SPG on blood, particularly agglutination of erythrocytes, would certainly alter some of the physical properties of blood. It is interesting to note that Rosenthal *et al.*(3) mention no effect, other than an "expected increase" in sedimentation rate, on human erythrocytes with their bacterial γ, D-glutamyl polypeptide. That the increase in sedimentation rate observed with SPG is not just a non-specific effect of all macromolecules, can be concluded from the fact that commercial dextran had negligible effects on ESR of human and dog bloods in our studies. Synthetic α, L-copolymers of glutamic acid and lysine have also been investigated with respect to their effects on blood *in vitro*. It has been found (Klein, Fasman and Blout, personal communication), that those copolymers containing more than 65 mole % of glutamic acid behaved similarly to SPG in that they caused agglutination of erythrocytes and an increase in viscosity of plasma; those in the 50-60 mole % range exhibited no such toxic effects. Determination of blood levels of SPG revealed that after the first 5 hours further elimination of the drug did not occur. Such a finding was compatible with the observations of urinary excretion of SPG, where it was noted that no appreciable amount was excreted after the first 6 hours. In contrast to the toxic effects of SPG reported here, Maurer (7) found in immunological studies that no detectable antibodies could be produced against the material (batch #S-2035-186C) either in man, rabbit, or guinea pig.

Summary. (1) A synthetic polypeptide, sodium poly-α, L-glutamate (SPG), has been evaluated as a plasma expander by comparing it with dextran in both normovolemic and hypovolemic dogs. (2) SPG at a concentration of 3% was equivalent to 6% dextran as a plasma expander. (3) Further studies with SPG proved it to be too toxic for clinical trial. It was observed to impair cardiac output in the dog, and to cause agglutination and an increase in sedimentation rate of both human and dog erythrocytes. (4) A considerable part of the injected dose remained in the blood at 5 hours after injection and no evidence of further elimination over the next 3 days was observed.

1. Bovarnick, M., Fieber, S., Bovarnick, M. R., Kazlowski, J., PROC. SOC. EXP. BIOL. AND MED., 1953, v83, 253.

2. Bovarnick, M., Eisenberg, F., O'Connell, D., Victor, J., Owades, P., *J. Biol. Chem.*, 1954, v207, 593.

3. Rosenthal, W. S., O'Connell, D. J., Axelrod, D. R., Bovarnick, M., *J. Exp. Med.*, 1956, v103, 667.

4. Blout, E. R., Idelson, M., *J. Am. Chem. Soc.*, 1956, v78, 497.

5. Krayer, O., Mendez, R., *J. Pharmacol. and Exp. Ther.*, 1942, v74, 350.

6. Wasserman, K., Mayerson, H. S., *Am. J. Physiol.*, 1952, v171, 218.

7. Mauer, P. H., PROC. SOC. EXP. BIOL. AND MED., 1957, v96, 394.

Received December 2, 1958. P.S.E.B.M., 1959, v100.

Effects of Cortisone and Herpes Simplex Virus on Metabolic Processes. I. Alterations in HeLa Cell Metabolism.* (24777)

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Recently various effects of cortisone and related steroids on cell growth and on carbohydrate and nitrogen metabolism have been reported(1-3). Gavosto *et al.*(2) suggested that cortisone increases gluconeogenesis in animals leaving a negative nitrogen balance due to enhanced transamination processes. Independently, Rosen and Roberts(3) substantiated these claims. As yet one can only speculate as to the effect of cortisone on metabolism of isolated tissue culture systems. Although cortisone treatment may be dramatically effective in limiting inflammatory re-

* Supported by research grants from U.S.P.H.S. Presented in part at 40th meeting of Am. Soc. of Immunol., Philadelphia, Apr. 1958.

sponse of non-infectious processes, it is contraindicated for diseases of viral etiology, since considerable evidence has accumulated suggesting that the steroid plays an important role enhancing the invasive properties of certain viruses(4-9). Our investigation was carried out to determine effect of herpes simplex virus on carbohydrate metabolism of HeLa cells grown in presence and absence of cortisone.

