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EVALUATION OF COOPERATIVITY FOR PHASE TRANSITIONS IN TWO- AND THREE-DIMENSIONAL SYSTEMS

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Use of the so-called "cooperative unit" (readily obtainable from the midpoint slope of phase transition curves) is discussed for the determination of cluster sizes and cooperative interaction energies. This quantity has been commonly employed in a rather empirical way since its correct interpretation is known only for some special cases (linear systems, all-or-none transitions). It is shown in the framework of a lattice model (Ising model) that the cooperative unit may be interpreted in terms of correlation functions and that it defines an average cluster corresponding to the patch size as obtained from scattering experiments. Relations between the cooperative unit and a cooperativity parameter are given for various lattices. Different types of transition curves are discussed using a simple analytical formalism, the quasichemical approximation. Some important nonideality effects are investigated which may lead to a "smearing-out" of first-order transitions.

1. Introduction

Cooperative systems appear to be important tools for the function of living organisms because of their ability for control and switching operations which require large changes of state as a consequence of rather small changes in the external conditions.

Cooperative systems are in fact characterized by very sharp transitions between different states (e.g. different molecular conformations, different types of aggregation or bound and unbound states of a substrate) within narrow intervals of temperature, pressure, ligand concentrations etc.

In order to obtain some quantitative information about the degree of cooperativity it thus seems most natural to examine the steepness of the sigmoidal transition curves. Commonly the midpoint of transition is taken as the reference point which allows the convenient definition of "van 't Hoff quantities" such as the apparent transition enthalpy ΔH_{vH}. This is frequently employed in the evaluation of thermotropic transitions [1,2]:

$$\Delta H_{\mathrm{vH}} = 4RT_{\mathrm{m}}^2(\partial\theta/\partial T)_{T_{\mathrm{m}}}. \quad (1)$$

Here T_{m} is the midpoint temperature, θ the degree of transition and R the gas constant.

In complete analogy we can define a "van 't Hoff transition volume" ΔV_{vH} [3]:

$$\Delta V_{\mathrm{vH}} = -4RT(\partial\theta/\partial p)_{p_{\mathrm{m}}}. \quad (2)$$

Yet to derive the information about cooperativity from the "midpoint slope" we need to normalize the van 't Hoff quantities by the molar transition quantities ΔH^0 and ΔV^0, respectively, as measured by means of calorimetry and compressibility. The thus obtained ratios define the well-known "cooperative unit" (c.u.) [1,2]:

$$\text{c.u.} = |\Delta H_{\mathrm{vH}}/\Delta H^0| = |\Delta V_{\mathrm{vH}}/\Delta V^0|. \quad (3)$$

For convenience, we shall restrict ourselves to thermal transitions in the following, but due to the outlined analogies all results will be applicable to other transitions in just the same way.

For systems with non-interacting particles, eq. (1) becomes identical to van 't Hoffs equation with $\Delta H_{\mathrm{vH}} = \Delta H^0$. The c.u. is then unity: this is the case of noncooperative transitions.

For cooperative systems, the simplest mechanism is described as the "all-or-none" transition, where a certain number, n, of subunits react simultaneously. In this case, ΔH_{vH} is the reaction enthalpy of that portion of the system which participates in the transition, i.e. n times ΔH^0 so that c.u. = n. (Here it is presumed that ΔH^0 is given per mole of subunits. If ΔH^0 refers to larger entities containing N subunits, c.u. must be n/N; this is the standard convention, e.g. in protein folding problems, where ΔH^0 is generally given per mole of protein. For proteins with N amino acid constituents c.u. = $1/N$ then means non-cooperativity whereas c.u. = 1 means the all-or-none case with $n = N$).

The question arises now whether the concept of the cooperative unit is equally useful for more complicated systems — as frequently encountered in practice — where the all-or-none approach is inadequate. A very general class of such systems is characterized by cooperative growth of clusters. We restrict our analysis to transitions where the individual elements can be assumed to adopt only two different states. Cluster growth for one state must then be accompanied by cluster shrinkage for the other state. It may be expected that the cooperative unit bears information on the average midpoint cluster size.

This view is strongly supported by the results found for linear lattices for which it was shown by means of the linear Ising model that the c.u. equals indeed the mean sequence length at T_m (aside from a term unity of no practical importance) [4]. Furthermore, a direct relation between c.u. and the cooperativity parameter σ can be established; this parameter indicates by how much the probability of nucleation is smaller than that of cooperative growth [4,5]:

$$\text{c.u.} = \sigma^{-1/2} \tag{4}$$

Encouraged by these results, many authors have used the c.u. even for nonlinear systems, e.g. membrane problems of a two-dimensional character [2,6,7].

