

SILVER ION CONCENTRATION STUDIES OF COLLOIDAL SILVER GERMICIDES.*¹

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There has been in the past much uncertainty as to the actual composition of the colloidal silver germicides. Some have thought that compounds of the solar-gentum type were colloidal metallic silver, others that they were colloidal silver oxide; some have thought that compounds of the Protargentum type were colloidal silver oxide, others that they were silver-protein compounds. Recently Pilcher and Sollmann² have published work on the inhibiting effect of various silver germicides on yeast fermentation and have drawn from their results certain conclusions regarding the silver ion content of these materials which seemed to us questionable.

In an effort to settle some of these vexed questions we decided to attack the problem by actually measuring by an electrometric method the silver ion concentrations given by solutions of these materials, which measurements might also give some clues as to the actual constitution of these materials. But this measurement told us only part of what we wished to know; we desired to know also how much of the silver was ionizable to this extent. Just as one can have weakly acidic solutions containing a small amount of a strong acid, or a large amount of a weak acid, so here it would have been impossible to distinguish by this method between a small amount of a highly ionized silver compound, or a large amount of a slightly ionized one. In order to do this we titrated the compounds with a material which would throw silver into an extremely ionized form, and followed the changes in silver ion concentration by our electrometric method. In this titration, the most highly ionized silver reacted first, so that our titrations showed how much of the silver was ionizable at each silver ion concentration level.

As titrating agents we used potassium and hydrogen iodides. For comparison, we also ran titrations with sodium and hydrogen chlorides, chlorides being frequently present in actual use of these materials. The materials titrated were silver nitrate, silver oxide, a compound of the Protargin Strong type (Protargentum), a compound of the Protargin Mild type (Solargentum), a compound of the Collargol type (Collargol) and a colloidal silver iodide (laboratory preparation).

In expressing our results we have found it convenient to use the symbol p_{Ag} , meaning the negative logarithm of the silver ion concentration. Thus, if a solution

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¹ In December, 1923, K. v. Neergaard published (*Arch. f. Exper. Pathol. u. Pharmakol.*, 100, p. 162) an article which we were unable to get hold of until after the presentation of this paper. He gave curves for potentiometric titrations of AgNO₃, Septacrol, Albargin, Protargol, Syrgol, Hegenon, Choleval, Elektrargol, Argentamin, and Collargol (N/500 with respect to total Ag in all cases) with N/10 KCl. On account of the difference in concentrations and materials used, it is impossible to compare our results directly with his. The use of KCl instead of HI or KI resulted in his not recognizing any ionizable silver in Collargol, which we found.

We are unable to agree that this method shows a difference between silver in true solution and in colloidal form. It only shows the amount of silver ionizable to any particular degree. Silver may be both colloidal and somewhat ionizable or in true solution (as a complex ion or compound) and much less ionizable.—Authors.

² *J. Lab. and Clin. Med.*, 8, 301 (1923); 9, 256 (1924); 10, 38 (1924).

is $N/10000$ with respect to silver ions, its p_{Ag} is 4. It will be recognized that p_{Ag} has the same meaning as regards silver ions as has p_H in regard to hydrogen ions.

Our results show that Solargentum contains from 11 to 14% of its silver in a form more highly ionized than AgI in the presence of excess soluble iodide. Its p_{Ag} is 6.7 to 7.1. Collargol has an initial p_{Ag} of 6.5, and 10% of its silver is more ionized than silver iodide. The remainder of the silver in these compounds is probably metallic silver. The ionizable silver is probably combined with protein material in some insoluble or slightly ionized form.

Protargentum has a p_{Ag} of from 2.1 to 2.4, and has 81% of its silver more ionizable than silver iodide. It is probably a mixture of colloidal silver oxide with silver protein compounds.

While no exhaustive search of the literature has been made, the authors believe that this is the first time that such ion concentration studies have been made except in the case of hydrogen ions. They desire to point out that the method is a perfectly general one. In making titration curves of acids (or bases), a titration agent is used which will throw the hydrogen (or hydroxyl) ions into a very slightly dissociated form, namely, water. The same principle can be used with any other ion. All that is needed to make a titration curve for an ion is a method of measuring the concentration of the ion, and a titrating agent which will throw the ion into a very slightly dissociated form.

General Part.

Discussion of Method.—The general method was to make up 100 cc. of a solution which was tenth-normal with respect to the silver content of the compound, to titrate this with $N/10$ HI, KI, HCl or NaCl and to follow the titration by means of the potential of a silver electrode in the solution.

