In addition to showing that compounds totally devoid of chemotherapeutic activity (*i.e.*, true sulfonamide action) give inhibition equal to or exceeding that given by sulfanilamide, these data also show that the inhibition of the hemolytic streptococcus is not greater than that of the staphylococcus which generally is more resistant to sulfonamides.

Further, the effect upon respiration of a more active sulfonamide, sulfacetimide, was compared with that of sulfanilamide. This compound was selected because it would dissolve in .04 M concentrations. Preliminary experiments with sulfathiazole indicated that significant reductions of the rate of respiration could not be obtained with concentrations up to 100 mg per cent., the upper limit of solubility.

TABLE II

EFFECT OF .04 M SULFANILAMIDE AND SULFACETIMIDE ON RESPIRATION OF *E. coli* ON GLUCOSE IN M/60 PHOSPHATE BUFFER IN AIR

	Q_{O_2}		
	Control	.04 M Sulfanilamide	.04 M Sulfacetimide
pH 6.2	155	126	125
Inhibition ..		19 per cent.	19 per cent.
pH 7.2	143	126	121
Inhibition ..		12 per cent.	15 per cent.

Sulfacetimide shows no greater activity in this experiment than sulfanilamide. When 10 mg per cent. *p*-aminobenzoic acid was added to some of the flasks containing .04 M sulfonamide it did not reverse the inhibition of respiration by either sulfanilamide or sulfacetimide.

Finally an attempt was made to compare inhibition of respiration and of growth, using resistant organisms. The relative sulfonamide resistance of a parent strain of *E. coli* and a resistant strain developed from it is given in Table III.

TABLE III

CONCENTRATIONS OF SULFONAMIDES PERMITTING ONE-HALF MAXIMUM GROWTH RATE OF *E. coli* IN SYNTHETIC MEDIUM AT PH 7.0

	Parent Strain	Resistant Strain
Sulfanilamide ...	3.4 mg per cent.	62 mg per cent.
Sulfaguanidine ..	3.4	63
Sulfapyridine ...	.17	1.6
Sulfadiazine	.077	.34
Sulfathiazole	.073	.35

Inoculum = 100,000 cells per ml.

However, when the effect of .04 M sulfanilamide on the respiration of resting cells of these organisms was compared, equal inhibition was obtained with both strains.

TABLE IV

EFFECT OF .04 M SULFANILAMIDE ON GLUCOSE RESPIRATION OF RESISTANT AND NON-RESISTANT *E. coli* IN M/60 PHOSPHATE BUFFER, PH 7.2 IN AIR

	Control Q_{O_2}	.04 M Sulfanilamide Q_{O_2}
Parent strain	61	52
Inhibition		15 per cent.
Resistant strain ..	69	58
Inhibition		16 per cent.

SUMMARY

These data indicate that the inhibition of bacterial respiration is not a suitable criterion for the presence or absence of true sulfonamide activity.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

MICROMETER BURETTE

THE microburette recently described by Scholander[1] in common with all other modifications of a Rehberg burette[2] can only be used with solutions that do not attack mercury. Burettes of this type get dirty quickly, probably because grease creeps along the mercury. They are also hard to clean. To avoid these difficulties, we have constructed and used a burette combining a micrometer with a syringe, as has been done in one imported microburette. The anvil of a micrometer is cut off and a glass syringe mounted on a simple clamp in line with the spindle. Rubber bands attached to two hooks near the knurled head of the micrometer and to the plunger hold the latter tight against the spindle. A delivery tube can be attached to the syringe with a No. 0 one-hole rubber stopper; or if necessary a broken syringe of the same glass can be drawn out and fused on to the orifice of the syringe. A brass washer should be cemented to the outer end of the plunger and accurately perpendicular to its axis to act as a thrust bearing against the spindle. This bearing should be oiled occasionally. A convenient support can be made by screwing the yoke of the micrometer to the boss of a universal burette clamp, which can then be attached to a ring stand. The clamps on the syringe should be lined with friction tape to protect the glass and prevent slippage. Extra clamps permit the use of syringes of different sizes. We have used a 1-inch micrometer which delivers about 0.4 cc from a 1 cc tuberculin syringe or 1.5 cc from a standard 2 cc syringe. The syringes can

