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The efficiency of chemical detectors

S. Manghani[1] and J. J. Ramsden[2,*]

[1]*Abteilung biophysikalische Chemie, Biozentrum der Universität, Basel, Switzerland*
[2]*Collegium Basilea (Institute of Advanced Study), Basel, Switzerland*

Chemical detectors ("sensors") usually consist of a two-dimensional array of receptors exposed to the solution to be tested, from whose output the bulk solution concentration of the analyte of interest can be determined. Both input and output—the number of analyte particles striking the array in a given interval of time, and the number captured—are countable events. The gain is the quotient of these two numbers, and the detectivity the quotient of their fluctuations. The gain and detectivity provide a universal framework for comparing different types of sensors, and in which the desirable properties of sensors, e.g. their ability to detect very weak signals ("sensitivity"), and to detect the analyte in the presence of a large excess of other molecules ("selectivity"), can be related to various physico-chemical parameters such as the packing density and size of receptors, and their affinity for the analyte. Analyte multivalence, although formally a source of inefficiency, is very useful for making the sensor more resistant to spurious chemical noise. An important result is that chemical fog engendered by a huge excess of nonspecifically binding particles has no effect on the detectivity, provided that the nonspecific interaction is reversible.

1. INTRODUCTION

In recent years a variety of devices for detecting atoms, molecules, ions or other particles present in solution or in the atmosphere have been devised and developed. In this paper, the species being detected will be referred to as the analyte. The goal of the detection procedure is to determine the ambient analyte concentration (a number per unit volume) c. These devices, which are called variously sensors, chemical sensors, biosensors, "capteurs", etc., usually possess similar design features: they consist of a layer of receptors (captors or traps) able to interact with the analyte but not with any other species which may be present in the ambient medium. The layer is mounted on a transducer, whose output (usually optical or electrical) is related to the number of analyte particles trapped by the receptors.

Since such devices offer considerable advantages (of accuracy, convenience, cost, miniaturization etc.) when applied to a wide range of engineering, chemical, medical and environmental problems, considerable effort is currently being expended in developing and improving them. Nevertheless, criteria measuring the performance of such chemically based sensors are presently of a rather *ad hoc* nature, often using vaguely defined notions of sensitivity. It is therefore difficult to compare one sensor with another, and it would be highly desirable to define the sensitivity, or efficiency of detection, of different types of chemical sensors on a common scale. Furthermore, in the absence of a formal framework, work to improve sensors is presently largely empirical. An understanding of the basic principles underlying chemical detection is a prerequisite for the rational improvement of sensors. The aim of this paper is to provide that framework.

*Correspondence to: Hochstrasse 51, 4053 Basel, Switzerland

2. THE DETECTOR ARRAY

The basic chemical detector consists of a monolayer array of N chemical receptors capable of reacting only with the analyte. Each individual receptor occupies an area $a > a_a$, the projected area of the analyte. If the detector surface (total area A) is wholly covered by a grid of N receptors, then

$$A = Na. \tag{1}$$

In a practical device, A is the area in which the presence of analyte molecules is actually transduced into an electrical or optical signal. In what follows we shall take $A = 1$. It may be the area of a quartz crystal microbalance (QCM), the gate or electrode area in a variety of electronic or electrochemical methods, or the area illuminated by the incoming light beam in various optical techniques such as ellipsometry, scanning angle reflectometry (SAR), surface plasmon resonance (SPR), integrated optics, etc.[1]. In some of them, notably those based on Fresnel's reflexion formulae (i.e. ellipsometry, SAR and IO), the sensor

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output is the absolute number of bound molecules per receptor, which offers certain advantages when analysing the performance of the devices. We shall denote the output per unit area as Y.

3. EXPOSURE

The exposure time τ is the quantity of practical interest—the interval during which the sensor is exposed to the sample. During this interval a certain number α of analyte molecules will impinge on the sensor, of which a lesser or equal number β of them will be captured. Unlike an optical detector [2] in which all photons not "captured" (in the sense of engendering a response) either pass through the sensor unchanged or are annihilated, any analyte molecules not captured on their first impingement tend to remain in the vicinity of the sensor surface and can make further attempts to be captured [3, 4].

