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APPENDICES

APPENDIX A

Weight loss of Thai silk fibroin/type B gelatin electrospun fiber mats after treatment

Table A-1 Treatment by soaking in ethanol

| SF/GB 50/50 | | | | | |
|-------------|---|--|-----------------|-------------------------|-------|
| No. | Dry weight of fiber mats before crosslink (g) | Dry weight of fiber mats after crosslink (g) | Weight loss (%) | Average weight loss (%) | SD |
| 1 | 48.90 | 21.60 | 55.83 | 53.00 | 10.96 |
| 2 | 49.30 | 21.00 | 57.40 | | |
| 3 | 40.10 | 18.10 | 54.86 | | |
| 4 | 47.90 | 30.30 | 36.74 | | |
| 5 | 41.50 | 18.70 | 54.94 | | |
| 6 | 50.30 | 22.00 | 56.26 | | |
| 7 | 41.10 | 23.60 | 42.58 | | |
| 8 | 48.80 | 30.10 | 38.32 | | |
| 9 | 47.60 | 12.90 | 72.90 | | |
| 10 | 52.20 | 20.80 | 60.15 | | |
| SF/GB 30/70 | | | | | |
| No. | Dry weight of fiber mats before crosslink (g) | Dry weight of fiber mats after crosslink (g) | Weight loss (%) | Average weight loss (%) | SD |
| 1 | 17.40 | 9.70 | 44.25 | 50.27 | 8.45 |
| 2 | 28.70 | 16.40 | 42.86 | | |
| 3 | 34.80 | 20.00 | 42.53 | | |
| 4 | 36.90 | 14.50 | 60.70 | | |
| 5 | 37.20 | 14.90 | 59.95 | | |
| 6 | 34.60 | 13.90 | 59.83 | | |
| 7 | 36.20 | 14.60 | 59.67 | | |
| 8 | 40.70 | 23.10 | 43.24 | | |
| 9 | 31.70 | 17.50 | 44.79 | | |
| 10 | 23.20 | 12.80 | 44.83 | | |

| SF/GB 10/90 | | | | | |
|-------------|---|--|-----------------|-------------------------|------|
| No. | Dry weight of fiber mats before crosslink (g) | Dry weight of fiber mats after crosslink (g) | Weight loss (%) | Average weight loss (%) | SD |
| 1 | 11.10 | 2.70 | 75.68 | 70.25 | 8.11 |
| 2 | 12.10 | 3.50 | 71.07 | | |
| 3 | 24.90 | 9.00 | 63.86 | | |
| 4 | 12.00 | 2.30 | 80.83 | | |
| 5 | 11.50 | 5.60 | 51.30 | | |
| 6 | 10.40 | 3.20 | 69.23 | | |
| 7 | 14.10 | 4.20 | 70.21 | | |
| 8 | 13.00 | 3.40 | 73.85 | | |
| 9 | 12.20 | 2.90 | 76.23 | | |
| 10 | 10.40 | 3.10 | 70.19 | | |

