A

Cross-sectional profile


Length (nm)

B

Autocorrelation

Lag (nm)

Supplementary Figure S1. Autocorrelation analysis of line profiles to calculate helical pitch. (A) shows an example line profile along a dsRNA. (B) shows the autocorrelation plot of the profile in (A). To calculate the periodicity in the profile, first the peaks in the autocorrelation plot were identified and then a threshold was applied to reject peaks with obvious shoulders (red circles in B show the selected peaks). The peak to peak distances between the selected peaks were then calculated. The above method was applied to all the profiles and the peak to peak distances from all the profiles were combined to determine the mean and the standard deviation of the periodicity. The same method was applied to determine the periodicity of both dsRNA and dsDNA. All the data analysis was performed using Matlab (Mathworks).

Supplementary Figure S1

A   Fraction 2   B   Fraction 3

C
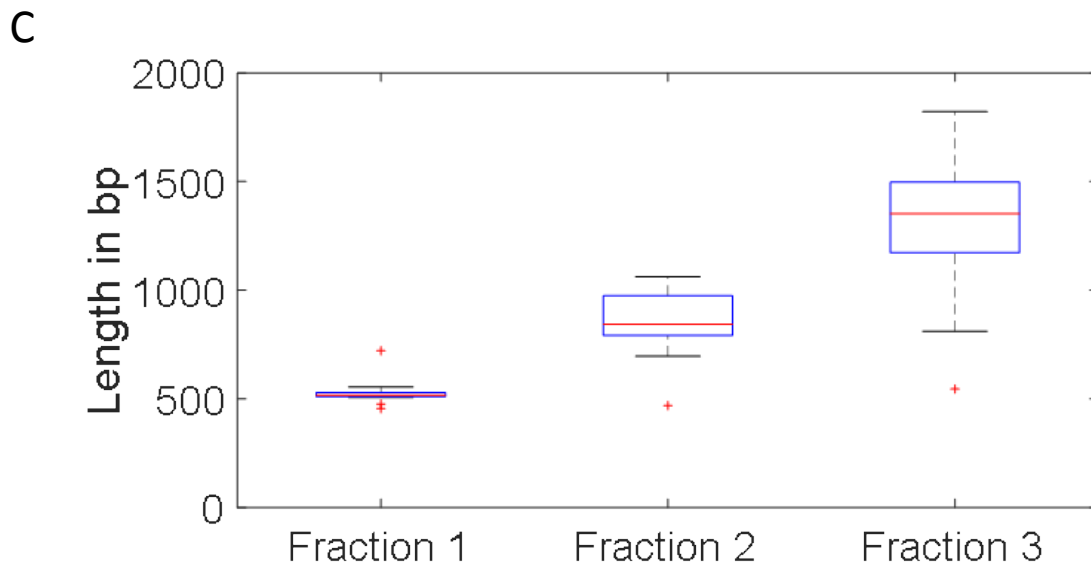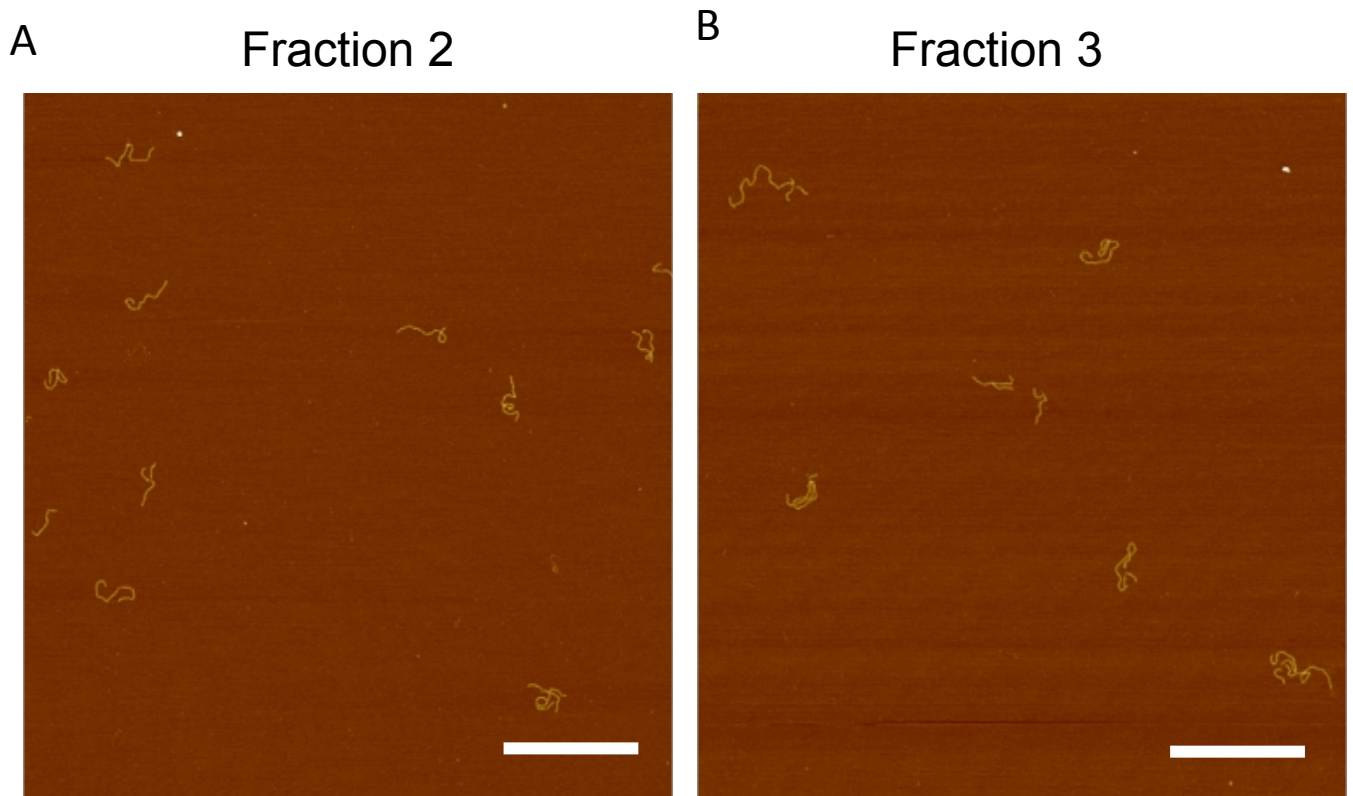
Supplementary Figure S2. AFM analysis of diluted HPLC fractions 2 and 3. (A) and (B) show the sparse surface coverage of fraction 2 and fraction 3 of HPLC purified *in vivo* dsRNA. The low surface coverage reduces the chance for intermolecular overlaps. (A) and (B) have the intensity range from black to white of 4 nm and have the same scale with the scale bar representing 400 nm. (C) AFM images of non-covalent dsRNA multimers were used to calculate their lengths in basepairs and the comparison of the distribution of lengths present in fractions 2 and 3 to that of dsRNA fraction 1 is shown as a boxplot.

Supplementary Figure S2