Modulation of Complement Activation and Amplification on Nanoparticle Surfaces by Glycopolymer Conformation and Chemistry

*Kai Yu,[†] Benjamin F.L. Lai,[†] Jonathan H. Foley,[§] Michael J. Krisinger,[§,#] Edward M. Conway[§] and Jayachandran N. Kizhakkedathu[†,‡,]**

Supporting Information

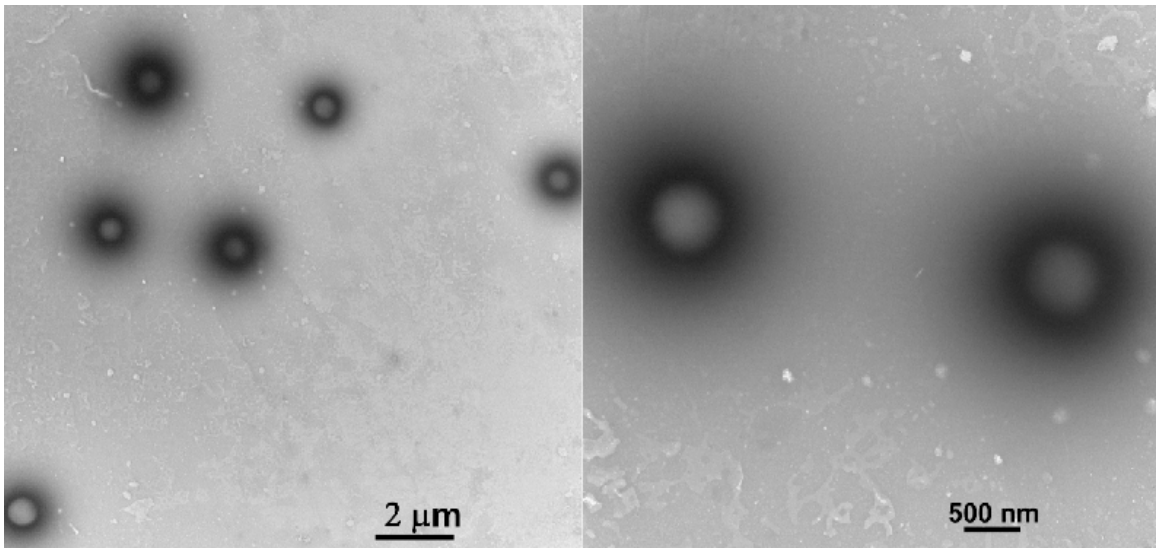

Figure S1. TEM images of nanoparticles grafted with glucose-containing polymer, stained by 2% phosphotungstic acid, which subjected to hydrolysis to produce glucose particles with different grafting density (Glc 1 to Glc 6)

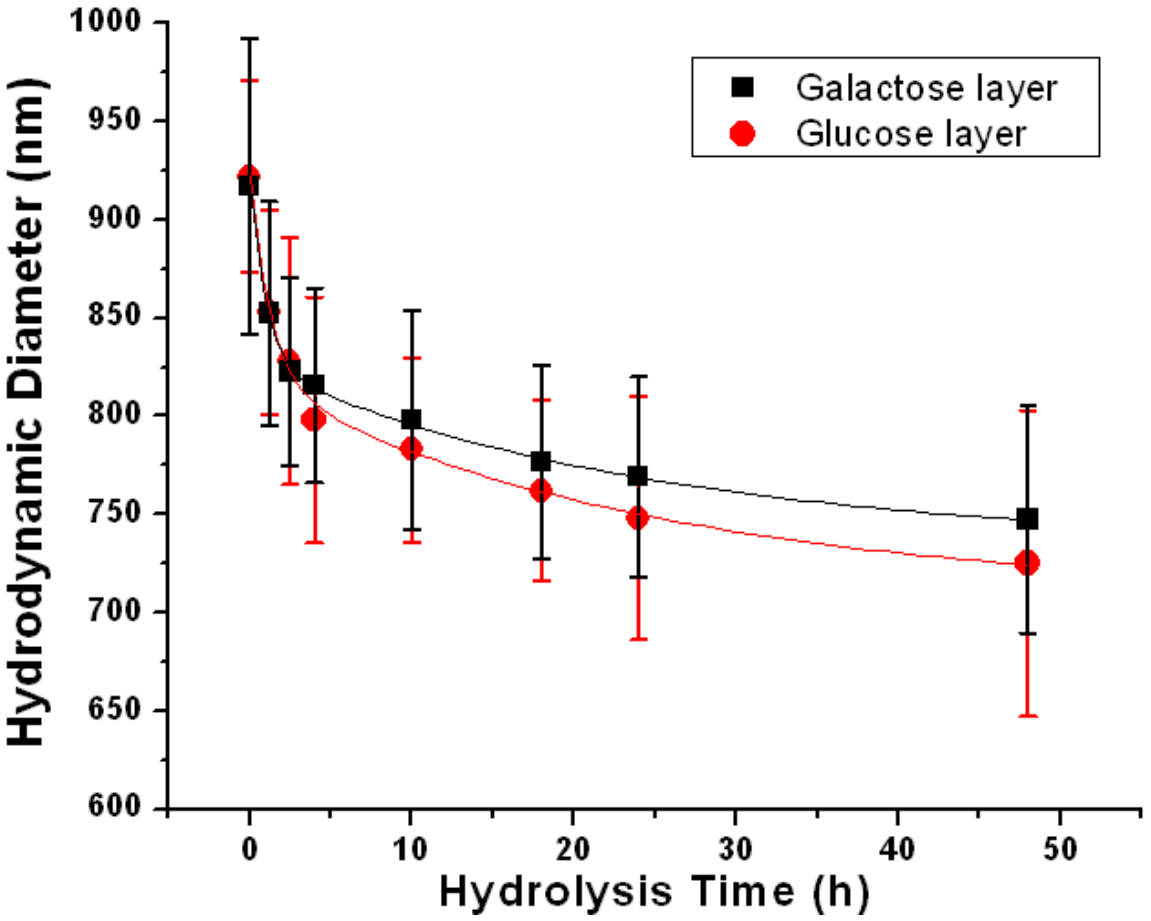

Figure S2. Dependence of hydrodynamic diameter of glycopolymer grafted nanoparticles on hydrolysis time.

