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LIQUID FILLING OF THERAPEUTIC SUBSTANCES

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This invention relates to the filling of definite quantities of solid materials into containers, and more particularly to such dispensing involving small quantities.

Considerable difficulty has been experienced in placing small amounts of solid material into a container. If a mechanical weighing of the solid is attempted, the machines must necessarily be extremely complicated and limited in precision. If manual handling is resorted to, it is a most time-consuming operation to obtain a satisfactory degree of accuracy. In some instances the solid material is dissolved in a suitable solvent and metered either mechanically or manually into containers as a solution, the solvent subsequently being removed by distillation or lyophilization, but this has many objections, as will appear.

Despite this difficulty of mechanical or manual filling of solids into containers, there are many instances in industry which require the dispensing of small amounts of a solid into individual containers. For example, in the pharmaceutical and biological fields, it is frequently necessary that an individual container include a small amount of the agent to be administered. This is because many therapeutic agents have such potent therapeutic value that a few milligrams will be sufficient for a single dosage, and a larger dosage may be injurious.

Moreover, there are many substances, particularly in the pharmaceutical and biological field, which do not readily lend themselves to handling by the conventional methods. If an attempt is made to dissolve the solid in a suitable solvent, it is frequently found that the solid is unstable in solution and its activity is destroyed or at least reduced. For example, there are labile antigens, antibiotics such as penicillin and aureomycin, and vitamins such as ascorbic acid, that oxidize quickly in solution or are otherwise destroyed by the water or other liquid.

The solution method of placing small amounts of a solid into a container is also found objectionable when the original form of the solid is to be preserved. For example, penicillin is very stable in crystalline form but not as an amorphous powder; when the liquid in which it is dissolved is removed, the penicillin appears as an amorphous powder. Even though lyophilization is resorted to as the only practical means of drying a penicillin-containing solution, the penicillin is left as an amorphous product which is in a less stable condition. Moreover, penicillin, or derivatives thereof, is often sold in definite crystal size, the latter condition being of paramount importance for controlled absorption of the drug when incorporated into repository products, such as penicillin or procaine penicillin in oil. Solution of the drug for the purpose of filling it in small quantities vitiates this intended use.

The solution method of filling small amounts of the solid is not useable when no suitable solvent is known for the material. For example, riboflavin cannot be dissolved in appreciable concentration in any acceptable

liquid and therefore does not lend itself to this conventional practice.

It should be emphasized that automatic weighing of the solids introduces many difficulties when the quantities desired are too minute to be handled properly by weighing machinery. This is true of certain vitamins and many immunizing antigens, such as allergens, dried vaccines and bacterial toxins. When the solid is dangerous to personnel, hazards may be created because automatic machines give rise to a certain amount of dust. In the case of streptomycin, which in the powdered state gives rise to allergies, this is a serious industrial problem. In the case of antigens, the allergy and toxicity problems are of primary importance.

Moreover, the limits of precision in automatic weighing are large, compared to the small amounts of material handled. With potent drugs (i. e. vaccines) such errors may lead to serious consequences, as reactions or lack of protection.

The usual mechanical or manual weighing involves the additional hazard of contamination. It is most difficult to maintain sterile conditions with the equipment which is required. The present invention readily lends itself to sterile operations because of the great ease with which it can be carried out.

The present invention obviates the many difficulties which have heretofore been encountered. In essence the invention consists in the dispensing of solid materials by suspending them in a liquid in which the solid material is insoluble and not chemically reactive, the liquid further having a specific gravity which is the same as or very close to that of the suspended solid. The suspension so prepared is filled into containers by transferring a measured amount of the suspension into individual containers. The suspending medium then may be removed by methods known to the art for accelerated conversion to a vapor as, for instance, evaporation, which includes evaporation both above and below the boiling point of the liquid used, and also sublimation.

This method completely eliminates the difficulties inherent with dry filling or filling as solutions. The desired material is not altered in form from its original state, it is not subjected to the type of decomposition (hydrolysis, oxidation) that so many sensitive materials undergo in solution, and it eliminates the search for a suitable solvent in those cases in which the material does not exhibit appreciable solubility in any conventional solvent (riboflavin). The difficulties inherent in dry weighing also are overcome since even minute quantities of material are easily handled by this invention. The danger of allergic or chemical irritation to personnel caused by fine dust during dry filling cannot occur in this method. Moreover, the precision is very great, being limited only to those errors involved in measuring out an amount of liquid, inasmuch as the concentration of the suspension can be varied over a wide range.

