IV. ADSORPTIVE STRATIFICATION IN GELS, II.

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It has been postulated [Bradford, 1916] that the formation of stratified deposits in gels may be explained by the action of the finely divided particles of the precipitate in adsorbing the solute from the gel in their immediate neighbourhood. The accompanying phenomena are, however, so numerous and of so varied a nature that it is necessary to examine them more in detail and to attempt to demonstrate their complete accordance with the theory.

Von Weimarn [1914] has shown that the form of the precipitate produced in a double decomposition of the type $AB + CD = AC + BD$ is dependent on the number, N , of crystallisation centres produced, determined by the formula :

$$N = \frac{P}{L} K_{AB} \cdot K_{CD} \cdot K_{BD} \cdot K_{AC} \cdot Z \dots,$$

where P is the mass of the substance AC to be deposited as precipitate,

L , the solubility of the precipitate,

K_{AB} , K_{CD} , etc., the degrees of aggregation, in solution, of the four components of the reaction, and

Z , the viscosity of the reaction medium.

The value of N is also proportional to the degree of aggregation of the reaction medium and of any other substance, or substances, present therein which do not enter into the double decomposition.

The occurrence of stratification in gels is therefore dependent on N , since it is only in cases where the precipitate is sufficiently finely divided that the adsorbing surface will be large enough to exhaust the solute from the adjacent region of gel. As the value of N decreases, in the presence of sufficient nutrient material, so the size of the individual aggregates, or crystals, will

increase, and the area of adsorbing surface diminish, until there is insufficient surface to produce stratification. On the contrary the greater the value of N , the more gelatinous the precipitate, and the slower the rate of diffusion of the precipitant through the layer of precipitate. So that, with very large values of N , the precipitant cannot permeate the precipitate in time to traverse the exhausted layer of gel immediately beyond, before this is replenished with solute by diffusion from the gel. Precipitation will then be continuous. When in this connection it is remembered that the solution in the gel must be sufficiently dilute to allow of its exhaustion from the region adjacent to the precipitate, giving a small value of P , it will be clear that zone formation can occur only in cases in which L is very small, and within somewhat narrow limits. This is fully confirmed by observation.

Since, however, the already dilute solutions meet in the gel only by diffusion, the value of P will be actually much smaller than that corresponding to the concentration of the hypotonic reagent. Consequently there will be a tendency for the formation of larger crystals than would result from the instantaneous mixing of aqueous solutions.

Under similar conditions Dreaper [1914] obtained large crystals of lead chloride and ferrocyanide by allowing the reacting solutions to diffuse in opposite directions through a capillary tube. Indeed the process of allowing the slow mixing of solutions has long been known as a method for the preparation of crystals of sparingly soluble substances.

The occurrence of such large crystals is a remarkable feature of reactions in gels. This is particularly noticeable in silicic acid gels, frequently observed in agar and more rarely in gelatin. It is thus evident that the gel medium has itself an extraordinary influence on the form of the precipitate resulting from a particular double decomposition, which tends to counteract, in a greater or less degree, the effect of the slow diffusion. Many examples of the influence of the gel medium on the value of N could be given, and it is convenient to consider this in connection with the effect of altering other physical conditions. The original Liesegang reaction with silver bichromate takes place readily in gelatin gels of various concentrations, while in 1 % agar remarkable ribbon-like crystals are formed which are much too large to cause stratification. If, however, the reagents are reversed, so that the silver salt is contained in the gel, beautiful laminae again result. These are, however, almost black and crystalline instead of red-brown and apparently amorphous as in gelatin. This may be attributed to an increased value of N resulting from the effect of a larger mass of that solute which is in the

state of greater aggregation, as in the case of the gelatinous precipitate of lead iodide, produced with basic lead acetate, mentioned below. If potassium chromate be substituted for the bichromate, only detached crystals are formed, without a trace of stratification; though here, of course, the crystals consist of silver chromate instead of bichromate. Returning to the case of the agar gel containing potassium bichromate, if the concentration of the solute be considerably diminished, the size of the crystals rapidly falls and an unmistakable tendency to stratification begins to appear, until, with a concentration of $N/100$ $K_2Cr_2O_7$, two separate bands of tiny rounded brownish black particles are formed at a distance of 2.5 mm. apart. With still smaller concentrations dark reddish brown colorations are noticeable in the weaker part of the gel below the bands, indicating sol formation. On reducing the solubility of the precipitate by mixing the agar gel with half its volume of absolute alcohol, an ordinary concentration of $N/20$ $K_2Cr_2O_7$ gives several bands of tiny feather-like crystals, 2 mm. thick, separated by clear spaces, with indications of sol formation in the lower part of the tube.

