TABLE II—RESULTS OF FUNGICIDAL TESTING

Compd. No. or Name	No. of Tubes Exhibiting Growth After 25 Days[a] 0.1%	0.5%	1.0%
1	10	8	6
2	10	10	10
3	4	4	3
4	6	4	4
5	4	4	0
6	6	4	2
7	8	6	6
8	4	4	4
Diphenolic acid	10	3	3
Undecylenic acid	8	4	2
10-Undecenoic hydroxamic acid	2	0	0

[a] The maximum number of tubes exhibiting growth for each concentration is 10.

CONCLUSION

From Table II it can be seen that 10-undecenoic hydroxamic acid exhibited the greatest amount of fungicidal activity. Propyl 4,4-bis(3,5-dichloro-4-hydroxyphenyl) pentanoate was the next most active fungicidal compound tested. Six of the ten compounds tested exhibited greater activity than undecylenic acid under the conditions of the test.

The undecylenic hydroxamic acid, because of its activity against *T. mentagrophytes*, was further tested and showed good activity against the following organisms: *Microsporum audouini* ATCC 9079, *Microsporum canis* ATCC 10241, *Trichophyton mentagrophytes (gypseum)* ATCC 9129, *Trichophyton mentagrophytes (interdigitale)* ATCC 9972, *Candida albicans* ATCC 10231, *Trichophyton rubrum* ATCC 10281, *Stemphylium solani* ATCC 11128, and *Stemphylium species* ATCC 9569.

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Activity of Different Segments of Rabbit Gastrointestinal Tract

By THEODORE H. EICKHOLT, ROGER H. BOX, and NOLAN D. COURVILLE

Five different segments of the rabbit intestinal tract, (duodenum, jejunum, ileum, ascending and descending colon), were used to record the normal activity of these segments using an isolated muscle bath technique. Kymograph recordings and transducer-recorder recordings were compared. Contractions per minute, interval between contractions, and amplitude of contractions were observed and evaluated. In the transducer recorded series, *t* tests indicated statistically significant difference for all comparisons of contractions per minute. From 60 comparisons of normal rabbit intestinal activity using *t* tests, only 5 comparisons were not found to be significantly different at *P* 0.05.

THE WEIGHTS and linear measurements of several segments of the gastrointestinal tract of the rabbit have been investigated by Latimer and Sawin (1). Other tissues and organs have been studied (2–5), but the variations in the normal activity of different segments of the rabbit gut so often employed in drug testing and drug experimentation require delineation.

It was, therefore, the purpose of this study to record some of the variations observed in the different segments of the rabbit intestine and subject them to statistical evaluation for reference when a similar preparation was used experimentally. Two systems, typical kymograph and transducer systems, were employed to further delineate differences in observations of activity of rabbit intestinal segments.

EXPERIMENTAL

Adult, albino rabbits were anesthetized with ether and 2–3 cm. segments of duodenum, jejunum, ileum, ascending and descending colon were expediently removed. The segments were individually maintained in oxygenated Tyrode's solution in a constant-temperature bath at 38° prior to their placement in the isolated muscle bath. The Tyrode's solution contained sodium chloride 0.8%, potassium chloride 0.02%, calcium chloride 0.02%, magnesium chloride 0.01%, sodium bicarbonate 0.1%, sodium diphosphate 0.005%, and glucose 0.1%.

The first series of experiments utilized constant-temperature muscle baths and kymographs. Tyrode's solution (100 ml.) was used to bathe the segment; temperature was maintained at 38° and the preparation was oxygenated using an air flow bubbled through the solution. The segment was attached to the muscle hooks, one stationary and the other being connected to a lever and stylus, such that the segment was completely submerged in the Tyrode's solution. The muscle lever was made to record on a slowly rotating carbon black kymograph drum. A synchronous timer recorded the time on the same tracing. The only constant tension placed upon the segment was that of the weight of the lever which in turn was centered on its fulcrum and

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raised only until the attachment *via* muscle hooks was tight enough to transfer the segment's movement to the lever and recording stylus.