Filling in suspension is known to the art but there the suspending liquid has not been of equal or even approximately equal density and the suspended material has been kept from settling by vigorous agitation or shaking. However, unless the material happens to be a slowly settling substance, such as a flocculated protein, settling in the feed lines occurs even if the main storage reservoir is violently agitated. The selection of a suspending liquid of density substantially equal to that of the solid suspended is decisive in overcoming this difficulty, and, in fact, enables the filling of suspensions that cannot be handled otherwise.

The suspending liquid of the invention process may be of a single molecular structure or it may be a mixture of two or moret liquids of different density in order to have the desired final composite density. The liquids se-

lected for the mixture should be miscible with each other and none should be a solvent or impair the properties of the solid material to be suspended therein.

Mention should be made of the fact that the invention is limited to the handling of a single material at a time unless the materials of a mixture are all of approximately the same specific gravity. The amount of difference in specific gravity of materials that may be filled together is dependent only upon the primary requirement that they not separate during the time consumed in the filling process. Moreover, the precision with which the solution and the solid must be matched in respect to their densities is dependent on this same consideration of time consumed in the filling process.

The concentration of the solid in the suspending liquid is not critical. If the amount of solid to be filled is to be in micrograms, a greater accuracy is obtainable if the concentration is very low, but this of course requires an increased time for subsequent removal of the large amount of liquid. If the amount of solid to be filled is to be in grams or large fractions thereof, the concentration may be the maximum obtainable compatible with pouring or other ease of handling.

It is preferable that the suspending liquid be of a volatile nature so that it may be removed easily, leaving the solid material in the container. As has been mentioned, the liquid may be removed by ordinary evaporation or by boiling, either at atmospheric or reduced pressure. The suspending liquid can be removed by a lyophilization process as well. When the suspending liquid is removed under vacuum, a controlled reduction of pressure in the vacuum chamber, for example, over a period of approximately one hour, may be found to be important to eliminate "bumping" within the container.

In the case of penicillin, it has been found to be particularly suitable to remove the suspending medium by placing the vials containing the solid material in the suspending medium in an oven heated to approximately 100° C. for approximately one hour. Although the suspending medium can be removed by maintaining the vials in an oven at approximately 100° C. for about one hour, it was found that spore-contaminated samples could not be sterilized by this procedure. However, raising and maintaining the temperature at approximately 150° C. for about one hour, after the original hour of heating at 100° C., produces a sterilized condition and the therapeutic activity of the solid material is not measurably affected.

Another suitable means of removing the liquid suspending the penicillin is to place the filled vials in a vacuum chamber and to evacuate the chamber over a period of approximately one hour to a pressure of about 10 mm. of mercury. This pressure is then maintained for a period of from approximately two to twenty-four hours to complete removal of the suspending medium. The time required can be varied by variance of the temperature and pressure at which the process is carried out.

The following examples of the application of the invention are in no sense limiting. They have been selected to demonstrate the filling of substances that are difficult to handle by methods known to the art.

Example 1.—A suspension of crystalline sodium penicillin G, assaying 1690 units per milligram, was suspended in a medium composed of equal parts of methylene chloride and chloroform so that two milliliters of the suspension represented sixty milligrams of penicillin. Two milliliters of this suspension were filled into several vials and the vials and contents were then subjected to vacuum. The pressure of the vacuum chamber was reduced over a period of approximately one hour to about ten millimeters of mercury and this pressure was maintained for twenty hours. The vacuum was then released, the vials capped and then assayed for penicillin activity. The penicillin in the vials assayed 1760 units per milligram. The filled penicillin was crystalline, free of any trace of the suspending medium and of essentially the same color, potency and appearance as before filling.

One of the vials was subjected to an accelerated stability test consisting of being exposed to a temperature of 100° C. for four days and then was assayed. The penicillin assayed 1770 units per milligram showing no deterioration in antibiotic activity.

Example 2.—Crystalline sodium penicillin G assaying 1690 units per milligram was suspended in a medium consisting of equal parts of methylene chloride and chloroform such that each milliliter contained 120 milligrams of penicillin. Five mls. of this suspension was filled into vials and the vials then put under vacuum. The pressure of the vacuum chamber was reduced over a period of approximately one hour to about ten millimeters of mercury and the vacuum maintained for approximately 24 hours. The vacuum was released and the vials of penicillin stoppered. A sample of the penicillin in the vial assayed 1840 units per milligram. The filled penicillin was crystalline, free of any trace of the suspending medium and of essentially the same color, potency and appearance as before filling.