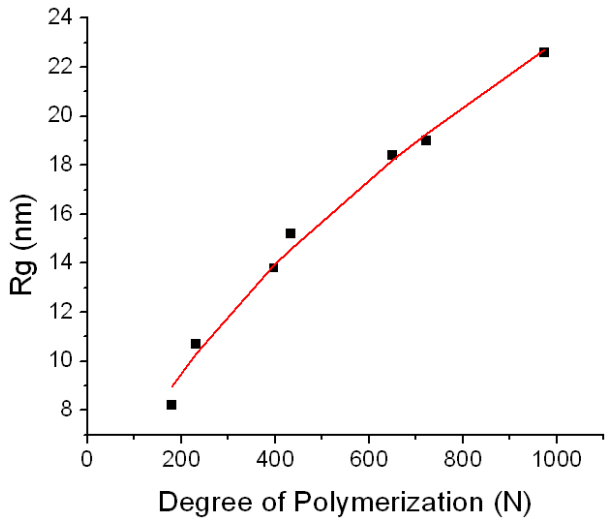

Figure S3. Dependence of radius of gyration of glycopolymer chain on degree of polymerization. The data was fitted using $Rg = aN^{v}$, where the v is derived as the efficient Flory exponent. Glycopolymer (containing galactose residues) having different degree of polymerization (N) was synthesized by ATRP with changing the initiator concentration. The obtained value of efficient Flory exponent is 0.55 ± 0.03, sitting between 0.5 (corresponding to polymer chain in θ solvent) and 0.6 (corresponding to linear polymer chain in good solvent). The result indicates that the polymer is swollen in PBS buffer (0.11 M), but not fully extended.

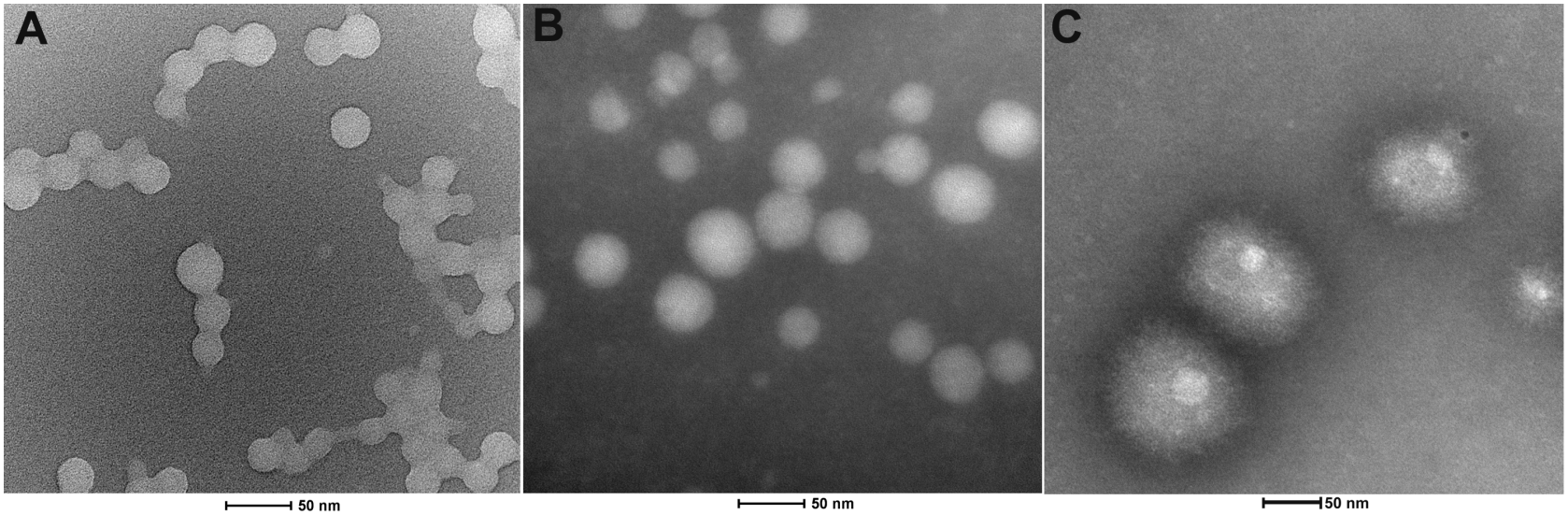

Figure S4. TEM images of initiator modified PS nanoparticles stained by 2% phosphotungstic acid (33.3 ± 6.7 nm) (A), nanoparticles grafted with galactose-containing polymer (38.5 ± 7.3 nm) with a grafting density of 0.026 chains/nm^2 (B), nanoparticles grafted with galactose-containing polymer (97.4 ± 26 nm) with a grafting density of 0.17 chains/nm^2 (C).

