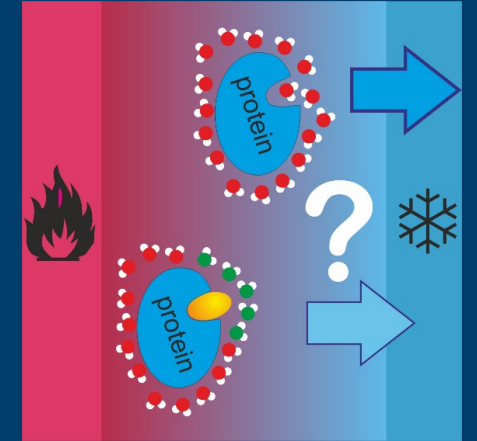
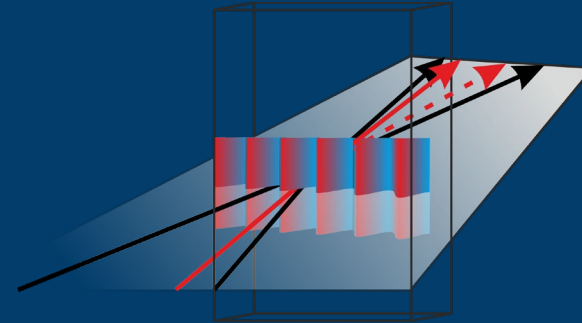


THERMOPHORESIS OF (BIO)-COMPOUNDS AND CHELATING AGENTS

Institute of Biological Information (IBI)

IBI-4: Biomacromolecular Systems and Processes



09.04.2024 | BINNY RUDANI

MOTIVATION

Thermophoresis

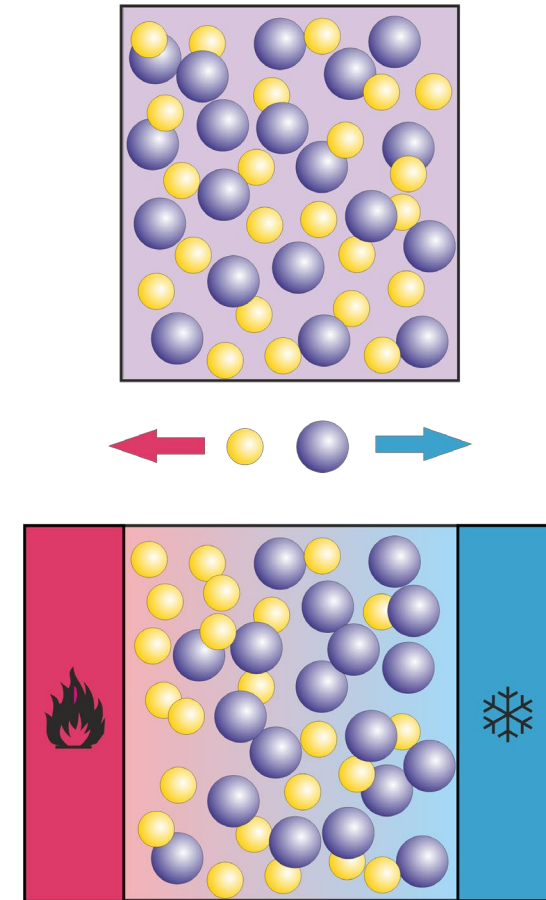
- Motion of solute particles induced by thermal gradient.
- Sensitive tool for probing molecular interactions.

$$\vec{j} = \underbrace{-\rho D \nabla c}_{\text{mixing}} - \underbrace{c(1-c) \rho D_T \nabla T}_{\text{demixing}}$$

Steady state defines Soret coefficient S_T .

$$S_T = \frac{D_T}{D} \propto \frac{\Delta c}{\Delta T}$$

\vec{j}	mass flux
c	concentration
D	diffusion coefficient
D_T	thermodiffusion coefficient
T	temperature
S_T	Soret coefficient



$$S_T = 10^{-3} \text{K}^{-1} \quad \text{molecules} \quad - \quad 1 \text{K}^{-1} \quad \text{colloids}$$