It would be possible to determine this potential by immersing a silver electrode in the solution and another in a silver solution of known concentration, but in practice it is much simpler and more accurate to use a saturated KCl-calomel half-cell as the reference electrode, as we did. When this is done the silver electrode in all solutions having an Ag ion concentration greater than 3.2×10^{-8} will be positive to the mercury of the calomel half-cell and the connection of the calomel half-cell to a potentiometer will be to the negative instead of to the positive terminal. When during the course of the titration the silver ion concentration drops below 3.2×10^{-8} then the leads from the concentration chain are interchanged. When these results were plotted the voltage readings corresponding to a high silver ion concentration were plotted as plus, so that the p_{Ag} scale would lie in the right direction.

In order to calculate p_{Ag} from the potential measurements obtained by the above method, it is necessary to know the potential difference between a silver electrode in a solution which is normal with respect to silver ions and a saturated KCl-calomel half cell. A somewhat incomplete search of the literature yielded no reference to the measurement of such a value, but it can however be calculated with sufficient accuracy from the potential of a silver electrode in $N/10$ AgNO₃ against a saturated KCl-calomel half cell. As will be seen later this potential is 0.497 volt as measured with the hook-up described above. The constant can be calculated as follows:

According to Nernst the relation of the ion concentration and the electromotive force is given by the equation:

$$E = E_0 + \frac{RT}{nF} \log_e C,$$

- where E = electrode potential corresponding to ionic concentration C
 E_0 = electrolytic potential (electrode potential at molar concentration of the ion)
 R = the gas constant (8.32 joules per degree)
 T = absolute temperature ($273^\circ + \text{temperature } C$)
 n = valency of the ion
 F = the faraday (96,500 coulombs)
 C = ionic concentration of the solution.

This formula applies to an electrode immersed in a solution containing ions of the element of which the electrode is composed.

Substituting in the Nernst formula numerical values wherever possible, and using 25° as our working temperature, we have:

$$E = E_0 + .0591 \log C$$

The ionization of $N/10$ AgNO_3 as determined by the conductivity method is 81% and the actual silver ion concentration will therefore be 0.081N.

$$\begin{aligned} E &= E_0 + .0591 \log .081 \\ &= E_0 + .0591 (8.91 - 10) \\ &= E_0 + .0591 (-1.09) \\ &= E_0 - .064 \\ E \text{ as measured for } N/10 \text{ AgNO}_3 &= 0.497 \\ 0.497 &= E_0 - .064 \\ E_0 &= 0.497 + .064 = .561 \end{aligned}$$

The p_{Ag} of a solution at 25° will therefore be given by the formula:

$$p_{\text{Ag}} = \frac{0.561 - E}{.0591}$$

During recent years other methods besides the conductivity and transference number methods have been developed for determining the actual ion concentration of the solutions and these tend to show that both the above methods give results which are too low. They indicate that even in normal solutions the ionization is almost 100%. If this be true and $N/10$ AgNO_3 is completely ionized, it is easy to show that our assumption, that it was only 81% ionized, can introduce an error of only 0.09 of a p_{Ag} unit. At the present stage of this work such an error is not serious and if it does exist may be legitimately neglected.

The following titrations were made:

- $N/10$ AgNO_3 titrated by $N/10$ HI, KI, HCl and NaCl.
- Silver Oxide by HI.
- $N/10$ Protargentum by $N/10$ HI, KI, HCl and NaCl.
- $N/10$ Solargentum by $N/10$ HI, KI, HCl and NaCl.
- $N/10$ Collargol by $N/10$ HI.
- $N/10$ Colloidal AgI (laboratory preparation) by $N/10$ HI.

None of the solutions used for titrating were exactly $N/10$, but all volumes were calculated to the exact $N/10$ basis before plotting the curves. The solutions of

the silver preparations were $N/10$ with respect to total Ag. In the case of Sol-argentum and Protargentum analyses were available; in the case of Collargol the published analysis was used to determine the amount to be used. Any slight variation in the actual material from the assumed composition would only have the effect of spreading the curve more to the right, or the reverse.

It was impossible to plot on the curves all the points at which readings were taken. But the points omitted in all cases lay as close to the curve as those which were plotted. Many of the titrations were run in duplicate. In these cases only one experiment has been plotted, as the duplicates in all cases agreed well with each other.

