Quantitative Estimation of Particulate Matter in Pharmaceutical Preparations Intended for Intravenous Administration

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Abstract □ A fast, reproducible, economical, and dependable automated counter method is recommended for the quality control of pharmaceutical preparations intended for intravenous administration. The USP gives no specifications on the limitations of particulate matter in intravenous products, while the BP specifies limitations on intravenous solutions of more than 500-ml volume. Out of 15 different marketed products, few passed the BP specifications. The microscopically identifiable particles included starch, cellulose fibers, glass, rubber, lacquer flakes, carbon black, and metal shavings. The proposed quality control method introduces a modification to the BP specifications. The method includes a standard log–log plot obtainable from a least-squares line fit to the data of the marketed products showing minimal particle contamination. The standard plot is compared to the experimental data obtained from any other product and, accordingly, it is decided whether the product is acceptable or not.

Keyphrases □ Particles—automated counter analysis, intravenous pharmaceutical preparations, proposed as quality control method □ Intravenous dosage forms—automated counter particle analysis proposed as quality control method □ Dosage forms—intravenous pharmaceutical preparations, automated counter particle analysis, proposed as quality control method □ Automated counter—particle analysis, intravenous pharmaceutical preparations, proposed as quality control method

The presence of particulate matter in parenteral preparations for intravenous administration gave rise to three important questions: the nature and origin of particulate matter (1–4), the hazards produced by these particles (5–12), and quality control of such products. Recent investigations (13–23) in this area have not covered quality control. The present work resulted in a quality control method for the analysis of particulate matter in any intravenous preparation.

EXPERIMENTAL

Sample Analysis—An automated counter[1] was employed. The test solutions were placed in carefully washed beakers to eliminate the possibility of contamination. The particulate matter concentrations in the tested products were small enough to allow the particles to pass through the aperture one at a time.

Calibration of Counter—According to a rough estimate of the particle-size distribution of the tested solution, a 30-, 50-, 70-, or 100-μm aperture tube opening was installed into the machine. Standard monosized calibration materials with a size range between 5 and 20% of the aperture diameter were used to estimate the calibration factor, K, calculated by:

$$K = \frac{D}{\sqrt[3]{t}} \qquad \text{(Eq. 1)}$$

where D is the particle diameter, and t is the threshold value setting on the machine.

Materials such as polyvinyl toluene latex (D = 1.87 μm), polystyrene divinyl benzene latex (D = 5.02 μm), and puff ball spores[1] (D = 3.36 μm) were used for calibration of the machine. The particles were dispersed in a particle-free normal saline solution (see *Preparation of Particle-Free Control Stocks*) in a beaker and placed on the counter stand for counting. The desired dilution of the calibration material was used such that at low threshold (t) settings, the machine was counting at a low coincidence factor.

Standard calibration curves were obtained from a plot of the number of particles oversize *versus* threshold. The threshold corresponding to 50% of the number of particles oversize was taken to be proportional to the volume of a sphere whose average diameter was that of the calibration material. The calibration factor, K, was then calculated from Eq. 1. During experimentation, the machine was occasionally recalibrated.

Preparation of Particle-Free Control Stocks—A particle-free stock solution for control experiments was prepared as follows. Nine grams of sodium chloride[2] was dissolved in 1000 ml of distilled water. A stainless filter holder[3] adapted with a 47-mm, 0.45-μm membrane filter[3] and attached to a vacuum flask was used for the filtration. This procedure resulted in the filtration of particulate matter whose diameter exceeded 0.45 μm. The filtrate was refiltered through a newly installed filter until it was almost particle free, as checked microscopically and by the automated counter.

Part of the freshly prepared particle-free stock was used to fill the reservoir of the machine to avoid leakage of particulate matter from the reservoir during flushing of the inner part of the aperture tube. Another part of the particle-free stock solution served to run the baseline control experiments for a particle-free solution. The control study was run in exactly the same manner as the study of the marketed products. The solution was poured into a clean beaker and then placed on the automated counter stand. Aluminum foil was placed around the stand to eliminate electrical noise pickup from the environment. The aperture tube and the outer electrode were then dipped into the solution, and the counts were taken at different thresholds. The counts obtained at specified thresholds from the control stock were considered as background counts and, therefore, were subtracted from the corresponding threshold counts when the marketed products were being counted for adulterated particles. During the experiments, the solution in the beaker was constantly stirred by a stirrer attached to the counter. The rest of the particle-free stock solution was used as a suspending fluid for the calibration material.

Testing Marketed Products for Particulate Matter—Three types of marketed products intended for intravenous administration were studied. First, intravenous infusion fluids packed in plastic bags and in glass containers with lacquered and nonlacquered rubber stoppers were chosen. Products containing an electrically conductive liquid were directly used in the counting process; those containing a nonconductive liquid, such as 5% dextrose solution, were diluted in a 1:1 ratio with 4% particle-free sodium chloride solution.