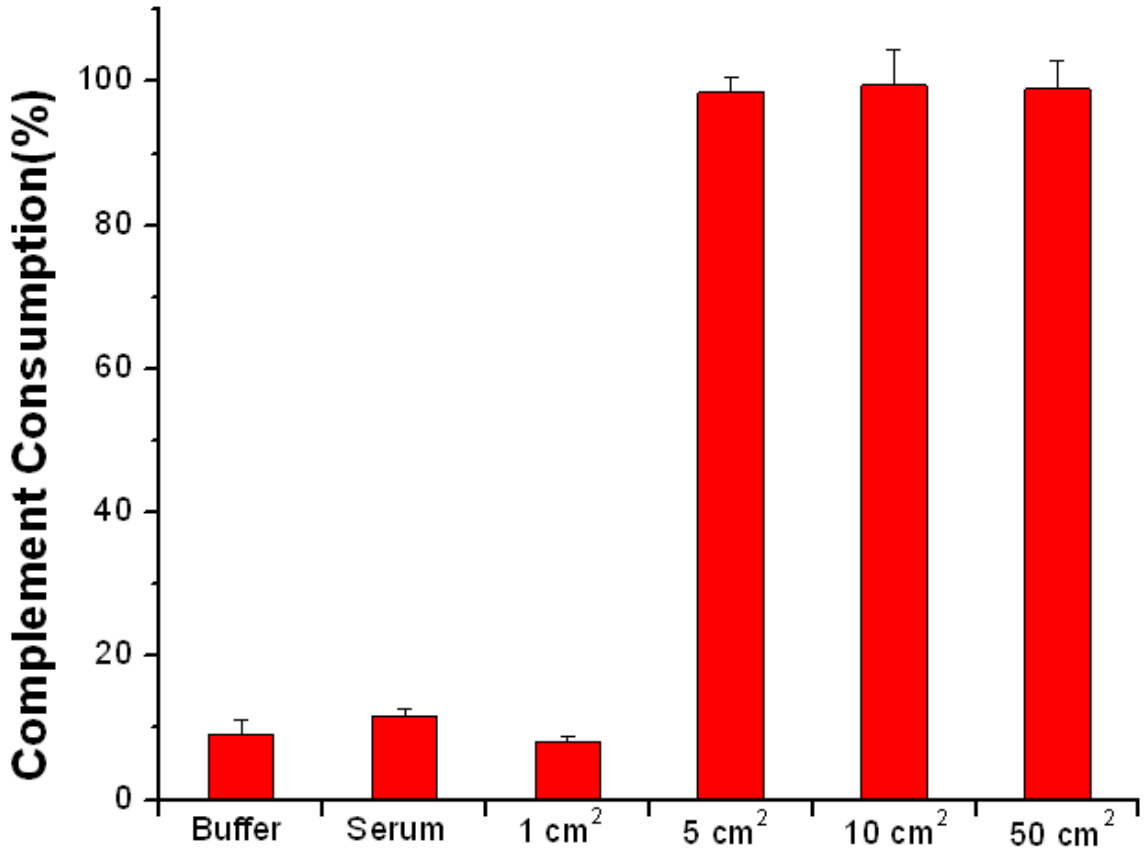

Figure S5. Effect of surface area of glycopolymer grafted nanoparticles on complement consumption. Nanoparticles grafted with glucose-containing polymer at a grafting density 0.083 chains/nm^2 were used for this study. Diluted fresh human serum (20%, 180 μL) was incubated with NP suspensions (different surface areas, on 669 nm particles) for 1 h at 37°C and the residual total complement activity of the exposed serum was measured using the sheep erythrocyte based hemolytic assay. Each data point represents an average ± SD (n = 3) of three experiments (3 donors) in fresh serum.

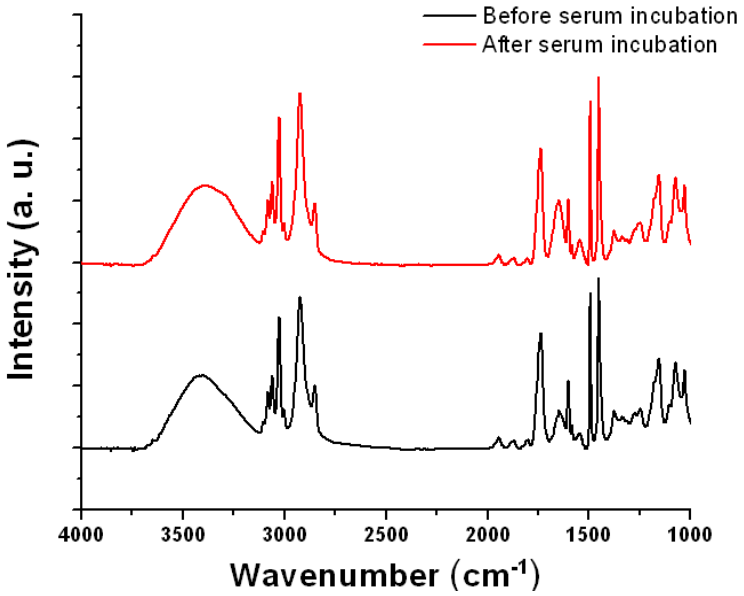

Figure S6. IR spectra of nanoparticles grafted with galactose-containing polymer (Galactose NP1) before and after incubation with serum (both spectra were normalized by the carbonyl peak at 1720 cm^{-1} from the bare particles). Fresh human serum (180 μL) was incubated with 20 μL of NP (Galactoe NP1) suspensions (50 cm^2, on 669 nm particles) for 1 h at 37°C. Then the particles were washed by PBS buffer solution twice (200 uL each), boiled at 90 °C with 2% SDS (100 uL) for 10 minutes, ultrasonicated in buffer for 10 min, finally washed with water and dried. Except the increase in the intensity at 1650 cm^{-1}, correspond to the C=O stretching vibration of the amide group due to the chemically fixation of complement C3,[1] there is no noticeable change of IR intensity. The consistent IR band intensity from 3600 to 3200 cm^{-1}, which is due to O–H stretch in the glycopolymer, imply that there is no hydrolysis during incubation.

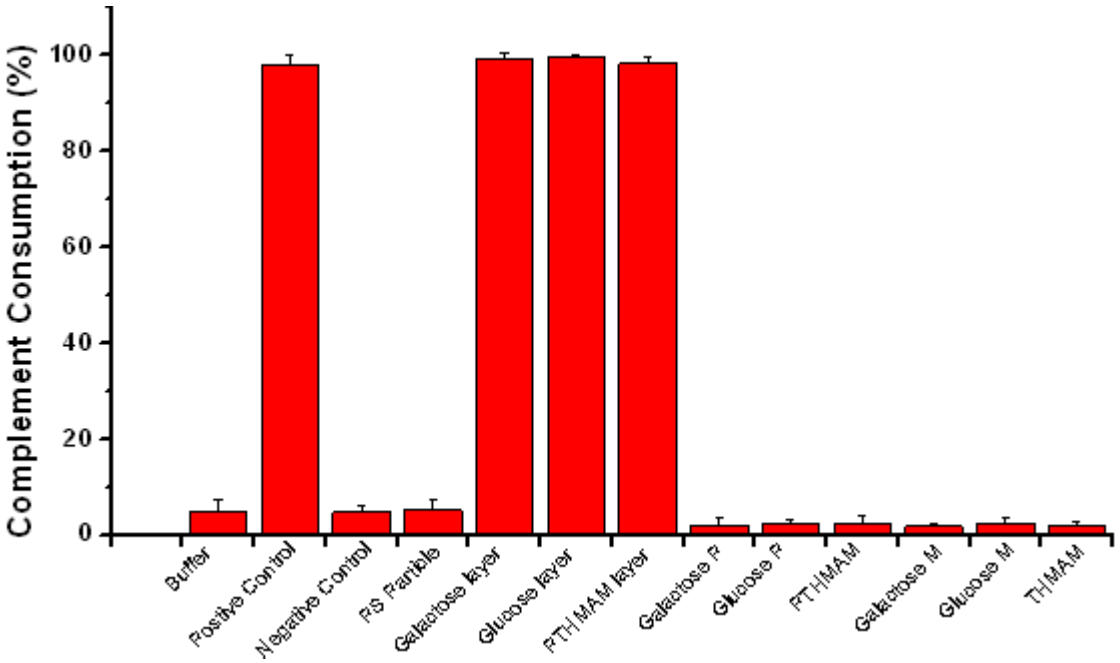

Figure S7. Complement consumption induced by bare PS particles, particles grafted glycopolymer (galactose layer, glucose layer) and control polymer (PTHMAM layer), soluble glycopolymer (Galactose P and glucose P) and PTHMAM, carbohydrate monomer (galactose M and glucose M) and control monomer (THMAM). IgG (5mg/mL) as a positive control. GVB-EDTA (1 mM) as a negative control. Diluted fresh human serum (20%, 180 µL) was incubated with 20 µL of NP suspensions (50 cm^2, on 669 nm particles) or 20 µL polymer solution (6.1 mg/mL) or 20 µL monomer solution (6.1 mg/mL) for 1 h at 37°C. The residual total complement activity of the exposed serum was measured using the sheep erythrocyte based hemolytic assay. Each data point represents an average ± SD (n = 3) of three experiments (3 donors) in fresh serum.

