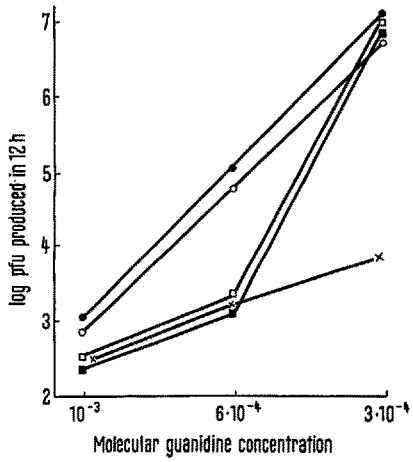

Fig. 1. Pattern of the antiguanidine effect of different compounds. Challenging a constant dose of antagonist against increasing guanidine concentrations. x——x guanidine; ●——● guanidine + L-methionine; o——o guanidine + L-leucine; □——□ guanidine + choline; ■——■ guanidine + ethanolamine. The concentration of the antagonists was $3.3 \times 10^{-4} M$. pfu, plaque forming units.

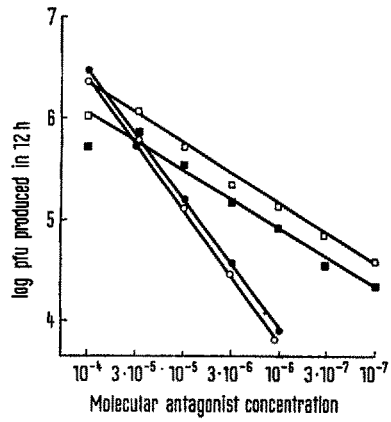

Fig. 2. Pattern of the antiguanidine effect of different compounds. Challenging increasing concentrations of each antagonist against a constant dose of guanidine. ●——● guanidine + L-methionine; o——o guanidine + L-leucine; □——□ guanidine + choline; ■——■ guanidine + ethanolamine. The guanidine concentration was $3.3 \times 10^{-4} M$. pfu, plague forming units.

Conversely, it is possible that guanidine might exert its antipolio action by reacting with the same structures present in an amino acid or in an amino alcohol essential for poliovirus growth (during replicase synthesis) but not for culture cells.

Consequently, a potential antipolio agent might be found among some analogs of those amino acids which most actively antagonize guanidine. It is very suggestive that a valine analog, D-penicillamine, inhibits very effectively and selectively polio virus growth[6,7].

Riassunto. L'inibizione da parte della guanidina sulla crescita del virus polio può essere antagonizzata da amino-acidi e amino-alcoli. Viene definita la struttura responsabile dell'effetto antiguanidinico negli amino-acidi attivi. Una struttura simile è presente anche negli amino-alcoli attivi. I risultati ottenuti suggeriscono che i composti antiguanidinici interagiscono con la molecola guanidinica sottraendola dai recettori connessi con l'effetto antivirale.

B. Loddo, G. L. Gessa,
A. Tagliamonte and W. Ferrari

Institutes of Pharmacology, University of Cagliari and Modena and Institutes of Hygiene, Virology and Microbiology, University of Cagliari (Italy),
24 April 1967.

[6] G. L. Gessa, B. Loddo, G. Brotzu, M. L. Schivo, A. Tagliamonte, A. Spanedda, G. Bo and W. Ferrari, Virology *30*, 618 (1966).

[7] This work was supported by the Consiglio Nazionale delle Ricerche, Roma.

The Influence of Chlorothiazide and Tolbutamide upon Intestinal Serotonin Levels in the Sprague-Dawley Rat

Rats pretreated with sulfamerazine and several antibiotics develop increased serotonin levels in some areas of the gastrointestinal tract[1-3]. The mechanism(s) of this increase is unknown, but it has been suggested that lumenal sterilization is responsible[2,3]. There are 2 possible mechanisms by which lumenal organisms could alter tissue serotonin levels. Firstly, amines may undergo an enterohepatic circulation with subsequent destruction by intestinal bacteria[4]; and secondly, bacterial utilization of dietary amino acids may reduce the availability of these amine precursors for tissue decarboxylase. However, it seems unlikely that the elevated serotonin levels observed following sulfamerazine are due primarily to an antibacterial effect because, the addition of sulfasuxidine to the diet leads to a greater recovery of intestinal serotonin in tryptophan deficient rats compared to controls[5]. Furthermore, serotonin levels following sulfamerazine are not uniformly elevated in those bowel areas normally inhabited by micro-organisms[1], and in man, urinary tyramine and tryptamine have been shown to be of tissue rather than of bacterial origin[4]. In order to study the possibility that the increased mucosal serotonin levels following sulfamerazine are due to a non-specific effect

[1] J. H. Thompson and L. B. Campbell, Nature *212*, 850 (1966).
[2] R. S. Stacey and T. J. Sullivan, J. Physiol. *137*, 63P (1957).
[3] T. J. Sullivan, Br. J. Pharmac. *16*, 90 (1961).
[4] V. L. DeQuattro and A. Sjoerdsma, Clinica chim. Acta *16*, 227 (1967).
[5] E. M. Gal and P. A. Drewes, Proc. Soc. exp. Biol. Med. *110*, 368 (1962).

of the basic 'sulfonamide' nucleus, the concentration of serotonin has been determined in several intestinal areas following pretreatment with the chemically related drugs, chlorothiazide and tolbutamide.

Male Charles River rats weighing between 280–310 g were randomly divided into 3 groups of 15 animals, housed in colony cages, and fed powdered Purina rat chow with the tryptophan content of 0.22%. Group 1 (control rats) received pure chow. Groups 2 and 3 received 30 mg chlorothiazide (Merck, Sharpe and Dohme) or tolbutamide (Upjohn)/20 g chow. All drugs were continued for 15–25 days and food consumption and weight gain were normal in all groups.