3.1. Transport

In the absence of an energy barrier (the perfect sink), the initial flux I (per unit area and per unit time) of analyte molecules arriving at the detector array depends solely on analyte concentration and the hydrodynamics of the system. For a flowing or stirred solution transport is via convective diffusion, i.e.

$$I = Dc/\delta_d \tag{2}$$

where D is the diffusion coeffcient of the analyte and δ_d the thickness of the diffusion boundary layer. For standard flow cell geometries formulae for computing δ_d are available [5]. For unstirred systems the flux is diffusion controlled and given by [6]:

$$I = cD^{1/2}(\pi t)^{-1/2}. \tag{3}$$

The exposure X per unit area (i.e. the number of analyte molecules impinging on the detector) is

$$\int_0^\tau I(t)dt \tag{4}$$

for an exposure time τ, i.e.

$$X = \tau Dc/\delta_d \tag{5}$$

for the flowing system, and

$$X = 2c(D\tau/\pi)^{1/2} \tag{6}$$

for the purely diffusive system. Note that in both cases X is linearly proportional to c.

3.2. Transport with an energy barrier

In the presence of an energy barrier of height U the initial flux I of analyte molecules to the detector array is determined by the proportion of analyte molecules surmounting the energy barrier. Hence the actual exposure X^* will be lower than that expected from the knowledge of c, τ and the hydrodynamic factors (eqns 5 or 6) by a factor $\sim e^{-U/kT}$; this is equivalent to placing a neutral density filter in front of a photodetector, i.e.

$$X^* = Xe^{-U/kT} = Xb. \tag{7}$$

A more complete treatment takes account of the fact that U is usually an interaction potential depending on the separation z between the analyte and the surface, and its integral then defines a reaction distance δ_a [7]:

$$\delta_a = \int_{z_0}^{\infty} (e^{U(z)/kT} - 1)\,dz, \tag{8}$$

where the lower boundary of integration is the distance at which $U(z)$ becomes zero for the first time when approaching the surface from infinity. δ_a in turn defines a chemical rate constant for binding [7]:

$$k_a = \frac{D}{\delta_a} \tag{9}$$

The probability of an approaching molecule actually binding is now less than unity. The flux is correspondingly reduced and is then

$$I = k_a c_\upsilon \tag{10}$$

where c_υ is the concentration of analyte molecules in the immediate vicinity of the interface, obtained by summing the fluxes to and from the layers [8]:

$$V\frac{\mathrm{d}c_\upsilon}{\mathrm{d}t} = \frac{A(c - c_\upsilon)D}{\delta_d} - Ak_a c_\upsilon \tag{11}$$

where V is unit volume. Putting the left hand side to zero yields

$$c_\upsilon = \frac{c}{1 + \delta_d k_a / D} \tag{12}$$

and hence

$$I = \frac{c}{1/k_a + \delta_d / D}. \tag{13}$$

Writing the actual exposure as a function of time and bulk analyte concentration gives finally:

$$X^* = \frac{c\tau}{1/k_a + \delta_d / D}. \tag{14}$$

Note that here we assume that $U(z)$ does not depend on the amount of analyte bound (such a dependance may arise if, for example, the analyte bears a net electrostatic charge [9], but it will become negligible at a suitably high ionic strength). For a low energy barrier, i.e. large k_a, this expression reduces to (5)/τ; conversely if k_a is small, hydrodynamic details become unimportant.

