Supporting information

# Bio-composite Nanogels Based on Chitosan and Hyaluronic Acid for the Treatment of Lung Infections

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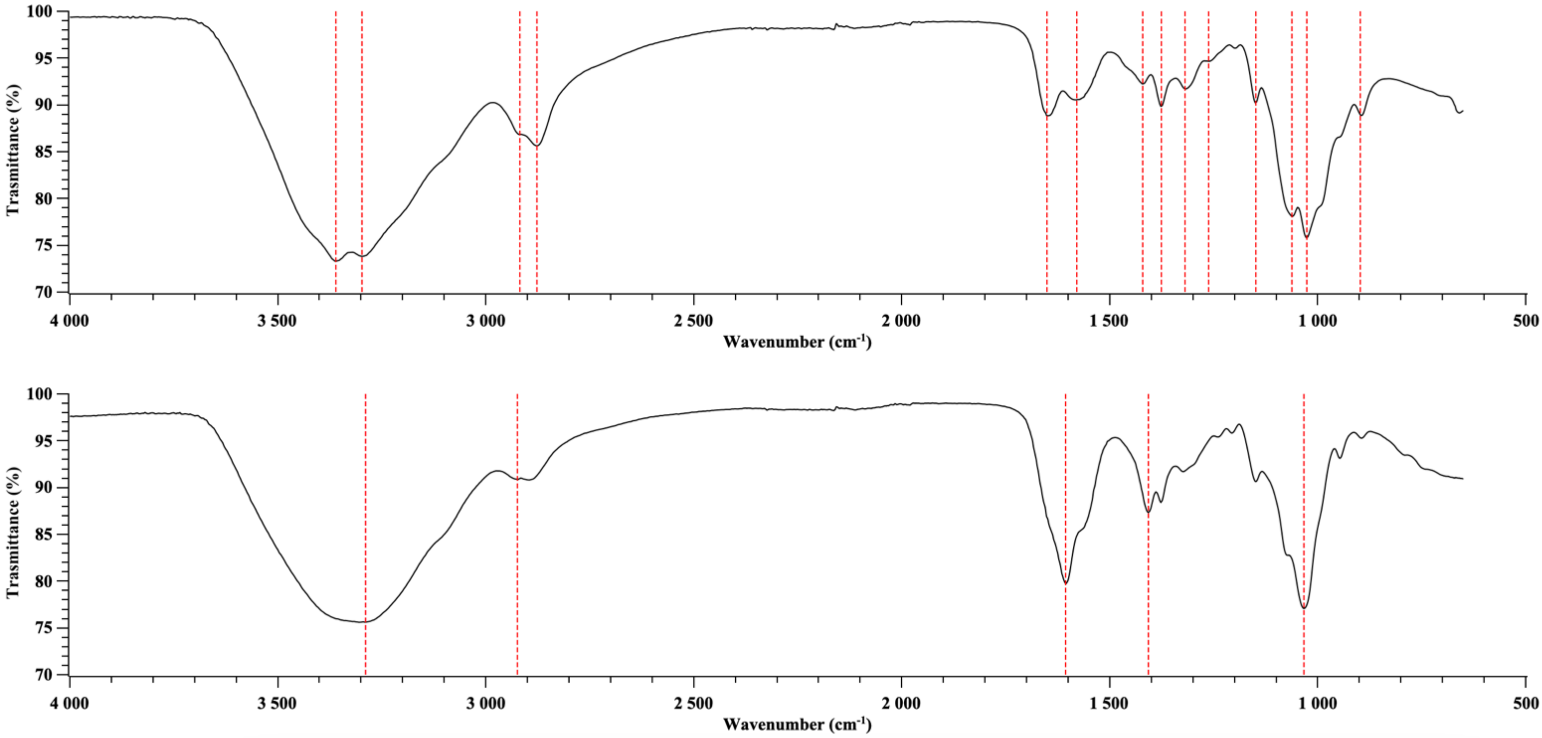


**Figure S1**: FTIR spectra of (a) CS and (b) HA powders. Characteristic peaks are highlighted. The acquisition range was from 4000 $cm^{-1}$ to 650 $cm^{-1}$ with a resolution of 1 $cm^{-1}$ and 64 scans for each sample.

