Modulation of aggregation with Electric Field; scientific roadmap for a potential non-invasive therapy against tauopathies

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Section 1

Figure S1. HPLC profile of synthesized Ac-VQIVYK (PhF6) peptide.

Figure S2. MALDI-TOF mass spectrum of synthesized Ac-VQIVYK (PhF6) peptide. Calculated mass for PhF6 is 789.956, observed mass is 791.197 [M+H]^+, 813.224 [M+Na]^-, and 829.242 [M+Ca]^+. 
Figure S3. HPLC profile of synthesized Ac-VQIINK (PhF6*) peptide.

Figure S4. MALDI-TOF mass spectrum of synthesized Ac-VQIINK (PhF6*) peptide. Calculated mass for PhF6* is 754.911, observed mass is 756.422 [M+H]⁺, 778.185 [M+Na]⁺, and 794.183 [M+Ca]⁺.
Section 2

Figure S5. Aggregation Kinetics profile of PhF6 peptide, determined by time dependent thioflavin T fluorescence assay.

Figure S6. Aggregation Kinetics profile of PhF6* peptide, determined by time dependent thioflavin T fluorescence assay.
Section 3

Figure S7. Estimation of percentage aggregation using tyrosine estimation, by measuring the absorbance intensity of PhF6 peptide in solution. Absorbance at 280 nm is measured after an incubation period of 16 hours in presence and absence of electric field.

Figure S8. Static right angle scatter plots. Graph represent comparative static light scattering by PhF6 peptide solution allowed to aggregate in ambient conditions (0 Vcm\(^{-1}\)), AC and DC electric fields. The 0 Vcm\(^{-1}\) showed a very high scattering intensity compared to field samples, thus indicating higher aggregation compared to treated samples.
Figure S9. Static right angle scatter plots. Graph represent comparative static light scattering by PhF6* peptide solution allowed to aggregate in ambient conditions (0 Vcm$^{-1}$), AC and DC electric fields. The 0 Vcm$^{-1}$ showed a very high scattering intensity compared to field samples, thus indicating higher aggregation compared to treated samples.
Figure S10. Schematic illustration of a potential electric field based therapeutic device against tauopathies.
Section 6

Table S1: Comparative thioflavin T fluorescence data for treated (electric field) and untreated PhF6 peptide samples.