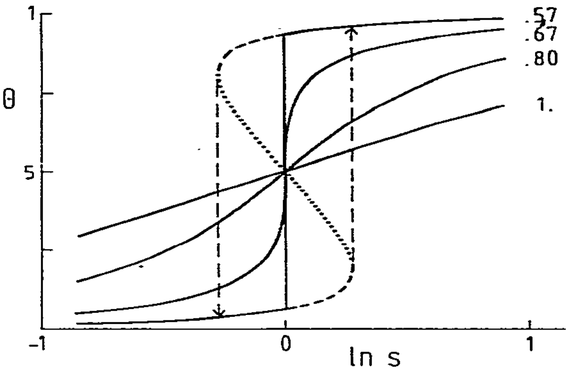

Fig. 1. Different types of transition curves for nonlinear systems as a function of the equilibrium constant s for different cooperativity parameters $\sqrt{\sigma}$ as indicated in the figure. The curves have been calculated according to the quasichemical approximation with a coordination number $z = 6$ (plane hexagonal or simple cubic lattice). $\sqrt{\sigma} = 1$ corresponds to non-cooperativity, $\sqrt{\sigma} = 0.67$ is the critical value. Dotted parts are instable, dashed parts are metastable and may lead to hysteresis.

To our knowledge, no strict justification was ever given for this practice. The question is in fact more difficult than one might think at first inspection. Two- and three-dimensional systems give rise to some peculiar features which make it impossible to simply extrapolate the arguments of the one-dimensional case to higher dimensions. For instance, first-order transitions (with a discontinuous part of the transition curve) may occur in lattices with more than one dimension. This makes questionable the attempt to adapt the formulae of the linear Ising model to a two-dimensional problem, as has been proposed by some authors [8]. In addition, the relation between the c.u. and the cooperativity parameter σ is strongly lattice-dependent, as will be shown in the next chapter. As a consequence, the simple formula c.u. = $\sigma^{-1/2}$ will yield meaningless results when applied to surface and space lattices.

In section 2, we offer a precise definition of clusters in terms of correlation functions. This definition allows a generalization of the "average cluster" interpretation of the c.u. including higher dimensional lattices. The use of the easily accessible, experimental quantity "c.u." may thus be justified for general cooperative systems.

Furthermore, the cooperative unit can be related to energy variables, especially the cooperativity parameter σ. This is done in section 3.

In nonlinear systems a critical value of the cooperativity parameter is known to exist (often referred to as the "critical point") where the midpoint slope of the transition curve just becomes infinite. For σ smaller than this critical value σ_c (which is lattice-dependent), part of the transition curve is discontinuous, with a strictly vertical portion at $\theta = 1/2$ (see fig. 1). In these cases, the concept of c.u. as defined by eq. (3) is meaningless, since the midpoint slope of the transition curve is always infinite. Instead, σ must be estimated from the length of the vertical portion of the transition curve.

In practice, however, it will often be difficult to measure the ideal situation where the midpoint part of the transition curve is vertical. In fact, even minor impurities of the used material or some other non-idealities may suffice to smear out such discontinuities in the measured signal. This seems to be the experimental situation e.g. in the case of phospholipid phase transitions (see e.g. ref. [9]).

In all these cases, a finite value of the c.u. is obtained from the experiments. This value is no longer primarily determined by the interaction energies of the pure system, but by the degree of nonideality, e.g. the concentration of impurities or lattice defects. This offers a somewhat unusual, but highly interesting application of cooperative units, namely the determination of the degree of perturbation in a nonideal system. We discuss this topic in section 4.

In order to make the presentation less abstract, some examples concerning the phospholipid phase transition are given. A justification for using the Ising approach to these problems may be found in the literature [10,11]. A more detailed treatment of lipid membranes containing impurities or intrinsic molecules is being prepared for separate publication.

2. Cooperative unit and correlations

In order to give an accurate interpretation of the cooperative unit in complex nonlinear systems and to relate it to other parameters, we shall use a lattice model. Thus the particles involved in the transition are assumed to be bound to the sites of a given lattice and to be able to diffuse only by hopping from one lattice site to the next. Such lattice models have proved to be extremely useful approximations even for liquid phases.

We imagine that the "particles" or "elements" at each lattice point are capable of a temperature-dependent transition between two states 1 and 2 and that they can interact cooperatively. We shall restrict our discussion to nearest-neighbour interactions. With these assumptions (which should be at least approximately adequate for a large number of experimentally interesting systems) the appropriate mathematical description is provided by the Ising theory. Although this theory is known to be very complex in the nonlinear case, the important results in our context can be given in a fairly simple form. In this section we shall first discuss the interpretation of the cooperative unit in terms of correlation functions.