It seemed important to learn whether or not cortisone actually affects the energy-yielding mechanism of the cell, and if so, whether it acts in conjunction with virus to enhance effects of the latter. Findings relevant to these questions will be described to understand something about biochemical injuries imposed on HeLa cell metabolism under such conditions.

Methods. Tissue culture. Initially HeLa cells were grown on glass (200 ml milk dilution bottles) in Eagle's medium containing 10% human serum; later, due to availability of adequate serum volumes, calf serum was substituted for human serum. Cortisone acetate was added when required, to final concentration of 2 μg/ml medium. Cultures, diluted initially to total of 14 x 10^5 cells in 14 ml growth medium, yielded cell sheets which appeared confluent and contained approximately 8 x 10^6 cells after 7 days at 37C. Cultures were washed with Hanks' balanced salt solution and wash fluids then replaced by Eagle's basal medium containing 10% rabbit serum (and cortisone if previously grown in cortisone-containing fluids). Four bottle cultures grown in the presence of cortisone and 4 others cultured without the steroid were then infected with 0.8 to 1 ml herpes virus suspension. Similarily, uninfected control bottles received 0.8 to 1 ml medium so that a total of 16 bottle cultures constituted routine growth experiment. Fluid volumes remained constant for all bottles as did cell numbers, the latter showing variation of less than 10%. Appropriate growth fluid aliquots were collected and pooled at 24-, 48- and 72-hour intervals following infection or change of medium and stored in screw cap test tubes at -20°C for future chemical or chromatographic analysis. A wild strain of virus isolated from recurrent herpetic lesions was used. This agent, following 2 passages in amniotic fluid of fertile eggs, received 12 passages on chorioallantoic (CA) membranes of 11-12 day eggs, as well as several passages in HeLa cells. Viral multiplication could be demonstrated by characteristic pock lesions on CA membranes and marked cytopathogenicity in HeLa cultures. Corneal inoculation of virus in rabbits frequently resulted in fatal encephalitis. In addition, it was possible to neutralize the virus strain with known herpes antiserum. Virus inoculum/bottle tissue culture consisted of material previously grown either in eggs or in HeLa cells. Test virus gave countable pocks at 1-2048 to 1-4096 dilution when 0.2 ml volumes were titered on egg CA membranes. Virus inocula and samples from infected bottles collected at termination of experiments were generally titrated by CA membrane technic(10); occasionally $TCID_{50}$ levels were checked in HeLa tube cultures. Ratios of 1 viral unit/800-1000 HeLa cells initially as compared to 1 viral unit/2-8 cells following 72 hours' growth could be detected by CA titrations. Cytopathic effect was frequently observed 48 hours following virus inoculation and was quite apparent at 72-hour interval. *Chemical and manometric methods.* Lactic acid(11) and glucose(12,13) determinations were made on infected and uninfected culture fluids in presence and absence of cortisone as well as on aliquots of uninoculated control media. The following modification of the Keilin and Hartree(13) glucose oxidase method, using glucostat enzymatic reagents of Worthington Biochemical Corp., proved particularly valuable in that the enzyme reagent is specific for glucose. *Test reagent.* 1 ml 1% o-dianisidine in methanol, 99 ml 0.01 M phosphate buffer pH 7.0, 125 mg glucose oxidase and 3 mg peroxidase. The mixture may be stored 1 week at 4°C or held frozen for longer intervals. *Determination.* 1) Prepare a 1.5 ml fluid volume of glucose standard (range 0 to 120 μg glucose), or samples diluted in water, as well as 1.5 ml water blank. 2) Add 3.5 ml oxidase test reagent. 3) Incubate 20 min. at 37°C. 4) Stop the reaction with 1 drop 4N HCl. 5) Read optical density at 401 μ in Beckman, or 405 on Coleman spectrophoto-

