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Self-diffusion of ions in gels

Thomas Caldwell Smith

SELF-DIFFUSION OF IONS IN GELS

BY

THOMAS CALDWELL SMITH, B. S.
UNIVERSITY OF RICHMOND
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Approved
Addison D. Campbell
Ernest G. Hu
Jack J. Fox
James N. Boyd
J. B. Leftwich
Billy W. Sloope
W. Allen Powell
H. Pearce Atkins

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ABSTRACT

Radioisotope tracer techniques were used to determine the self-diffusion coefficients of the ions of sodium, potassium, cesium, chloride, bromide, and iodide in 3% agar gel, and the self-diffusion coefficient of the sodium ion in gelatin-agar gels at differing pH values of the gel.

The coefficients in agar gels were found to be in agreement with the Nernst equation for the diffusion coefficients of ions in aqueous solution. The diffusion coefficients in gelatin-agar gels were found to be higher in the presence of non-active NaCl than in its absence. The value of the coefficient was lower at the isoelectric point of the gelatin-agar gel than at pH values on the acid and basic sides of the isoelectric point. The value of the self-diffusion coefficient for the sodium ion found in gelatin-agar gel of pH 3.1 and non-active NaCl concentration of 110 microequivalents per milliliter was larger than the coefficient value in aqueous solution.

INTRODUCTION

The purpose of this experimental investigation is to determine the coefficients of self-diffusion of the ions of sodium, potassium, cesium, chloride, bromide, and iodide in agar gel and to study the effect of varying the pH of a gelatin gel on the coefficient of self-diffusion of sodium.

Diffusion is a transport process in which mass is transferred across a concentration gradient. Thus, the movement of a particle from one equilibrium position to another constitutes diffusion. This redistribution of matter leads to a homogenization of the system. The term self-diffusion is applied to any diffusion measured by use of a tracer in the absence of a chemical concentration gradient. In this case the gradient that exists is an isotopic gradient.

Every living cell is dependent upon diffusion for the procurement and distribution of necessary nutrients and for the removal of the products of metabolism. For this reason the subject has been one of considerable interest to science for over two centuries.

The earliest studies in this field were attempts to explain certain physiological processes in plants and animals. These experiments (for example, Nollet[23] 1748, Fischer[13] 1822, Poisson[25] 1827, Ludwig[19] 1849) were concerned with the diffusion of water and solutes across animal membranes. This work led to studies carried out by Fick[12] (1855) in diffusion in the absence of a membrane or "free diffusion" upon which the current theory of diffusion may be said to be based.

Robert Brown[4] (1828) observed that small suspended particles exhibited a continuous irregular motion. Brownian movement was subsequently shown by Einstein[9] (1905), Perrin[24] (1909) and others to be a visual manifestation of the diffusion mechanism.

Einstein also showed through an application of Stokes' Law that diffusion under the influence of gravity was dependent upon the radius of the diffusing particles if the particles were uncharged. Similar studies tried to relate the diffusion coefficient to the molecular weight of the particles. This relation is discussed by Riecke[26] (1890), Euler[10] (1897), Zeile[31] (1933), and others, and the relation $D(M)^{\frac{1}{2}} =$ constant is usually obtained, although many exceptions are found, particularly when the particles being compared are not closely related chemically.

The above relations, however, cannot be successfully applied to the diffusion of ions. The diffusion coefficient of an electrolyte is dependent upon the behavior of both of its ions, and the determinations of the coefficient of diffusion of one of the ions without taking into account the effect of the other ion have been shown by Bruins[5] (1931) to give values much larger than those reported by most investigators. Nernst[20] (1926) introduced a general discussion of the theory of ion diffusion in aqueous solution. He concluded that the coefficient of diffusion of an ion was proportional to the ion mobility.

Gels are found to have much to offer in the study of

diffusion. The use of gels eliminates the problems of convection and thermal and mechanical mixing in the system being studied. The rigidity of the gels also allows for the application of sharp boundary conditions. Voigtlander[30](1889) studied the diffusion coefficients of four electrolytes in agar gel and concluded that any differences observed in the coefficients of diffusion in different concentrations of gel were within experimental error. Later studies by Bechhold and Ziegler[3](1906) and Friedman[14](1930) have indicated, however, that the more concentrated the gel the slower the diffusion. Between the concentrations of 0.8% and 5.15% agar gel the diffusion of urea was found to be slowed by 36%.

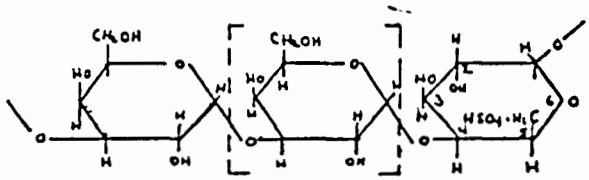
In more recent work Fujii and Thomas[15](1958) made a study of the self-diffusion of sodium ions in agar gels. They allowed Na(22)Cl to diffuse out of a small rod of agar gel through a thin membrane of unwaterproofed cellophane into a solution of inactive sodium chloride. The diffusion coefficient was determined by analyzing the amount of radioactive isotope that had entered the NaCl solution.

Schantz and Lauffer[27](1962) have reported an improved method of studying diffusion in gels. Their technique involves bathing the end of a cylinder of gel, free of solute, with a solution of the substance (salts and non-electrolytes) to be studied. The gel is then sliced and the slices are chemically analyzed.

The use of gels, however, does introduce some complexities into the diffusion studies. The gel substance reduces

the effective volume of the solvent, that is, in every unit volume of the gel, there is less than a unit volume of the solvent present because of the space occupied by the gel substance. The gel substance also presents obstructions to the diffusing ion and may exert attractive or repulsive forces on the ions.

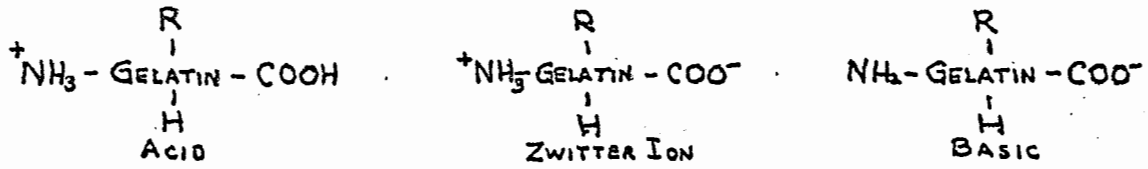
Agar is a dried hydrophilic colloidal substance which is extracted from Gelidium cartilagineum and related red algae. It is generally considered to be composed mainly of the calcium salt of the sulfuric ester of a linear polygalactose for which the following chemical structure has been suggested:[32]



Chemically it can be written $(R\text{-}O\text{-}SO_2\text{-}O)Ca$, where R is the carbohydrate. Agar gels have been shown by Araki[2] (1956) to have a small cation exchange capacity. The cation exchange capacities of different agars have been studied by Currie[7] (1955), who found that the exchange capacity varied directly with the sulfur content. Agar goes into aqueous solution at about 100°C and upon cooling sets in a stiff homogeneous gel.