FACTORS INFLUENCING S_T

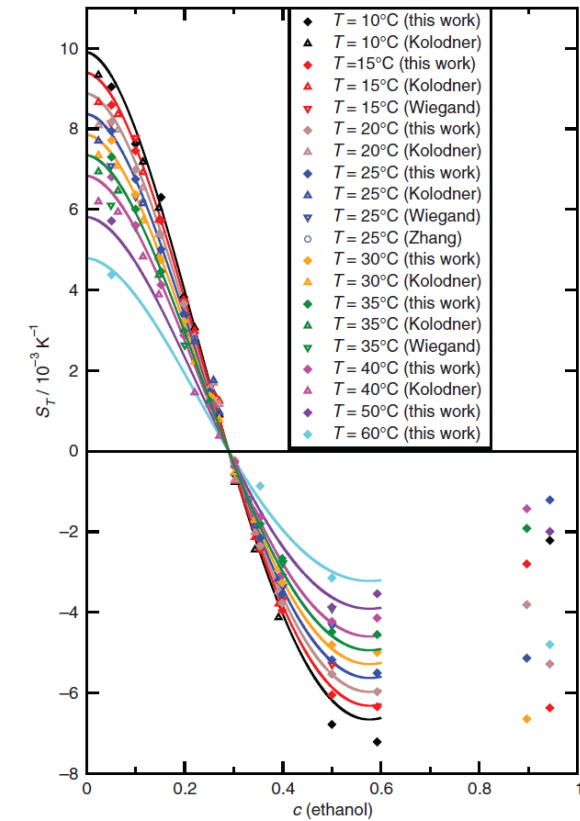
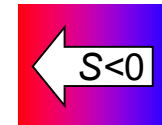
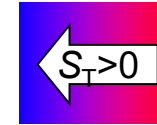
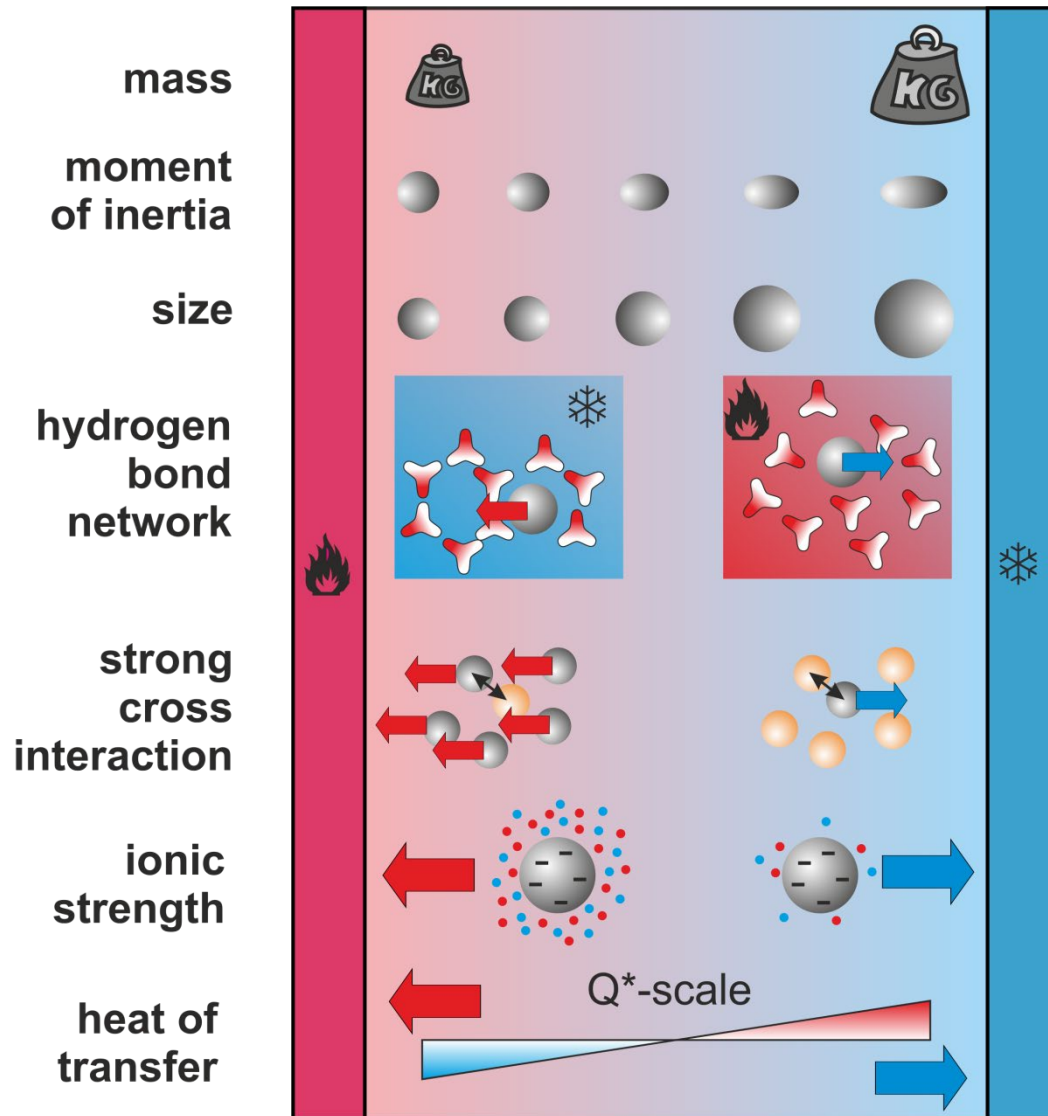


