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# Theory of multivalent binding in one and two-dimensional lattices

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**Abstract**

Ligand binding to a linear lattice composed of $N$ sites, under general conditions of cooperativity and number of sites covered upon binding, $m$, is approached in terms of the theory of contracted partition functions. The partition function of the system obeys a recursion relation leading to a generating function that provides an exact analytical solution for any case of interest. Site-specific properties of the lattice are derived from simple transformations of the analytical expressions. The McGhee–von Hippel model is obtained as a special case in the limit $N \to \infty$. The derivation is straightforward and involves no combinatorial arguments. Partition functions and site-specific properties are also derived for the case of non-cooperative binding to a two-dimensional torus of length $N$, containing $s$ sites in its section for a total of $sN$ sites. The torus provides a relevant model for ligand binding to double-stranded DNA ($s = 2$) or protein helices ($s = 3,4$). It is proved that non-cooperative binding to the two-dimensional torus can mimic cooperative binding to a one-dimensional linear lattice when $m = s$. The dimensional embedding of the lattice and the geometry of interaction of its sites play a crucial role in defining the binding properties of the system accessible to experimental measurements. Hence, caution must be exercised in the interpretation of Scatchard plots in terms of the one-dimensional McGhee–von Hippel model, especially when $m \leq 4$ and the geometry of the system is clearly two-dimensional.

*Keywords:* Binding; Cooperativity; Ising problem; Protein-DNA interactions; Scatchard plot

## 1. Introduction

For a variety of biologically relevant problems, including but not limited to the binding of small ligands or macromolecules to nucleic acids or to clusters of receptors on the surface of a cell, the description of binding equilibria requires extension of the theories of binding and linkage [1,2]. When the molecular surface of recognition spans multiple elementary binding sites, ligand binding to a given domain composed of multiple sites prevents binding of other ligands to the same domain or parts of it. This phenomenon, first surfaced in the study of protein-nucleic acid interactions [3,4], is however far more general. In fact, practically all binding domains in a protein contain multiple epitopes in the form of individual amino acid residues and the specificity pocket of several enzymes can also be partitioned among distinct recognition subsites. Hence, understanding

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multivalent binding, that is the binding of a ligand to multiple individual sites of a lattice, may shed light on the molecular basis of macromolecular interactions in general.

Multivalent binding to a one-dimensional linear lattice of infinite length has been treated by McGhee and von Hippel [4] using an ingenious, though somewhat lengthy combinatorial approach. A treatment of similar effects in infinitely long lattices has been offered by Schwarz [5]. This treatment applies standard techniques of matrix algebra pioneered by Zimm [6] and Lifson [7] and endorsed by many others [8–10]. The treatment of multivalent binding to a linear lattice of finite length is considerably more complex. Epstein has offered a combinatorial analysis [11], while Szabo has developed a more interesting approach using a transfer matrix [12]. All these approaches, however, lack the generality necessary to dissect the effects at the site-specific level. More importantly, none of the previous approaches offers ways to extend the analysis of multivalent binding to two-dimensional lattices, that model more closely the properties of biological systems of interest.

In this article we develop an entirely new approach to multivalent binding based on the principles of site-specific thermodynamics and the properties of contracted partition functions [13,14]. We derive a generating function for the partition function of the system in the case of a one-dimensional linear lattice of arbitrary length and cooperativity and derive the classical McGhee–von Hippel model as a special case in a straightforward manner. We also show how this approach naturally lends itself to consideration of site-specific properties of the lattice and to extension to the analysis of two-dimensional lattices.

## 2. Recursion relation for the partition function

We are interested in the description of multivalent binding to a linear lattice containing $N$ identical sites that can be treated as a one-dimensional Ising network [15] as shown in Fig. 1. When the multivalent ligand binds to the lattice, it covers $m$ adjacent sites. The equilibrium binding constant for the transition free $\rightarrow$ bound at each site is $K$, while $x$ denotes the ligand activity. These two parameters can be merged to define a scaled activity variable $\omega = Kx$. When two ligands contact each other upon binding to the linear lattice, they experience a nearest-neighbor interaction parameterized by the dimensionless term $\sigma$. The value of $\sigma$ signals the presence of positive ($\sigma > 1$) or negative ($\sigma < 1$) coupling, with $\sigma = 1$ referring to the absence of cooperativity. In the case $m > 1$, a distinction should be made between the number of ligated sites in the lattice, $X$, and the number of bound ligands, $L$. These quantities coincide only if binding of the ligand results in the occupation of one site (univalent binding). In the case of multivalent binding ($m > 1$), the value of $L$ is always less than $X$. In general, while $X$ is bound from 0 to $N$, the quantity $L$ is bound from 0 to $N/m$.

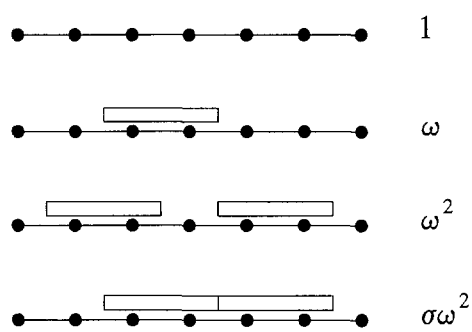


Fig. 1. Multivalent binding to a one-dimensional lattice. The ligand covers $m$ sites of the lattice upon binding. Two adjacent ligands experience an interaction $\sigma$ when bound to neighbor sites. The four configurations in the lattice map into corresponding terms in the partition function, as shown at right.

The problem we consider next is to find the analytical form of the partition function of the system for given $N$ and $m$. We cast our analysis in terms of site-specific thermodynamics [13,14]. Consider multivalent binding to a one-dimensional ring obtained from the linear lattice by connecting the end sites. The solutions of the problem for the ring and the linear lattice are connected through a simple transformation, first documented for the case of $m = 1$ [14]. Let ${}^{R}\Psi_{(N)}$ be the partition function for multivalent binding to the ring of $N$ sites. Then, the average number of ligands bound to the lattice is evidently $L = \mathrm{d}\ln {}^{R}\Psi_{(N)}/\mathrm{d}\ln\omega$. Since a bound ligand covers exactly $m$ sites, the average number of ligated sites is $X = Lm$. There are $N$ sites in the lattice that are identical and equivalent since the ring is homogeneous [14,15]. Therefore, the probability that the $j$th site is bound is simply $X_{j(N)} = X/N = Lm/N$. Using contracted partition functions [13,14], the probability that the $j$th site in the ring is bound is given by

$$X_{j(N)} = 1 - \frac{{}^{0,R}\Psi_{j(N)}}{{}^{R}\Psi_{(N)}} = \frac{X}{N} = \frac{Lm}{N} = \frac{m}{N}\frac{\mathrm{d}\ln {}^{R}\Psi_{(N)}}{\mathrm{d}\ln\omega} \tag{1}$$

where ${}^{0,R}\Psi_{j(N)}$ is the contracted partition function of the ring when site $j$ is kept unligated. This contracted partition function is the same as the partition function of a linear lattice of $N-1$ sites, because keeping a site unligated under the stated interaction rule (see Fig. 1) is equivalent to eliminating that site from the lattice [14]. Hence,

$${}^{L}\Psi_{(N-1)} = {}^{R}\Psi_{(N)} - \frac{m}{N}\frac{\mathrm{d}{}^{R}\Psi_{(N)}}{\mathrm{d}\ln\omega} \tag{2}$$

is a very basic transformation that connects the partition functions of the ring and the linear lattice. This relation generalizes the result for univalent binding reported previously [14]. From Eq. (2) it follows that if

$${}^{R}\Psi_{(N)} = \sum_{j=1}^{m+1} \lambda_j^N \tag{3}$$

is the solution for the ring, where the $\lambda$s are proper eigenvalues independent of $N$, then the solution for the linear lattice of $N$ sites is

$${}^{L}\Psi_{(N)} = \sum_{j=1}^{m+1} \lambda_j^N\left(\lambda_j - m\lambda_j'\right) \tag{4}$$

where $\lambda_j' = \mathrm{d}\lambda_j/\mathrm{d}\ln\omega$.

