



Bare iron oxide nanoparticles as drug delivery carrier for the short cationic peptide lasioglossin

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Particle characterization

Table S1: BION Size calculation with the Scherrer equation from XRD data, measured with Mo K α . The lattice constant is calculated from the Bragg equation.

2θ	16.01	13.72	19.40	Average
Size	9.07 nm	8.71 nm	9.74 nm	9.17 nm

Scherrer equation:

$$L = \frac{0.89-0.07093 \text{ nm}}{\Delta 2\theta \cdot \cos(\theta)} \quad (\text{S2})$$

Table S2: Modified Langevin fit for SQUID Analysis

	y0	xc	C	s
Magnetization	0.13902±0.66446	2.96189±25.77472	58.02718±0.79117	352.84062±22.54824

Boltzmann fit for Zeta Potential Analysis:

$$y = -39.61 + (15.46 - (-39.61)) / (1 + (e^{x-8.21})^{0.25}) \quad (\text{S1})$$

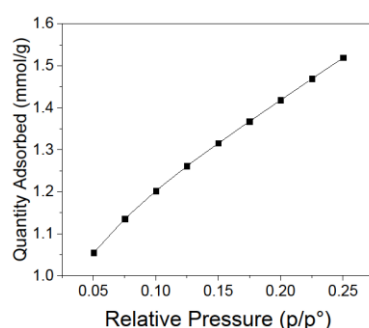
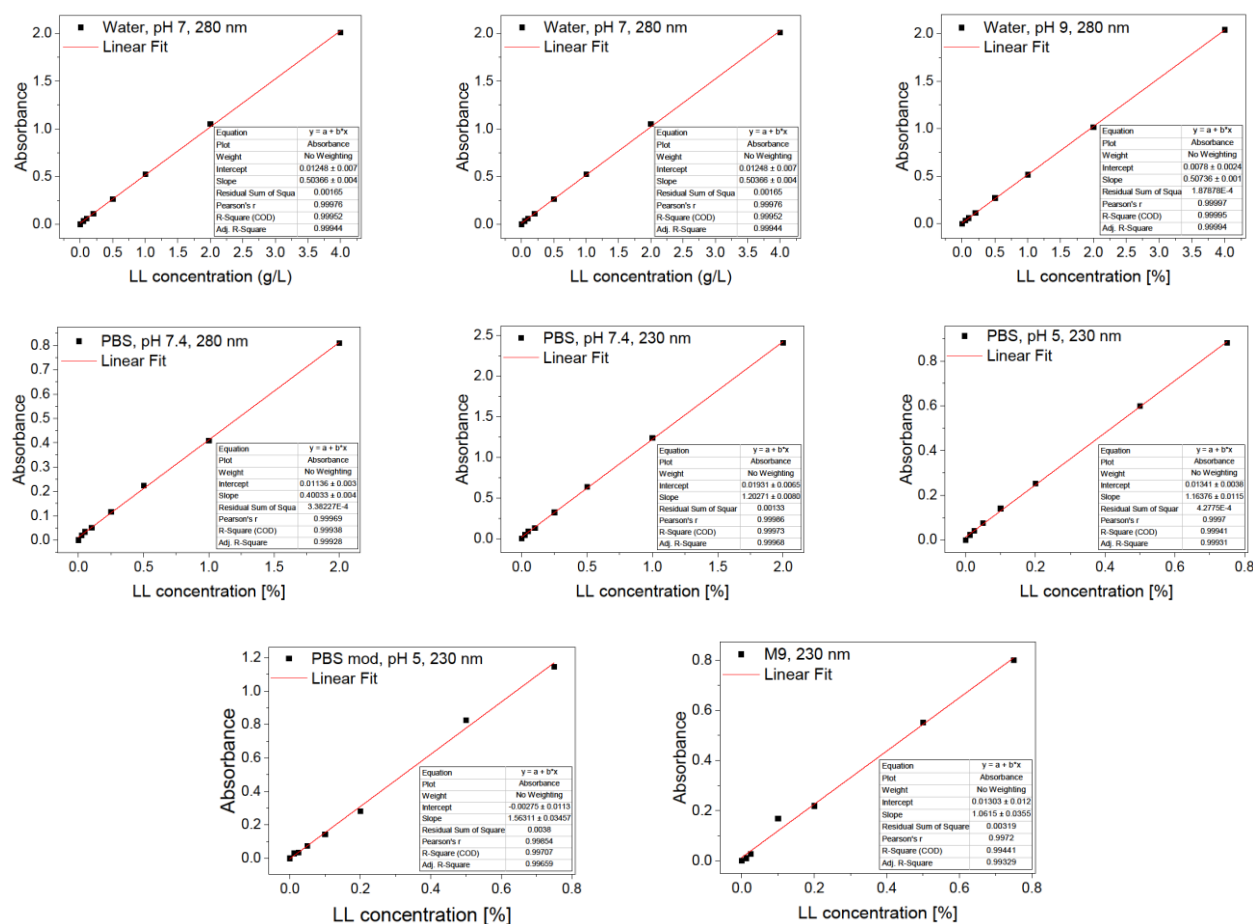