3.3. Capture statistics

We shall denote the mean exposure per receptor as x, i.e.

$$x = Xa = X / N, \tag{15}$$

and the proportion P_n of receptors struck by n molecules is

$$P_n = x^n e^{-x} / n!. \tag{16}$$

Since the variance of a Poisson distribution equals its mean, we have

$$\sigma_x = x^{1/2}. \tag{17}$$

4. GAIN AND SATURATION

We define the gain, γ, of the sensor as the quotient of the input X (*were the sensor a perfect sink for the analyte*) and output Y, both of which are in principle countable numbers of molecules, i.e.

$$\gamma = Y / X. \tag{18}$$

When an analyte molecule strikes a vacant receptor, it will be bound and remain to be counted by the transducer part of the sensor. The variance of the output signal σ_Y^2, a measure of how compactly it is concentrated around the mean output signal, will depend on the mechanical, electrical, optical, thermal etc. stability of the transducer and is an empirically determined quantity.

For monodentate analyte molecules and monodentate receptors that bind analyte irreversibly and independently from each other, once the binding site is occupied by one analyte molecule no further molecules can be captured, i.e., each receptor has a maximum (saturation) count level $s = 1$.

The probability of no analyte molecules being bound to a given receptor is, from eqn (16),

$$P_0 = e^{-x} \tag{19}$$

and hence the probability of the receptor being saturated (i.e. binding one ligand) is:

$$P_1 = 1 - P_0 = 1 - e^{-x} \tag{20}$$

as P_2, P_3 etc. are all equal to zero. This is the mean output y per receptor, and for the transducer completely covered with monodentate receptors in the presence of an energy barrier,

$$Y = yN = (1 - e^{-X^*/N})N. \tag{21}$$

Recalling that $X^* = Xb$ with $b = e^{-U/kT}$, the gain is

$$\gamma = \frac{(1 - e^{-Xb/N})N}{X}. \tag{22}$$

Figure 1 shows the output Y versus exposure X, with and without an energy barrier, and Figure 2 shows the response (gain) versus exposure.

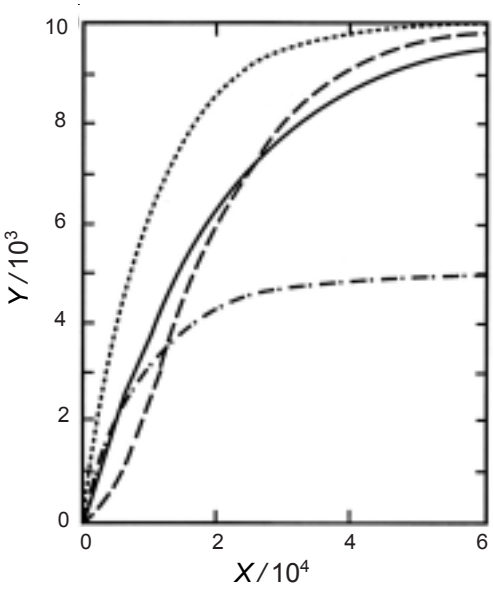

Figure 1. Y vs X. The following apply to all figures unless otherwise stated: $N = 10^4$, $b = 1$, $\theta_r = 1$, $a = 1/N$; curves: dotted line: monodentate; solid line: $b = 0.5$ or $\theta = 0.5$, monodentate analyte; dashed line: $N = 2 \times 10^4$, bidentate analyte; dashed-dotted line: $N = 5 \times 10^3$, monodentate analyte.

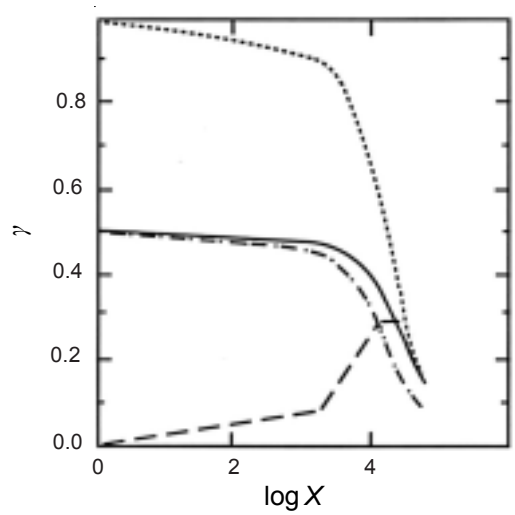

Figure 2. γ vs log$X$ (see legend to Fig. 1 for details).