The case of lead iodide differs from that of silver bichromate in that beautiful laminations are produced in 1 % agar, and, as stated by Hatschek [1914], only a continuous gelatinous precipitate is formed in gelatin. The statement appears, however, to refer to the concentrations of the reagents used in 1 % agar, viz., 4 % of KI in the gel treated with concentrated $Pb(NO_3)_2$. The value of N in this case is too large. By using $N/20$ KI, and $N/1$ $Pb(NO_3)_2$, or $N/1$ $Pb(C_2H_3O_2)_2$, the precipitate becomes more permeable and stratification occurs in the lower half of the tube, although the precipitate is still much more gelatinous than in agar. A similar gelatinous precipitate is also obtained in agar by treating a gel containing $N/20$ KI with basic lead acetate solution. The number of crystallisation centres is increased by the greater aggregation of the precipitant and the precipitate becomes almost impermeable, so that only a few bands are produced. When the reaction takes place in silicic acid gel containing about 3.5 % of silica, which allows about the same rate of diffusion as in 5 % gelatin and 1 % agar, no stratification is visible, but the salt is deposited in large crystals and growth and dendritic forms. Von Weimarn has shown that growth forms and spherical crystals are conditioned by a value of N which is intermediate between those corresponding to complete crystals and so-called amorphous precipitates.

Lead chromate forms a gelatinous precipitate in agar so that it is only in very dilute concentrations, about $N/100$, that stratification occurs, the

result being then very beautiful. In gelatin the precipitate is too gelatinous to stratify, while in silicic acid crystalline needles are produced. Cupric phosphate gives a gelatinous precipitate in agar, which however becomes distinctly stratified in very dilute concentrations, whereas in silicic acid it yields remarkably beautiful deep blue discs. The basic hydroxide also gives good bands in silicic acid, but has not been obtained, so far, other than as a continuous gelatinous precipitate in test tubes containing agar gel. However, by working with U-tubes, half filled with pure agar gel between the reacting solutions in the limbs above, the value of P is so much reduced where the liquids meet that stratification takes place even with an initial concentration of $N/20$. Zone formation is assisted in this case by the increased facility with which the gel can be exhausted of solute owing to the great dilution in the neighbourhood of the meeting solutions.

From these considerations it will be clear that the formation of a banded precipitate is dependent on the number of crystallisation centres produced, and that this quantity is affected by the properties of the gel in which the reaction takes place. This influence of the reaction medium may be explained by the fact that it is actually a dilute aqueous solution of the gel substance in a high degree of aggregation. The number of crystallisation centres is therefore proportionally increased. Under conditions of sufficient dilution the production of sols would be favoured.

The well-known "protective values" of gelatin, agar and silicic acid are in the proportion which would explain the observed variations in the form of the precipitate. In the masterly paper previously quoted, in which he collects together the great variety of facts which require explanation, Hatschek suggests that this protective action may eventually be found to be connected with the behaviour of different gels.

Sol formation is however unnecessary for the production of a stratified precipitate, and could hardly occur, except momentarily, in normal cases. Dreaper [1914] obtained well-defined bands of copper sulphide in columns of sand, which has no protective action. Although in gelatin, particularly, the instantaneous formation of a sol is possible on the first meeting of the diffusing solutions, it would be coagulated immediately by the rapid increase in the value of P leading merely to a more gelatinous precipitate; and it is noticeable that in cases where sol formation is most probable it is usually most difficult to induce stratification. As Hatschek has clearly demonstrated, supersaturation, in any form, is not adequate to explain the many varied cases of zone formation. Nor does it afford an explanation of the very different

results obtained when the solutions in and above the gel are interchanged, as for example with lead iodide, as previously recorded [Bradford, 1916].

The microscopic observation of reactions in gels has already been carried out by Liesegang [1913] for silver bichromate, and by Hatschek [1914] for lead iodide and chromate. The experiments were made in the plate form. The original descriptions are here reproduced. "On observing the region beyond the last line, for some time one perceives nothing, although, as remarked, silver chromate must be forming. Suddenly a new line appears at a distance of half a millimetre. In distinction from the previous line, which appears dark red by reflected light and black by transmitted light, the new line is yellow and very tenuous. In the course of the next ten seconds it becomes considerably intensified and fiery red in colour, but does not attain the density of the older line for some time. This solidification, which is coupled with a darkening of the colour, is reached in about three minutes." It is convenient to consider here a point raised by Hatschek [1914], in connection with the same reaction. "The drop of silver nitrate diffuses into the bichromated gelatin producing, at first, a band of silver bichromate two or three millimetres wide before ring formation commences, and, even if the concentration of silver nitrate be reduced to 5 %, a similar wide band is still built up." According to the adsorption theory, the silver nitrate diffuses into the gel containing potassium bichromate, forming a ring of precipitate of silver bichromate. This precipitate immediately begins to adsorb solute from the gel, which is precipitated as it collects, with constantly growing surface and increasing adsorbing power. In this way the region of gel next the precipitate tends to become gradually exhausted of potassium bichromate and fresh material diffuses towards this region from the more distant parts of the gel. A concentration gradient is thus set up towards the zone of gel in contact with the adsorbing precipitate, and this region eventually becomes so dilute that no further precipitate is formed. The wide band then ceases to grow. It is evident, therefore, that the formation of this wide band depends on the concentration of the hypotonic reagent in the gel, rather than on that of the precipitant. In the case of U-tube experiments this concentration gradient is already in existence when the diffusing solutions meet, and these are so dilute that band formation generally commences immediately. Considering now Liesegang's description: the silver nitrate slowly diffuses through the previous rings of precipitate into a region from which the potassium bichromate has been adsorbed, and no further silver bichromate is formed (not even as a sol, since this would