Table A-2 Treatment by soaking in EDC/NHS in ethanol

| SF/GB 50/50 | | | | | |
|--------------------|--|---|------------------------|--------------------------------|-----------|
| No. | Dry weight of fiber mats before crosslink (g) | Dry weight of fiber mats after crosslink (g) | Weight loss (%) | Average weight loss (%) | SD |
| 1 | 28.80 | 13.80 | 52.08 | 44.94 | 8.08 |
| 2 | 29.80 | 19.50 | 34.56 | | |
| 3 | 31.50 | 20.60 | 34.60 | | |
| 4 | 39.60 | 19.50 | 50.76 | | |
| 5 | 32.80 | 15.60 | 52.44 | | |
| 6 | 32.80 | 15.50 | 52.74 | | |
| 7 | 40.40 | 19.60 | 51.49 | | |
| 8 | 26.10 | 15.60 | 40.23 | | |
| 9 | 32.10 | 17.40 | 45.79 | | |
| 10 | 37.50 | 24.50 | 34.67 | | |
| SF/GB 30/70 | | | | | |
| No. | Dry weight of fiber mats before crosslink (g) | Dry weight of fiber mats after crosslink (g) | Weight loss (%) | Average weight loss (%) | SD |
| 1 | 22.30 | 11.20 | 49.78 | 50.59 | 0.99 |
| 2 | 26.80 | 13.50 | 49.63 | | |
| 3 | 20.90 | 10.50 | 49.76 | | |
| 4 | 27.70 | 13.40 | 51.62 | | |
| 5 | 33.00 | 16.20 | 50.91 | | |
| 6 | 23.70 | 11.50 | 51.48 | | |
| 7 | 31.30 | 15.10 | 51.76 | | |
| 8 | 33.30 | 16.50 | 50.45 | | |
| 9 | 26.10 | 13.30 | 49.04 | | |
| 10 | 23.90 | 11.60 | 51.46 | | |
| SF/GB 10/90 | | | | | |
| No. | Dry weight of fiber mats before crosslink (g) | Dry weight of fiber mats after crosslink (g) | Weight loss (%) | Average weight loss (%) | SD |
| 1 | 18.90 | 8.20 | 56.61 | 58.23 | 3.02 |
| 2 | 15.60 | 7.00 | 55.13 | | |
| 3 | 12.70 | 5.00 | 60.63 | | |
| 4 | 18.96 | 8.70 | 54.11 | | |
| 5 | 18.00 | 6.30 | 65.00 | | |
| 6 | 14.40 | 6.00 | 58.33 | | |
| 7 | 11.70 | 4.80 | 58.97 | | |
| 8 | 11.70 | 4.90 | 58.12 | | |
| 9 | 14.80 | 6.20 | 58.11 | | |
| 10 | 11.70 | 5.00 | 57.26 | | |

Table A-3 Treatment by spraying and soaking in EDC/NHS in ethanol

| SF/GB 50/50 | | | | | |
|--------------------|--|---|------------------------|--------------------------------|-----------|
| No. | Dry weight of fiber mats before crosslink (g) | Dry weight of fiber mats after crosslink (g) | Weight loss (%) | Average weight loss (%) | SD |
| 1 | 0.0514 | 0.0502 | 2.33 | 2.03 | 1.18 |
| 2 | 0.0550 | 0.0548 | 0.36 | | |
| 3 | 0.0641 | 0.0623 | 2.81 | | |
| 4 | 0.0556 | 0.0554 | 0.36 | | |
| 5 | 0.0645 | 0.0628 | 2.64 | | |
| 6 | 0.0561 | 0.0540 | 3.74 | | |
| 7 | 0.0505 | 0.0496 | 1.78 | | |
| 8 | 0.0465 | 0.0458 | 1.51 | | |
| 9 | 0.0578 | 0.0558 | 3.46 | | |
| 10 | 0.0394 | 0.0389 | 1.27 | | |
| SF/GB 30/70 | | | | | |
| No. | Dry weight of fiber mats before crosslink (g) | Dry weight of fiber mats after crosslink (g) | Weight loss (%) | Average weight loss (%) | SD |
| 1 | 0.0324 | 0.0314 | 3.09 | 4.04 | 0.70 |
| 2 | 0.0520 | 0.0501 | 3.65 | | |
| 3 | 0.0478 | 0.0458 | 4.18 | | |
| 4 | 0.0369 | 0.0353 | 4.34 | | |
| 5 | 0.0439 | 0.0423 | 3.64 | | |
| 6 | 0.0624 | 0.0598 | 4.17 | | |
| 7 | 0.0330 | 0.0320 | 3.03 | | |
| 8 | 0.0364 | 0.0348 | 4.40 | | |
| 9 | 0.0509 | 0.0482 | 5.30 | | |
| 10 | 0.0392 | 0.0374 | 4.59 | | |
| SF/GB 10/90 | | | | | |
| No. | Dry weight of fiber mats before crosslink (g) | Dry weight of fiber mats after crosslink (g) | Weight loss (%) | Average weight loss (%) | SD |
| 1 | 0.0559 | 0.0511 | 8.59 | 8.17 | 2.56 |
| 2 | 0.0605 | 0.0561 | 7.27 | | |
| 3 | 0.0350 | 0.0338 | 3.43 | | |
| 4 | 0.0557 | 0.0517 | 7.18 | | |
| 5 | 0.0610 | 0.0547 | 10.33 | | |
| 6 | 0.0656 | 0.0593 | 9.60 | | |
| 7 | 0.0568 | 0.0501 | 11.80 | | |
| 8 | 0.0419 | 0.0399 | 4.77 | | |
| 9 | 0.0598 | 0.0541 | 9.53 | | |
| 10 | 0.0586 | 0.0532 | 9.22 | | |

