SUPPLEMENTARY INFORMATION

RNA Aptamers with Specificity for Heparosan and Chondroitin Glycosaminoglycans

Megan Kizer[1,6], Peiqin Li[2,3,6*], Brady F. Cress[4,6], Lei Lin[1,6], Tom T Jing[1,6], Xing Zhang[1,6], Ke Xia[1,6], Robert J. Linhardt[1,3,4,5,6*], and Xing Wang[1,6*]

[1] Department of Chemistry and Chemical Biology, Rensselaer Polytechnic Institute[1]

[2] Department of Forest Pathology, College of Forestry, Northwest A&F University[2]

[3] Department of Biology, Rensselaer Polytechnic Institute[1]

[4] Department of Chemical and Biological Engineering, Rensselaer Polytechnic Institute[1]

[5] Department of Biomedical Engineering, Rensselaer Polytechnic Institute[1]

[6] Center for Biotechnology and Interdisciplinary Studies, Rensselaer Polytechnic Institute[1]

* Corresponding Authors: lipg@nwafu.edu.cn, linhar@rpi.edu, wangx28@rpi.edu

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[1] Rensselaer Polytechnic Institute, 110 8th Avenue, Troy, New York 12180
[2] Northwest A&F University, Yangling 712100, Shaanxi, China

Table of Contents

Tables:

Methods:

Figures:

Table S1. Sequences of the candidate RNA aptamers recovered post SELEX against heparosan (HE) GAG. The constant primer binding regions are indicated by italic letters.

Group	Aptamer RNA No.	No. of hits	Representative sequences	Molecular weight (g/mol)
A	HE-01	1	*GGGAAGAGAAGGACAUAUGAU*CCGCAUAAAGUGUAUCCGAUUUGGUUGCUG*UUGACUAGUACAUGACCACUUGA*	22686.4
	HE-02	1	*GGGAAGAGAAGGACAUAUGAU*CGAAUAGAAACUCGAGGUAAAAGCGGAGUG*UUGACUAGUACAUGACCACUUGA*	22904.7
	HE-03	20	*GGGAAGAGAAGGACAUAUGAU*GAAUGGAGGGAGGGAUAGCCUUCGUGAGGG*UUGACUAGUACAUGACCACUUGA*	22985.7
	HE-04	1	*GGGAAGAGAAGGACAUAUGAU*UCGGAAUGAGCGCAGAAGUAGCGCAUUGGC*UUGACUAGUACAUGACCACUUGA*	22849.6
	HE-05	1	*GGGAAGAGAAGGACAUAUGAU*GCCUGGCCCAAUAAAGUUCAUCGUCCGGGG*UUGACUAGUACAUGACCACUUGA*	22722.5
	HE-06	3	*GGGAAGAGAAGGACAUAUGAU*CCCCAUUGCGGCCAAAAUGACAUAGGCGUG*UUGACUAGUACAUGACCACUUGA*	22729.5
	HE-07	1	*GGGAAGAGAAGGACAUAUGAU*UCGUUCAAGACGGCCUCUGGUUGCGGAUGGC*UUGACUAGUACAUGACCACUUGA*	22755.5
B	HE-08	2	*GGGAAGAGAAGGACAUAUGAU*GCGGGAUGUGGUGUACCCGCUAUCCCAGGC*UUGACUAGUACAUGACCACUUGA*A	22745.5
	HE-09	2	*GGGAAGAGAAGGACAUAUGAU*UCGCACUCAAUGAAAGCGGGUCAAUUGCGCG*UUGACUAGUACAUGACCACUUGA*	22769.5
	HE-10	2	*GGGAAGAGAAGGACAUAUGAU*CCAACAGUGGGACAGAGACGACUUAGCGCG*UUGACUAGUACAUGACCACUUGA*	22831.6
	HE-11	2	*GGGAAGAGAAGGACAUAUGAU*GUAGGCAUGAGAGCACUGUGGCUGACCGGA*UUGACUAGUACAUGACCACUUGA*	22865.6
	HE-12	5	*GGGAAGAGAAGGACAUAUGAU*GGAGGGAGGAGGGAAGGGUGAGGGAGAUGU*UUGACUAGUACAUGACCACUUGA*	22791.6
	HE-13	1	*GGGAAGAGAAGGACAUAUGAU*CCAACGGUGGGCAGGAUAUCUCAUGCAGUG*UUGACUAGUACAUGACCACUUGA*	22786.5
	HE-14	1	*GGGAAGAGAAGGACAUAUGAU*CUCAAUAAGAGACAGCUUCCGGUGGCUUGG*UUGACUAGUACAUGACCACUUGA*	22747.5
	HE-15	1	*GGGAAGAGAAGGACAUAUGAU*CCCUGGUCUGGCCUUAACUACUCUCAGAGC*UUGACUAGUACAUGACCACUUGA*	22574.3
	HE-16	2	*GGGAAGAGAAGGACAUAUGAU*CCCAAACAGGAAAAGGGAUGGGGUCAGCCG*UUGACUAGUACAUGACCACUUGA*	22894.7
	HE-17	1	*GGGAAGAGAAGGACAUAUGAU*CCAAAACAACGUGGUCGAAGCAUGAUCCGC*UUGACUAGUACAUGACCACUUGA*	22736.6
	HE-18	1	*GGGAAGAGAAGGACAUAUGAU*CCUAUGCGAAAGCUCACUACCGAAGUCGGG*UUGACUAGUACAUGACCACUUGA*	22729.5
	HE-19	3	*GGGAAGAGAAGGACAUAUGAU*GGAUCAGUAUCCCACAACGAAAGCGCCGGG*UUGACUAGUACAUGACCACUUGA*	23167.9

Note: RNAs in the group A (GA) were recovered post SELEX with only using blank beads during the negative selection. RNAs in the group B (GB) were obtained post SELEX with using the DNA oligonucleotides, complementary to ‘background-binding’ RNAs, during the negative selection.

Table S2. Sequences of the candidate RNA aptamers candidates recovered post SELEX against chondroitin (CH) GAG. The constant primer binding regions are indicated by italic letters.

RNA No.	No. of hits	Representative Sequences (5'-3')	Molecular weight, g/mol
CH-01	1	*GGGAAGAGAAGGACAUAUGAU*CUCGCUCCGGGGGCGGCCCUGAGCGGCGC*UUGACUAGUACAUGACCACUUGA*	22470.3
CH-03	1	*GGGAAGAGAAGGACAUAUGAU*UGCCAAUGGGAUCGCACAAGGAAUGGCCGG*UUGACUAGUACAUGACCACUUGA*	22848.6
CH-04	3	*GGGAAGAGAAGGACAUAUGAU*CCUGGGGCGGCGAGAGUAUGCGCGCGGCGG*UUGACUAGUACAUGACCACUUGA*	22927.6
CH-05	1	*GGGAAGAGAAGGACAUAUGAU*GCUGUAUCAAACAGUGUAGCGGCACCUGCC*UUGACUAGUACAUGACCACUUGA*	22393.3
CH-07	1	*GGGAAGAGAAGGACAUAUGAU*CGCGUGUCGGCUCUUUGCCUCGUUGUGCCA*UUGACUAGUACAUGACCACUUGA*	22606.3
CH-08	1	*GGGAAGAGAAGGACAUAUGAU*CAUUCGCGCAAGUUUGUCGGCCGCGUGGUG*UUGACUAGUACAUGACCACUUGA*	22732.4
CH-09	1	*GGGAAGAGAAGGACAUAUGAU*CGGAUUGGGCAGAGGCUCGUACGUAUCGGC*UUGACUAGUACAUGACCACUUGA*	22818.5
CH-10	1	*GGGAAGAGAAGGACAUAUGAU*AGCCGGGCACCAUCUGGCAGAACGCUCCCG*UUGACUAGUACAUGACCACUUGA*	22719.5
CH-11	1	*GGGAAGAGAAGGACAUAUGAU*ACAACUGCUCUGAGGGCAGCUAGUUGGGCC*UUGACUAGUACAUGACCACUUGA*	22762.5
CH-12	1	*GGGAAGAGAAGGACAUAUGAU*CGGGAACGGUAAUCUAUCGUGUGGCCUGCA*UUGACUAGUACAUGACCACUUGA*	22763.5
CH-13	1	*GGGAAGAGAAGGACAUAUGAU*CCCAAUGGGAUCGCACAAGGAAUGGCCGG*UUGACUAGUACAUGACCACUUGA*	22518.4
CH-17	1	*GGGAAGAGAAGGACAUAUGAU*AGGGAAGGUGCGGGUUGUCCAGUAGUGCAG*UUGACUAGUACAUGACCACUUGA*	22922.6
CH-18	2	*GGGAAGAGAAGGACAUAUGAU*UCCGCGGGAGGUCGGAAUGCGCUCAUG*UUGACUAGUACAUGACCACUUGA*	21885.9
CH-20	5	*GGGAAGAGAAGGACAUAUGAU*CUGGAAUCGACGGGCAGGGCUCAGUUGCGC*UUGACUAGUACAUGACCACUUGA*	22817.5
CH-21	1	*GGGAAGAGAAGGACAUAUGAU*UAGGGCAGGUGUAGGGUUGGUCCUUCGGCG*UUGACUAGUACAUGACCACUUGA*	22852.5
CH-23	1	*GGGAAGAGAAGGACAUAUGAU*CAGGCUCCGGGGGCGGCCCUGAGCGGCGC*UUGACUAGUACAUGACCACUUGA*	22556.4
CH-26	1	*GGGAAGAGAAGGACAUAUGAU*GCAUAAGAUCCACAGUACGAGCGGUUGCAG*UUGACUAGUACAUGACCACUUG*	22480.4
CH-27	1	*GGGAAGAGAAGGACAUAUGAU*CCACGAUGAAGUGCGAAGGGGGCGGUCUCC*UUGACUAGUACAUGACCACUUGA*	22840.6
CH-29	1	*GGGAAGAGAAGGACAUAUGAU*UUACGCAGAGCAGAUUAGCCGAGCCGCACC*UUGACUAGUACAUGACCACUUGA*	22728.5
CH-31	1	*GGGAAGAGAAGGACAUAUGAU*ACCGCCGGAGUGGAUAGGCAGGGGUGGUAG*UUGACUAGUACAUGACCACUUGA*	22960.7
CH-32	1	*GGGAAGAGAAGGACAUAUGAU*CCCGAUGUACGCGGUUUGGGGGCGUCAGGC*UUGACUAGUACAUGACCACUUGA*	22810.5
CH-33	1	*GGGAAGAGAAGGACAUAUGAU*AGGGACAAAGGUUGGGGUUGUCCGAGGCGG*UUGACUAGUACAUGACCACUUGA*	22961.7
CH-34	1	*GGGAAGAGAAGGACAUAUGAU*GAGGUACUGGGGGGGAGGCCGAAUGUGCAU*UUGACUAGUACAUGACCACUUGA*	22961.7
CH-35	10	*GGGAAGAGAAGGACAUAUGAU*GGAGGGGAUUGGGGGCAUGUUGGGGCGUCC*UUGACUAGUACAUGACCACUUGA*	22970.6
CH-36	1	*GGGAAGAGAAGGACAUAUGAU*CCUGUCAUGGGGGGAGUCGGACGCGGUGA*UUGACUAGUACAUGACCACUU*	21942.0
CH-37	1	*GGGAAGAGAAGGACAUAUGAU*GCGUUUCCGUGCGCCUUUCAGACGUCUGUG*UUGACUAGUACAUGACCACUUGA*	22630.3
CH-38	1	*GGGAAGAGAAGGACAUAUGAU*CCUCAGCUCGUUGGACCCUGCCGGCCAUCC*UUGACUAGUACAUGACCACUUGA*	22570.3
CH-39	2	*GGGAAGAGAAGGACAUAUGAU*CCGCGGUGCUCACGGGGAGGAAGCUGUUAG*UUGACUAGUACAUGACCACUUGA*	22857.6

CH-41	1	*GGGAAGAGAAGGACAUAUGAU*UCGUUCUGGAAUAGGGCAAGGGUCAGCCAG*UUGACUAGUACAUGACCACUUGA*	22826.6
CH-48	1	*GGGAAGAGAAGGACAUAUGAU*AGGAGGGGAUUGGGGCAUAUAGGGGUGUCC*UUGACUAGUACAUGACCACUUGA*	22962.7
CH-52	1	*GGGAAGAGAAGGACAUAUGAU*CAGGUUAGCAAUCUUGGAAAGCGAUCACUG*UUGACUAGUACAUGACCACUUGA*	22755.5
CH-56	1	*GGGAAGAGAAGGACAUAUGAU*CUUGAGGGGCGGGGAGGUAAAGCAGUUUGA*UUGACUAGUACAUGACCACUUGA*	22946.7
CH-57	8	*GGGAAGAGAAGGACAUAUGAU*GAGCGCGUCGCCGUGUUACCGCGGGGGGUG*UUGACUAGUACAUGACCACUUG*	22552.3
CH-59	1	*GGGAAGAGAAGGACAUAUGAU*CCGGUUCGUACGGCGCGGCGUCGUCCUCCG*UUGACUAGUACAUGACCACUUGA*	22682.4
CH-62	1	*GGGAAGAGAAGGACAUAUGAU*CGCCAACUAAGCAACCAAUUCAUGCGCGGC*UUGACUAGUACAUGACCACUUGA*	22672.5
CH-64	2	*GGGAAGAGAAGGACAUAUGAU*GGAGGGGUAGUGGGGCUUAGAGGGGCGUCC*UUGACUAGUACAUGACCACUUGA*	22993.7
CH-65	3	*GGGAAGAGAAGGACAUAUGAU*GGGACGGCGAGCCCGGCGUGCUAUCCCAGC*UUGACUAGUACAUGACCACUUGA*	22791.5
CH-69	1	*GGGAAGAGAAGGACAUAUGAU*GACAGGGAAUUGAUGCGUUUGGCCCGGCGG*UUGACUAGUACAUGACCACUUGA*	22858.6

Note: All RNAs were obtained post SELEX with using the DNA oligonucleotides, complementary to ‘background-binding’ RNAs, during the negative selection.

