Supplementary Information for Soft Matter

Charge screening in RNA: An integral route for dynamics enhancements

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Preparation of tRNA samples

The wheat-germ tRNA mixtures (R7876) used in the present study was purchased from Sigma-Aldrich. RNA solution of 5 mg/ml was prepared with RNase-free nanopure H₂O. To further purify this RNA, we first added saturated phenol in equal volume. The resultant emulsion was mixed thoroughly with a vortex mixer and centrifuged at 10,000 rpm for 4 min at 4 °C. The aqueous and boundary phase were collected and further workup with chloroform. To this purified RNA solution, 100% EtOH and 250 mM NaCl were again added with a volume ratio 1:3. The mixture was incubated at -80 °C for 1 hour. It was centrifuged at 10,000 rpm for 45 min at 4 °C. A pellet was obtained by carefully decanting the supernatant and it was further washed with 70% EtOH and then dried in a speed vacuum pump.

The pellet was dissolved in RNase-free nanopure H_2O and dialyzed using a membrane (10K MWCO) against RNase-free nanopure H_2O with volume ratio 20:1 at 4 °C for 24 hours with constant stirring to remove salts. We lyophilized this salt-free RNA sample under 0.3 mbar at -40 °C for 24 hours and obtained dry RNA powder. The dry powder was dissolved (5mg/ml) in deuterated water (D₂O) to exchange labile H-atoms with D-atoms.

To prepare salt containing RNA samples, NaCl, KCl or MgCl₂ (Stock solutions prepared in D₂O) was added into the RNA-D₂O (molar ratio of MgCl₂ to RNA = 30:1). The mixed solution was incubated at room temperature for 30 min to complete folding transition. Then the sample was lyophilized again under 0.3 mbar at -40 °C for 24 hours. The dried RNA powder was rehydrated under 100 % relative humidity of D₂O. First hydration layer consisting of 42 wt % water was chosen for all wet samples.

Supplementary figures

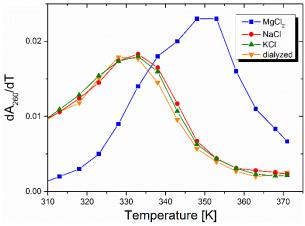


Figure S1. Temperature variation of the derivatives of UV-absorption at 260 nm, reflecting thermal denaturation of tRNA samples mixed with MgCl₂, NaCl, or KCl in double deionized H₂O (18 Ω) at pH 7.5. The mole ratio of tRNA to salt was 1:30. UV scanning was performed by heating the tRNA samples by 5 K/min and after 10 min at each temperature for thermal equilibration.

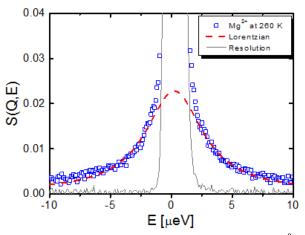
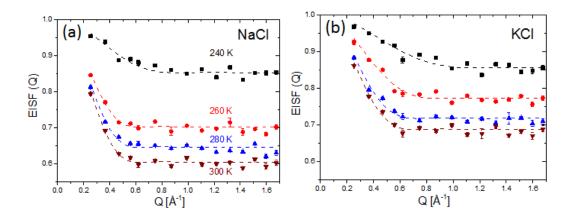


Figure S2. Dynamic structure factor, $S(Q=0.56 \text{ Å}^{-1},\text{E})$, of Mg²⁺-tRNA at 260 K. Resolution functions are $S(Q=0.56 \text{ Å}^{-1},\text{E})$ at 10 K. Dashed line is a representative Lorentzian fit of the quasielastic scattering using Eq. 2.



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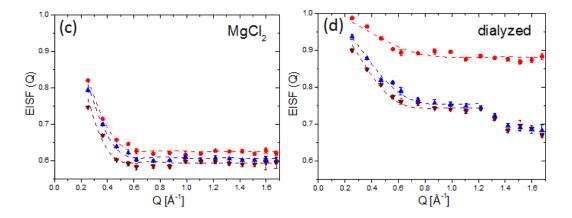


Figure S3. Elastic incoherent structure factor (EISF) of salted and dialyzed tRNA at 0.65 h at 240, 260, 280, and 300 K. Dashed lines represent fits of the EISF to a model for diffusive motions in a sphere (Eq. 4).

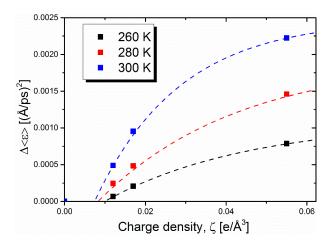


Figure S4. $\Delta < \varepsilon >$ of salted tRNA at 260, 280, and 300 K. Symbols and dash lines are experimental data and their fitting to exponential functions. The arrows indicates the charge density at zero point $\Delta \varepsilon$ at each temperature.

Note: Certain commercial material suppliers are identified in this paper to foster understanding. Such identification does not imply recommendation or endorsement by the National Institute of Standards and Technology, nor does it imply that the materials or equipment identified are necessarily the best available for the purpose.