Simple Syringe Burette[1]

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The use of a syringe for rapid and highly accurate delivery of fluids was introduced by Krogh and Keys (*1*), and their syringe pipette has since come into wide use. The less accurate syringe burette, described herein, and the titration procedure were developed primarily for field use. The equipment is compact and easy to operate, and gives an accuracy within 0.5 per cent for conventional titrations. The burette can also be used as a constant delivery pipette with a routine accuracy for total delivery of 0.1 per cent. The plunger can be removed at any time for cleaning without disturbing the setting of the instrument.

The burette (Fig. 1) consists of a standard 1-cc. tuberculin syringe on which is fused a 5-cm.-long, tapered delivery tip. The barrel of the syringe is provided with a split plastic block which can be clamped onto it by means of two screws (A). A straight-angle bow of stainless spring steel wire, 0.5–0.7 mm. in diameter, is press fitted into the block. The bow is so adjusted as to press slightly onto the plunger of the syringe to give it the necessary friction for a smooth operation. The plunger is notched (C) to engage the spring wire clip in order to furnish the automatic zero point. This notch is cut perpendicular to the axis of the plunger by means of a sharp-edged, triangular, medium or fine India oil stone. The cut must be sharp and straight and fit the spring steel bow without play. The notch should be located in line with the number on the plunger in order to orient the plunger regularly.

When used as a burette, it is necessary to grind the end of the plunger flat and perpendicular to the axis in order to obtain a parallax-free reading of the instrument. The grinding is performed in a block of bakelite provided with a series of holes of slightly different size. The plunger is placed in the best-fitting hole, and the block, together with the plunger, is ground against a piece of emery paper resting on a flat surface.

A split cork on the syringe barrel serves to unite the burette with the titration vessel (B).

To adjust the automatic zero setting of the burette, the syringe burette is filled with water and the block (A) loosened; while the spring engages the notch (C), the block is set so that the end of the plunger reads exactly 100. A lens should be used for reading. The graduation is calibrated by weighing the water delivered by, for example, 95 divisions, evaporation losses being most simply avoided by delivery into a tall and narrow, open weighing vial which is handled by forceps to avoid temperature gradients (*2*).

For constant delivery the plunger is moved down from the notch until it bottoms with the notch (and number) facing the bow. The end of the plunger must be ground flat, as described above, but it is left with a blunt edge so that it does not stick when moved to the bottom. The delivery is determined by weighing as described. Seven different syringe burettes were tested in this way. Out of 35 consecutive weighings, 32 agreed within ± 0.1 per cent or better.

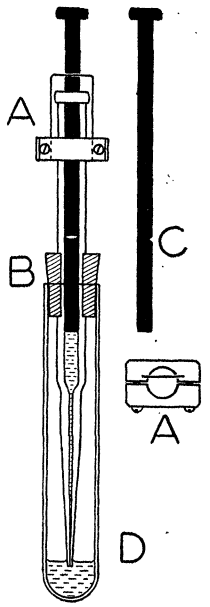

FIG. 1. Syringe burette attached to titration vial: A, plastic block with spring clip; B, split cork for uniting burette with titration vial (D); C, notch in plunger which engages spring wire (A) when the end of the plunger is at zero point of the syringe scale.

The sample to be titrated is delivered in the desired volume from a syringe or other accurate pipette directly to the bottom of the titration vial, without leaving drops sticking to the wall.

With the burette full of titration fluid (*i.e.* with the spring engaged in the notch), it is then inserted in the titration vial and fastened by means of the split cork. The tip of the burette is moved close to the surface of the sample, or slightly below, if the specific gravity of the burette fluid is lower than that of the sample.

While the whole unit is being agitated, the plunger is held by the button and moved slowly downwards by a screwing motion until titration is complete.

[1] This method was developed in connection with work on Cost Reimbursement Scientific Research and Development Contract No. W33-038-ac-14985, between the Air Materiel Command, Wright Field, and Swarthmore College (Department of Zoology).

[2] The writer is indebted to Dr. Laurence Irving for help and advice during this work. The tuberculin syringes were adapted for the present use by J. D. Graham, Department of Physiology, University of Pennsylvania.