Table S3. DNA oligonucleotides used for negative selection

No.	Sequence (5’-3’)
1	CCCTTCCC
2	CCCTCCCT
3	CCCTCCTT
4	CCCCTCCA
5	CCCACCCG
6	CGC ACC CCT CCG CCC TCC
7	CCT TCC TCC TCC TCC TTG
8	CTA TCC CTC CCT CC
9	CCC CTC CAC CCT CCT
10	TCC CGC CCT CCT
11	TAC CCT ACC CTC CA

Table S4. Sequence of the RNA Library and Selection Primers (DNA) used in this study

Name	Sequence (5’-3’)
Random RNA Library	UAGGGAAGAGAAGGACAUAUGAU (N_{30}) UUGACUAGUACAUGACCACUUGA
T7 Forward Primer	TAG GGA AGA GAA GGA CAT ATG AT
T7 Reverse Primer	TCA AGT GGT CAT GTA CTA GTC AA

N_{30} indicates 30 nucleotides with equimolar incorporation of A, U, G and C at each position.

Table S5. Non-linear regression analysis for K_D calculation of GAG-RNA binding complexes.

GAG	RNA Aptamer	Fitted Equation	R^2	B_{max}	K_D (μM)
Heparosan	HE-08	$B = 54.526\ln(x) + 107.87$	0.9870	184	0.75
	HE-13	$B = 26.586\ln(x) + 47.686$	0.9476	95	1.0
	HE-14	$B = 24.408\ln(x) + 51.972$	0.9965	87	0.71
Chondroitin	CH-09	$B = 36.791\ln(x) + 53.94$	0.9361	110	1.0
	CH-20	$B = 17.242\ln(x) + 33.161$	0.9287	54	0.76
	CH-32	$B = 23.042\ln(x) + 38.433$	0.9549	71	0.89
Heparin	HE-14	$B = 8.2488\ln(x) + 16.638$	0.8403	31	0.87

"*B*" indicates the fluorescence intensity of GAG-RNA binding complex.

Table S6. Quantification of glycosaminoglycan immobilized on the beads or microplate.

GAG	Total Amount of Carbohydrate	
	Beads (ng/μL)	Microplate (ng/well)
Heparosan (HE)	87.0	21.2
Chondroitin (CH)	5.2	78.4
Hyaluronan (HA)	83.3	ND

ND = not determined; HA GAG was not immobilized on microplates.

Methods

1. Nuclear Magnetic Resonance for confirmation of GAG purity and modification.

All the GAGs and biotinylated-GAGs were dissolved in 0.5 mL of 99.6% D_2O centrifuged at 5000 x *g* for 2 min and lyophilized. The process was repeated twice, and the final samples were dissolved in 0.5 mL of 99.96% D_2O. ^1H spectroscopy experiments were carried out at 298 K on a Bruker 800 MHz NMR spectrometer recorded for 32 scans with Topspin 2.1.6 software.

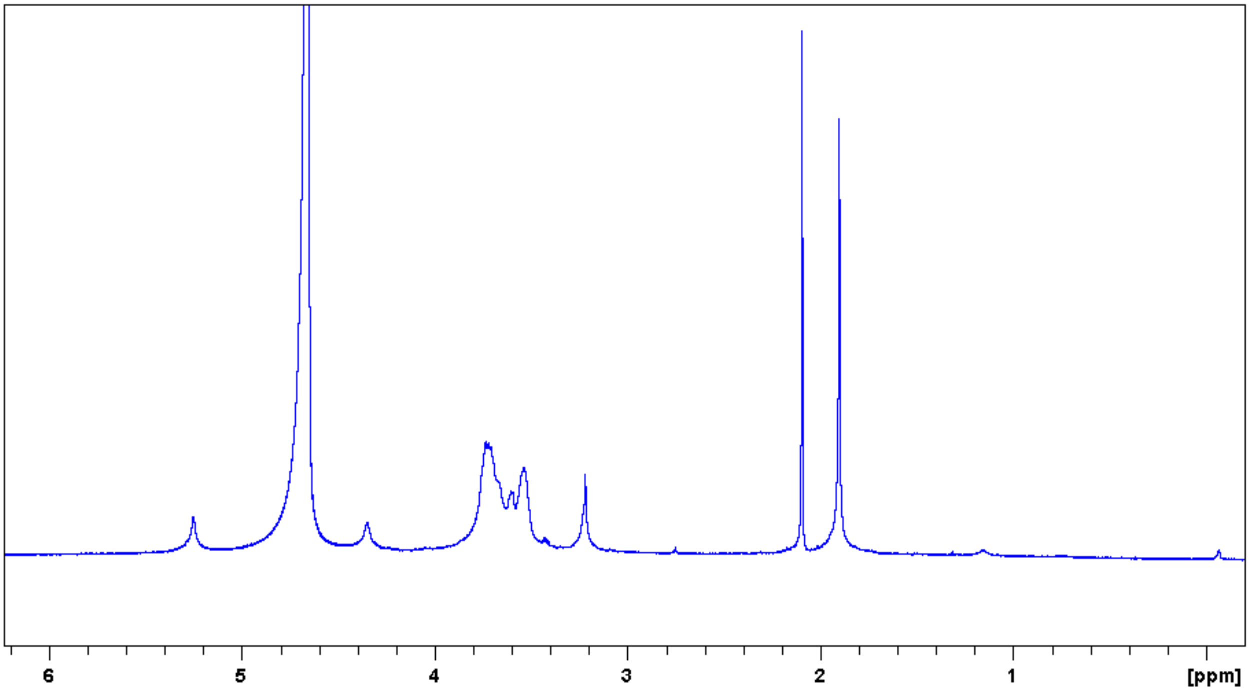
2. Liquid chromatography-mass spectrometry for GAG purity.

GAGs were digested and AMAC labeled following the method in the main text.

LC was performed on an Agilent 1200 LC system at 45 °C using an Agilent Poroshell 120 ECC18 (2.7 µm, 2.1 × 150 mm) column. Mobile phase A (MPA) was 50 mM ammonium acetate aqueous solution, and the mobile phase B (MPB) was methanol. The mobile phase passed through the column at a flow rate of 150 µL/min. The gradient start from 10% B to 46% B in 36 mins.

An Ion Trap mass spectrometry system equipped with an ESI source (Agilent 6340) was used as detector. Full scan used from 350-900 m/z. In-software integration was used to calculate purity.

A.



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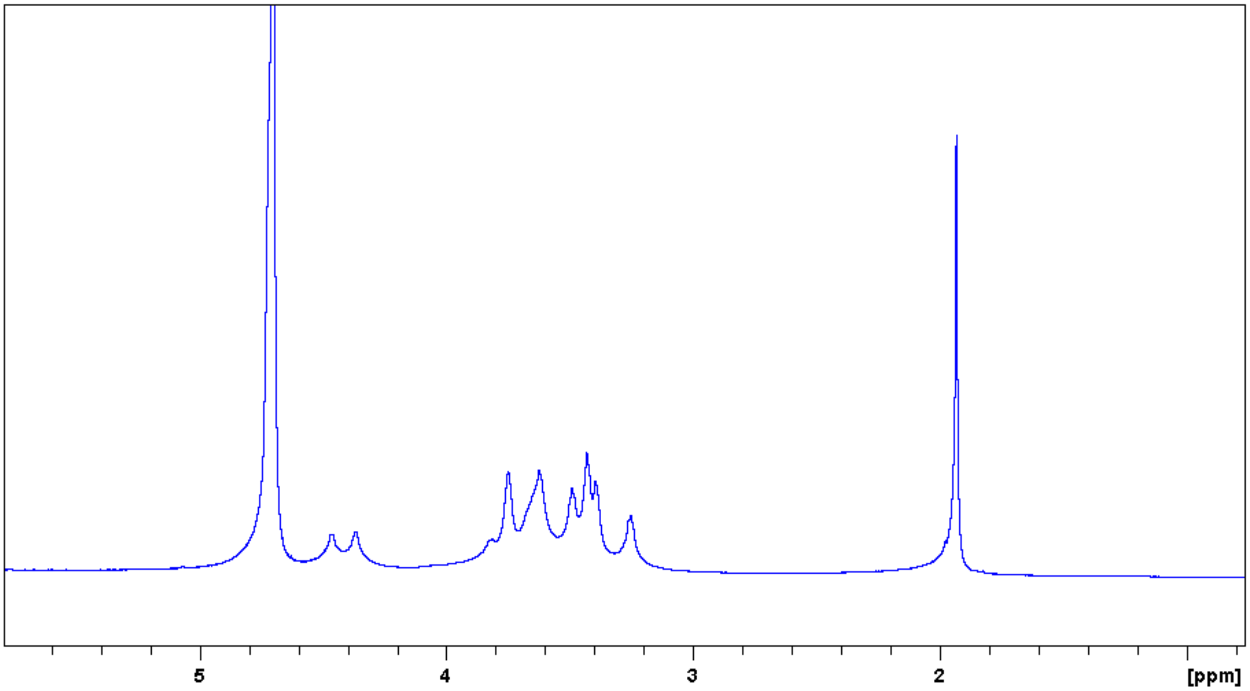

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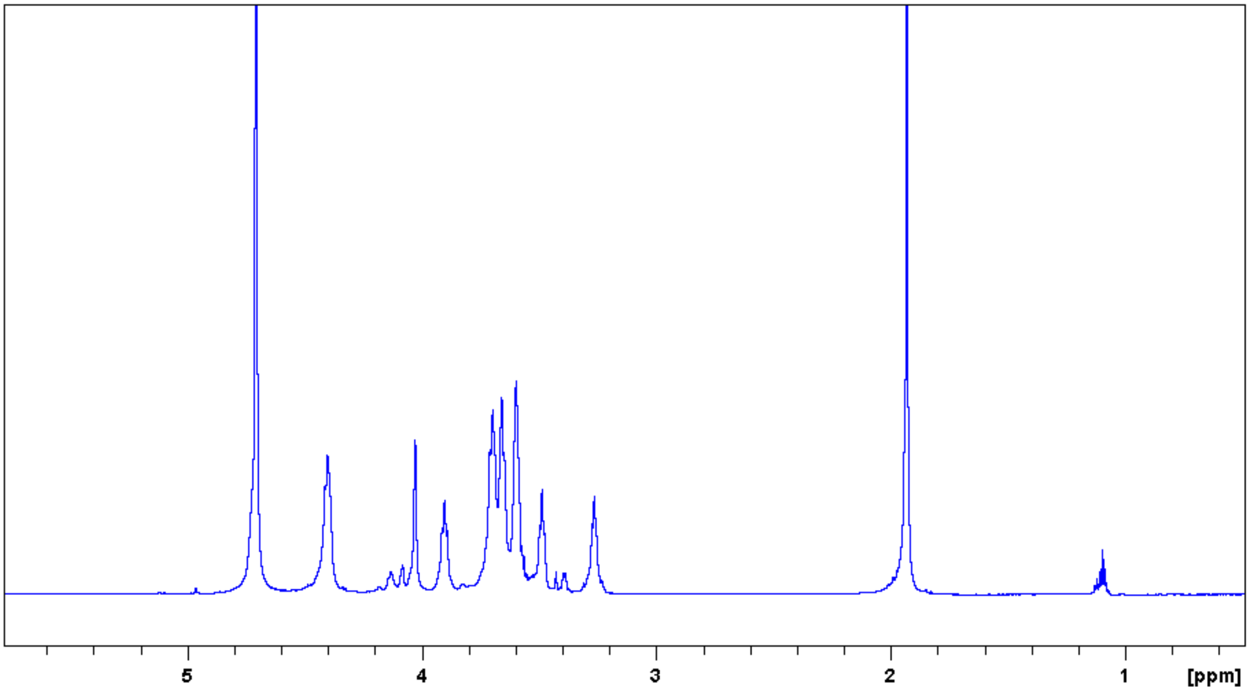


Figure S1. NMR Spectra of pure heparosan (A), hyaluronan (B) and chondroitin (C).