Figure S1: BET measurement: Adsorbed nitrogen to relative pressure.

Drug loading

$$\text{Drug loading} = \frac{m(\text{drug})}{m(\text{drug}) + m(\text{nanocarrier})} \quad (\text{S3})$$

Figure S2: Exemplary calibration curves for the photometric measurements for the adsorption and release experiments.

A new calibration curve was generated for each measurement, so these curves are exemplary for all the used conditions.

Table S3: Data of Adsorption and release experiments.

LL Conc.	Adsorption: Water, pH 7, 280 nm			Washing 1: Water, pH 7, 280 nm			Adsorption: Water, pH 9, 280 nm			Washing 1: Water, pH 9, 280 nm		
	Absorbance			Absorbance			Absorbance			Absorbance		
[g/L]	Value 1	Value 2	Value 3	Value 1	Value 2	Value 3	Value 1	Value 2	Value 3	Value 1	Value 2	Value 3
0	0.0727	0.059	0.0773	0.0414	0.0427	0.0414	0.0409	0.0432	0.0414	0.0403	0.0401	0.0403
0.025	0.0558	0.0972	0.127	0.0422	0.0426	0.0815	0.0656	0.0756	0.0555	0.0938	0.0767	0.0593
0.05	0.0839	0.0622	0.0822	0.0532	0.045	0.0452	0.0816	0.0698	0.0819	0.052	0.056	0.0496
0.1	0.107	0.103	0.162	0.0429	0.0478	0.0465	0.062	0.073	0.101	0.0441	0.046	0.0558
0.25	0.163	0.155	0.156	0.0496	0.0499	0.0455	0.113	0.0975	0.114	0.0423	0.0422	0.0465
0.5	0.262	0.274	0.237	0.0478	0.0496	0.0497	0.223	0.224	0.235	0.0501	0.0565	0.071
1	0.505	0.459	0.487	0.0648	0.0574	0.0739	0.452	0.444	0.427	0.0842	0.0744	0.0845
2	1	1.01	1.01	0.124	0.0969	0.11	0.834	0.892	0.93	0.127	0.136	0.15

LL Conc.	Adsorption: PBS, pH 7.4, 280 nm			Washing 1: PBS, pH 7.4, 280 nm			Adsorption: PBS, pH 7.4, 230 nm			Washing 1: PBS, pH 7.4, 230 nm		
	Absorbance			Absorbance			Absorbance			Absorbance		
[g/L]	Value 1	Value 2	Value 3	Value 1	Value 2	Value 3	Value 1	Value 2	Value 3	Value 1	Value 2	Value 3
0	0.0446	0.0429	0.0439	0.0456	0.0456	0.0455	0.082	0.0794	0.0803	0.0809	0.0809	0.0814
2	0.581	0.683	0.648	0.141	0.161	0.171	1.74	2.01	1.93	0.349	0.387	0.418
1	0.261	0.241	0.25	0.123	0.126	0.136	0.688	0.637	0.641	0.293	0.297	0.305
0.5	0.121	0.113	0.12	0.097	0.0943	0.105	0.274	0.263	0.214	0.209	0.194	0.199
0.25	0.0794	0.0849	0.0989	0.0833	0.12	0.0863	0.148	0.17	0.216	0.147	0.194	0.159
0.1	0.0642	0.0873	0.0895	0.0781	0.0855	0.0968	0.118	0.148	0.15	0.131	0.139	0.152
0.05	0.0693	0.0628	0.0921	0.0643	0.073	0.0915	0.119	0.114	0.148	0.114	0.122	0.144
0.025	0.0571	0.0532	0.0593	0.054	0.0601	0.0786	0.102	0.0958	0.104	0.0942	0.0992	0.125