Gelatin is a protein and is prepared from the skin of mammals. As is the case for all proteins, gelatin is an amphoteric electrolyte and can be given either a positive or negative net electrical charge by adjusting the pH of the aqueous solution. The isoelectric point of a protein is by definition that pH value of its solution at which it does not

migrate in an electric field. Reference to Figure (1), the titration curve for gelatin, shows that the isoelectric point for gelatin is pH 4.8. Above that pH value it is seen that there are bound OH^- groups and below that pH value there are bound H^+ groups. Thus by increasing the pH of a solution of protein to a value above the isoelectric point, the protein can be charged negatively, and by decreasing the pH value, the protein can be charged positively. This can be represented as below:



This fact suggests the study of the effect of the pH of a protein gel upon the coefficient of diffusion of sodium.

Crank[6](1956) studied the effect of binding in gelatin gels on the diffusion coefficients of metal ions. He observed that the diffusion coefficient is given by the expression

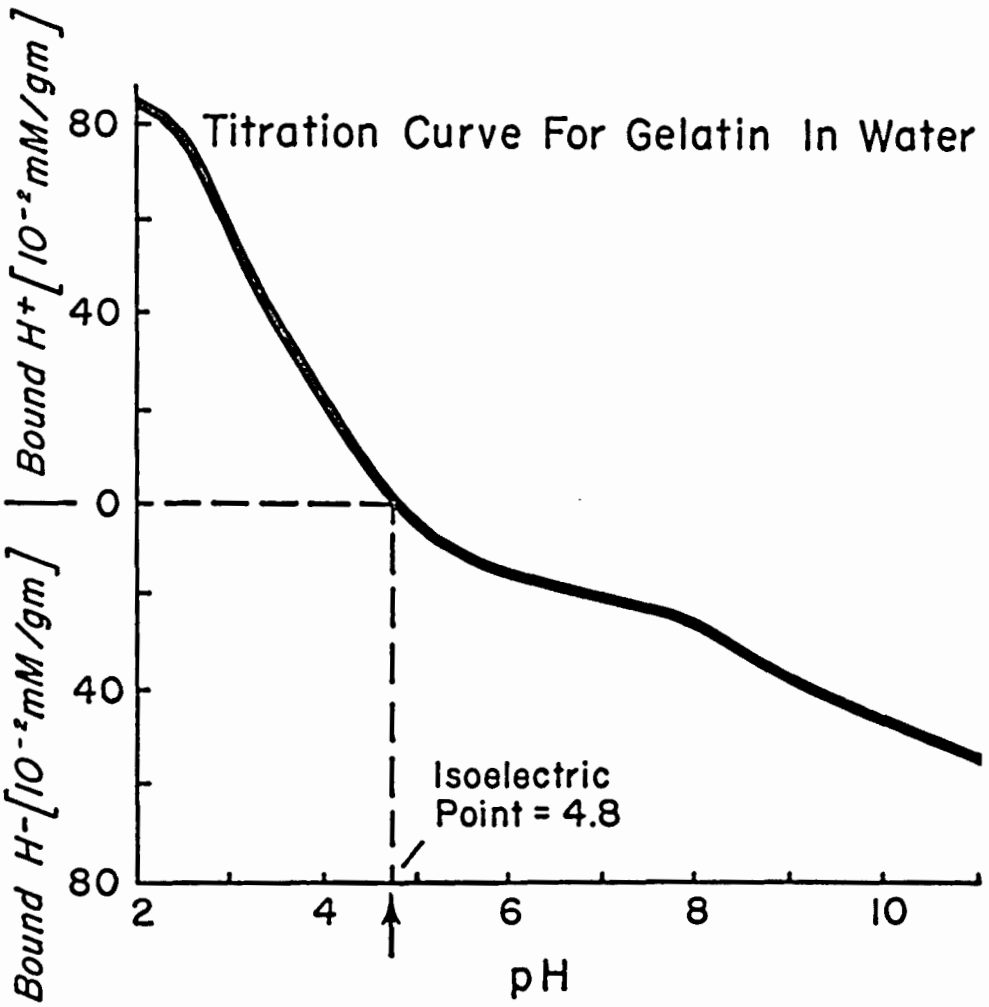
$$D_g = D_g^o/(R + 1)$$

where D_g is the coefficient determined in the protein gel, D_g^o is the coefficient in a similar gel in which there are no electrical interactions between the protein and the ions, and R is the ratio of the number of ions bound by the charged protein to the number not bound.

Using the results of Crank's work as a basis, Newsom and Gilbert[22](1964) studied the effect of binding on the coefficient of diffusion of zinc ions in a gel made from collagen derived from rat skin. All of their determinations were made at

Figure 1.

Titration Curve for Gelatin. Reproduced from Netter[21], p. 316.



pH values on the basic side of the isoelectric point. They concluded that three different sites in the collagen molecule had the capacity to bind the zinc ion to the collagen.

The investigations sited are but a few of the vast number in the literature relating to diffusion, but they give an indication of the course that the study has followed and are the ones deemed most pertinent to the work reported here.

THEORY

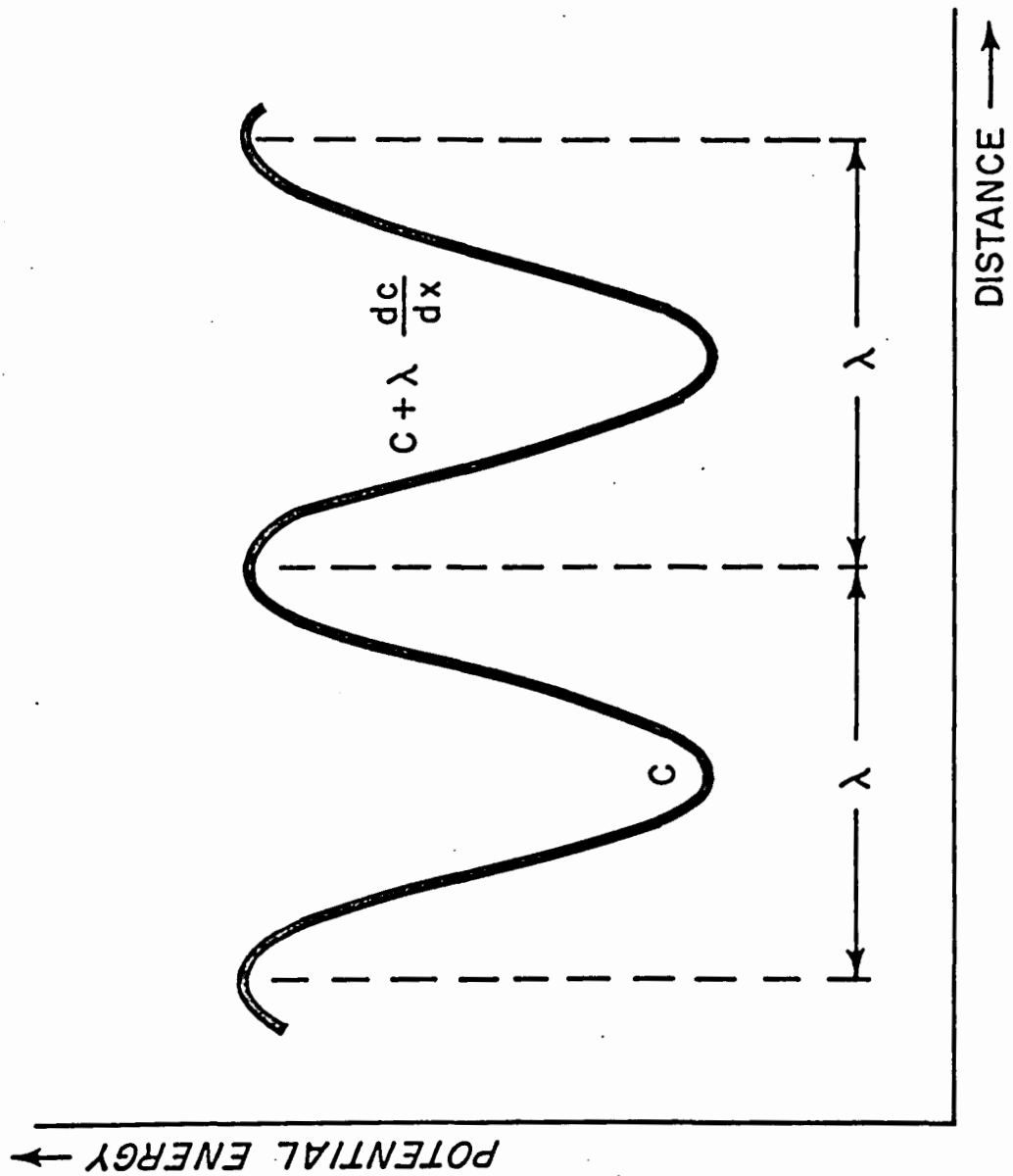
It is well known from the theory of specific heat that atoms in a lattice vibrate about an equilibrium position. Each atom is surrounded by a sheath of nearest neighbors. Occasionally the vibrations become energetic enough so that an atom can slip through its sheath of neighbors to occupy a new lattice position. It is movement such as this that leads to diffusion in solids.