In the widely used immunoassays of the ELISA type [10], the amount of captured analyte is estimated by binding a further ligand to it, which is capable of enzymatically transforming a substrate into a coloured product. The measured output is proportional to the number of product molecules generated, which depends on the substrate concentration and duration of the enzymatic transformation. Hence the gain γ can be arbitrarily large. Moreover any adventitious binding of the enzymatic ligand will lead to an output signal even in the absence of captured analyte. Hence gain does not appear to be a useful basis for comparing different assays and sensors.

5. DETECTIVITY

Suppose we are measuring a certain output Y, which corresponds to an input X, i.e. $Y = \gamma X$, where γ is the gain of the sensor. With both input and output there is associated a certain uncertainty (noise) characterized by variances σ_X^2 and σ_Y^2. We then notice that following a change of analyte solution, the output has increased to $Y + \Delta Y$. Does this correspond to a real increase in X? More pertinently, what is the least increment $\Delta X_{\min}$ which can be detected? These questions require explicit consideration of the errors associated with the input and the output signals, and we ask, how big does the input signal increment have to be in order to be able to detect an increment in the output signal? To answer this question it is necessary to separate the contributions to the overall system noise: input signal noise and noise due to the measuring device. The sensor will detect an incremental signal ΔX on top of an existing input X at the input only if $\Delta X \geq m\sigma_X$ where m is a multiplier depending on the ultimate resolution of the measuring instrument, typically equal to unity. This signal will then be carried through to the output (amplified by the sensor's gain γ) and will be detected at the output as a signal ΔY only if it is greater than σ_Y. The gain Y/X incorporates features of both signal and detector; in order to compare the performance of different detectors the contribution from the signal has therefore to be separated out. A true measure of the input signal takes into account the noise contribution and is given by input signal to noise power ratio S_i:

$$S_i = (\Delta X/\sigma_X)^2. \tag{23}$$

Similarily a true measure of the output signal is given by the output signal to noise power ratio S_o:

$$S_o = (\Delta Y/\sigma_Y)^2. \tag{24}$$

The detectivity [11] ε of a sensor is defined as the quotient of the normalized output to the normalized input:

$$\varepsilon = \frac{S_o}{S_i} = \left(\frac{\sigma_X \Delta Y}{\sigma_Y \Delta X}\right)^2 \tag{25}$$

Differentiating equation (21) with respect to X and equating the result with the difference ratio in eqn (25) gives

$$\Delta Y/\Delta X = be^{-bX/N}. \tag{26}$$

For the ideal (perfect sink) detector

$$\sigma_X^2 = xN = X, \tag{27}$$

i.e. the variance of the input is equal to its mean. The detectivity of the chemical detector is

$$\varepsilon_1 = \frac{Xb^2 e^{-2bX/N}}{\sigma_Y^2} \tag{28}$$

where the subscript 1 signifies monodentate analyte. The detectivity is in essence the statistical efficiency (s.e.) of the actual estimate of X. The s.e. is the ratio of the best possible estimate—i.e. $\sigma_X^2 = X$—to the output variance σ_Y^2 projected back onto the input axis, i.e. $\sigma_Y^2/(dY/dX)^2$. Figure 3 shows the detectivities versus exposure.

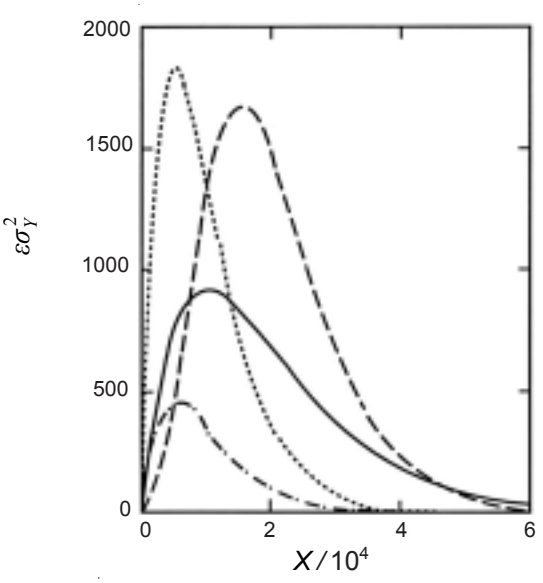

Figure 3. $\varepsilon\sigma_Y^2$ vs X. (see legend to Fig. 1 for details).