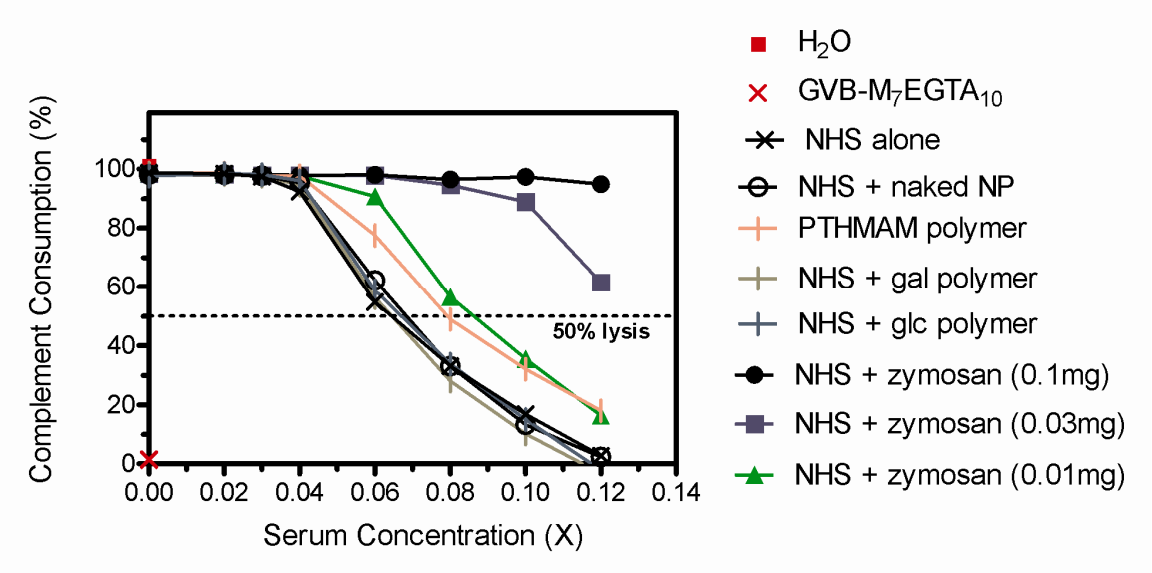

Figure S8. Effect of complement consumption measured by rabbit erythrocyte lysis upon incubation with zymosan, glycopolymer containing galactopyranoside or glucopyranoside and PTHMAM at different dilutions of human serum. Pooled human serum (150 µL) was incubated with zymosan (0.01, 0.03, 0.1 mg) for 1 h at 37°C and the residual complement activity of the exposed serum was measured using the rabbit erythrocyte based hemolytic assay at different serum concentrations. Results are representative of 2 independent experiments, each with similar results.

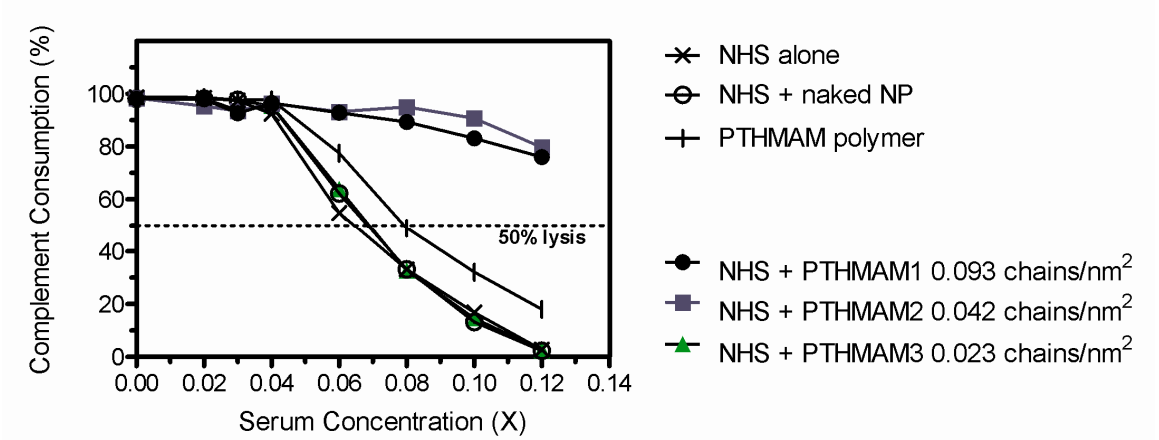

Figure S9. Effect of complement consumption measured by rabbit erythrocyte lysis upon incubation with particles grafted with PTHMAM at different concentrations of human serum. Pooled human serum (150 µL) was incubated with 100 µL of NPs suspension (150 cm^2, on 669 nm particles) for 1 h at 37°C and the residual complement activity of the exposed serum was measured using the rabbit erythrocyte based hemolytic assay at different serum concentrations. Zymosan and GVB-CM (GVB containing 0.15 mM Ca^{2+} and 1 mM Mg^{2+}) were used as positive and negative controls. Results are representative of 2 independent experiments, each with similar results.