The situation in semi-solids, such as gels, is similar though not as well ordered. The structure of semi-solids is much less rigid than that of solids and the particles are not fixed to definite lattice sites. However, the immediate surroundings of any given particle are ordered for a short distance from the particle, resembling the arrangement in a crystal. The particles oscillate in "cages" formed by their nearest neighbors. An energy configuration of the cage may occur which allows a particle to slip through into a new equilibrium position. This is similar to the process in a crystal except that the average distance moved in a semi-solid is not a full lattice distance and the energy barrier that a particle must overcome may be smaller than in a crystal.

For the sake of simplicity assume that diffusion is taking place in one dimension and that a particle has an equal probability to move in either direction. Eyring[11](1953) suggests the diagram given in Figure (2) as the potential energy diagram for simple diffusion. Let c be the concentration of particles per unit length at the first minimum and

Figure 2.

Plot of Potential Energy versus Distance. Reproduced from Eyring[11], page 104.



$(c + \lambda dc/dx)$ that at the second minimum. λ is the distance between successive potential energy maxima. If p is the probability per second of a jump in either direction, then the following expression for the number of molecules per second, q, crossing the barrier is obtained:

$$q = \lambda pc - (c + \lambda dc/dx)\lambda p \qquad (1)$$
$$= -\lambda^2 p dc/dx$$

The factor $\lambda^2 p$ with dimensions of $(\text{length})^2(\text{time})^{-1}$ is defined as the coefficient of diffusion D, and equation (1) becomes

$$q = -D\, dc/dx \qquad (2)$$

which is Fick's First Law of Diffusion.

This equation gives only the steady state condition for diffusion. The change in concentration in any region with time can be found by considering a region of unit cross-sectional area and of length dx. The increase in concentration in this region per unit time is the amount of material diffusing out, divided by the volume of the region. This can be expressed mathematically as

$$dc/dt = \frac{1}{dx}\left[-D(dc/dx)_x + D(dc/dx)_{x+dx} \right] \qquad (3)$$

assuming D is independent of concentration. The subscripts indicate the points of evaluation.

Now,

$$(dc/dx)_{x+dx} = (dc/dx)_x + (d/dx)(dc/dx)dx \qquad (4)$$

so that equation (3) becomes

$$dc/dt = D(d^2c/dx^2) \qquad (5)$$

which is Fick's Second Law of Diffusion.

A solution of this second order differential equation is found to be of the form

$$c = (a/t^{\frac{1}{2}}) \exp(-x^2/4Dt) \qquad (6)$$

where a is a constant.

Shewmon[28] (1963) considers the solution of this second order equation when the initial distribution of concentration is

$c = 0$ for $x < 0$, at $t = 0$
$c = c_o$ for $x > o$, at $t = 0$.

The solution for this case can be obtained by imagining the region of $x > 0$ to consist of n slices, each of thickness Δx_i and unit cross-sectional area. Consider one particular slice. It initially contains $c_o \Delta x_i$ of solute. If the region surrounding this slice were considered solute free, the diffusion after time, t, would be that given by the thin film solution, that is, equation (6) with $a = 2\sqrt{\pi Dt}$ or

$$c = c_o/2\sqrt{\pi Dt} \exp(-x^2/4Dt). \qquad (7)$$

This result is in no way affected by the fact that there is solute in the adjacent slices. The solution thus is a superposition of the solution for a thin film. If x_i is the distance to the center of the ith slice from $x = 0$, the concentration at any given value of x after time t will be

$$c = c_o/2\sqrt{\pi Dt} \sum_{i=1}^{n} \Delta x_i \exp -(x-x_i)^2/4Dt. \qquad (8)$$

From the definition of an integral, as the number of slices approaches infinity and the thickness approaches zero, this is

$$c(x,t) = c_o/2\sqrt{\pi Dt} \int_o^{\infty} \exp -\frac{(x-x_i)^2}{4Dt} \, dx_i . \qquad (9)$$

Substituting $(x-x_i)/2\sqrt{Dt} = y$, this becomes

$$c(x,t) = c_o/\sqrt{\pi} \int_{-\infty}^{x/2\sqrt{Dt}} \exp(-y^2)dy . \qquad (10)$$

By putting

$$\text{erf}\ (x) = 2/\sqrt{\pi} \int_{o}^{x} \exp(-y^2)dy, \ -\text{erf}(-x) = \text{erf}(x),$$

$$\text{erf}\ (\infty) = 1,$$

where erf(x) is Gauss' error function, this can be written as

$$c(x,t) = c_o/2\ (1 + \text{erf}(x/2\sqrt{Dt})) . \qquad (11)$$

This equation can be rearranged to the form

$$\text{erf}(x/2\sqrt{Dt}) = (1 - 2c/c_o) . \qquad (12)$$

The boundary conditions for which the above solution was determined can be closely approximated by using two cylinders, one of which is filled with a radioactive isotope of the ion to be studied, which serves as the source, and the second filled with non-radioactive gel into which the ions diffuse. It is seen from equation (12) and the definition of the error function that the value of $\frac{x}{2(Dt)^{\frac{1}{2}}}$ can be found from a determination of the ratio c/c_o where c is the concentration of radioactive ions at a distance x into the second cylinder after diffusion has proceeded for a time t, and c_o is the original concentration of radioactive ions in the source. By plotting this value against the distance into the cylinder for which the ratio was determined, the self-diffusion coefficient can be obtained.

The form of the solution shows that the concentration gradient is proportional to $\exp(-x^2/4Dt)$ which is the Gauss' error curve. Thus a check on the system used would be to make a plot of the concentration gradient of the system versus

the distance from x = 0, or the interface between the two cylinders. This plot should yield a curve of the Gaussian form.

EXPERIMENTAL

The method employed was a variation of the one described by Schantz and Lauffer. It was developed independent of any knowledge of their paper and appears to offer some improvements. Instead of using chemical analysis to determine the coefficient of diffusion, radiotracer techniques are employed and, in place of the solution as a source, a second agar cylinder is used.