APPENDIX B

In vitro biodegradation of Thai silk fibroin/type B gelatin electrospun fiber mats

Table B-1 *In vitro* biodegradation in collagenase solution

| Degradation time (hour) | SF/GB 50/50 | | | | | |
|----------------------------|-------------|---|--|-------------------------|---------------------------------------|-------|
| | No. | Dry weight of fiber mats before degradation (g) | Dry weight of fiber mats after degradation (g) | Remaining weight (%) | Average Remaining weight (%) | SD |
| 2 | 1 | 0.0036 | 0.0028 | 77.78 | 86.11 | 7.25 |
| | 2 | 0.0029 | 0.0026 | 89.66 | | |
| | 3 | 0.0033 | 0.0030 | 90.91 | | |
| 4 | 1 | 0.0057 | 0.0041 | 71.93 | 85.21 | 12.05 |
| | 2 | 0.0066 | 0.0063 | 95.45 | | |
| | 3 | 0.0068 | 0.0060 | 88.24 | | |
| 8 | 1 | 0.0052 | 0.0046 | 88.46 | 61.54 | 35.92 |
| | 2 | 0.0061 | 0.0046 | 75.41 | | |
| | 3 | 0.0053 | 0.0042 | 20.75 | | |
| 12 | 1 | 0.0049 | 0.0037 | 75.51 | 78.62 | 4.67 |
| | 2 | 0.0050 | 0.0042 | 84.00 | | |
| | 3 | 0.0055 | 0.0042 | 76.36 | | |
| 16 | 1 | 0.0074 | 0.0069 | 93.24 | 82.73 | 10.82 |
| | 2 | 0.0074 | 0.0053 | 71.62 | | |
| | 3 | 0.0072 | 0.0060 | 83.33 | | |
| 20 | 1 | 0.0087 | 0.0048 | 55.17 | 45.81 | 12.02 |
| | 2 | 0.0064 | 0.0032 | 50.00 | | |
| | 3 | 0.0062 | 0.0042 | 32.26 | | |
| 24 | 1 | 0.0053 | 0.003 | 56.60 | 55.34 | 3.57 |
| | 2 | 0.0074 | 0.0043 | 58.11 | | |
| | 3 | 0.0076 | 0.0039 | 51.32 | | |
| 28 | 1 | 0.0035 | 0.0019 | 54.29 | 50.45 | 4.53 |
| | 2 | 0.0031 | 0.0016 | 51.61 | | |
| | 3 | 0.0033 | 0.0015 | 45.45 | | |
| 32 | 1 | 0.0050 | 0.0026 | 52.00 | 50.71 | 3.20 |
| | 2 | 0.0049 | 0.0026 | 53.06 | | |
| | 3 | 0.0068 | 0.0032 | 47.06 | | |

| Degradation time (hour) | SF/GB 30/70 | | | | | |
|----------------------------|-------------|--|--|-------------------------|------------------------------------|-------|
| | No. | Dry weight of fiber mats before degradation (g) | Dry weight of fiber mats after degradation (g) | Remaining weight (%) | Average Remaining weight (%) | SD |
| 2 | 1 | 0.0110 | 0.0045 | 40.91 | 41.87 | 3.89 |
| | 2 | 0.0104 | 0.0048 | 46.15 | | |
| | 3 | 0.0083 | 0.0032 | 38.55 | | |
| 4 | 1 | 0.0050 | 0.0025 | 50.00 | 55.39 | 16.28 |
| | 2 | 0.0080 | 0.0034 | 42.50 | | |
| | 3 | 0.0057 | 0.0042 | 73.68 | | |
| 8 | 1 | 0.0057 | 0.0041 | 71.93 | 56.97 | 13.62 |
| | 2 | 0.0053 | 0.0024 | 45.28 | | |
| | 3 | 0.0054 | 0.0025 | 53.70 | | |
| 12 | 1 | 0.0048 | 0.0022 | 45.83 | 47.06 | 3.40 |
| | 2 | 0.0063 | 0.0028 | 44.44 | | |
| | 3 | 0.0055 | 0.0028 | 50.91 | | |
| 16 | 1 | 0.0050 | 0.0023 | 46.00 | 45.96 | 7.13 |
| | 2 | 0.0067 | 0.0026 | 38.81 | | |
| | 3 | 0.0049 | 0.0026 | 53.06 | | |
| 20 | 1 | 0.0068 | 0.0025 | 36.76 | 37.48 | 5.62 |
| | 2 | 0.0076 | 0.0033 | 43.42 | | |
| | 3 | 0.0062 | 0.0020 | 32.26 | | |
| 24 | 1 | 0.0047 | 0.0017 | 36.17 | 30.47 | 5.39 |
| | 2 | 0.0047 | 0.0014 | 29.79 | | |
| | 3 | 0.0055 | 0.0014 | 25.45 | | |
| 28 | 1 | 0.0095 | 0.0028 | 29.47 | 27.65 | 3.46 |
| | 2 | 0.0093 | 0.0022 | 23.66 | | |
| | 3 | 0.0057 | 0.0017 | 29.82 | | |
| 32 | 1 | 0.0036 | 0.0010 | 27.78 | 25.30 | 4.59 |
| | 2 | 0.0032 | 0.0009 | 28.12 | | |
| | 3 | 0.0035 | 0.0007 | 20.00 | | |