Elution	LL Start Concentration	Absorbance: 1 h			Absorbance: 17 h			Absorbance: 27 h		
	[g/L]	Value 1	Value 2	Value 3	Value 1	Value 2	Value 3	Value 1	Value 2	Value 3
230 nm										
PBS, pH 5, 25	1	0.18	0.159	0.164						
°C	0	0.0801	0.0807	0.08						
PBS, pH 5, 40	1	0.167	0.159	0.168	0.171	0.164	0.181	0.189	0.177	0.196
°C	0	0.085	0.0859	0.0853	0.0842	0.0842	0.0844	0.0861	0.086	0.0861
PBS, pH 5, 60	1	0.33	0.268	0.205						
°C	0	0.088	0.0879	0.0888						
PBS mod, pH	1	0.23	0.221	0.234						
5, 25 °C	0	0.088	0.0889	0.0905						
PBS mod, pH	1	0.24	0.245	0.22						
5, 60 °C	0	0.0905	0.0909	0.0907						
M9, 37 °C	1	0.2	0.199	0.199	0.219	0.214	0.207	0.236	0.232	0.218
	0	0.185	0.189	0.182	0.201	0.215	0.218	0.211	0.211	0.212

Elution kinetic	LL Start Concentration							
230 nm	[g/L]	1 min	5 min	15 min	30 min	60 min	180 min	810 min
	1	0.23	0.226	0.227	0.233	0.225	0.232	0.264
PBS, pH 5, 25	1	0.24	0.247	0.221	0.228	0.226	0.238	0.343
°C	1	0.234	0.234	0.229	0.262	0.227	0.222	0.26
	0	0.113	0.112	0.102	0.106	0.102	0.103	0.107

Adsorption kinetic	LL Starting Concentration							
	[g/L]	1 min	6 min	16 min	31 min	61 min	181 min	1441 min
230 nm	1	0.316	0.287	0.259	0.287	0.27	0.275	0.263
PBS, pH 5, 25 °C	1	0.324	0.267	0.229	0.26	0.276	0.278	0.278
	1	0.321	0.277	0.244	0.275	0.272	0.277	0.271
	0	0.0453	0.0461	0.0453				

For the calculation of all experiments the blank is subtracted from the adsorption or desorption data.

Table S4: Influence of Washing on the peptide loading in PBS buffer.

Starting peptide concentration [g/L]	Equilibrium peptide concentration [g/L]	Loading Adsorption [g/g]	Loading Washing 1 [g/g]	Loss 1 [%]	Loading Washing 2 [g/g]	Loss 2 [%]	Loading Washing 3 [g/g]	Loss 3 [%]
2.00	1.45	0.55	0.29	46.1	0.24	17.7	0.20	15.5
1.00	0.49	0.51	0.33	34.8	0.27	20.2	0.21	22.2
0.50	0.17	0.34	0.24	30.4	0.15	37.7	0.06	58.4

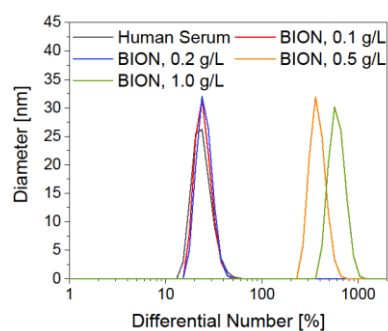


Figure S3: Agglomeration behavior of BIONs in Human Serum depending on the concentration.

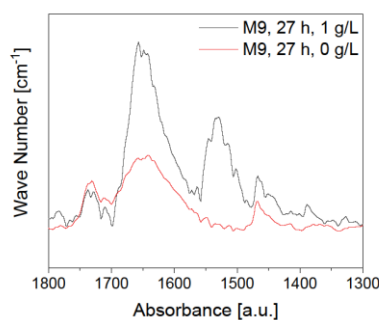


Figure S4: IR spectra of BION@LL and only BIONs after 27 h desorption in M9 medium.