We now prove that Eqs. (3) and (4) are the solutions for the multivalent binding problem. To derive the partition function we consider the linear lattice in Fig. 1. It follows from site-specific thermodynamics [13,14] that for any partition function $\Psi$

$$\Psi = {}^{0}\Psi_j + {}^{1}\Psi_j K_j x \tag{5}$$

${}^{0}\Psi_j$ denotes the contracted partition function with site $j$ kept unligated and ${}^{1}\Psi_j$ is the analogous partition function with site $j$ kept ligated. $K_j$ is the site-specific binding constant for site $j$. Eq. (5) applies regardless of the site $j$ chosen and also holds for any contracted partition function of first or higher order [14]. If Eq. (5) is applied to the linear lattice in Fig. 1 by contracting over the end site $N$ (contracting over site 1 leads of course to the same result) the following relation holds

$${}^{L}\Psi_{(N)} = {}^{0,L}\Psi_{N(N)} + \omega^{1,L}\Psi_{N(N)} = {}^{L}\Psi_{(N-1)} + \omega^{1,L}\Psi_{N(N)} \tag{6}$$

${}^{0,L}\Psi_{N(N)}$ is the same as the partition function of the linear lattice of $N-1$ sites, ${}^{L}\Psi_{(N-1)}$. The contracted partition function ${}^{1,L}\Psi_{N(N)}$ can be evaluated by contracting over site $N-m$ as follows

$${}^{1,L}\Psi_{N(N)} = {}^{01,L}\Psi_{N-m,N(N)} + \sigma\omega^{11,L}\Psi_{N-m,N(N)} = {}^{L}\Psi_{(N-m-1)} + \sigma\omega^{11,L}\Psi_{N-m,N(N)} \tag{7}$$

The factor $\sigma$ reflects the interaction between the bound ligands covering sites $N$ and $m$. In fact, when these sites are both bound, then two ligands must necessarily be bound to the end of the lattice and be in contact with each other. Clearly, ${}^{01,L}\Psi_{N-m,N(N)} = {}^{L}\Psi_{(N-m-1)}$ because keeping site $N-m$ unligated cuts the original lattice into two smaller lattices of length $N-m-1$ and $m$, with the latter being bound to the ligand. Likewise,

$$\omega^{11,L}\Psi_{N-m,N(N)} = \omega^{1,L}\Psi_{N-m(N-m)} = {}^{L}\Psi_{(N-m)} - {}^{L}\Psi_{(N-m-1)} \tag{8}$$

In fact, ${}^{11,L}\Psi_{N-m,N(N)} = {}^{1,L}\Psi_{N-m(N-m)}$ because the bound ligand at the end of the lattice does not influence the other sites when a second ligand molecule covers site $N-m$. Substitution of Eq. (8) into Eq. (7) and then into Eq. (6) leads to

$${}^{L}\Psi_{(N)} = {}^{L}\Psi_{(N-1)} + \sigma\omega{}^{L}\Psi_{(N-m)} - (\sigma-1)\,\omega{}^{L}\Psi_{(N-m-1)} \tag{9}$$

which is a recursion relation for the partition function of the linear lattice. The same relation holds for the ring, since Eq. (2) is a linear transformation of the partition functions. Hence,

$${}^{R}\Psi_{(N)} = {}^{R}\Psi_{(N-1)} + \sigma\omega{}^{R}\Psi_{(N-m)} - (\sigma-1)\,\omega{}^{R}\Psi_{(N-m-1)} \tag{10}$$

The solution of Eqs. (9) and (10) is of the form

$$\Psi_{(N)} = \sum_{j=1}^{m+1} \alpha_j \lambda_j^N \tag{11}$$

where the $\alpha s$ are independent of $N$ and the $\lambda$s are solutions of the expression

$$\lambda^{m+1} - \lambda^m - \sigma\omega\lambda + (\sigma-1)\,\omega = 0 \tag{12}$$

It is easy to verify that for the ring $\alpha_1 = \alpha_2 = \ldots = \alpha_{m+1} = 1$, since ${}^{R}\Psi_{(1)}$ is $1 + \sigma\omega$ for $m = 1$ and 1 otherwise, and so is the sum of the $\lambda$s. The analogous values of the $\alpha$s for the linear lattice are derived from Eq. (2), so that Eqs. (3) and (4) apply.

We have so derived the partition functions of the ring and the linear lattice from application of contracted partition functions, without any consideration of lengthy combinatorial arguments or the use of a transfer matrix. The importance of Eq. (8) is twofold. First, it offers a relation for the partition function of the system from which all eigenvalues $\lambda$s can be derived directly. Eq. (12), which is a consequence of Eq. (8), generalizes the result obtained previously on the largest eigenvalue derived from matrix approaches [5] or the method of sequence generating functions [16]. Second, it can be used to derive the partition function of the system without any knowledge of the eigenvalues.

## 3. Alternative solution from the transfer matrix

It is quite instructive to derive Eq. (12) using a transfer matrix, which echoes a previous analysis of multivalent binding by Szabo [12]. We will then merge the site-specific approach and the transfer matrix method in the analysis of multivalent binding to a two-dimensional lattice. To construct the transfer matrix, we consider two neighbor sites on the lattice, as shown in Fig. 2. If the ligand covers $m$ sites upon binding, then there are $m+1$ possible states for each site, 1 unligated and $m$ ligated. The simple case $m = 1$ leads to the following $2 \times 2$ transfer matrix

$$\mathbf{W} = \begin{pmatrix} 1 & 1 \\ \omega & \sigma\omega \end{pmatrix} \tag{13}$$

The first column reflects the possible states of a site when the neighbor site is unligated and the second column gives the states of the site when the neighbor site is ligated. When $m = 2$, there are two possible ligated states

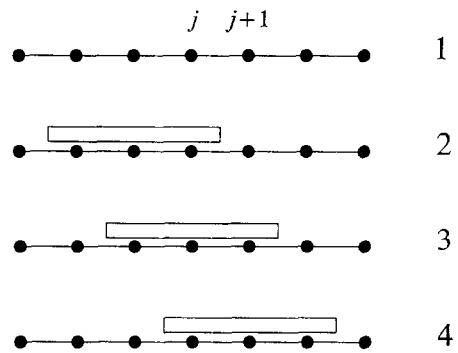


Fig. 2. Possible binding modes for a trivalent ($m = 3$) ligand interacting with a one-dimensional lattice. The allowable configurations for site $j$ are shown and can be compared with those of site $j+1$. These configurations, when translated into proper energetic terms, define the transfer matrix for multivalent binding (see Eq. (15) in the text).