For any lattice point i, we define a parameter u_i which can assume the values -1 (if the element at site i is in the 1 state) or $+1$ (if it is in the 2 state). The thermodynamic average over the whole lattice, $\langle u \rangle$, is then directly related to the degree of transition, i.e. the probability of finding state 2 at a randomly picked site. Since θ varies between zero and unity, we have

$$\theta = \tfrac{1}{2}(1 + \langle u \rangle) \tag{5}$$

Direct information about the cooperativity of the system is provided by the pair correlation function Γ_r which may now be defined by [12]:

$$\Gamma_r = \langle u_i\, u_{i+r} \rangle - \langle u_i \rangle^2 , \tag{6}$$

where the lattice site i is chosen at random and site $i + r$ is r sites away from i. Note that $\langle u_i \rangle = \langle u \rangle$ vanishes for $\theta = 1/2$. The correlation function essentially measures the probability that two particles separated by r site-to-site distances, are found in the same state. In a non-cooperative system the lattice sites are independent from each other and the pair correlation vanishes for $r \neq 0$. In a cooperative system, neighbouring particles tend to be in the same state, and this tendency is propagated through the lattice from neighbour to neighbour. For large distances r, the decay of the pair correlation is dominated by an exponential [13]: $\exp(-r/\xi)$. The parameter ξ is called the correlation length.

These quantities must now be related to the energy parameters of the system. Taking into account nearest-neighbour interactions we write w_1 and w_2 for the free energies of the two states and w_{11}, w_{22} and $w_{12} = w_{21}$ for the pair interaction energies. The system hamiltonian may then conveniently be formulated using the projection operators $P_{1i} = \frac{1}{2}(1 - u_i)$ and $P_{2i} = \frac{1}{2}(1 + u_i)$ which select the correct contributions of states 1 and 2 at the lattice site i:

$$\mathcal{H} = \sum_i [P_{1i}w_1 + P_{2i}w_2] + \sum_{i,j} [P_{1i}P_{1j}w_{11} + (P_{1i}P_{2j} + P_{2i}P_{1j})w_{12} + P_{2i}P_{2j}w_{22}] \tag{7}$$

The second sum is taken over all nearest-neighbour pairs. By summing up the different u_i terms, eq. (7) may be written in the following form (cf. [10]):

$$\mathcal{H} = A - B\sum_i u_i - C\sum_{i,j} u_i u_j, \tag{8}$$

$$B = \tfrac{1}{2}[(w_1 - w_2) + \tfrac{1}{2}z(w_{11} - w_{22})], \tag{9}$$

$$C = \tfrac{1}{4}[2w_{12} - (w_{11} + w_{12})], \tag{10}$$

where z stands for the number of nearest neighbours of a lattice point (coordination number).

The parameter A is of little importance due to the freedom in choosing the energy reference. Symbol C obviously represents the exchange energy which the system gains by replacing a pair of neighbouring elements in different states (12) by neighbouring elements in equal states (11 or 22).

A cooperativity parameter σ may therefore readily be defined in the usual way [4,5]:

$$\ln \sigma = -4C/kT. \tag{11}$$

Assuming that C is not temperature dependent, σ can be considered a constant if the transition occurs in a narrow temperature range. On the other hand, the right-hand side of eq. (9) is readily recognized as the energy difference for the pure 1 and 2 states. An equilibrium constant s can be introduced by setting [14]:

$$B = \tfrac{1}{2}kT \ln s. \tag{12}$$

The parameter s, by definition, obeys the thermodynamical relation

$$\ln s = -\Delta H^{\ominus}/RT + \Delta S^{\ominus}/R \tag{13}$$

As usual $\Delta H^{\ominus}$ can be identified (approximately) with the calorimetric transition enthalpy. Then $\Delta S^{\ominus}$ is given by

$$\Delta S^{\ominus} = \Delta H^{\ominus}/T_m. \tag{14}$$

Using eq. (5) and the definition (3) it may be shown now by simple transformations that the cooperative unit reads:

$$\text{c.u.} \equiv \left|\frac{\Delta H_{vH}}{\Delta H^0}\right| = kT\left(\frac{\partial\langle u\rangle}{\partial B}\right)_{B=0} = 4\left(\frac{\partial\theta}{\partial \ln s}\right)_{s=1} \tag{15}$$

(The midpoint of transition is determined by the condition $s = 1$ and is independent of σ. Therefore $(\partial\theta/\partial \ln \sigma)_{s=1}$ vanishes and we have used $(\partial\theta/\partial T) = (\partial\theta/\partial \ln s)(\partial \ln s/\partial T)$ in eq. (15)).

