



# Interactions in dopamine and indole loaded thermosensitive hydrogels seen by high sensitivity microDSC. Implications for drug delivery

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Received: 31 January 2022 / Accepted: 26 April 2022 / Published online: 3 June 2022  
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## Abstract

Controlled and targeted drug delivery systems consist of a carrier matrix and one or more active ingredients. One of the roles of the matrix is to regulate the release of the drug. Uptake, release, differential scanning microcalorimetry (DSC) and powder X-ray diffraction (XRD) techniques were used to reveal the interactions governing the release of dopamine and indole from a thermoresponsive model polymer gel. The dopamine can be completely recovered from the loaded polymer matrix. The self-assembling affinity of the dopamine molecules reduces their interaction with the polymer, and the water molecules can form a uniform protecting water sheath. Thus neither the kinetics nor the temperature of the phase transition of the carrier matrix are influenced by the dopamine. The DSC results imply the formation of polymer and dopamine-rich domains above the phase transition. Indole, on the other hand, readily substitutes for the water molecules through the interaction between the C=O sites of the polymer and the NH groups of the drug. The loss of the protecting hydrophobic water and the decelerated fluctuation of the indole decorated polymer chains result in a much slower phase transition and a depleted phase transition temperature. The interaction between the carrier matrix and the indole results in a uniform distribution of the drug and after drying the indole is found in amorphous form. Dopamine, on the contrary, forms crystalline regions.

**Keywords** Responsive polymers · Poly(n-isopropylacrylamide) gel · Dopamine · Indole · Calorimetry · Host · Guest interactions

## Introduction

The increasing demand for personalized medical care has brought drug formulation into the focus of research. Controlled and targeted drug delivery systems are among the fastest developing research areas. Such systems consist of a

carrier matrix and an active ingredient. The main role of the matrix is to bring the active ingredient to reach the desired temporal and/or spatial distribution in the body, to protect it from external influences, and to regulate its release. The active ingredient can be discharged according to a predetermined release profile, in a certain amount and at a certain time. These systems can be used to ensure long-term, constant concentration of therapeutic agents, to optimize therapy, and to avoid side effects.

Polymers, particularly hydrogels, are frequently employed delivery matrices. Temperature triggered targeted delivery studies are often modelled with poly(N-isopropylacrylamide) (PNIPA) hydrogels. Its special reputation is due to the fact that it shows a nonlinear volume phase transition with a lower critical solution temperature ( $T_{VPT}$ ) around 34 °C, i.e. close to the human body temperature. Its use as a controlled drug release model vehicle is also justified by its reversible deformability and liquid absorption capacity. There are various means to tune both the temperature and the rate of the phase transition

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in such systems. The interactions formed in the drug—gel matrix—swelling liquid system are among the factors to be considered in the development of drug delivery systems. Their nature and the strength of the interaction have a fundamental effect on the kinetics and the efficiency of controlled delivery. As the phase transition is related to the polymer—solvent interaction, both the matrix and the solvent quality may participate in the tuning. Therefore, in a wider sense any change in the matrix and/or in the swelling medium may influence the conditions and kinetics of the phase transition. Combined nano- and macroscale studies on carbon nanotube and graphene oxide doped PNIPa nanocomposites swollen in water revealed that the kinetics of the temperature response in these carbon nanoparticle (CNP) doped systems strongly depends on the morphology, the hydrophilic/hydrophobic character and the concentration of the CNPs. Interestingly, it is also affected by the temperature gradient inducing the phase transition [1].

In hydrogels the influence of the surrounding liquid can be tuned through the nature and the concentration of auxiliary compounds, including inorganic salts or buffers [2]. Last but not least, the drug to be delivered may also affect the phase transition. It has been observed that addition of small guest molecules, even at low concentration, noticeably influences the transition behaviour of the PNIPa hydrogel. Several additives, e.g. inorganic salts, phenols and benzene derivatives (salicylaldehyde, 3,4-dimethoxybenzaldehyde, hydroxy-benzaldehyde, ethylvanillin, benzoic acid, methyl-*p*-hydroxybenzoate), saccharide and organic solvents (methanol, ethanol, dimethyl sulfoxide) were found to reduce the  $T_{VPT}$  of PNIPa. The transition temperature can be increased by the addition of surfactants (e.g. sodium dodecyl sulphate) or organic quaternary ammonium salts [3–6]. Second-order interactions between the gel matrix and the drug molecules have a fundamental effect on the structure and properties of the system. In a systematic study, it was demonstrated how the number and location of the hydroxyl groups in different phenolic derivatives affect the swelling properties and  $T_{VPT}$  of this polymer gel [7–10]. Although the  $T_{VPT}$  shift depends on the structure and the concentration of the additive, no correlation was found with hydrophobicity or solubility, suggesting that specific additive–polymer interactions may be the major factors in controlling the  $T_{VPT}$ .

Understanding the mechanism of all these complex and often interconnected effects is unavoidable for the design of enhanced and tailor-made drug delivery systems. It is well known that drugs, generally having low water solubility, are preferentially delivered in amorphous form. The matrix applied in the formulation may play an active role in the amorphization. Phenol, as an example, dries in amorphous form when embedded into a PNIPa matrix, as their strong interaction prevents development of phenol crystals during

slow removal of the water [10]. On the other hand, the same effect blocks the full release of the phenol after re-swelling [11].

Differential scanning calorimetry (DSC) is among the most frequently applied techniques for drug delivery studies in thermally sensitive systems [12–15]. Recent improvements in the instrumentation including enhancement of detection sensitivity and the continuous development of the software justify the still-growing attention. The interactions between the gel matrix and the drug can be successfully characterized by changing the thermal properties of the matrix, which can be easily followed by calorimetry. The most obvious parameters obtained are the temperature of the phase transition and the heat involved in the overall process. With a more careful and thorough evaluation of the response signal, however, we can go beyond these primary data in order to extract more information about the complex processes, particularly if systematic studies are performed.