for each site, depending on whether the ligand covers it with its beginning or ending portion. The $3 \times 3$ transfer matrix is

$$\mathbf{W} = \begin{pmatrix} 1 & 0 & 1 \\ \omega^{1/2} & 0 & \sigma\omega^{1/2} \\ 0 & \omega^{1/2} & 0 \end{pmatrix} \tag{14}$$

The term $\omega$ appears with the power of $1/2$ because each site is covered by $1/2$ ligand. The terms in the first column reflect the allowable states for a site when the neighbor site is unligated. The $\omega^{1/2}$ term indicates that the ligand can bind at the site with its ending portion, while the 0 indicates the impossibility of binding with the beginning portion if site $j+1$ is unligated. The term $\sigma\omega^{1/2}$ in the third column indicates that when site $j+1$ has a ligand with the beginning portion on it, another ligand can bind with its ending portion to site $j$ and a nearest-neighbor interaction is experienced for the bound pair. For $m = 3$ (see Fig. 2) one has

$$\mathbf{W} = \begin{pmatrix} 1 & 0 & 0 & 1 \\ \omega^{1/3} & 0 & 0 & \sigma\omega^{1/3} \\ 0 & \omega^{1/3} & 0 & 0 \\ 0 & 0 & \omega^{1/3} & 0 \end{pmatrix} \tag{15}$$

Now there are three bound states for each site, depending on whether the ligand covers it with its beginning, middle or ending portion. In general, the following $(m+1) \times (m+1)$ transfer matrix contains all possible ligation states pertaining to the neighbor sites

$$\mathbf{W} = \begin{pmatrix} 1 & 0 & 0 & \dots & 0 & 1 \\ \omega^{1/m} & 0 & 0 & \dots & 0 & \sigma\omega^{1/m} \\ 0 & \omega^{1/m} & 0 & \dots & 0 & 0 \\ 0 & 0 & \omega^{1/m} & \dots & 0 & 0 \\ \vdots & \vdots & \vdots & \vdots & \vdots & \vdots \\ 0 & 0 & 0 & \dots & \omega^{1/m} & 0 \end{pmatrix} \tag{16}$$

Given the periodic boundary conditions of the ring, its partition function is [6,8,10]

$${}^{R}\Psi_{(N)} = Tr\mathbf{W}^{N} = Tr\mathbf{\Lambda}^{N} = \sum_{j=1}^{m+1} \lambda_j^N \tag{17}$$

where $\Lambda$ is the diagonal form of $\mathbf{W}$ and the $\lambda$s are the eigenvalues derived from the secular equation

$$|\lambda\mathbf{I} - \mathbf{W}| = \lambda^{m+1} - \lambda^m - \sigma\omega\lambda + (\sigma - 1)\omega = 0 \tag{18}$$

which is identical to Eq. (12). Also, Eq. (17) is the same as Eq. (3). Once the partition function of the ring is known, that of the linear lattice is derived from Eq. (2).

## 4. Generating function

A generating function can be derived from the basic Eq. (10) to obtain the analytical form of the partition function in the general case. Let

$$G(\omega,\zeta) = \sum_{N=0}^{\infty} {}^{R}\Psi_{(N)}\zeta^N \tag{19}$$

be the generating function so that the partition function for the ring of $N$ sites is

$${}^{R}\Psi_{(N)} = \frac{1}{N!}\left(\frac{\partial^N G(\omega,\zeta)}{\partial\zeta^N}\right)_{\zeta=0} \tag{20}$$

Once this partition function is known, that of the linear lattice is obtained from Eq. (2). Multiplication of all terms in Eq. (10) by $\zeta^N$ and summation over all possible values of $N$ yields

$$\begin{aligned} &G(\omega,\zeta)\left[1 - \zeta - \sigma\omega\zeta^m + (\sigma - 1)\omega\zeta^{m+1}\right] \\ &\quad = {}^{R}\Psi_{(0)} + \left({}^{R}\Psi_{(1)} - {}^{R}\Psi_{(0)}\right)\zeta + \left({}^{R}\Psi_{(m)} - {}^{R}\Psi_{(m-1)} - \sigma\omega{}^{R}\Psi_{(0)}\right)\zeta^m \\ &\qquad + \left({}^{R}\Psi_{(m+1)} - {}^{R}\Psi_{(m)} - \sigma\omega{}^{R}\Psi_{(1)} + (\sigma - 1)\omega{}^{R}\Psi_{(0)}\right)\zeta^{m+1} \end{aligned} \tag{21}$$

where ${}^{R}\Psi_{(N)} = 1$ for $0 \le N \le m - 1$, ${}^{R}\Psi_{(m)} = 1 + \sigma m\omega$ and ${}^{R}\Psi_{(m+1)} = 1 + (m + 1)\omega$. Substitution into Eq. (21) finally leads to

$$G(\omega,\zeta) = \frac{1 + (m - 1)\sigma\omega\zeta^m - m(\sigma - 1)\omega\zeta^{m+1}}{1 - \zeta - \sigma\omega\zeta^m + (\sigma - 1)\omega\zeta^{m+1}} \tag{22}$$

Similar arguments lead to a generating function for the linear lattice as follows

$$G(\omega,\zeta) = \frac{1 - (\sigma - 1)\omega\zeta^m}{1 - \zeta - \sigma\omega\zeta^m + (\sigma - 1)\omega\zeta^{m+1}} \tag{23}$$

Eqs. (22) and (23) are the general solutions for multivalent binding to a one-dimensional lattice. The partition functions for the ring and linear lattice of $N$ sites can be derived from Eqs. (22) and (23) for any value of $m$, $\sigma$ and $N$. This represents a substantial improvement over earlier approaches to the same problem [11,12].

## 5. Site-specific properties of the lattice

We now turn to the site-specific properties of the linear lattice and compute the probability that a given site is covered by the ligand. From the general theory it follows that [14]

$${}^{L}X_{j(N)} = 1 - \frac{{}^{0,L}\Psi_{j(N)}}{{}^{L}\Psi_{(N)}} \tag{24}$$

where ${}^{L}X_{j(N)}$ is the site-specific binding curve of site $j$ in the linear lattice of length $N$, or the probability that this site is covered by the ligand. ${}^{L}\Psi_{(N)}$ is the partition function of the lattice and ${}^{0,L}\Psi_{j(N)}$ is its contracted form with site $j$ kept unligated. This contracted partition function can be cast in terms of the partition functions of linear lattices of smaller length. When site $j$ is kept unligated, the original lattice is cut into two smaller lattices containing $j-1$ and $N-j$ sites respectively. Hence, ${}^{0,L}\Psi_{j(N)} = {}^{L}\Psi_{(j-1)}\,{}^{L}\Psi_{(N-j)}$ and

$$ {}^{L}X_{j(N)} = 1 - \frac{{}^{L}\Psi_{(j-1)}\,{}^{L}\Psi_{(N-j)}}{{}^{L}\Psi_{(N)}} \tag{25} $$