(1) Preparation of gels: The agar gel was prepared by adding three grams of Difco Bacto-Agar to 100 milliliters of deionized water. If the ion for which the diffusion coefficient was being determined was a cation, enough chloride salt of the ion was added to the gel to give a salt concentration of 110 microequivalents per milliliter of solution. If the ion were an anion, enough of the sodium salt of the ion was added to give a salt concentration of 330 microequivalents per milliliter. The salt concentrations were arrived at arbitrarily. The gel was then divided into two parts. To one of these parts was added the radioactive ion, contained in either the sodium or chloride salt of the ion of interest. The solution of the radioactive salt which was added had been previously neutralized, and, in each case, the amount added to the gel did not effectively alter the gel or salt concentration. Experiments in the agar gel were run for the ions of sodium, potassium, cesium, chloride, bromide, and iodide. One set of experiments was run for the bromide ion in agar in which the salt concentration had not been adjusted.

The gelatin gel was prepared in a similar manner. Na(22) was the only isotope used in this case. Three grams of Eastman purified calfskin gelatin and three grams of agar were added to 100 milliliters of de-ionized water. The addition of the agar was found necessary since the gelatin alone did not give a gel which would harden sufficiently to permit slicing. Experimental determinations of the coefficient of diffusion were made in gelatin in which the salt concentration was set at 110 microequivalents per milliliter.

The isoelectric point of the gelatin was given by the manufacturer to be at pH 4.8. Three batches of the gelatin gel were prepared and the pH values were adjusted so that one was charged positively, pH 3.1 at 24°C, one was charged negatively, pH 6.5 at 24°C, and one was neutral, pH 4.8. This was done by reading the pH of the batch being adjusted on a Radiometer pH Meter and adding NaOH or HCl as was needed to obtain the proper pH value. After the pH of a batch was adjusted to the desired value the batch was divided into two and to one of these was added the radioactive ion. Experimental determinations were made for the coefficients of self-diffusion for sodium at all three pH values in both salted and unsalted gelatin.

According to specifications by the manufacturers, both the agar and the gelatin contained small amounts of minerals. The precise minerals and amounts, however, are not known to the investigator.

(2) Procedures of measurements: The cylinders which were

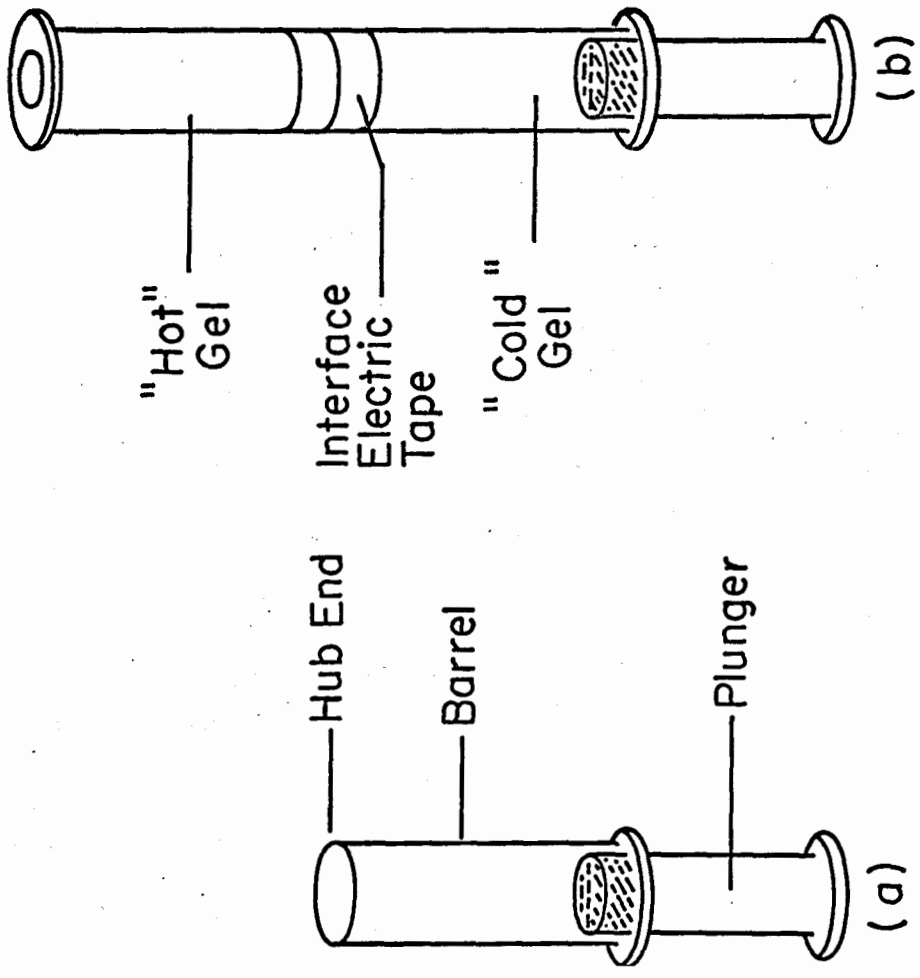
to contain the gel during the experiments were prepared by cutting off the needle ends of two dozen two-milliliter syringes. The end of the syringe barrel that had been cut was ground and fire polished to form a smooth surface. For each experiment the plunger of one of the syringes was placed in the barrel, and friction was allowed to hold it in place near the end of the barrel. (Refer to Figure (3-a)). The syringe was then placed in an upright position resting on the end of the plunger.

Non-radioactive gel containing the ion being studied was poured into the syringe from a ten-milliliter pipette. Care was taken in this procedure to eliminate all bubbles from the gel. The gel was allowed to protrude above the syringe because the gels shrink as they solidify. After the gel had hardened sufficiently, the protruding part was carefully sliced off. This slicing was done with a razor blade in the case of agar gels. A thin taut wire had to be used to slice the protein gel since the protein gel had a tendency to stick to the razor blade and consequently not give a clean cut.

Next a second syringe was mounted end to end atop the filled syringe and held firmly in place with electrical tape. (Refer to Figure (3-b)). Gel containing the radioactive ion was introduced into this second syringe by an eyedropper which had been altered by drawing the end into a thin capillary. Again care was taken not to introduce bubbles. The radioactive gel was always poured only after its temperature had

Figure 3.

Cylinder Arrangements.



fallen below 60°C to guard against melting the surface of the non-radioactive gel and thus mixing the two. The depth to which the radioactive gel was poured in the second syringe was adjusted so that it was equal in depth to the depth of the non-radioactive gel (about three centimeters). This was done so that equivalent boundary conditions would be obtained.

After the gel in the second syringe had hardened the plunger was carefully placed in the end. Undue pressure was avoided in this operation to prevent extruding the gel from between the syringes. These two syringes were placed in a plastic bag which was sealed and immersed in a water bath maintained at a temperature of 18.8 ± .1°C. After a period of from five to ten hours they were removed from the bath and separated.