[1] P. F. Scholander, SCIENCE, 95: 177, 1942.
[2] P. B. Rehberg, *Biochem. Jour.*, 19: 270, 1925.

easily be dismounted and cleaned and even sterilized if necessary, permitting the use of several solutions in the same burette. The micrometer burette can be conveniently calibrated by titrating a dilute base with

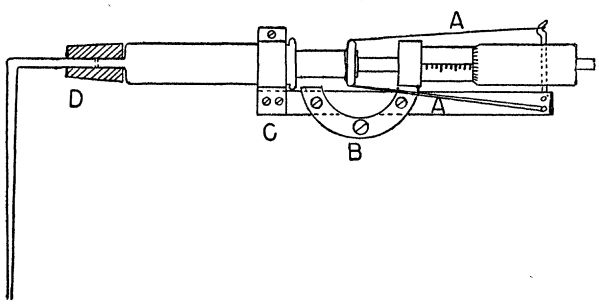

FIG. 1. A, Rubber bands; B, head of screw into boss of burette clamp; C, clamp around syringe barrel; D, rubber stopper.

constant boiling HCl. We have used cheap micrometers[3] and found linear calibrations to one part in 1,000 independent of the speed of delivery. The rest of our procedures did not warrant greater accuracy, but since some micrometers are accurate to 1 part in 10,000 and at least equal accuracy can be obtained with a Krogh syringe pipette,[4] the combination could doubtless be used with a corresponding accuracy.

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A STABLE HYDROGEN PEROXIDE AEROSOL

THE work of Twort and co-workers,[1] of others[2] as well as the recent work of Robertson[3] and his co-workers on the effect of propylene glycol aerosols on the decontamination of virus-infected air has led us to investigate the production and stability of hydrogen peroxide aerosols. Applying principles previously described[4] and using commercial nebulizers, hydrogen peroxide aerosols have readily been formed.

As described previously, the droplet vapor pressure was controlled by 50 per cent. glycerol. A solution of 0.1 per cent. hydrogen peroxide containing a stabilizing agent was nebulized at low pressure for forty-five minutes. During this time the weight decrease of the original solution was about 50 per cent. The hydrogen peroxide titre of the residual solution after nebulization was more than 0.1 per cent. (the original value) in spite of the fact that the solution was filled with bubbles resulting from the aeration. This increase in peroxide content following nebulization will be subsequently explained.

A stronger solution (3 per cent.) of hydrogen peroxide was vigorously nebulized in a closed room, $10 \times 10 \times 15$ feet, for forty-five minutes. The room was continuously filled with a fog produced by our technic of nebulization. Both normal and allergic individuals did not feel any discomfort or irritation while remaining in the room for as long as five minutes. Samples of the air were positive for peroxide. During the forty-five-minute period of nebulization, the volume of the solution decreased one half, but the peroxide content *increased* about 25 per cent. This increase in peroxide content was probably due to evaporation of water. In any event, it was surprising to find that the concentration of peroxide increased after nebulization. This makes the nebulization procedure practical. It is of interest that one may repeatedly breathe in dense mists of this concentration of peroxide without any irritation.

By inverting a two-liter bottle and forming a mist inside, the stability of a sample of a mist in this vessel was followed as well as the stability of the hydrogen peroxide droplets themselves. Potassium iodide starch papers were thrust quickly under the bottle at various intervals and the change in color followed. In this simple fashion it was found that hydrogen peroxide mists formed by nebulization show excellent peroxide activity (gaseous or droplet) for at least as long as one and one-half hours after the mist has been formed.

An investigation of additional biological and chemical properties of these stable hydrogen peroxide aerosols is in progress.

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[3] These can be obtained, for example, at Sears Roebuck and Company, or radio supply houses for about one dollar.

[4] A. Krogh. *Ind. and Eng. Chem. Anal. Ed.*, 7: 130, 1935.

[1] D. C. Twort, A. H. Baker, S. R. Finn and E. O. Powell, *Jour. Hyg. Camb.*, 40: 253, 1940.

[2] An excellent review of the literature: A. H. Baker, *Chem. Prod.*, January, 1941, p. 25.

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