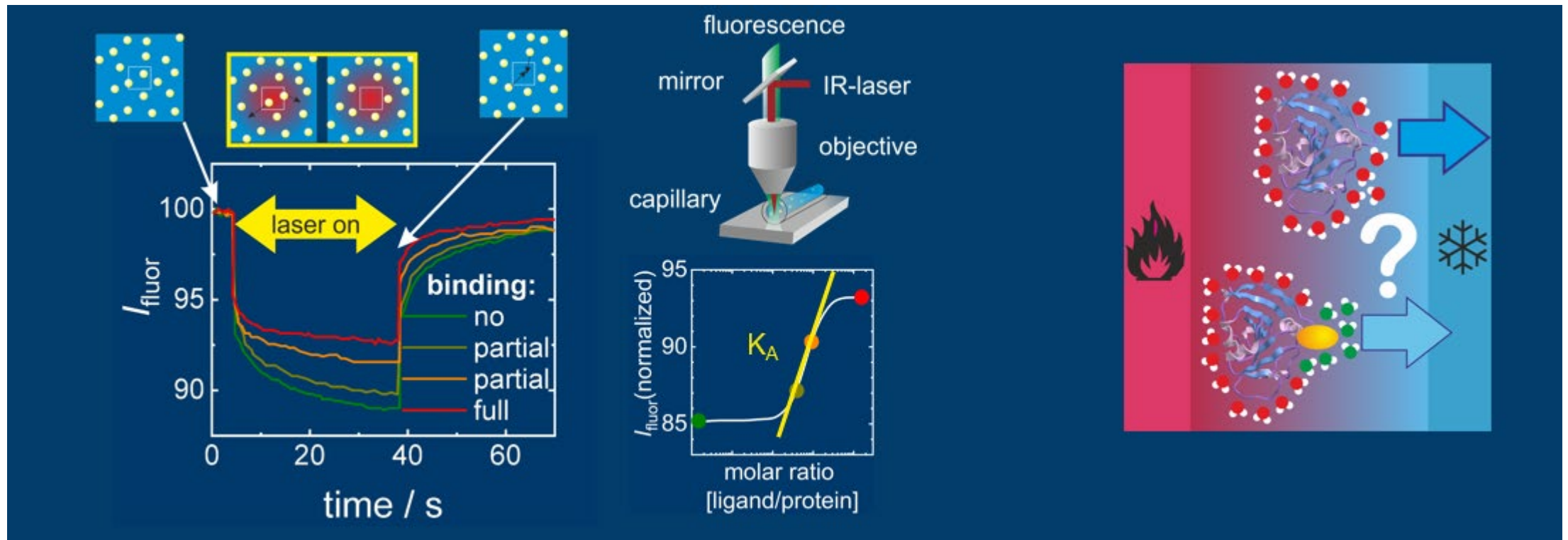
Figure 5. (Colour online). Soret coefficient S_T of ethanol–water as a function of ethanol mass fraction at various temperatures. See text for references.

A. Königer et al. *Philos Mag* **89**, (2009)907–923.

THERMOPHORESIS

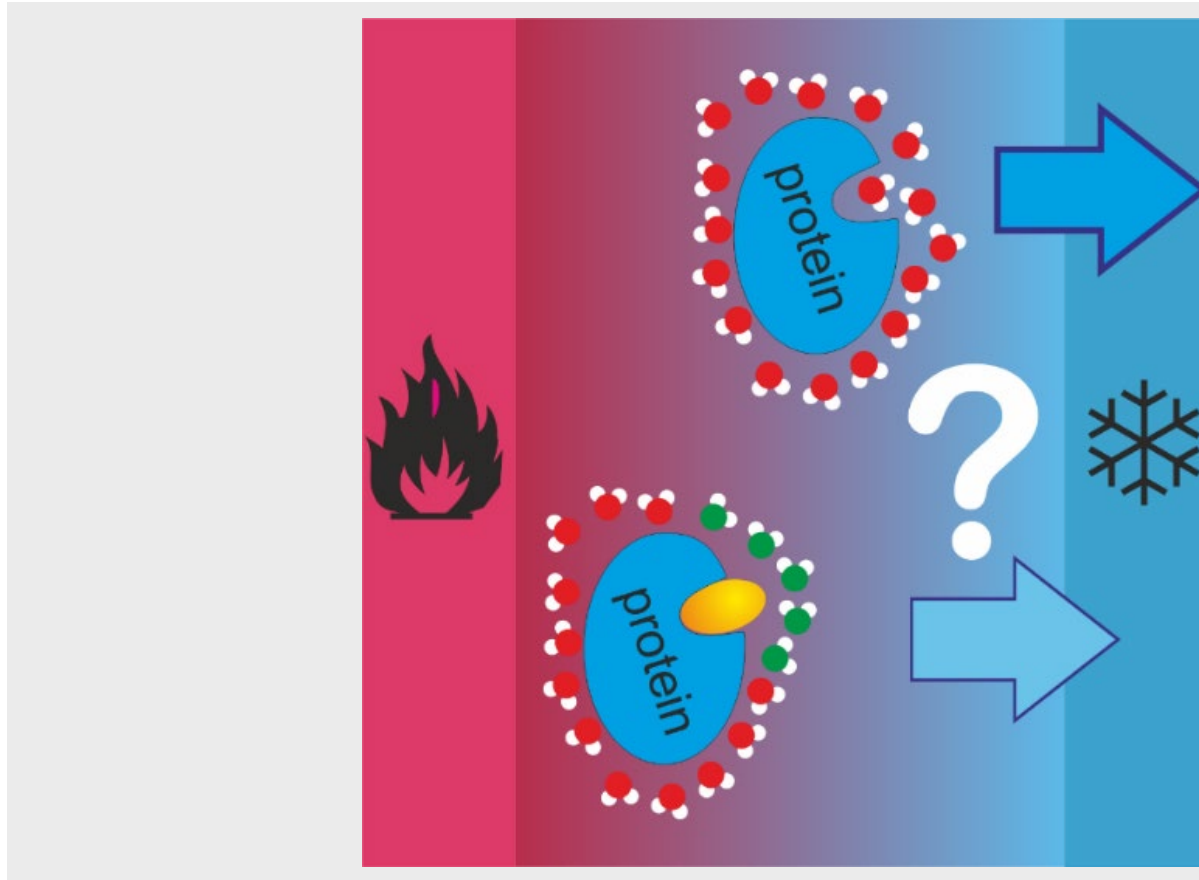
Microscale thermophoresis (MST) □ binding constant K_A

