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Theoretical study of temperature induced phase transitions in poly (β-benzyl-L-aspartate) and it's copolymer

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Abstract

In polypeptides and proteins, the phenomenon of conformational transitions has been successfully explained on the basis of Zimm and Bragg model, as a function of temperature, pressure, solvent concentration and pH etc. The Zimm and Bragg theory of helix ↔ coil transition has been modified to explain the temperature induced helix → helix transitions in solid films of poly(β-benzyl-L-aspartate) and in its copolymer copoly(β-stearyl-L-aspartate-β-benzyl-L-aspartate) with varying degrees of stearylation. An expression for the degree of order is obtained from the grand partition function for the entire chain in terms of nucleation parameter, which controls the transition width. At low degree of stearylation, an α → ω transition occurs with the increase in temperature, whereas, at degree higher than 40%, the reversal of α helix sense from right-handed to left-handed takes place. The theoretical transition curves are found to be in good agreement with the experimental data.

Keywords: Phase transition; α Helix; ω Helix

1. Introduction

The synthetic polymers have been attracting a great deal of interest in the recent times both theoretically and experimentally, because of their ability to act as simplest model for the conformational analysis of polypeptides and proteins. Many of them undergo an order–order or order–disorder transition under appropriate conditions i.e. change in solvent composition, pH, temperature, pressure, electric or magnetic field [1–9]. This results in large changes in their mechanical properties and morphological structure as well. Watanabe and coworkers [10] have reported in this journal, a Circular Dichroic study on solid films of copoly(β-sterayl-L-aspartate-β-benzyl-L-aspartate) [copoly(SLAsp-BLAsp)] as a function of degrees of stearylation and temperature. Copoly(SLAsp-BLAsp) was synthesized by ester exchange reaction of parent polymer poly(β-benzyl-L-aspartate) [PBLAsp]. They found an α → ω phase transition and reversal of helix sense corresponding to low and high degree of stearylation.

As a part of our ongoing research work on vibrational analysis, phonon dispersion [11–17] and phase transition [18–21] in a variety of macromolecules, here in the present communication Zimm and Bragg theory of helix ↔ coil transition has been modified to explain the phenomenon of temperature induced phase transition in copoly(SLAsp-BLAsp) as well as in its parent polymer α-helical poly(β-benzyl-L-aspartate) [PBLAsp]. This is an extension of our earlier work on the heat capacity and Vibrational Dynamics of the α helical poly(β-benzyl-L-aspartate) [16]. PBLAsp an unusual synthetic homopolypeptide, is capable of adopting four different conformations [22–27], namely, left-handed α-helix, right-handed α-helix, ω-helix, and β pleated sheet under varying conditions of preparation. The conformational versatility of PBLAsp has made this polymer, a model of great interest from the viewpoint of molecular dynamics of polypeptides and proteins [16].

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2. Theory

The present theoretical approach is a modification of Zimm and Bragg model [9] and has earlier been used to explain an order–order phase transition in poly-L-proline [20] and order–disorder phase transition in case of polyethylene [19]. An expression for degree of order Q is obtained from the grand partition function for the entire chain in terms of nucleation parameters. The segments in the form I (α helix) and the form II (ω helix) have been represented by h_1 and h_2. Whereas, k_1 and k_2 are the boundary states which is the first of the sequence of segments in states I and II respectively. The nucleation and the growth parameters have been represented by σ_1, σ_2 and s_1, s_2 respectively. The transition matrix for order–order transition is written as following,

$$M = \begin{array}{c|cccc} & h_1 & k_1 & k_2 & h_2 \\ h_1 & s_1 & 0 & \sigma_2 s_2 & 0 \\ k_1 & s_1 & 0 & 0 & 0 \\ k_2 & 0 & 0 & 0 & s_2 \\ h_2 & 0 & \sigma_1 s_1 & 0 & s_2 \end{array}$$

The eigenroots are given by secular equation,

$$|M - \lambda I| = 0 \tag{1}$$

$$\lambda^2(\lambda - s_1)(\lambda - s_2) - \sigma_1\sigma_2 s_1^2 s_2^2 = 0 \tag{2}$$