Growth studies

Table S5: Components of M9 Medium for 1 L (867 ml sterile water).

Solution	Composition	Amount of substances	Volume
M9 salt solution (10x)	Na ₂ HPO ₄	33.7 mM	100 mL
	KH ₂ PO ₄	22.0 mM	
	NaCl	8.55 mM	
	NH ₄ Cl	9.35 mM	
20% glucose solution	Glucose monohydrate	0.40%	20 mL
1 M MgSO ₄ solution	MgSO ₄ ·7 H ₂ O	1.00 mM	1 mL
1 M CaCl ₂ solution	CaCl ₂ ·2 H ₂ O	0.30 mM	0.3 mL
1 M mg/mL biotin solution	Biotin	1.00 µg	1 mL
1 M mg/mL thiamin solution	Thiamin	1.00 µg	1 mL
Trace elements solution (100x)	EDTA	13.4 mM	10 mL (1x)
	FeCl ₃ ·6 H ₂ O	3.10 mM	
	ZnCl ₂	0.62 mM	
	CuCl ₂ ·2 H ₂ O	76.0 µM	
	CoCl ₂ ·2 H ₂ O	42.0 µM	
	H ₃ BO ₃	162 µM	
	MNCl ₂ · 4 H ₂ O	8.10 µM	

Microscopy

For each measurement, five group squares of biological triplicates are counted.

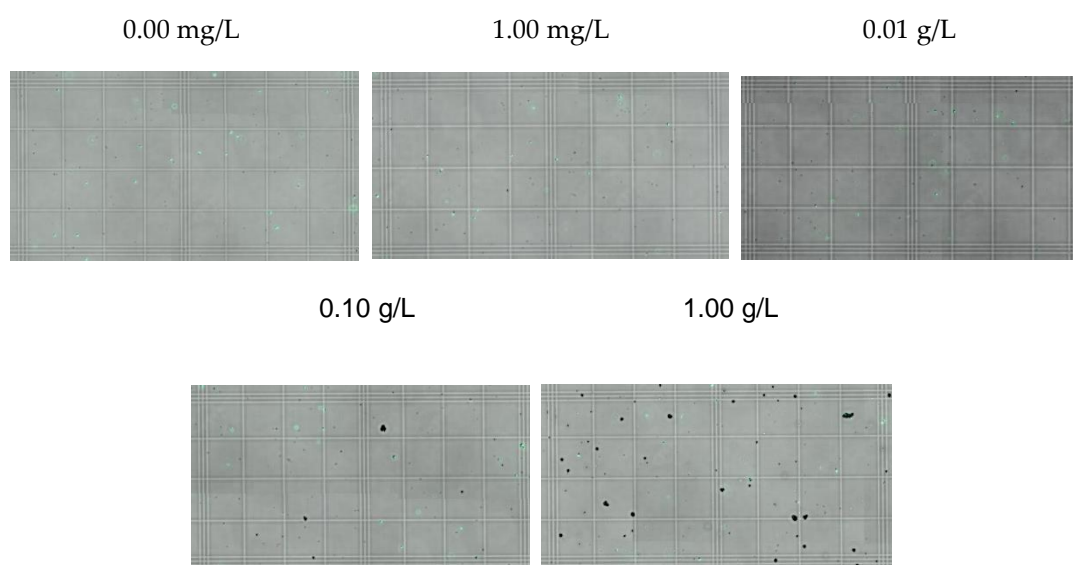


Figure S5: *E. coli* colony grown in M9 media with ampicillin, incubated with different concentrations of BIONs (37 °C, addition of IPTG after 5 h).