| Time (min.) | Fluorescence Intensity (a.u.) | Std. Dev. | Fluorescence Intensity (a.u.) | Std. Dev. | Fluorescence Intensity (a.u.) | Std. Dev. | Fluorescence Intensity (a.u.) | Std. Dev. | Fluorescence Intensity (a.u.) | Std. Dev. |
|-------------|-------------------------------|-----------|-------------------------------|-----------|-------------------------------|-----------|-------------------------------|-----------|-------------------------------|-----------|
| 0 V/cm      | 3.78466                       | 0.11362   | 3.78466                       | 0.11362   | 3.78466                       | 0.11362   | 3.78466                       | 0.11362   | 3.78466                       | 0.11362   |
| 150 V/cm (DC) | 3.49173                      | 0.0706    | 3.16595                       | 0.06933   | 5.54196                       | 0.09839   | 3.94184                       | 0.07166   | 4.63786                       | 0.07586   |
| 300 V/cm (DC) | 7.64945                      | 0.32159   | 4.14102                       | 0.16139   | 3.89939                       | 0.11331   | 4.62679                       | 0.09555   | 6.62668                       | 0.08811   |
| 150 V/cm (AC) | 8.21572                      | 0.17852   | 4.82133                       | 0.11965   | 6.58798                       | 0.08255   | 4.35337                       | 0.24532   | 7.54103                       | 0.27692   |
| 300 V/cm (AC) | 10.23876                     | 0.3023    | 5.17981                       | 0.07746   | 7.02221                       | 0.08574   | 5.96412                       | 0.07929   | 7.37728                       | 0.21763   |
| 100         | 11.39967                      | 0.51601   | 5.40583                       | 0.14331   | 7.94105                       | 0.08985   | 5.65875                       | 0.12512   | 7.2354                        | 0.33141   |
| 120         | 17.93171                      | 0.16115   | 4.83675                       | 0.1694    | 6.93912                       | 0.28923   | 7.361                         | 0.0902    | 6.44631                       | 0.3605    |
| 150         | 30.61325                      | 0.25589   | 7.17733                       | 0.08934   | 6.63268                       | 0.54475   | 8.55075                       | 0.1079    | 8.78657                       | 0.10024   |
| 240         | 36.1836                       | 0.36474   | 7.38816                       | 0.31358   | 8.42042                       | 0.21639   | 7.66766                       | 0.30546   | 11.63167                      | 0.0989    |
| 360         | 74.44483                      | 0.32742   | 7.91511                       | 0.42257   | 9.31676                       | 0.39744   | 8.71079                       | 0.17352   | 10.07957                      | 0.14558   |
| 480         | 90.13458                      | 0.48547   | 18.95891                      | 0.15037   | 14.97963                      | 0.17174   | 14.58304                      | 0.19087   | 20.59567                      | 0.15189   |
| 600         | 101.0842                      | 1.68668   | 21.00824                      | 0.34084   | 16.97724                      | 0.11511   | 16.18648                      | 0.85326   | 20.30977                      | 0.62177   |
| 720         | 121.6417                      | 4.31804   | 26.6759                       | 0.55251   | 18.54152                      | 0.75316   | 19.32805                      | 0.55964   | 24.71845                      | 0.31217   |
| 960         | 127.1251                      | 2.36654   | 33.37058                      | 0.29541   | 23.54854                      | 0.33164   | 32.07641                      | 0.68747   | 20.21911                      | 0.48271   |
**Table S2:** Comparative thioflavin T fluorescence data for treated (electric field) and untreated PhF6* peptide samples.

| Time (min.) | Fluorescence Intensity (a.u.) | Std. Dev. | Fluorescence Intensity (a.u.) | Std. Dev. | Fluorescence Intensity (a.u.) | Std. Dev. | Fluorescence Intensity (a.u.) | Std. Dev. | Fluorescence Intensity (a.u.) | Std. Dev. |
|-------------|-----------------------------|----------|-----------------------------|----------|-----------------------------|----------|-----------------------------|----------|-----------------------------|----------|
| 0           | 3.65543                     | 0.19374  | 3.65543                     | 0.19374  | 3.65543                     | 0.19374  | 3.65543                     | 0.19374  | 3.65543                     | 0.19374  |
| 20          | 11.61434                    | 0.1326   | 4.83019                     | 0.07256  | 4.91645                     | 0.29585  | 4.53997                     | 0.5867   | 5.2249                      | 0.2176   |
| 40          | 8.06531                     | 0.16883  | 4.40173                     | 0.66074  | 4.23282                     | 0.88534  | 3.95734                     | 0.40161  | 3.75338                     | 0.20492  |
| 60          | 8.9155                      | 1.17015  | 4.81716                     | 0.21556  | 4.987                       | 0.06713  | 3.61079                     | 0.2402   | 3.83191                     | 0.29964  |
| 90          | 13.65369                    | 0.95364  | 6.10656                     | 0.27155  | 5.26822                     | 0.78605  | 6.10953                     | 0.52616  | 5.02986                     | 0.45221  |
| 120         | 43.64246                    | 0.50713  | 5.35128                     | 0.46102  | 6.16659                     | 1.42783  | 4.59655                     | 0.28731  | 4.70611                     | 0.56348  |
| 240         | 48.95573                    | 0.81686  | 6.13732                     | 1.27464  | 4.90916                     | 0.3253   | 8.5425                      | 0.3875   | 5.48153                     | 0.41479  |
| 300         | 54.12469                    | 0.25612  | 5.98967                     | 0.84064  | 6.70051                     | 1.349    | 7.24982                     | 1.49903  | 7.06711                     | 1.26756  |
| 360         | 49.62951                    | 1.40278  | 6.46123                     | 1.4182   | 7.56686                     | 1.16942  | 7.19581                     | 0.32818  | 5.70447                     | 0.64689  |
### Table S3: Summary of MTT assay data for SH-SY5Y cell viability in presence of treated and untreated PhF6 and PhF6* peptide samples.