| Degradation time (hour) | SF/GB 10/90 | | | | | |
|----------------------------|-------------|--|--|-------------------------|------------------------------------|-------|
| | No. | Dry weight of fiber mats before degradation (g) | Dry weight of fiber mats after degradation (g) | Remaining weight (%) | Average remaining weight (%) | SD |
| 2 | 1 | 0.0058 | 0.0026 | 44.83 | 55.68 | 9.58 |
| | 2 | 0.0054 | 0.0034 | 62.96 | | |
| | 3 | 0.0054 | 0.0032 | 59.26 | | |
| 4 | 1 | 0.0060 | 0.0019 | 31.67 | 26.88 | 6.89 |
| | 2 | 0.0079 | 0.0015 | 18.99 | | |
| | 3 | 0.0050 | 0.0015 | 30.00 | | |
| 8 | 1 | 0.0046 | 0.0011 | 23.91 | 42.51 | 29.97 |
| | 2 | 0.0048 | 0.0011 | 77.08 | | |
| | 3 | 0.0049 | 0.0013 | 26.53 | | |
| 12 | 1 | 0.0025 | 0.0006 | 24.00 | 13.61 | 14.69 |
| | 2 | 0.0031 | 1E-04 | 3.23 | | |
| | 3 | 0.0047 | 0.0006 | 87.23 | | |
| 16 | 1 | 0.0064 | 0.0023 | 35.94 | 33.16 | 3.93 |
| | 2 | 0.0079 | 0.0024 | 30.38 | | |
| | 3 | 0.0057 | 0.0020 | 35.09 | | |
| 20 | 1 | 0.0041 | 0.0005 | 12.20 | 42.52 | 45.63 |
| | 2 | 0.0054 | 0.0011 | 20.37 | | |
| | 3 | 0.0040 | 0.0002 | 95.00 | | |
| 24 | 1 | 0.0055 | 0.0006 | 10.91 | 11.20 | 1.59 |
| | 2 | 0.0038 | 0.0005 | 13.16 | | |
| | 3 | 0.0042 | 0.0004 | 9.52 | | |
| 28 | 1 | 0.0043 | 0.0004 | 9.30 | 10.60 | 1.68 |
| | 2 | 0.0040 | 0.0005 | 12.50 | | |
| | 3 | 0.0060 | 0.0006 | 10.00 | | |
| 32 | 1 | 0.0022 | 0.0002 | 9.09 | 11.90 | 3.97 |
| | 2 | 0.0034 | 0.0005 | 14.71 | | |
| | 3 | 0.0029 | 0.0001 | 3.45 | | |