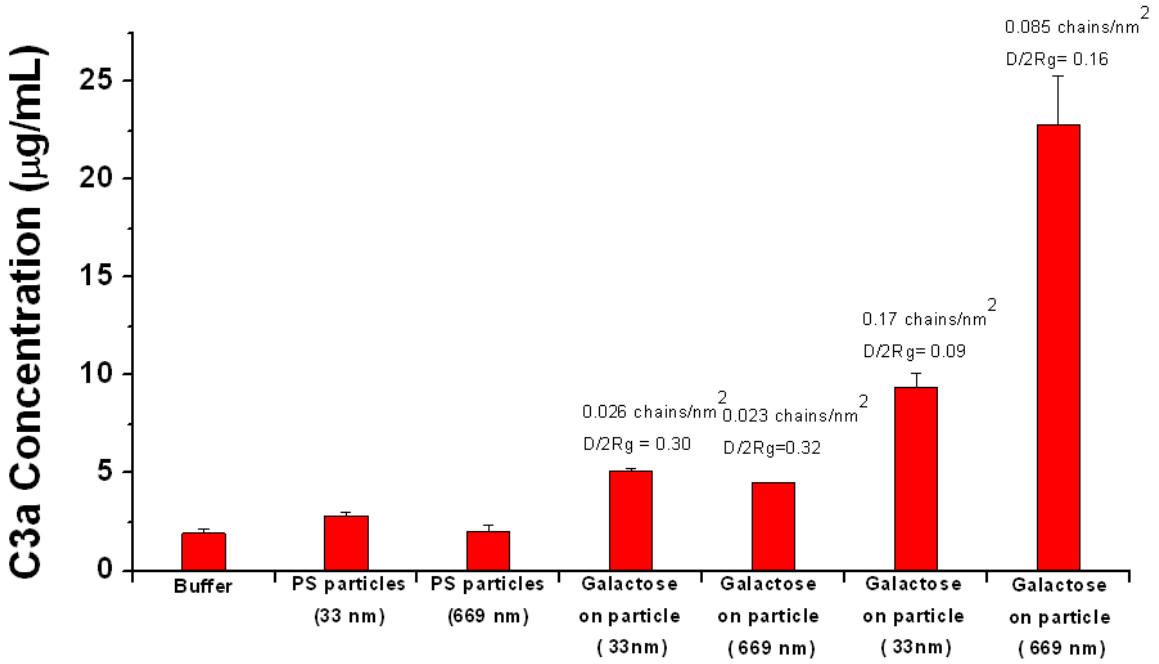

Figure S10. Effect of particle size (surface curvature) on the generation of C3a. Fresh human serum (180 μL) was incubated with 20 μL of NP suspensions (50 cm^2, on 33 and 669 nm particles) for 1 h at 37°C. C3a levels in the serum were quantified by Microvue C3a Plus ELISA. Each data point represents an average ± SD (n = 3) of concentration of complement activation product C3a generated in fresh serum.

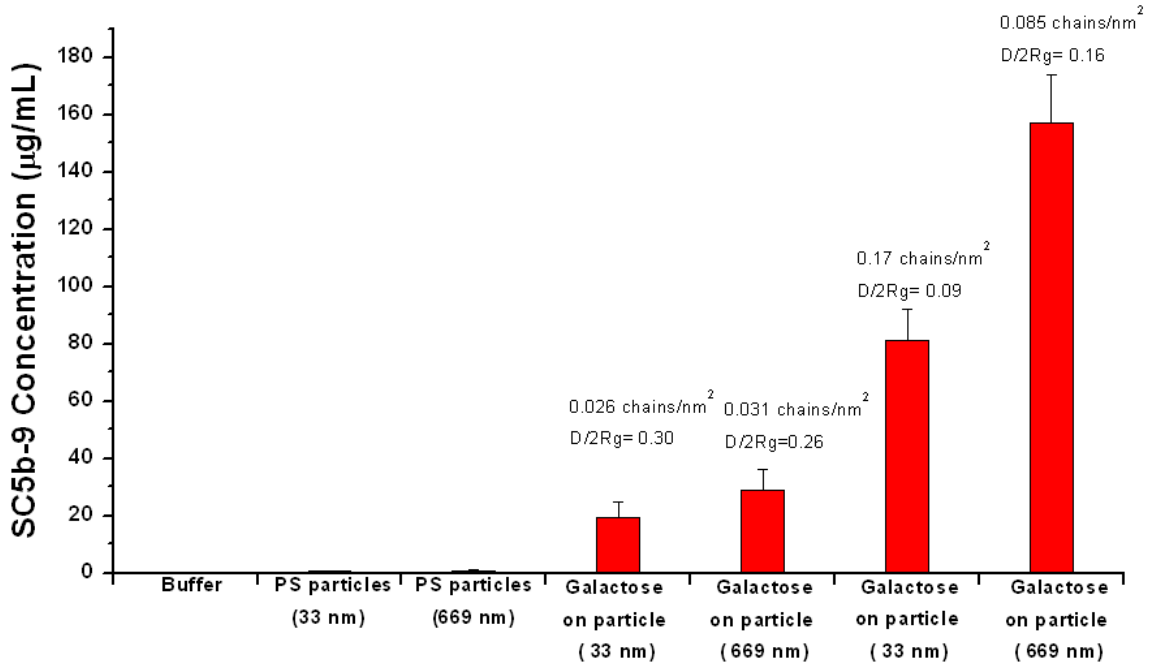

Figure S11. Effect of particle size (surface curvature) on the generation of SC5b-9. Fresh human serum (180 μL) was incubated with 20 μL of NP suspensions (50 cm^2, on 33 and 669 nm particles) for 1 h at 37°C. SC5b-9 levels in the serum were quantified by MicroVue SC5b-9 Plus ELISA. Each data point represents an average ± SD (n = 3) of concentration of complement activation product SC5b-9 generated in fresh serum.