The last expression in eq. (15) may be identified as 4 times the Γ-factor related to the amplitude of kinetic measurements [3] at the midpoint of transition.

Now we have arrived at the point where we can merge our considerations about correlation and energetic interactions. This is accomplished by virtue of the dissipation–fluctuation theorem. It states that the system cannot decide whether a small change in state arises from spontaneous fluctuations (as expressed by the correlation function) or from a small change in external variables (as expressed by $\partial/\partial B$). Using this theorem [13] together with eq. (15), it is found that [14]

$$\text{c.u.} = \sum_r \Gamma_r \tag{16}$$

where the sum is taken over all lattice points.

This result offers a precise interpretation of the c.u. in terms of the correlation function. We may say in a simplified manner, that the c.u. represents an average cluster of strongly correlated molecules at the midpoint of transition.

The physical meaning of the cooperative unit becomes even more evident if we notice that eq. (16) is the extrapolation to $k \to 0$ of the Fourier transform

$$\sum_r \Gamma_r \exp(-\mathrm{i}\boldsymbol{k}\cdot\boldsymbol{r})$$

This, in turn, is known to equal the normalized intensity $I(k)/I^0(k)$ obtained in a scattering experiment, at a scattering vector k [15]. Symbol I^0 is the intensity which would be observed if all particles were independent. Therefore the c.u. is just the extrapolation of the scattering intensity to zero scattering angle (i.e. $k \to 0$):

$$\text{c.u.} = \lim_{k\to 0} I(k)/I^0(k). \tag{17}$$

This is currently interpreted as the cluster size "seen" in the scattering experiment.

Furthermore, eq. (16) represents a generalization of the concept of all-or-none transitions: In the all-or-none case groups of n particles change state simultaneously. These n particles are therefore strongly correlated, whereas the correlation with the rest of the lattice vanishes. Summing up Γ_r over the whole lattice thus yields c.u. = n.

For the linear Ising model, the general cluster definition of eq. (16) essentially coincides with the average sequence length as defined e.g. by Lifson [16]. (Strictly speaking the two definitions differ by unity due

Table 1
Relations between cooperative unit, cooperativity parameter and correlation length for various lattices

lattice a)	coord. number z	$\sqrt{\sigma_c}$	series expansions b)		quasichemical approx.	
			c.u.$^{-1}$	c.u.	$\sqrt{\sigma_c}$	c.u.$^{-1}$
linear	2	0	y	$2.00\ \xi$	0	y
sq	4	$\sqrt{2}-1$	$4.86\ y^{1.75}$	$2.60\ \xi^{1.75}$	1/2	$2y$
hex	6	$1/\sqrt{3}$	$7.41\ y^{1.75}$	$2.85\ \xi^{1.75}$	2/3	$3y$
sc	6	0.642	$4.35\ y^{1.25}$	$4.44\ \xi^{1.944}$	2/3	$3y$
bcc	8	0.730	$6.21\ y^{1.25}$	$4.77\ \xi^{1.944}$	3/4	$4y$
fcc	12	0.815	$9.57\ y^{1.25}$	$4.93\ \xi^{1.944}$	5/6	$6y$

a) Abbreviations are: sq = plane square lattice; hex = plane hexagonal ("triangular") lattice; bcc = body-centered cubic lattice; fcc = face-centered cubic lattice.

b) Calculated from the results of ref. [13] with some simplifications. The c.u.$^{-1}$ and c.u. relations in columns 4 and 5 are valid for very small y and large ξ, resp., roughly corresponding to c.u. of the order of some hundreds or larger. For smaller values of c.u. the formulae still provide a good estimate. Note that $\sigma \approx \sigma_c$ in most practical cases, as a rough first estimate.

to negative correlations with the elements next to the ends of the sequence).

In fact, the linear lattice correlation function is of the form [15]:

$$\Gamma_r = e^{-r/\xi} \tag{18}$$

The correlation length is an expression of the average range of correlations. It is given by [15]:

$$\xi^{-1} = \ln \frac{1+\sqrt{\sigma}}{1-\sqrt{\sigma}} \approx 2\sqrt{\sigma}; \tag{19}$$

Summing up Γ_r over the whole lattice, we obtain:

$$\text{c.u.} = 1/\sqrt{\sigma} = 2\xi, \tag{20}$$

ξ is thus the "radius" or the half-length of a sequence.