Table B-2 *In vitro* biodegradation in phosphate buffer saline

| Degradation time (hour) | SF/GB 50/50 | | | | | |
|-------------------------|-------------|---|--|----------------------|------------------------------|------|
| | No. | Dry weight of fiber mats before degradation (g) | Dry weight of fiber mats after degradation (g) | Remaining weight (%) | Average remaining weight (%) | SD |
| 2 | 1 | 6.90 | 6.90 | 100.00 | 99.44 | 0.96 |
| | 2 | 6.70 | 6.70 | 100.00 | | |
| | 3 | 6.00 | 5.90 | 98.33 | | |
| 12 | 1 | 3.80 | 3.70 | 97.37 | 98.29 | 1.48 |
| | 2 | 4.00 | 3.90 | 97.50 | | |
| | 3 | 3.80 | 3.80 | 100.00 | | |
| 24 | 1 | 6.20 | 6.00 | 96.77 | 97.99 | 1.06 |
| | 2 | 6.70 | 6.60 | 98.51 | | |
| | 3 | 7.70 | 7.60 | 98.70 | | |
| 72 | 1 | 6.80 | 6.50 | 95.59 | 94.96 | 0.70 |
| | 3 | 6.10 | 5.80 | 95.08 | | |
| | 5 | 6.90 | 6.50 | 94.20 | | |
| 168 | 1 | 4.80 | 4.50 | 93.75 | 92.13 | 2.81 |
| | 2 | 2.70 | 2.40 | 88.89 | | |
| | 3 | 6.40 | 6.00 | 93.75 | | |
| 336 | 1 | 3.40 | 3.10 | 91.18 | 90.87 | 0.48 |
| | 2 | 4.50 | 4.10 | 91.11 | | |
| | 3 | 3.10 | 2.80 | 90.32 | | |
| Degradation time (hour) | SF/GB 30/70 | | | | | |
| | No. | Dry weight of fiber mats before degradation (g) | Dry weight of fiber mats after degradation (g) | Remaining weight (%) | Average remaining weight (%) | SD |
| 2 | 1 | 7.80 | 7.80 | 100.00 | 100.00 | 0.00 |
| | 2 | 9.10 | 9.10 | 100.00 | | |
| | 3 | 7.20 | 7.20 | 100.00 | | |
| 12 | 1 | 3.70 | 3.70 | 100.00 | 100.00 | 0.00 |
| | 2 | 3.60 | 3.60 | 100.00 | | |
| | 3 | 4.10 | 4.10 | 100.00 | | |
| 24 | 1 | 3.80 | 3.80 | 100.00 | 99.07 | 1.60 |
| | 2 | 3.40 | 3.40 | 100.00 | | |
| | 3 | 3.60 | 3.50 | 97.22 | | |
| 72 | 1 | 3.60 | 3.60 | 100.00 | 99.02 | 1.70 |
| | 2 | 3.40 | 3.30 | 97.06 | | |
| | 3 | 3.70 | 3.70 | 100.00 | | |
| 168 | 1 | 3.30 | 3.30 | 100.00 | 98.33 | 1.45 |
| | 2 | 3.90 | 3.80 | 97.44 | | |
| | 3 | 4.10 | 4.00 | 97.56 | | |
| 336 | 1 | 3.40 | 3.30 | 97.06 | 96.97 | 0.09 |
| | 2 | 3.20 | 3.10 | 96.88 | | |
| | 3 | 3.30 | 3.20 | 96.97 | | |

| Degradation time (hour) | SF/GB 10/90 | | | | | |
|----------------------------|-------------|--|--|-------------------------|------------------------------------|------|
| | No. | Dry weight of fiber mats before degradation (g) | Dry weight of fiber mats after degradation (g) | Remaining weight (%) | Average remaining weight (%) | SD |
| 2 | 1 | 4.10 | 4.00 | 97.56 | 99.19 | 1.41 |
| | 2 | 4.10 | 4.10 | 100.00 | | |
| | 3 | 4.00 | 4.00 | 100.00 | | |
| 12 | 1 | 3.60 | 3.50 | 97.22 | 98.60 | 1.20 |
| | 2 | 3.50 | 3.48 | 99.43 | | |
| | 3 | 3.50 | 3.47 | 99.14 | | |
| 24 | 1 | 9.40 | 9.10 | 96.81 | 96.75 | 0.78 |
| | 2 | 7.40 | 7.10 | 95.95 | | |
| | 3 | 8.00 | 7.80 | 97.50 | | |
| 72 | 1 | 5.60 | 5.30 | 94.64 | 94.93 | 1.40 |
| | 2 | 5.90 | 5.69 | 96.44 | | |
| | 3 | 11.10 | 10.40 | 93.69 | | |
| 168 | 1 | 6.10 | 4.60 | 75.41 | 80.04 | 4.01 |
| | 2 | 6.70 | 5.50 | 82.09 | | |
| | 3 | 4.60 | 3.80 | 82.61 | | |
| 336 | 1 | 2.60 | 1.10 | 42.31 | 40.16 | 1.90 |
| | 2 | 3.80 | 1.50 | 39.47 | | |
| | 3 | 3.10 | 1.20 | 38.71 | | |

