soever. Upon cessation of breathing ammonia, the NPN drops regularly to the normal value, but at a slower rate than it was absorbed. It was thought at first that there would be an increase in the urea content of the blood through conversion of ammonium carbonate into urea, but such does not seem to be the case. An examination of the urine for excess urea and ammonium salts over the normal gave no further clue. The amount of increase in the urea or ammonium salt formed by the ammonia inhaled would be insignificant compared to the normal amount excreted in a 24-hr sample.

TABLE 2

CARBON DIOXIDE-COMBINING POWER OF THE BLOOD PLASMA

Time (min)	Volume (% CO_2)
Series I	
0	56
120	57
180	57
Series II	
0	52
90	48
Series III	
0	52
60	53
120	50

The carbon dioxide-combining power of the blood plasma apparently is not impaired by the accumulation of ammonia, as is illustrated in Table 2.

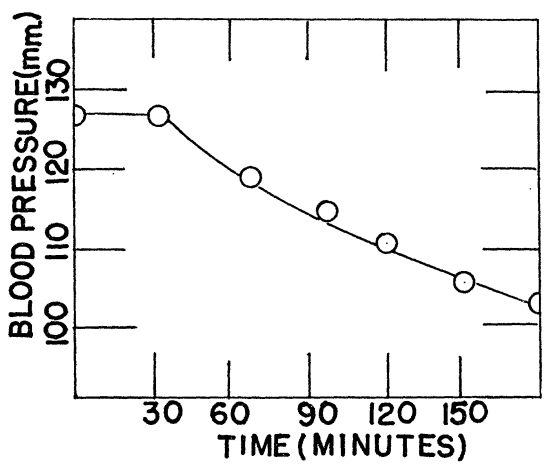

FIG. 2. Change in blood pressure with time upon exposure to gaseous ammonia.

The pulse rate, determined at regular intervals during each experiment, was found to be constant. Blood pressures taken on the subject showed a regular drop after the first 35 min of inhalation. A typical determination is shown in Fig. 2.

No attempt is made to explain the mechanism of the absorption of ammonia by the blood. The authors feel sure, however, that it is a chemical process and not a physical one. It seems to be a second-order reaction. The data are presented in the hope that more work will be done on this problem.

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Inhibition of Mitotic Poisoning by *meso*-Inositol[1]

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The studies briefly reported here stem from several observations recorded in the literature. The strong insecticidal agent, γ-hexachlorocyclohexane (Gammexane) has a cytological effect on *Allium Cepa* similar to that of colchicine. It produces the phenomenon known as c-mitosis, which is characterized by the arrest of nuclear division in the metaphase and, on longer exposure, formation of c-tumors. Other isomeric hexachlorocyclohexanes are either ineffective or only slightly active (*5*). The

TABLE 1

TUMOR FORMATION

Medium* (mM/liter)		c-Tumors†	Medium* (mM/liter)		c-Tumors†
Mitotic poison	Inhibiting agent		Mitotic poison	Inhibiting agent	
0.025 C	...	− (2)	0.035 G	...	+ (4)
0.25 C	...	+ (4)	0.35 G	...	+ (3)
0.25 C	0.33 mI	± (2)	0.035 G	0.33 mI	± (3)
0.25 C	3.3 mI	− (6)	0.035 G	3.3 mI	− (5)
0.25 C	2.0 S	+ (2)	0.35 G	3.3 mI	± (3)
0.25 C	3.3 dI	+ (2)	0.035 G	3.3 dI	+ (2)
....	...	− (5)	0.035 G	2.0 S	+ (3)
....	0.66 mI	− (2)	....	0.25 S	− (2)

* C = colchicine; G = Gammexane; mI = *meso*-inositol; dI = *d*-inositol; S = D-sorbitol.

† + = tumors on all roots; ± = small tumors on some of the roots; − = no tumors. The figures in parentheses indicate the number of bulbs examined.

[1] This work was supported in part by a grant from the American Cancer Society on the recommendation of the National Research Council Committee on Growth.