In the nonlinear cases the situation is more complex. In three dimensions e.g. the classical expression of the correlation function reads for large values of r [15]:

$$\Gamma_r = e^{-r/\xi}/r \tag{21}$$

The factor $1/r$ causes a more rapid decay of the correlation function than a pure exponential. In particular, at the critical point where ξ goes to infinity [13] and the exponential approches unity, Γ_r still decays as $1/r$. The correlation length is therefore systematically larger than the radius of an average "correlation cluster". This explains the otherwise astonishing fact that the c.u. is found to be proportional to ξ^2 and not to ξ^3:

$$\text{c.u.} = \alpha\xi^2 \tag{22}$$

where α varies only weakly with the critical parameters. The two-dimensional case resembles the linear one in the sense that in the classical (approximate) theories the c.u. is found to be proportional to ξ^2 [13], making it possible to interpret ξ as an average cluster radius.

In the modern development of the Ising theory it has been shown that the simple ξ^2 behaviour has to be slightly modified. Near the critical point, i.e. for large ξ, it can generally be written according to Fisher [17]:

$$\text{c.u.} = \alpha\xi^{2-\eta} \tag{23}$$

with α nearly constant, $\eta = 0.25$ in two dimensions and $\eta \approx 0$ in three dimensions. (In the last case an exact value of η is unknown, but current estimates are of the order of $\eta \approx 0.055$). The formula (23) may be extended to linear lattices by defining $\eta = 1$ for this case.

The concept of correlation lengths plays an important role in the discussion of nonideal systems [18,19] (see also section 4). Therefore we list eq. (23) in table 1 for some important lattices. The numerical values for α are taken from ref. [13].

3. Cooperative unit and cooperativity parameter

In order to obtain direct information about the interaction energies between lattice elements, the c.u. has to be related to the cooperativity parameter σ as defined in eq. (11). For linear lattices, the well-known relation c.u. = $\sigma^{-1/2}$ holds implying that large cooperative units correspond to small values of σ. The situation is different in two- and three-dimensional systems due to the existence of a finite critical value σ_c of the cooperativity parameter. For $\sigma = \sigma_c$ the midpoint slope becomes infinite, for $\sigma < \sigma_c$ the transition curve is discontinuous (cf. fig. 1). From ref. [13] we find typical σ_c-values for nonlinear lattices in the range between 0.17 (plane square) and 0.66 (face-centered cubic) whereas $\sigma_c = 0$ in the linear case. Thus large cooperative units may be obtained (in fact infinite ones at $\sigma = \sigma_c$) even for comparatively large values of σ. For the face-centered cubic lattice, for instance, $\sigma = 0.67$ leads to nearly infinite c.u., because σ is very close to σ_c. For the linear lattice, the same value of σ would be barely distinguishable from non-cooperativity ($\sigma = 1$). It appears that the relevant variable is not σ itself but its difference from the critical value σ_c. Therefore we introduce the new variable

$$y = \sqrt{\sigma} - \sqrt{\sigma_c} \qquad (24)$$

(The reason for using square roots will become evident later). For the linear lattice y coincides with $\sqrt{\sigma}$.

We now want to express c.u. as a function of y. An exact and general formula does not exist for plane and space lattices, but we shall present some useful relations valid in the limit of small y as well as a less exact, but general and analytical approximation method.

In table 1, columns 3 and 4, we give $\sqrt{\sigma_c}$ for various lattices and the inverse cooperative unit as a function of y. These relations have been calculated from the very accurate results of Fisher and Burford [13] arranging them into a simplified form which we think to be well adapted for application in practical work. The relations of column 4 are valid for σ very close to, but still larger than σ_c — *this is just the most interesting* case of large (but finite) c.u.. Despite their high precision, the results of Fisher and Burford have the disadvantage of being derived from very sophisticated series expansions which makes it difficult to see how they come about. To fill this gap and to provide a more intuitive understanding of the phenomena occurring in nonlinear cooperative systems, we shall briefly present the quasichemical approximation of the Ising model which allows a general and analytical description. The most important advantage of the method is that it gives information not only about the midpoint of transition, but that the complete transition curve can be calculated as a function of the cooperativity parameter σ and the equilibrium constant s defined in eqs. (12) and (13).

The dimensionality of the lattice is not explicitly incorporated into this formalism, but the lattice character is taken into account by the number of nearest neighbours of a lattice point (coordination number), z. In the linear case, for $z = 2$, the exact results are found.

Let $p_2 = \theta$ be the probability for finding an element in state 2, and $p_1 = 1 - \theta$ for finding it in state 1. Similarly, let p_{11}, p_{22} and $p_{12} = p_{21}$ be the probabilities for finding a pair of neighbours both in state 1, both in state 2 and one in state 1, the other in state 2, resp. The equality $p_{12} = p_{21}$ must hold for symmetry reasons in infinitely large systems and in lattices with periodic boundary conditions. In principle, the consideration of triplet, quadruplet etc. probabilities would be required. But the basic assumption of the quasichemical approximation is that this series of probabilities can be reduced to single-element and pair probabilities as defined above. This "doublet closure" is clearly an approximation for general lattices, but can be shown to hold rigorously for linear lattices [20].