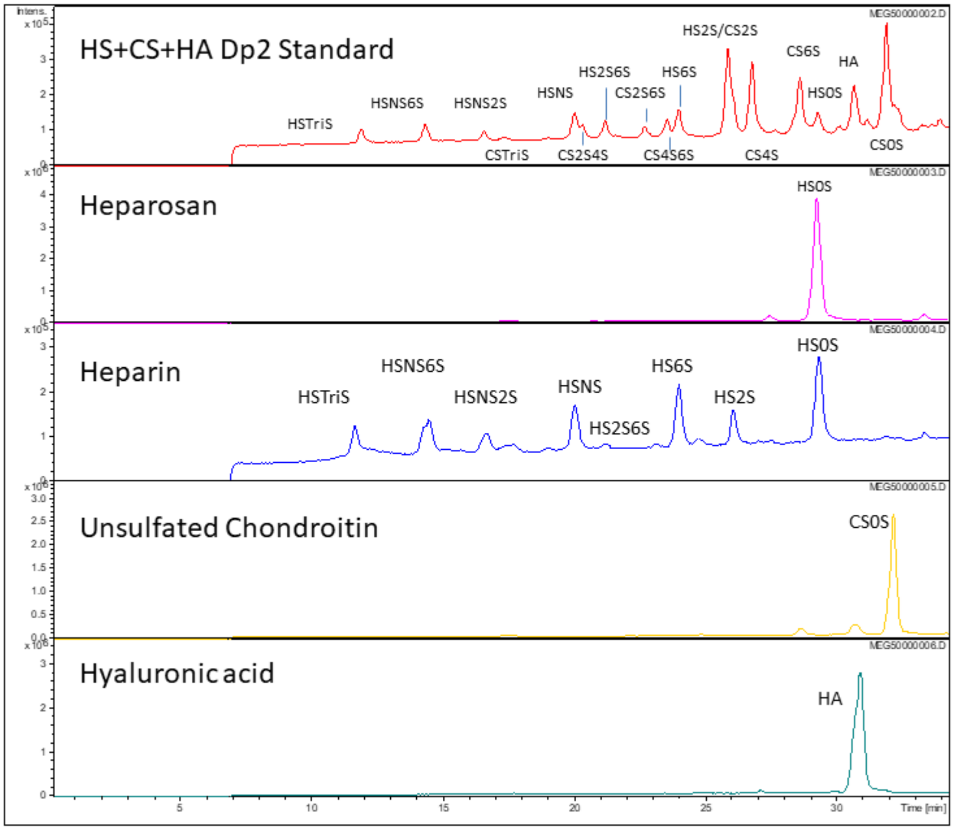
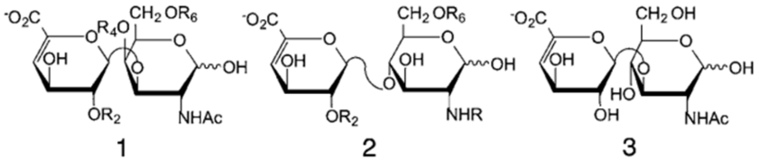
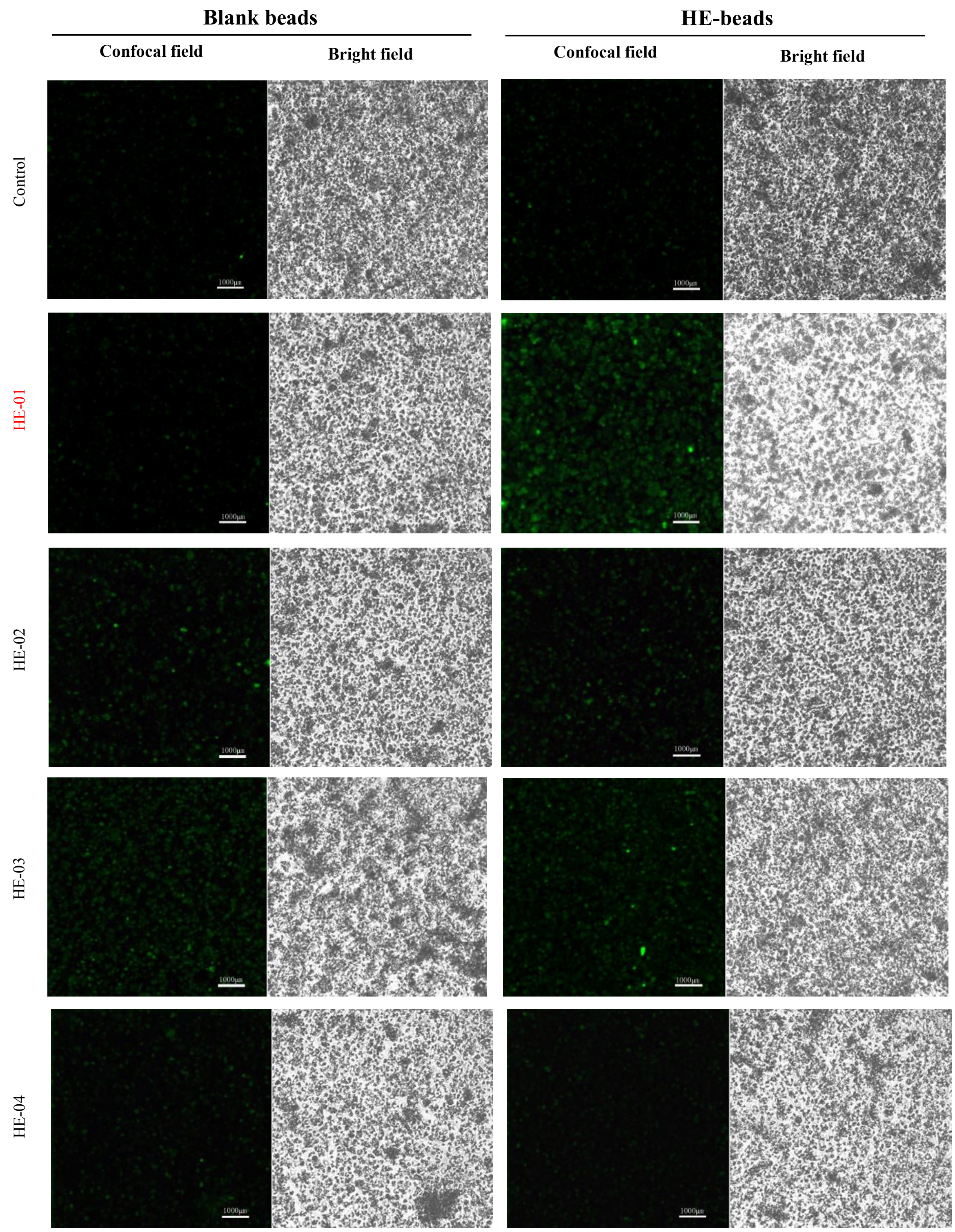


Figure S2. LC/MS full scan of disaccharide (Dp2) generated from GAG samples comparing with HS, CS, HA Dp2 standard. Samples tested are almost pure. Heparosan>97%, Heparin>99%, Chondroitin >95%, Hyaluronic acid>99%.



CS disaccharides	structure **1**	R_2	R_4	R_6
TriS_{CS}	ΔUA2S(1,3)GalNAc4S6S	SO_3^-	SO_3^-	SO_3^-
2S4S_{CS}	ΔUA2S(1,3)GalNAc4S	SO_3^-	SO_3^-	H
2S6S_{CS}	ΔUA2S(1,3)GalNAc6S	SO_3^-	H	SO_3^-
4S6S_{CS}	ΔUA(1,3)GalNAc4S6S	H	SO_3^-	SO_3^-
2S_{CS}	ΔUA2S(1,3)GalNAc	SO_3^-	H	H
4S_{CS}	ΔUA(1,3)GalNAc4S	H	SO_3^-	H
6S_{CS}	ΔUA(1,3)GalNAc6S	H	H	SO_3^-
0S_{CS}	ΔUA(1,3)GalNAc	H	H	H
HS disaccharides	structure **2**	R_2	NR	R_6
TriS_{HS}	ΔUA2S(1,4)GlcNS6S	SO_3^-	SO_3^-	SO_3^-
NS6S_{HS}	ΔUA(1,4)GlcNS6S	H	SO_3^-	SO_3^-
NS2S_{HS}	ΔUA2S(1,4)GlcNS	SO_3^-	SO_3^-	H
NS_{HS}	ΔUA(1,4)GlcNS	H	SO_3^-	H
2S6S_{HS}	ΔUA2S(1,4)GlcNAc6S	SO_3^-	Ac	SO_3^-
6S_{HS}	ΔUA(1,4)GlcNAc6S	H	Ac	SO_3^-
2S_{HS}	ΔUA2S(1,4)GlcNAc	SO_3^-	Ac	H
0S_{HS}	ΔUA(1,4)GlcNAc	H	Ac	H
HA disaccharides	structure **3**			
0S_{HA}	ΔUA(1,3)GlcNAc			