be indicated by its colour¹), until the precipitant meets the bichromate diffusing in from the gel. A faint new ring is then generated. The new precipitate commences to adsorb fresh solute from the gel, which is precipitated as it collects. The density of the ring, and incidentally the adsorbing surface, increases. As the solute is rapidly adsorbed from the thin layer of gel immediately in contact with the precipitate, fresh solute slowly diffuses towards this region, until the layer of gel adjacent to the precipitate again becomes exhausted of solute. The new ring will then cease to grow, and the whole process will be repeated until the available solute in the gel has been precipitated.

Since the adsorbed solute collects on the surfaces of the freshly deposited particles of the precipitate, the value of P/L will be greater there than elsewhere. Consequently precipitation will take place on the surfaces of the grains of the precipitate, which will grow equally in all directions. A further increase in concentration may result in the formation of an additional number of smaller centres, but the individual particles will grow in the same way. Microscopic examination of the rings and bands invariably reveals the spherical or approximately spherical shape of the constituent grains of the precipitate, which frequently reach macroscopic size. In many cases larger particles are seen imbedded in a finer grained precipitate. Hatschek's description of the lead iodide reaction records the observation of the actual growth of such spherical aggregates, which are characteristic of reactions in gels. "Here also the reaction begins with the appearance of a light yellow coloured strip. However, at the same time, numerous points of lead iodide appear on the inner side of the band which grow remarkably while the strip is only increasing slightly in intensity and width. When the isolated grains seem to have reached their full size, the band appears to grow suddenly and become opaque, but it has an essentially coarser structure than the silver chromate ring."

The progress of the lead chromate reaction is exactly the same as that of the lead iodide. The coarser structure of the bands is due to a smaller value of P/L , and allows the precipitant to diffuse through more quickly than in the case of silver bichromate. This has the same effect as if the diffusion gradient of the solute in the gel were less. The precipitant enters a region where the resulting value of P/L is very small, and comparatively few crystallisation

¹ Cases of sol formation which have been observed have always occurred at the end of the reaction, after the solvent has been almost completely extracted from the gel. The appearance is then merely of a diffuse coloration without any indication of stratification.

centres are produced. The excess of precipitant, flowing on, meets with constantly increasing quantities of solute, N grows proportionally, until the precipitated particles appear to form a band, and as their number increases, so their size diminishes. This corresponds exactly to the microscopic appearances, and may be taken as a further direct proof of the existence of a concentration gradient. The isolated particles appear simultaneously with the ring, because it is not until they have reached a certain size that they become separately visible. Actually they may be formed somewhat earlier. They grow by adsorption of nutrient material from the adjacent gel, until this is exhausted, and thickening of the ring cuts off further supplies. By this time the region of the new band has increased in concentration of both the inflowing reagents, resulting, at first, in a denser precipitate and a quicker rate of adsorption and precipitation, until, in consequence of the adsorption, the concentration of the adjacent gel falls once more.

One other point must be mentioned in conclusion. It has been insisted that stratification is a rare phenomenon limited to a few insoluble precipitates. This is not in accordance with the writer's experience. Although experiments have been directed to the examination of the conditions underlying zone formation in a limited number of cases, rather than to the multiplication of specimens, successful attempts have been made to prepare stratified deposits of those substances such as silver chloride, barium sulphate, copper phosphate in agar, and lead iodide in gelatin which have been specifically mentioned as refusing to stratify. No serious attempt to obtain a sufficiently insoluble precipitate in the stratified form has failed, except in the case of aluminium hydroxide in agar in U-tubes, and this was to be expected on account of its extreme insolubility. There appears to be no evidence to show that stratification will not occur provided a precipitate can be obtained in a suitable state of subdivision from solutions of a sufficient dilution: or that the production of such precipitate is other than a question of the influence of the various known physical factors. As opportunity offers, it is hoped, by increasing or diminishing the number of crystallisation centres in various ways, to prepare well stratified precipitates of substances in gels in which they are not ordinarily so obtained.

REFERENCES.

- Bradford (1916), *Biochem. J.* 10, 169.
Dreaper (1914), *Kolloid Zeitsch.* 14, 163.
Hatschek (1914), *Kolloid Zeitsch.* 14, 115.
Liesegang (1913), *Geologische Diffusionen*, p. 89.
v. Weimarn (1914), *Zur Lehre von den Zuständen der Materie*.