The expected symmetry of the lattice around the midpoint $M = (N+1)/2$ is evident. Sites equidistant from $M$ have the same binding curve, since the replacement $j \rightarrow N-j+1$ leaves Eq. (25) unchanged. The binding curve of the end site ($j = 1$ or $N$) is particularly important because it provides a connection between the partition function of the lattice and that of a lattice one unit shorter. It follows from Eq. (25) that

$$ {}^{L}X_{N(N)} = 1 - \frac{{}^{L}\Psi_{(N-1)}}{{}^{L}\Psi_{(N)}} \tag{26} $$

Hence,

$$ \begin{aligned} {}^{L}X_{j(N)} &= 1 - \frac{{}^{L}\Psi_{(j-1)}\,{}^{L}\Psi_{(N-j)}}{{}^{L}\Psi_{(N)}} \\ &= 1 - \frac{\left(1-{}^{L}X_{N-j+1(N-j+1)}\right)\left(1-{}^{L}X_{N-j+2(N-j+2)}\right)\cdots\left(1-{}^{L}X_{N(N)}\right)}{\left(1-{}^{L}X_{1(1)}\right)\left(1-{}^{L}X_{2(2)}\right)\cdots\left(1-{}^{L}X_{j-1(j-1)}\right)} \\ &= 1 - \frac{\sum_{i=1}^{m+1}\lambda_i^{j-1}(\lambda_i - m\lambda_i')\sum_{i=1}^{m+1}\lambda_i^{N-j}(\lambda_i - m\lambda_i')}{\sum_{i=1}^{m+1}\lambda_i^{N}(\lambda_i - m\lambda_i')} \end{aligned} \tag{27} $$

The properties of any site in the lattice depend on the properties of the end site of lattices of different length. Hence, knowledge of the behavior of the end site completely defines the properties of any other site in the lattice, as also implied by the probe theorem [17]. This fundamental property of the linear lattice can be exploited in practical applications. Binding isotherms obtained with linear lattices of different length yield the various partition functions in Eq. (27) by numerical integration. Hence, the site-specific properties of the original linear lattice can be constructed using Eq. (27), from measurements of global binding curves and without a need for site-specific probes. In contrast, binding to any site on the ring obeys the expression

$$ {}^{R}X_{j(N)} = 1 - \frac{\sum_{i=1}^{m+1}\lambda_i^{N-1}(\lambda_i - m\lambda_i')}{\sum_{i=1}^{m+1}\lambda_i^{N}} = {}^{R}X_{(N)} \tag{28} $$

independent of the particular site, as expected.

We now consider the behavior of individual sites in the linear lattice in the limit $N \rightarrow \infty$. In this limit we need only worry about the largest eigenvalue, $\lambda_1$, of the expansions 3 and 4. The end site obeys the expression

$$ {}^{L}X_{\infty(\infty)} = 1 - \frac{1}{\lambda_1} \tag{29} $$

while for any other site in the lattice one has

$$ {}^{L}X_{j(\infty)} = 1 - \frac{\lambda_1 - m\lambda_1'}{\lambda_1} = \frac{m\lambda_1'}{\lambda_1} = {}^{R}X_{(\infty)} \tag{30} $$

Since there are infinite such sites in the limit $N \to \infty$, the global binding properties of the ring and linear lattice, like the average number of ligands bound $L$, become identical in the asymptotic limit. However, the end site of the linear lattice always behaves differently, as first noted by Epstein [11]. In the asymptotic limit, the binding probability to the end site defines unequivocally the largest eigenvalue of the transfer matrix and vice versa. In the case of any other sites, the binding probability is defined by the largest eigenvalue and its derivative. Again, knowledge of the behavior of the end site completely and uniquely defines the site-specific and global properties of the entire lattice.

## 6. Derivation of the McGhee–von Hippel model

The properties of multivalent binding to a linear lattice of infinite length can now be derived in a straightforward manner using the foregoing definitions and the basic Eq. (10). Here we derive the McGhee–von Hippel model as a special case in the limit $N \to \infty$. The quantity of interest is ${}^{R}X_{(\infty)}$ since it reflects the average binding probability to a site in the linear lattice (see Eq. (30)) and connects to the average number of bound ligands, $L$, accessible to experimental measurements. In the asymptotic limit, Eqs. (12) and (29) lead to

$$ {}^{L}X_{\infty(\infty)} = \omega\left(1 - {}^{L}X_{\infty(\infty)}\right)^{m}\left[1 + (\sigma - 1)\,{}^{L}X_{\infty(\infty)}\right] \tag{31} $$

To obtain the derivative of the largest eigenvalue we differentiate Eq. (31), so that

$$ {}^{R}X_{(\infty)} = \frac{m\,{}^{L}B_{\infty(\infty)}}{1 - {}^{L}X_{\infty(\infty)}} = \frac{m\,{}^{L}X_{\infty(\infty)}}{1 - {}^{L}X_{\infty(\infty)}} + \frac{m(\sigma - 1)\,{}^{L}B_{\infty(\infty)}}{1 - {}^{L}X_{\infty(\infty)}}\,\frac{{}^{L}X_{\infty(\infty)}}{1 + (\sigma - 1)\,{}^{L}X_{\infty(\infty)}} - \frac{m^2\,{}^{L}B_{\infty(\infty)}\,{}^{L}X_{\infty(\infty)}}{\left(1 - {}^{L}X_{\infty(\infty)}\right)^2} \tag{32} $$

where ${}^{L}B_{\infty} = \mathrm{d}\,{}^{L}X_{\infty(\infty)}/\mathrm{d}\ln\omega$ is the binding capacity of the end site. Introducing the variables $z = {}^{L}X_{\infty}/(1 - {}^{L}X_{\infty(\infty)})$ and $y = {}^{R}X_{(\infty)}/(1 - {}^{R}X_{(\infty)})$ yields the quadratic expression in $z$

$$ \sigma m z^2 + (m - y)\,z - y = 0 \tag{33} $$

which can be solved to obtain the properties of the ring from those of the end site of the linear lattice. The solution is

$$ z = \frac{y - m + \Delta}{2\sigma m} \tag{34} $$

where

$$ \Delta^2 = (m - y)^2 + 4\sigma m y \tag{35} $$

Substitution into Eq. (31) and rearrangement yields the final result

$$ \frac{y}{\omega} = \frac{m + (2\sigma - 1)\,y + \Delta}{2}\left[\frac{2\sigma m}{(2\sigma - 1)m + y + \Delta}\right]^{m} \tag{36} $$

This is equivalent to the relation obtained by McGhee and von Hippel through lengthy combinatorial arguments [4]. The relation is valid for all finite values of $\sigma$. The expression in bracket tends to $(m - y)/m$ for $\sigma = 0$. A

more convenient form for Eq. (36) is given by the transformation ${}^{R}X_{(\infty)} = mv$, where $v = L/N$ is the fraction of ligands bound per site accessible to experimental measurements. The Scatchard form of Eq. (36) is then

$$\frac{v}{x} = \sigma K(1 - mv)\frac{1 + (2\sigma - m - 1)v + Q}{(2\sigma - 1)(1 - mv) + v + Q}\left[\frac{2\sigma(1 - mv)}{(2\sigma - 1)(1 - mv) + v + Q}\right]^{m-1} \tag{37}$$

where

$$Q = \sqrt{[1 - (m + 1)v]^2 + 4\sigma v(1 - mv)} \tag{38}$$

The form in Eq. (37) becomes identical to that derived previously by McGhee and von Hippel [4], after simple transformations (see Appendix).