Second, plastic bags employed for blood collection in blood banks were selected. They contained anticoagulant citrate dextrose solution. Since the label stated that they should be made up to 500 ml with blood, they were diluted in a 1:4 ratio with 0.9% particle-free sodium chloride solution.

Third, powdered unexpired antibiotic products (some of them lyophilized), packed in glass containers with rubber-like stoppers, were randomly selected from the market. These products were dissolved in 250 ml of particle-free normal saline solution, exactly as recommended by the manufacturers.

All of the products were tested for particulate matter utilizing the automated counter and microscopy for possible particle identification. Each container was lightly shaken before opening to assure uniform distribution of particles. A sample of 200 ml was then placed into a very clean beaker for counting. The procedure followed was exactly the same as that described in the previous sections.

Flocculation Experiments—Flocculation experiments were done to study the particle-size distribution of flocculated and deflocculated systems and to correlate the results with those obtained using the mar-

[1] Coulter counter model A, Coulter Electronics, Hialeah, Fla.; puff ball spores were also obtained from Coulter Electronics.

[2] Sodium chloride crystals G.R., E. Merck, Darmstadt, W. Germany.

[3] Millipore Corp., Bedford, MA 01730.

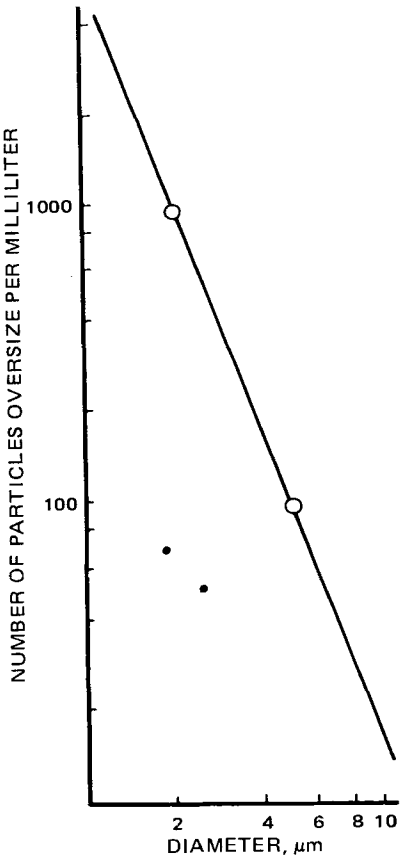

Figure 1—*Logarithmic plot of log (number of particles oversize per milliliter) versus log (diameter) for the extrapolated BP specification.*

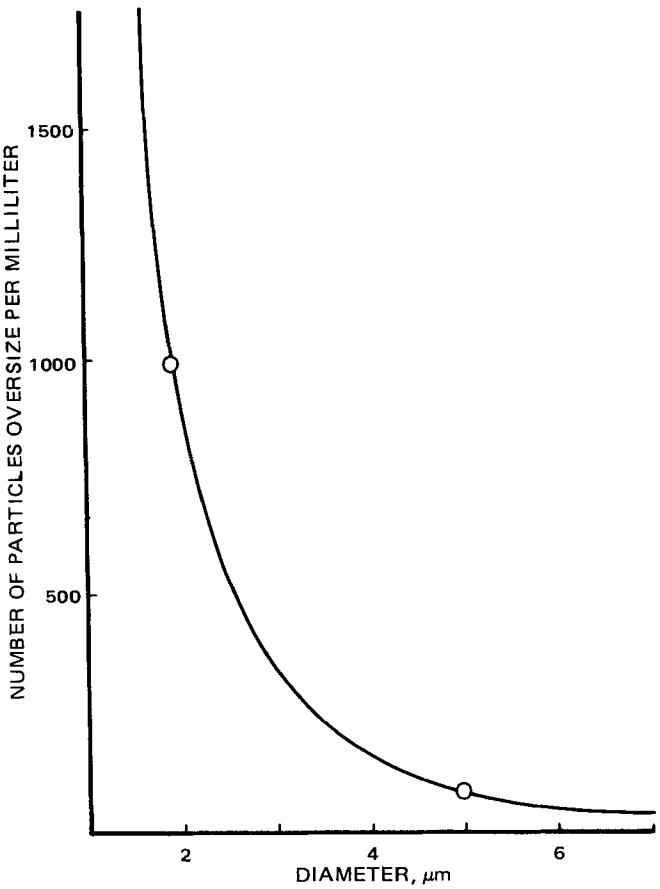

Figure 2—*Rectangular coordinate plot of log (number of particles oversize per milliliter) versus log (diameter) for the extrapolated BP specification.*

keted products. A 0.9% particle-free sodium chloride solution was prepared as described under *Preparation of Particle-Free Stock Solutions.* A small quantity of puff ball spores (D = 3.36 μm) was suspended in 100 ml of this solution with the aid of 2 ml of 1% aqueous dioctyl sodium sulfosuccinate[4] solution. This served as a nonflocculated suspension stock. Then 35 ml of this nonflocculated suspension stock was diluted to 250 ml with particle-free normal saline solution, resulting in a nonflocculated suspension sample for counting. To another 35-ml sample, 10 drops of saturated magnesium chloride[5] solution were added, providing the counterion effect for flocculation. The suspension was then made up to 250 ml with particle-free normal saline solution.