Figure S3. Structure of unsaturated disaccharide standards.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HE-03
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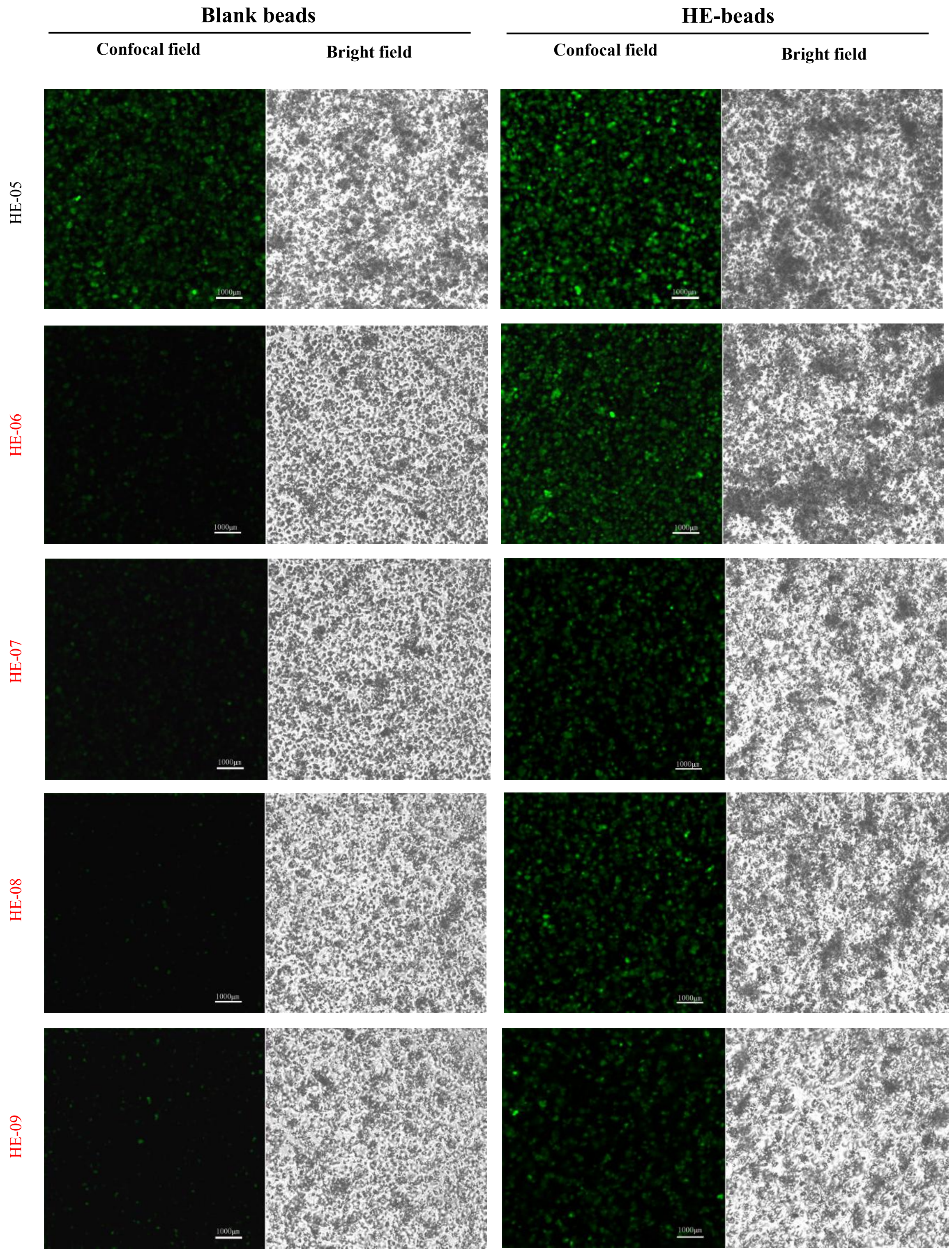
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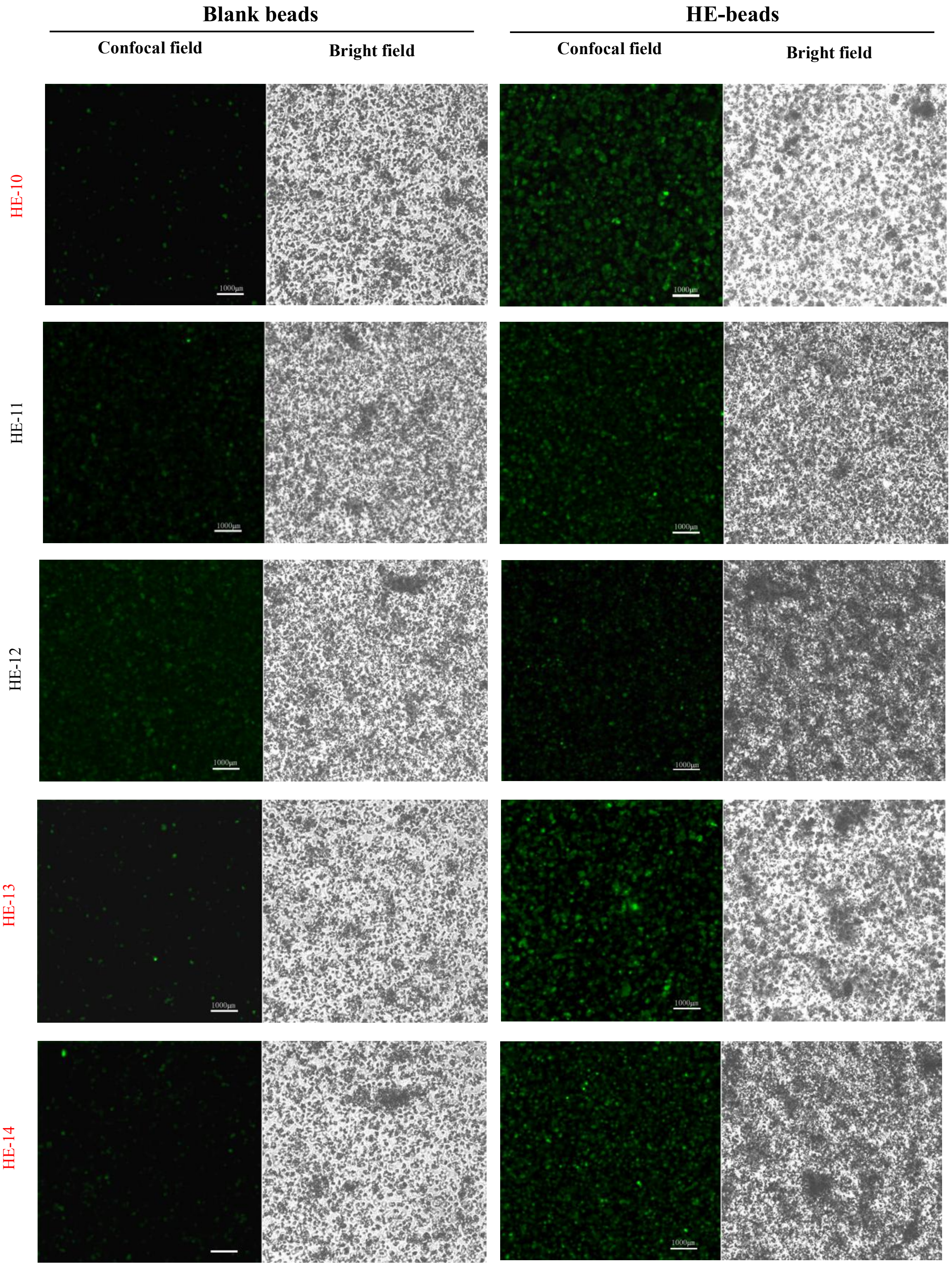
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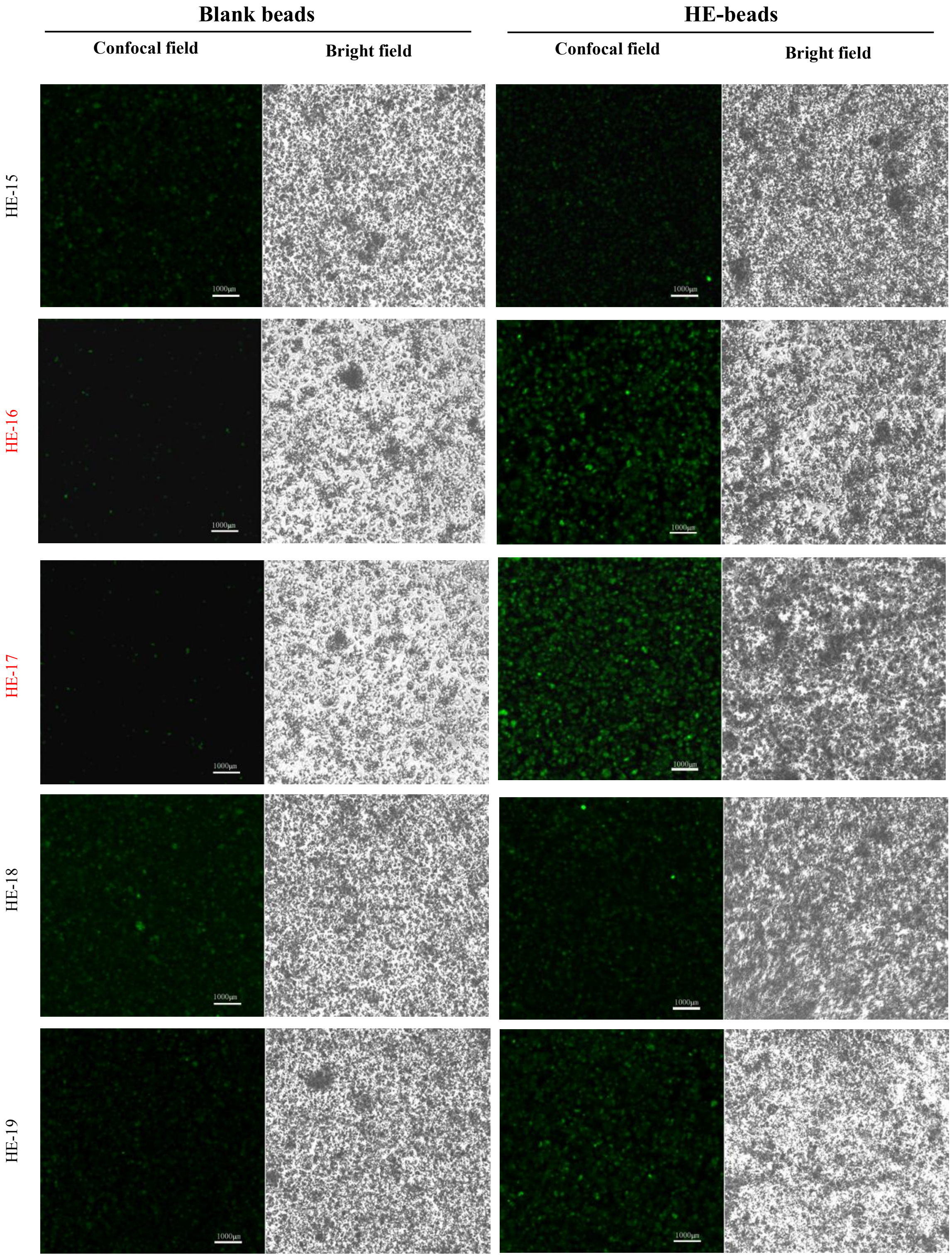
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HE-16
HE-17
HE-18
HE-19
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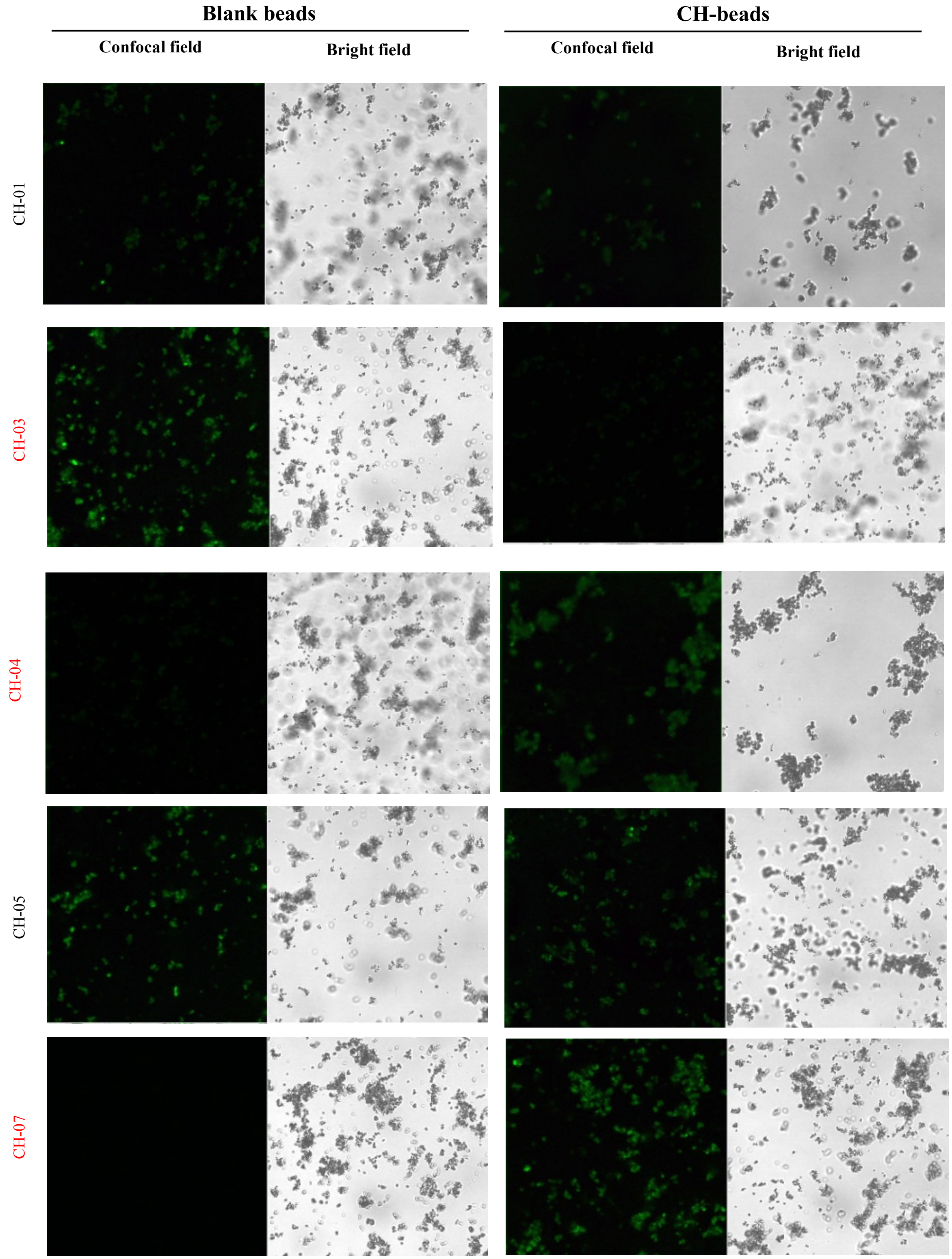