- Monitors protein-ligand interaction



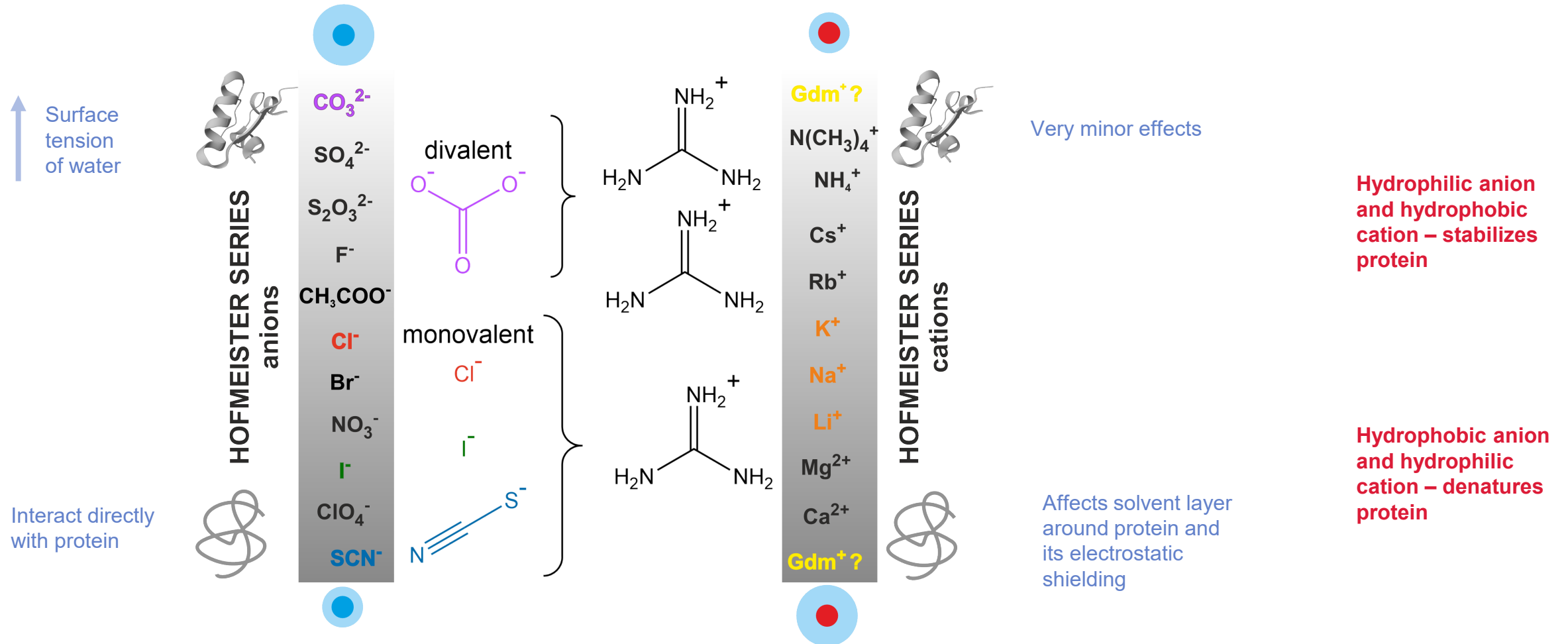
OBJECTIVE

Can we quantify the relation between thermodiffusion and hydration



Hypothesis:
Movement in a temperature
gradient is sensitive to changes
in the hydration layer

WE START WITH A COMPLEX SALT

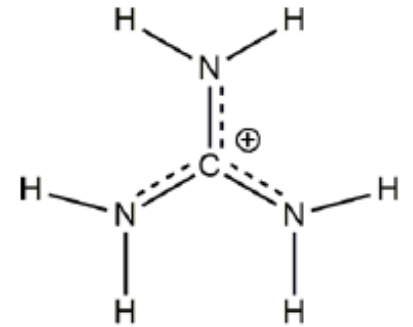


Kunz, W. Curr Opin Colloid In 15, 34–39 (2010).

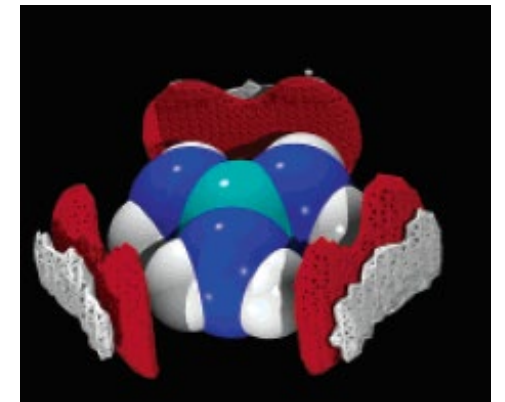
ONGOING STUDY OF GUANIDINIUM

Why guanidinium salts?