**Table S1**: Peak assignment for CS FTIR spectrum.

| Peak center ($cm^{-1}$) | Molecular vibration |
|---|---|
| 3361 | $O-H$ and $N-H$ stretching |
| 3298 | $O-H$ and $N-H$ stretching |
| 2919 | $C-H$ symmetric stretching |
| 2877 | $C-H$ asymmetric stretching |
| 1650 | $C=O$ stretching of amide I |
| 1580 | $N-H$ bending of primary amine |
| 1421 | $-CH_2$ bending |
| 1376 | $-CH_3$ symmetrical deformations |
| 1319 | $C-N$ bending of amine III |
| 1261 | $O-H$ bending |
| 1149 | $C-O-C$ asymmetric stretching |
| 1061 | $C-O$ stretching |
| 1027 | $C-O$ stretching |
| 897 | $C-H$ bending out of the plane |

**Table S2**: Peak assignment for HA FTIR spectrum.

| Peak center ($cm^{-1}$) | Molecular vibration |
|---|---|
| 3289 | $O-H$ and $N-H$ stretching |
| 2924 | $C-H$ symmetric stretching |
| 1606 | $C=O$ stretching of amide I |
| 1407 | $C=O$ stretching of carboxyl groups |
| 1033 | $C-O$ stretching oh alcohol |

**Table S3**: Size and PDI over time of NGs at different amount of HA (2.5%, 3.3%, 5.0%) and stored at 4 °C. Data are the mean ± standard deviation of three independent experiments.

| | $d_H$ (nm) | | | PDI | | |
|---|---|---|---|---|---|---|
| **t (day)** | **2.5%** | **3.3%** | **5.0%** | **2.5%** | **3.3%** | **5.0%** |
| **0** | 92 ± 4 | 83 ± 5 | 98 ± 2 | 0.34 ± 0.04 | 0.33 ± 0.05 | 0.267 ± 0.004 |
| **1** | 94 ± 2 | 90 ± 10 | 117 ± 7 | 0.31 ± 0.04 | 0.30 ± 0.03 | 0.28 ± 0.03 |
| **2** | 96.6 ± 0.9 | 90 ± 12 | 120 ± 11 | 0.33 ± 0.03 | 0.31 ± 0.03 | 0.30 ± 0.06 |
| **3** | 92.9 ± 1.2 | 90 ± 14 | 130 ± 10 | 0.35 ± 0.02 | 0.31 ± 0.03 | 0.31 ± 0.05 |
| **4** | 95 ± 2 | 90 ± 11 | 130 ± 13 | 0.32 ± 0.05 | 0.32 ± 0.05 | 0.32 ± 0.05 |
| **7** | 95.3 ± 1.2 | 100 ± 13 | 140 ± 10 | 0.35 ± 0.03 | 0.34 ± 0.09 | 0.33 ± 0.06 |
| **8** | 97 ± 2 | 90 ± 15 | 130 ± 20 | 0.34 ± 0.04 | 0.32 ± 0.02 | 0.30 ± 0.05 |
| **9** | 98 ± 2 | 100 ± 20 | 140 ± 14 | 0.34 ± 0.04 | 0.31 ± 0.06 | 0.30 ± 0.05 |
| **10** | 103.6 ± 1.5 | 100 ± 14 | 130 ± 20 | 0.35 ± 0.04 | 0.32 ± 0.03 | 0.31 ± 0.07 |
| **11** | 104 ± 2 | 100 ± 13 | 130 ± 20 | 0.35 ± 0.06 | 0.36 ± 0.05 | 028 ± 0.03 |
| **14** | 107 ± 6 | 100 ± 10 | 140 ± 20 | 0.34 ± 0.05 | 0.34 ± 0.05 | 0.30 ± 0.04 |
| **15** | 105 ± 3 | 100 ± 11 | 140 ± 20 | 0.36 ± 0.04 | 0.35 ± 0.06 | 0.31 ± 0.04 |

**Table S4**: ζ-potential over time of NGs at different amount of HA (2.5%, 3.3%, 5.0%) and stored at 4 °C. Data are the mean ± standard deviation of three independent experiments.

| | **ζ-potential (mV)** | | |
|---|---|---|---|
| **t (day)** | **2.5%** | **3.3%** | **5.0%** |
| **0** | 21.6 ± 0.4 | 22 ± 2 | 16.8 ± 0.9 |
| **1** | 22.8 ± 0.6 | 21.2 ± 0.4 | 19.2 ± 0.8 |
| **2** | 22 ± 1 | 21.2 ± 0.6 | 20.0 ± 0.3 |
| **3** | 22 ± 1 | 22 ± 1 | 19.9 ± 0.5 |
| **4** | 22.6 ± 0.8 | 20.9 ± 1.3 | 19 ± 4 |
| **7** | 23.1 ± 0.6 | 21.6 ± 1.1 | 22 ± 2 |
| **8** | 23 ± 2 | 23.8 ± 1.2 | 22 ± 2 |
| **9** | 23.0 ± 0.3 | 23 ± 2 | 22.1 ± 0.6 |
| **10** | 23 ± 3 | 22 ± 3 | 20.7 ± 0.9 |
| **11** | 21 ± 3 | 22 ± 3 | 22.3 ±1.4 |
| **14** | 22 ± 2 | 23 ± 2 | 24 ± 2 |
| **15** | 23.6 ± 0.9 | 24.4 ± 1.1 | 24.8 ± 0.6 |