The originally non-radioactive or "cold" gel was then sliced into thin sections by using the razor blade or taut thin wire. The slices were always taken from the cold side since it was found that a Gaussian distribution of concentration gradient was more closely obeyed on that side. This fact will be pointed up more clearly in the presentation of the results. The sectioning was done by exuding the gel from the syringe by applying pressure on the plunger. Ten slices were taken from each cold syringe. The slices were kept in a moist chamber to prevent evaporation while the weighing procedure was carried out. The slices were removed from the chamber one at a time and weighed on a Federal

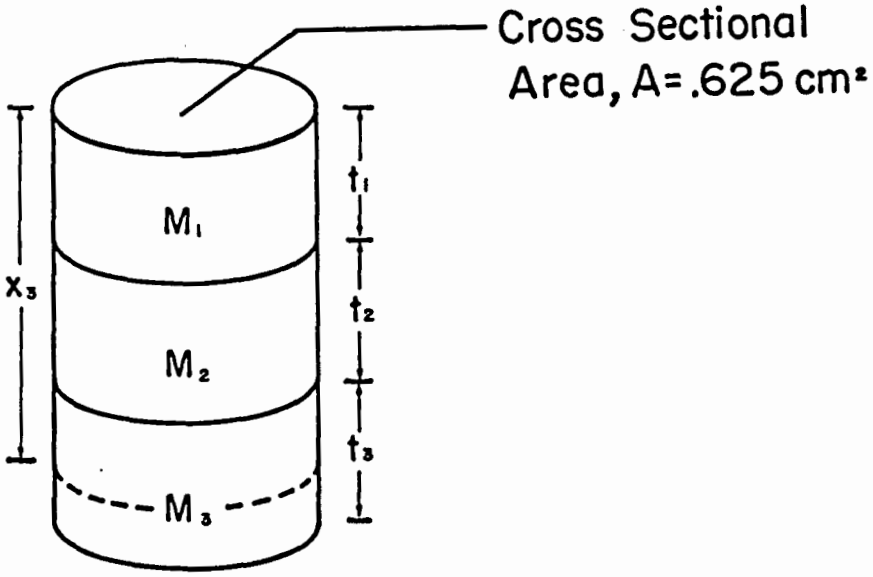
Pacific precision torsion balance to the nearest tenth of a milligram. As the slices were weighed they were placed in individual test tubes. The relative radioactivity of each slice was than determined by counting with a Baird Atomic model 132 scaler using a shielded well in which the scintillating crystal and photocell were mounted.

The original concentration of the radioactive ion was determined by a similar process. A single syringe was filled with radioactive gel and allowed to set for several hours. The gel was then sectioned and weighed. The slices were placed in test tubes and counted under the same conditions as were the experimental slices. No concentration gradient was found in this process indicating that the source was uniformly distributed throughout the syringe.

The distance into the plug from which each slice was taken was determined from the weight of the slice and the cummulative weight of all of the previous sections of the plug. Reference to Figure (4) will indicate the procedure used.

The data obtained from the weighing and counting were analyzed by use of the IBM model 1620 computer. By use of equation (12) and a plot of the distance into the plug versus the value of the limit of the error function, the value of the coefficient of self-diffusion can be obtained.

Figure 4.



$$P_{AGAR} = 1.003\ gm/cm^3$$

$$p = Mass/Volume$$

$$p = Mass/Area \cdot Thickness$$

$$\text{Thickness of Slice} = Mass\ of\ Slice/.625 \times 1.003$$

$$\text{Thickness of Slice} = 1.60 \times 10^{-3}\ Mass\ of\ Slice$$

The distance of a slice into the cylinder is taken as the distance to the middle of the slice. Therefore,

$$x_i = \frac{t_i}{2} + \sum_{j=0}^{i-1} t_j$$

$$x = 1.60 \times 10^{-3} \left(\frac{M_i}{2} + \sum_{j=0}^{i-1} M_j\right)$$

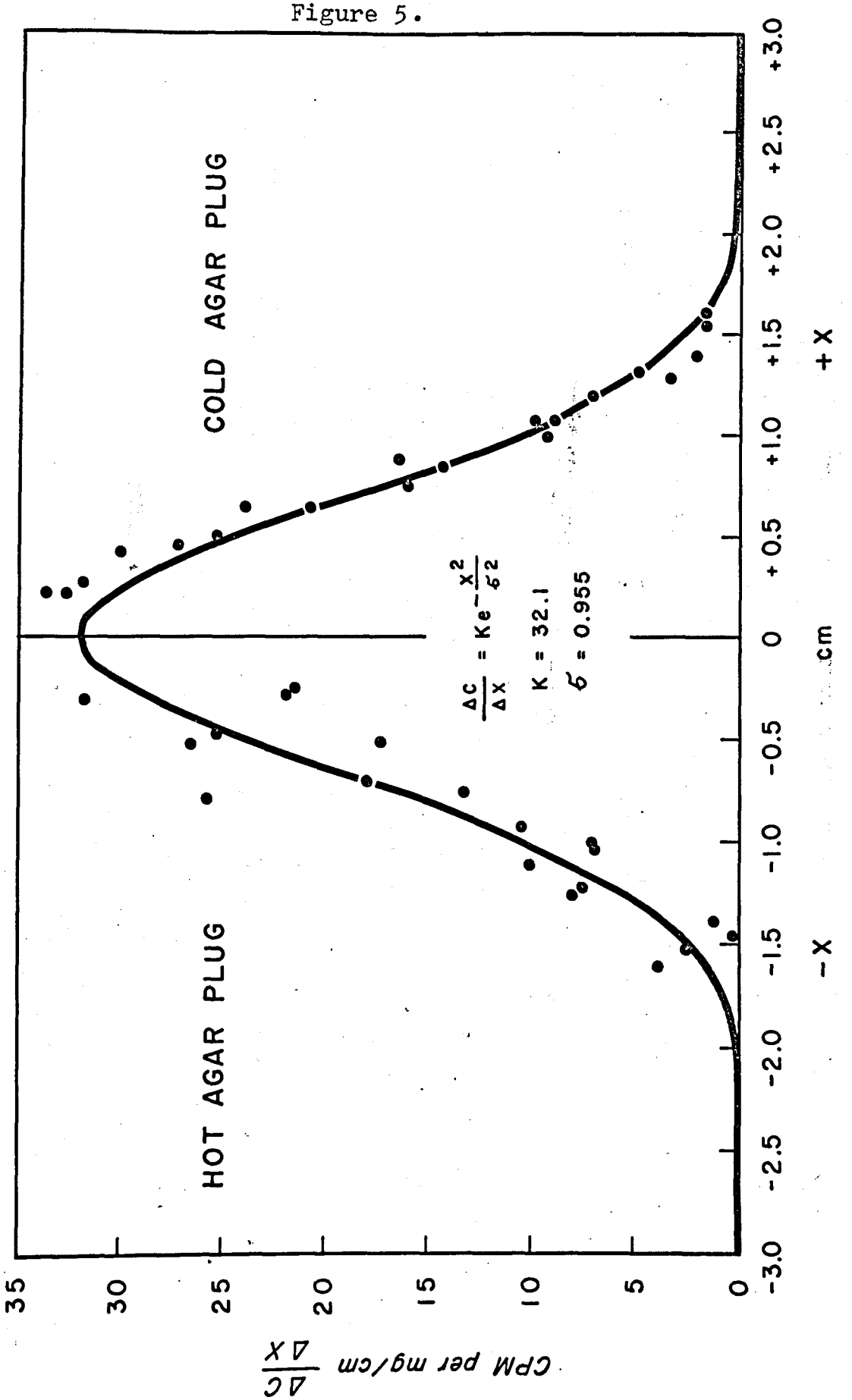
RESULTS

When diffusion took place from an initially radioactive cylindrical source into an initially non-radioactive cylinder of gel, a gradient was found that fitted a Gaussian distribution curve. The results of three five-hour experiments for sodium are shown in Figure (5). The gradient is plotted against distance into the cylinders. It can be seen from the figure that there is relatively little scattering of points on the cold side. Since this was the case, the data from the cold sides were used in calculating the self-diffusion coefficients.