- study the hydration of complex molecular species
- highly symmetric and rigid structure
- has a unique disk-shaped structure characterized by flat hydrophobic surfaces and three amine groups that allow directional hydrogen bonding along the edges
- Thus, interact favorably with both water and hydrophobic side chains of protein
- simulation and thermodynamic study of protein folding and denaturation



Guanidinium cation

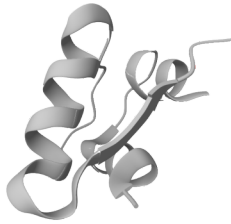


Mason, P. E. *et al. J. Am. Chem. Soc.* **126**, (2004) 11462–11470;

ONGOING STUDY OF GUANIDINIUM

Properties of the four studied guanidinium salts

	pH	M_o (g/mol)	Charge density (anion) C/m ²	Log P	Anion radius (pm)	Hydration enthalpy (kJ/mol)
GdmCO₃	~11.5	180.17	-0.80	-44.6	178	-1395
GdmCl	~5	95.53	-0.39	-11.36	181	-365
GdmI	~7.6	186.98	-0.26	-10.4	220	-290
GdmSCN	~7.6	118.16	-0.25	-19.49	213	-310



HOFMEISTER SERIES anions

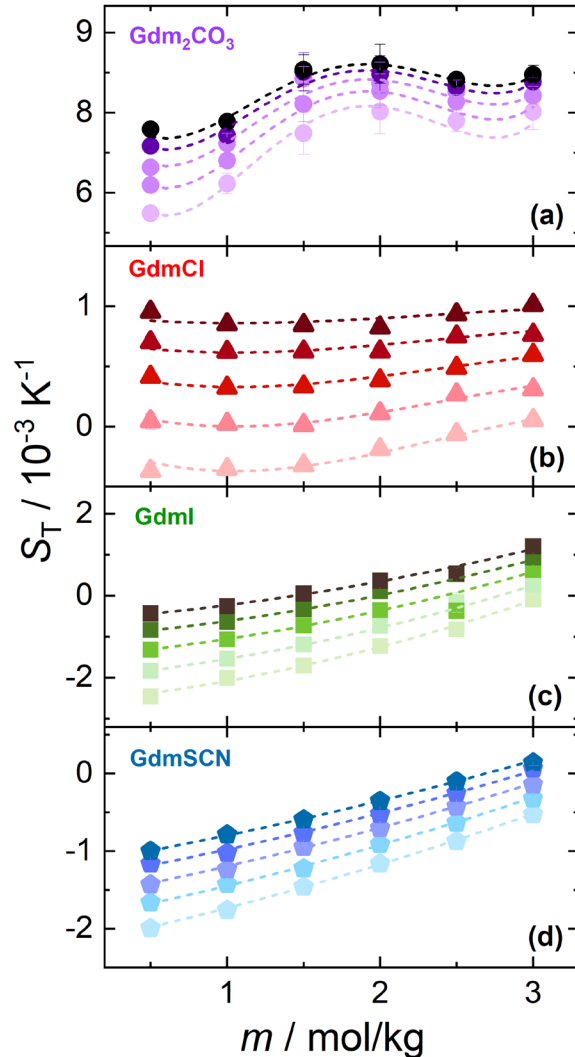



 CO_3^{2-} SO_4^{2-} $\text{S}_2\text{O}_3^{2-}$ F^- CH_3COO^- **Cl^-** Br^- NO_3^- I^- ClO_4^- **SCN^-**

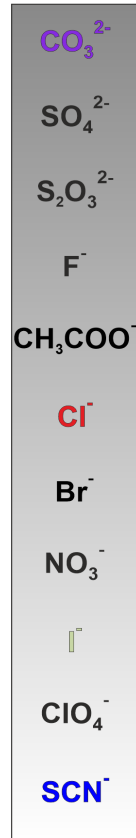

Kunz, W. Curr Opin Colloid In 15, 34–39 (2010).
Radii & hydration enthalpy: Marcus, Y., Biophys Chem 51, 111–127 (1994).

ONGOING STUDY OF GUANIDINIUM

Systematic studies of guanidinium salts (0.5m-3m; 15°C-35°C)



more
hydrophilic



more
hydrophobic

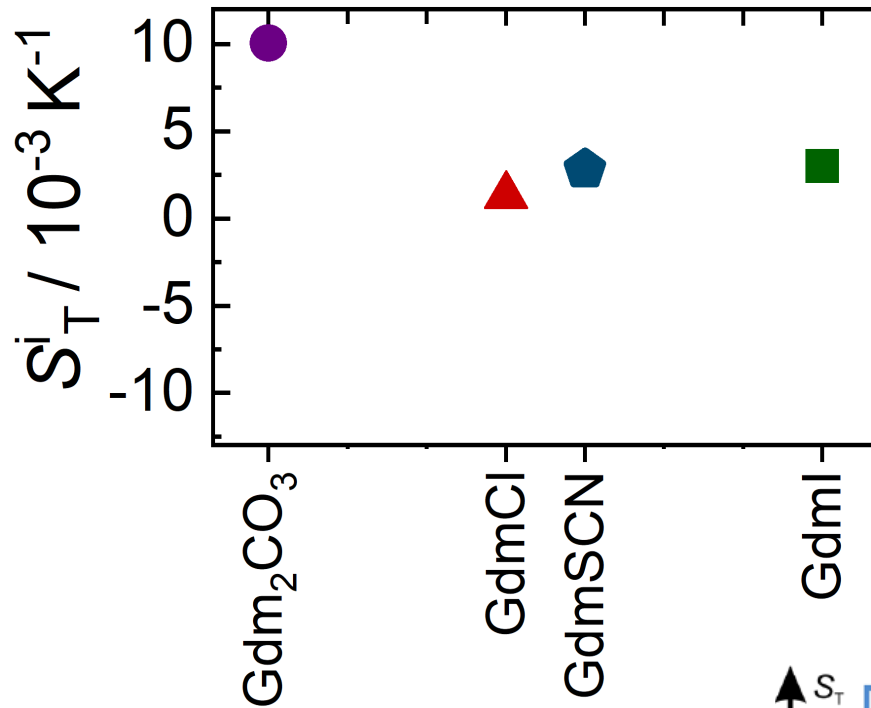
- Gdm_2CO_3 : Most pronounced temperature *and* concentration-dependence
- GdmCl : Minimum in S_T
- GdmI : Sign change in S_T with concentration
- GdmSCN : Negative S_T at most temperatures and concentrations