Upon being subjected to an accelerated stability test, consisting in exposure to a temperature of 100° C. for four days, the penicillin in a sample vial assayed 1830 units per milligram.

Example 3.—Crystalline sodium penicillin G assaying 1690 units per milligram was suspended in a medium consisting of four parts of ethylene chloride and six parts chloroform so that each milliliter of the suspension represented 30 milligrams of penicillin. Two milliliters of this suspension was filled into various vials and the vials subjected to vacuum. The pressure in the vacuum chamber was reduced over a period of approximately one hour to approximately ten millimeters of mercury and the vacuum maintained for approximately 24 hours. The vacuum was then released, the vials capped and assayed. The vials assayed 1760 units per milligram. The filled penicillin was crystalline, free of any trace of the suspending medium and of essentially the same color, potency and appearance as before filling.

After an accelerated stability test consisting in exposure to a temperature of 100° C. for four days, the penicillin assayed 1690 units per milligram.

Example 4.—Crystalline sodium penicillin G assaying 1690 units per milligram was suspended in a medium consisting of four parts of ethylene chloride and six parts of chloroform such that each milliliter of the suspension represented 125 milligrams of penicillin. Five milliliters of this suspension was measured into vials, and the vials subjected to vacuum. The pressure in the vacuum chamber was reduced over a period of approximately one hour to approximately ten millimeters of mercury and the vacuum maintained for approximately twenty-four hours. The vacuum was released, the vials capped and assayed. A sample vial assayed 1810 units per milligram. The filled penicillin was crystalline, free of any trace of the suspending medium and of essentially the same color, potency and appearance as before filling.

After an accelerated stability test consisting in exposure to a temperature of 100° C. for four days, a sample vial assayed 1760 units per milligram.

Example 5.—Crystalline sodium penicillin G assaying 1623 units per milligram was suspended in a medium consisting of six parts of chloroform and four parts ethylene chloride in such quantity that each two milliliters of the suspension represented 100,000 units of penicillin. Two milliliters of this suspension was filled into a number of containers and the containers were subjected to vacuum. The pressure in the vacuum chamber was reduced over a period of approximately one hour to about ten milliliters of mercury and the vacuum maintained for twenty-four hours. The vacuum was then released and the containers stoppered and assayed. Assay of 10% of the vials (10 vials) gave an average of penicillin content per vial of

112,100 units. The filled penicillin was crystalline, free of any trace of the suspending medium and of essentially the same color, potency and appearance as before filling.

Example 6.—Crystalline sodium penicillin G assaying 1650 units per milligram was suspended in a mixture of two parts ethylene chloride and three parts of chloroform such that each milliliter of the suspension represented 50,000 units of penicillin. Two milliliters of this suspension were filled into various containers and the containers then placed in an oven at 100° C. for approximately one hour. At the end of one hour the temperature was raised to 150° C. in order to sterilize the vials, all of the solvent having been removed during the first hour of heating. After exposure at 150° C. for one hour, the vials were stoppered and assayed for penicillin content and tested for sterility. All the vials were sterile and the penicillin content averaged 94,000 units per vial.

Example 7.—Example 6 was repeated using a suspending medium consisting of four parts of carbon tetrachloride and one part n-hexane. Substantially similar results were obtained.

Example 8.—Example 6 was repeated using a suspending medium consisting of 20 parts carbon tetrachloride and seven parts benzene. Substantially similar results were obtained.

Example 9.—Example 6 was repeated using a suspending medium consisting of 10 parts carbon tetrachloride and four parts ethyl acetate. Substantially similar results were obtained.

Example 10.—Example 6 was repeated using a suspending medium consisting of 10 parts carbon tetrachloride and three parts of di-isopropyl ether. Substantially similar results were obtained.