The rats were killed by decapitation between 08.30 and 10.00 on the day of assay, and half inch segments of the upper duodenum, mid-jejunum, terminal ileum and appendix were rapidly removed and prepared as previously described[6]. Serotonin was assayed by the method of BOGDANSKI et al.[7]. Data are presented as mean values ± 1 standard error, and differences between mean values were determined using the *t* test. Results are indicated in the Table.

The serotonin concentration in μg/g mucosa of control and drug treated rats

Tissue	Control	Chlorothiazide	Tolbutamide
Upper duodenum	6.6 ± 0.3	6.2 ± 0.5	6.9 ± 0.3
Mid-jejunum	3.6 ± 0.2	3.6 ± 0.3	3.8 ± 0.2
Terminal ileum	4.2 ± 0.4	4.2 ± 0.2	4.6 ± 0.3
Appendix	14.3 ± 0.09	14.0 ± 0.4	14.7 ± 0.5

Data are expressed as means ± 1 standard error. There are 15 animals/group.

There were no significant differences in the serotonin levels from comparable bowel areas examined between the control group and those rats receiving chlorothiazide or tolbutamide. Thus, it is apparent that the elevated mucosal serotonin levels previously observed in the gastrointestinal tract following sulfamerazine[1] are not related to a non-specific effect of the basic 'sulfonamide' nucleus[8].

Zusammenfassung. Der Serotoningehalt der Schleimhaut im obern Duodenumteil, im mittleren Jejunum, im Ileumendteil und im Appendix von Sprague–Dawley Charles River Rattenmännchen wurde ohne Vorbehandlung und mit Chlorothiazid- und Tolbutamidvorbehandlung spektrophotofluorimetrisch bestimmt. Die Serotoningehalte für die 3 Gruppen vergleichbarer Gewebe waren ähnlich, was zeigt, dass die früher beschriebene Erhöhung des Serotoningehaltes der Schleimhaut nach Sulfamerazinvorbehandlung wahrscheinlich nicht mit einer unspezifischen Wirkung des Sulfonamidteils verbunden ist.

J. H. THOMPSON, CH. SPEZIA and M. ANGULO

Department of Pharmacology, U.C.L.A. School of Medicine, Los Angeles (California 90024, USA), 31 July 1967.

[6] J. H. THOMPSON and L. B. CAMPBELL, Ir. J. med. Sci. *490*, 411 (1966).
[7] D. F. BOGDANSKI, A. PLETSCHER, P. A. SHORE and B. B. BRODIE, J. Pharmac. exp. Ther. *117*, 82 (1956).
[8] This study was supported in part by an award from the National Science Foundation to J. H. THOMPSON, M.D., M.R.C.P.I., Department of Pharmacology, U.C.L.A. School of Medicine, Los Angeles, California 90024.

Comparison of the Cell Cycle and Cell Migration in the Intestinal Epithelium of Suckling and Adult Mice

It has been recently demonstrated that the rate of cell migration is considerably slower in the small intestinal epithelia of suckling rats than in adult animals. This difference was attributed to a slower rate of cellular proliferation and division in the younger rats[1]. However, studies on the age changes in the mean duration of the duodenal cell cycle have shown that the cycle is shortest in suckling mice and gradually lengthens with increasing age[2,3]. Therefore, this investigation was undertaken in an attempt to show that the difference in cell migration is assignable to rapid postnatal growth rather than to great dissimilarities in the cell cycle and proliferative rates.

Methods. Two groups of male Swiss albino mice, aged 10 days (suckling) and 1 year (adult), were utilized. The animals were sacrificed in pairs from $^1/_2$ to 48 h after a single dorsal s.c. injection of tritium thymidine (H^3-T), specific activity 6.4 C/m*M*, methyl labeled, New England Nuclear Corp., at a concentration of $^1/_2$ μc/g body weight. The duodenum was removed, fixed and processed for autoradiography as previously described[4].

The cell cycle for each group was obtained from constructed labeled metaphase curves[2]. The rate of cell migration was determined by measuring the percentage of the villus height (total of 10 villi) that labeled daughter cells had reached at $^1/_2$, 8, 12, 24 and 48 h after H^3-T administration. The average cryptal size (total of 20) and villus height (total of 20) were estimated with a 50-division ocular grid and a B & L stage micrometer. The values were then converted to millimeters (mm). In the analyses of cell migration and sizes, both the crypto-villal junction and extrusion zone were present for villus measurements[5], and measured crypts were longitudinally sectioned[3]. An estimate of cell proliferation was assessed by scoring the number of labeled nuclei/1000 cells in the lamina propria and the cryptal epithelium at $^3/_4$ h after H^3-T injection.

Results. The mean duration of the cell cycle was 12 h in the suckling mice and 14 h in the adult animals. The

[1] O. KOLDOVSKY, P. SUNSHINE and N. KRETCHMER, Nature *212*, 1389 (1966).
[2] J. D. THRASHER and R. C. GREULICH, J. exp. Zool. *159*, 39 (1965).
[3] J. D. THRASHER and R. C. GREULICH, J. exp. Zool. *159*, 385 (1965).
[4] J. D. THRASHER, in *Methods in Cell Physiology* (Ed. D. M. PRESCOTT; Academic Press, New York 1966), Vol. 2, p. 323.
[5] M. R. LORAN and T. L. ALTHAUSEN, J. biophys. biochem. Cytol. 7, 667 (1960).