These solutions were gently shaken and left to sediment for a few days. At the time of counting, the suspension was shaken and counted by the automated counter.

Microscopic Identification of Particulate Matter—A 13-mm membrane filter[3] (0.45-μm pore size) adapted to a filter holder[3] was used for the collection of particulate matter. A 10-ml syringe was fitted to the filter holder, and the sample containing particulate matter was poured into the syringe. A vacuum flask attached with a rubber tube to the end of the filter holder was used for the filtration of the solution. The particulate matter was collected on the filter. A filter through which particle-free normal saline solution was filtered was used as a control. The filter was carefully removed from the filter holder, placed in a petri dish, dried in an oven, and then mounted on a slide.

To use transmitted polarized light, it is necessary to make the filter transparent by immersing it in a liquid of the same refractive index as that of the filter. Immersion oil[6] with a refractive index of 1.515 was used. Several drops were placed on a clean glass slide, and the filter was lowered onto the oil drops and allowed to soak up the oil. A scrupulously clean cover glass was placed over the filter. Subsequently, the mounted filters were systematically scanned with a polarizing microscope so that the entire 13-mm diameter filtration area was observed. Finally, a photomicrograph was taken for each sample, using a camera attached to a polarizing microscope[7].

Calculations and Treatment of Data—Groves (14) reported the following log–log relationship between the number of particles oversize *versus* diameter:

$$\log_{10}N = a + k \log_{10}D \qquad \text{(Eq. 2)}$$

Furthermore, the BP reported the following on the limit test of intravenous solutions of more than 500-ml volume: "The mean counts of particles per 1.0 ml do not exceed 1000 equal to or greater than 2 μm and 100 equal to or greater than 5 μm" (24). Application of Eq. 2 to the BP specification would result in a two-point plot on log–log graph paper, namely the points (2, 1000) and (5, 100). The equation of the straight line joining these two points was then deduced to be:

$$\log_{10}N = \log_{10} 5630 - 2.5 \log_{10}D \qquad \text{(Eq. 3)}$$

where N is the number of particles oversize, and D is the diameter. Figure 1 shows the log–log plot of the BP recommendation using Eq. 3. Also, Eq. 3 can be simplified into the following relationship:

$$N = \frac{5630}{D^{2.5}} = \frac{a}{D^b} \qquad \text{(Eq. 4)}$$

Equation 4 is an exponential (to the base D) type of relationship that would fit with any automated counter data when the value of a is 5630 and that of b is 2.5. A rectangular coordinate plot of Eq. 4 is shown in Fig. 2.

RESULTS

The results are presented as graphs. Particulate matter identification and measurement are presented in Table I, which was formulated from the photomicrographs taken of these samples. Because of lack of space, photomicrographs were not included.

[4] Aerosol OT.
[5] E. Merck, Darmstadt, W. Germany.
[6] Cargille type B, Bausch & Lomb Inc., Rochester, N.Y.
[7] Reichert Photo-Automatic, Wien Austria, A1171 Vienna, Austria.

Table I—Compilation of Some Identifiable Particles in the Marketed Products with Their Approximate Sizes[a]

Product[b]	Particles Identified and Size
1H	Rubber (2–100 μm), carbon black (1–5 μm), pigments (1–5 μm), calcium carbonate crystals (2 μm), cellulose fibers (up to 100 μm), glass (5–10 μm), starch grains (10 μm), *etc.*
1G	Carbon black (1–5 μm), starch grains (15 μm), unidentified particles (1–3 μm), *etc.*
1P	Rubber (5–40 μm), carbon black (1–5 μm), lacquer coating (5–100 μm), pigments (3 μm), calcium carbonate crystals (2 μm), *etc.*
1AL	Carbon black (1–5 μm), pigments (1–5 μm), calcium carbonate crystals (2 μm), lacquer flakes (5–50 μm), rubber (5–50 μm), starch grains (8 μm), metal shavings (5 μm), *etc.*
1S	Unidentified particles (1–2 μm), few particle contaminants
2AL	Carbon black (1–10 μm), starch grains (8–15 μm), lacquer flakes (20 μm), rubber (5–100 μm), unidentified particles (1–5 μm), *etc.*
2G	Carbon black (1–3 μm), unidentified particles (1–5 μm), *etc.*
3AL	Carbon black (1–5 μm), pigments (1–3 μm), rubber (5–50 μm), starch grains (15 μm), fibers (100 μm), *etc.*
3G	Unidentified particles (1–3 μm), few particle contaminants
4G	Unidentified particles (1–5 μm)
1E	Unidentified particles (1–5 μm)
1B	Carbon black (1–5 μm), starch grains (10–50 μm), fibers (100 μm), unidentified particles (1–5 μm), *etc.*
4AL	Starch grains (25 μm), carbon black (1–10 μm), fibers (up to 100 μm), unidentified particles (1–4 μm), *etc.*
1A	Unidentified particles (1–5 μm)
2A	Fibers (up to 120 μm), carbon black (1–10 μm), unidentified particles (1–5 μm), *etc.*

