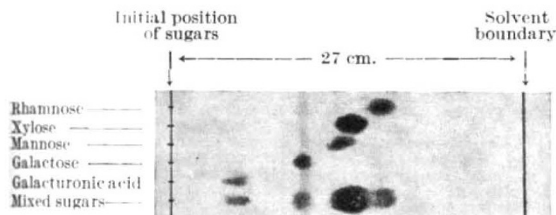

FILTER PAPER CHROMATOGRAM OF FIVE MONOSACCHARIDES SHOWING THE SEPARATION OF THE MIXTURE. SOLVENT, COLLIDINE. PAPER, WHATMAN No. 1. TEMP. 21–23° C.

The photograph shows the separation of a number of sugars in s-collidine. The difference in R_F value between xylose and mannose is not great enough to result in separation between these two sugars. With phenol as the moving phase, three groups of sugars could not be separated; these were: rhamnose, ribose, glucosamine (R_F 0·59–0·62); galactose, mannose, xylose (R_F 0·44–0·45); sorbose, glucose (R_F 0·39–0·41). The use of collidine or butanol-ethanol mixtures permitted the separation of the first and second groups, provided the amount of sugar taken was not too large. Sorbose and glucose could not be separated in any of the three solvents investigated, although small differences in R_F values could be demonstrated by use of the individual sugars.

This work forms part of the programme of the Food Investigation Board, of the Department of Scientific and Industrial Research.

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July 12.

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Alginic Acid-acetate

VARIOUS attempts have been described[1] to acetylate alginic acid by means of acetic anhydride or acetic acid, without or with catalysts, for example, pyridine, acids, etc. Some of the products thus obtained were probably degraded, and it appeared of interest, therefore, to find out whether the acetylation of alginic acid can be done by a relatively mild method; for this reason the interaction with ketene was studied.

Alginic acid fully swollen with acetone reacts with ketene at room temperature to form a colourless insoluble ester acid which can be converted into a sodium and calcium salt. These derivatives could also be prepared by direct reaction of sodium or calcium alginate and ketene. The analysis of the various products indicates that approximately one acetyl grouping has been introduced into each repeating unit of these chain polymers. Viscosity determinations were made with solutions containing sodium alginate obtained by hydrolysis of the acetyl derivatives, and these tests lead to the following conclusions.

During the acetylation of calcium alginate, no substantial degradation appears to occur; but during the reaction with sodium alginate and particularly with free alginic acid, some degradation takes place. The viscosity of solutions containing the 'recovered' sodium alginate is in all cases far higher, however, than that of solutions of salts of low molecular uronic acids. The following polymers were tested with regard to their swelling in water:

	Moles of water × 10^{-2} bound at 20° by 1 gm.-equivalent
Alginic acid	7
Sodium alginate	∞
Calcium alginate	6
Alginic acid-acetate	5
Sodium salt of alginic acid-acetate	25
Calcium salt of alginic acid-acetate	12

It should be noted that, in contrast to sodium alginate, which is soluble in water, the sodium salt of alginic acid-acetate swells to a limited extent only. If the free ester-acid is dried and the sodium salt is made by adding, at room temperature, an equivalent amount of sodium hydroxide solution, a non-transparent gel is obtained which binds only $\sim 8 \times 10^2$ mol. water per gm.-equivalent; if, on the other hand, the sodium hydroxide solution and the fully swollen ester acid are mixed at about 60°, a colourless, transparent jelly is obtained, with a high degree of swelling (see table). An equally highly swollen transparent jelly can be made by dispersing the moderately swollen modification of the sodium salt in glycerol and adding excess water to the dispersion.

It has been found that alginic acid-acetate and its sodium salt are cation-exchange materials, that is, these colourless gels can be used either to remove certain cations, such as calcium, from dilute solutions, or as adsorption materials for inorganic chromatography.

A full account of these experiments will be published elsewhere.

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July 27.