## 7. Multivalent binding in two dimensions

The foregoing treatment of multivalent binding can be extended to the more realistic and complex scenario of binding to a two-dimensional lattice. Examples of such lattices are given in Figs. 3 and 4. The ladder represents a good model for double-stranded DNA, while the torus with triangular section can serve as a model of a polypeptide helix in a $3_{10}$-conformation. Similar two-dimensional lattices are dealt with in lattice theories of protein folding [18]. There is currently no general solution for the Ising problem in two dimensions [19], except for Onsager's solution when the lattice is infinite and subject to periodic boundary conditions [20]. Computational methods have been developed to tackle the Ising problem for finite lattices [21]. Applicability of these methods remains confined to systems of unrealistically small size. Hence, an analytical approach to the problem of multivalent binding in two dimensions is not only relevant to biology, but is also central to the statistical thermodynamics of the Ising problem.

When dealing with multivalent binding in two dimensions the valency of the ligand becomes decoupled from its geometry. For example, a ligand covering three sites may be linear or triangular in shape and each of these cases leads to different partition functions. As a first step to the approach of multivalent binding in two-dimensions, we consider the simple case where binding to a given site excludes binding to all neighbor sites in contact with it. This case is mathematically equivalent to univalent binding in two dimensions with $\sigma = 0$.

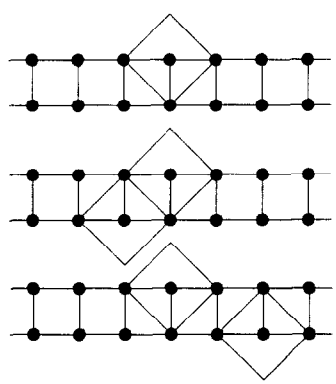

Fig. 3. Multivalent binding to a two-dimensional ladder. The ligand has a diamond shape and binds to a site with its center. When bound, it covers the adjacent sites and precludes binding of the center of a second ligand to these sites. Shown are possible configurations of the lattice.

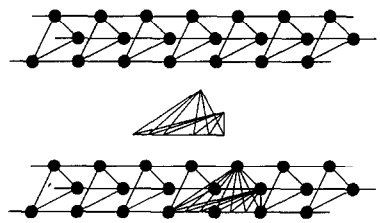

Fig. 4. Multivalent binding to a two-dimensional torus with triangular section. The ligand (shown in the middle) has a diamond shape and binds to a site with its center (bottom). When bound, it covers the adjacent sites and precludes binding of the center of a second ligand to these sites.

Also in one dimension, the case of a divalent ligand ($m = 2$) binding without cooperativity is mathematically identical to the case of a univalent ligand binding with $\sigma = 0$ (see Eq. (10)).

The partition function of a two-dimensional torus of length $N$ with $s$ sites in contact with each other in its section, ${}^{L}\Psi_{(N)}$, can be solved exactly in the general case. This problem includes the lattices in Figs. 3 and 4 as special cases. Also, the one-dimensional case is obtained for $s = 1$. The transfer matrix for the general solution can be constructed by induction starting with the simple case of $s = 2$ (ladder shown in Fig. 3). The relevant form in the general case of cooperative univalent ligand binding to the ladder is

$$\mathbf{W} = \begin{pmatrix} 1 & 1 & 1 & 1 \\ \omega & \sigma\omega & \omega & \sigma\omega \\ \omega & \omega & \sigma\omega & \sigma\omega \\ \sigma\omega^2 & \sigma^2\omega^2 & \sigma^2\omega^2 & \sigma^3\omega^2 \end{pmatrix} \tag{39}$$

Each column enumerates the energetic states of two vertically connected sites in the ladder once the configuration of the neighbor pair is specified. Under the assumption that $\sigma = 0$, binding to a given site excludes binding to all neighbor sites. This is the case for binding a diamond-shaped ligand that covers a site with its center and overlaps with its vertices other sites, thereby precluding binding of a second ligand to these sites (see Fig. 3). The transfer matrix is a special case of Eq. (39), i.e.,

$$\mathbf{W} = \begin{pmatrix} 1 & 1 & 1 & 1 \\ \omega & 0 & \omega & 0 \\ \omega & \omega & 0 & 0 \\ 0 & 0 & 0 & 0 \end{pmatrix} \tag{40}$$

The next case of interest is the torus with triangular section, as shown in Fig. 4, for which $s = 3$. The ligand, hypothetically of a diamond-like shape, binds with its center to a given site and sterically hinders binding of a second ligand to the neighbor sites. The transfer matrix for the general solution of univalent binding is $8 \times 8$, but simplifies considerably when $\sigma = 0$ to yield

$$\mathbf{W} = \begin{pmatrix} 1 & 1 & 1 & 1 & 1 & 1 & 1 & 1 \\ \omega & 0 & \omega & \omega & 0 & 0 & \omega & 0 \\ \omega & \omega & 0 & \omega & 0 & \omega & 0 & 0 \\ \omega & \omega & \omega & 0 & \omega & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \end{pmatrix} \tag{41}$$

Hence, in the case of a torus containing $s$ sites all in contact with each other in its section, the transfer matrix is $2^s \times 2^s$, because there are $2^s$ possible ligated configurations for the $s$ sites in the section. These configurations

can easily be listed to write down explicitly the transfer matrix. The ligand in this case encircles the torus at the level of its section, which is reminiscent of the binding mode of a polymerase to DNA [22]. For $\sigma = 0$, which is the case of interest here, the transfer matrix is

$$\mathbf{W} = \left(\begin{array}{ccccc|c} 1 & 1 & 1 & \cdots & 1 & \\ \omega & 0 & \omega & \cdots & \omega & \\ \omega & \omega & 0 & \cdots & \omega & \mathbf{A} \\ \vdots & \vdots & \vdots & \vdots & \vdots & \\ \omega & \omega & \omega & \cdots & 0 & \\ \hline & & \mathbf{O} & & & \mathbf{O} \end{array}\right) \tag{42}$$

Most of the eigenvalues vanish and the non-zero eigenvalues are obtained from the secular equation associated with the $(s+1)\times(s+1)$ determinant of the block in the top left corner.

To find the analytical expressions for the $s+1$ eigenvalues we define

$$\Delta_n = \begin{vmatrix} 1-\lambda & 1 & 1 & \ldots & 1 & 1 \\ \omega & -\lambda & \omega & \ldots & \omega & \omega \\ \omega & \omega & -\lambda & \ldots & \omega & \omega \\ \vdots & \vdots & \vdots & \vdots & \vdots & \vdots \\ \omega & \omega & \omega & \ldots & -\lambda & \omega \\ \omega & \omega & \omega & \ldots & \omega & -\lambda \end{vmatrix} \tag{43}$$

where the index $n = 0,1,\ldots s-1$, enumerates the number of columns and rows bordering the matrix associated with the determinant

$$\Delta_0 = \begin{vmatrix} 1-\lambda & 1 \\ \omega & -\lambda \end{vmatrix} = -\lambda(1-\lambda) - \omega \tag{44}$$

so that

$$\Delta_1 = \begin{vmatrix} 1-\lambda & 1 & 1 \\ \omega & -\lambda & \omega \\ \omega & \omega & -\lambda \end{vmatrix} \tag{45}$$

$$\Delta_2 = \begin{vmatrix} 1-\lambda & 1 & 1 & 1 \\ \omega & -\lambda & \omega & \omega \\ \omega & \omega & -\lambda & \omega \\ \omega & \omega & \omega & -\lambda \end{vmatrix} \tag{46}$$

and so forth. Subtracting the $s$th row from the last one in Eq. (43) leaves the value of the determinant unchanged [23] and yields

$$\Delta_n = \begin{vmatrix} 1-\lambda & 1 & 1 & \ldots & 1 & 1 \\ \omega & -\lambda & \omega & \ldots & \omega & \omega \\ \omega & \omega & -\lambda & \ldots & \omega & \omega \\ \vdots & \vdots & \vdots & \vdots & \vdots & \vdots \\ \omega & \omega & \omega & \ldots & -\lambda & \omega \\ 0 & 0 & 0 & \ldots & \lambda+\omega & -(\lambda+\omega) \end{vmatrix} \tag{47}$$