meters. Celite chromatography as described by Swim and Krampitz(14) was the method of choice for separation of organic acids released into culture fluids. Compounds were further identified qualitatively by ascending paper chromatography(15). Aerobic glycolysis was measured manometrically. Warburg cup contents were as follows: 3.5 x 10^7 HeLa cells, 25 μM glucose, 30 μM $NaHCO_3$, pH 7.6, 20 μM phosphate, 10 μM $MgSO_4$ and water to total volume of 3.2 ml.

Results. Resting cell experiments. For short term studies HeLa cells were harvested approximately 40 hours following infection so that cell destruction could be minimized, allowing intact enzyme systems, supposedly directed by virus, to function. Large amounts of virus, therefore, remained within cells when no obvious cytopathic effects were evident. The inoculum was prepared as follows: Virus was spun down from tissue culture fluids at 54,450 g for 180 min. and concentrated 6-fold by volume on resuspending in Hanks' solution containing 10% rabbit serum. In one experiment the inoculum consisted of at least 1 pock-forming viral unit/34 to 68 HeLa cells; after 43-hour growth, 1 pock-forming unit/13 cells could be measured in supernatant growth fluids by the CA technic.

Enumeration of pock lesions initiated by herpes simplex virus may yield low values. More recently, a plaque technic involving growth of virus in rabbit kidney cell system has been reported to increase sensitivity of titration 10- to 100-fold(16) over values obtained from CA titrations. Our preliminary experiments using this technic confirm these findings. However, assayed titers of microscopic plaques formed by herpes in HeLa cells, as reported by Farnham(17), do not show this increased sensitivity possibly due to differences in virus strain and host cell susceptibility. It seems possible that most, if not all, cells tested in the Warburg experiment were infected, thereby implicating both virus and cortisone as co-responsible agents for increased acid accumulation.

Fig. 1 presents manometric evidence regarding acid production by infected HeLa cells grown in presence and absence of low cortisone concentration. In aerobic Warburg experiments more CO_2 was released by infected cells grown either in presence or absence of cortisone, than by uninfected HeLa cells grown routinely in untreated Eagle's medium. Virus-infected cultures grown in presence of cortisone released considerably more CO_2 from $NaHCO_3$ buffer than infected cells grown without cortisone, suggesting formation of even larger amounts of acid or acids. Values for CO_2 evolution were slightly lower than those obtained for lactic acid production (Table I) since Warburg cup contents were heavily buffered, a concentration of 20 μM phosphate/flask being required to realize maximum activity in this system. Table I lists data obtained when lactic acid determinations were made on cup contents following completion of 80-minute total reaction interval. Under conditions of the resting-cell experiment, virus suppressed the endogenous rate while cortisone increased it. When endogenous lactic acid was subtracted from test flask values, HeLa cells previously grown in routine medium produced more lactic acid than those grown with cortisone. Herpes-infected cell cultures accumulated more lactic acid than uninfected controls, while infected cells grown

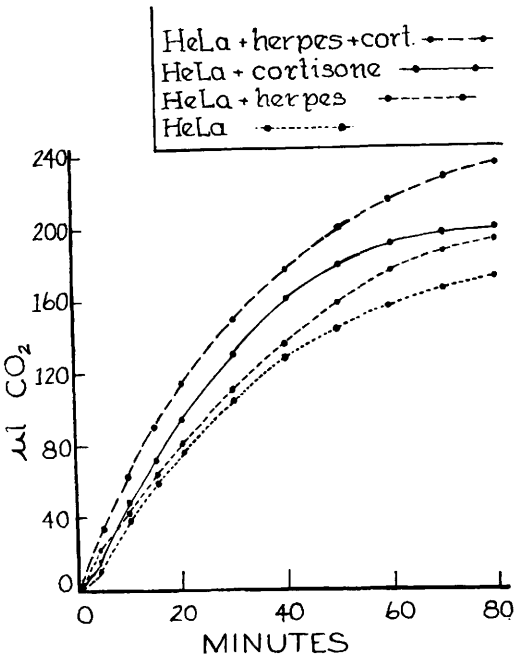

FIG. 1. Effect of cortisone and herpes simplex virus on aerobic glycolysis of HeLa cells as measured manometrically by CO_2 evolution.