Figure S4. Screening of RNA-HE GAG interaction. Confocal microscopy imaging of the candidate HE aptamers interacting with either the blank beads or the HE-beads. The HE candidate RNA aptamers in red show positive interaction with HE-beads in comparison with the interaction with the blank beads. “Confocal field” indicates the fluorescent channel image of the confocal microscopy while “Bright field” indicates the optical channel image.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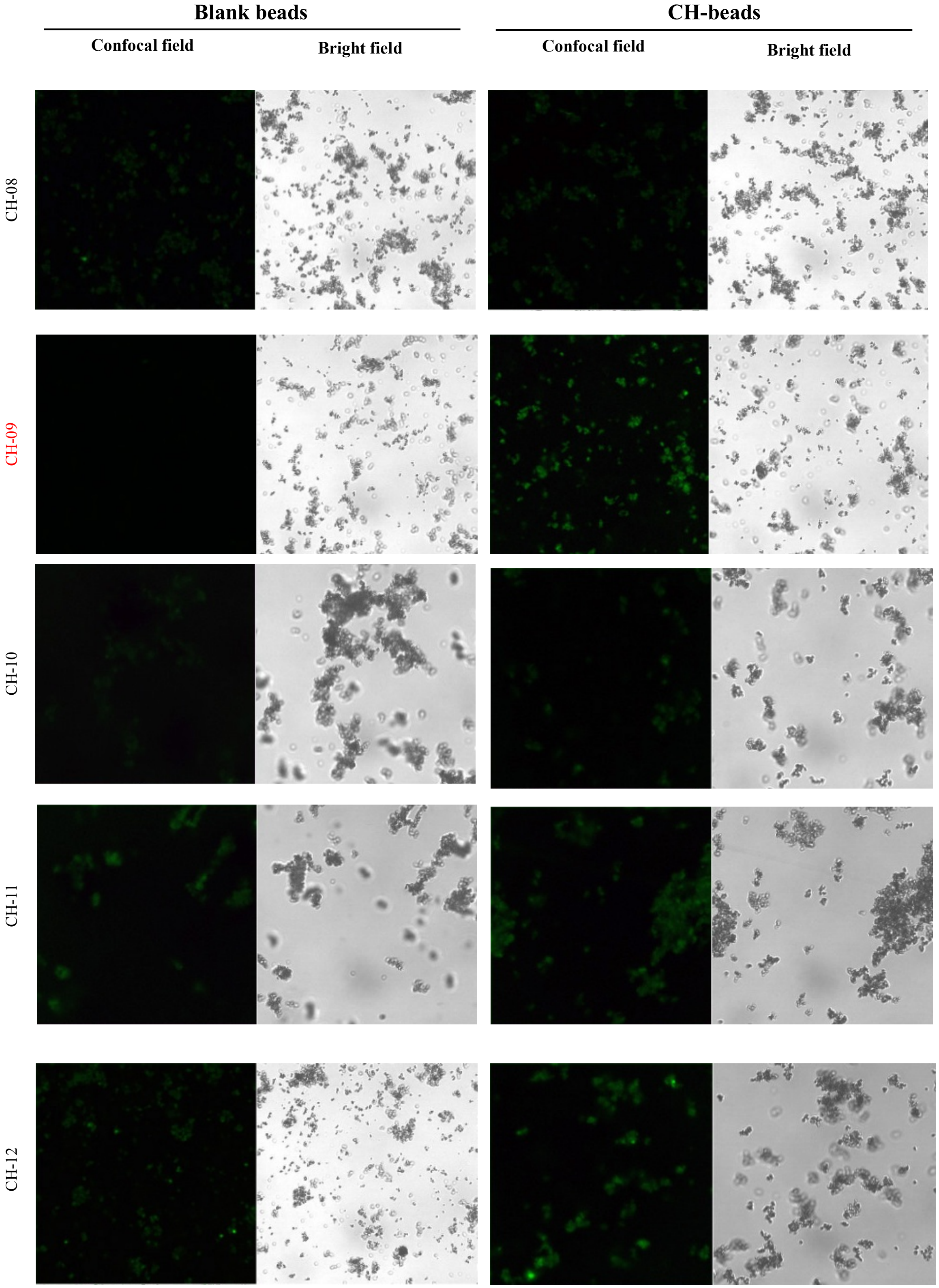