The four eigenroots of the Eq. (2) depend only on the product $\sigma_1\sigma_2$ ($=\sigma$ say). If $\sigma_1\sigma_2 = 0$, the eigenroots are s_1, s_2, 0, 0 and if the product $\sigma_1\sigma_2 \ll 1$, the two smaller eigenroots are of the order of $\sqrt{(\sigma_1\sigma_2)}$ and the larger eigenroots are of the order of s_1 and s_2 that is unity in the transition range. Thus the contribution of smaller eigenroots to the partition function is negligible and the main contribution comes from the larger eigenroots denoted by λ_1 and λ_2. These eigenroots can be obtained on iteration and are given by the following relations:

$$\lambda_1 = s_1 + \frac{\sigma_1\sigma_2 s_1^2 s_2^2}{\lambda^2(\lambda - s_2)} \tag{3}$$

and

$$\lambda_2 = s_2 + \frac{\sigma_1\sigma_2 s_1^2 s_2^2}{\lambda^2(\lambda - s_1)} \tag{4}$$

The partition function Z of a chain of N segments is then given by

$$Z = \sigma' \sum A_i \lambda_i^N \quad (i = 1 \text{ to } 4) \tag{5}$$

where

$$A_i = \left\{ \frac{\lambda_i^2(1+x) - \lambda_i(s_2 + s_1 x) + s_1 s_2(\sigma_1 x + \sigma_2)}{4\lambda_i^2 - 3\lambda_i(s_1 + s_2) + 2 s_1 s_2} \right\} \tag{6}$$

with $x = \dfrac{\sigma''}{\sigma'}$

Factor x is defined as the end effect parameter. The parameters σ' and σ'' give the interaction of the end segments in states h_1 and h_2 with the surroundings.

Hence Q_{I}, the fraction of the state in the form I can thus be calculated as following;

$$= \left(\frac{1}{N}\right)\left(\frac{\partial \ln Z}{\partial \ln s_1}\right) = \left(\frac{1}{N}\right)\left(\frac{s_1}{Z}\right)\left(\frac{\partial Z}{\partial s_1}\right) \tag{7}$$

$$\cong \left[\frac{(s_1/\lambda_1)(\partial\lambda_1/\partial s_1) + (s_1/\lambda_2)(\partial\lambda_2/\partial s_1)B + (1/N)\{(s_1/A_1)(\partial A_1/\partial s_1) + (s_1/A_2)(\partial A_2/\partial s_1)B\}}{1+B}\right], \text{ where } B = \left(\frac{A_2}{A_1}\right)\left(\frac{\lambda_2}{\lambda_1}\right)^N \tag{8}$$

The equilibrium constant s and nucleation parameter σ for transition are given by

$$s = \frac{s_1}{s_2},\ s_1 s_2 = 1 \text{ and } \sigma = \sigma_1\sigma_2$$

Where

$$s = \exp\left(\frac{\Delta H}{R}\right)\left[\left(\frac{1}{T} - \frac{1}{T_{\mathrm{f}}}\right)\right] \tag{9}$$

Since the growth parameter s and subsequently s_1 ($s_1 = \sqrt{s}$) depend on the temperature T as given by the Eq. (9). The variation in values of s or s_1 parameter is reflected by the corresponding change in temperature T. Hence variation of one parameter takes care of the other. ΔH is the molar change in enthalpy, T_{f} is the transition temperature and R is the constant given by 2 cal/mol.

For very large value of N, fraction Q_{I} becomes

$$Q_{\mathrm{I}} = \frac{(s_1/\lambda_1)(\partial\lambda_1/\partial s_1) + (s_1/\lambda_2)(\partial\lambda_2/\partial s_1)B}{1+B} \tag{10}$$

The value of B goes to zero for infinitely large N, hence

$$Q_{\mathrm{I}} = \left(\frac{s_1}{\lambda_1}\right)\left(\frac{\partial\lambda_1}{\partial s_1}\right) \tag{11}$$

λ_1 denoting the largest eigen root.