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same agent interferes with nuclear division in *Paramecium caudatum* and produces distorted forms of growth (*3*). Regardless of the exact sterical configuration of the various hexachlorocyclohexanes, they are all analogues of the corresponding hexahydroxy compounds,

For the study of the effects on mitosis, 10 bulbs with a large number of roots were selected. From 2 to 5 roots were removed from each bulb for each of the 16 individual experiments recorded in Table 2. All roots were exposed for 4 hrs at 19° C to mixtures of the various

TABLE 2

CYTOLOGICAL EFFECTS*

Media (mM/-liter)	Experiment No.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
	Mitotic poison	0.25 C	2.5 C	0.25 C	0.25 C	2.5 C	0.25 C	0.25 C	0.035 G	0.35 G	0.035 G	0.35 G	0.35 G	—	—	—	—
	Inhibiting agent	—	—	3.3 mI	33 mI	33 mI	33 S	33 dI	—	—	3.3 mI	33 S	33 dI	33 mI	33 S	33 dI	—
Bulb No.		Per cent affected metaphases per root															
I		82	90	18	24	94	72	100	74	94	26	86	90	2	4	14	10
II		76	94	12	22	82	18†	94	84	82	20	90	94	6	2	12	8
III		80	96	6	14	72	86	96	86	92	14	84	86	10	6	10	6
IV		82	86	12	10	18	84	88	84	92	18	86	84	6	10	16	12
V		94	94	20	6	30	84	90	86	90	20	96	86	12	4	6	10
VI		80	92	16	6	18	82	92	80	94	14	94	94	10	12	4	6
VII		76	92	20	10	30	78	94	82	90	10	90	90	6	10	6	10
VIII		86	96	18	2	18	80	90	78	96	10	96	96	12	14	4	4
IX		86	90	18	10	66	86	96	82	86	18	90	94	12	4	10	8
X		84	94	16	6	16	78	90	84	96	18	94	90	4	6	6	8
Average		83	92	16	11	44	75	93	82	92	17	91	90	8	7	9	8

* See Table 1 for abbreviations used.

† Other roots of the same bulb showed c-mitosis.

the inositols, which are of great biological interest. The remarkable specificity of *meso*-inositol as an essential agent in the growth of microorganisms has been discussed repeatedly (*6, 9*). The important specific influence of the relative sterical positions of the substituent hydroxyls in the various inositols was demonstrated recently with respect to the enzymatic oxidation of these stereoisomers (*4*).

It has now been found that *meso*-inositol is able to inhibit the metaphase arrest and tumor formation induced in *Allium Cepa* by colchicine or by Gammexane. This effect appears to be specific; *d*-inositol and D-sorbitol are inactive.

Onion bulbs were placed with their bases resting on the rims of glass jars filled with tap water. Roots approximately 2.5 cm long were formed in 3 days. For the observation of tumor formation, the entire bulbs with roots still attached were transferred to jars containing the various mixtures of mitotic poisons and substances being tested as inhibiting agents and kept at 18°–25° C for 48–72 hrs (Table 1). Colchicine, U.S.P. (Inland Alkaloid Co., Tipton, Indiana, or Merck & Co.), and the substances tested as inhibiting agents were employed as aqueous solutions. γ-Hexachlorocyclohexane (obtained through the courtesy of Dr. J. P. Baxter, Imperial Chemical Industries, Ltd., Widnes, England) was made up as a 0.033 M solution in ethanol and diluted with tap water to the desired concentrations.

mitotic poisons and inhibiting agents in small vials. The roots were then killed with absolute ethanol and acetic acid (3:1), and propiono-carmine smears were prepared for microscopic examination. The first 50 metaphases were counted and classified. All metaphases in which the chromosomes were split, but still attached at the centromere, were considered as affected, *i.e.* as c-mitotic (see Figs. 1 and 2).

Whereas γ-hexachlorocyclohexane, because of chemical similarities, could have been suspected of acting as a biological antagonist of *meso*-inositol, this does not apply to colchicine. Even in the case of Gammexane the evidence is not entirely clear. In some instances *meso*-inositol has been reported to overcome the inhibition of microbial growth by this agent (*1, 2*). In other biological systems, however, no such effects were observed (*7, 8*). It is possible that these discrepancies would disappear if different amounts and proportions were tested. That the ratio of mitotic poison to inhibiting agent is not the only factor is shown by a comparison of Experiments 3 and 5 in Table 2.