| Experimental Condition | % Cell Viability (SH-SY5Y) Ac-VQIVYK-NH2 | Std. Dev. Ac-VQIVYK-NH2 | % Cell Viability (SH-SY5Y) Ac-VQIINK-NH2 | Std. Dev. Ac-VQIINK-NH2 |
|------------------------|------------------------------------------|--------------------------|------------------------------------------|--------------------------|
| 0V                     | 55.64322                                 | 1.768472                 | 60.80617                                 | 5.274733                 |
| 150 AC                 | 75.04288                                 | 3.484566                 | 73.27616                                 | 5.860087                 |
| 150 DC                 | 73.92796                                 | 5.787593                 | 69.26244                                 | 5.920028                 |
| 300 AC                 | 82.43568                                 | 5.734737                 | 67.49571                                 | 6.215325                 |
| 300 DC                 | 83.15609                                 | 3.480342                 | 69.25386                                 | 5.518031                 |
| Staurosporine          | 69.7084                                 | 7.887985                 | 69.7084                                 | 7.887985                 |
| Untreated              | 99.97141                                 | 11.8114                  | 99.97141                                 | 11.8114                  |

### Table S4: Summary of MTT assay data for IMR-32 cell viability in presence of treated and untreated PhF6 and PhF6* peptide samples.

| Experimental Condition | % Cell Viability (IMR-32) Ac-VQIVYK-NH2 | Std. Dev. Ac-VQIVYK-NH2 | % Cell Viability (IMR-32) Ac-VQIINK-NH2 | Std. Dev. Ac-VQIINK-NH2 |
|------------------------|------------------------------------------|--------------------------|------------------------------------------|--------------------------|
| 0V                     | 68.16654                                 | 12.90302                 | 91.57246                                 | 4.90684                  |
| 150 AC                 | 83.72123                                 | 4.24394                  | 93.13977                                 | 8.43164                  |
| 150 DC                 | 101.7585                                 | 3.33622                  | 88.30935                                 | 7.29277                  |
| 300 AC                 | 84.94957                                 | 8.32598                  | 96.61271                                 | 4.25072                  |
| 300 DC                 | 102.8704                                 | 5.46537                  | 93.51019                                 | 6.13001                  |
| Staurosporine          | 66.58273                                 | 5.74858                  | 66.58273                                 | 5.74858                  |
| Untreated              | 99.98715                                 | 8.50592                  | 99.98715                                 | 8.50592                  |
Table S5: Summary of MTT assay data for PC-12 cell viability in presence of treated and untreated PhF6 and PhF6* peptide samples.

| Experimental Condition | % Cell Viability (PC-12) Ac-VQIVYK-NH2 | Std. Dev. Ac-VQIVYK-NH2 | % Cell Viability (PC-12) Ac-VQQINK-NH2 | Std. Dev. Ac-VQIQINK-NH2 |
|------------------------|---------------------------------------|--------------------------|---------------------------------------|--------------------------|
| 0V                     | 50.50437                              | 5.01388                  | 76.26093                              | 3.92647                  |
| 150 AC                 | 61.71262                              | 6.52366                  | 76.6308                               | 3.15261                  |
| 150 DC                 | 59.49899                              | 3.73679                  | 78.70433                              | 7.20627                  |
| 300 AC                 | 57.17552                              | 2.11028                  | 79.11231                              | 7.91041                  |
| 300 DC                 | 72.42771                              | 6.21394                  | 85.44048                              | 4.6816                   |
| Staurosporine          | 49.58529                              | 3.12509                  | 49.58529                              | 3.12509                  |
| Untreated              | 104.33759                             | 6.58416                  | 104.33759                             | 6.58416                  |