Fitting: $S_T(m, T) = \alpha(m)\beta(T) + \textcircled{S_T^i}$

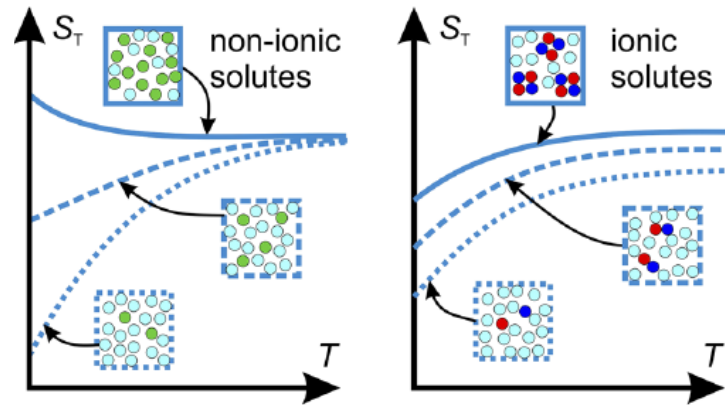
$$\alpha(m) = a_0 + a_1m + a_2m^2 + \dots,$$

$$\beta(T) = 1 + b_1(T - T_0) + b_2(T - T_0)^2 + \dots,$$

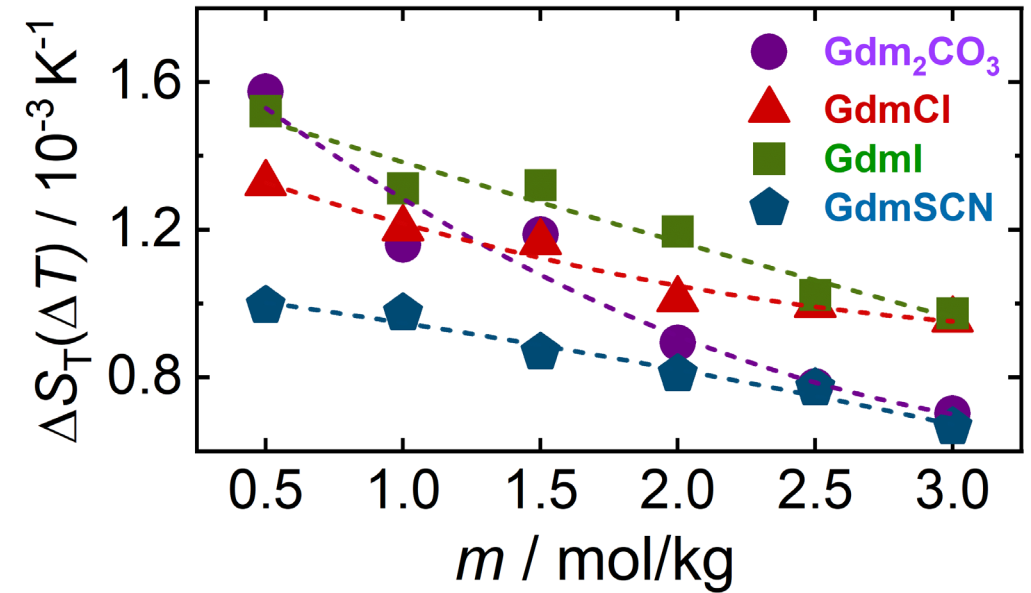
Concentration and temperature dependence



- Anions showed deviation in Hofmeister sequence:

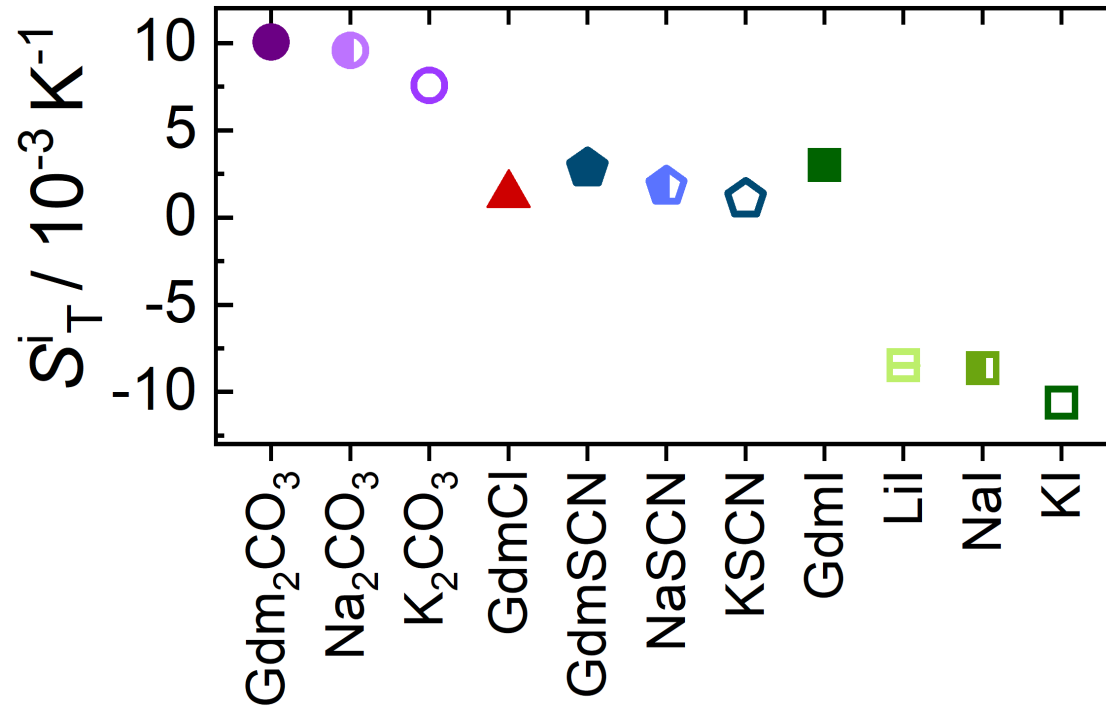


Temperature dependence



- $\Delta S_T(\Delta T) = S_T(35^\circ\text{C}) - S_T(15^\circ\text{C})$
- Order of $\Delta S_T(\Delta T)$ at 0.5 mol/kg:**
 $\text{Gdm}_2\text{CO}_3 > \text{GdmI} > \text{GdmCl} > \text{GdmSCN}$

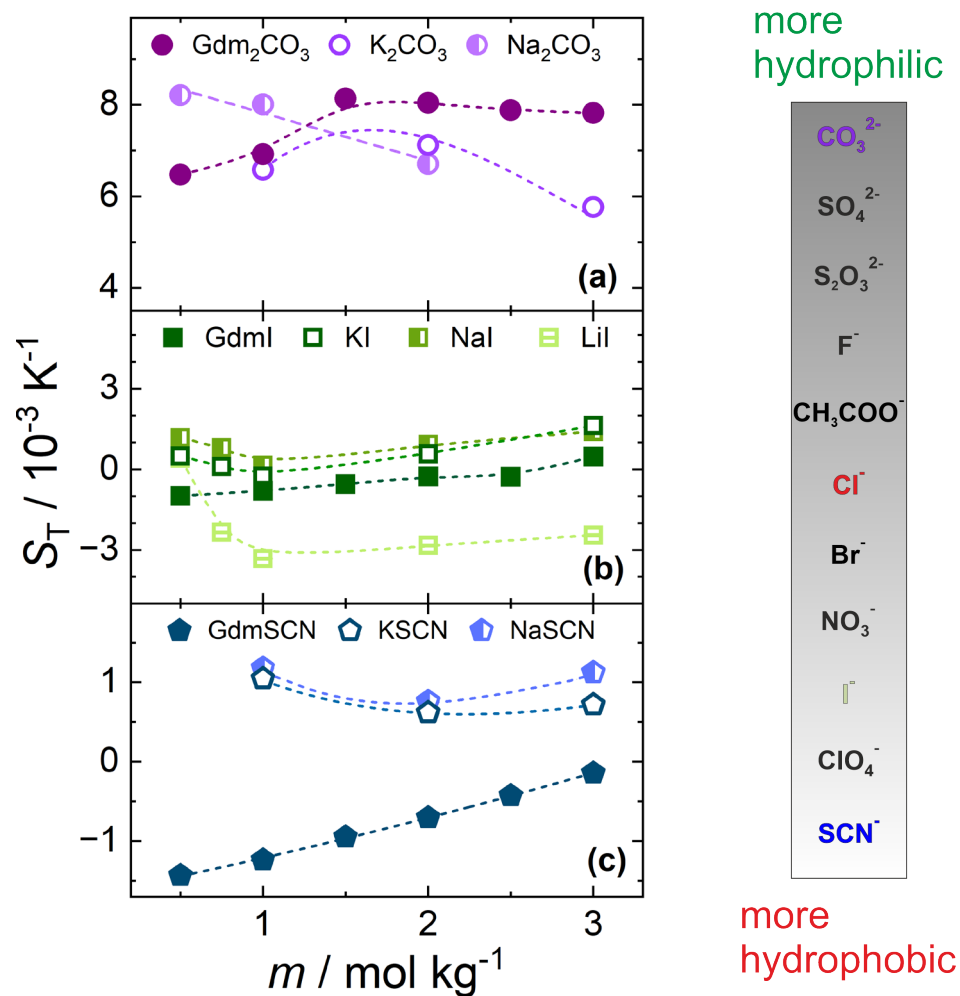
COMPARISON OF s_T^i FOR DIFFERENT SALTS



- Cations followed Hofmeister series: $\text{Gdm}^+ > \text{Li}^+ > \text{Na}^+ > \text{K}^+$
- Anions showed deviation at hydrophobic end: $\text{CO}_3^{2-} > \text{Cl}^- > \text{SCN}^- > \text{I}^-$

ANIONS DETERMINE THE MAGNITUDE OF S_T

Concentration dependence at 25°C (Comparison with previously investigated salts)