Silver Nitrate.—The p_{Ag} of $N/10$ $AgNO_3$ is 1.08 as calculated on a basis of 81% ionization. When it is titrated with KI (Curve "A"), the curve has the same general form as that of the titration of a strong acid by a strong base. The mid-point of the break falls at a p_{Ag} of 7.95. This checks fairly well with the value 7.76, calculated from the solubility of AgI given by Seidell¹ as 0.000 0028 Gm. per liter. If HI is used instead of KI, the same curve is obtained. With HCl (Curve "K") the same type of curve is obtained, but the break does not extend so low, on account of the much greater solubility of AgCl. The mid-point of the break gives a p_{Ag} of 4.9 for pure saturated AgCl solution, which checks well with the value 4.72, calculated from the solubility given by Seidell,² 0.0020 Gm. per liter.

Silver Oxide.—Silver oxide was titrated only with $N/10$ HI (Curve "B"). An attempt was made to titrate the silver oxide with $N/10$ KI, but the rate at which equilibrium was established was so slow that it was not practical to make this titration. When silver oxide is titrated with $N/10$ HI equilibrium is rapidly obtained and a smooth curve results. The silver oxide as prepared evidently contained traces of sodium hydroxide, which slightly depressed the solubility. Varying quantities of nitric acid were added to small samples before titration until the ionization level of pure silver oxide was pretty definitely established. A titration was then made on a large sample of silver oxide to which just enough nitric acid had been added to bring it slightly below this ionization level. As the HI was added the potential slowly rose during the first 25% of the titration to a voltage of 0.393, which corresponds to a p_{Ag} of 2.85. This flat portion of the curve extended to 92 cc. where it started to break sharply. The main part of the break was very sharp.

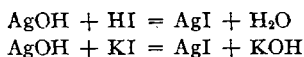
Protargentum.—This was titrated with $N/10$ HI (Curve "C"), $N/10$ KI (Curve "E"), $N/10$ HCl (Curve "L"), and $N/10$ NaCl (Curve "D").

The curves with HI and HCl are practically identical, except that the final ionization levels are different because of the differences in solubility of the iodide and the chloride. The curves of the titration with KI and NaCl are similar in a like respect. Protargentum when titrated did not always give the same original p_{Ag} , the variation being from 2.17 to 2.81. When Protargentum was titrated with HI or HCl the slope of the first 65% of the curve was not great, then the break started. The sharp part of the break started when 75% of the silver had been titrated and this part of the break ended at a point corresponding to 87% of the silver content. The end-point for ionizable silver as determined from the curve was 81.5%. When

¹ "Solubilities of Inorganic and Organic Compounds," 2nd Ed.

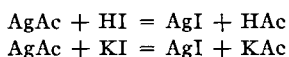
Protargentum was titrated with KI or NaCl the curve was of a different character. Here a definite slope started almost from the first and ended at a point corresponding to 77% of the silver content, giving a p_{Ag} at this point of about 8.0. From this point on the curves followed those of HI and HCl.

This difference in the character of the curves obtained in titrating Protargentum with acids and salts having ions which form insoluble silver salts indicates that the ions formed during the course of the titration affect ionization or solubility of the untitrated silver. If the ionizable silver be present in the form of silver oxide the reactions when titrating with an acid and a salt will be as follows:



in the first reaction the hydroxyl ion is used to form water and since this is practically undissociated it will have no effect upon the solubility of the Ag_2O which can only go into solution as $AgOH$. In the second reaction, however, KOH is formed and since in the dilutions used this will be more than 90% dissociated, then by the law of mass action there must be a steady drop in the $AgOH$ ionization as the titration proceeds.

It may be assumed, however, that the ionizable silver is present in the form of silver salts of the amino acids present as a result of the hydrolysis of the gelatine. With this assumption in mind the following equations may be written, in which Ac is used to indicate the acid ion of a silver amino acid complex.



In this case the effect on the ionization of the untitrated $AgAc$ will be, to all intents, the same as where the ionizable silver was assumed to be in the form of Ag_2O . The HAc formed as per the first equation will in this case be an extremely weak acid and will therefore not repress the ionization of the $AgAc$. The KAc formed as per the second equation is, however, the alkali salt of a weak acid. Such salts are known to be highly dissociated and the ionization of the untitrated $AgAc$ will therefore be more and more repressed as the titration proceeds. The fact that the effect on the ionization of the untitrated $AgAc$ or Ag_2O is the same when titrated respectively with KI or HI makes it impossible to tell definitely by this method the exact constitution of Protargentum.