In this work, we demonstrate how high sensitivity DSC measurements can provide valuable information about the thermodynamics of the interactions governing the phase transitions in ternary delivery systems using two small drug molecules, dopamine and indole. The release profile of these two molecules, as well as their influence on the phase transition behaviour of the model gel matrix are fundamentally different.

Dopamine (3,4-dihydroxyphenethylamine), a chemical released by nerve cells, acts as a neurotransmitter in the brain. Outside the central nervous system it serves as a local messenger in cellular communication. In therapy it is used as a stimulant drug in the treatment of severe low blood pressure, slow heart rate, and cardiac arrest.

Indole is widely distributed in the natural environment and can be produced by a variety of bacteria. In the human body indole is a result of tryptophan metabolism. It is one of the platform molecules in modern drug discovery, thanks to its broad-spectrum biological activity. Nowadays, indoles and their derivatives are mainly studied for their analgesic and anti-inflammatory effect, against tuberculosis, malaria, diabetes, certain bacterial infections, and even viruses.

In this paper we demonstrate how high sensitivity scanning calorimetric measurements can contribute to the understanding of drug–polymer matrix interactions and support the development of drug delivery systems. We focus on the analysis of the DSC response curves of a thermosensitive gel matrix loaded with the two biologically active molecules. By decomposing the phase transition signals we attempt to reveal the mechanism and to distinguish sub-processes occurring during the phase transition.

## Materials and methods

### Materials

Poly(N-isopropyl acrylamide) (PNIPA) gel films with thickness of 2 mm were prepared in aqueous phase with the radical polymerization of N-isopropyl acrylamide (NIPA, Tokyo Chemical Industry Co., LTD., Tokyo, Japan) and N,N'-methylene bis(acrylamide) (BA, Sigma-Aldrich) in aqueous medium with nominal molar ratio of [NIPA]/[BA] = 150 at 20 °C by free radical polymerization. The reaction was initiated by ammonium persulphate (APS, Sigma-Aldrich) and N,N,N',N' tetramethylethylenediamine (TEMED, Fluka). All chemicals were used as received except NIPA. High purity NIPA was obtained from the purchased material after recrystallization from toluene-hexane mixture [8]. The films were dialyzed in water to remove unreacted chemicals, cut into discs of diameter 5 mm, air-dried and stored in a desiccator above sulphuric acid. A detailed characterisation of the swelling properties of this gel in water is reported elsewhere [16]. Dopamine hydrochloride (98%) and indole were purchased from Sigma-Aldrich, Their physico-chemical properties are listed in Table 1.

### Methods

#### Swelling and uptake

Swelling measurements were carried out by equilibrating dry PNIPA gel disks with excess aqueous solutions for one week at  $20.0 \pm 0.2$  °C. The ratio of the liquid phase volume and the dry gel mass was 83. The swelling ratio at equilibrium was calculated from the mass balance as

$$\text{Swelling ratio} = \frac{m_{\text{gel, swollen}}}{m_{\text{gel, dry}}} \quad (1)$$

where  $m_{\text{gel, dry}}$  and  $m_{\text{gel, swollen}}$  are the mass of the dry and the equilibrated gel disks, respectively.

The swelling degree in water was used as a reference when the relative swelling degree was calculated. The uptake of the guest molecules  $n_a$  ( $\text{mmol g}_{\text{dry gel}}^{-1}$ ), was determined from the initial ( $c_0$ ) and equilibrium concentrations ( $c_e$ ) of the swelling liquid as

$$n_a = \frac{c_0 V_0 - c_e V_e}{m_0} \quad (2)$$

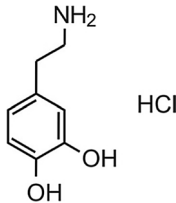
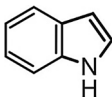
where  $c_0$  and  $V_0$  are the initial concentration and volume of the swelling medium, and  $c_e$  and  $V_e$  their corresponding values when the gel is at equilibrium with the drug containing medium. Concentrations were measured with UV-Vis spectrophotometry. The concentration of the active substance within the gel matrix was calculated as

$$c_{\text{gel}} = \frac{c_0 V_0 - c_e V_e}{V_0 - V_e} \quad (3)$$

#### Release experiments

The rate of drug release was examined with a Cary 60 UV-Vis spectrophotometer (Agilent Technologies) at  $\lambda_{\text{max}}$  given in Table 1. The gels were equilibrated in 35, 70, and 500 mM dopamine and 5, 15, and 20 mM indole solutions, respectively. The loaded gel discs, dried in a desiccator to constant mass (ca 0.03.g), were placed into 45 mL distilled water and the drug release was monitored through the absorbance of the solution for 12 h at 25 °C with constant stirring. The release curves were derived from the mass balance.

**Table 1** Selected properties of dopamine and indole

Property	Dopamine hydrochloride (3,4-Dihydroxy-phenethylamine hydrochloride)	Indole (1H-Indole)
Structure		
Chemical formula	$\text{C}_8\text{H}_{11}\text{NO}_2 \cdot \text{HCl}$	$\text{C}_8\text{H}_7\text{N}$
Molecular weight/ $\text{g mol}^{-1}$	189.64	117.15
Solubility/ $\text{g L}^{-1}$	600 (20 °C) [17]	3.5 (25 °C) [20]
$\text{pK}_a$ (25 °C)	9.0, 10.6 and 12.1 [18]	16.2 [21]
logP	-0.98 [19]	-2.4 [22]
$\lambda_{\text{max}}/\text{nm}$	280	270

### High sensitivity differential scanning calorimetry (microDSC)

Scanning microcalorimetry measurements were performed on a MicroDSCIII or a MicroSC 1A apparatus (both from SETARAM). About 5–10 mg of dry powdered gel sample was immersed into 500  $\mu\text{L}$  aqueous solution of the test molecule. After 2 h incubation at constant temperature the samples were heated with a ramp rate of  $0.02\text{ }^\circ\text{C min}^{-1}$  or  $0.05\text{ }^\circ\text{C min}^{-1}$  for the dopamine (D) and indole (I) series, respectively.