6. BIDENTATE ANALYTE

In this case a bidentate analyte molecule (e.g. an antibody of the IgG class) binds irreversibly to two adjacent monodentate receptors but will not be retained by only one receptor and hence P_n is the proportion of receptor pairs binding n bidentate analyte molecules. This effectively halves the number of receptors, i.e. each pair will be considered as a single receptor; to a first approximation there are thus $N/2$ receptors.[1] We now have

$$P_2 = 1 - P_0 - P_1 = 1 - e^{-x}(1 + x), \tag{29}$$

and the output of the detector is then:

$$Y_2 = [1 - e^{-2bX/N}(1 + 2bX/N)]N/2 \tag{30}$$

[1] With random addition of bidentate ligands some of the monodentate receptors will remain unreacted. For a 1D arrangement, the fraction θ_1 which reacts is 0.86 [12]. No solution for 2D exists, but we may estimate the corresponding fraction making use of the Palásti conjecture for the jamming limit of random sequential absorption [13], namely that for 2D the fraction θ_2 is equal to θ_1^2, i.e. 0.75.

and its detectivity is:

$$\varepsilon_2 = \frac{4X^3 b^4 e^{-4bX/N}}{\sigma_Y^2 N^2} \tag{31}$$

where the subscript 2 signifies bidentate analyte (See Figures 1–3).

7. INCOMPLETE RECEPTOR COVERAGE

In this case $Na < A$, and an inefficiency is introduced whose magnitude depends upon the fraction θ_r of area actually covered, i.e.

$$\theta_r = Na/A. \tag{32}$$

7.1. a unchanged

This is equivalent to simply reducing the number of receptors by θ_r; the response is linearly reduced by a fraction equivalent to the area covered and hence the detectivity is reduced by a factor equivalent to θ_r^2, i.e for monondentate receptors and analyte molecules:

$$\varepsilon_1 = \frac{X(b\theta_r)^2 e^{-2bX/N}}{\sigma_Y^2}. \tag{33}$$

7.2. N unchanged

The area per receptor is now reduced by the factor θ_r. This inefficiency is identical to that imposed by an energy barrier (cf. eqn 28), i.e. b is everywhere replaced by the product $b\theta_r$:

$$\varepsilon_1 = \frac{X(b\theta_r)^2 e^{-2b\theta_r X/N}}{\sigma_Y^2}. \tag{34}$$

8. REVERSIBLE BINDING

In the irreversible cases considered hitherto the receptors have an extremely high affinity for the analyte, which, once bound, remains so essentially forever: relaxation (dissociation) is infinitely slow compared with the time scale of the measurement. We now consider reversible binding applicable to receptors with lower affinity for the analyte. Here we restrict the treatment to rapid relaxation for which both association and dissociation are fast on the time scale of the measurement, viz.,

$$\frac{\mathrm{d}\theta_a}{\mathrm{d}t} \gg \frac{\mathrm{d}c}{\mathrm{d}t}, \tag{35}$$

where θ_a is the fractional occupancy of the receptors by bound analyte, and the system is therefore always in equilibrium. X is no longer dependent on τ but only on c which now becomes the input parameter, and eqn (10) is extended by explicitly including desorption:

$$I = k_a \varphi c_v - k_d \theta_a / A \tag{36}$$

where φ is the fractional occupation of receptors and k_d is the desorption rate constant (hence $\tau_d = 1/k_d$ is the mean residence time of the analyte at the receptor). In our limiting case relaxation is so fast θ_a instantaneously monitors c, i.e. a steady state obtains with $k_a \varphi c_v = k_d \theta_a$. Since $\varphi = 1 - \theta_a$ for the grid,

