30mM MIM. All solutions had pH=7.4. MIM addition resulted in increase of the broad spectrum component, therefore, the amount of dissolved nanoparticles decreased.

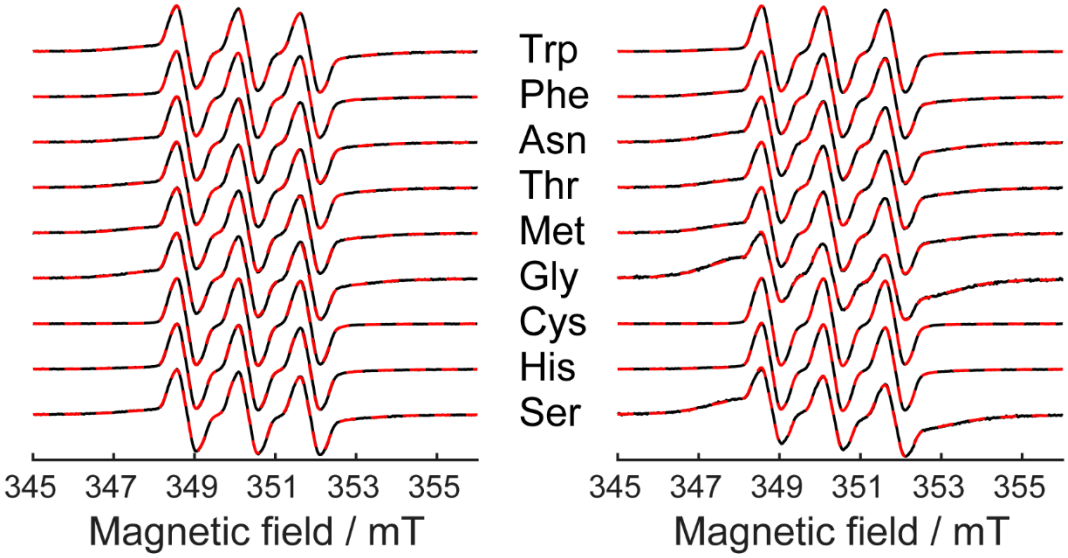

Figure S8. CW EPR spectra of R@ZIF-8 with addition of amino acids (5mM concentration). On the left - spectra of samples without MIM, on the right - samples with addition of 10mM MIM. Red dashed lines show spectra simulation. All spectra are normalized by double integral.

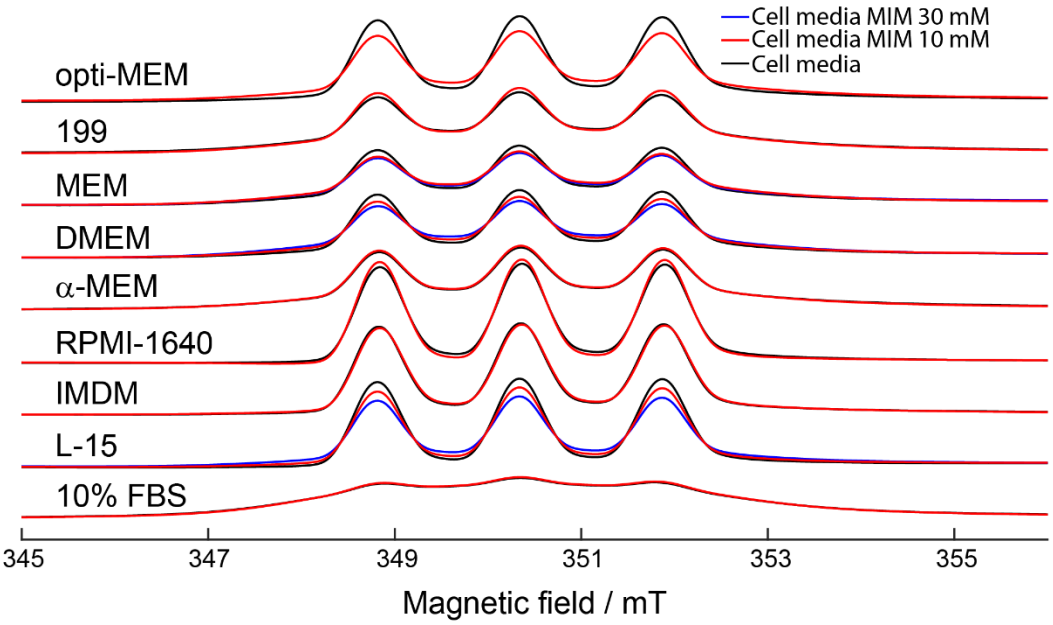

Figure S9. First integrals of CW EPR spectra of R@ZIF-8 after culture media addition, with and without MIM. All spectra are normalized by double integral.

Cytotoxic activity of MIM in A549 cells.

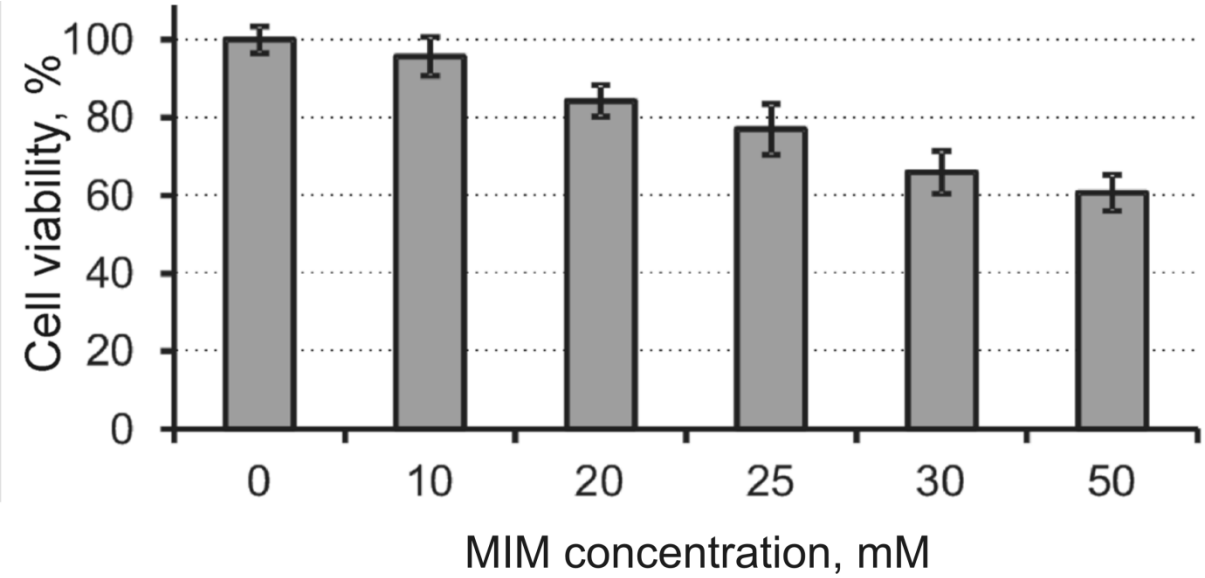

Figure S10. Cytotoxic activity of MIM in A549 cells. Cells were treated with different MIM concentration, and after 24 h of incubation, cells viability was analyzed by MTT assay.