The probability of finding an element in state i with n of its nearest neighbours in state 2 and $(z - n)$ in state 1 then is expressed by

$$(p_i)_n = p_i (p_{i2}/p_i)^n (p_{i1}/p_i)^{z-n}, \qquad (25)$$

where the terms in brackets on the right-hand side are the conditional probabilities of forming $i1$ and $i2$ pairs, presuming that the central particle is in the i state. According to thermodynamics, the equilibrium probabilities for $(p_1)_n$ and $(p_2)_n$ obey

$$(p_2)_n/(p_1)_n = \exp\{-(1/kT)[w_2 - w_1 + n(w_{22} - w_{12}) + (z - n)(w_{12} - w_{11})]\}, \qquad (26)$$

where the energy parameters w_i, w_{ij} are the same as in section 2, eqs. (7)–(10), if we neglect the difference between Helmholtz and Gibbs free energies. Upon separation of the n-dependent terms, this equation reads:

$$(p_2)_n/(p_1)_n = K_0 q^n, \tag{27}$$

with

$$kT \ln K_0 = (w_1 - w_2) - z(w_{12} - w_{11}), \tag{28}$$

$$kT \ln q = -(w_{11} + w_{22}) + 2w_{12}. \tag{29}$$

A comparison with eqs. (9)–(12) yields that $q = \sigma^{-1}$ and $K_0 = \sigma^{z/2} s$.

Using eq. (25) to compute eq. (27) for $n = 0$ and $n = 1$, one finds the so-called "quasichemical equation"

$$\sigma^{-1} = p_{11} p_{22} / p_{12}^2 \tag{30}$$

and

$$s = (p_{22}/p_{11})^{z/2} (p_1/p_2)^{z-1}. \tag{31}$$

The five probability parameters may finally be reduced to two independent ones using the logical relations [21]:

$$p_1 + p_2 = 1, \qquad p_{11} + p_{12} = p_1,$$

$$p_{22} + p_{12} = p_2. \tag{32a,b,c}$$

Choosing $p_2 = \theta$ and p_{12} as the independent parameters, we finally find [22]:

$$p_{12} = \frac{(\beta - 1)\sigma}{2(1 - \sigma)} \tag{33}$$

$$s = \left(\frac{1 - \theta}{\theta}\right)^{(z/2)-1} \left(\frac{\beta - 1 + 2\theta}{\beta + 1 - 2\theta}\right)^{z/2}, \tag{34}$$

$$\text{with } \beta = [1 + 4\theta(1 - \theta)(1 - \sigma)/\sigma]^{1/2}. \tag{35}$$

Using these formulae together with eq. (13) complete thermal transition curves may be computed, with σ as a parameter.

For σ larger than a critical value σ_c, eq. (34) has only one real solution. For $\sigma < \sigma_c$, a region with three real solutions is found (see fig. 1).

An analysis of the free energies then shows that the loops are instable (dotted lines) or metastable (dashed lines). This is the domain of first-order transitions. If upon change of thermodynamic conditions the system moves along the metastable branches, hysteresis effects will occur. For $\sigma = \sigma_c$ the midpoint slope of the transition curve is infinite, but no loops appear (second-order transition).

From eq. (34) we find that $(\partial\theta/\partial \ln \sigma)_{s=1} = 0$.

Calculating $(\partial\theta/\partial \ln s)$ and $(\partial \ln s/\partial T)$ from eqs. (34) and (13) we obtain

$$\text{c.u.} = \left(\frac{z}{2}\sqrt{\sigma} - \left(\frac{z-2}{2}\right)\right)^{-1} \tag{36}$$

This is the generalization of the expression c.u. = $\sigma^{-1/2}$ of the linear Ising model (which is obtained by setting $z = 2$). The critical value σ_c is determined by the condition that c.u. becomes infinite. Thus

$$\sqrt{\sigma_c} = (z - 2)/z \tag{37}$$

For the linear lattice σ_c vanishes, i.e. first-order transitions are impossible.

We find it very instructive to rearrange eq. (36) into the following form:

$$y = \sqrt{\sigma} - \sqrt{\sigma_c} = \left(\frac{z}{2}\,\text{c.u.}\right)^{-1} \tag{38}$$

Thus the inverse cooperative unit is directly proportional to the difference between $\sqrt{\sigma}$ and the critical value $\sqrt{\sigma_c}$.