Hence,

$$\Delta_n = -(\lambda+\omega)\Delta_{n-1} - (\lambda+\omega)\Delta_{n-1,\omega} \tag{48}$$

where

$$\Delta_{n,\omega} = \begin{vmatrix} 1-\lambda & 1 & 1 & \dots & 1 & 1 \\ \omega & -\lambda & \omega & \dots & \omega & \omega \\ \omega & \omega & -\lambda & \dots & \omega & \omega \\ \vdots & \vdots & \vdots & \vdots & \vdots & \vdots \\ \omega & \omega & \omega & \dots & -\lambda & \omega \\ \omega & \omega & \omega & \dots & \omega & \omega \end{vmatrix} \tag{49}$$

differs from $\Delta_n$ only in so far as the last diagonal element is $\omega$ instead of $-\lambda$. Subtracting the $s$th row from the last one in Eq. (49) leaves again the determinant unchanged and leads to the following recursion relation

$$\Delta_{n,\omega} = -(\lambda+\omega)\Delta_{n-1,\omega} \tag{50}$$

Hence,

$$\Delta_{n,\omega} = (-1)^{n-1}(\lambda+\omega)^{n-1}\Delta_{1,\omega} = (-1)^{n-1}(\lambda+\omega)^{n-1}\begin{vmatrix} 1-\lambda & 1 & 1 \\ \omega & -\lambda & \omega \\ \omega & \omega & \omega \end{vmatrix}$$

$$= (-1)^{n-1}(\lambda+\omega)^n\lambda\omega \tag{51}$$

Substitution into Eq. (48) and iteration gives

$$\Delta_n = (-1)^n(\lambda+\omega)^n\Delta_0 + n(-1)^{n-1}(\lambda+\omega)^n\lambda\omega \tag{52}$$

The non-zero eigenvalues of the transfer matrix in Eq. (42) are therefore solutions of the equation

$$\Delta_{s-1} = (-1)^{s-1}(\lambda+\omega)^{s-1}\{\lambda^2 - [1+(s-1)\omega]\lambda - \omega\} = 0 \tag{53}$$

Hence,

$$\lambda_{1,2} = \frac{1+(s-1)\omega \pm \sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}{2} \tag{54}$$

$$\lambda_3 = \lambda_4 = \dots = \lambda_{s+1} = -\omega \tag{55}$$

Under periodic boundary conditions (ring), the partition function for the two-dimensional torus with $s$ sites all in contact in its section is therefore

$${}^{R}\Psi_{(N)} = \lambda_1^N + \lambda_2^N + (s-1)(-1)^N\omega^N \tag{56}$$

This expression deserves much consideration. For $s = 1$ the torus degenerates into a one-dimensional ring and the partition function becomes identical to Eq. (3), as expected. The simplest case of a two-dimensional lattice is obtained for $s = 2$, which is the ladder with periodic boundary conditions (see Fig. 3). In this case, a term proportional to $\omega$ is added to the two eigenvalues in Eq. (54). This correction is borne out by the change in dimensionality of the lattice and is peculiar of the two-dimensional embedding. For $s = 3$, which is the case given in Fig. 4, the correction is weighted by a factor of two in the partition function and, in general, when $s$ sites are in the section of the torus, there are $s-1$ identical terms proportional to $\omega$ in the partition function. Since there are $sN$ total sites in the torus and only a maximum of $N$ ligands can be bound at saturation ($\omega \to \infty$), the correction in Eq. (56) affects the last term in the partition function expressed as a polynomial in the scaled ligand activity $\omega$. The coefficient $A_N$ of $\omega^N$ can be calculated directly and is given by

$$A_N = (s-1)\left[(s-1)^{N-1} + (-1)^N\right] \tag{57}$$

Wyman's median ligand activity [24] is therefore

$$x_m = \frac{(s-1)^{-1/N}\left[(s-1)^{N-1}+(-1)^N\right]^{-1/N}}{K} \tag{58}$$

and tends to $1/sK$ for large $s$, as intuitively obvious.

The binding properties of the torus in the absence of periodic boundary conditions (two-dimensional linear torus) can be derived from those outlined above for the two-dimensional ring using arguments already mentioned for the one-dimensional case. Contraction over the $s$ sites in a section of the torus generates $s$th-order contracted partition functions [14]. The partition function of the system expressed relative to these contracted forms is

$${}^{R}\Psi_{(N)} = \sum_{j=0}^{s}\binom{s}{j}{}^{j,R}\Psi_{(N-1)}\sigma^{j(j-1)/2}\omega^{j} \tag{59}$$

where ${}^{j,R}\Psi_{(N-1)}$ indicates a ring where $s$ sites of one of its sections are frozen in a particular ligation state, and specifically $j$ in the ligated state and $N-j$ in the unligated state. When $\sigma = 0$ the expansion in Eq. (59) contains only two contracted partition functions, as follows

$${}^{R}\Psi_{(N)} = {}^{0,R}\Psi_{(N-1)} + s\omega\,{}^{1,R}\Psi_{(N-1)} \tag{60}$$

${}^{0,R}\Psi_{(N-1)}$ is the same as ${}^{L}\Psi_{(N-1)}$, the partition function of a torus of length $N-1$ without periodic boundary conditions. ${}^{1,R}\Psi_{(N-1)}$ is the contracted partition function of the torus with periodic boundary conditions when one site is kept ligated. This partition function obeys the following relation analogous to Eq. (1)

$${}^{R}X_{j(N)} = {}^{R}X_{(N)} = \frac{\omega\,{}^{1,R}\Psi_{(N-1)}}{{}^{R}\Psi_{(N)}} = \frac{1}{sN}\frac{\mathrm{d}\ln{}^{R}\Psi_{(N)}}{\mathrm{d}\ln\omega} \tag{61}$$

The binding probability is the same for any site of the torus in the presence of periodic boundary conditions. Hence,

$${}^{L}\Psi_{(N-1)} = {}^{R}\Psi_{(N)} - \frac{1}{N}\frac{\mathrm{d}\,{}^{R}\Psi_{(N)}}{\mathrm{d}\ln\omega} \tag{62}$$

is a very basic transformation, analogous to Eq. (2), that connects the partition functions of the torus in the presence and absence of periodic boundary conditions. The relation 62 is identical to that found in the one-dimensional case [14] and is independent of the value of $s$. The explicit form of ${}^{L}\Psi_{(N)}$ is obtained from Eqs. (56) and (62) as

$${}^{L}\Psi_{(N)} = \lambda_1^N(\lambda_1 - \lambda_1') + \lambda_2^N(\lambda_2 - \lambda_2') \tag{63}$$

and the $s-1$ corrections in Eq. (56) cancel out. Two eigenvalues suffice to define the binding properties to the torus in the absence of periodic boundary conditions.