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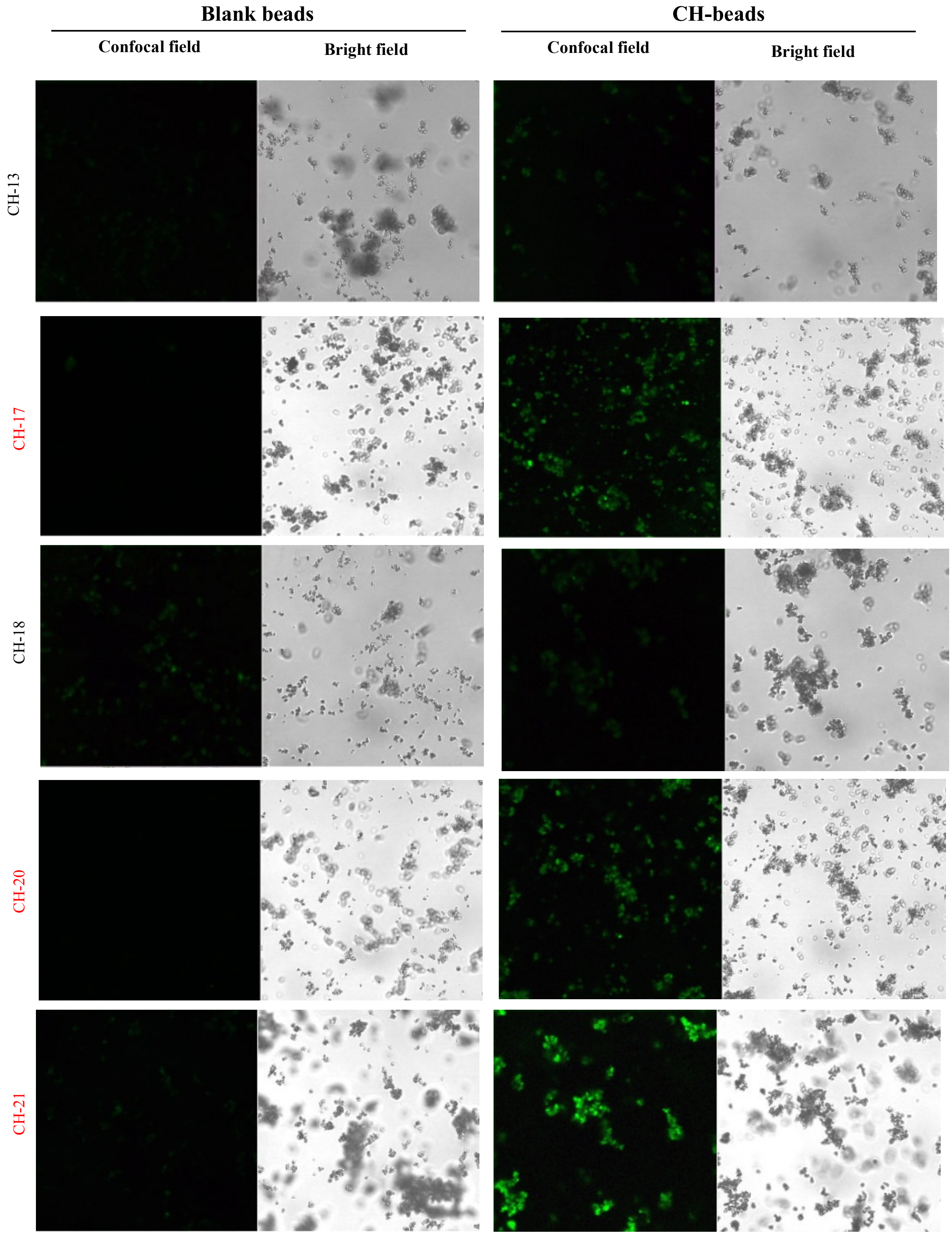
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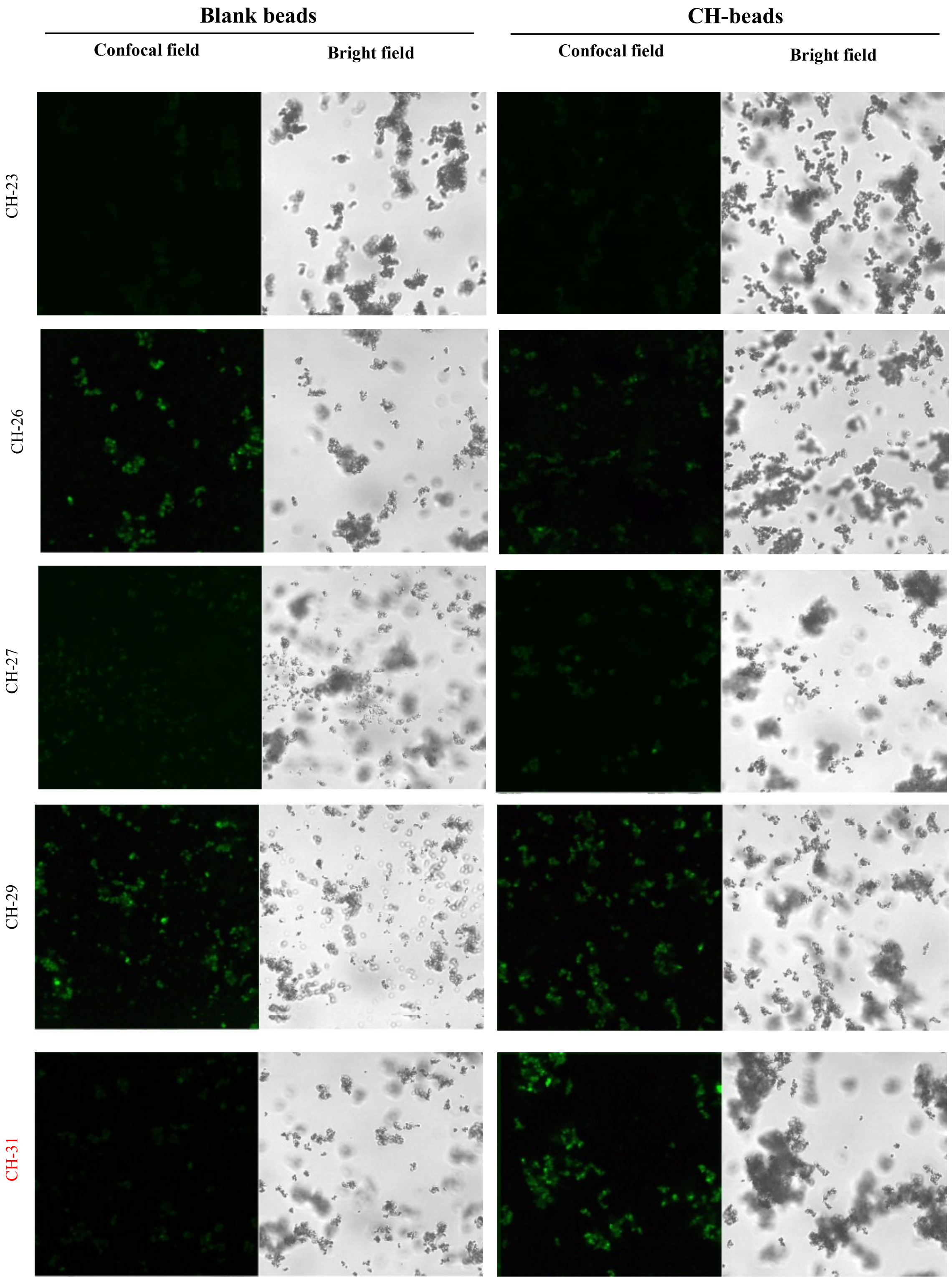
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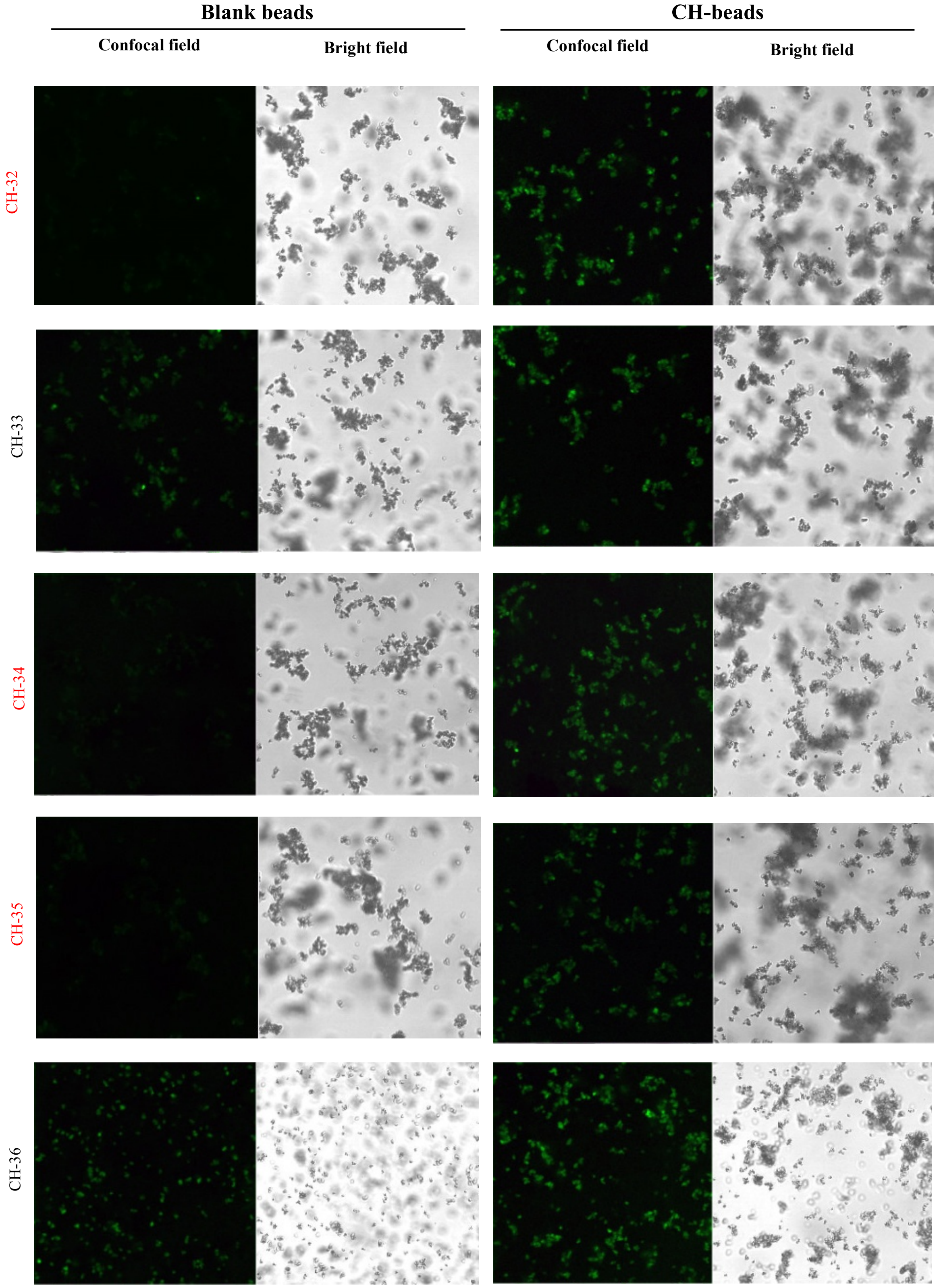
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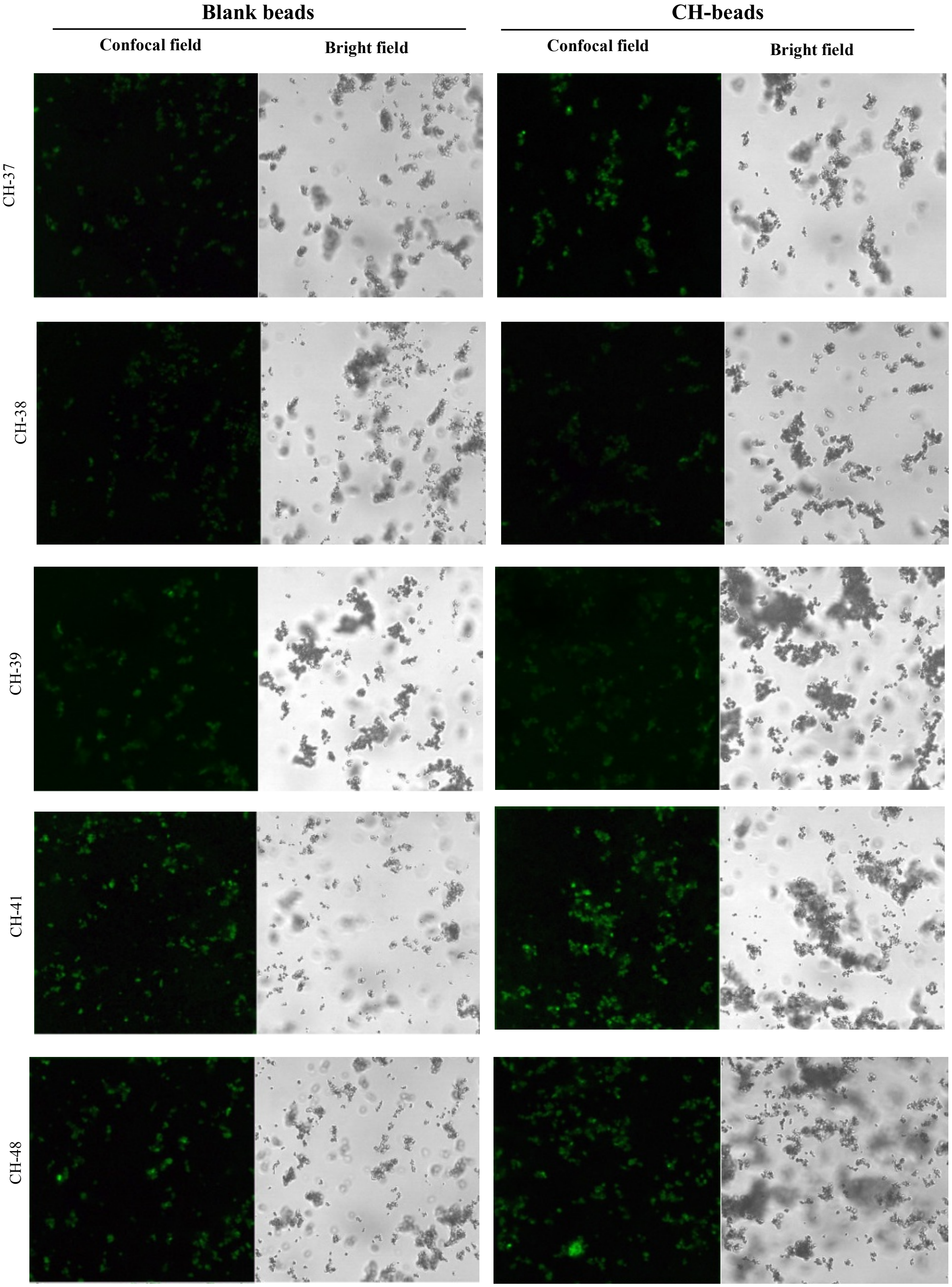
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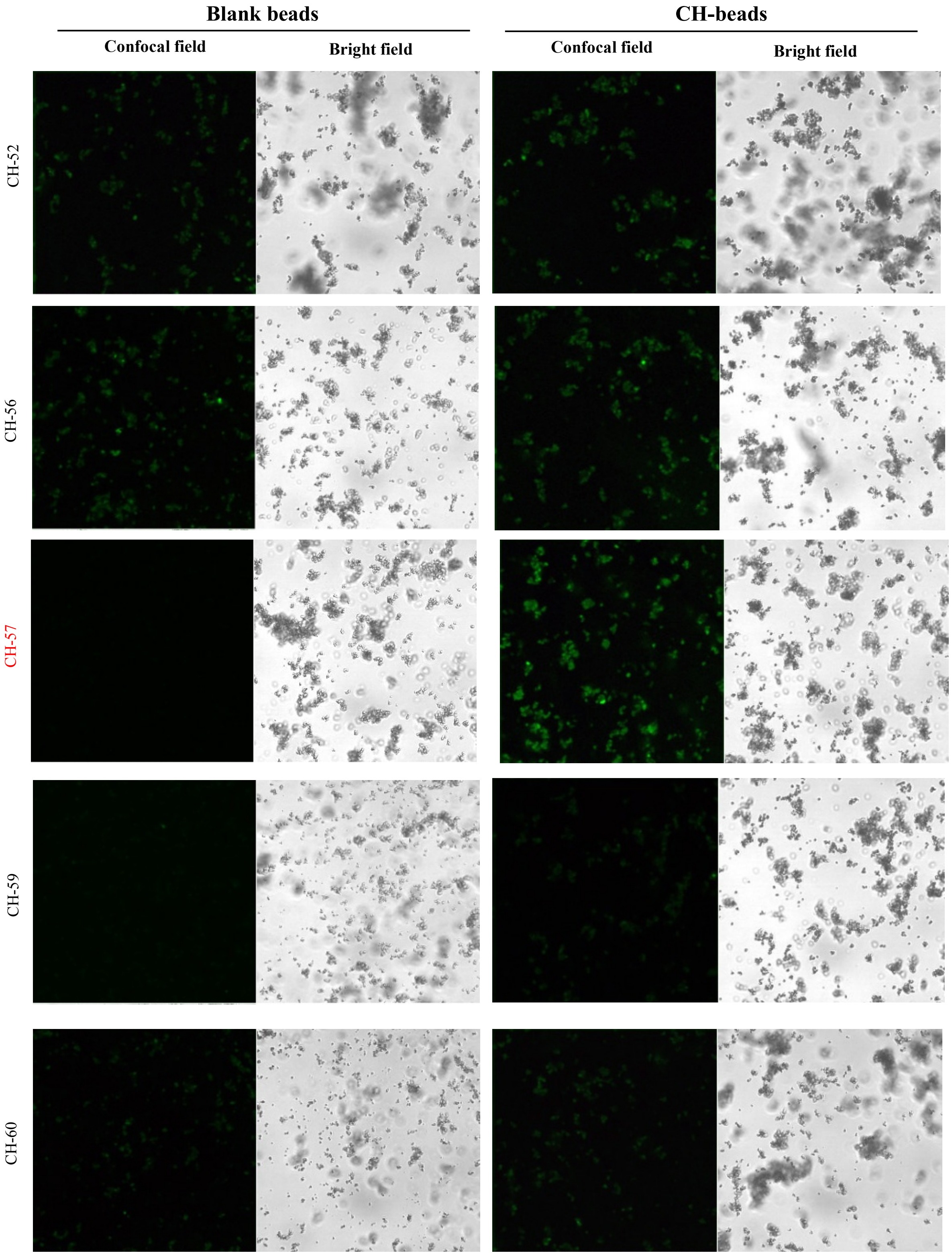
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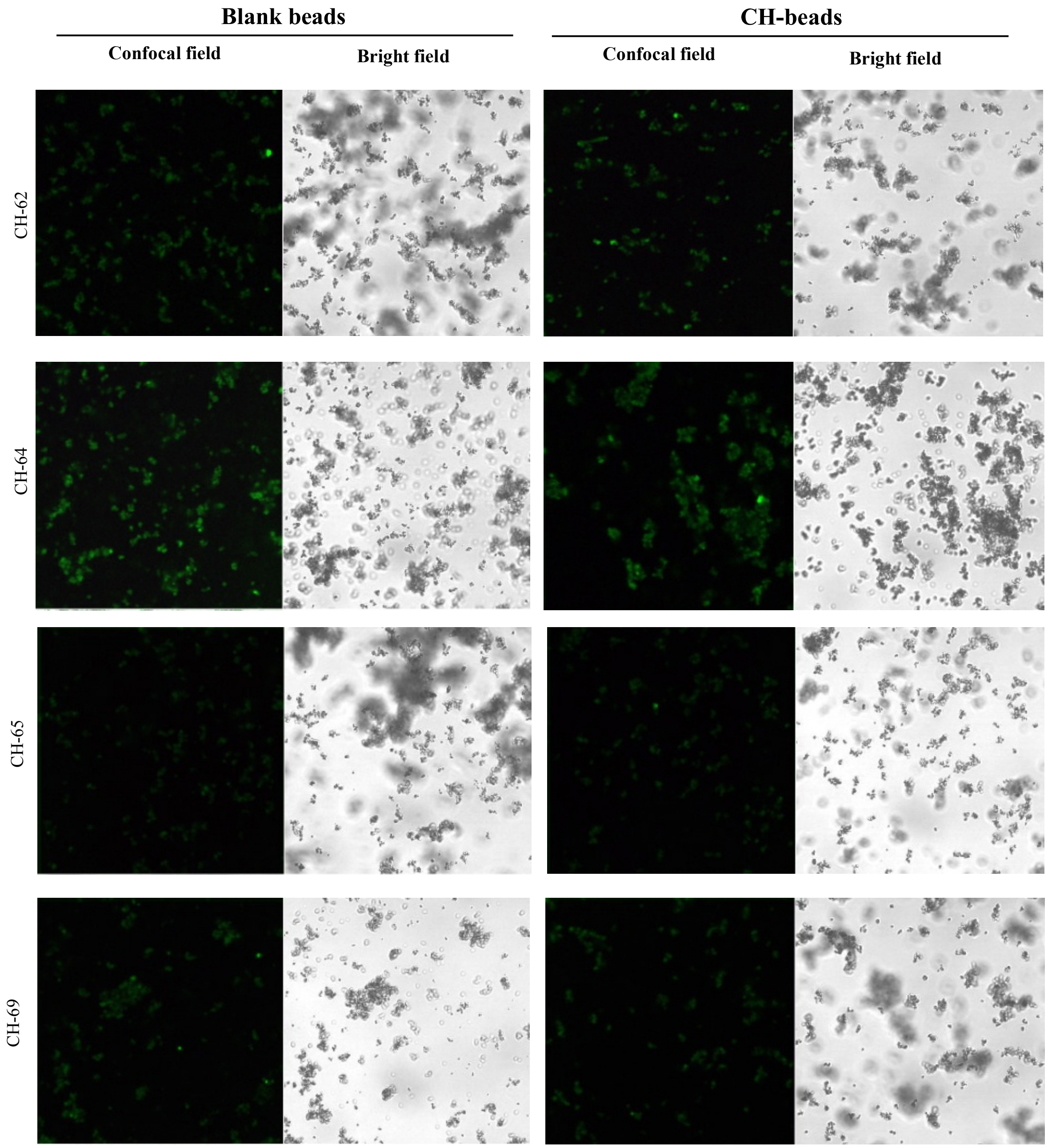
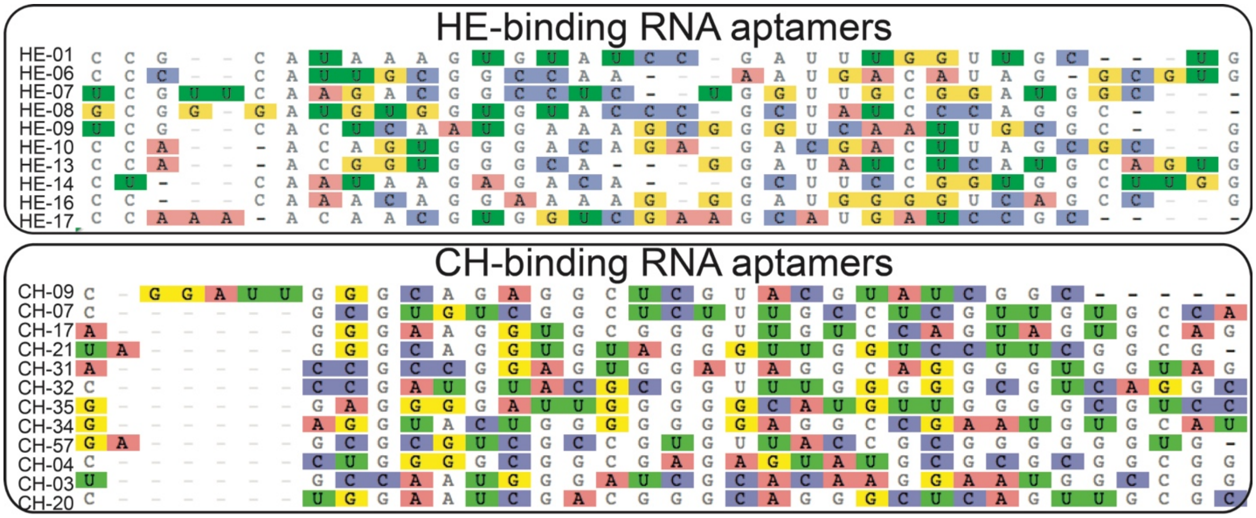