The fact that the p_{Ag} of saturated silver oxide and that of Protargentum lay very close together, made us think at first that Protargentum was colloidal silver oxide. But the characters of the titration curves with HI are quite different. That of Ag_2O is typical of a rather insoluble pure compound, while that of Protargentum resembles the titration of a buffer mixture. We therefore believe that Protargentum is mainly a mixture of silver compounds of hydrolyzed protein substances, perhaps mixed with some silver oxide. A p_H determination should show whether this is present, but we have not yet succeeded in making such a determination. A hydrogen electrode gave about the same voltage as had the silver electrode, probably due to precipitation of silver on the electrode. The color of the solution makes the direct indicator method inapplicable.

The results of a dialysis experiment rather confirm this idea. The first three diffusates contained about one-quarter of the total silver, an amount corresponding

quite closely to a saturated solution of silver oxide. But the next three contained only half as much silver as the first three. This would indicate that the bulk of the silver was not present as the oxide, but probably in some more insoluble form. Twenty per cent. of the silver is practically unionized and is probably in the form of colloidal metallic silver.

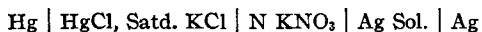
Solargentum.—This was titrated with $N/10$ HCl (Curve "M"), $N/10$ NaCl (Curve "F"), $N/10$ KI (Curve "E"), and $N/10$ HI (Curve "H"). The addition of the first cc. of $N/10$ HI or KI causes a rapid drop in the silver ion concentration. From here on the drop in ion concentration is rapid until a point corresponding to about 11% of the total silver content is reached. Here the curve breaks off rapidly into the ionization level of silver iodide. This indicates that only about 11% of the silver in Solargentum is present in a form which will ionize to a p_{Ag} of 12.5 or less. The rest of the silver is probably colloidal metallic silver. With NaCl, the curve shows only the mass action of the excess of Cl ions. The original p_{Ag} is less than that of pure, saturated AgCl, but greater than that of AgCl in the presence of an excess of Cl ions, hence this effect. The curve with HCl is so irregular as to be meaningless. We do not know why this is so.

When the ionization level of Solargentum is considered, it appears probable that the ionizable silver of Solargentum is in the form of a silver amino acid complex. We have no definite proof that this is the case, but such compounds are known to form and from the method of manufacture, it seems probable that they could exist in Solargentum. The ionization level of Solargentum does not correspond to any simple compound which could possibly be present.

Colloidal Silver Iodide.—This was prepared by treating $AgNO_3$ with a solution of I in KOH in the presence of hydrolyzed gelatine, dialyzing, and evaporating. Its titration with $N/10$ HI (Curve "J") showed the presence of some soluble iodide. Its initial p_{Ag} was 12.4.

Experimental Part.

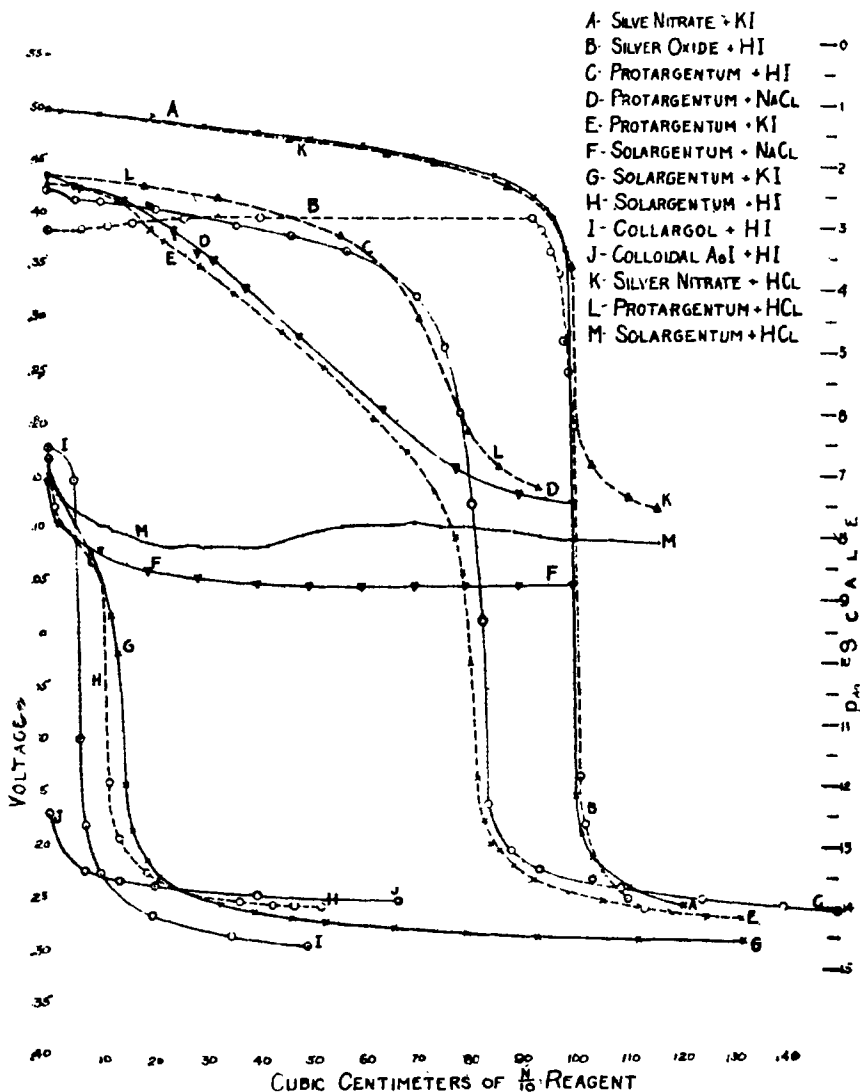
The concentration chain used in the work was made up as follows:



The potential measurements were made with a Leeds & Northrup portable potentiometer at room temperature. No attempt was made to carry out the work under constant temperature conditions. The temperature of the solutions ranged from 20° to 30° during the time the work was being carried out, but was generally not more than three degrees removed from 25° C. As was noted before, electrodes in all solutions having a silver ion concentration greater than 3.2×10^{-8} will be positive to the mercury of the calomel half cell, and in solutions below this concentration will be negative. When the silver ion concentration is greater than 3.2×10^{-8} the calomel half cell is connected to the negative terminal, and the silver electrode to the positive terminal of the potentiometer; and when the silver ion concentration drops below this value the connections must be reversed.

The silver electrodes were prepared by soldering pieces of silver wire to leads of copper wire, and cementing these into glass tubes with de Khotinsky cement so that only silver was exposed to the solution. They were then electroplated with a

platinum anode, using 1.5 volts and a $N/100$ solution of $AgNO_3$ containing about three drops of nitric acid in 50 cc. This did not bring them to equilibrium, so they were scraped and made the anode of a cell for about half an hour. They were then replated as before, using fresh solution. After washing, they were in equilibrium. They were kept short-circuited in the plating solution, and tested before use for equilibrium. At no time did they show a potential difference of over 1 millivolt.



The materials were prepared for titration, in all cases except $AgNO_3$ and Ag_2O , by weighing directly into a beaker the correct amount of material to give 100 cc. of $N/10$ silver and adding 95 cc. of water. The titrations were carried out by adding one or more cc. of reagent at a time and determining the potential of the above concentration chain after each addition. In almost all cases mechanical stirring was used to insure constant equilibrium. In the case of silver nitrate a mea-

sured volume of standard solution was used. In the case of silver oxide a suspension of an unknown quantity of Ag_2O was titrated; at the end of the titration the amount was calculated from the titration curve. With the silver oxide as prepared the titration curve rose slowly till the break came, indicating the presence in the solution of alkali, which repressed the ionization of the silver oxide. By adding varying amounts of nitric acid a curve was finally obtained which was flat until it broke. This showed the absence of excess of either alkali or silver salts, and gave the true p_{Ag} of Ag_2O . For making the titration which has been plotted, HNO_3 was added to the Ag_2O suspension till the p_{Ag} was just below the correct value of Ag_2O .

None of the solutions used were exactly tenth-normal. They were standardized by titrating against a known solution of AgNO_3 , in the same manner in which all the other titrations were carried out. The results of these titrations were plotted and the end-points read from the curves. The silver nitrate was standardized by precipitating with a slight excess of $N/10$ HCl and weighing the AgCl on a Gooch crucible. After standardization of the solutions the results of the titrations were calculated to a tenth-normal basis by multiplying the reading in cc. by the tenth-normal factor of the solution.

Stable readings on the potentiometer were obtained with no difficulty in all cases, except when the reading of a point on the break of a curve was being made. Here there is no buffer effect, and large voltage changes occur with only small additions of reagent, so complete stability could not be expected.

The original silver ion concentration exhibited by Solargentum seemed to be greatly influenced by the character of the water used in making up the solutions. It was always noticed that redistilled CO_2 -free water gave the highest and most regular initial silver ion concentrations.

