

antibodies or reagin. However, one should bear in mind that the general composition of the serum concentrated by the gelatin method is significantly altered, particularly with respect to the constituents present in colloidal state. The apparent discrepancy in the results obtained with the quantitative Kahn test and the Vernes test is noticeable. In 1926 Baylis, *et al.* (1) concluded in their paper dealing with the Vernes test: "The Vernes flocculation reading evidently depends, in part at least, upon other features of the patient's blood than does the Wassermann reading . . ." (p. 335). Epstein and Rubinstein (2) investigated the flocculation phenomena with syphilitic cerebrospinal fluids and expressed the opinion that the reagin is a relatively simple substance, unrelated to lipids or proteins. On the other hand, Vernes (3), who calls the reagin "Pallidine" and claims to have separated it from syphilitic serum, found that the substance did not pass through an ultrafilter, but that all of the active principle remained in the supernatant fluid after ultracentrifugalization of a solution for 17 hours at 86,000 r.p.m., while half of the solute was assembled at the bottom.

SUMMARY

A method is presented whereby certain constituents of serum or other liquids may be concentrated by the swelling of undissolved gelatin. Applications to chemistry, to the study of antibodies, and to the serology of syphilis have been indicated.

Colloids, such as proteins and cholesterol, were found to be concentrated as expected. Chlorides also complied with expectation, fully entering the swelling gelatin. An anomaly was found with calcium in blood serum; all of it behaved like nondiffusible material, in contrast with known facts pertaining to dialysis and ultrafiltration.

References

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A Method for Obtaining Standard Suspensions of Tubercle Bacilli in the Form of Single Cells

W. F. WELLS

*Laboratories for the Study of Air-borne Infection
School of Medicine, University of Pennsylvania*

A method devised to separate masses of organisms into single cells during culture (1) has proven singularly appropriate to the growth of the tubercle

bacillus. Suspensions of singled tubercle bacilli produced by this method develop quantitatively on appropriate media. Aerosol suspensions of these singled organisms of the bovine strain, sampled by the air centrifuge and grown on these media, give counts of colonies corresponding quantitatively to the number of tubercles developing in the lungs of rabbits inhaling these suspensions. The production of standard suspensions of singled organisms is, however, only one example of the usefulness of the method. It provides favorable cultural conditions which adapt it to *in vitro* techniques such as the study of the effect of antibiotics upon this bacillus.

The apparatus used in this process simulates a tiny ball mill (Fig. 1). A 250-cc. Erlenmeyer flask, con-

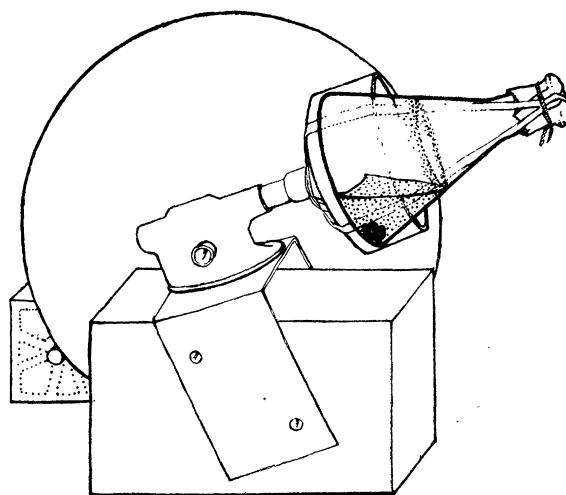


FIG. 1

taining 50 pyrex glass beads 4 mm. in diameter and 50 cc. of culture fluid, is rotated about its axis at an angle such that the top surface of the flask is horizontal. This angle exposes a maximum surface of the medium which becomes continuous with the film lining the upper interior surface of the flask as it revolves. Floating cultures of tubercle bacilli are carried, as on a traveling belt, over the surface of the liquid itself, picked up on the film lining of the flask, and returned to the other edge of the liquid surface. An extensive, well-aerated, cultural surface supplied with nutritive fluid is thus provided; the culture itself is broken up into a myriad of small patches.

After several days growth the culture begins to settle into the liquid, where it is subjected to a gentle grinding by the beads. This combination of subsurface culture with the gentle grinding produces an abundance of single cells which can be separated from the clumps by filtration through a No. 4 Whatman filter. The filtrate of singled organisms resembles broth cultures.