APPENDIX C

Standard curve

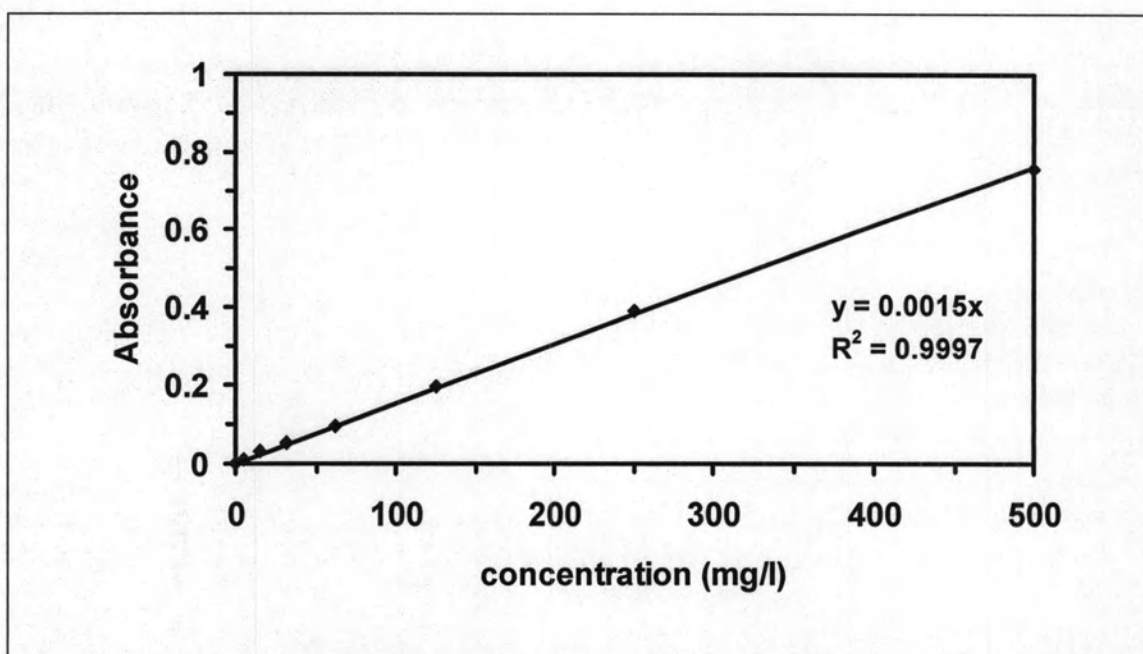


Figure C-1 Standard curve of azo-casein

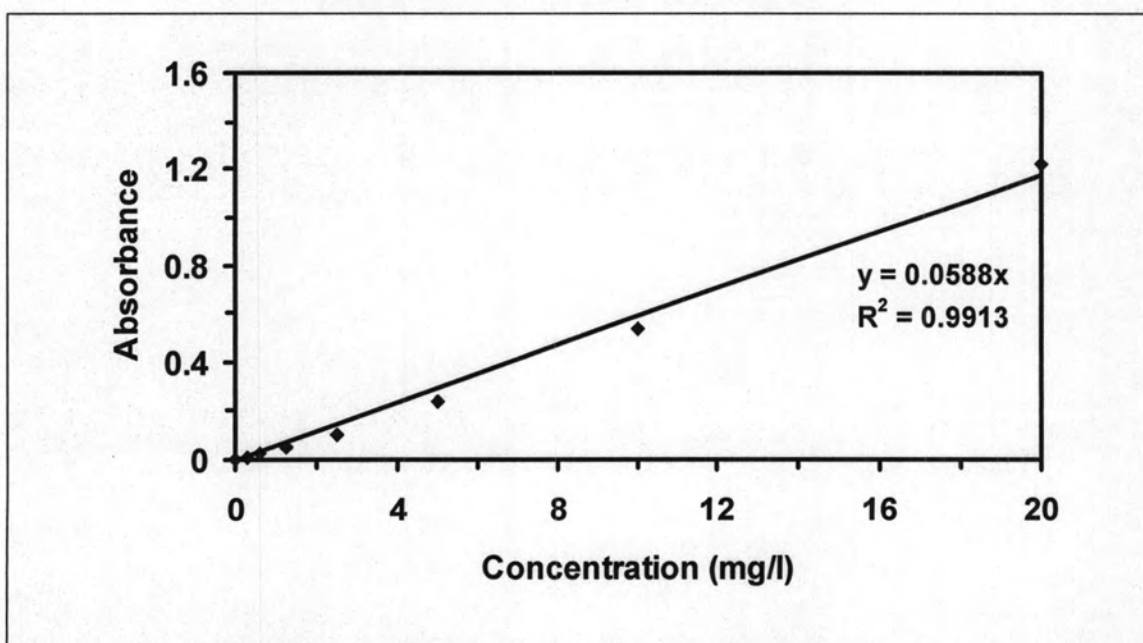


Figure C-2 Standard curve of methylene blue

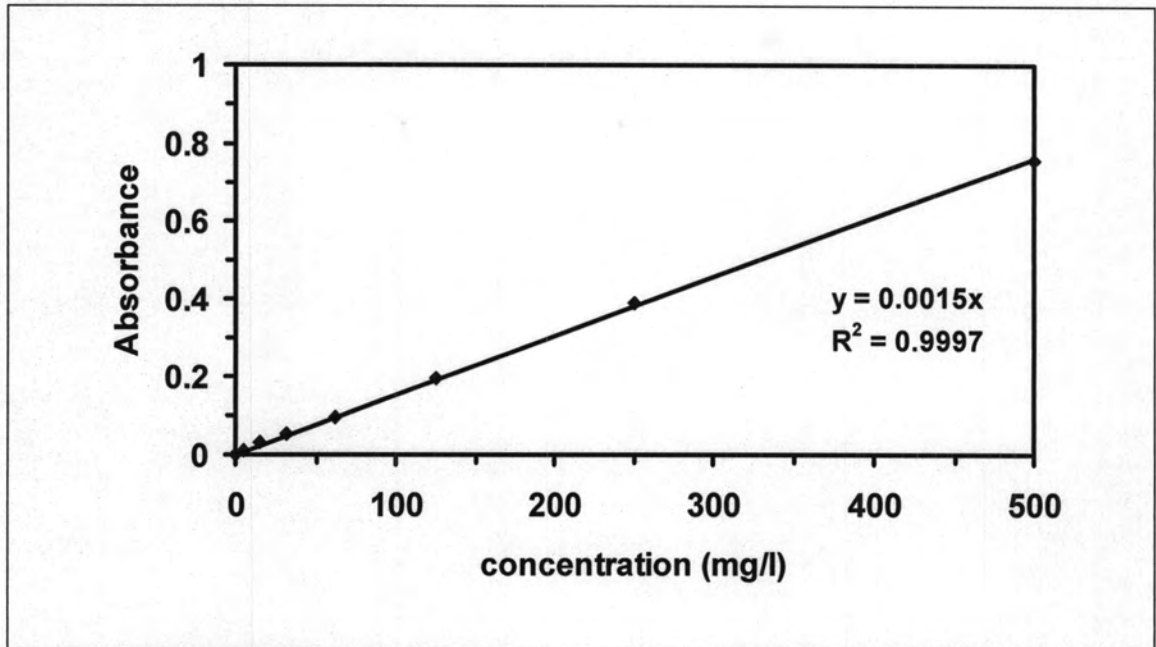


Figure C-3 Standard curve of nerve growth factor for ELISA assay

BIOGRAPHY

Miss Manunya Okhawilai was born in Bangkok, Thailand on April 2, 1985. She finished the high school education in 1996 from Saipanya school. In 2000, she received her Bachelor Degree of Engineering with a major of Chemical Engineering from Faculty of Engineering, King Mongkut's University of Technology Thonburi. After the graduation, she pursued her graduate study in Master of Engineering (chemical engineering), Faculty of Engineering, Chulalongkorn University.

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