$$\theta_a = (1/(Kc) + 1)^{-1} \tag{37}$$

where $K(= k_a/k_d)$ is the affinity of the analyte for the receptor. This expression gives us the sensor output $Y = \theta_a N$ directly as a function of c:

$$Y = \frac{KNc}{1 + Kc}. \tag{38}$$

Figure 4 shows the output versus analyte concentration. The gain is obviously

$$\gamma = \frac{Y}{c} = \frac{KN}{1 + Kc} \tag{39}$$

and the detectivity is (differentiating eqn (38) with respect to c, and taking $\sigma_c = c^{1/2}$):

$$\varepsilon = \frac{S_o}{S_i} = \left(\frac{\sigma_c \Delta Y}{\sigma_Y \Delta c}\right)^2 = \frac{c(KN)^2}{\sigma_Y^2 (1 + Kc)^2} \tag{40}$$

which is also plotted in Figure 4. For cases of intermediate relaxation, an approximate solution (see ref. 8 for further discussion) for the amounts of analyte bound can be obtained by summing the fluxes to and from the layer immediately above the sensor surface, giving:

$$V \frac{\mathrm{d}c_v}{\mathrm{d}t} = A\left[\frac{(c - c_v)D}{\delta_d} + k_d \theta_a / A - k_a c_v \varphi\right] \tag{41}$$

and equating the left hand side to zero to solve for c_v yielding:

$$c_v = \frac{cD + k_d \theta_a \delta_d}{D + k_a \delta_d \varphi}. \tag{42}$$

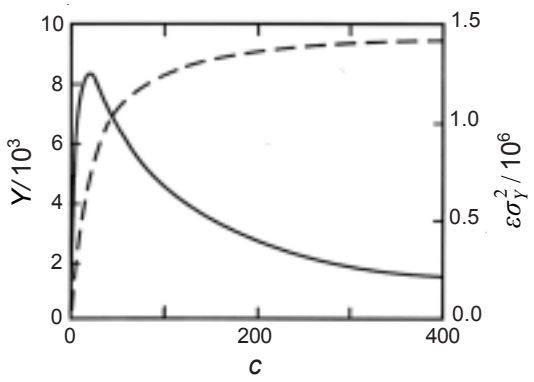

Figure 4. Y (solid line) and ε (dotted line) vs c for reversible analyte-receptor binding, characterised by an affinity K=0.05.

9. CHEMICAL FOG

Most clinical trials are carried out using biological fluids such as serum, in which even in the case of a heavily infected patient the analyte (antibody) may only constitute a fraction of a percent of the total protein content of the sample. All these proteins are potentially able to bind to the detector, generating output Y even in the absence of analyte ("chemical fog"). Typically the proteins (mostly albumin, fibronectin and non-analyte immunoglobulin) have a very weak affinity K_f for that part of the detector not actually covered by receptors, and no affinity at all for the receptors themselves. Under these conditions, clearly the detector completely covered by receptors will be unaffected. For incomplete receptor coverage the fraction of detector uncovered is $1 - \theta_r$, and therefore the contribution of chemical fog to the output is $(1 - \theta_r)c_f K_f$ where c_f is the concentration of fog proteins. The output is then:

$$Y = f(X) + (1 - \theta_r)c_f K_f. \qquad (43)$$

where $f(X)$ is for example $(1 - e^{Xb/N})N\theta_r$ for incomplete detector coverage due to diminished N. Note that the detectivity will be unaffected by chemical fog as can be seen by differentiating eqn (43).

