International Journal of Applied Radiation and Isotopes, 1967, Vol. 18, pp. 327–329. Pergamon Press Ltd. Printed in Northern Ireland

Determination of Gold Sol Concentrations from Spectrophotometric Data

(*Received* 19 *October* 1966)

Introduction

A MORE complete knowledge of the characteristics of the colloidal radioactive gold is needed in view of the growing use of this compound in the clinical research. The mean size of the particles, the dispersion degree and the number of particles per volume unit are between the more important parameters. The latter can be easily calculated from the value of the particle mean size and the gold concentration of the suspension.

CARO and INGRAND have recently reported[(1)] a simple spectrophotometric procedure for determining the particle mean size of the colloidal gold preparations. The procedure is routinely used in our laboratories with good results.

On the contrary, the conventional chemical determination of the gold concentration in gold sols is a slow procedure and, for this reason, it is unsuitable to the analysis of the radioactive preparations, due to the short half life of the Au^{198}.

In this note we study a procedure to determine the gold sols concentration by absorption spectrophotometry; the determination is based on the MIE theory[(2)] of the optical behaviour of gold sols with different particle sizes. By determining at 526 mμ the optical density of the colloidal suspension, we can estimate with an accuracy of approximately 10 per cent, the concentration of gold sols with particle diameters between 30 and 400 Å. These sizes are those of the commonly used radiogold colloids.

Principle of the method

The concentration of gold sols c can be calculated from the values of its optical density, (o.d.) = $\log (I_0/I)$, determined by spectrophotometry, using the following equation:

$$c = \frac{(\text{o.d.})}{0{\cdot}43Kl} \qquad (1)$$

where K is the coefficient of volume extinction and l the thickness of the solution. In this equation c is the ratio of total volume of the gold particles/volume of the suspension. If we wish to give the concentration C as μg of gold per ml of the colloidal suspension, the equation is:

$$C = \frac{4{\cdot}49 \times 10^7 \, (\text{o.d.})}{Kl} \qquad (2)$$

The coefficient of volume extinction K, for particles with a diameter smaller than 300 Å, is given by:

$$K = \frac{36\pi}{\lambda} n_0^3 \frac{nk}{(n^2 - k^2 + 2n_0^2)^2 + 4n^2k^2} \qquad (3)$$

λ, n_0 and n are respectively the wavelength of the incident light in the vacuum, the refractive index of the dispersing medium, the refractive parameter of the colloidal particle and k is its absorption parameter. n and k are a function of both the wavelength of the incident light and the particle size. ROEMER and FRAGSTEIN[(3)] have determined experimentally the n and k values for gold colloids with different particle diameters at several wavelengths. (The particle diameter can be obtained from the ratio of the optical densities measured at two different wavelengths.) The values for solid gold have been given by HAGENS and RUBENS[(4)] and by OTTER.[(5)]

If the K value is needed for sols with a particle diameter greater than 300 Å, it is necessary to introduce in the calculations the second and third terms of the Mie series.

In the present paper we have selected the wavelength of 526 mμ for which the n and k values are well known, as a convenient one for determining the colloid concentration. Shorter wavelength values are unsuitable, because, for these, the plasma effect described by SERAPHIN,[(6)] which has not been taken into account in equation 3, becomes much more important. As the experimental determination of the optical density at large wavelengths can be affected by light scattering in some spectrometers,

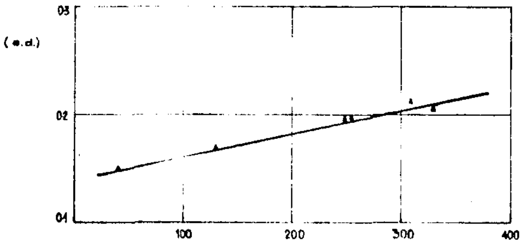

FIG. 1. Optical density (o.d.) at 526 mμ of a 10 μg/ml gold colloid as a function of the particle mean size.
——— Theoretical.
△ Experimental values

we have not used wavelengths larger than 550 mμ either.

By using the equations (2) and (3) and the n and k values for gold colloids, we have calculated (function I) the optical density at 526 mμ of a 10 μg/ml gold sol as a function of the particle diameter. A similar function (function II) was also calculated with the n and k values for solid gold by using the data given by JEPPESEN and BARLOW.$^{(7)}$ These authors have introduced into the calculations the second and third terms of the Mie series.

We expected that the first function (n and k for colloids) would give a better fit with the experimental values. However, the plasma effect, which is difficult to evaluate quantitatively, produces a rise of the actual optical density of gold sols with a particle size smaller than 150 Å. Therefore we have made a rough estimation of the contribution of the plasma effect together with an evaluation of the errors involved by adopting the equations with n and k for solid gold and those with n and k for colloids. We have in this way obtained a new function which, by counterbalancing the errors, agrees better with the experimental data. In this function (solid line in the Fig. 1) the values of the optical density were obtained by averaging, for each particle size, the values obtained by the function I and II.