For reading, the syringe is detached from the vial and the scale read by sighting across the flat end of the plunger and estimating to the nearest fifth or quarter of a division, again using a lens.

References

1. KROGH, A., and KEYS, A. B. *J. chem. Soc.*, 1931, Part II, 2436.
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Improved Technique for Enumeration of *Escherichia coli* on Black Walnut Meats[1]

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The enumeration of microorganisms found on solid materials is, in the main, based on their removal by washing a known weight of the solids with a known volume of washing fluid and then making serial dilutions from this fluid prior to the use of one of the common modes for determining numbers.

In the course of an investigation on the effectiveness of pasteurization for killing *Escherichia coli* on inoculated black walnut meats, it was found that washing the meats to remove the bacteria was not satisfactory when the Halvorson-Ziegler (*1*) method of determining the "most probable number" (MPN) of bacteria was used. The number of positive tubes found in the 10 replicate tubes of each of the three lowest dilutions gave codes which, when applied to the probability table, were equivalent to such "MPN" values as to make it evident that the seeding of the solid itself was needed if more accurate codes were to be secured.

Accordingly, an attempt was made to use the meats themselves in the replicate plantings, so that one member of the resultant code would be of no dilution. This was accomplished

TABLE 1

Test	MPN/gram	
	Washed meats	Blendor-treated meats
1	0	0.7
2	0.9	275.0
3	0	240.0
4	0	1.6
5	0	7.9
6	0	16.9
7	3.2	130.0

by cutting a known weight of the meats into small particles in a sterile Waring blendor container. After a homogeneous mass was secured and 10 replicate 1-gram samples had been removed for planting into lactose broth fermentation tubes, a known volume of sterile water was added to the container and the mixture further agitated until a creamy mass was secured. This material could be pipetted easily in making the 10 replicate plantings from this dilution, and it could also serve as the starting point for making subsequent dilutions.

[1] Published with the approval of the director, West Virginia Agricultural Experiment Station, as Scientific Paper No. 369.

The results of typical tests made on inoculated meats which had received various heat treatments are shown in Table 1. This table contrasts the "MPN" of *E. coli* per gram of meats when washing was used for the removal of these organisms with the "MPN" gained by the use of the Waring blendor procedure. Meats from the same batch were used when comparing the procedures.

TABLE 2

Test	MPN/gram	
	Washed meats	Blendor-treated meats
1	6,220	11,600
2	24,400	39,900
3	2,900	4,930
4	3,990	9,200
5	11,400	32,900
6	1,960	4,840
7	7,420	34,900

The superiority of the Waring blendor procedure is shown also in Table 2. Inoculated nut meats which had not been heat treated served as source material for contrasting the two modes of removal. Meats from the same batch were used when comparing the procedures.

The results of these tests suggest that, where the solid lends itself to subdivision, the use of the Waring blendor for this process permits the seeding of homogeneous, undiluted solid material into the tubes of broth. This procedure, together with the actual "carrying over" of the solid into subsequent dilutions, makes for a greater recovery of the organisms than that secured by washing the solid materials.

Reference

1. HALVORSON, H. O., and ZIEGLER, N. R. *J. Bact.*, 1933, **26**, 559-567.

Cultivation of Microorganisms With the Aid of Cellophane Membranes

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This brief report describes the use of thin sheets of cellophane for the cultivation of bacteria and fungi. We first used cellophane for this purpose during the German occupation, when Petri dishes were unobtainable.

A sheet of cellophane was rolled around a rod and sterilized by autoclaving. It was then unrolled on a table, and melted agar medium was poured out over the sheet. The surface was inoculated by spraying, and the agar immediately covered by a second cellophane sheet. To save space, the whole culture was finally rolled up again loosely around the rod and incubated.

Recently we have taken up the problem from another side, using the cellophane sheets not only as a substitute for glass but as a dialysing membrane. These sheets are impermeable to bacteria and viruses, but they allow the passage of water and solutes (except those of very large molecular weight). Thus, colonies of bacteria can be grown on one side of the cellophane,