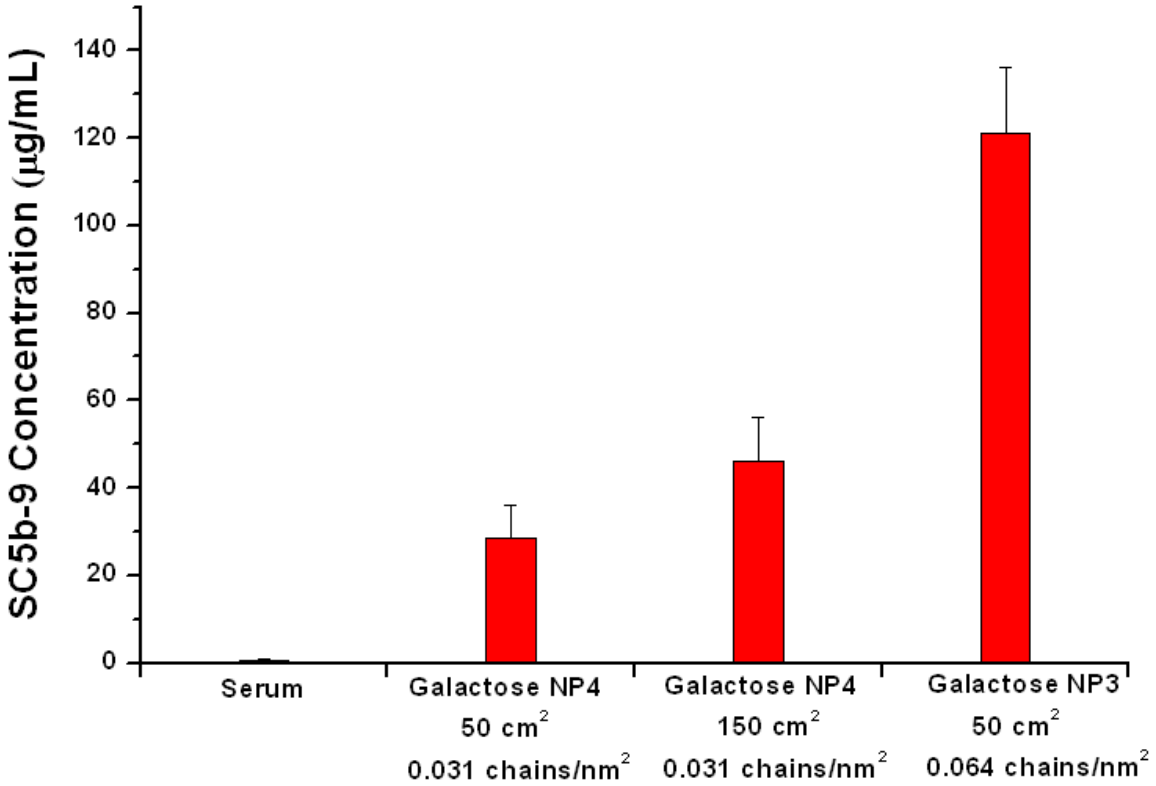

Figure S12. Effect of surface area of glycopolymer grafted nanoparticles on the generation of SC5b-9. Nanoparticles grafted with galactose-containing polymer with grafting densities of 0.031 and 0.064 chains/nm^2 were used for this study. Fresh human serum (180 μL) was incubated with 20 μL of NP suspensions (50 and 150 cm^2, on 669 nm particles) for 1 h at 37°C. SC5b-9 levels in the serum were quantified by MicroVue SC5b-9 Plus ELISA. Each data point represents an average ± SD (n = 3) of concentration of complement activation product SC5b-9 generated in fresh serum.

Table S1. Characteristics of PTHMAM brush grafted NPs

	PTHMAM NP1	**PTHMAM NP2**	**PTHMAM NP3**
Hydrodynamic size	804 ± 55 nm	769 ± 67 nm	749.2 ± 43 nm
Grafting density (chains/nm^2)	0.093	0.042	0.023
D/2Rg	0.16	0.24	0.33

The molecular weight of the grafted PTHMAM was 72000 (PDI =1.3). The error estimate for the grafting density calculation is less than 20% of the mean value.

Table S2. List of proteins detected in the corona on the Galactose-containing glycopolymer layer with grafting density 0.064 chains/nm^2

Accession number	Protein	emPAI	Abundance (%)
P01024	Complement C3	16.25	19.20
P02647	Apolipoprotein A-I	7.64	9.03
P02649	Apolipoprotein E	5.26	6.21
P02768	Serum albumin	4.06	4.80
P02654	Apolipoprotein C-I	3.89	4.60
P02748	Complement component C9	3.22	3.80
P0DJI8	Serum amyloid A-1 protein	2.85	3.37
P06727	Apolipoprotein A-IV	2.54	3.00
B0YIW2	Apolipoprotein C-III	2.25	2.66
P02652	Apolipoprotein A-II	1.89	2.23
P01031	Complement C5	1.69	2.00
P01857	Ig gamma-1 chain C region	1.59	1.88
P10909-2	Isoform 2 of Clusterin	1.4	1.65
P04264	Keratin, type II cytoskeletal 1	1.28	1.51
P04004	Vitronectin	1.25	1.48
P01834	Ig kappa chain C region	1.16	1.37
P01625	Ig kappa chain V-IV region Len	1.04	1.23
P01764	Ig heavy chain V-III region VH26	1.04	1.23
P07360	Complement component C8 gamma chain	1.01	1.19
P13671	Complement component C6	0.98	1.16
P01766	Ig heavy chain V-III region BRO	0.98	1.16
P13645	Keratin, type I cytoskeletal 10	0.92	1.09
P00734	Prothrombin	0.88	1.04
F8W6P5	LVV-hemorphin-7	0.85	1.00
P01859	Ig gamma-2 chain C region	0.84	0.99
P02766	Transthyretin	0.78	0.92
P10643	Complement component C7	0.7	0.83
P0CG05	Ig lambda-2 chain C regions	0.69	0.82
F5GY80	Complement component C8 beta chain	0.68	0.80
Q96IY4	Carboxypeptidase B2	0.68	0.80
G3V1N2	HCG1745306, isoform CRA_a	0.66	0.78
P01593	Ig kappa chain V-I region	0.65	0.77
Q0VAC5	HP protein	0.63	0.74
P35527	Keratin, type I cytoskeletal 9	0.59	0.70
P01860	Ig gamma-3 chain C region	0.57	0.67
P01871-2	Isoform 2 of Ig mu chain C region	0.53	0.63
P35542	Serum amyloid A-4 protein	0.51	0.60
G3V3A0	Alpha-1-antichymotrypsin	0.49	0.58
I3L1H9	Zymogen granule protein 16 homolog B	0.48	0.57
P02790	Hemopexin	0.44	0.52
P35908	Keratin, type II cytoskeletal 2 epidermal	0.41	0.48
P07357	Complement component C8 alpha chain	0.4	0.47
Q99969	Retinoic acid receptor responder protein 2	0.39	0.46
P04259	Keratin, type II cytoskeletal 6B	0.38	0.45

P02538	Keratin, type II cytoskeletal 6A	0.38	0.45
P02533	Keratin, type I cytoskeletal 14	0.36	0.43
B1AKG0	Complement factor H-related 1	0.35	0.41
E7EPG1	Multimerin-1	0.33	0.39
P05109	Protein S100-A8	0.32	0.38
P12259	Coagulation factor V	0.31	0.37
P04196	Histidine-rich glycoprotein	0.3	0.35
P02655	Apolipoprotein C-II	0.3	0.35
P13647	Keratin, type II cytoskeletal 5	0.29	0.34
Q13103	Secreted phosphoprotein 24	0.29	0.34
B4E1Z4	Complement factor B	0.28	0.33
J3KN47	Serotransferrin	0.28	0.33
F5H1A8	Gelsolin	0.27	0.32
P18428	Lipopolysaccharide-binding protein	0.27	0.32
P01008	Antithrombin-III	0.27	0.32
P01023	Alpha-2-macroglobulin	0.22	0.26
P07737	Profilin-1	0.22	0.26
P10124	Serglycin	0.19	0.22
C9JV77	Alpha-2-HS-glycoprotein	0.17	0.20
Q9Y251	Heparanase	0.17	0.20
C9JEX1	Kininogen-1	0.15	0.18
Q5VYL6	Complement factor H-related 5	0.15	0.18
P08603	Complement factor H	0.14	0.17
B0YJC4	Vimentin	0.14	0.17
P19652	Alpha-1-acid glycoprotein 2	0.14	0.17
P27918	Properdin	0.13	0.15
P31946-2	Isoform Short of 14-3-3 protein beta/alpha	0.12	0.14
P24593	Insulin-like growth factor-binding protein 5	0.11	0.13
F5H5M6	Hyaluronan-binding protein 2 50 kDa heavy chain	0.11	0.13
Q5T985	Inter-alpha (Globulin) inhibitor H2	0.1	0.12
P10646	Tissue factor pathway inhibitor	0.09	0.11
B0UZ83	Complement C4 gamma chain	0.09	0.11
P02766	Serum paraoxonase/arylesterase 1	0.08	0.09
E7EQ48	Proteoglycan 4	0.06	0.07
P07196	Neurofilament light polypeptide	0.05	0.06
E7EMV2	Neurofilament medium polypeptide	0.04	0.05
P19021-2	Isoform 2 of Peptidyl-glycine alpha-amidating monooxygenase	0.03	0.04
F8W7G7	Anastellin	0.01	0.01

The absolute protein abundance in proteomics was estimated with the Exponentially Modified Protein Abundance Index through HCD fragmentation. Protein abundance (mol %) = emPAI/∑(emPAI) X100. Protein score 50 was taken as the identity threshold as the confidence level is higher than 95%.