3. Results and discussion

We report here, an extension of the Zimm and Bragg model [9] to explain the order–order transition through the two parameters theory. The theoretical transition curves for a polymer chain of length N have been obtained from Eqs. (7)–(10). However, the Eq. (10) is the final equation for calculation of crystallinity. These curves are found to be linear in the transition region. The linear part when extrapolated can give the value of the temperature corresponding to the $Q = 0$. This is known as the melting temperature T_{m} at which the last crystallite disappears. The value of melting temperature depends on the slope of transition curves, which in turn depends on the value of nucleation parameter σ. In contrast to the significant variation in values of growth parameter s with temperature, the nucleation parameter σ has a weak dependence on temperature. Therefore, for all the theoretical purposes the values of nucleation parameter are assumed to be constant and independent of temperature as well as side-chain interactions. The end effect parameter x has been chosen to be equal to unity due to the end effects being negligible for the long chains.

Circular Dichroic study on copoly(SLAsp-BLAsp) with varying degree of stearylation by Watanabe et al. [10] shows an irreversible $\alpha \rightarrow \omega$ phase transition occurred around 140 °C for low degree of stearylation (SA = 0–30%). Whereas, at degree of stearylation higher than 40% the reversal of helix sense from right to left handed took place. They have also plotted the maximum molar ellipticity $[\Theta]_{\max}$ against temperature for SA = 0, 41, 63, 72 in case of solid films. The maximum molar ellipticity $[\Theta]_{\max}$ may be used as a convenient parameter to follow the temperature induced phase transition [1] in the parent polymer PBLAsp (SA = 0) and in its copolymer copoly(SLAsp-BLAsp). The temperature induced $\alpha \rightarrow \omega$ phase transition in PBLAsp and reversal of helix sense from right-handed copoly(SLAsp-BLAsp) → left-handed copoly(SLAsp-BLAsp) are discussed under separate headings.

3.1. The temperature induced $\alpha \rightarrow \omega$ phase transition in poly(β-benzyl-L-aspartate)

The transition parameters which provide the best fit to the experimental data of Watanabe et al. [10], have been given in Table 1. The experimental points (represented by ▲) and theoretical points (represented by ● and ■) corresponding to the various degree of crystallinity versus temperature for ω and α forms have been plotted in the Fig. 1. In case of polymers such as polyglycine, polyproline, polyethylene the σ values are typically small in the range of 10^{-3}–10^{-4} (refer to the Table 2) [19,20,28]. The values of nucleation parameter for the PBLAsp, are taken to be on the higher side, on account of the fact that larger the value of σ, the smaller will be the free energy change in creating the transition/boundary interface [29]. This is also reflected as a broader width of transition in experimental data of PBLAsp (SA = 0). The consequence of requirement of $\sigma_1 > \sigma_2$ for best fitted theoretical σ values implies that within the chain, nucleation of α helix is more probable than the nucleation of ω helix. The $\alpha \rightarrow \omega$ transition could be induced by the stacking formation of side chain benzene ring. The gain in the energy from stacking of side chain benzene ring is sufficient enough to offset the energy loss, which occurs when α helix is thermally deformed to ω helix.

3.2. Reversal of helix sense in copoly(SLAsp-BLAsp)

The transition parameters for copoly(SLAsp-BLAsp) have been given in Table 1. The experimental points (represented by ▲) and theoretical points (represented by ● and ■) corresponding to the various degrees of crystallinity in left-handed and right-handed form, have been plotted in the Figs. 2–4 (crystallinity versus temperature curve). These curves represent the different degree of stearylation in copoly(SLAsp-BLAsp). Comparison of the values of σ arrived at theoretically (refer to Table 1), in both the kinds of conformational behaviour, reveals the larger probability of phase transition from the right to the left-handed helical form as compared to $\alpha \rightarrow \omega$ transition $[\sigma(\mathrm{r} \rightarrow \mathrm{l}) < \sigma(\alpha \rightarrow \omega)]$. This also makes helix sense transition sharper in copoly(SLAsp-BLAsp) as compared to transition in case of PBLAsp. The increase in the values of nucleation parameter σ for copoly(SLAsp-BLAsp) with the increasing degree of stearylation is observed theoretically. The order of theoretical values of σ i.e. $\sigma(\text{SA-72}) > \sigma(\text{SA-63}) > \sigma(\text{SA-41})$, explains the experimentally observed comparative degree of stability in copoly(SLAsp-BLAsp) with increasing degree of stearylation. This stabilization is manifested by the shifts in the helix sense transition temperatures, i.e. $T_f(\text{SA-72}) > T_f(\text{SA-63}) > T_f(\text{SA-41})$, as reported by Watanabe et al. The best fitted theoretical values $\sigma_1 > \sigma_2$, implies that within the chain, nucleation of right-handed helical copoly(SLAsp-BLAsp) is more probable than the nucleation of left-handed helical copoly(SLAsp-BLAsp), because the variation of σ_1 and σ_2 (where $\sigma = \sigma_1\sigma_2$) reflects the variation of probabilities of nucleation of conformation I in a sequence of segments II and the nucleation of conformation II in a sequence of segments I, respectively [20]. The reason for the higher stability of right-handed helix at low temperatures and left-handed helix of copoly(SLAsp-BLAsp) at higher temperatures is probably due to the difference in the side chain–main chain orientations in right and left-handed helical forms. This in turn causes the difference in dipole–dipole interaction