The onset temperature of the phase transition was defined as the deviation of the baseline and the response signal. The onset temperature of the peak was taken as  $T_{\text{VPT}}$ . The temperature corresponding to the peak maximum, the width of the peak at half maximum and the peak area related to the heat of the phase transition were also determined from the recorded DSC curves (Fig. 1). Evaluation of the peaks including the deconvolution was performed with the of the SETARAM software (Data Processing by AKTS).

Owing to the low scanning rate the heat effect can be considered as the overall enthalpy  $\Delta H$  of the phase transition. Its error is about 1–5% ( $\sim 3\text{ J g}^{-1}$ ). The impact of the drug molecule on the overall entropy of the phase transition ( $\Delta S_d$ ) was estimated as

$$\Delta S_d = \frac{\Delta H_d - \Delta H_{\text{water}}}{T} \quad (4)$$

where  $\Delta H_d$  and  $\Delta H_{\text{water}}$  refer to the overall enthalpy measured with the corresponding drug solution and pure water, respectively.

### Powder X-ray diffraction (XRD)

Dry gel disks were equilibrated in dopamine hydrochloride and indole solutions with initial concentrations 500 mM and 20 mM, respectively. After drying at room the diffractograms were measured on a PANanalytical X'pert Pro MPD multi-purpose powder X-ray diffractometer.

## Results and discussion

### Drug uptake

The swelling behaviour of the hydrogels in the aqueous indole and dopamine solutions is different. While the transparent gel in pure water gradually turns opaque and later white with increasing indole concentration (Fig. 2), no such change was observed, even in 1000 mM dopamine solution at  $20\text{ }^\circ\text{C}$ . In dopamine the relative swelling degree is well in excess of 1, and is practically not affected further even at 500 mM concentration. By contrast, the gel swollen in indole solutions exhibited moderate shrinkage at lower concentrations, and around 20 mM an abrupt, concentration-triggered volume phase transition occurred (Fig. 3). That is, at or above this concentration, indole reduces the temperature of the volume phase transition from ca.  $34\text{--}20\text{ }^\circ\text{C}$ . Within the pH range determined by the  $\text{p}K_a$  values of the corresponding drugs the swelling degree of the gel and the onset temperature of the phase transition are not influenced by pH [2].

To obtain the uptake isotherms the hydrogels were equilibrated in the aqueous solutions of the drug molecules of different initial concentrations (Fig. 4a). The initial region of both equilibrium uptake isotherms is linear and the Henry constants are similar,  $0.25$  and  $0.28\text{ L g}^{-1}$  for the dopamine and indole, respectively. On approaching the critical phase transition concentration, ca 16 mM,