[a] Particles identified with help of Ref. 25. [b] For type of formulation, check Figs. 3–14. The letter H after a number signifies a product manufactured by a local hospital in Lebanon. The letter P designates a local Lebanese product, G designates a German product, S designates a Swiss product, B designates a Belgian product, and E designates an Egyptian product. The letter A signifies a United States product, and AL indicates a product manufactured by a United States company under license in a European or Middle Eastern country.

Cumulative Size Distribution in Solutions Intended for Intravenous Administration—Figures 3 and 4 are representative plots on rectangular and logarithmic coordinates for particulate matter in Products 1AL, 1P, and 1S. Similarly, Figs. 5 and 6 are representative rectangular and logarithmic plots for data obtained for Products 1H and 1G. In both plots, a profile was drawn of data obtained on particulate matter existing in the filtrate of Product 1H after filtration through a 0.45-μm membrane filter.

Figures 7 and 8 represent data for Products 2AL and 2G, while Figs. 9 and 10 represent data for anticoagulant citrate dextrose solutions of Products 4G and 1E. Figures 3–10 include a comparison between the results of the marketed products and the smooth curves obtained from the extrapolated BP specifications as already described.

Cumulative Size Distribution in Powdered Antibiotic Products Intended for Intravenous Administration—The dilutions of these products were already discussed. Figures 11 and 12 represent rectangular and logarithmic plots for particulate matter in Products 1B and 4AL containing ampicillin sodium. Figures 13 and 14 are similar plots for Products 1A and 2A for cephapirin sodium and cephalothin sodium, respectively. The smooth curves appearing in Figs. 11–14 are the extrapolated BP limit test plots. The BP does not mention any limit test for the quality control of adulterated particles in antibiotic products.

Cumulative Size Distribution in Deflocculated and Flocculated Puff Ball Spores—Figures 15 and 16 represent typical rectangular and logarithmic plots on deflocculated and flocculated systems. The plots show the monosized distribution in the deflocculated systems and the deviation from monosized distribution in flocculated systems. Similar deviations resulted in the data obtained for some marketed products as shown in Figs. 5–12 and 14.

Microscopic Identification of Particulate Matter in Marketed Products—Table I represents a compilation of most of the identifiable particles, with their approximate sizes, in the tested products.

DISCUSSION

A thorough literature survey revealed that previous findings concerning quantifying and identifying particulate matter in intravenous solutions did not result in substantial recommendations for quality control. There appeared to be agreement that particulate matter in intravenous products was hazardous to patients and that the pharmacopeias should include specific and strict recommendations for its limitations.

Proposed Analysis of Results on Basis of BP Specifications—The advantages of this method of analysis based on the extrapolated BP specifications, as described under *Experimental*, are that it is fast, reproducible, economical, and dependable for the quality control of pharmaceutical preparations intended for intravenous administration. As previously reported, the particle-size distribution of the particulate matter in an intravenous infusion under storage may change with time due to particle aggregation or deaggregation. The BP has given only two specific particle numbers with their corresponding sizes, with no mention of possible aggregation or deaggregation.

The proposed analysis, based on the extrapolated BP limitations, gives a whole range of size distributions, permitting quality control personnel to check possible particle flocculation or deflocculation instantaneously. This check is in addition to a general analysis of the particle-size distribution in the products. A simple comparison analysis was needed. To support this method, Figs. 15 and 16 showed the general trend of the curve for flocculated and deflocculated systems. Any deviation from this curve due to flocculation would result from a change in the particle-size distribution. Therefore, in an intravenous product, any behavior similar to that of these plots could be explained on the basis of particle aggregation or deaggregation.

Recommendations—Some drug companies have been able to produce products for intravenous administration in which the particulate matter is far below BP limitations. Some marketed products, especially the powdered antibiotic products, far exceed the proposed limitations. The comparison of the results obtained in this work with the BP specifications for powdered antibiotic and blood bank bag products was done to show the importance of limitations for the quality control of these pharmaceutical preparations.