Table 1
Transition parameters for PBLAsp (SA = 0) and it's copolymer copoly(SLAsp-BLAsp)

S. No.	Degree of stearylation 'SA'	σ_1	σ_2	$\sigma = \sigma_1\sigma_2$
01	00	5.0×10^{-1}	1.0×10^{-1}	5.0×10^{-2}
02	41	6.0×10^{-2}	2.0×10^{-2}	1.2×10^{-3}
03	63	9.0×10^{-2}	4.0×10^{-2}	3.6×10^{-3}
04	72	1.0×10^{-1}	6.0×10^{-2}	6.0×10^{-3}

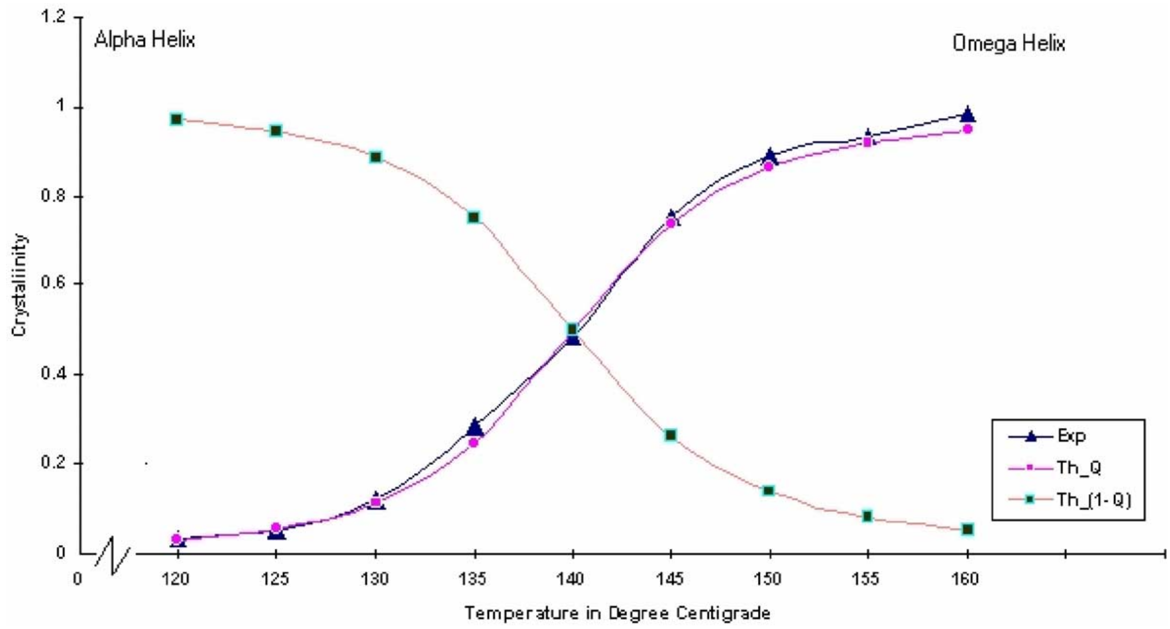

Fig. 1. Schematic representation of crystallinity as a function of temperature in PBLAsp (SA = 0).