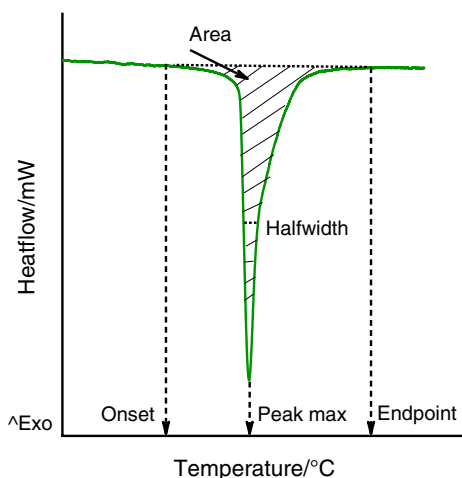


Fig. 1 Primary parameters derived from the DSC response

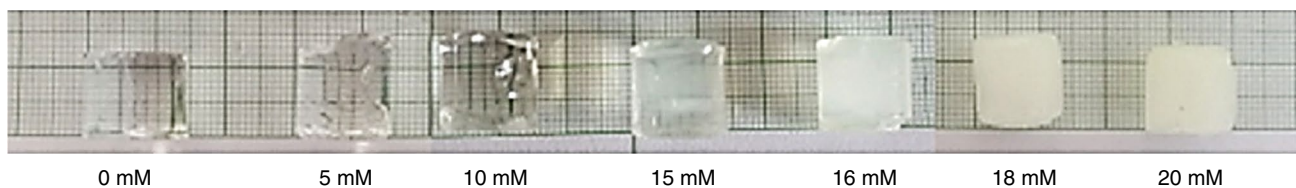
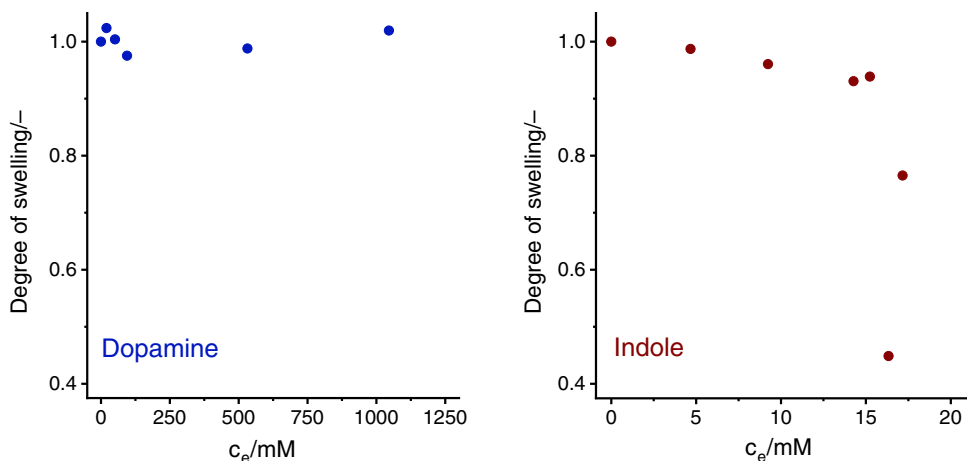
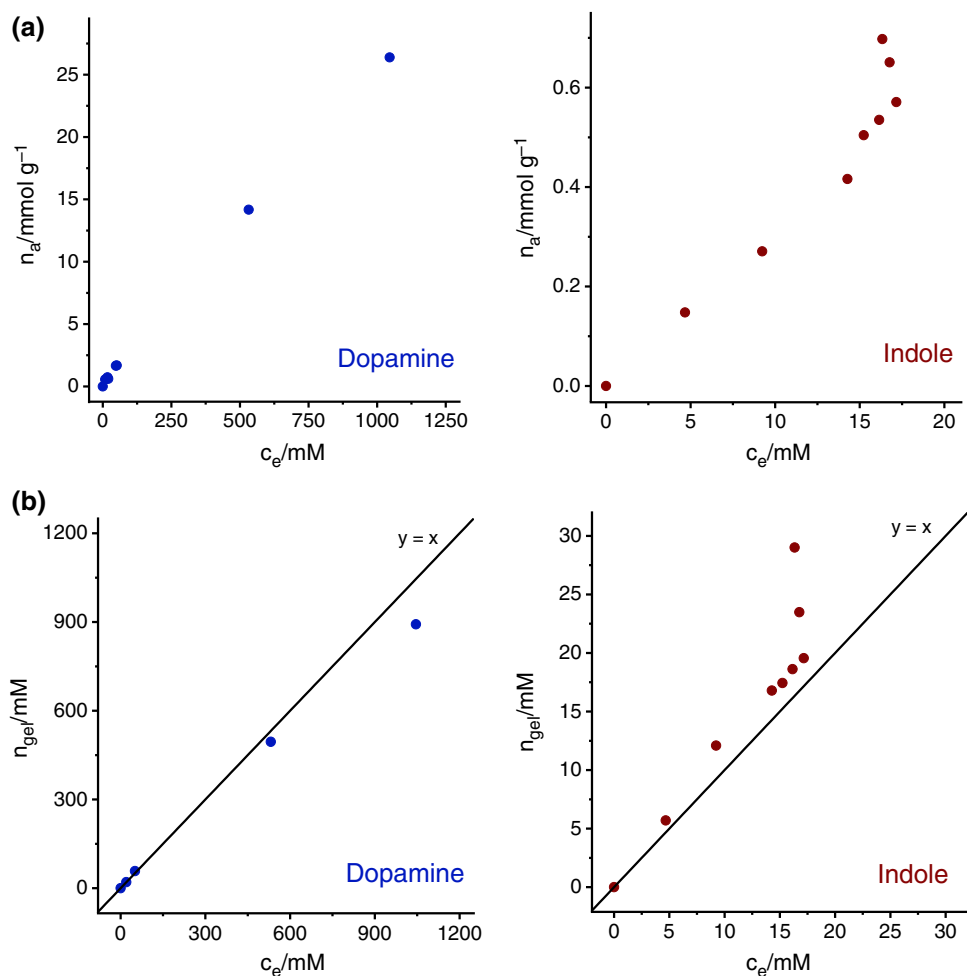


Fig. 2 PNIPA gel cylinders swollen in indole solution of different initial concentration at  $20\text{ }^\circ\text{C}$

**Fig. 3** Swelling of PNIPAM hydrogel at equilibrium in aqueous dopamine and indole solutions at 20 °C



**Fig. 4** Uptake **a** and distribution **b** of the drugs in equilibrium at 20 °C



the shape of the indole isotherm shows an upturn. No such feature is seen in the dopamine isotherm. The distribution curves of the two drugs between the gel and aqueous phases in Fig. 4b also imply that the two probe molecules behave differently in the polymer gel. The concentration of dopamine is slightly depleted in the gel phase over the

whole concentration range covered, while the indole first shows a slight then later a significant enrichment within the gel phase.

## Release kinetics at 25 °C

The release kinetics of the drug molecules was studied from gel samples fully dried after loading. According to the release curves in Fig. 5, practically 100% of the dopamine can be recovered at any loading concentration. The shape of the response curves however indicates that the kinetics is slightly slower when the concentration of the loading medium was 500 mM. But only a limited part, 10–20%, of the indole was released, hindered by the loading concentration. The recovery from freshly loaded swollen gels, however, was complete. This behaviour implies that the two drugs interact with the polymer in a different way when the solvent is removed.

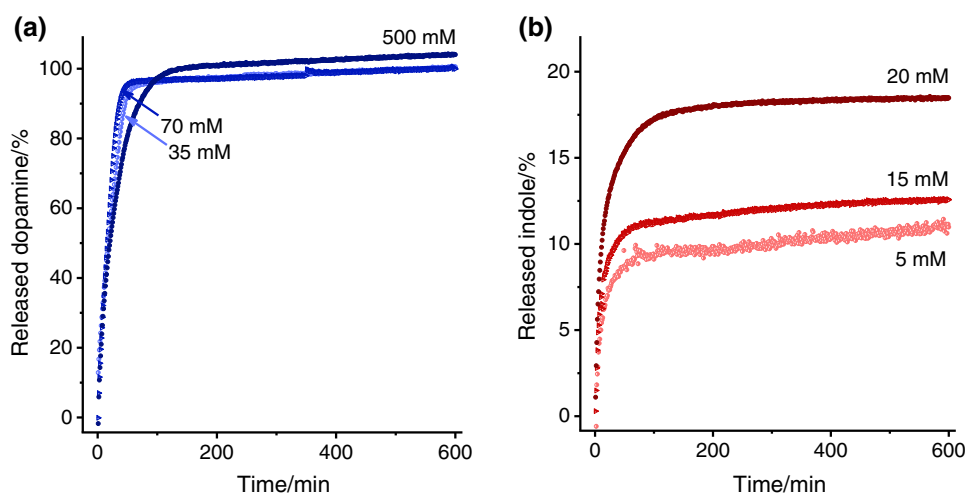
## Differential scanning microcalorimetry (DSC)

The deswelling process was also monitored by microcalorimetric measurements using unusually slow scanning rates.