Eqs. (37) and (38) are listed in table 1 for various lattices. Comparison with the more exact series expansion results shows that the direct proportionality of c.u.$^{-1}$ and y is a strong simplification. However, for σ not too close to σ_c the series expansion formulae of table 1 (column 4) become less accurate whereas the quasichemical approximation becomes better. The discrepancies of the $\sqrt{\sigma_c}$ values are of less importance since the relevant variable is $y = (\sqrt{\sigma} - \sqrt{\sigma_c})$. For consistency, $\sqrt{\sigma_c}$ must be taken from eq. (37) or column 6 of table 1 when working with the quasichemical approximation.

It is easy to see from eq. (37) that σ_c increases with increasing z: steep transitions may be obtained even for comparatively large σ if the cooperative effect is multiplied by numerous nearest neighbours. As a consequence, the transition curves may show a very slow approach to the $\theta = 0$ and $\theta = 1$ asymptotes, as demonstrated by the curves of fig. 1. (This effect remains the same when we correct for a temperature dependence of the cooperativity parameter such that $\ln \sigma = const/T$. Cf. e.g. the transition curves in ref. [23] to make sure that the phenomenon does not depend upon a special method). For a more quantitative understanding consider a particle which goes from state 1 to state 2 with a) all its neighbours in state 1, b) half of its neighbours in state 1, the other half in state 2. The situation a) corresponds to the nucleation

Table 2
Theoretical estimates and experimental values of the cooperative unit for small vesicles of dimyristoylphosphatidylcholine

R_m [Å]	ξ_{max}	c.u. (ξ_{max})	c.u. ($\xi_{max}/5$)	c.u.$_{exp}$ a)	c.u.$_{exp}$ b)
162.5	30.75	1145	68	132	47
182.5	34.54	1402	84	149	58
222.5	42.11	1984	119	142	64
262.5	49.68	2649	158	149	77
302.5	57.25	3396	203	133	77
342.5	64.82	4220	252	152	98

Theoretical values are calculated according to the finite-size concept using eqs. (39) and (40). For the determination of ξ_{max} we use the "average radius" R_m which equals the measured outer vesicle radius minus 17.5 Å, and the lattice site distance $d = 8.3$ Å corresponding to an average area per lipid molecule of 60 Å^2. Experimental values are from the light-scattering data of ref. [25] using (a) the radius-dependent transition enthalpies given in the same article and (b) the radius-independent value $\Delta H^0 = 27.2$ kJ mole^{-1}.

of a cluster, b) is an example of cooperative growth. The relative probability of the two processes is found from eq. (27) to be

$$\frac{\text{probab. of nucleation}}{\text{probab. of growth}} = \sigma^{z/2}$$

For a planar hexagonal lattice ($z = 6, \sqrt{\sigma_c} = 2/3$) this yields only 0.09 at the critical point, i.e. the probability of nucleation is still remarkable even for an infinitely steep transition. These results seem to be supported e.g. by some experiments on lipid bilayer phase transitions showing substantial variations of membrane parameters even rather far above the main transition [24].

Consequently, special attention has to be paid to baseline problems in nonlinear systems. Otherwise the transition enthalpy ΔH^0 will tend to be systematically underestimated in the evaluation of calorimetric data. Also, ΔH_{vH} may easily be overestimated if the reference lines for $\theta = 0$ and $\theta = 1$ are chosen too close to the experimental transition curves.

The problem is much less important for first-order transitions for which σ is generally smaller (at least, $\sigma < \sigma_c$).

4. Nonideal systems

It has become clear in the preceding sections that two- and three-dimensional systems may show some characteristics which are fundamentally different from those of linear systems. Most important is the existence of first-order transitions (for $\sigma < \sigma_c$) where a strictly vertical step connects the two continuous branches of the transition curve (cf. fig. 1). In the following, we shall assume that this step occurs (at least ideally) at T_m, thereby neglecting the case of hysteresis. We will show that some nonidealities of the systems may cause the strict discontinuity to be smeared out. The transition curve will then be continuous, with an apparently finite midpoint slope and thus finite c.u.. In these cases, the c.u. yields information about the degree of perturbation rather than about the interaction energies of the ideal system [14].

One such nonideality which has already aroused some interest [8,23] is the smallness of the observed systems. Strictly speaking, no finite system can ever display a discontinuity, because its partition function is a finite sum of analytical functions of the type $\exp(-E/kT)$. But the region of deviation from ideality will usually be extremely small and escape detection even by the most precise instruments. In the theory of finite Ising models it has been estimated that the "range of rounding" is of the order of $1/n$ for a lattice containing n particles in each dimension ($n \times \ldots \times n$ – lattice) [18]. This corresponds to a range of $\Delta T/T = 10^{-6}\%$ if the system contains one millimole of substance. However, small lipid vesicles form bilayers which contain only some ten to hundred thousand lipid molecules: finite size effects should then be observable.