To find the explicit expression for binding to any site in the $j$th section of the torus in the absence of periodic boundary conditions, we expand again the partition function in terms of $s$th-order contracted partition functions, so that

$${}^{L}\Psi_{(N)} = {}^{L}\Psi_{(N-1)} + s\omega\,{}^{L}\Psi_{(N-2,s-1)} \tag{64}$$

Hence,

$${}^{L}X_{N(N)} = \frac{\omega\,{}^{L}\Psi_{(N-2,s-1)}}{{}^{L}\Psi_{(N)}} = \frac{1}{s}\left(1 - \frac{{}^{L}\Psi_{(N-1)}}{{}^{L}\Psi_{(N)}}\right) \tag{65}$$

This is the binding probability to any of the $s$ sites in the end section of the torus. Note that this probability reaches an asymptotic value of $1/s$ for $\omega \to \infty$, because binding of a ligand to any of the $s$ sites precludes binding to the other $s-1$ sites. In the limit $N \to \infty$, this binding probability becomes

$$\lim_{N\to\infty} {}^{L}X_{N(N)} = {}^{L}X_{\infty(\infty)} = \frac{1}{s}\left(1 - \frac{1}{\lambda_1}\right) \tag{66}$$

Binding to any site in the $j$th section of the torus is therefore

$$\begin{aligned}
{}^{L}X_{j(N)} &= 1 - \frac{{}^{0,L}\Psi_{j(N)}}{{}^{L}\Psi_{(N)}} = 1 - \frac{{}^{L}\Psi_{(j-1)}\,{}^{L}\Psi_{(N-j)}}{{}^{L}\Psi_{(N)}} - \frac{(s-1)\,\omega\,{}^{L}\Psi_{(j-2,1)}\,{}^{L}\Psi_{(N-j-1,1)}}{{}^{L}\Psi_{(N)}} = \frac{\omega\,{}^{L}\Psi_{(j-2,1)}\,{}^{L}\Psi_{(N-j-1,1)}}{{}^{L}\Psi_{(N)}} \\
&= \frac{1}{s}\left(1 - \frac{{}^{L}\Psi_{(j-1)}\,{}^{L}\Psi_{(N-j)}}{{}^{L}\Psi_{(N)}}\right) \\
&= \frac{1}{s}\left[1 - \frac{\left(1 - s^{L}X_{N-j(N-j)}\right)\left(1 - s^{L}X_{N-j+1(N-j+1)}\right)\cdots\left(1 - s^{L}X_{N(N)}\right)}{\left(1 - s^{L}X_{1(1)}\right)\left(1 - s^{L}X_{2(2)}\right)\cdots\left(1 - s^{L}X_{j-1(j-1)}\right)}\right] = \\
&\frac{1}{s}\left\{1 - \frac{\left[\lambda_1^{j-1}(\lambda_1 - \lambda_1') + \lambda_2^{j-1}(\lambda_2 - \lambda_2')\right]\left[\lambda_1^{N-j}(\lambda_1 - \lambda_1') + \lambda_2^{N-j}(\lambda_2 - \lambda_2')\right]}{\lambda_1^{N}(\lambda_1 - \lambda_1') + \lambda_2^{N}(\lambda_2 - \lambda_2')}\right\}
\end{aligned} \tag{67}$$

In the asymptotic limit

$$\lim_{N\to\infty} {}^{L}X_{j(N)} = \frac{1}{s}\frac{\lambda_1'}{\lambda_1} = {}^{R}X_{(\infty)} \tag{68}$$

follows directly from Eqs. (56) and (61), as expected. The explicit expressions for binding probabilities and eigenvalues are summarized in Table 1.

Table 1
Eigenvalues and asymptotic binding properties of a two-dimensional torus

| | Periodic boundary conditions | No periodic boundary conditions |
|---|---|---|
| $\lambda_1$ | $\frac{1+(s-1)\omega+\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}{2}$ | $\frac{1+(s-1)\omega+\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}{2}$ |
| $\lambda_2$ | $\frac{1+(s-1)\omega-\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}{2}$ | $\frac{1+(s-1)\omega-\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}{2}$ |
| $\lambda_{3,4,s+1}$ | $-\omega$ | 0 |
| $\Psi_{(N)}$ | $\lambda_1^N + \lambda_2^N + (s-1)(-1)^N\omega^N$ | $\lambda_1^N(\lambda_1 - \lambda_1') + \lambda_2^N(\lambda_2 - \lambda_2')$ |
| $\lambda_1'$ | $\frac{\omega}{2}\left[(s-1) + \frac{(s+1)+(s-1)^2\omega}{\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}\right]$ | $\frac{\omega}{2}\left[(s-1) + \frac{(s+1)+(s-1)^2\omega}{\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}\right]$ |
| $X_{j(\infty)}$ | $\frac{\omega}{s}\frac{(s-1)+\frac{(s+1)+(s-1)^2\omega}{\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}}{1+(s-1)\omega+\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}$ | $\frac{\omega}{s}\frac{(s-1)+\frac{(s+1)+(s-1)^2\omega}{\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}}{1+(s-1)\omega+\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}$ |
| $X_{\infty(\infty)}$ | $\frac{\omega}{s}\frac{(s-1)+\frac{(s+1)+(s-1)^2\omega}{\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}}{1+(s-1)\omega+\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}$ | $\frac{1}{s}\frac{(s-1)\omega-1+\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}{1+(s-1)\omega+\sqrt{1+2(s+1)\omega+(s-1)^2\omega^2}}$ |

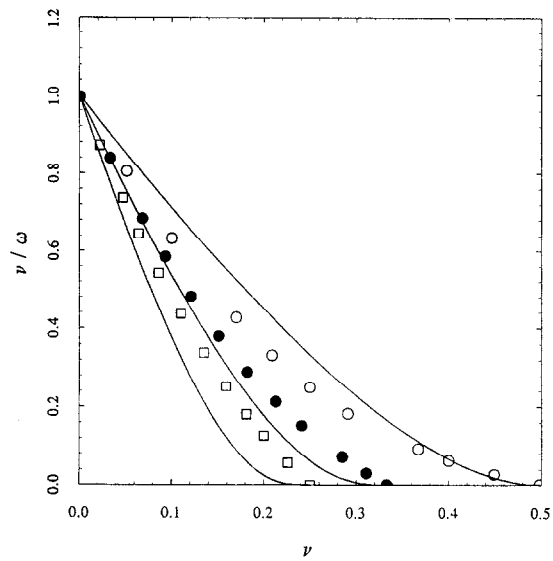


Fig. 5. Schatchard plot for the fraction of ligands bound per site, $\upsilon$, in the one-dimensional (continuous lines) and two-dimensional (○,●,□) case. Curves for the one-dimensional case were plotted using the McGhee–von Hippel model [4] for $m = 2$ (right), $m = 3$ (middle) and $m = 4$ (left), keeping $\sigma = 1$ (see Eq. (37) in the text). Points depicting the two-dimensional ladder (○, $s = 2$) and torus (●, $s = 3$; □, $s = 4$) were plotted using the expressions listed in Table 1. ${}^{R}X_{(\infty)}$ is the same as $\upsilon$ for these models, because $m = 1$ and the fraction of ligands bound per site coincides with the binding probability to the site. Note how binding in two dimensions is characterized by an apparent degree of cooperativity when compared with the non-cooperative one-dimensional McGhee–von Hippel model with the same intercepts in the Scatchard plot.