The conditions for the second series of experiments for comparison were the same as above except intestinal activity was recorded *via* an E & M physiograph isotonic myograph transducer on an E & M physiograph. A timer was an integral part of the recorder and the paper speed was 0.05 cm./sec. The only constant tension placed upon the segment was that of the weight of the movable armature (2.2 Gm.) and of the lever which in turn was centered on its fulcrum and raised only until the attachment *via* muscle hooks was tight enough to transfer the segment's movements to the transducer and recording physiograph. In both systems relaxation was attempted with minimal stretching to best approximate normal conditions.

The myograph had amplitude and calibration controls and all observed pen deflections were equilibrated to the standard pen deflection displacements.

In each experiment, the segments were allowed to acclimate to the muscle bath environment. Acclimation was determined by consistency and/or rhythmicity of contractions.

Following acclimation in both series of experiments, normal intestinal activity for the different intestinal segments was recorded. Five to seven different rabbits were used to provide different individual samples of duodenum, jejunum, ileum, ascending and descending colon. Tracings of a minimum of 15 min. were obtained for each of the 5 different segments in each series. From each tracing of each individual gut segment, amplitude of contraction, interval between contractions, and contractions per minute were measured. Three separate measurements of the three parameters above were made on each tracing providing an average of the three for that particular parameter of that single gut segment. These averages were in turn averaged for the 5 to 7 samples of each individual gut segment. This provided 15 to 21 actual measurements for the averages of the three parameters. Standard deviations were calculated and t tests were made between all possible combinations of intestinal segments.

RESULTS

Table I summarizes the averages and standard deviations of the amplitude of contraction, interval between contractions, and contractions per minute obtained in the first series of experiments recording on the smoked kymograph.

The t test values from the kymograph recorded series for all combinations of intestinal segments on the three parameters, contractions per minute, interval between contractions, and amplitude of contractions, indicated significant differences at P 0.05 for 28 of the total 30 comparisons.

Table II summarizes the averages and standard deviations of the amplitude of contraction, interval between contractions, and contractions per minute obtained in the second series of experiments recording with the myograph transducer.

The t test values from the transducer recorded series for all combinations of intestinal segments on the three parameters indicated significant differences at P 0.05 for 27 of the total 30 comparisons.

SUMMARY AND CONCLUSIONS

Five different segments of the rabbit intestinal tract, duodenum, jejunum, ileum, ascending and descending colon, were utilized in recording normal activity of these segments using an *in vitro* isolated muscle bath technique. One series of experiments was recorded using a typical smoked kymograph, while the second series for comparison employed an isotonic myograph transducer and multichannel recorder.

Contractions per minute, interval between contractions, and amplitude of contractions were recorded, determined, averaged, standard deviations calculated, and t tests performed on all possible segment combinations. In the kymograph recorded series, the contractions per minute decreased with progression to the lower segments of the intestinal tract. The interval between contractions increased which would correlate with the decreased contractions per minute. All t values for comparing

TABLE I—SUMMARY OF AVERAGES AND STANDARD DEVIATIONS OF MEASUREMENTS OF RABBIT INTESTINAL ACTIVITY OBTAINED FROM KYMOGRAPH RECORDINGS

	No. of Samples	Contractions/min.	Interval Between Contractions, sec.	Amplitude of Contraction, mm.
Duodenum	6	16.9 ± 2.01	3.1 ± 0.6	22.0 ± 7.40
Jejunum	6	13.8 ± 1.63	3.7 ± 0.7	36.1 ± 20.10
Ileum	6	13.3 ± 3.21	4.6 ± 1.1	12.0 ± 7.55
A. colon	6	6.2 ± 3.97	6.0 ± 4.1	2.8 ± 1.84
D. colon	6	4.9 ± 1.56	10.0 ± 4.3	19.5 ± 1.98

TABLE II—SUMMARY OF AVERAGES AND STANDARD DEVIATIONS OF MEASUREMENTS OF RABBIT INTESTINAL ACTIVITY OBTAINED FROM TRANSDUCER RECORDINGS

	No. of Samples	Contractions/min.	Interval Between Contractions, sec.	Amplitude of Contraction, mm.
Duodenum	7	16.7 ± 1.68	3.9 ± 0.1	2.56 ± 2.50
Jejunum	7	12.4 ± 2.31	5.0 ± 1.5	6.53 ± 3.46
Ileum	7	10.5 ± 1.27	6.2 ± 1.1	9.17 ± 0.68
A. colon	5	2.1 ± 0.83	15.0 ± 8.2	1.68 ± 0.57
D. colon	5	4.3 ± 1.68	13.4 ± 5.0	3.91 ± 2.38

all possible combinations of intestinal segments showed statistically significant differences at P 0.05 except two. The jejunum–ileum and A. colon–D. colon contractions per minute comparisons were not significantly different; however, the two other adjacent segments comparisons here were significantly different.