It is too early to speculate on the mechanism of the remarkable cytological effect of *meso*-inositol reported here. Additional poisons of mitosis, including those affecting other phases of nuclear division, will have to be examined before the conclusion that *meso*-inositol is involved specifically in the metaphase, or perhaps in the formation of the spindle, is justified. It is not unlikely

that the cellular component with which colchicine and Gammexane interfere is not *meso*-inositol itself, but a

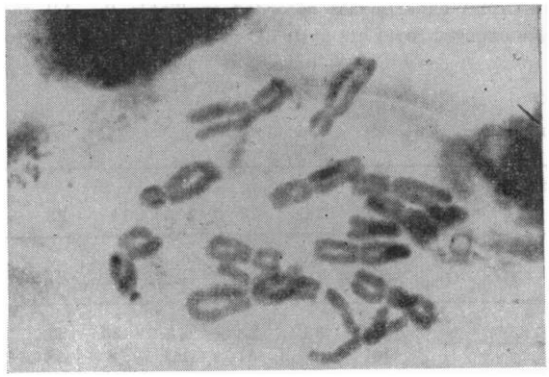

FIG. 1. Metaphase in *Allium Cepa* treated with 0.00035 M Gammexane and 0.033 M *d*-inositol solution, showing failure of the *d*-inositol to inhibit c-mitosis. (Magnification, approximately 1,000 ×.)

substance to which it gives rise and the formation of which is prevented by the mitotic poisons in question.

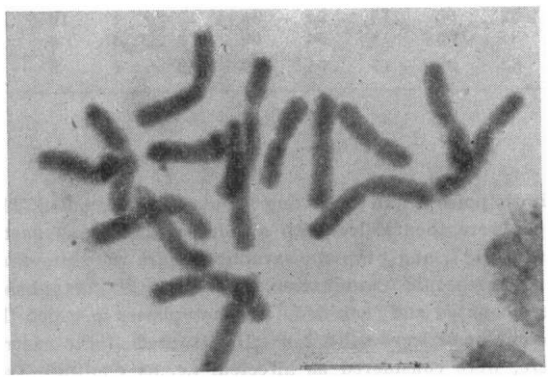

FIG. 2. *Allium Cepa* treated with 0.000035 M Gammexane and 0.0033 M *meso*-inositol solution, showing inhibition of c-mitosis. (Magnification, approximately 1,000 ×.)

Experiments on these and other aspects will form the subject of a detailed communication which will appear at a later date.

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Biosynthesis of Radioactive Drugs Using Carbon 14[1]

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The therapeutic dose of many important drugs is so small that neither the compound itself nor its possible breakdown products can be detected in the body by ordinary chemical or biological methods. Incorporation of radioactive isotopes into such drugs makes possible studies of their distribution and fate in the animal organism because of the great sensitivity of the isotope tracer technique. Obtaining the organic compound containing the isotope constitutes the major practical difficulty in the application of this technique to biological problems. At the present time many of the needed compounds can be prepared only by biological synthesis.

This report deals with our experiences in obtaining radioactive digitoxin and nicotine from the medicinal plants *Digitalis purpurea* and *Nicotiana rustica*.

Plants were grown from seed and transplanted into suitable containers containing soil, sand, or crushed mica. Growth was supported by an inorganic nutrient solution containing calcium nitrate (0.1%), magnesium sulfate (0.06%), potassium nitrate (0.05%), potassium acid phosphate (0.04%), ammonium sulfate (0.01%), and smaller amounts of other salts needed in trace amounts (*2*). As soon as the young transplanted plants were sufficiently well established, they were sealed in a closed system, usually consisting of two battery jars placed with their open ends in apposition. Suitable holes were drilled in the glass jars for the introduction of nutrient solution and of radioactive carbon dioxide, for the attach-

[1] This work was aided by grants from the Life Insurance Medical Research Fund, the U. S. Public Health Service, and the Dr. Wallace C. and Clara A. Abbott Memorial Research Fund of the University of Chicago.

Because work of this kind involves many techniques unusual for the pharmacologist and because certain precautions must be taken when radioactive substances are used, we consulted numerous colleagues in our own University and elsewhere. Among the many who aided us, we should acknowledge especially the help of Dr. Austin Brues, chief of the Biological Division, Argonne National Laboratory, and Drs. Wright Langham and Lloyd Roth, Los Alamos Scientific Laboratory, who instructed us in the procedures used in the assay of radioactive carbon and in the necessary precautions. Dr. Crooks, of the U. S. Department of Agriculture, offered helpful hints concerning the culture of these plants and also furnished us with seeds. Dr. Burris, of the University of Wisconsin, kindly gave us the benefit of his experience with technics in plant culture and in the study of plant respiration. Prof. Ezra Kraus, chairman of the Department of Botany, University of Chicago, gave us many suggestions and also placed at our disposal the facilities of the University's greenhouses, where the chief gardener, Mr. Michael Costello, was indispensable to us in the seeding, transplanting, and care of the plants in the greenhouse. We are also grateful to the Lilly Research Laboratories and to the Sandoz Chemical Company for generous supplies of the pure glycosides.

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