Recommendation I—The following model is proposed for all products manufactured for intravenous administration. This model is very arbitrary and flexible and is not mentioned as a replacement for any limitations proposed by a pharmacopeia. The emphasis is directed toward a similar applicability in any quality control laboratory.

Since the particle-size analysis of some marketed products showed low particle contamination and since any limit test is arbitrary, the best products (1G, 1S, and 3G) were used to set out the proposed limit test. The number of particles oversize per 1.0 ml *versus* diameter was plotted on log–log paper (Fig. 17). The best straight line through all points was drawn utilizing the method of least squares (26). The following is the equation of the straight line obtained as a result of the least-squares line method:

$$\ln N = \ln 225 - 1.80 \ln D \qquad \text{(Eq. 5)}$$

where N is the number of particles oversize per 1.0 ml, -1.8 is the slope of the line, D is the particle diameter, and 225 is the intercept when $D = 1$ μm. Equation 5 can be reduced to:

$$N = \frac{225}{D^{1.80}} \qquad \text{(Eq. 6)}$$

A plot of N *versus* D of Eq. 5 on rectangular coordinates is shown in Fig. 18.

The best least-squares line fit shown in Fig. 17 was used in this work as the basis for the proposed limitations. However, some products had particle contaminants (as seen in Fig. 17) that did not pass the specifications. To take this into account, it was suggested that a line be drawn parallel to the least-squares line and that products (1S, 1G, and 3G) fitting this line be considered acceptable. This procedure also took into account flocculated systems where the size distribution caused a change in the linearity of the plot (Fig. 16). The straight line passed through the points (1, 700), (2, 200), (3, 100), and (5, 40). The following is the equation of the straight line passing through these points:

$$\ln N = \ln 700 - 1.80 \ln D \qquad \text{(Eq. 7)}$$

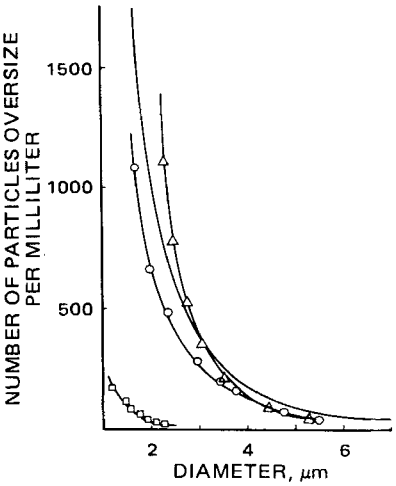

Figure 3—*Cumulative particle-size distribution in 0.9% sodium chloride solutions for injection. Key: △, Product 1AL packed in 500-ml glass containers with lacquer-coated rubber stopper; ○, Product 1P packed in 1-liter glass containers with lacquer-coated rubber stopper; □, Product 1S packed in 500-ml plastic bags; and smooth curve, BP specification.*

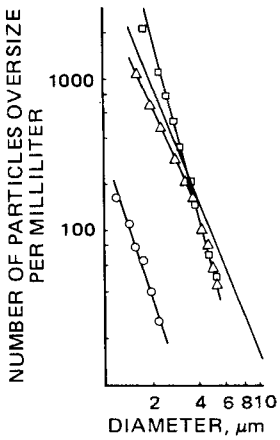

Figure 4—*Logarithmic plot of cumulative particle-size distribution in 0.9% sodium chloride solutions for injection. Key: □, Product 1AL; △, Product 1P; ○, Product 1S; and smooth curve, BP specification.*

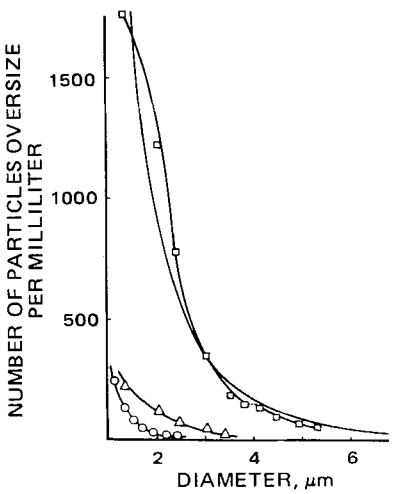

Figure 5—*Cumulative particle-size distribution in 0.9% sodium chloride solutions for injection. Key: □, Product 1H packed in 1-liter glass containers with rubber stopper; △, Product 1H after filtration through a 0.45-μm filter; ○, Product 1G packed in 500-ml plastic bags; and smooth curve, BP specification.*

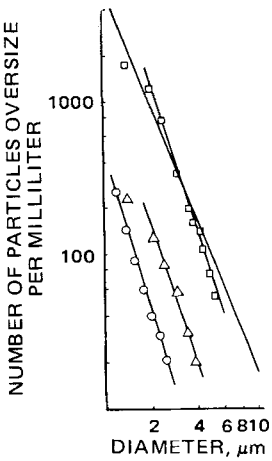

Figure 6—*Logarithmic plot of cumulative particle-size distribution in 0.9% sodium chloride solutions for injection. Key: □, Product 1H; △, Product 1H after filtration; ○, Product 1G; and smooth curve, BP specification.*