Table (1) shows a typical data sheet. The value for the weight of the slice and the radioactive counts per five minutes were determined experimentally as previously described. All of the remainder of the data was rendered by the IBM 1620 with the exception of the value of the limit of the error function. This value was found from mathematical tables.

To determine the D for each experiment a plot was made of the values of the limit of the error integral versus the distance of the corresponding slice from the interface of the hot and cold plugs. The slope of the line obtained in this plot is inversely proportional to the square of D. The values of the slopes were obtained using the IBM 1620 and a least squares program. Only the first six experimental points were used to get these values since it was found that in most of the experiments the slope of the line changed at high values of the distance into the cylinder. Figure (6) illus-

Figure 5.


COLD AGAR PLUG
HOT AGAR PLUG
$\frac{\Delta C}{\Delta X} = Ke^{-\frac{X^2}{\sigma^2}}$
K = 32.1
σ = 0.955
$\frac{\Delta C}{\Delta X}$ CPM per mg/cm
35
30
25
20
15
10
5
0
-3.0
-2.5
-2.0
-1.5
-1.0
-0.5
0
+0.5
+1.0
+1.5
+2.0
+2.5
+3.0
cm
-X
+X

Diffusion Experiments

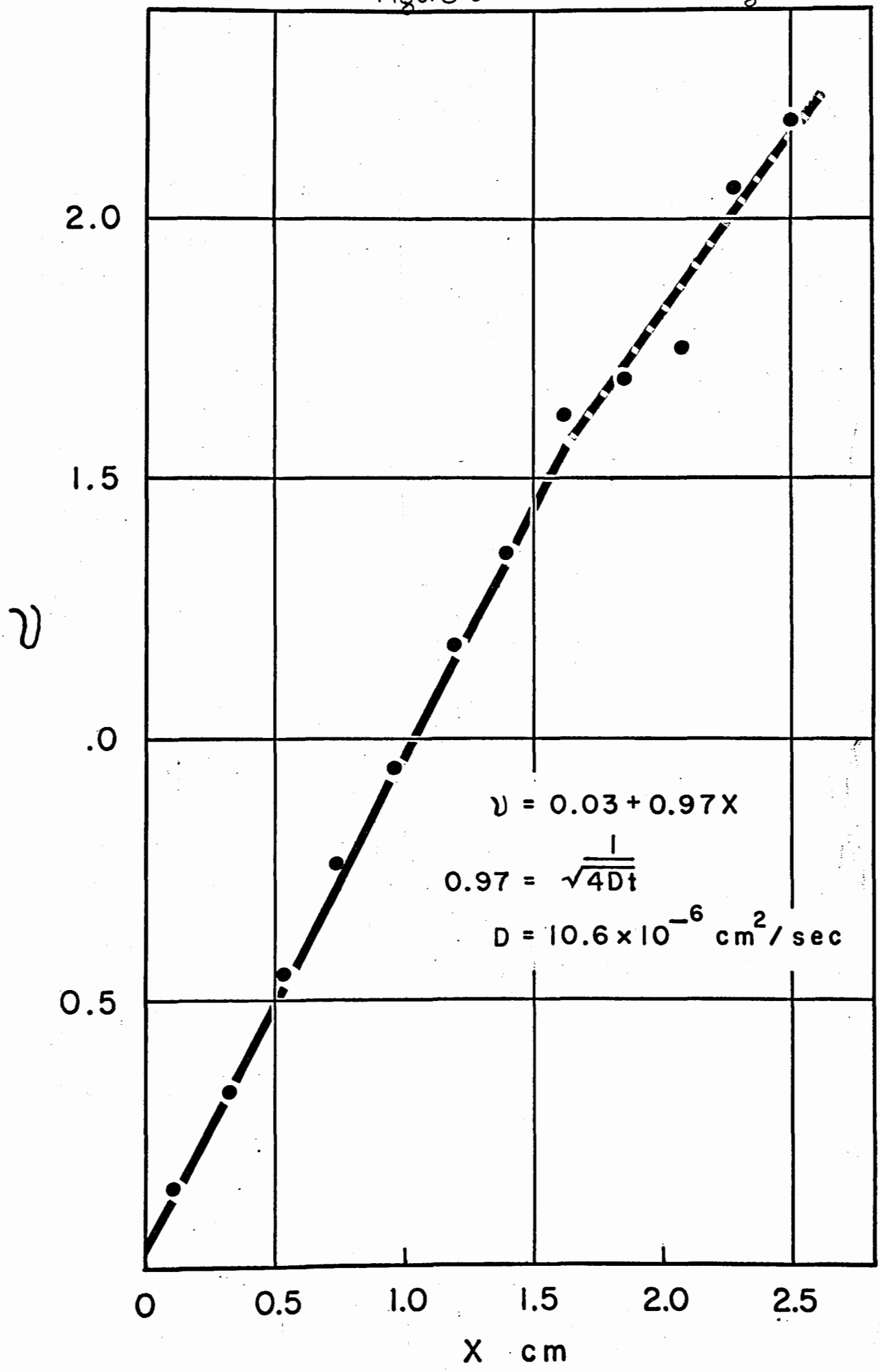
Experiment Number 2-Cl Date 7/17/64

Time of Experiment 5hr

Original Concentration 71.55 ct/min/mg

Comments Background 690 cts/min

No.	Wt.	Cts/Min	Cts/Min/Mg	Cum. Wt.	X	c_x/c_o	Erf	η
1	137.5	4939	30.90	68.75	.1100	.4320	.136	.121
2	172.3	4444	21.79	218.65	.3498	.3046	.391	.369
3	113.8	2379	14.84	356.7	.5707	.2075	.585	.576
4	148.6	2168	9.95	487.9	.7806	.1391	.723	.769
5	137.2	1571	6.42	630.8	1.009	.0898	.820	.948
6	152.6	1216	3.45	775.7	1.241	.0482	.904	1.177
7	144.1	950	1.80	924.05	1.478	.0252	.950	1.386
8	160.8	836	.91	1076.5	1.722	.0127	.975	1.585
9	137.4	737	.34	1225.6	1.961	.0048	.990	1.821
10	125.8	712	.17	1357.2	2.172	.0024	.995	1.985


2.0
1.5
.0
0.5
ν
$\nu = 0.03 + 0.97X$
$0.97 = \frac{1}{\sqrt{4Dt}}$
$D = 10.6 \times 10^{-6}$ cm²/sec
0
0.5
1.0
1.5
2.0
2.5
X cm

trates the results of one experiment. From four to ten experimental determinations were made of the coefficient of self-diffusion for each ion of interest. The average results of these determinations are shown in Tables (2 and 3). The errors resulting from weighing, counting, and temperature fluctuations were found to be much smaller than the statistical error introduced by averaging the values obtained for several determinations. The uncertainties tabulated are standard errors of means and were calculated from the formula

$$\sigma = \pm\left[\sum_{i=1}^{n} d_i^2/n(n-1)\right]^{1/2}$$

where d_i is the deviation of the ith value from the mean and n is the number of values used in the average.

Table 2: Self-diffusion coefficients of several ions in 3% agar gel at 18.8° ± .1°C. In the case of the cations the agar gel had a salt concentration of 110 micro-equivalents per milliliter. For the anions the salt concentration of the gel was adjusted to 330 micro-equivalents per milliliter. One determination was made for bromide in which the gel was unsalted.