In section 2 it has been shown that the cooperative unit represents the average size of a "correlation cluster". Remembering that at transition midpoint half of the lattice elements are in each state 1 and 2, we understand that the largest possible cluster on the surface of such a vesicle would cover half of the sphere of radius R. For the correlation length an analogous upper bound is given by a figure of the order of $\pi R/2$. Since we have introduced ξ as a dimensionless parameter for the counting of lattice sites (see sect. 2), we must divide by the site distance d to obtain

$$\xi_{\max} = \pi R/2d. \tag{39}$$

Applying the relation of table 1 for a hexagonal lattice, we find the following upper bound for the c.u.:

$$\text{c.u.} \lesssim 2.85\,(\xi_{\max})^{1.75} = 2.85\,(\pi R/2d)^{1.75} \tag{40}$$

Similar formulae may easily be derived for other lattices and other shapes of the system, using the relations of table 1.

It should be noted that the bounds calculated from the correlation length are much lower than those obtained directly from setting c.u. $\leqslant$ half of the vesicle surface (cf. [8]). This is due to the exponential 1.75 in eq. (40).

We have applied eq. (40) to the experimental data known from small dimyristoyl-phosphatidylcholine vesicles [25]. The results are listed in table 2. The calculated upper bounds are clearly larger than the observed values. Since the average correlation length must be much smaller than the maximum one, we also have tentatively set $\xi = \xi_{\max}/5$ instead of $\xi_{\max}$ in eq. (40). It may be seen from table 2 that the calculated cooperative units increase much faster with R than the experimentally obtained ones. Therefore we think that other effects, especially lattice defects due to the constraints of a strongly curved surface, will play an important role. These may be treated similarly as impurities which represent the major source of non-ideality in most of the interesting biological systems. Indeed a significant increase of c.u. with the improvement of chemical purification has been reported for liposomes [9].

"Smearing-out" of first-order discontinuities will certainly occur if the system contains impurities which have a strong preference for one of the two phases. It is well known from general thermodynamics that the transition temperature T_m is shifted to lower values if the impurity partitions preferentially into the more liquid environment (and to higher values in the opposite case). During the transition the impurity accumulates in this phase, giving rise to a continuous shift of the transition curve to the lower (higher) temperature region. As a consequence, the c.u. is found to be finite and to decrease with increasing concentration of impurity. A simple thermodynamical treatment of this phenomenon [26] has been applied to the evaluation of lipid phase transition data by Albon and Sturtevant [9]. In this calculation, infinite cooperativity is assumed for the pure material, i.e. a transition proceeding in one vertical step from $\theta = 0$ to $\theta = 1$, at a certain ideal transition temperature.

It would certainly exceed the scope of this article to present a general treatment of the different sorts of impurities and lattice inhomogeneities for systems of finite cooperativity. Local distributions of lattice perturbations and specific interactions between the impurities and those lattice elements which are subject to transition need to be considered in most cases. Some important aspects will be discussed in a separate article with special application to lipid bilayers containing intrinsic molecules.

In the following, we shall give some simple relations which hold in the case of "impurities" which show no partitioning between the phases, but are essentially randomly distributed throughout the lattice. This may be considered as an approximate model for certain types of lattice defects. It also holds for some probe molecules inserted into systems to monitor the phase transition (e.g. DPH in lipid bilayers [27]).

For these lattice perturbations, no shift of transition temperature occurs. Nevertheless, the c.u. can decrease, because the presence of the perturbation abolishes some interactions between the particles of the pure material. This is easily formulated in the framework of the quasichemical approximation. We simply assume that the average number of nearest neighbours is smaller than in the ideal case, and introduce an effective coordination number, z_{eff} [28].

If the impurities occupy a fraction x of the lattice sites, we have:

$$z_{\text{eff}} = z_{\text{ideal}}(1 - x) \tag{41}$$

The critical value of the cooperativity parameter then becomes a function of x, namely (cf. eq. (37))

$$(\sigma_c^{\text{eff}})^{1/2} = (z_{\text{eff}} - 2)/z_{\text{eff}} \tag{42}$$

The effective critical value of σ decreases with decreasing z_{eff}, i.e. with increasing x. For $z_{\text{eff}} = 2$, $\sigma_c^{\text{eff}} = 0$ and a critical behaviour is no longer possible. This corresponds to the case of the linear lattice. Remembering that the c.u. is inversely proportional to the difference $y = (\sqrt{\sigma} - \sqrt{\sigma_c})$, according to eq. (38) an effective decrease of the critical σ will make this difference larger and thus yield smaller cooperative units. These results remain qualitatively correct, at least for small x, even in a more rigorous treatment of the Ising model with impurities (see e.g. [29]).

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