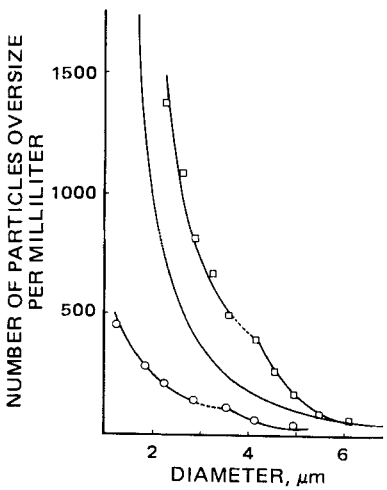

Figure 7—*Cumulative particle-size distribution in 5% dextrose solutions for injection. Key: □, Product 2AL packed in 500-ml glass containers with lacquer-coated rubber stopper; ○, Product 2G packed in 500-ml plastic bags; and smooth curve, BP specification.*

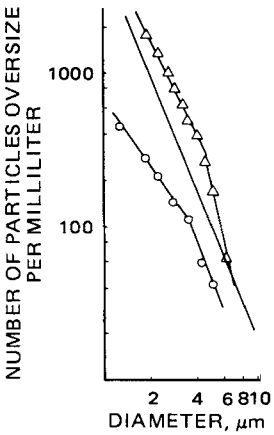

Figure 8—*Logarithmic plot of cumulative particle-size distribution in 5% dextrose solutions for injection. Key: △, Product 2AL; ○, Product 2G; and smooth curve, BP specification.*

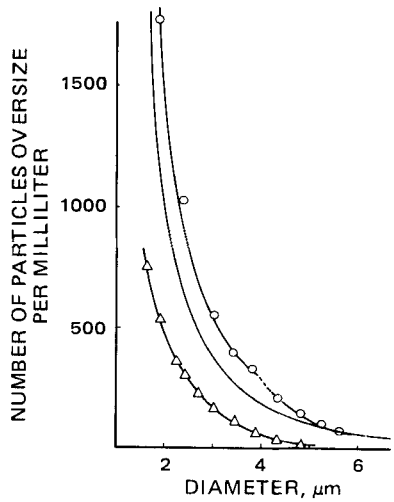

Figure 9—*Cumulative particle-size distribution in anticoagulant citrate dextrose solutions intended for blood collection. Key: ○, Product 4G packed in 500-ml plastic bags; △, Product 1E packed in 500-ml plastic bags; and smooth curve, BP specification.*

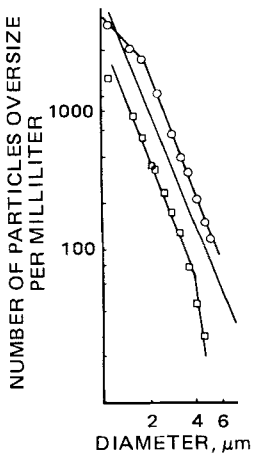

Figure 10—*Logarithmic plot of cumulative particle-size distribution in anticoagulant citrate dextrose solutions intended for blood collection. Key: ○, Product 4G; □, Product 1E; and smooth curve, BP specification.*

Therefore, the proposed limitations should be: "For any product intended for intravenous administration, regardless of its volume, the average counts per 1.0 ml of five to 10 samples should not be more than 700 equal to or greater than 1 μm, 200 equal to or greater than 2 μm, 100 equal to or greater than 3 μm, and 40 equal to or greater than 5 μm."

The plots in Figs. 17 and 18 can be used as standard curves for the quality control of any product intended for intravenous administration, utilizing for counting an electronic counting device that can count the cumulative number of particles in a solution. Five to 10 products from the production line could be randomly picked up and sent to the quality control laboratory. By using a precalibrated counting device with an aperture tube of 30, 50, or 70 μm and counting the number of particles oversize *versus* particle diameter, a similar plot could be drawn. Comparison of the standard plots (Figs. 17 and 18) to the experimentally obtained plot will indicate whether the product is acceptable or not. Such a quality control method will take only a few minutes, provided the machine is kept clean, precalibrated, and ready for use. The handling and maintenance of the machine should not be more difficult than for any equipment utilized in a quality control laboratory.

Recommendation I has the disadvantage that large particles may not be counted by the counter. Microscopic analysis could concurrently be done on the product. This does not mean that microscopic analysis, which is very tedious and time consuming, is a better tool.

Recommendation II—Figures 5 and 6 present the cumulative size distribution of particulate matter when Product 1H was filtered through a 0.45-μm membrane filter. It appeared obvious that the number of particle contaminants was far below the BP specification. Therefore, filtration prior to administration of the product was recommended as an efficient method of removing particulate matter. This could be accomplished by using an in-line final filtration device.