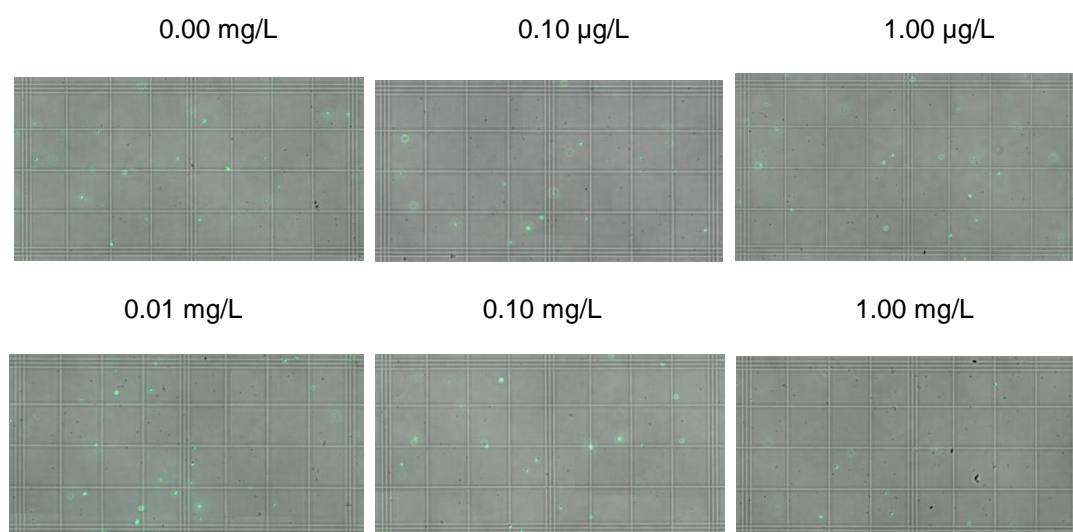
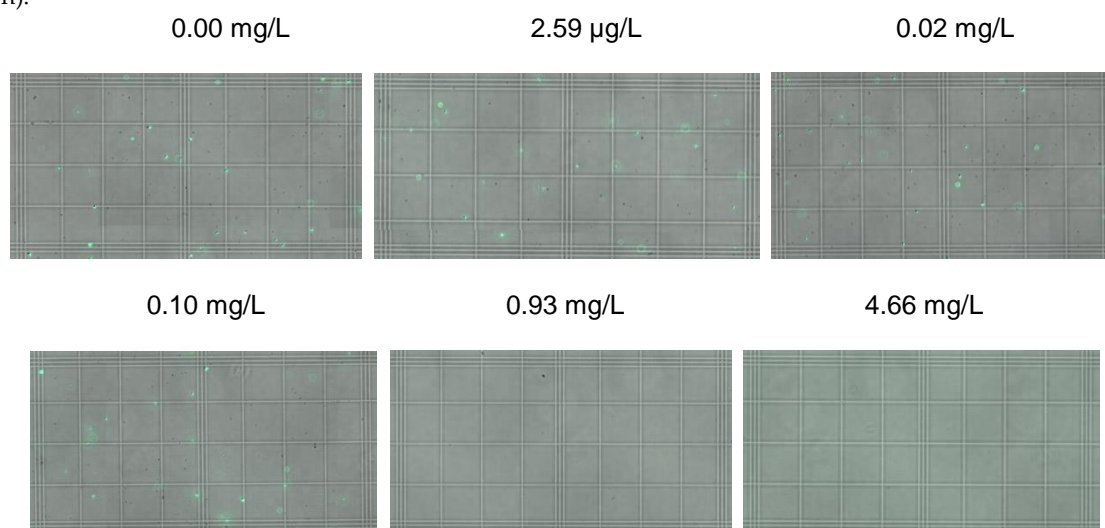


Figure S6: *E. coli* colony grown in M9 media with ampicillin, incubated with different concentrations of LL (37 °C, addition of IPTG after 5 h).



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Figure S7: *E. coli* colony grown in M9 media with ampicillin, incubated with different concentrations of BION@LL (37 °C, addition of IPTG after 5 h).

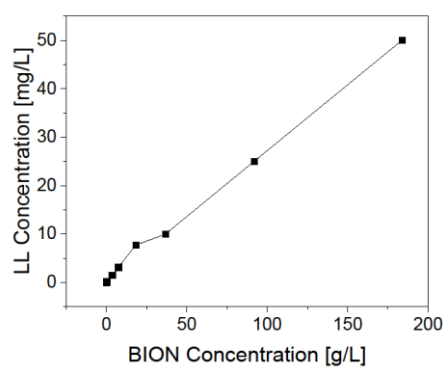


Figure S8: Amount of LL loaded on BIONs.