In the transducer recorded series, the contractions per minute again decreased with progression to the lower intestinal tract and the intervals between contractions again increased. The A. colon was the only exception to this trend in these two cases and may be due to more sensitive recording.

In this series the t values indicated statistically significant differences for all comparisons of contractions per minute which would be very important for drug comparison effects upon the intestinal tract.

The comparison of interval between contractions between A. colon and D. colon, however, was not significantly different. The reverse was true in the kymograph series in the comparisons between jejunum–ileum and A. colon–D. colon; the interval between contractions was significant but the contractions per minute were not significantly different. The transducer system would appear to better record the contractions per minute.

Two comparisons of amplitude of contractions in the transducer series also were not significantly different. They were duodenum–A. colon and duodenum–D. colon.

It would appear that this study and evaluation of normal rabbit intestinal activity could serve as a basis for further drug activity studies employing the rabbit intestinal tract and furthermore, in determining the preferential intestinal segment of a drug's activity.

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Inhibition of Cortisone-Induced Cleft Palate in Mice by Cobaltous Chloride

By GILBERT KASIRSKY, RONALD F. GAUTIERI, and DAVID E. MANN, JR.

Young primipara CF-1 mice were mated overnight and those with vaginal plugs (day zero) were considered pregnant. Animals were divided into five groups of 26 each and subjected to the following treatment: group *A* received 2.5 mg. of cortisone acetate i.m. on days 11, 12, 13, and 14; group *B* received cobaltous chloride, 25 mg./Kg. i.p. on day 11 only followed by cortisone acetate, 2.5 mg. i.m. on days 11, 12, 13, and 14; group *C* received physiological saline, 0.1 ml. i.m. on days 11, 12, 13, and 14; group *D* represented untreated controls; group *E* received cobaltous chloride, 25 mg./Kg. i.p. on day 11 only followed by saline, 0.1 ml. i.m. on days 11, 12, 13, and 14. Mice were laparotomized and fetuses were examined on day 18. Group *A* (cortisone), 140 of 185 fetuses developed cleft palate (75.6 per cent incidence); group *B* (cobaltous chloride–cortisone), 24 of the 190 fetuses had clefts (12.6 per cent incidence); group *C* (saline), none of the 201 fetuses showed clefts; group *D* (untreated controls), none of the 194 fetuses had clefts; group *E* (cobaltous chloride–saline), 27 of the 208 fetuses had cleft palate (12.9 per cent incidence). Also, the incidence of cleft palate by litters was group *A*, 100 per cent; group *B*, 23.1 per cent; group *C*, 0 per cent; group *D*, 0 per cent; group *E*, 50 per cent.

DURING A recent evaluation of the relative antitumorigenic properties of cortisone (1), cobaltous chloride (2), and sodium cobaltinitrite (3, 4) in CF-1 mice subjected to the minimal carcinogenic dose$_{50}$ (MCD$_{50}$) of methylcholanthrene, several striking similarities became apparent in the character of the histological responses evoked by each drug, for not only was there a significant reduction of tumor incidence, but also the remarkable absence of cutaneous inflammation. The latter observation may be related to a report of an accentuation of anti-inflammatory activity of δ-1 hydrocortisone by the addition of a cobalt atom to the C-20 carbonyl function (5). Because interference with normal closure of the palatine shelves in certain strains of mice by cortisone is well documented (6), and furthermore, because previous studies in this laboratory have uncovered certain cortisone-like characteristics of two inorganic cobalt compounds, it thus became the primary objective of this investigation to discern the nature and extent of any cobalt–steroid interactions in terms of greater or lesser anomalies in the mouse fetus.

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EXPERIMENTAL

Groups of 30 female mice of the CF-1 strain, weighing approximately 25 Gm., were individually