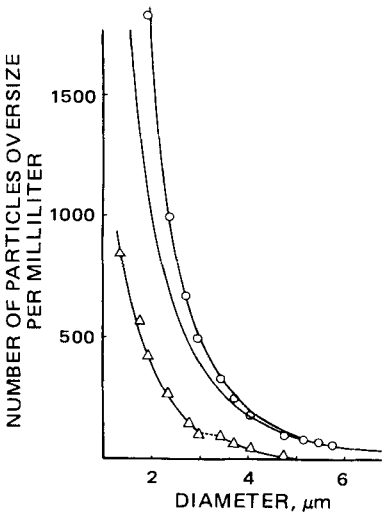

Figure 11—*Cumulative particle-size distribution in injection vials containing ampicillin sodium. Key: ○, Product 1B in 500-mg ampicillin sodium vials; △, Product 4AL in 1-g ampicillin sodium vials; and smooth curve, BP specification.*

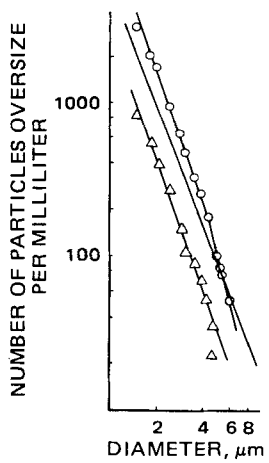

Figure 12—*Logarithmic plot of cumulative particle-size distribution in injection vials containing ampicillin sodium. Key: ○, Product 1B; △, Product 4AL; and smooth curve, BP specification.*

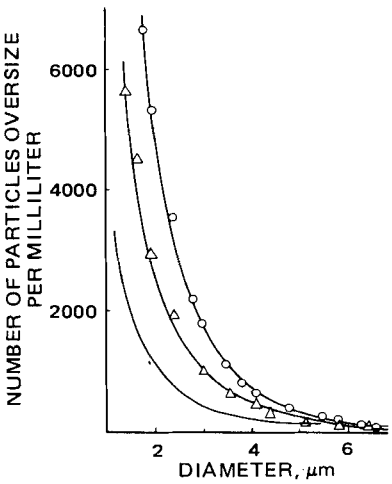

Figure 13—*Cumulative particle-size distribution in injection vials containing cephalothin sodium. Key: △, Product 2A in 1-g cephalothin sodium vials; ○, Product 1A in 500-mg cephapirin sodium vials; and smooth curve, BP specification.*

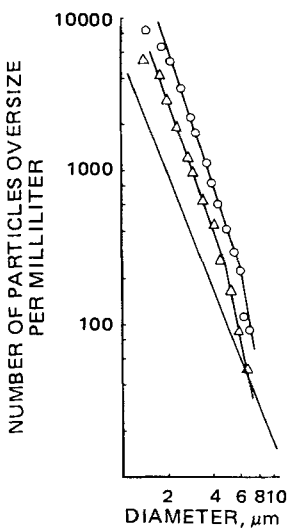

Figure 14—*Logarithmic plot of cumulative particle-size distribution in injection vials containing cephalothin sodium. Key: △, Product 2A; ○, Product 1A; and smooth curve, BP specification.*

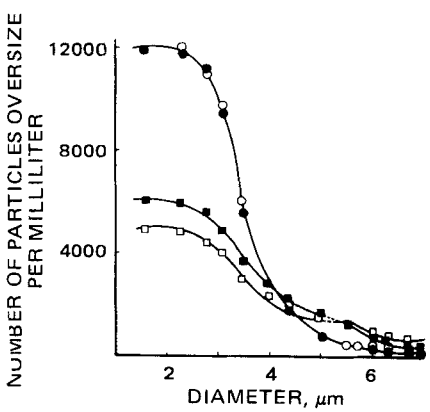

Figure 15—*Cumulative particle-size distribution in flocculated and deflocculated suspensions. Key: ●, deflocculated Suspension A (counts taken after 1 day); ○, deflocculated Suspension B (counts taken after 2 days); ■, flocculated Suspension A; and □, flocculated Suspension B.*

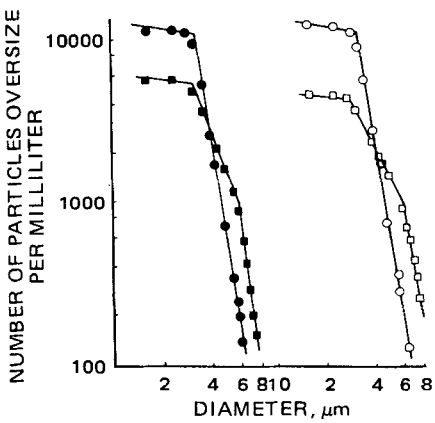

Figure 16—*Logarithmic plot of cumulative particle-size distribution in flocculated and deflocculated suspensions. Key: ●, deflocculated Suspension A; ○, deflocculated Suspension B; ■, flocculated Suspension A; and □, flocculated Suspension B.*