Table S3. List of proteins detected in the corona on the galactose-containing glycopolymer layer with grafting density 0.031 chains/nm^2

Accession number	Protein	emPAI	Abundance (%)
P01024	Complement C3	12.11	13.93
P02647	Apolipoprotein A-I	10.75	12.37
P02649	Apolipoprotein E	6.46	7.43
P02654	Apolipoprotein C-I	5.72	6.58
P0DJI8	Serum amyloid A-1 protein	2.85	3.28
P06727	Apolipoprotein A-IV	2.79	3.21
P02748	Complement component C9	2.63	3.03
B0YIW2	Apolipoprotein C-III	2.25	2.59
P02768	Serum albumin	2.23	2.57
P04264	Keratin, type II cytoskeletal 1	2.05	2.36
P07360	Complement component C8 gamma chain	2.05	2.36
P10909-2	Isoform 2 of Clusterin	1.99	2.29
P00734	Prothrombin	1.94	2.23
P01857	Ig gamma-1 chain C region	1.38	1.59
P55056	Apolipoprotein C-IV	1.28	1.47
Q99969	Retinoic acid receptor responder protein 2	1.26	1.45
P04004	Vitronectin	1.25	1.44
P01031	Complement C5	1.23	1.42
P02652	Apolipoprotein A-II	1.22	1.40
P02655	Apolipoprotein C-II	1.22	1.40
P35527	Keratin, type I cytoskeletal 9	1.16	1.33
P01834	Ig kappa chain C region	1.16	1.33
P01781	Ig heavy chain V-III	1.03	1.18
P0DJI9	Serum amyloid A-2 protein	0.96	1.10
P13671	Complement component C6	0.92	1.06
F5GY80	Complement component C8 beta chain	0.87	1.00
P35542	Serum amyloid A-4 protein	0.85	0.98
P04196	Histidine-rich glycoprotein	0.79	0.91
P13645	Keratin, type I cytoskeletal 10	0.72	0.83
G3V1N2	HCG1745306, isoform CRA_a	0.66	0.76
P10643	Complement component C7	0.65	0.75
P01765	Ig heavy chain V-III region TIL	0.63	0.72
P35908	Keratin, type II cytoskeletal 2 epidermal	0.55	0.63
P01871-2	Isoform 2 of Ig mu chain C region	0.53	0.61
Q96IY4	Carboxypeptidase B2	0.48	0.55
G3V2M1	Plasma serine protease inhibitor	0.44	0.51
P01859	Ig gamma-2 chain C region	0.42	0.48
P10124	Serglycin	0.42	0.48
P12259	Coagulation factor V	0.41	0.47
P07357	Complement component C8 alpha chain	0.4	0.46
E7EPG1	Multimerin-1	0.37	0.43
P06396	Gelsolin	0.35	0.40
P01008	Antithrombin-III	0.35	0.40
P18428	Lipopolysaccharide-binding protein	0.35	0.40
H0YIN9	Keratin, type II cytoskeletal 5	0.33	0.38
E7EQ48	Proteoglycan 4	0.32	0.37

A0M8Q6	Ig lambda-7 chain C region	0.3	0.35
P02763	Alpha-1-acid glycoprotein 1	0.3	0.35
P01593	Ig kappa chain V-I region AG	0.28	0.32
P01614	Ig kappa chain V-II region Cum	0.27	0.31
P08697	Alpha-2-antiplasmin	0.26	0.30
P01042	Kininogen-1	0.25	0.29
B4E1Z4	Complement factor B	0.23	0.26
B7Z8R6	Trypstatin	0.23	0.26
J3KN47	Serotransferrin	0.22	0.25
B1AKG0	Complement factor H-related 1	0.22	0.25
P24593	Insulin-like growth factor-binding protein 5	0.22	0.25
J3KMX7	Matrix Gla protein	0.22	0.25
F5GXS5	Apolipoprotein F	0.21	0.24
P02766	Transthyretin	0.21	0.24
P08603	Complement factor H	0.2	0.23
Q5VYL6	Complement factor H-related 5	0.2	0.23
P03950	Angiogenin	0.2	0.23
D6REX5	Selenoprotein P (Fragment)	0.19	0.22
P10646	Tissue factor pathway inhibitor	0.19	0.22
D6RAQ1	Heparanase 8 kDa subunit	0.18	0.21
C9JV77	Alpha-2-HS-glycoprotein	0.17	0.20
Q53RD9-2	Isoform 2 of fibulin-7	0.15	0.17
P01137	Transforming growth factor beta-1	0.15	0.17
P02790	Hemopexin	0.13	0.15
P00742	Coagulation factor X	0.12	0.14
P04259	Keratin, type II cytoskeletal 6B	0.11	0.13
G8JL88	Apolipoprotein L1	0.11	0.13
G8JLA8	Transforming growth factor-beta-induced protein ig-h3	0.09	0.10
P01876	Ig alpha-1 chain C region	0.09	0.10
P09871	Complement C1s subcomponent	0.09	0.10
P01023	Alpha-2-macroglobulin	0.08	0.09
Q0VAC5	HP protein	0.08	0.09
P27918	Properdin	0.06	0.07
E7EV71	Latent-transforming growth factor beta-binding protein 1	0.06	0.07
E7EQB2	Lactoferroxin-C	0.04	0.05
Q5T985	Inter-alpha (Globulin) inhibitor H2	0.03	0.03

The absolute protein abundance in proteomics was estimated with the Exponentially Modified Protein Abundance Index through HCD fragmentation. Protein abundance (mol %) = emPAI/$\sum$(emPAI) X100. Protein score 50 was taken as the identity threshold as the confidence level is higher than 95%

Table S4. List of proteins detected in the corona on the glucose-containing glycopolymer layer with grafting density 0.034 chains/nm^2