10. LATITUDE

Another important sensing parameter is the latitude, i.e. the exposure range over which the detector is useful, i.e. where $\varepsilon \geq \varepsilon_{\min}$. $\varepsilon_{\min}$ can be defined by drawing an analogy between the frequency response of an electrical circuit and the plot of detectivity versus exposure (which are similar in shape). For electrical circuits the latitude is conventionally the frequency bandwidth over which the gain is 3 dB (a factor of $1/\sqrt{2}$) below the maximum gain. Hence one may define the latitude of the chemical detector as being the exposure range over which the detectivity is not less than 3 dB below the maximum detectivity. Note in particular that increasing the valence of the analyte increases the latitude, and hence bidentate IgG antibodies have a higher latitude than multivalent IgM, which should have an even higher latitude than bidentate IgG. One might speculate that nature makes use of multivalence in this way to prevent an immoderately large response to a minor infection which would saturate the response of a monodentate detection system.

11. CONCLUSIONS

The detectivity, defined as the ratio of input to output signal to noise power ratios, is a universal measure of detector response. A chemical sensor responds to a stream of analyte particles in an analogous though somewhat different way from the response of a photodetector to light quanta.

Different sources of detection inefficiency may give similar responses but very different detectivities. Multivalence is a particularly useful source of "inefficiency" which may render the detector more resistant to spurious noise. Notice that the valence of an analyte is always defined with respect to a given receptor; the designer of a sensor has in principle considerable freedom in choosing a capture layer which can interact with more than one zone of an analyte molecule.

Chemical fog engendered by huge excesses of non-specifically binding particles has no effect on the detectivity, provided the nonspecific interaction is reversible.

REFERENCES

1. J. J. Ramsden. *Q. Rev. Biophys.* **27** (1994) 41.
2. H.J. Zweig. *J. Opt. Soc. Am.* **51** (1961) 310.
3. E. Rabinowitch. *Trans. Faraday Soc.* **33** (1937) 1225.
4. P. O. Luthi, J. J. Ramsden, & B. Chopard. *Phys. Rev. E* **55** (1997) 3111.
5. V. G. Levich. *Physicochemical Hydrodynamics*. Englewood Cliffs, NJ: Prentice Hall (1962).
6. M. von Smoluchowski, *Phys. Z.* **27** (1916) 557.
7. L. A. Spielman & S. K. Friedlander. *J. Colloid Interface Sci.* **46** (1974) 22.
8. J. J. Ramsden. In: *Biopolymers at Interfaces* (ed. M. Malmsten), New York: Dekker (1998).
9. J. J. Ramsden & M. Máté. *J. Chem. Soc. Faraday Trans.* **94** (1998) 783.
10. *ELISA in the Clinical Microbiology Laboratory* (ed. T. G. Wreghitt & P. Morgan-Capner). London: Public Health Laboratory Service (1990).
11. R. C. Jones. *Nature* (Lond.) **170** (1952) 937.
12. P. J. Flory. *J. Am. Chem. Soc.* **61** (1939) 1518.
13. I. Palásti. *Publ. Math. Inst. Hung. Acad. Sci.* **5** (1960) 353.

A. NOTATION

- γ — Gain of the sensor
- δ_a — Reaction distance
- δ_d — Diffusion boundary layer thickness
- ε — Detectivity
- θ_a — Fraction of receptors occupied by analyte
- θ_r — Fraction of detector surface occupied by receptors
- σ_Y^2 — Output variance per receptor
- σ_X^2 — Input variance per receptor
- τ — Exposure time

τ_d — Residence time of analyte at a receptor
a — Area of one receptor
A — Total area of detector (= unity)
b — $e^{-U/kT}$
c — Bulk analyte concentration
c_v — Analyte concentration above the interface
D — Analyte diffusion coefficient
I — Analyte flux per unit area
k — Boltzmann's constant
k_a — Adsorption rate constant
k_d — Desorption rate constant
N — Total number of chemical receptors on the detector
P_n — Proportion of receptors struck by n analyte molecules
r — Radius of the receptor
t — Time
T — Absolute temperature
U — Analyte—receptor interaction potential
V — Unit volume
x — Mean number of analyte molecules incident on one receptor
X — Maximum possible number of analyte molecules incident on the detector over a time τ
X^* — Actual number of analyte molecules incident in the detector over a time τ
y — Number of analyte molecules captured and counted per receptor
Y — Total output signal
z — Distance perpendicular to the sensor surface