Figure S5. Screening of RNA-CH GAG screening. Confocal microscopy imaging of the candidate CH aptamers interacting with either the blank beads or the CH-beads. The CH candidate RNA aptamers in red show positive interaction with CH-beads in comparison with the interaction with the blank beads. "Confocal field" indicates the fluorescent channel image of the confocal microscopy while "Bright field" indicates the optical channel image.



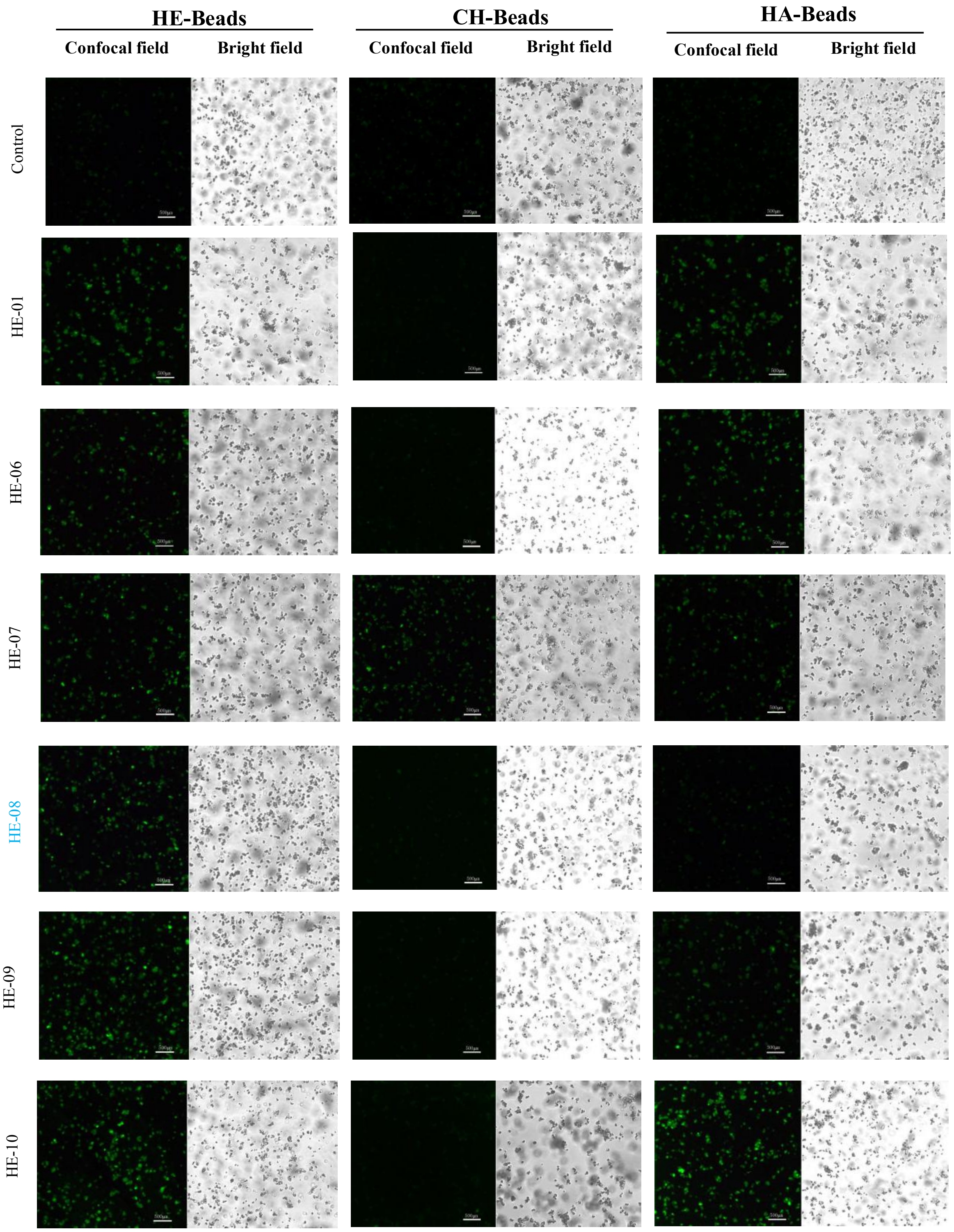
HE-binding RNA aptamers

HE-01	C	C	G	-	-	C	A	U	A	A	A	G	U	G	U	A	U	C	C	-	G	A	U	U	U	G	G	U	U	G	C	-	-	-	U	G
HE-06	C	C	C	-	-	C	A	U	U	G	C	G	G	C	C	A	A	-	-	-	A	A	U	G	A	C	A	U	A	G	-	G	C	G	U	G
HE-07	U	C	G	U	U	C	A	A	G	A	C	G	G	C	C	U	C	-	-	U	G	G	U	U	G	C	G	G	A	U	G	G	C	-	-	-
HE-08	G	C	G	G	-	G	A	U	G	U	G	G	U	G	U	A	C	C	C	-	G	C	U	A	U	C	C	C	A	G	G	C	-	-	-	-
HE-09	U	C	G	-	-	C	A	C	U	C	A	A	U	G	A	A	A	G	C	G	G	G	U	C	A	A	U	U	G	C	G	C	-	-	-	G
HE-10	C	C	A	-	-	-	A	C	A	G	U	G	G	G	A	C	A	G	A	-	G	A	C	G	A	C	U	U	A	G	C	G	C	-	-	G
HE-13	C	C	A	-	-	-	A	C	G	G	U	G	G	G	C	A	-	-	-	G	G	A	U	A	U	C	U	C	A	U	G	C	A	G	U	G
HE-14	C	U	-	-	-	C	A	A	U	A	A	G	A	G	A	C	A	-	-	-	G	C	U	U	C	C	G	G	U	G	G	C	U	U	G	G
HE-16	C	C	-	-	-	C	A	A	A	C	A	G	G	A	A	A	A	G	-	G	G	A	U	G	G	G	G	U	C	A	G	C	C	-	-	G
HE-17	C	C	A	A	A	-	A	C	A	A	C	G	U	G	G	U	C	G	A	A	G	C	A	U	G	A	U	C	C	G	C	-	-	-	-	-

CH-binding RNA aptamers

CH-09	C	-	G	G	A	U	U	G	G	G	C	A	G	A	G	G	C	U	C	G	U	A	C	G	U	A	U	C	G	G	C	-	-	-	-	-
CH-07	C	-	-	-	-	-	-	G	C	G	U	G	U	C	G	G	C	U	C	U	U	U	G	C	C	U	C	G	U	U	G	U	G	C	C	A
CH-17	A	-	-	-	-	-	-	G	G	G	A	A	G	G	U	G	C	G	G	G	U	U	G	U	C	C	A	G	U	A	G	U	G	C	A	G
CH-21	U	A	-	-	-	-	-	G	G	G	C	A	G	G	U	G	U	A	G	G	G	U	U	G	G	U	C	C	U	U	C	G	G	C	G	-
CH-31	A	-	-	-	-	-	-	C	C	G	C	C	G	G	A	G	U	G	G	A	U	A	G	G	C	A	G	G	G	G	U	G	G	U	A	G
CH-32	C	-	-	-	-	-	-	C	C	G	A	U	G	U	A	C	G	C	G	G	U	U	U	G	G	G	G	G	C	G	U	C	A	G	G	C
CH-35	G	-	-	-	-	-	-	G	A	G	G	G	G	A	U	U	G	G	G	G	G	C	A	U	G	U	U	G	G	G	G	C	G	U	C	C
CH-34	G	-	-	-	-	-	-	A	G	G	U	A	C	U	G	G	G	G	G	G	G	A	G	G	C	C	G	A	A	U	G	U	G	C	A	U
CH-57	G	A	-	-	-	-	-	G	C	G	C	G	U	C	G	C	C	G	U	G	U	U	A	C	C	G	C	G	G	G	G	G	G	U	G	-
CH-04	C	-	-	-	-	-	-	C	U	G	G	G	G	C	G	G	C	G	A	G	A	G	U	A	U	G	C	G	C	G	C	G	G	C	G	G
CH-03	U	-	-	-	-	-	-	G	C	C	A	A	U	G	G	G	A	U	C	G	C	A	C	A	A	G	G	A	A	U	G	G	C	C	G	G
CH-20	C	-	-	-	-	-	-	U	G	G	A	A	U	C	G	A	C	G	G	G	C	A	G	G	G	C	U	C	A	G	U	U	G	C	G	C

Figure S6. Sequence alignment of HE- and CH-binding RNA aptamers. Color-highlighted bases indicate the RNA regions showing no consensus sequence motif among the aptamers within each group. Dash lines indicate the artificial gaps generated by the sequence alignment algorithm to maximize the matched sequence alignment.


HE-Beads
CH-Beads
HA-Beads
Confocal field
Bright field
Confocal field
Bright field
Confocal field
Bright field
Control
HE-01
HE-06
HE-07
HE-08
HE-09
HE-10

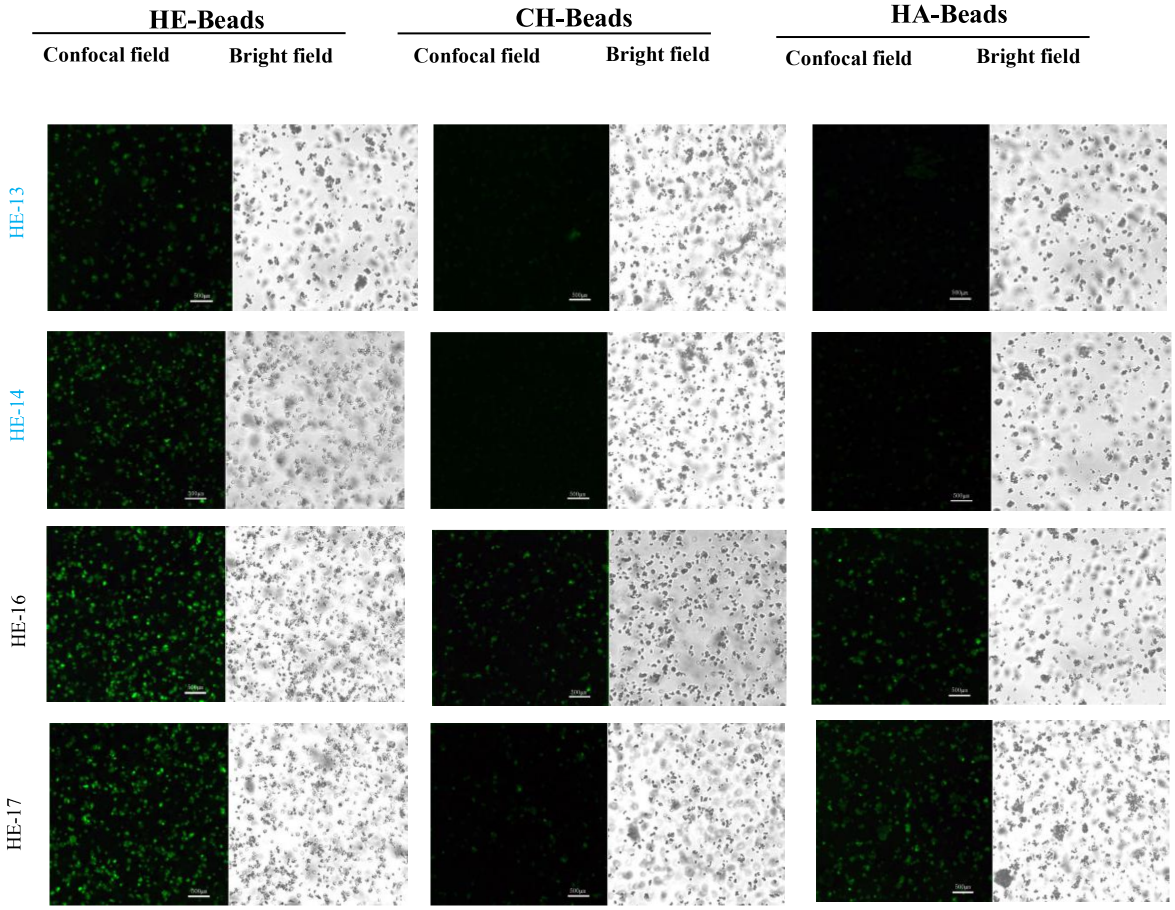
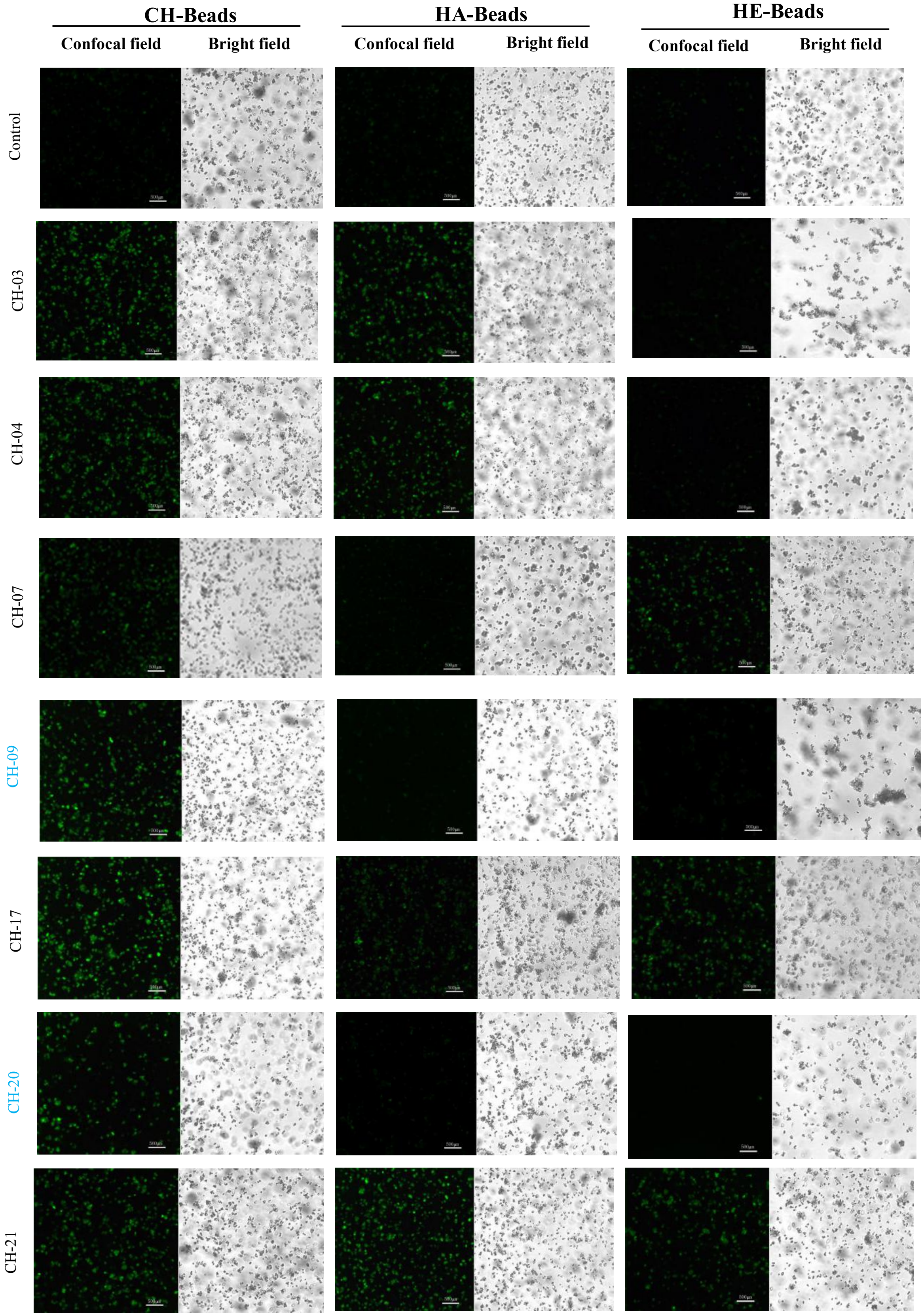