Accession number	Protein	emPAI	Abundance (%)
P01024	Complement C3	14.04	15.28
P02647	Apolipoprotein A-I	13.43	14.61
P02649	Apolipoprotein E	10.54	11.47
P02654	Apolipoprotein C-I	5.72	6.22
P02768	Serum albumin	3.23	3.51
P06727	Apolipoprotein A-IV	3.07	3.34
P07360	Complement component C8 gamma chain	3.03	3.30
P02748	Complement component C9	3.01	3.28
P10909-2	Isoform 2 of Clusterin	2.52	2.74
P04264	Keratin, type II cytoskeletal 1	2.05	2.23
P0CG05	Ig lambda-2 chain C regions	1.87	2.03
P35527	Keratin, type I cytoskeletal 9	1.8	1.96
P01834	Ig kappa chain C region	1.79	1.95
B0YIW2	Apolipoprotein C-III	1.57	1.71
P04004	Vitronectin	1.53	1.66
P01031	Complement C5	1.47	1.60
P0DJI8	Serum amyloid A-1 protein	1.46	1.59
P00734	Prothrombin	1.24	1.35
P02655	Apolipoprotein C-II	1.22	1.33
P35908	Keratin, type II cytoskeletal 2 epidermal	1.18	1.28
P13671	Complement component C6	0.98	1.07
Q99969	Retinoic acid receptor responder protein 2	0.92	1.00
F5GY80	Complement component C8 beta chain	0.87	0.95
P13645	Keratin, type I cytoskeletal 10	0.82	0.89
P10643	Complement component C7	0.7	0.76
P02652	Apolipoprotein A-II	0.7	0.76
P01857	Ig gamma-1 chain C region	0.68	0.74
G3V1N2	HCG1745306, isoform CRA_a	0.66	0.72
P01766	Ig heavy chain V-III region BRO	0.58	0.63
P0DJI9	Serum amyloid A-2 protein	0.57	0.62
P01008	Antithrombin-III	0.52	0.57
P02766	Transthyretin	0.47	0.51
P06396	Gelsolin	0.45	0.49
P02533	Keratin, type I cytoskeletal 14	0.36	0.39
P07357	Complement component C8 alpha chain	0.33	0.36
C9JEX1	Kininogen-1	0.33	0.36
P12259	Coagulation factor V	0.31	0.34
P01859	Ig gamma-2 chain C region	0.3	0.33
Q96IY4	Carboxypeptidase B2	0.3	0.33
P19652	Alpha-1-acid glycoprotein 2	0.3	0.33
P13647	Keratin, type II cytoskeletal 5	0.29	0.32
P01620	Ig kappa chain V-III region SIE	0.29	0.32
Q13103	Secreted phosphoprotein 24	0.29	0.32
P01871-2	Isoform 2 of Ig mu chain C region	0.28	0.30
J3KN47	Serotransferrin	0.28	0.30
C9JV77	Alpha-2-HS-glycoprotein	0.27	0.29

D6RAK8	Vitamin D-binding protein	0.25	0.27
G5E9R0	Actin	0.25	0.27
P02538	Keratin, type II cytoskeletal 6A	0.24	0.26
E7EPG1	Multimerin-1	0.22	0.24
B1AKG0	Complement factor H-related 1	0.22	0.24
B4E1Z4	Complement factor B	0.2	0.22
P02790	Hemopexin	0.2	0.22
F8VXB4	Keratin, type II cytoskeletal 8	0.18	0.20
P01876	Ig alpha-1 chain C region	0.18	0.20
P08603	Complement factor H	0.14	0.15
C9JF17	Apolipoprotein D	0.14	0.15
P02763	Alpha-1-acid glycoprotein 1	0.14	0.15
P27918	Properdin	0.13	0.14
E7EQ48	Proteoglycan 4	0.13	0.14
P06858	Lipoprotein lipase	0.13	0.14
P08697	Alpha-2-antiplasmin	0.12	0.13
B0UZ83	Complement C4 gamma chain	0.11	0.12
P01023	Alpha-2-macroglobulin	0.1	0.11
Q5T985	Inter-alpha (Globulin) inhibitor H2	0.1	0.11
Q5VYL6	Complement factor H-related 5	0.1	0.11

The absolute protein abundance in proteomics was estimated with the Exponentially Modified Protein Abundance Index through HCD fragmentation. Protein abundance (mol %) = emPAI/$\sum$(emPAI) X100. Protein score 50 was taken as the identity threshold as the confidence level is higher than 95%.

Table S5. Adsorbed HSA and C3-dep serum concentration onto galactose nanoparticles with different grafting density prepared from 669 nm PS particles.

Samples	Grafting density (chains/nm^2)	Complement consumption by CH50 assay	Non-specific protein adsorption	
			Adsorbed HSA concentration (mg/mL)	Adsorbed C3-dep serum concentration (mg/mL)
Galactose NP1	0.11	100.07 ± 0.24	0.11 ± 0.01	0.11 ± 0.02
Galactose NP3	0.064	82.73 ± 10.22	0.14 ± 0.03	0.17 ± 0.02
Galactose NP5	0.023	13.57 ± 5.55	0.17 ± 0.02	0.18 ± 0.01
PS	0	8 ± 2.5	0.29 ± 0.02	0.27 ± 0.03

Nanoparticle suspension (20 uL, 50 cm^2) was incubated with C3-depleted serum (180 uL) or HSA solution (180 uL, 40 mg/mL) at 37 °C for 1h. Then the particles were washed by PBS buffer solution twice (200 uL each). The adsorbed proteins were eluted from the surface by boiled at 90 °C with 2% SDS (100 uL) for 10 minutes. The eluted samples were then centrifuged, and the supernatants were collected. The protein concentration in the supernatant was analyzed by NanoDrop 1000 Spectrophotometer through the measurement of adsorption at 280 nm.

Table S6. Ions scores of proteins identified by quantification MS run (between galactose polymer with different grafting density).

Threshold value of Individual ions scores for the quantification MS run (between galactose polymer with different grafting density): 37	
Protein	Score
Complement C3	8502
Complement C5	962
Complement C6	526
Complement C7	722
Complement C8	225
Complement C9	1042
Factor B	185
Factor H	120

Table S7. Ions scores of proteins identified by quantification MS run (between galactose and glucose nanoparticle with similar grafting density).

Threshold value of Individual ions scores for the quantification MS run (between galactose and glucose nanoparticle with similar grafting density): 37	
Protein	Score
Complement C3	8348
Complement C5	849
Complement C6	298
Complement C7	563
Complement C8	286
Complement C9	898
Factor B	129
Factor H	82

References

1. Arima, Y.; Kawagoe, M.; Toda, M.; Iwata, H. Complement Activation by Polymers Carrying Hydroxyl Groups. *ACS Appl. Mater. Interfaces* **2009**, *1*, 2400-2407.