Transfer is also simplified: a bent glass rod dipped into the culture picks up patches from the surface which, because of the surface tension, immediately disperse over the surface of the inoculated fluid. A blend of equal parts of Difco nutrient broth, tryptose-phosphate broth, and brain-heart broth with 5 per cent glycerin was used as a culture medium.

In providing a standard suspension, a week-old cul-

ture is revolved a week. By such a schedule, 10,000,000–100,000,000 singled organisms per cubic centimeter are obtained in the filtrate. A steady biological state can thus be maintained for *in vitro* and *in vivo* experiments.

Reference

1. WELLS, W. F., and WELLS, M. W. *J. Amer. med. Ass.*, 1936, **107**, 1698–1703; 1805–1809.

Letters to the Editor

Comparative Digestibilities of Plastic Shortenings Made From Lard and From Hydrogenated Vegetable Oils

One of the most common popular beliefs which are stated as fact is that lard and other pork fats are less digestible than fats from other sources, such as beef fat, hydrogenated vegetable oils, etc. This misconception has persisted despite a tremendous amount of scientifically controlled experimental observations to the con-

groups receiving the bland lard as the shortening portion and the other two groups receiving a well-known, all-vegetable shortening purchased at a retail store for this purpose. After a three-day orientation period on the experimental diets, all feces were collected and food consumption measured for seven days. The two groups receiving the bland lard diet were then changed over to the vegetable shortening diet and *vice versa*. Again

TABLE 1
FAT INGESTION AND EXCRETION OF RATS ON DIETS CONTAINING PLASTIC SHORTENINGS MADE FROM LARD AND FROM ALL-HYDROGENATED VEGETABLE OILS

Animal group	A		B		C		D		All animals	
Shortening type	Lard	Veg.	Lard	Veg.	Lard	Veg.	Lard	Veg.	Lard	Veg.
Food consumption (grams)	444	494	480	484	493	472	430	431	1847	1881
Wt. fat in food (grams)	66.6	74.0	72.0	72.6	74.0	70.8	64.5	64.7	277.1	282.1
Wt. acidic extract (grams)	4.9	8.1	6.6	5.7	6.6	4.6	5.2	6.8	23.3	25.2
Corrected for low fat diet	3.7	6.9	5.4	4.5	5.4	3.4	4.0	5.6	18.5	20.4
X 1.045	3.9	7.2	5.6	4.7	5.6	3.5	4.2	5.8	19.3	21.3
Total fat undigested (%)	5.9	9.7	7.8	6.5	7.6	4.9	6.5	9.0	7.0	7.6
Digestibility (%)	94.1	90.3	92.2	93.5	92.4	95.1	93.5	91.0	93.0	92.4

trary. The most comprehensive studies, reported in Technical Bulletins from the Animal Nutrition Division of the U. S. Department of Agriculture (see R. Hoagland and G. G. Snider, *Tech. Bull.* 321), have shown that lard is significantly more digestible than either vegetable shortenings or blended vegetable and animal shortenings.

In the past two years a new type of shortening has been introduced which is made entirely of lard plasticized by the addition of hydrogenated lard and then deodorized to an entirely bland product, as is done in the case of vegetable shortenings. To prevent any suspicion that this bland lard shortening might be less digestible than a comparable all-vegetable shortening, the following feeding experiment was conducted.

The basic diet contained crude casein (18 per cent), dextrose (56 per cent), salt mixture (7 per cent), liver extract concentrate (3 per cent), brewers' yeast (1 per cent) and shortening (15 per cent). A control experiment using the above proportion of components without the shortening was run to determine the basal excretion of fat by the experimental animals.

Four groups of five albino male rats were started, two

there was a two-day transition period followed by a seven-day experimental period. Thus, each of the four groups was maintained for seven days on each of the two shortenings.

The collected feces were crushed, saponified in methanol, acidified with 35 per cent H_2SO_4 , and then extracted thoroughly with ether according to methods previously reported (see Hoagland and Snider; also K. F. Mattil and J. W. Higgins. *J. Nutrition*, 1945, 29, 255–260). The extracts were washed with water, dried, freed of solvent, and then dried to constant weight. From the weight of the acidic residue obtained in each case was subtracted the corresponding amount of lipid obtained in the feces on the low fat diet. The difference was multiplied by the factor 1.045 to convert to glyceride weight. The digestibility coefficients were determined from the calculated weight of excreted glyceride and the total amount of experimental fat ingested (Table 1).

Examination of the literature indicates that two fats which are equally digested by rats will also be equally digested by humans. (This point will be elaborated further in a forthcoming review on the subject.) From