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Physiology of Pure Culture of *Trichomonas vaginalis:* III. Fermentation of Carbohydrates and Related Compounds.

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This study is a continuation of a survey of the physiologic activities of a bacteria-free culture of *Trichomonas vaginalis*.[1-5] Knowledge of the fermentation reactions of this organism is desirable to serve as a basis for the comparison of this species with other pure cultures when they may become available. Moreover, since compounds containing carbohydrates are extensively used in the therapy of vaginal trichomoniasis, the demonstration of any trichomonas-growth-stimulating action of a carbohydrate suggests that it would probably be of limited clinical value.

Experimental. All experiments were performed in duplicate and for those compounds utilized by the protozoa a third test was made.

Utilization of the various substances was determined by comparing the population increase and pH shift in a basic medium with and without the test compounds. Four tubes with the test compound were used in most of the experiments and the results averaged. Population counts were made in hemocytometers (one cubic mm counted per tube). pH measurements were made with glass electrode equipment standardized each time against 2 known buffers.

The basic medium employed and recommended, which gives very poor growth in the absence of utilizable carbohydrates, is prepared as follows:

Two percent proteose peptone No. 3 (Difco), 0.5% NaCl, and 0.1% agar in distilled water adjusted to pH 6, is tubed in 9 cc amounts and autoclaved. After cooling, 0.5 cc of sterile filtered undiluted human serum (adjusted to pH 6 with N/1 HCl) and sufficient autoclaved concentrated sodium thioglycollate solution to give a final concentration of 0.1% are added. The latter is neces-

1 Trussell, R. E., *J. Iowa M. Soc.*, 1940, **30**, 66.

2 Trussell, R. E., and Plass, E. D., *Am. J. Obst. and Gynec.*, 1940, **45**, 883.

3 Kupferberg, A. B., PROC. SOC. EXP. BIOL. AND MED., 1940, **45**, 220.

4 Johnson, G., PROC. SOC. EXP. BIOL. AND MED., 1940, **45**, 567.

5 Johnson, G., *Anat. Rec.*, 1940, **78** (suppl.), 179.

TABLE I.
Representative Data Taken from Duplicate or Triplicate Experiments Testing the Fermentation Activities of a Bacteria-free Culture of *Trichomonas vaginalis*.

	A. Experimental cultures in basic medium + test compound				B. Control cultures in basic medium without test compound			Result
Test compound	Uninoculated pH	pH after 3-4 days culture	No. tubes	Avg population per mm³	Uninoculated pH	pH after 3-4 days culture	Avg population per mm³	Compound utilized
Monosaccharides								
Arabinose	5.96	6.22	4	1	6.00	6.19	61	0
Rhamnose	5.93	6.10	4	8	6.00	6.16	9	0
Xylose	5.64	5.65	4	3	5.69	5.66	5.5	0
Glucose	6.18	5.22	4	378	6.21	6.17	9	+
Fructose	6.18	6.07	8	104	6.21	6.17	9	±
Galactose	6.12	6.05	3	58	6.21	6.17	9	±
Mannose	6.33	6.27	5	7	6.35	6.40	8	0
Sorbose	6.34	6.38	5	7	6.35	6.40	8	0
Disaccharides								
Lactose	6.02	6.23	4	87	6.00	6.19	61	0
Sucrose	6.00	6.19	4	64	6.00	6.19	61	0
Maltose	6.18	4.69	4	1037	6.21	6.17	9	+
Trehalose	6.00	6.06	4	0.5	6.00	6.08	3	0
Melibiose	6.00	6.10	4	1	6.00	6.16	4	0
Cellobiose	6.00	6.05	3	7	6.00	6.16	4	0
Polysaccharides								
Raffinose	5.70	5.67	4	2	5.69	5.66	5.5	0
Melezitose	6.00	6.20	5	16	6.00	6.08	3	0
Starch (Sol.)	6.18	4.47	3	1260	6.21	6.17	9	+
Inulin	5.79	5.76	4	8	5.69	5.66	5.5	0
Dextrin	6.15	4.66	3	1055	6.21	6.17	9	+
Glycogen	6.15	4.65	3	1268	6.21	6.17	9	+
Pectin	6.13	6.21	5	5	6.35	6.40	8	0
Xylan	6.35	6.36	5	7	6.35	6.40	8	0
Alcohols								
Glycerin	5.73	5.70	4	3	5.69	5.66	5.5	0
Erythitol	5.68	5.70	4	4.6	5.69	5.66	5.5	0
Adonitol	6.00	6.11	5	4	6.00	6.16	4	0
Mannitol	5.66	5.65	3	6	5.69	5.66	5.5	0
Sorbitol	5.99	6.16	5	34	6.00	6.08	3	0
Dulcitol	6.35	6.44	5	8	6.35	6.40	8	0
Glucosides								
Salicin	5.71	5.63	4	17	5.69	5.66	5.5	0
Aesculin	5.70	5.86	5	4	6.00	6.08	3	0
α Methyl Glucoside	6.00	6.15	4	13	6.00	6.16	7	0
Polyhydric Phenol								
Inositol	6.37	6.45	5	8	6.35	6.40	8	0

sary because it was found that "aerobic" fermentation did not occur in its absence.

The 32 compounds listed in Table I were added to the basic medium to a final concentration of 0.5%. The test substances in concentrated solution were sterilized either by filtering or by autoclaving 8 minutes. The basic medium without any additions served as the control.

The inoculums used in test and control tubes were the same in any given set of experiments. Population counts and pH measurements were determined simultaneously. Routine bacteriologic cultures were made from all tubes and no data are included from contaminated specimens.

Under the conditions of these experiments glucose, and its polymers, maltose, soluble starch, glycogen, and dextrin alone were utilized to any considerable extent. Fructose and galactose are in a borderline group as evidenced by a slight stimulation of growth but no appreciable pH shift.

These results differ somewhat from those recorded by Cailleau[6] for *T. foetus* and *T. columbae* and offer one more argument against the identity of *T. vaginalis* and the lower animal trichomonads.

It is interesting clinically that growth of the strain of *T. vaginalis* tested is not stimulated by lactose. This is also true of the common vaginal fungi ("monilia"). Hesseltine[7] contends that lactose should be used in the therapy of vaginitis to prevent the growth stimulation of the fungi which may occur if dextrose is employed. The data here recorded indicate that a similar situation prevails among the trichomonads, although it is obvious that other strains must be studied before such conclusions are warranted.

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Susceptibility of Syrian Hamster (*Cricetus auratus*) to Viruses of St. Louis and Japanese B Encephalitis.

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Because of the lack of other susceptible animal hosts, experimental studies on the virus of St. Louis encephalitis have been confined entirely to the use of the mouse and the monkey. Similarly, although the host-range for the Japanese B encephalitis virus appears to be larger,[1] only the mouse and the monkey have proved suitable for laboratory purposes. In view of the desirability, therefore, of having available a fairly large, inexpensive, and readily procurable

[6] Cailleau, R., *Ann. Inst. Pasteur*, 1937, **59**, 137, 293.

[7] Adair, F. L., and Hesseltine, H. C., *Am. J. Obst. and Gynec.*, 1936, **32**, 1.

[1] Kasahara, S., *et al.*, *Kitasato Arch. Exp. Med.*, 1936, **13**, 248.