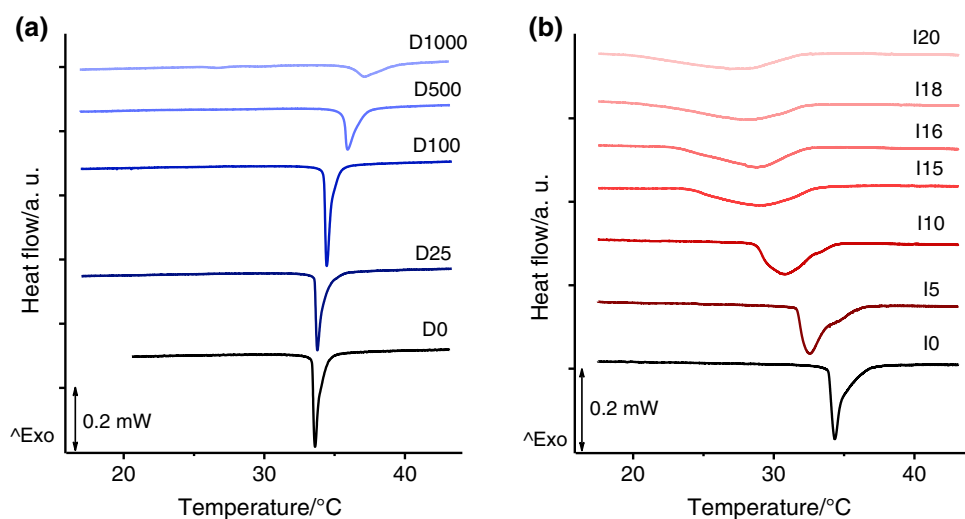
Due to the high sensitivity of the method the dissimilarities of the two systems mentioned earlier are clearly reflected by the sets of response curves recorded during swelling of the dry PNIPA samples in the various media (Fig. 6). The samples are denoted by the initial of the drug molecule (D for dopamine or I for indole) and the concentration of the swelling medium in mM, e.g. I10 is the sample swollen in a 10 mM aqueous indole solution. The scanning program was launched after the samples were equilibrated in the swelling medium applied in abundant excess. Direct comparison of any numerical values from the two series of curves, however, must be viewed with caution as the two sets of curves were recorded with different scanning rates. That is why the response curves of the unloaded samples are also shown for both sets.

The response curves reveal the difference of influence of the two probe molecules with much greater sensitivity than in the gravimetric swelling experiments. Dopamine displays sharp peaks, very similar to that of water. Only a slight

**Fig. 5** Drug release profiles determined at different drug concentrations at 25 °C; **a** dopamine; **b** indole



**Fig. 6** DSC responses of the PNIPA—dopamine ( $dT/dt = 0.02 \text{ } ^\circ\text{C min}^{-1}$ ) **a** and the PNIPA—indole ( $dT/dt = 0.05 \text{ } ^\circ\text{C min}^{-1}$ ) **b** systems. D and I stand for dopamine and indole, respectively, while the numbers refer to the concentration of the drug in the swelling medium in mM. Curves are shifted vertically for clarity





increase is detected in  $T_{VPT}$  (Fig. 6a, Table 2): the onset and the maximum temperature of the peak exhibit a modest but systematic upward shift, with limited peak broadening. The latter becomes obvious only from 500 mM on.

The effect of indole is much more pronounced (Fig. 6b, Table 2). It produces a marked and systematic decrease in the characteristic temperatures and substantial peak broadening. The gap between the onset and the peak maximum increases gradually and significantly.

In the case of dopamine no systematic change in the heat occurs at lower concentrations, but at 500 mM, and even more so at 1000 mM, the heat recorded becomes less endothermic, which implies considerable additional exothermic effects. In contrast, the heat of transition increases moderately (and linearly) with indole concentration, implying that indole causes further endothermic processes during the phase transition.

## Discussion

The total release of dopamine and indole from the freshly swollen gels indicates that any interaction between the small molecules and the gel can be excluded. Even the gel swollen in a 20 mM indole solution yielded a 100% release. While the dopamine is fully released at any concentration, even from the filled and then dried samples, the indole loaded gels, once kept in the desiccator, withhold the majority of the stored indole, e.g. the gel swollen in 5 mM indole releases about 11% of the stored drug.

To gain more information about the mechanism occurring during the temperature induced phase transition of the equilibrated gels a deeper analysis of the DSC curves was performed (Figs. 7 and 8, Tables 3 and 4). Deconvolution

