

APPENDIX A
Calculation of percent carbon conjugate

A.1 Calculation of percent carbon conjugate at 350µg/ml of anti-NGAL antibody in 0.2% colloidal carbon suspension

Table A.1 Absorbance at 562 nm of BSA standard curve for measuring protein at 350µg/ml of anti-NGAL antibody in 0.2% colloidal carbon suspension

Concentration (µg/ml)	Absorbance round 1	Absorbance round 2	Absorbance average
0	0	0	0
0.5	0.032	0.03	0.031
1	0.06	0.056	0.058
2.5	0.134	0.142	0.138
5	0.255	0.261	0.258
10	0.437	0.428	0.4325
20	0.8	0.825	0.8125

$$\% \text{Carbon conjugate} = \frac{c_1 - c_2}{c_1} \times 100 \text{ (A.1)}$$

Where;

C1 = Initial NGAL concentration

C2 = Free NGAL concentration

Table A.2 Percent carbon conjugate at 350µg/ml in 0.2% colloidal carbon suspension

% Colloidal carbon suspension	C1 (µg/ml)	C2 (µg/ml)	%Carbon conjugate
0.2%	350	37.71	89.23

A.2 Calculation of percent carbon conjugate at 350 µg/ml of anti-NGAL antibody in 1.0% and 2.0% colloidal carbon suspension

Table A.3 Absorbance at 562 nm of BSA standard curve for measuring protein at 350µg/ml of anti-NGAL antibody in 1.0% and 2.0% colloidal carbon suspension

Concentration (µg/ml)	Absorbance round 1	Absorbance round 2	Absorbance average
0	0	0	0
0.5	0.057	0.056	0.0565
1	0.160	0.038	0.099
2.5	0.106	0.111	0.1085
5	0.271	0.277	0.274
10	0.429	0.511	0.47
20	0.806	0.804	0.805
40	1.567	1.544	1.5555

$$\% \text{Carbon conjugate} = \frac{C_1 - C_2}{C_1} \times 100 \quad (\text{A.1})$$

Where;

C1 = Initial NGAL concentration

C2 = Free NGAL concentration

Table A.4 Percent carbon conjugate at 350µg/ml in 1.0% and 2% colloidal carbon suspension

% Colloidal carbon suspension	C1 (µg/ml)	C2 (µg/ml)	%Carbon conjugate
1.0%	350	161.87	53.75
2.0%	350	320.79	8.34

APPENDIX B

Size of carbon conjugate after sonication

Table 4.3 Size of carbon conjugate after sonication

Day after sonication (day)	Size of carbon (nm)
60	382.07
30	378.43
15	260.17
7	355.8
3	211.23
1	187.43

APPENDIX C

The data of the intensity from an Image J program

Table C.1 The intensity from an Image J program in section 4.2.2.1.1

Number of membrane	Intensity			
	Spot 1	Spot 2	Spot 3	Spot 4
1	5.13	5.80	3.89	7.29
2	5.66	6.85	4.56	8.25
3	-	-	-	-
4	-	-	-	-
5	4.37	5.91	8.28	4.45
6	4.84	7.56	4.33	9.67
7	6.36	8.41	7.20	4.88
8	-			
9	4.59	3.92	4.72	3.10
10	5.55	6.97	5.68	6.36
11	-	-	-	-
12	-	-	-	-

*membrane number: 3, 4, 8, 11 and 12 were tested by lateral flow.

Table C.2 The intensity from an Image J program in section 4.2.2.1.2

Number of membrane	Intensity			
	spot 1	spot 2	spot 3	spot 4
13	30.16	172.66	174.88	172.18
14	86.04	17.57	46.41	37.90
15	77.52	11.77	40.14	36.67
16	-	-	-	-
17	-	-	-	-
18	17.18	9.69	4.43	9.50
19	11.49	7.06	17.84	8.04
20	10.39	10.14	12.91	13.25
21	-	-	-	-
22	-	-	-	-
23	10.12	8.36	17.20	14.99
24	5.90	3.40	5.54	4.00
25	5.65	7.08	7.02	10.35
26	-	-	-	-
27	-	-	-	-

*membrane number 16, 17, 21, 22, 26 and 27 were tested by lateral flow.

Table C.3 The intensity from an Image J program in section 4.2.2.1.3

Number of membrane	Intensity			
	spot 1	spot 2	spot 3	spot 4
1	183.24	88.22	61.57	72.59
2	62.67	56.87	65.12	69.26
3	88.30	89.23	76.25	77.69
4	92.29	80.50	85.12	67.24
5	80.11	74.02	76.95	70.60
6	89.45	87.60	84.28	74.40
7	106.30	91.05	93.48	92.92
8	87.81	93.84	91.82	82.71
9	66.37	70.61	85.26	86.57
10	106.97	99.50	95.28	110.93
11	106.30	166.61	98.69	103.13
12	95.91	94.43	103.82	109.94
X1	87.05	67.19	206.81	107.62
X2	84.39	69.17	205.71	106.82