On this basis we have prepared, in order to simplify the calculations, a nomograph for finding the concentration of the gold sols from its optical density (Fig. 2). The first column corresponds to the particle mean diameter in Å, the second to the concentrations in μg Au/ml and the third to the optical densities of the colloid at 526 mμ. Moreover, on the right side of the first column we have also plotted the corresponding values of the $(\text{o.d.})_{526\text{m}\mu}/(\text{o.d.})_{492\text{m}\mu}$ ratios. These ratios are used for calculating the mean particle diameter.$^{(1)}$

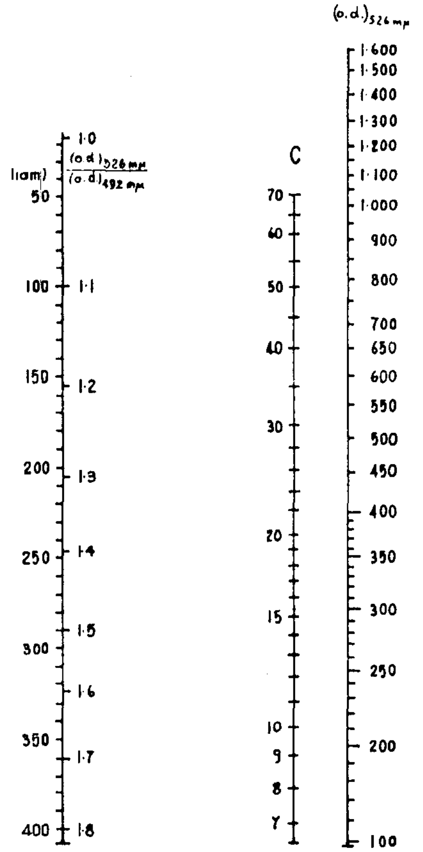

FIG. 2. Nomograph for calculating the concentration of gold sols. The diameter is given in Å, the concentration in μg Au/ml. The optical density is indicated by (o.d.).

Experimental

We have prepared gold colloids of different particle sizes, by the procedure given by Henry *et al.*,$^{(8)}$ from small gold wires irradiated in the reactor RA1. Their concentrations were determined radiometrically as follows. We have prepared, with equal specific activity gold, a solution of known content of chloroauric acid. We have then compared, with a well-type ionization chamber, the activity of this solution to the activity of the colloidal preparations.

At the same time, for checking the absence of ionic gold in the colloids, the preparations were analysed by paper chromatography with the solvent—acetone:water:conc hydrochloric acid (7:2:1). In all cases the ionic gold content was smaller than 0·5 per cent.

After the radiometric determination, the colloidal preparations were diluted with distilled water to a concentration between 10 and 70 μg Au/ml. The absorption spectra of the sols were determined against blanks of distilled water. In our case a distilled water blank, instead of a solution with the same composition as the dispersing medium of the colloid, does not produce serious errors.$^{(1)}$ All the preparations were analysed in several spectrophotometers: the optical density values obtained in the different instruments were in agreement within 2 per cent.

The mean particle size was calculated from the ratio of the optical densities at 526 mμ and 492 mμ. With this value, a straight line was plotted from the first column of the nomograph to the point corresponding to the optical density of the preparations at

526 mμ on the third column. The intersection of the straight line with the second column gives the concentration of the sol.

Results

The solid line in the Fig. 1 represents the calculated optical density of a 10 μg Au/ml sol of gold as a function of the particle mean size. On the same graph are plotted the experimental values of $\frac{(\text{o.d.})_{526\,\text{m}\mu}}{0{\cdot}1c}$ where c is the gold concentration of each preparation as obtained by radiometric analysis. We can observe that the experimental values agree with the theoretical ones within 10 per cent. The difference may be explained either by possible experimental errors—in the determination of the optical density and in the radiometric analysis—or by the intrinsic limitations of the theoretical treatment. These limitations have been discussed in the paper by CARO and INGRAND[1] on the determination of the particle size.

We think, however, that the simple and fast procedure we propose can be used as a routine method for determining the concentration of radiogold sols, since its accuracy is suitable to the accuracy required by most of the uses of the colloidal gold in the biological and clinical research.

R. A. CARO*
J. O. NICOLINI*†
R. RADICELLA*†

*Cátedra de Física
Facultad de Farmaciay Bioquímica
Universidad de Buenos Aires
Argentina
†Departamento de Producción
Comisión Nacional de Energía Atómica
Buenos Aires, Argentina

References

1. CARO R. A. and INGRAND J. *Rev. Opt.* **44,** 281 (1965).
2. MIE G. *Ann. Phys.* **25,** 377 (1908).
3. ROEMER H. and v. FRAGSTEIN C. *Z. Phys.* **163,** 27 (1961).
4. HAGENS E. and RUBENS H., *Ann. Phys.* **8,** 432 (1902).
5. OTTER W. *Z. Phys.* **161,** 163 (1961).
6. SERAPHIN B. *Ann. Phys.* **10,** 1 (1952).
7. JEPPESEN M. A. and BARLOW R. B. *J. Opt. soc. Am.* **52,** 99 (1962).
8. HENRY R., HERCZEG C. and FISHER C. *Int. J. appl. Radiat. Isotopes* **2,** 136 (1957).