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Influence of Glucose in the Assay of Streptomycin

DURING the course of our investigations into the production and properties of streptomycin, we have confirmed the majority of the observations recorded by Waksman *et al.*[1,2], Denkelwater *et al.*[3], and by Abraham and Duthie[4]. These confirmations cover thermal stability, *p*H at which optimum activity is apparent, stability in solutions on storage, and methods of inactivation; but we have obtained results which suggest that the effect of glucose, to which attention has been directed, may be more complex than has hitherto been thought.

In their work on the properties of streptomycin, Waksman *et al.*[1] indicated that two methods of assay were used: (*a*) a serial dilution method using *B. coli* as the test organism; and (*b*) a cup-plate method using spores of *B. subtilis*; and they showed that in the latter test the addition of 2 mgm. of glucose to 10 ml. of agar reduced the potency of streptomycin by one half. The suggestion put forward for this reduction[2] was that "This may be due to the reducing properties of glucose or to the production of some acid by the test organism". In this laboratory we have used mainly a ring-plate method of assay with *B. coli* as the test organism and a bile-salt-lactose agar medium. We have preferred to use *B. coli* instead of a spore-bearing bacterium, such as *B. subtilis*, because our original interest in streptomycin arose from the claims that it was active against Gram-negative bacteria. We use bile-salt-lactose agar because it gives a better defined zone of inhibition than ordinary nutrient agar made from tryptic digest broth.

Under our conditions of assay, the addition of glucose to the medium did not result in any apparent change in potency of the streptomycin. When, however, nutrient agar was substituted for bile-salt agar, a decline in potency was observed, the decline being dependent on the amount of glucose added to the medium. On the other hand, when *B. subtilis* was used in nutrient agar, a reduction in apparent potency, similar to that obtained by Waksman, was found (see table).

EFFECT OF GLUCOSE ON THE PLATE ASSAY OF STREPTOMYCIN USING VARIOUS MEDIA AND ORGANISMS

Glucose added (mgm.) per plate (15 ml. agar)	Assay (u./ml.) using *B. coli*		Assay (u./ml.) using *B. subtilis*		
	Nutrient agar	Bile-salt-lactose agar	Nutrient agar		Quoted by Waksman[1]
			Sample *A*	Sample *B*	
0 (Control)	60	60	82	80	1200
2	56	62	43	—	—
3	—	—	—	—	600
10	36	63	35	51	—
15	—	—	35	—	620
20	17	61	38	54	—
40	8	55	—	55	—

In addition to the above tests, a series of assays was carried out in which glucose was added to solutions of streptomycin (70–80 u./ml.) instead of to the culture medium. The results showed that the activity was reduced when assayed against *B. subtilis*, the activity falling from 85 u./ml. with glucose absent to 46 u./ml. when 20 mgm. glucose per 10 ml. solution were added. In the tests with *B. coli*, however, no reduction was observed in either nutrient agar or in bile-salt-lactose agar.

From these results it is evident that the effect of glucose on the apparent potency of streptomycin depends on the organism and the culture medium used in the assay. It seems doubtful, therefore, that acid production by the organism or reducing action is solely responsible for the phenomena encountered.

We have pleasure in acknowledging the active interest of Sir Jack Drummond, and the help given by Mr. C. E. Coulthard.

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July 22.

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Strong Magnetic Fields

IT is well known that with strong electromagnets one can produce unlimited magnetic fields, since the field increases with the logarithm of the dimensions of the polepieces; but in reality the field obtainable is limited by the enormous expense involved by the building of big electromagnets.

In the case of solenoids, the limit of the field is determined by the way in which the heat developed is carried away, and by the cost of the energy source; the experiments of Deslandres and Pérot have shown the order of magnitude of the strongest fields obtainable in this way.

In 1924 Kapitza succeeded in producing in an original way very strong fields of short duration by short-circuiting a battery by a small solenoid during a fraction of a second; in 1927 he improved his method by using, instead of a battery, a generator which could stand the enormous shock caused by the short-circuiting. In these experiments the small solenoid had to be reinforced, as without this precaution it tended to explode. In this way fields of short duration (about 1/100 sec.) could be produced.

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