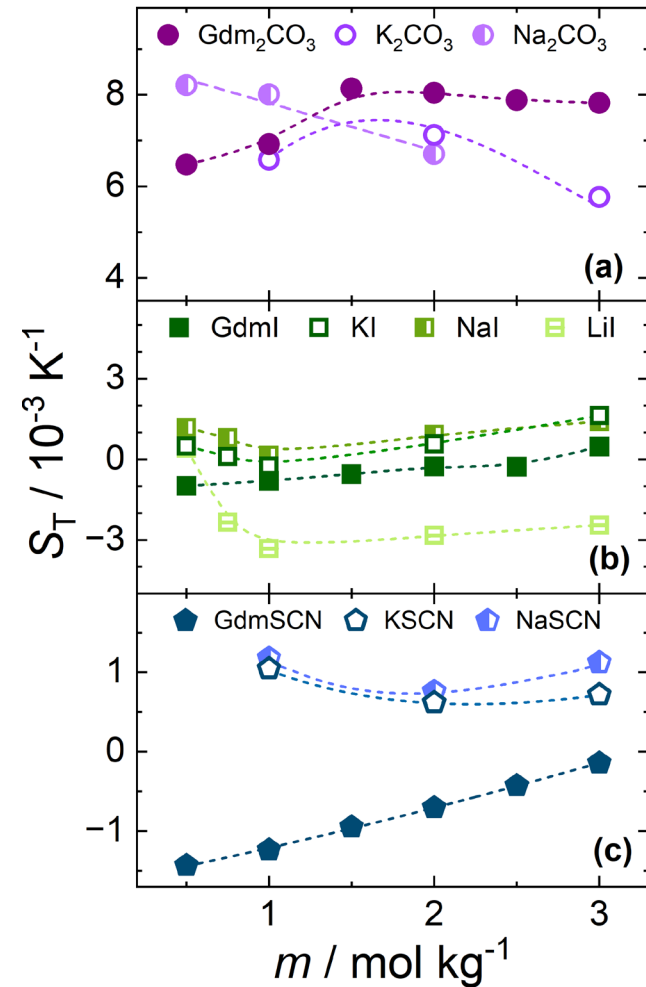


- Increasing the hydrophilicity of anions, increases S_T (follows Hofmeister series)
- Minimum in S_T :
 Gdm_2CO_3 and K_2CO_3 (intricate behaviour)
 KI , NaI , and LiI (1 mol/kg)
 KSCN and NaSCN (2 mol/kg)
- No minimum in S_T :
 Na_2CO_3 (S_T decreases),
 GdmI and GdmSCN (S_T increases)

ANIONS DETERMINE MAGNITUDE OF S_T

Concentration dependence at 25°C (Comparison with previously investigated salts)

WHY?



Anions are better hydrated than cations. Thermodiffusion is strongly related to hydration.

“For a cation and an anion with a same radius, the anion has stronger hydration ability.” (MD simulation)

J. Zhou et al., Fluid Phase Equilibr. **257**(2002) 194-197.

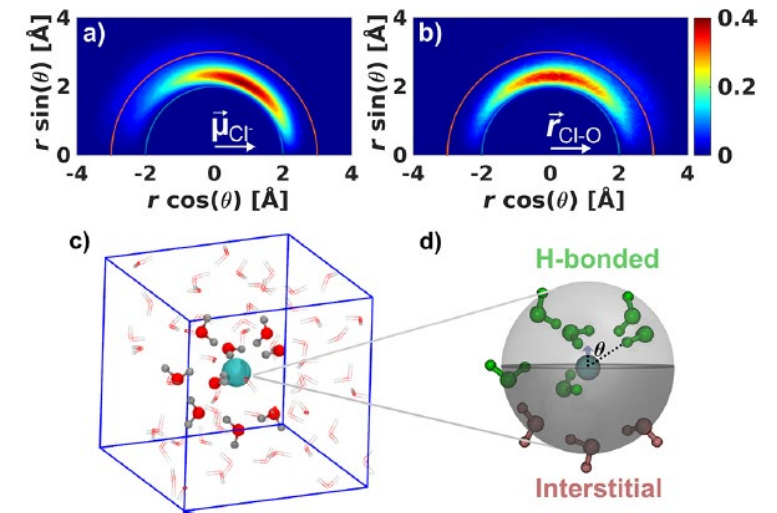
“In general, the cations’ effects on the molecular organization of liquid H_2O have been recognized to be weaker than those of anions..” (partial molar enthalpy)

T. Morita et al., J. Phys. Chem. B **118** (2014) 8744.

“...indicating that the anions are more strongly hydrated than the cations ...” (chromatography)

K. D. Collins, PNAS **92** (1995) 5553.

Dipole moment distributions around anions shows an asymmetry, which is not observed for cations



Jindal, A et al. J. Phys. Chem. Lett. **15**, (2024) 3037–3042

No microscopic picture yet how those differences influence the thermophoretic behaviour?

CONCLUSIONS

- Structural changes in the solvent, in particular the hydrogen bonding network influence the thermophoretic behavior.
- Thermophoretic behavior of salts is majorly influenced by changes of the anion. **The reason is not clear yet (ongoing).**
- S_T of several salts shows a minimum with concentration.
- S_T^i of various salts does not always follow the Hofmeister series.

OUTLOOK

1. Organic salts – first experiments completed



- Analysing experimental data and interpreting (manuscript JCP in preparation)
- additional experiments of ammonium salts (Gohlke)

2. Salts + chelating agents (TDFRS+ITC)

3. Proteins (TDFRS, ITC and QENS) cooperation A. Stadler (JCNS-1)

Thanks