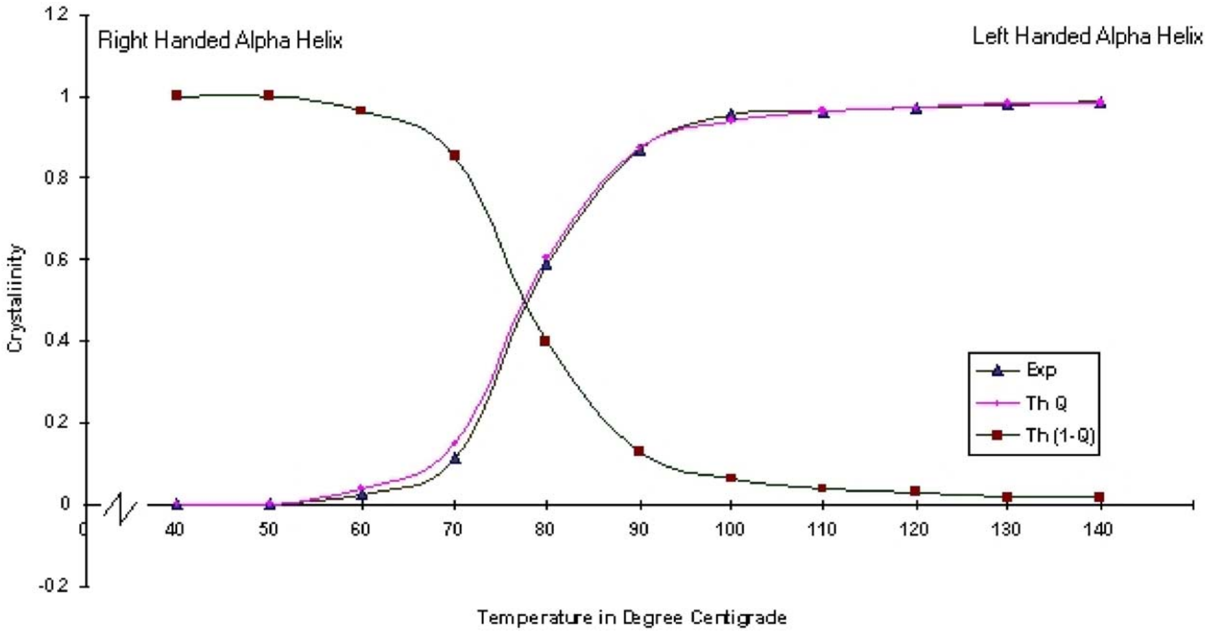

Fig. 2. Schematic representation of crystallinity as a function of temperature in copoly(SLAsp-BLAsp)(SA = 41).

Table 2
Fit parameters of different polymers in case of temperature/pressure-induced phase transition

S. No.	Biopolymers/ polymers	Nucleation parameter σ	Growth parameter s
01	Polyglycine [28]	1.0×10^{-5}	0.62
02	Polyalanine [28]	8×10^{-4}	1.06
03	Polyserine [28]	7.5×10^{-5}	0.79
04	Polyleucine [28]	33×10^{-4}	1.14
05	Polyproline [20]	5×10^{-5}	1.00
06	Polyethylene [19]	1×10^{-4}	1.00

between the ester carbonyl groups of side chain and amide groups of main chain for the two-helix forms.

4. Conclusions

The present theoretical work on temperature induced phase transition in the synthetic polypeptide PBLAsp and in its copolymer copoly(SLAsp-BLAsp) with varying degrees of stearylation, clearly demonstrates the ability of our modified Zimm and Bragg treatment to successfully