Figure S7. HE-aptamer selectivity screening. Confocal microscopy imaging of the candidate HE aptamers interacting with HE-, CH- and HA-beads. The candidate RNA aptamers in blue show high binding specificity HE in comparison with the interaction with the other two GAGs, CH and HA. “Confocal field” indicates the fluorescent channel image of the confocal microscopy while “Bright field” indicates the optical channel image.


CH-Beads
HA-Beads
HE-Beads
Confocal field
Bright field
Confocal field
Bright field
Confocal field
Bright field
Control
CH-03
CH-04
CH-07
CH-09
CH-17
CH-20
CH-21

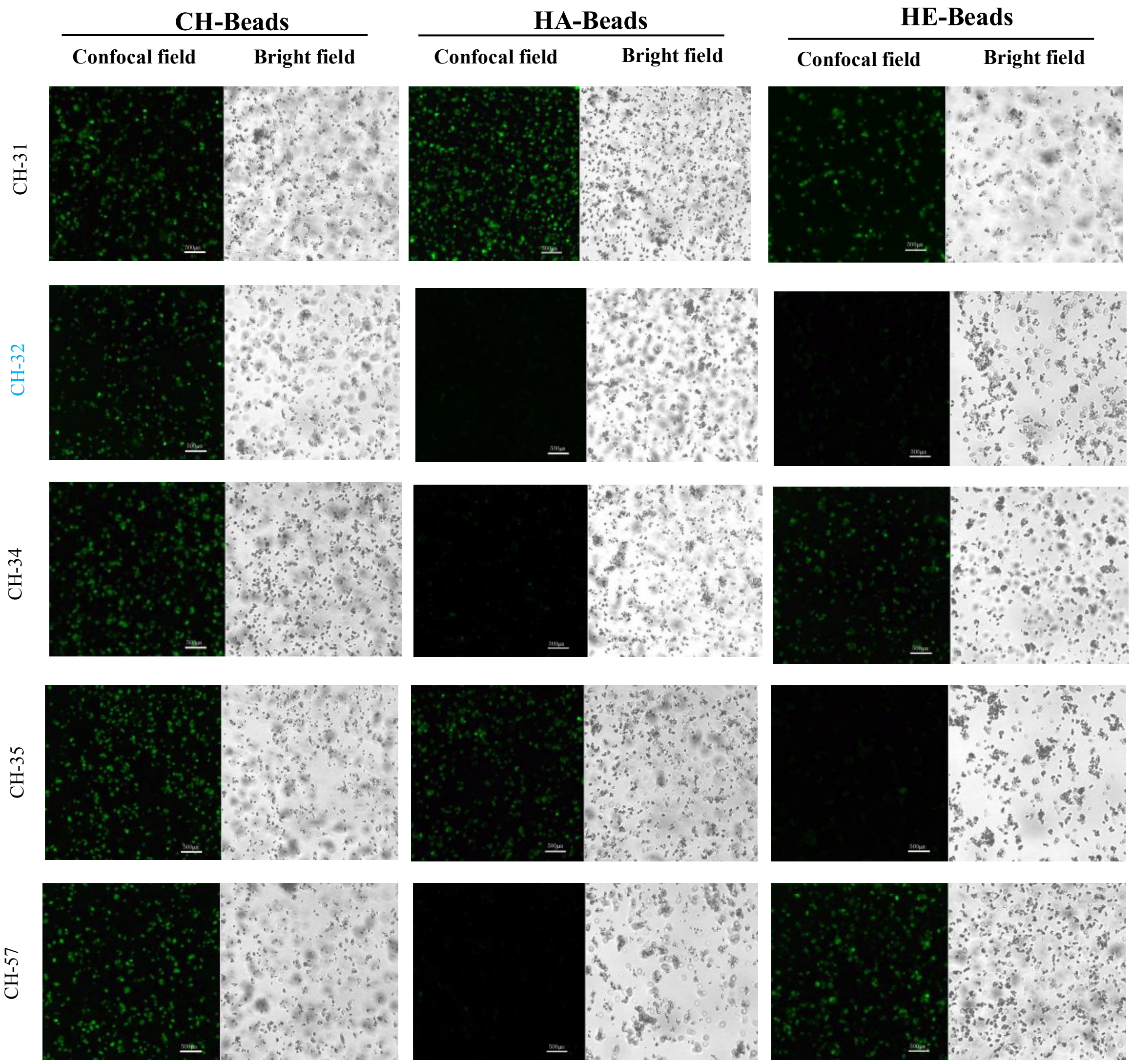


Figure S8. CH-aptamer selectivity screening. Confocal microscopy imaging of the candidate CH aptamers interacting with CH-, HE- and HA-beads. The candidate RNA aptamers in blue show high binding specificity CH in comparison with the interaction with the other two GAGs, HE and HA. “Confocal field” indicates the fluorescent channel image of the confocal microscopy while “Bright field” indicates the optical channel image.

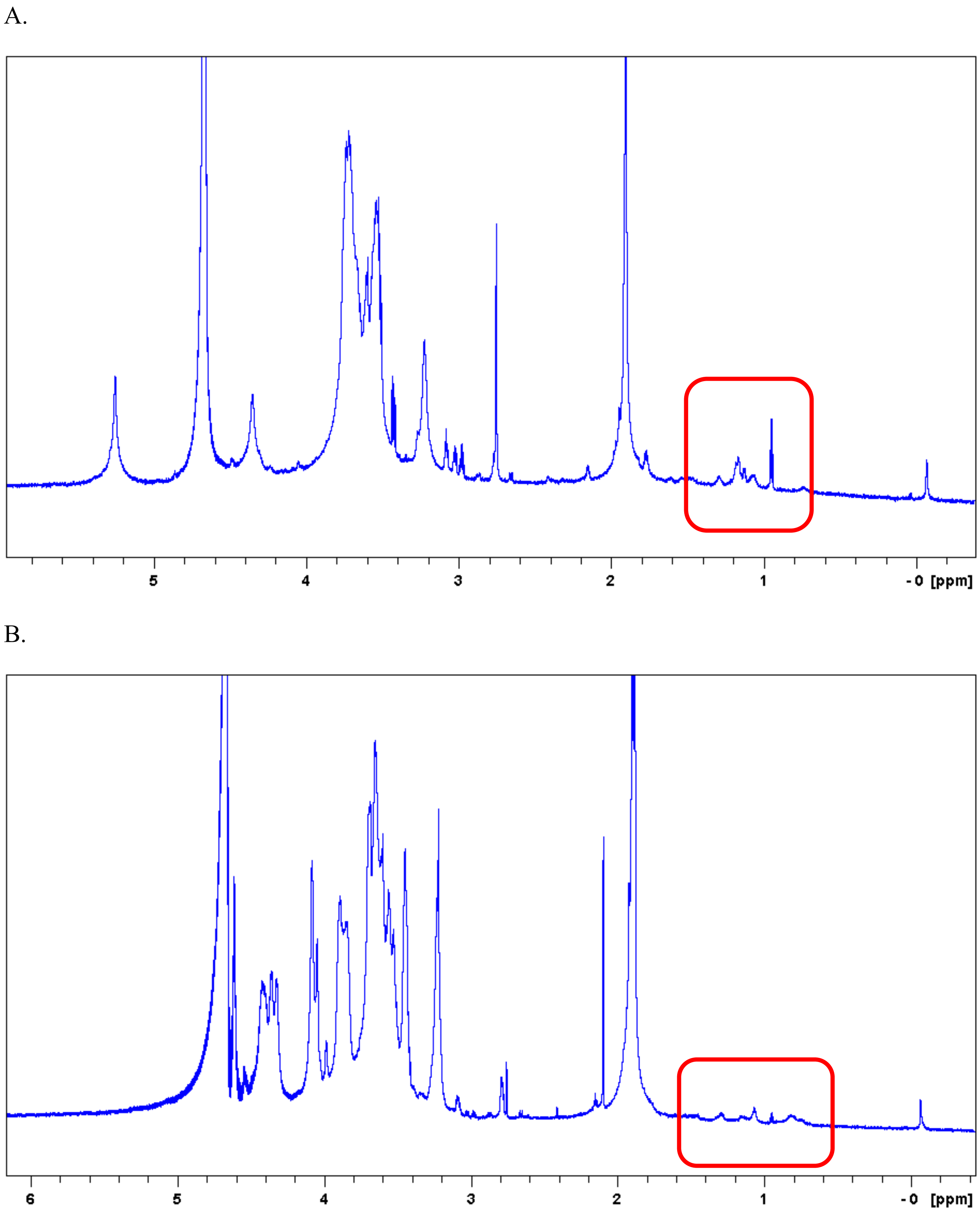


Figure S9. NMR Spectra of biotin-heparosan (A) and biotin-chondroitin (B). Biotin modification signal is highlighted by red boxes.

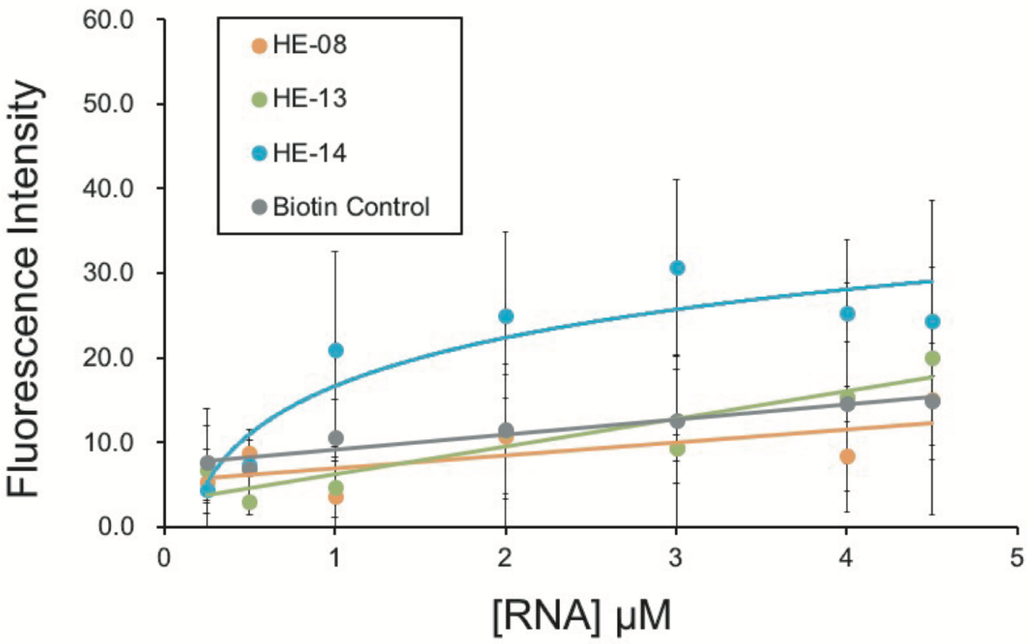


Figure S10. Screening of specific HE aptamer candidates against heparin. HE-08 and HE-13 show fluorescence levels close to the biotin used as a background control, while HE-14 shows slight binding to heparin.