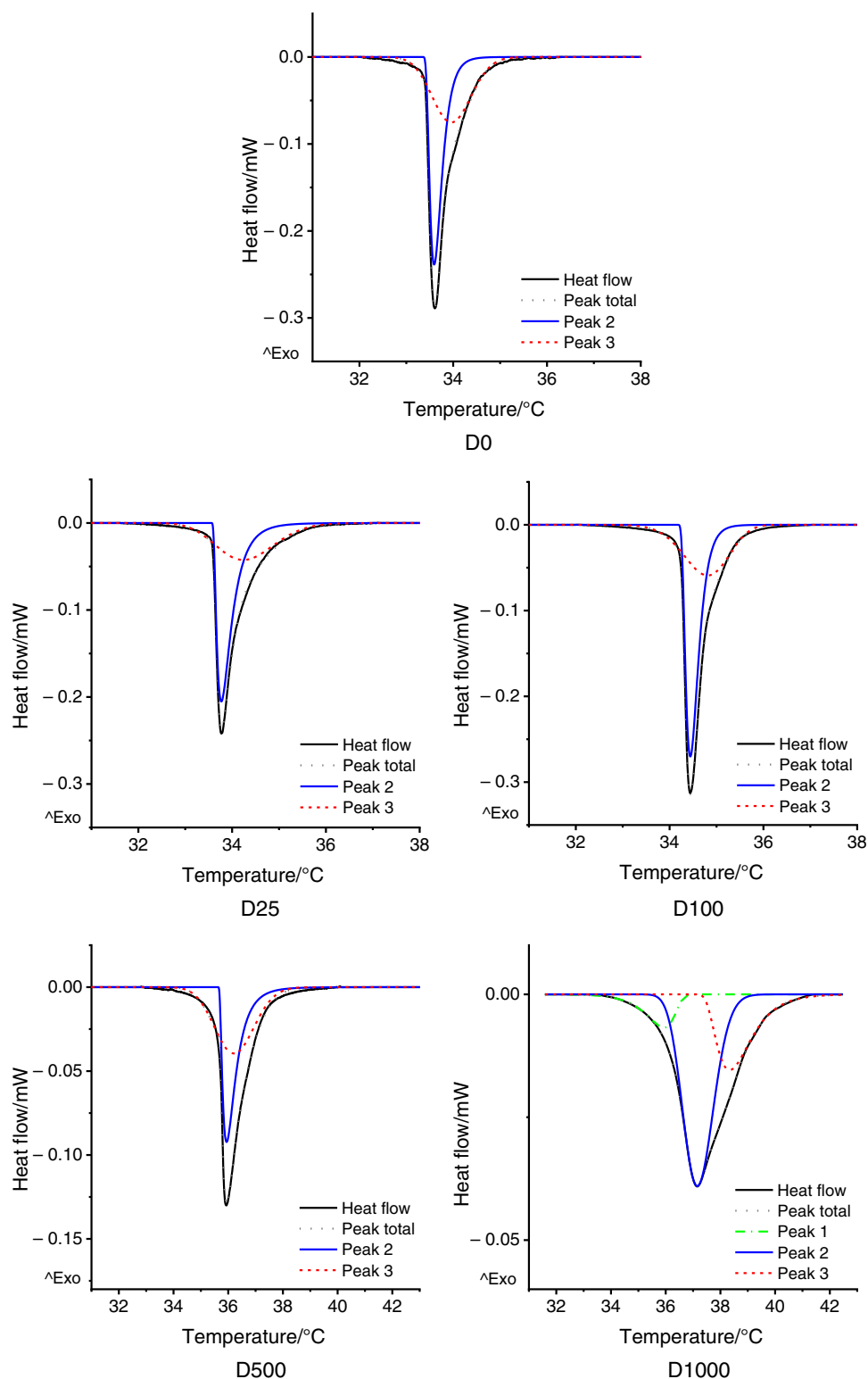
makes it possible to distinguish between effects having sufficiently different time constants.

In pure water the equilibrated swollen polymer chains are decorated with a sheath of water. The space between them is filled with “confined” water while the rest of the water molecules are “free” [23]. When this gel sample is slowly heated a complex series of processes occurs. On reaching the phase transition temperature the water layer that adheres by hydrophobic interaction is released, thereby allowing the naked polymer chains to assemble. The syneresis triggered in this way expels the water and prevents the stiffened polymer chains from relaxing, a process often called jamming (we are much below the glass transition temperature of the gel) [16]. According to the DSC signals the first step is fast and results in a pulse-like endothermic heat release. The following slower process corresponds to the relaxation of the polymer chains and intrinsically involves dissipation of the heat. The fast and slow processes can be distinguished in the deconvolution of the DSC signal of the samples swollen in pure water (Fig. 7, D0; Fig. 8, I0). The comparison of the response curves of these otherwise identical samples reveals the significance of the slow heating rate during the DSC studies. The shape of the two fitted curves, e.g. the half widths, clearly reflect the kinetics of the abrupt deswelling and the slower relaxation. It was found earlier that in such systems width of the broader DSC response stems from the structural complexity of the relaxation [9]. In the case of dopamine the response curves can be similarly fitted by two curves of almost equal contribution up to 500 mM (Fig. 7, D25-D500). The signal of the 1000 mM sample, however, is broadened. To achieve a satisfactory fit a third peak is required at the beginning of the decelerated initial section of the phase transition, implying that the multiple processes in the “fast” region split into two sets at high dopamine

**Table 2** Parameters of DSC response curves of PNIPA gel in different dopamine and indole solutions

	Symbol	Onset/ $^{\circ}\text{C}$	Peak max/ $^{\circ}\text{C}$	Full width of the peak/ $^{\circ}\text{C}$	$\Delta H_d/\text{J g}^{-1}$	$\Delta S_d/\text{mJ g}^{-1} \text{K}^{-1}$
Dopamine, $0.02 \text{ }^{\circ}\text{C min}^{-1}$	D0 (water)	32.1	33.6	3.7	57	0
	D25	31.6	33.8	5.6	55	-7
	D100	32.0	34.4	5.0	60	10
	D500	32.8	35.9	7.4	50	-23
	D1000	33.5	37.2	7.9	33	-77
Indole, $0.05 \text{ }^{\circ}\text{C min}^{-1}$	I0 (water)	31.4	34.3	6.3	48	0
	I5	27.7	32.6	7.9	57	29
	I10	26.4	30.7	9.8	58	34
	I15	22.2	29.0	13.8	61	44
	I16	21.5	29.0	14.3	62	49
	I18	17.9	28.0	16.7	64	54
	I20	11.5	28.3	20.9	65	56