ABSTRACT OF DISCUSSION.

H. C. Wood: Referring to the curves, I would like to ask what the proportion of total silver in solargentum, for example, is as compared to protargentum? There are evidently three distinct types of curves for these colloidal silvers: one represented by protargentum, the second by solargentum and the third by the colloidal iodide. What are the relative proportion of total silver in these compounds?

I would also like to ask if I correctly understood Dr. Giesy's point that in the protargentum the silver was probably chiefly in the form of a proteinate, whereas in the solargentum it was probably chiefly in the form of metallic silver, but neither of them contain appreciable quantities of peroxide or oxide.

I. M. Kolthoff: Dr. Giesy will agree with me that the difference between the ionogenic silver values in the second column gives an indication about the preferability of these preparations. "Silver that is more ionogenic than silver chloride" gives us an indication of the total silver content and the silver ion concentration. The figures mentioned in the second column give us an indication about the silver ion concentration itself, so the difference between those two gives an indication of the buffer value of the silver ions, and also one can judge about the valuation of these preparations as remedies.

Do you think that the silver which is more ionogenic than silver iodide and less than silver chloride is present as silver chloride? You don't mention that, but I suggest that the difference between the figures in the third and fourth columns is due to the presence of silver chloride.

In Germany very extensive study has been given to this work, not yet published, by Theodore Paul. He told me two months ago when in Germany attending a meeting, that he had given a very extended study to this subject that you have treated, so you can see it is of international importance.

Lyman F. Kebler: Dr. Woods Hutchinson in his book states that the average human being generates in the course of twenty-four hours about two quarts of alcohol. Of course, he said there were experiments under way to show that. However, he gives the impression in his book that this actually occurs, and that is used by some of the people in connection with propaganda I come in contact with.

The question with me is whether these silver preparations will actually inhibit the action of the cells in the stomach. We have these cells in the stomach and they are used every day. If this actually inhibits the growth of these cells, it might be objectionable to some people, while not to others.

P. M. Giesy: Taking up first Dr. Wood's question, I think we can state definitely that compounds of the solargentum and collargol types contain no silver oxide. The ionization of silver oxide you will note is about p_{Ag} 3. None of these compounds ionize more than p_{Ag} 6.5. That, I think, definitely excludes the possibility of silver oxide being present in those materials.

In the case of the protargentum type of compounds, I don't think we can be sure. The curves can be interpreted equally well by assuming that silver salts of weak organic acids are present there, and until we have actually made p_H measurements, we cannot say whether there is any silver oxide present in the protargentum type of compounds or not.

Solargentum contains in the neighborhood of 20 per cent. total silver; protargentum around 8 per cent.

Our solutions were made up so that they were always normal with respect to total silver, so that we used about two and one-half times as much protargentum for titration as solargentum.

Taking up Dr. Kolthoff's questions, I think that the silver more ionizable than silver chloride is a very good indication of the silver buffer effect of these preparations, but I can tell a whole lot more about that from looking at the curves than from any single numerical values. The flatness of the protargentum type of curve shows very well the buffer effect. Of course, there is a slight amount of buffer effect in the case of solargentum and still less in the case of collargol, but there is a little. I think if silver chloride were there we should get a flattening out of the curve at the silver chloride ionization level, that is, at about p_{Ag} 5. We don't get that at all, you see. The curves go on right past that point without any break at all, while you did actually obtain a break there.

I. M. Kolthoff: I carried out my experiments in acid solutions. I think you get it without any acid. I think that gives a great difference. I didn't give all of my results.

I have titrated in alkalized solutions and obtained silver oxide. I don't know how much, but they all had alkali. You get no great difference, but a far better result if you titrate in acid solutions. I think your results are obtained in the solutions as they are without adding anything.

P. M. Giesy: These are in the solutions without any addition at all. I can see this objection to a titration in acid solution. In the case of protargentum we probably have silver salts of weak organic acids. We add a strong acid, such as sulphuric acid, to that and it is going to raise the p_{Ag} , and we won't have exactly the same material that we would have in a neutral solution. And the presence of that acid is going to keep that p_{Ag} up right straight along, so that the whole curve will lie higher than the curves which we have obtained.

I. M. Kolthoff: My intention was not to show that you have, by titrating in acid solutions, a very easy method to determine all the silver present, but only to show how the silver ionization changes. I can also show you the curves I obtained and they are quite different. The silver concentration is increased by adding acid. It behaves as if there were present silver acid salts of a weak kind.

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