Example 11.—Crystalline potassium penicillin G assaying 1580 units per milligram was suspended in a medium consisting of six parts of chloroform and four parts ethylene dichloride in such quantity that each milliliter of the suspension represented 30 milligrams of penicillin. Two milliliters of this suspension was filled into various vials and the vials subjected to vacuum. The pressure in the vacuum chamber was reduced over a period of approximately one hour to approximately 10 millimeters of mercury and the vacuum maintained for approximately 24 hours. The vacuum was then released, the vials capped and assayed. The vials assayed 1565 units per milligram. The filled penicillin was crystalline, free of any trace of the suspending medium and of essentially the same color, potency and appearance as before filling.

After an accelerated stability test consisting in exposure to a temperature of 100° C. for four days, the penicillin assayed 1590 units per milligram.

Example 12.—Example 6 was repeated using crystalline potassium penicillin G instead of sodium penicillin. Substantially similar results were obtained.

Example 13.—1300 ml. of a mixture of 2¼ parts of carbon tetrachloride and one part acetone were added to 39.0 grams of the ground cells of *S. typhosa.* The specific gravity of the liquid mixture without the cells was 1.35 and the cells remained in a stable suspension. This suspension of cells was filtered through a 200 mesh nylon filter and then filled in 2 ml. volumes in 30 ml. vials so that each vial contained 60 milligrams of the cells. The vials were covered with process stoppers and placed in a vacuum chamber for removal of the filling fluid mixture. This was done by applying 60–110 mm. of pressure for 35 minutes. At this time the vacuum was increased to regular lyophilizing condition (100μ pressure) and applied for 20 hours. The vials were then removed, stoppered under vacuum and sealed with "roll-on" enclosures. The material made by this process was found to retain its Vi antigen content when tested in both mice and rabbits.

Example 14.—The process of Example 13 was carried out using *S. paratyphi* instead of *S. typhosa* and the same results were obtained.

Example 15.—The process of Example 13 was carried out using *S. schottmulleri* instead of *S. typhosa* and the same results were obtained.

Example 16.—The process of Example 13 was carried out using an equal part mixture of the cells of *S. typhosa, S. paratyphi* and *S. schottmulleri,* and the same result was obtained as they have approximately equal densities.

Example 17.—A liquid suspension mixture was made up of 20% chloroform and 80% ethylene dichloride, having a specific gravity of 1.280. To this was added crystalline vitamin B_{12} so that each milliliter of the liquid contained 250 micrograms of the vitamin B_{12}. This was measured out by placing 2 ml. in individual ampuls and the liquid was withdrawn by subjecting to a vacuum of about 10 mm. mercury for twenty-four hours.

Example 18.—A liquid suspension mixture was made up of 90% chloroform and 10% ethylene dichloride, having a specific gravity of 1.454. To this was added ascorbic acid so that each milliliter of the liquid contained 500 mg. of the ascorbic acid. This was measured out by placing 2 ml. in individual ampuls and the liquid was withdrawn by subjecting to a vacuum of about 10 mm. mercury for twenty-four hours.

Example 19.—To ethylene dichloride having a specific gravity of 1.250 was added the sedative, sodium 5-ethyl-5-(1-methyl-1-butenyl) barbiturate, sold under the trade name "Delvinal." Each milliliter of the mixture contained 60 mg. of the "Delvinal." This was measured out in 2 ml. quantities into individual ampuls and the liquid withdrawn by subjecting to a vacuum of 10 mm. mercury for twenty-four hours.

This is a continuation-in-part application of my application Serial No. 92,499, filed on May 10, 1949, entitled Liquid Filling of Antibiotic Substances.

What I claim is:

1. The process of filling a solid substance into a container comprising suspending the substance in a continuous phase of a relatively volatile body of liquid which is inert to the substance and is of a density substantially equal to that of the substance whereby to minimize any settling tendency due to differences in density, measuring into a container the quantity of the suspension which contains the desired amount of the substance and removing the suspending liquid as a vapor.

2. The process according to claim 1 in which the solid substance is penicillin.

3. The process according to claim 1 in which the solid substance is typhoid vaccine.

4. The process according to claim 1 in which the solid substance is ascorbic acid.

5. The process according to claim 1 in which the solid substance is dihydrostreptomycin.

6. The process according to claim 1 in which the solid substance is crystalline vitamin B_{12}.

7. The process of filling a solid substance into a container comprising suspending the substance in a continuous phase of a relatively volatile, organic body of liquid which is inert to the substance and is of a density substantially equal to that of the substance whereby to minimize any settling tendency due to differences in density, measuring into a container the quantity of the suspension which contains the desired amount of the substance and removing the suspending liquid as a vapor.

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