**Fig. 7** Deconvoluted DSC peaks of PNIPA-dopamine systems



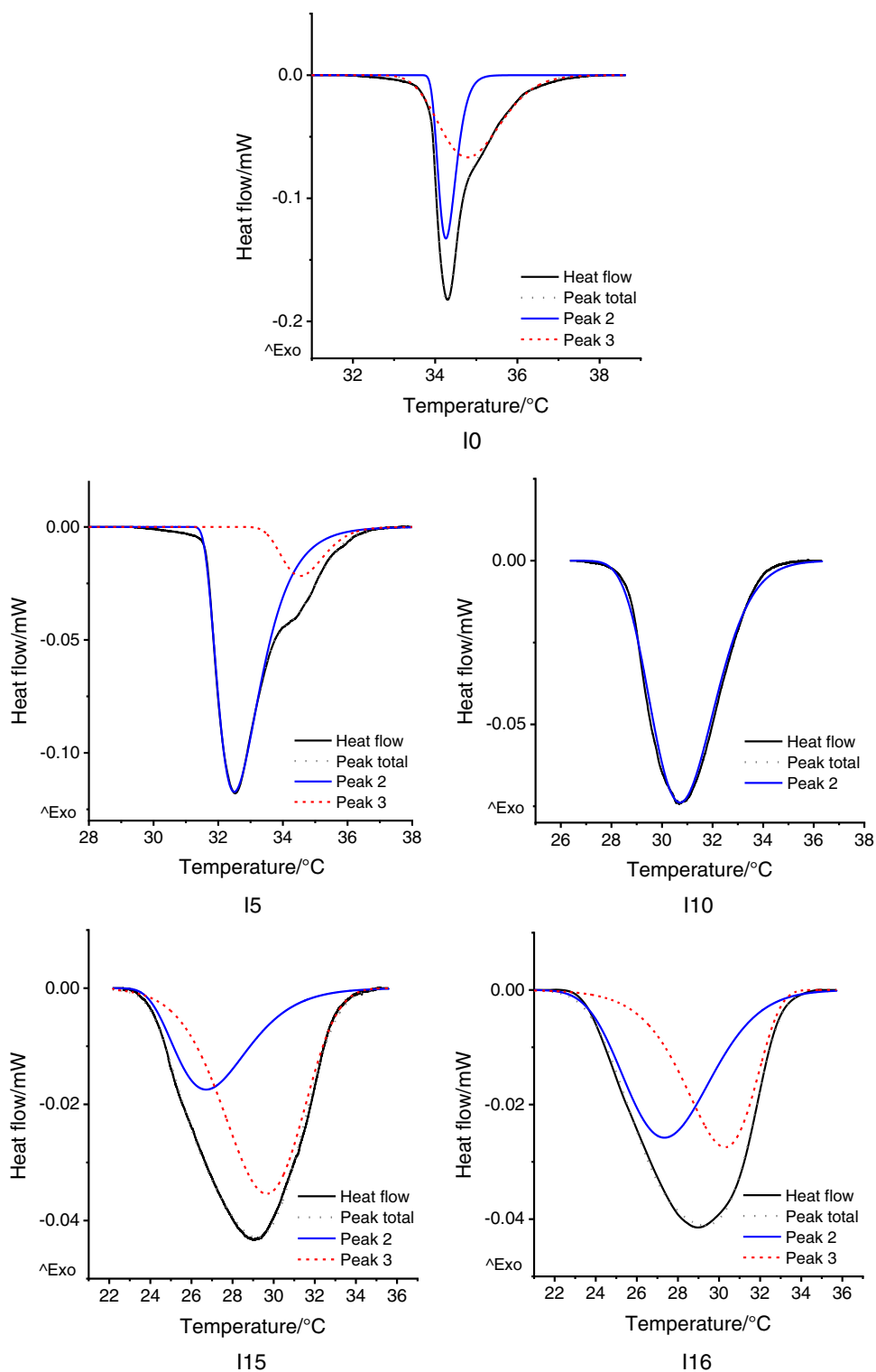
concentration (Fig. 7, D1000). The parameters obtained after deconvolution are shown in Table 3.

Former TG/DTG and NMR investigations revealed that aqueous dopamine does not interact with the polymer chain but instead may undergo self-assembly, even leading

to polymerization under certain conditions [24–26]. This process may be exothermic, thus reducing the overall endothermic enthalpy of the phase transition [27]. The entropy decrease could be an additional sign of the separation of the polymer gel and the self-assembled dopamine “phases”



**Fig. 8** Deconvoluted DSC peaks of PNIPA-indole systems

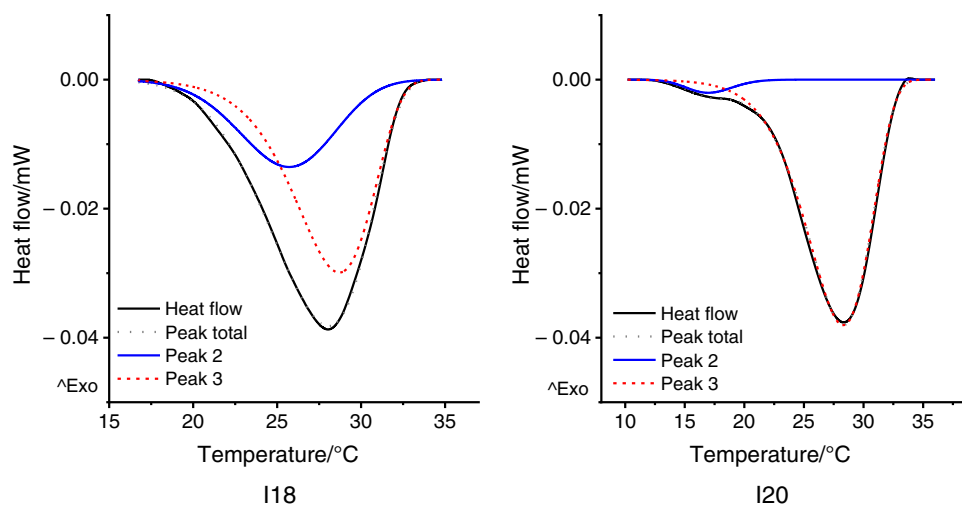


(Table 3). The lower mobility of the bulkier dopamine assemblies may result not only in retarded relaxation but also in a slowing down of the temperature induced expulsion of the hydrophobic water. The strong dopamine–dopamine interaction inhibiting the dopamine–PNIPA interaction was also found to foster crystallization of the drug as the loaded

gel was dried [10]. This indicates that such a scenario is not advantageous if the drug is required in amorphous form.