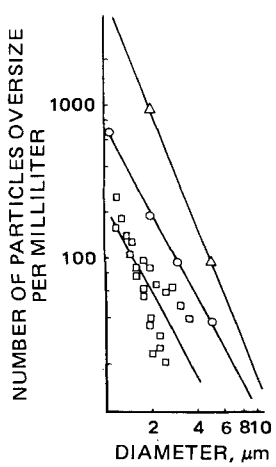

Figure 17—*Cumulative particle-size distribution indicating the least-squares line fit through data points of Products 1G, 1S, and 3G. Key: □, data points for Products 1G, 1S, and 3G; smooth curve, least-squares line; ○, proposed specification; and △, BP specification.*

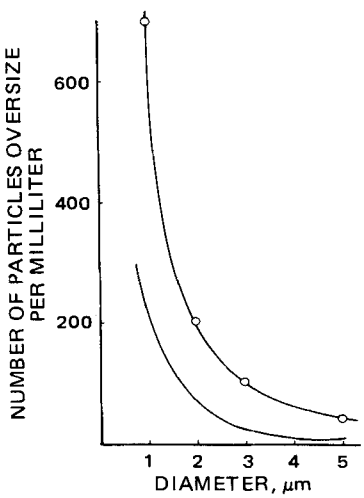

Figure 18—*Plot of number of particles oversize* versus *diameter. Key: smooth curve, least-squares line; and ○, proposed specification to fit* $N = 225/D^{1.8}$.

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Oxidation Photosensitized by Tetracyclines

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Abstract □ Irradiation with 365-nm UV light of aerated aqueous solutions of tetracycline gives rise to oxygen uptake when the pH of the solution is above 7.5. The kinetics of the reaction were followed using a polarographic oxygen electrode at a range of pH values for seven currently prescribed tetracyclines. Variation of tetracycline concentration, UV light intensity, and temperature showed the characteristics normally associated with a sensitized photo-oxygenation mechanism rather than a free-radical process. Copper(II) ions inhibited the photo-oxidation of tetracycline, apparently by complex formation. The tetracyclines were tested for photosensitizing capability with oxidizable acceptors. In aqueous solution, no photosensitizing effect could be seen, but methanol solutions of 2,5-dimethylfuran and *dl*-limonene were oxidized at considerably increased rates when small amounts of tetracyclines were present. This observation has implications for the mechanism of *in vivo* photosensitivity reactions that occur when tetracyclines are taken internally.

Keyphrases □ Tetracyclines, various—photosensitized oxidation, reaction kinetics followed using polarographic oxygen electrode, effect of pH and copper(II) ions □ Photosensitized oxidation—various tetracyclines, reaction kinetics followed using polarographic oxygen electrode, effect of pH and copper(II) ions □ Oxidation, photosensitized—various tetracyclines, reaction kinetics followed using polarographic oxygen electrode, effect of pH and copper(II) ions □ Polarography—determination, rate of oxygen uptake by solutions of various tetracyclines irradiated with UV light, effect of pH and copper(II) ions □ Antibacterial agents—various tetracyclines, photosensitized oxidation, reaction kinetics followed using polarographic oxygen electrode, effect of pH and copper(II) ions

Photosensitivity reactions have been reported for the tetracyclines after exposure of the patient to strong sunlight (1). The phenomenon of "photodynamic action" has been recognized since 1900, when it was discovered that microorganisms can be killed when exposed to light in the presence of oxygen and sensitizing dyes (2). The basic mechanism is the photosensitized oxidation of the adsorbate or substrate by molecular oxygen (3, 4). All tetracycline antibiotics have a strong absorption in the near UV region at about 365 nm, the first requirement for a sensitizer of photodynamic action.

Little information is available, however, concerning the reactions of tetracyclines following absorption of UV light. Chlortetracycline, tetracycline, and oxytetracycline lost significant antibiotic potency when irradiated with visible light in solution with riboflavin, which probably acted as a photosensitizer (5). Leeson and Weidenheimer (6) reinvestigated this system at pH 4.5 and concluded that the loss of tetracycline activity could be suppressed by the addition of ascorbic acid. Thus, an oxidative pathway was implied for degradation of tetracycline after irradiation.

This paper reports a more detailed study of the photooxidation of seven currently prescribed tetracyclines, together with some experiments that indicate that the tetracyclines can act as photosensitizers for the oxidation of suitable acceptor molecules.

EXPERIMENTAL

Samples of tetracycline[1] (I), oxytetracycline[1], doxycycline[1], metha-

[1] Pfizer Laboratories.