TABLE I. Effect of Herpes Simplex Virus and Cortisone on HeLa Cell Glycolysis* Resting-Cell Experiments.†

Warburg flasks	μM lactic acid produced	μM lactic acid: endogenous subtracted
‡HeLa	6.8	
§ "	21.1	14.3
‡ + herpes‖	5.8	
§ + " ‖	21.1	15.3
‡ + cortisone	10.3	
§ + "	22.3	12
‡ + herpes‖ + cortisone	6.8	
§ + herpes‖ + cortisone	23.0	16.2

* Aerobic glycolysis—Warburg measurements.
† Short term: 80 min.
‡ Endogenous flask contents: 3.5×10^7 HeLa cells, 10 μM $MgSO_4$, 20 μM PO_4, 30 μM $NaHCO_3$, H_2O to 3.2 ml.
§ Test flasks: endogenous contents + 25 μM glucose.
‖ No cytopathogenicity when cells harvested 43 hr after infection. At least 1 pock-forming viral unit/13 cells in growth fluid supernates at this time.

with cortisone produced even greater quantities of lactic acid than other systems considered. Even so, accumulation of acids other than lactic acid was suspected since HeLa cells grown with cortisone released more CO_2 than could be accounted for as lactate. Also, more glucose was utilized than is required for straightforward glycolytic pathway. This will be considered later.

Growth studies. To resolve some incompatibilities, growth experiments were designed so that large samples might be taken for column chromatographic analysis. It was hoped that identification and quantitation of any accumulating organic acids might be readily measured by this method. Since there are great differences between short term resting-cell experiments and growth studies, due chiefly to processes of cellular assimilation and dissimilation, one cannot hope to approach the theoretical balance of glucose disappearance *vs.* organic acid accumulation in a given metabolic pathway. Despite this fallacy it is often possible to obtain clues regarding cellular interactions and alterations by various agents. The experiment under consideration as well as supporting studies were conducted as follows: HeLa cells were grown 7 days in presence or absence of cortisone acetate and appropriate bottles were infected with herpes virus as indicated in the methods section. One ml aliquots of growth fluids were removed from each of 4 similarly treated bottle cultures and pooled at 24-, 48- and 72-hour intervals. All cultures showed definite cytopathogenicity 48 hours following infection. After 72 hours, at least 1 viral unit/2-8 HeLa cells could be demonstrated by pock counts. Table II lists the various acids separated from these samples on celite columns and the quantities of each acid obtained when titrated with .01 N NaOH. Lactic and succinic acids were re-

TABLE II. Effects of Herpes Simplex Virus and Cortisone Acetate on HeLa Cell Metabolism-Growth Experiment.

Sample	Sampling time, hr	Microequivalents acids produced—Celite column data: Acetate	Pyruvate	Lactate & succinate
*HeLa	24	.6	.4	4.5
	48	.9	.1	4.7
	72	.7	.2	4.6
* " + herpes	24	1.0	1.0	4.6
	48‡	.9	.4	5.3
	72‡	.9	.9	5.4
†HeLa	24	.4	.3	3.5
" + herpes	48	.2	1.0	4.0
" + cortisone	72	1.0	.3	3.7
†HeLa	24	1.2	1.6	6.6
" + herpes	48‡	2.0	2.4	7.9
" + cortisone	72‡	2.8	2.5	7.0

* Cells grown and maintained in Eagle's medium.
† *Idem*, containing total conc. of 2 μg cortisone acetate/ml.
‡ Marked cytopathogenicity.