At low indole concentration the shape of the curves as well as their deconvolution are very similar to those of the dopamine: a faster and a slower set of processes can be distinguished (Fig. 8). However, the continuously increasing

Fig. 8 (continued)

**Table 3** Parameters of fitted curves – PNIPA-dopamine systems

Peak	Area/mJ			Area/%			Peak position/°C			Half width/°C		
	1	2	3	1	2	3	1	2	3	1	2	3
D0	–	249.0	233.1	–	51.6	48.4	–	33.6	34.0	–	0.3	1.0
D25	–	274.1	196.5	–	58.2	41.8	–	33.8	34.2	–	0.3	1.4
D100	–	296.2	212.8	–	58.2	41.8	–	34.4	34.8	–	0.3	1.1
D500	–	190.8	213.5	–	47.2	52.8	–	35.9	36.2	–	0.5	1.7
D1000	28.8	163.5	77.3	10.7	60.6	28.7	35.9	37.2	38.3	1.2	1.3	1.4

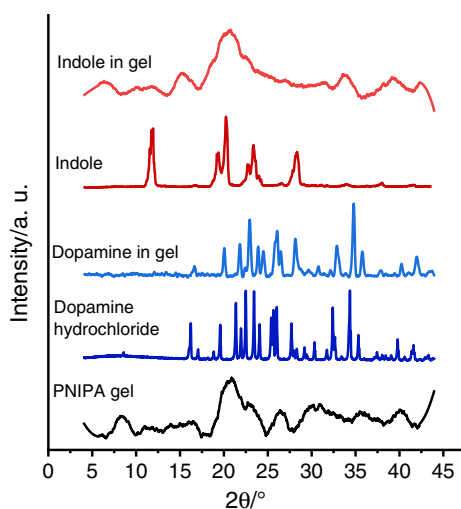
**Table 4** Parameters of fitted curves – PNIPA-indole systems

Peak	Area/mJ		Area/%		Peak position/°C		Halfwidth/°C	
	2	3	2	3	2	3	2	3
I0	89.1	150.1	37.2	62.8	34.2	34.8	0.5	1.7
I5	250.1	43.2	85.3	14.7	32.5	34.5	1.5	2.6
I10	286.0	–	100	–	30.7	–	3.0	–
I15	97.0	209.0	31.7	68.3	26.7	29.7	4.2	5.4
I16	165.8	146.1	53.2	46.8	27.3	30.3	5.1	3.9
I18	116.7	220.3	34.6	65.4	25.7	28.7	6.6	5.4
I20	12.0	323.8	3.6	96.4	17.0	28.3	4.5	6.3

half width of both fitted curves indicates the increasing complexity of the thermodynamic processes taking place with increasing indole concentration (Table 4). Up to I10 the contribution of the faster processes gradually increases, and above this concentration the slower processes become dominant. The non-monotonic trend in the contribution of the “fast” and “slow” processes marks the critical concentration of indole. As a result of the interaction between the C=O regions of the polymer and the NH group of the indole revealed by FTIR [28], the drug molecules are gradually able to expel and substitute the water molecules from the hydrophobic sheath, leading to break down of the gel structure and syneresis. The increasing entropy values may imply that the drug molecules are

distributed along the polymer chains. The bulkier indole molecules decelerate the motion of the polymer segments and are unable to prevent adhesion among the chains, as they are deprived of their protecting water layer. These effects result in a more rigid system where all the motion and relaxation slow down drastically.

Figure 9 compares the powder X-ray diffractograms of the dry polymer gel samples loaded with 500 mM dopamine hydrochloride and 20 mM indole, respectively, confirming the relation between the interactions within the gel and the degree of crystallinity of the drug. The self-assembled dopamine regions are converted to crystalline dopamine. HCl once the water has evaporated, while the similarity of the diffractograms of the neat gel and indole filled samples



**Fig. 9** XRD patterns of the crystalline drugs and the dried loaded gel samples. Curves are normalized to their highest intensity and shifted vertically for clarity

implies that the evenly distributed indole is in an amorphous state after drying.

## Conclusions

The various techniques applied here to reveal the interactions governing the release of dopamine and indole from a responsive polymer gel consistently confirm that the two molecules act dissimilarly. The dopamine can be completely recovered from the loaded polymer matrix at 25 °C. The self-assembling affinity of the dopamine molecules obstructs their interaction with the polymer, and the water molecules can form a uniform protective water sheath even at elevated concentration. Therefore the swelling is not affected by dopamine. The kinetics and the temperature of the phase transition is not influenced by dopamine up to 500 mM. At 1000 mM, however, a few dopamine molecules may stick to the polymer chains, thus slowing down syneresis. The DSC results also imply that above the phase transition temperature polymer and dopamine-rich domains are formed. Indole, on the other hand, readily replaces the water molecules through the interaction between the C=O sites of the polymer and the NH groups of the drug, thereby depriving the chains of the protecting hydrophobic water layer. At 20 °C this already leads to an indole induced phase transition at ca 16 mM initial indole concentration. The temperature of the phase transition gradually decreases with increasing indole concentration. Owing to the heavier indole decorated polymer chains the rate of the phase transition decelerates. The strong interaction between the carrier matrix and the indole results

in an even distribution of the drug along the polymer chains, and after drying the indole is found in amorphous form.

**Acknowledgements** We extend our warm thanks to Ms. V. Bérczes and Mr. G. Bosznai for their invaluable assistance. This research was supported by OTKA K115939 and also funded by the National Research, Development, and Innovation Fund of Hungary under Grant TKP2021-EGA-02. We are grateful to the Soft Matter Group of BME and Prof. B. Iván for access to their instrumentation.

**Funding** Open access funding provided by Budapest University of Technology and Economics.

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