Ion	Number of Determinations	Self-diffusion Coefficient ($D \times 10^6$) cm^2/sec	Ionic Radii[18] (Hydrated) ($r \times 10^8$) cm
Sodium 22	5	11.39 ± .30	1.49-7.90*
Potassium 42	5	16.16 ± .44	1.17-5.32
Cesium 134	6	15.50 ± .21	1.11-5.05
Chloride 36	10	17.10 ± .40	-
Iodide 131	5	18.02 ± 2.78	-
Bromide 82	5	16.13 ± .11	-
Bromide 82 (in unsalted agar)	10	14.28 ± .17	-

Table 3: Self-diffusion coefficients of sodium 22 in 3% purified calfskin gelatin-3% agar gel at several pH values of the gel. The isoelectric point of the gel is pH 4.8. The salt concentration is 110 micro-equivalents per milliliter.

Gelatin-agar	Number of Determinations	Self-diffusion Coefficient ($D \times 10^6$) cm^2/sec
pH 3.1 Unsalted	5	10.69 ± .45
pH 4.8 Unsalted	6	7.15 ± .15
pH 6.5 Unsalted	5	9.26 ± .17
pH 3.1 Salted	4	15.92 ± .29
pH 4.8 Salted	6	11.19 ± .37
pH 6.5 Salted	5	11.30 ± .57

* Depending upon method used.

DISCUSSION

(1). Observance of boundary conditions in the experiments:

Some explanation is needed for the break in the plot of the limit of the error function integral versus the distance into the cylinder. In almost every case, with the exception of five hour experiments using sodium, the value of the limit is found to abandon its linearity after about the seventh of the ten slices of the gel and to maintain a nearly constant value to the last slice.

The most probable explanation of this phenomenon seems to be that the system is not truly infinite but that the diffusing ions in the cases where the break is observed reach the end of the cylinder before the diffusion experiment is terminated. If this happened there would be effectively another source of radioactive ions established at the end of the cylinder which would result in the superposition of another concentration distribution upon the one expected for an infinite system. This second distribution would prevent the value of the limit of the error function from increasing in the manner which it theoretically should and would cause the leveling out of the plot that is manifested.

This hypothesis can be checked by calculating the distance that diffusion should proceed into the cylinder. The maximum value that any diffusing particle should travel can be found by determining the distance at which the value of the error integral is one. At this point the value of the limit is approximately three. Therefore the relation is

obtained

$$x_{max} = 3 \times 2\sqrt{Dt}$$

Using this equation and the value of $D = 17.0 \times 10^{-6} cm^2/sec$ which is approximately the value of D for all ions in agar except sodium and a time of five hours, x_{max} is found to be 3.3 cm. The average length of the cylinders was about 3 cm, so that it is seen that these diffusing ions would reach the end of the cylinder.

If the value of $D = 11.4 \times 10^{-6} cm^2/sec$ for sodium is used and the time is taken as five hours, x_{max} is found to be 2.7 cm. Thus in five hours the sodium would not have reached the end of the cylinder. This is what is observed and is indicated in Figures (5 and 6).

This explanation seems to be acceptable, and since the first six slices in every case were found to be uneffected, it was assumed that the theory for diffusion in an infinite cylinder could be safely applied if only these were used in the determinations.

(2). Comparison of experimental with theoretical values of D:

The results obtained in agar can be compared with the theoretical values for the ions in aqueous solution from the Nernst equation

$$D_i = \frac{RT}{F^2} \lambda i$$

where λi is the equivalent ionic conductance of the ions. Table (4) shows the comparison of the experimental values and the calculated theoretical values.

Table 4: Comparison of experimental values for the self-diffusion coefficient of several ions in agar-agar gel with the theoretical values calculated from the Nernst relation.

Ion	Experimental Value ($D \times 10^6$) cm^2/sec	λ_i	Theoretical Value ($D \times 10^6$)
Sodium 22	11.39 ± .30	42.8	11.0
Potassium 42	16.16 ± .44	64.2	16.5
Cesium 134	15.50 ± .21	67.1	17.2
Chloride 36	17.10 ± .40	64.3	16.5
Bromide 82	16.13 ± .11	66.3	16.9
Iodide 131	18.02 ± 2.78	65.3	16.7

The theoretical values tabulated are based on a theory which is expected to give good agreement only in the case of very dilute solutions. At higher concentrations when the solutions vary from ideality, the frictional forces and ion mobilities, which are considered to be constant at infinite dilution, are by no means constant. Thus, good agreement is not expected in this case, but it is seen that the experimental values show the correct order of magnitude and are in fair agreement with the theoretical values.

(3). Comparison of D with ionic radii:

From Table (2) it can be seen that no correlation can be drawn between the coefficients of diffusion of the ions and their ionic radii. This is in agreement with the results reported by Bruins and the theory of Nernst for diffusion of ions in aqueous solutions.

(4). Effect of salt concentration on D:

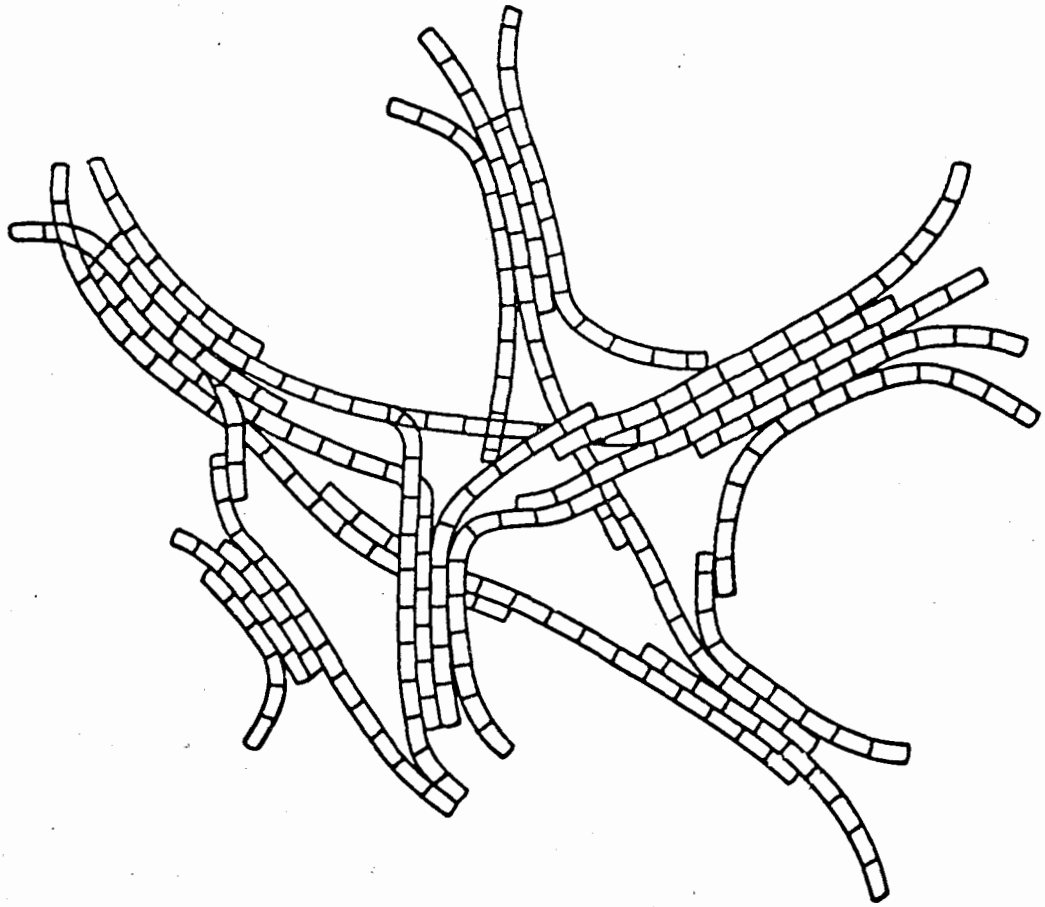
It is seen from Table (2) that the addition of NaBr to the agar gel increases the value of the diffusion coefficient

by 12.9%, from 14.28 x 10^{-6} cm^2/sec to 16.13 x 10^{-6} cm^2/sec. A similar increase in the self-diffusion coefficient of sodium is noted when salt is added to the gelatin gel. This is illustrated in Table (3). From the nature of self-diffusion, that is the replacement of one isotope by another, it can be understood why increased diffusion coefficient values are observed when salt is added to the gel. However, there may be other possible explanations for it.