TABLE III. Influence of Herpes Simplex Virus and Cortisone on Glucose Utilization with Acid Production by HeLa Cells.

Sample	Sampling time, hr	Growth experiment: Lactic acid,† μM	Total acids* produced as μM triose	Glucose† used, μM	Theoretical μM triose obtainable from glucose
‡HeLa	48	4.1	5.7	3	6
‡ " + herpes	48	5.1	6.6	3	6
§ " + cortisone	48	3.6	5.2	2.7	5.4
§ " + herpes + cortisone	48	4.9	12.3	5.1	10.2

* Total acids: lactate, acetate, pyruvate, succinate titration values from column data.
† Lactic acid and glucose – determined by chemical analysis.
‡ Cells grown and maintained in Eagle's medium.
§ *Idem*, containing final conc. 2 μg cortisone acetate/ml.
‖ Marked cytopathogenicity.

corded together as may be observed in last column of Table II since complete separation of these acids was difficult using the chloroform-butanol solvent. Actually, lactic acid leaves the column in tubes 33 through 38, while succinic acid is removed in tubes 35 through 42. It may be emphasized that in general maximum amount of stable acids accumulated by the 48-hour interval where cytopathogenicity is definite.

Table III includes 48 hour lactic acid and glucose values as determined by chemical methods. To evaluate better previous observations, lactic acid values were subtracted from microequivalents of combined titrated lactic acid and succinic acids (Table II) listed at 48-hour testing interval. It was then possible to observe whether or not the bulk of accumulated acid was lactic or another organic intermediate such as succinic acid. The greatest difference occurred when infected HeLa cells were grown in cortisone-containing medium. Total accumulated acids (Table III) expressed as micromoles of triose were then compared with the theoretical quantity of triose obtainable from glucose. Repeated experiments bear out the quantitative data.

Further identification of various carboxylic acids accumulating in growth fluids was made employing ascending paper chromatographic technics as described previously(15). Acetate, pyruvate and lactate were accounted for in this way. Samples thought to contain succinic acid were removed from the column and lyophilized to dryness to concentrate the previously titrated, and therefore, sodium salt of the acid. The preparation was acidified with small volume of 10 N H_2SO_4 before spotting on filter papers. In some experiments permanganate oxidation was used to decompose contaminating lactic acid; this material was then lyophilized, acidified and spotted on paper chromatograms. Table IV records R_f values for the partially identified compound and various known controls. Known succinic acid controls allowed to migrate in ethanol: NH_4OH: H_2O solvent gave the following R_f values as calculated from 3 experiments: 0.15, 0.16 and 0.17. The small degree of variation may be a result of temperature fluctuation and other physical features. The unknown acid had an R_f of 0.13, 0.15 and 0.17 in the same experi-

TABLE IV. Paper Chromatographic Identification of An Unknown Acid Accumulating in Herpes-Infected Cortisone-Grown HeLa Cultures. Comparison of R_f values of known acids and the unknown compound.

Exp.	Solvent	Acid determined	R_f values: Known control	Unknown control*
1	Ethanol-NH_4OH-water†	Succinic	.16	.17
		Fumaric	.30	
		Pyruvic	.11	
		Acetic	.11	
		Malic	.22	
2		Succinic	.17	.15
		Fumaric	.33	
3		Succinic	.15	.13
		Lactic	.49	
		Pyruvic	.09	
		Acetic	.11	
4	Methanol-saturated $(NH_4)_2CO_3$‡	Succinic	.39	.39
		Lactic	.57	
		Pyruvate (Na salt)	.43	
		Acetate	.42	

* Removed from celite columns in tubes 35-42.
† 80% ethanol, 5% NH_4OH, 15% water.
‡ 75% methanol and 25% saturated $(NH_4)_2CO_3$.

ments. Other investigators(18) have reported R_f's of 0.14 for succinic acid, 0.17 for fumaric, 0.08 for α-ketoglutaric, 0.48 for lactic acid and 0.09 for malic acid. R_f values of the unknown compound agree very closely with textbook R_f data reported for either succinic or fumaric acids(18). Fumaric acid controls in Experiments 1 and 2 had R_f values of 0.30 and 0.33 which implies a considerable difference in migratory rates of our succinic and fumaric acids on paper chromatograms.