**Table S5**: Size and PDI over time of NGs at different amount of HA (2.5%, 3.3%, 5.0%) and stored at 37 °C. Data are the mean ± standard deviation of three independent experiments.

| | **$d_H$ (nm)** | | | **PDI** | | |
|---|---|---|---|---|---|---|
| **t (day)** | **2.5%** | **3.3%** | **5.0%** | **2.5%** | **3.3%** | **5.0%** |
| **0** | 92 ± 4 | 83 ± 5 | 98 ± 2 | 0.34 ± 0.04 | 0.33 ± 0.05 | 0.267 ± 0.004 |
| **1** | 111 ± 3 | 105 ± 4 | 120 ± 8 | 0.30 ± 0.01 | 0.32 ± 0.04 | 0.28 ± 0.03 |
| **2** | 140 ± 20 | 120 ± 11 | 123 ± 9 | 0.33 ± 0.03 | 0.29 ± 0.03 | 0.30 ± 0.06 |
| **3** | 119 ± 4 | 103 ± 9 | 124 ± 8 | 0.33 ± 0.01 | 0.33 ± 0.02 | 0.31 ± 0.05 |
| **4** | 121 ± 7 | 100 ± 8 | 129 ± 8 | 0.33 ± 0.03 | 0.29 ± 0.04 | 0.32 ± 0.05 |
| **7** | 127 ± 3 | 140 ± 30 | 130 ± 10 | 0.36 ± 0.01 | 0.23 ± 0.03 | 0.33 ± 0.06 |
| **8** | 126 ± 3 | 105 ± 5 | 140 ± 15 | 0.23 ± 0.02 | 0.31 ± 0.03 | 0.30 ± 0.05 |
| **9** | 125 ± 3 | 106 ± 7 | 133 ± 8 | 0.24 ± 0.01 | 0.32 ± 0.02 | 0.30 ± 0.05 |
| **10** | 127 ± 6 | 113 ± 3 | 132 ± 9 | 0.24 ± 0.01 | 0.29 ± 0.01 | 0.31 ± 0.07 |
| **11** | 133 ± 4 | 120 ± 20 | 137 ± 5 | 0.26 ± 0.02 | 0.27 ± 0.05 | 0.28 ± 0.03 |
| **14** | 130 ± 3 | 121 ± 7 | 144 ± 12 | 0.254 ± 0.04 | 0.29 ± 0.04 | 0.30 ± 0.04 |
| **15** | 136 ± 8 | 124 ± 11 | 148 ± 8 | 0.26 ± 0.02 | 0.27 ± 0.04 | 0.31 ± 0.04 |

**Table S6**: ζ-potential over time of NGs at different amount of HA (2.5%, 3.3%, 5.0%) and stored at 37 °C. Data are the mean ± standard deviation of three independent experiments.

| | ζ-potential (mV) | | |
|---|---|---|---|
| **t (day)** | **2.5%** | **3.3%** | **5.0%** |
| **0** | 21.6 ± 0.4 | 22 ± 2 | 16.8 ± 0.9 |
| **1** | 21.8 ± 0.3 | 20 ± 1 | 19.1 ± 0.9 |
| **2** | 22.2 ± 0.6 | 18 ± 4 | 18.4 ± 1.2 |
| **3** | 24.3 ± 0.6 | 22.8 ± 0.8 | 20.0 ± 0.7 |
| **4** | 24.5 ± 0.9 | 23.2 ± 2 | 14 ± 2 |
| **7** | 25.9 ± 0.7 | 25.6 ± 0.4 | 24 ± 2 |
| **8** | 25.8 ± 0.7 | 24.7 ± 0.3 | 24.9 ± 1.2 |
| **9** | 26.8 ± 0.8 | 25.7 ± 0.8 | 26.4 ± 0.4 |
| **10** | 26.4 ± 0.8 | 26.5 ± 0.9 | 26 ± 2 |
| **11** | 27.5 ± 0.2 | 26.3 ± 0.6 | 27 ± 2 |
| **14** | 28 ± 2 | 28.2 ± 1.2 | 29.4 ± 0.2 |
| **15** | 27 ± 2 | 30 ± 2 | 28.8 ± 0.8 |