The binding probability in Eq. (68) goes to $1/s$ as $\omega \to \infty$ because a maximum of $N$ ligands can be bound at saturation and there are $sN$ total sites. The binding probability reflects directly the quantity $\upsilon$ accessible to experimental measurements and can be compared to the analogous quantity derived from the McGhee–von Hippel model when $m = s$. This comparison is particularly important because it provides a means to test the effect of lattice dimensionality on binding properties. Relevant cases are shown in Fig. 5. The cases $\sigma = 1$, $m = 2$, $m = 3$ and $m = 4$ in the McGhee–von Hippel model are compared with the two dimensional torus behaving according to Eq. (68) when $s = 2$, $s = 3$ and $s = 4$. In all cases, the two-dimensional lattice produces a Scatchard plot that implies cooperative interactions when interpreted in terms of the McGhee–von Hippel model. Hence, non-cooperative multivalent binding in two dimensions can mimic cooperative multivalent binding in one dimension. This feature of multivalent binding is revealed here for the first time and clearly bears on the interpretation of Scatchard plots obtained for a variety of processes, encompassing protein-nucleic acid and ligand-receptor interactions.

## 8. Discussion

There is a considerable advantage in treating multivalent binding to a one-dimensional lattice in terms of contracted partition functions. The eigenvalues of the transfer matrix can be derived from the site-specific approach. The recursion relation for the partition function of the lattice provides an analytical and practical tool for understanding the properties of the system under any condition of interest. The McGhee–von Hippel model [4] can be derived in a straightforward manner in the limit $N \to \infty$, which proves that the combinatorial arguments used by these authors are exact.

The approach presented in this article lends itself to extension to the analysis of multivalent binding to two-dimensional lattices, that provide more realistic models of double-stranded polynucleotides, membrane surfaces or protein helices. Binding to a two-dimensional lattice mimics binding to a one-dimensional lattice with cooperativity, thereby introducing a complexity in the interpretation of Scatchard plots pertaining to ligand binding to double-stranded DNA, membrane surfaces or protein helices. The dimensional embedding of the lattice must be borne in mind when interpreting such data. The analysis of multivalent binding in two-dimensions introduced here represents an important first step toward further investigation of this problem in the general case, where bound ligands experience cooperative interactions. The mathematical complexity of the problem is conveyed immediately by the size of the transfer matrix needed to define the partition function of the system in the general case. Contracted partition functions offer alternative approaches that may come in quite handy, as will be discussed at length elsewhere.

## 9. List of symbols

| | |
|---|---|
| ${}^{L}B_{j(N)}$ | site-specific binding capacity of site $j$ in a linear lattice of $N$ sites |
| $L$ | average number of ligands bound |
| $m$ | valency of the ligand |
| $N$ | number of binding sites in the lattice |
| $s$ | number of sites in the section of the torus |
| ${}^{L}X_{j(N)}$ | site-specific binding curve of site $j$ in a linear lattice of $N$ sites |
| ${}^{R}X_{j(N)}$ | site-specific binding curve of site $j$ in a ring of $N$ sites |
| $\lambda$ | eigenvalue of the transfer matrix |
| $\upsilon$ | fraction of ligands bound per site |
| $\Psi$ | partition function |
| ${}^{0}\Psi_{j}$ | contracted partition function with site $j$ unligated |
| ${}^{1}\Psi_{j}$ | contracted partition function with site $j$ bound |
| ${}^{L}\Psi_{(N)}$ | partition function for a linear lattice of $N$ sites |
| ${}^{R}\Psi_{(N)}$ | partition function for a ring of $N$ sites |
| ${}^{0,L}\Psi_{N(N)}$ | contracted partition function for a linear lattice of $N$ sites with site $N$ unligated |
| ${}^{1,L}\Psi_{N(N)}$ | contracted partition function for a linear lattice of $N$ sites with site $N$ bound |
| ${}^{01,L}\Psi_{N-m,N(N)}$ | contracted partition function for a linear lattice of $N$ sites with site $N-m$ unligated and site $N$ bound |
| ${}^{11,L}\Psi_{N-m,N(N)}$ | contracted partition function for a linear lattice of $N$ sites with sites $N-m$ and $N$ bound |
| $\sigma$ | site-site interaction constant |
| $\omega$ | scaled ligand activity variable |

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## Appendix A

Eq. (37) is identical to that derived by McGhee and von Hippel as Eq. (15) in their article [4]. We start by noting that

$$[(2\sigma-1)(1-m\upsilon)+\upsilon+Q][(2\sigma-1)(1-m\upsilon)+\upsilon-Q]=4\sigma(\sigma-1)(1-m\upsilon)^2 \quad \text{(A1)}$$

This allows Eq. (38) to be rewritten as

$$\frac{v}{x} = K(1-mv)\frac{[1+(2\sigma-m-1)v+Q][(2\sigma-1)(1-mv)+v-Q]}{4(2\sigma-1)(1-mv)^2} \times\left[\frac{(2\sigma-1)(1-mv)+v+Q}{2(\sigma-1)(1-mv)}\right]^{m-1}$$
$$= K(1-mv)\frac{[1-(m+1)v+2\sigma v+Q][2\sigma mv-1+(m+1)v-Q]}{4(\sigma-1)(1-mv)^2} \times\left[\frac{(2\sigma-1)(1-mv)+v-Q}{2(\sigma-1)(1-mv)}\right]^{m-1} \quad (A2)$$

Define

$$\varphi = 1-(m+1)v \quad (A3)$$

so that, from Eq. (38)

$$Q^2 = \varphi^2 + 4\sigma v(1-mv) \quad (A4)$$

Substitution into Eq. (A2) yields

$$\frac{v}{x} = K(1-mv)\frac{[\varphi+2\sigma v+Q][2\sigma mv-\varphi-Q]}{4(\sigma-1)(1-mv)^2}\left[\frac{(2\sigma-1)(1-mv)+v-Q}{2(\sigma-1)(1-mv)}\right]^{m-1} =$$
$$K(1-mv)\frac{\sigma(Q^2-\varphi^2)+2\sigma\varphi(\varphi+Q)-(\varphi+Q)^2}{4(\sigma-1)(1-mv)^2}\left[\frac{(2\sigma-1)(1-mv)+v-Q}{2(\sigma-1)(1-mv)}\right]^{m-1} =$$
$$K(1-mv)\frac{(\varphi+Q)^2}{4(1-mv)^2}\left[\frac{(2\sigma-1)(1-mv)+v-Q}{2(\sigma-1)(1-mv)}\right]^{m-1}$$
$$= K(1-mv)\left[\frac{1-(m+1)v+Q}{2(1-mv)}\right]^2\left[\frac{(2\sigma-1)(1-mv)+v-Q}{2(\sigma-1)(1-mv)}\right]^{m-1} \quad (A5)$$

which is the same expression derived by McGhee and von Hippel [4].

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