Additional evidence regarding identity of the unknown compound was available from column chromatographic titrations. The acid was removed from celite columns in maximum range of tubes 35-42 depending on concentration of the compound. When known mixtures of given quantities of organic acids were treated chromatographically in the same manner as the unidentified substance(14), fumaric acid was removed from celite in tubes 24-30 while α-ketoglutaric acid could be recovered in eluates collected in tubes 58-65. These observations tend to implicate succinic acid as the unknown compound which conspicuously accumulates in herpes-infected HeLa cultures grown in Eagle's medium containing cortisone acetate.

Additional experiments were done in which all HeLa cell cultures were grown for first 7 days in Eagle's medium alone (cortisone acetate excluded). Eight of the bottle cultures were then fed Eagle's medium containing rabbit serum and cortisone; 4 of these were infected with herpes virus. Another series of 8 cultures were fed medium containing rabbit serum but lacking cortisone and 4 of these were similarly infected. Pooled aliquots of culture fluids were collected and tested as described for previous experiments. Findings regarding accumulated organic acids and glucose disappearance in culture fluids had now a familiar trend, though very marked differences as observed in Tables II and III could not be demonstrated. Evidently early cortisone treatment of uninfected tissue cultures followed by infection with herpes simplex virus plays a significant role in accentuating metabolic alterations.

Discussion. HeLa cells infected with herpes virus produce more lactate than uninfected cells. When infected cells are grown in medium containing cortisone, acid accumulation is marked. Large quantities of lactate and succinate appear as well as increased amounts of acetate and pyruvate. During the phase when increase in viral units may be demonstrated, stimulation of metabolic processes also occurs particularly if cortisone acetate, in concentration of 2 μg/ml, has been included in the growth medium. In the latter system more acid appears than can be accounted for by glucose utilization indicating that a portion of the accumulated acids results from dissimilation of other compounds, possibly proteins or amino acids. Obviously, lactic acid production is not always a valid indicator of glucose utilization. Why succinate or a very close chemical relative of this compound appears is not known but one may hypothesize and tentatively suggest some interesting alternatives. Perhaps an active decarboxylase enzyme removes CO_2 from a less stable precursor leaving succinate as end product; or, it may be that a specific enzyme system is blocked at this level. Of even greater interest is the question concerning transamination reactions such as the well known examples where glutamic acid plus oxaloacetic or pyruvic acids yield α-ketoglutaric acid plus aspartic acid or alanine. These possibilities have been mentioned by Rosen and Roberts(9).

There is no doubt that virus is implicated in the biochemical alterations; the extent of these changes is uncertain. Heavy inocula were used throughout to insure infection of the majority of HeLa cells. Much more virus could be detected at termination of each experiment than was introduced initially and marked cytopathogenicity was apparent at the conclusion of each growth experiment. Even so, one does not know the absolute percentage of infected cells in the culture, or the number of completely formed infectious viral units present in the inoculum. It would be of great interest to implicate conclusively viral replication with biochemical change. Because of the many unknowns the possibility of incomplete viral effect in conjunction with multiplication of infectious particles must at the present be considered. Whether the observed effect is due to infectious virus alone or a mix-

ture of incomplete and infectious virus remains to be studied.