One possible explanation can be found in the work of Abitz and Gerngross[1](1930). They determined that in a gelatin gel the ends of the gelatin molecules were tightly woven together. The addition of salt to the gel caused the gelatin molecules (Figure 7) to experience a loosening of the binding between them. This in turn opens more paths to diffusing particles and results in a higher value for the coefficient of diffusion.

This explanation, however, does not account for the coefficient of diffusion being higher in pH 3.1 salted gelatin than in aqueous solution. The reason for this phenomenum may be contained in the work of Durbin, et al. (1964). They studied the diffusion of the chloride ion across both animal and artificial membranes. They found that the addition of chloride to the side of the membrane to which diffusion was taking place enhanced the rate of diffusion by a factor of two in most cases. This phenomenon is referred to as exchange diffusion. Ussing[29](1948) postulated a mechanism for exchange diffusion in which a carrier for the diffusing ion traveled

Figure 7.



Gelatin Molecule. Reproduced from Netter[21], page 365.

back and forth across the membrane aiding in the transport of the ion. If the carrier itself could only cross the membrane in a complex with the diffusing ion, then the addition of an isotope of the ion to the solution to which diffusion was taking place would allow the carrier to make several return trips across the membrane and consequently increase the diffusion rate.

Although the exact machanism is not understood, the addition of salt to the gel might activate such an exchange diffusion and increase the value of the coefficient as is found in this investigation.

(5). Comparison of D_{Na} in agar with D_{Na} in gelatin-agar gel:

Comparison of the self-diffusion coefficient for sodium in agar (Table 2) with the values obtained and catalogued in Table (3) show that there is no difference between the coefficient in uncharged (pH 4.8) salted gelatin and negatively charged (pH 6.5) salted gelatin, and the value obtained in agar. However, on the acid side (pH 3.1) of the isoelectric point in the salted gelatin, the self-diffusion coefficient is found to be greater than in agar by 40%, $15.92 \times 10^{-6} cm^2/sec$ compared to $11.39 \times 10^{-6} cm^2/sec$.

It should also be noted that the value obtained in acid gelatin is greater than the value for sodium diffusion in aqueous solution. This violates all known theories of diffusion in gels. Lauffer has indicated that in a gel substance the coefficient of diffusion must always be less than diffusion in the solvent involved in the gel, so some new explanation

must be sought for diffusion as carried out in the pH 3.1 salted gelatin-agar gel. It seems likely that this explanation lies in the realm of exchange diffusion.

(6). Effect of pH on D_{Na} in gelatin-agar gel:

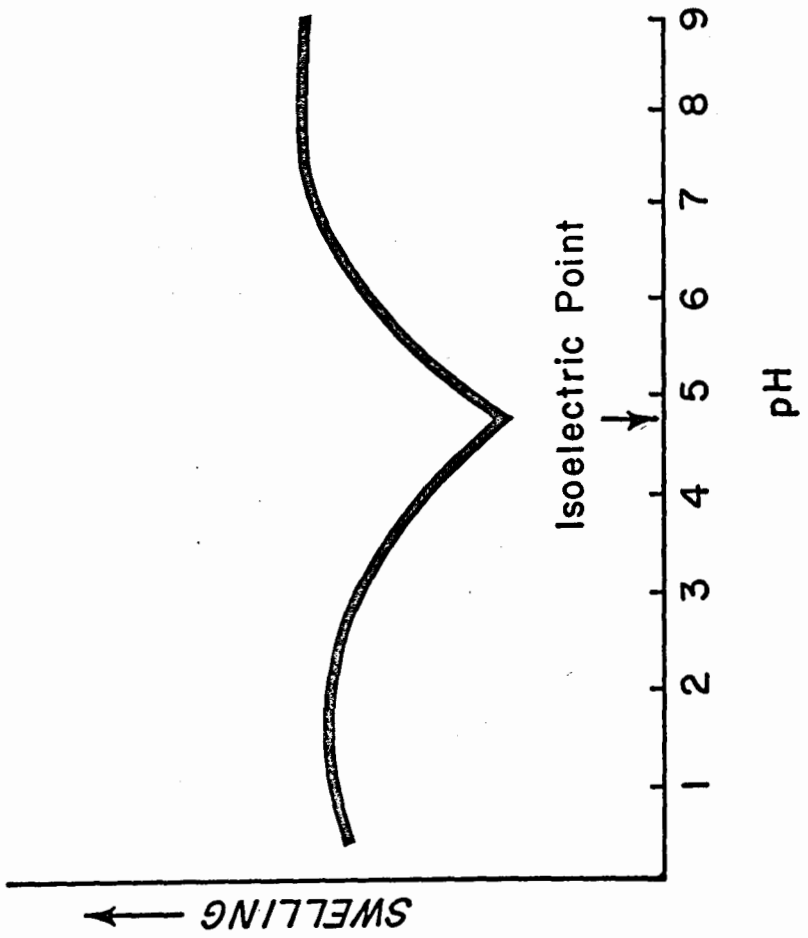
(a) Unsalted gelatin

Study of the self-diffusion coefficient for sodium in unsalted gelatin gel (Table 3) shows that the coefficient is lower at the isoelectric point than at pH values on either side of the point. A plot of the swelling in a gelatin gel as a function of pH (Figure 8) and reference to Table (3) indicate a direct relation between the swelling of the gelatin and the coefficient of self-diffusion. The swelling is at a minimum at the isoelectric point and increases at pH values on either side of the point. This same relation was noted in the discussion of the effect of salt concentration on the coefficient of self-diffusion.

(b) Salted gelatin

No clear conclusions concerning the effect of pH in salted gelatin gel can be drawn. The swelling effect noticed in the unsalted gelatin is reduced since the addition of salt reduces the swelling by repressing the Donnan effect. Hence, the increase found in the values cannot be attributed to swelling.

Figure 8.



Plot of Swelling versus pH. Reproduced from Netter[21], page 320.

CONCLUSIONS

(1). The results indicate that the coefficients of self-diffusion in agar gels are good approximations of the value of the coefficients in aqueous solution.

(2). The effect of the addition of a salt to the gel is to increase the value of the coefficient of self-diffusion in the gel.

(3). It is deduced that the inducement of swelling of the gel substance increases the coefficient of self-diffusion in the gel.

(4). No correlation is found between the coefficient of self-diffusion of an ion and the radii of the ion.

Further studies are indicated to determine the effect of the addition of salt to a gelatin-agar gel on the coefficient of self-diffusion.

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