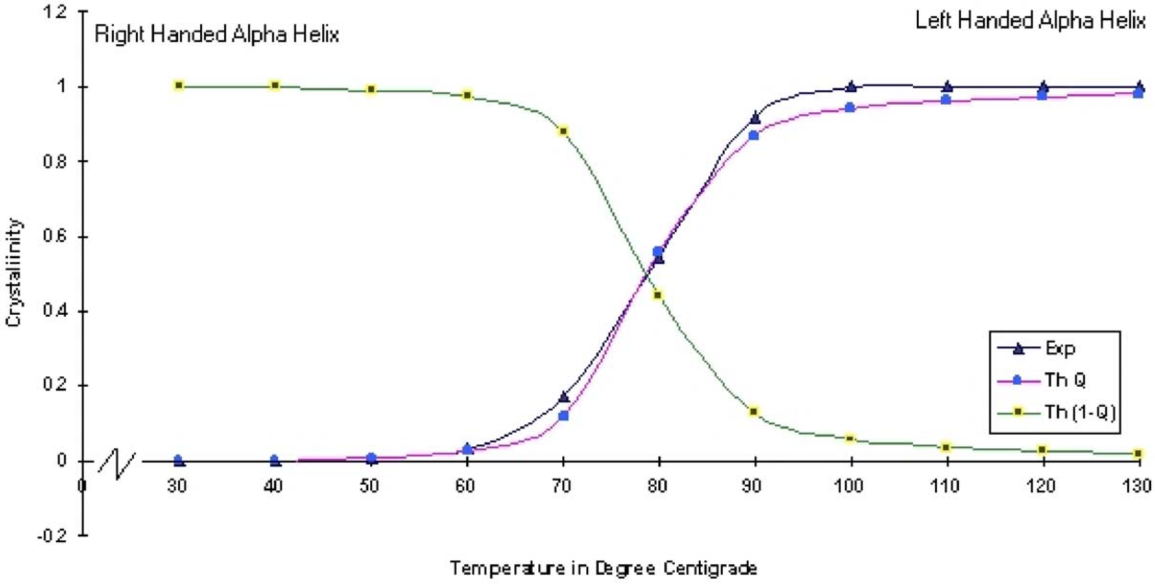

Fig. 3. Schematic representation of crystallinity as a function of temperature in copoly(SLAsp-BLAsp)(SA = 63).

organize and interpret the experimental data on the equilibrium properties of helix → helix transitions. The same treatment can also be applied to explain the helix ↔ coil transitions in a variety of macromolecules. One of the merits of the present approach leads to the explicit arrival of values of the σ_1 and σ_2, which are consistent with the stabilization/destabilization of the ordered/disordered states subjected to various kinds of environmental conditions. As a further extension of the present model, a general model for phase transition in macromolecular systems which incorporates the intermediate state also, is currently underway and will be reported shortly elsewhere.

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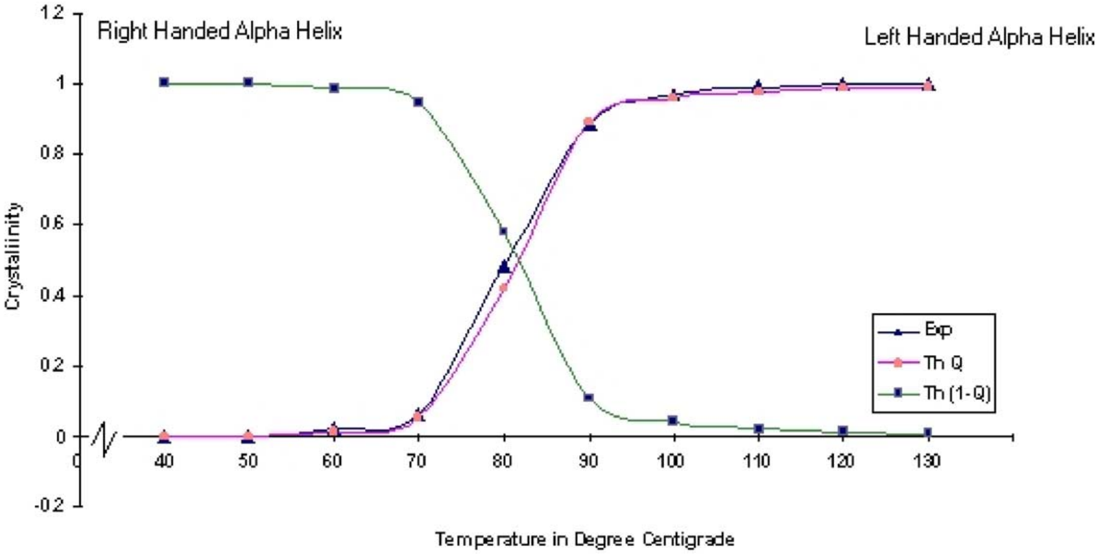

Fig. 4. Schematic representation of crystallinity as a function of temperature in copoly(SLAsp-BLAsp)(SA = 72).

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