Summary. HeLa cells infected with herpes simplex virus produced more organic acids than uninfected control cultures when glucose was the principal substrate. The organic acids which accumulated in growth fluids were separated by celite chromatography and further identified by ascending paper chromatography. Infected cells grown with cortisone formed as much lactic acid as infected cells grown in absence of cortisone, but in addition, the former cells produced large amounts of acetate, pyruvate and another compound tentatively identified as succinate. Glucose utilization was stimulated by virus and cortisone in the HeLa cell system resulting in accumulation of Krebs cycle intermediates. Concomitant with marked acid production definite cytopathogenicity and marked increases in virus titers occurred in the virus-infected cultures. It may be concluded that the herpes virus-cortisone association alters carbohydrate metabolism of HeLa cells in a striking pattern.

1. Crossfeld, H., *Science,* 1958, v127, 148.

2. Gavosto, F., Pileri, A., Brusca, A., *Biochem. et Biophys. Acta,* 1957, v24, 250.

3. Rosen, F., Roberts, N. R., *Science,* 1958, v127, 287.

4. Kass, E. H., Lundgren, M. M., Finland, M., *J. Exp. Med.,* 1954, v99, 89.

5. Kilbourne, E. D., Horsfall, F. L., Jr., PROC. SOC. EXP. BIOL. AND MED., 1951, v77, 135.

6. Schwartzman, G., *ibid.,* 1952, v79, 573.

7. Sigel, M., Beasley, A. R., *ibid.,* 1955, v88, 86.

8. Southam, C. M., Babcock, V. I., *ibid.,* 1951, v78, 105.

9. Syverton, J. T., Garabedian, G. A., Friedman, J., *J. Infect. Dis.,* 1955, v96, 9.

10. Buddingh, G. J., *Viral and Rickettsial Infections of Man,* 1952, J. B. Lippincott Co., Philadelphia, Chap. 5.

11. Hullin, R. P., Noble, R. L., *Biochem. J.,* 1953, v55, 280.

12. Park, J. T., Johnson, M. V., *J. Biol. Chem.,* 1943, v147, 415.

13. Keilin, D., Hartree, E. F., *Biochem. J.,* 1948, v42, 230.

14. Swim, H. F., Krampitz, L. O., *J. Bact.,* 1954, v67, 419.

15. Fisher, T. N., Ginsburg, H. S., PROC. SOC. EXP. BIOL. AND MED., 1957, v95, 47.

16. Kaplan, A. S., *Virology,* 1957, v4, 435.

17. Farnham, A. E., *ibid.,* 1958, v6, 317.

18. Block, R. J., Durrum, E. L., Zweig, G., *Paper Chromatography and Electrophoresis,* 2nd Ed., 1958, Academic Press, N. Y., p233.

Received December 29, 1958. P.S.E.B.M., 1959, v100.

Effect of 1-(3-Hydroxyphenyl) 2-Methylaminoethanol (Phenylephrine) and 1-(3-Hydroxyphenyl) 2-Aminoethanol (Norphenylephrine) on Intestinal Motility. (24778)

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Norphenylephrine (differing from phenylephrine in that it has a hydrogen atom instead of a methyl group on the nitrogen atom) has not been fully investigated with regard to its effects on smooth muscle of the type which is inhibited by epinephrine. Kuschinsky and Oberdisse(1) reported that norphenylephrine has an inhibitory action on rat uterus with a potency 1/10 that of epinephrine, and Boyd (2) reported that norphenylephrine has an excitatory action on rabbit uterus. According to Lands(3) norphenylephrine is inactive as a bronchodilator. As an inhibitor of the peristaltic reflex in the guinea pig ileum phenylephrine and norphenylephrine were found to be 1/5000 and 1/6000 as potent, respectively, as epinephrine, while they were 1/10 and 1/50 as potent as epinephrine in causing inhibition of the "pendular movements" in the isolated rabbit ileum(4). In the present study a comparison is made between the rabbit intestine-inhibiting actions of phenylephrine and norphenylephrine; and differences in potency of these compounds are found to be much less