broncho-dilatation on intrapleural pressure are mediated by a sequence of events, causally related, as follows:

Broncho-constriction	Broncho-dilatation
↓	↓
Increased alveolar and blood CO_2 tension	Diminished alveolar and blood CO_2 tension
↓	↓
Stimulation of inspiratory medullary center	Relative depression of inspiratory medullary center
↓	↓
Increased inspiratory muscular activity	Diminished inspiratory muscular activity
↓	↓
Increased average thoracic size	Reduced average thoracic girth
↓	↓
More negative intrapleural pressure	More positive intrapleural pressure

While our experimental evidence presented in this and previous reports does not prove that a causal relationship exists between the sequence of events as listed in these schema, we feel that it is sufficient to establish a working theory in the absence of any significant evidence otherwise.

5559

Iodochloroxyquinoline (Vioform, N.N.R.) as an Amebacide in Macaques.*

HAMILTON H. ANDERSON AND DOROTHY A. KOCH.
(Introduced by C. D. Leake.)

From the Pharmacological Laboratory, University of California Medical School, and the Pacific Institute of Tropical Medicine, Hooper Foundation for Medical Research, San Francisco.

In a previous study of a series of halogenated oxyquinoline derivatives, iodochloroxyquinoline (vioform, N.N.R.) was shown to be an effective balanticide in guinea pigs, with relatively low toxicity on repeated administration to monkeys.[1] The soluble hydrochloride of the basic ether of this compound was found to kill amoebae *in vitro* in high dilutions. On this basis we began to investigate the

* Part of an extended cooperative study of the chemotherapy of amebiasis, supported in part by the Ciba Co., Inc., New York City, and Eli Lilly and Co., Indianapolis.

[1] Anderson, H. H., David, N. A., and Koch, D. A., PROC. SOC. EXP. BIOL. AND MED., 1931, **28**, 484.

amebacidal effect of vioform, N.N.R., in macaques naturally infested with *Entamoeba histolytica.* Similar studies were made by Kessel[2] with sodium iodoxyquinoline sulphonate ("yatren" or chinofon, N.N.R.) in monkeys. Doses of from one-half to 2 gm. were given daily, by this worker, to 4 macaques for 4 weeks. Two animals died in the second week of treatment when given 2 gm. of this drug daily. The 2 surviving monkeys received one-half gm. daily for 4 weeks and their stools remained negative for *Entamoeba histolytica* for a 3 months period.[3]

Dobell[4] has demonstrated the effectiveness of emetine bismuth iodide in monkey amebiasis. Twelve mgm. per kilo orally given daily for about a week cleared 4 of 5 macaques of *Entamoeba histolytica.* Four of these animals, however, exhibited signs of emetine toxicity during the course of this rather vigorous treatment. This fact was not sufficiently emphasized by Dobell but should be remembered before attempting to apply these results to human therapy. His suggestion "that macaques . . . . be utilized . . . . in place of men in future chemotherapeutic experiments" is certainly timely.

Eight monkeys, naturally infested with *Entamoeba histolytica* and various other intestinal parasites, were used in this study. In an effort to reduce reinfection to a minimum, animals were kept singly or in pairs in wire cages, which were fitted with false bottoms of 1 inch heavy mesh wire, so that droppings could fall through to the floor below. The cages were scoured daily and every attempt was made to prevent contamination of the food and water given the animals. They had a varied diet of high vitamine content rich in carbohydrate. The temperature of the room in which the monkeys were kept was between 18° and 22°C. Fresh stools were examined daily or every second day, during both a control period of observation and the course of treatment, and for 3 months afterwards. Fixed wet iron haemotoxylin preparations were examined routinely, and the stools also were cultured for amoeba and for organisms of the colon typhoid group. Three naturally infested but untreated monkeys were kept under identical conditions. Their stools remained positive for amoebae during the period in which the treated animals were studied.

Three monkeys *(Macacus rhesus)* were given powdered vioform, N.N.R., in gelatine capsules by mouth in divided doses over a 6 weeks' period. A total dosage of 900 to 1200 mgm. per kilo ren-

[2] Kessel, J. F., *Univ. Calif. Publ. in Zool.*, 1928, **31**, 275.

[3] Kessel, J. F., personal communication.

[4] Dobell, C., and Bishop, A., *Parasitology*, 1929, **21**, 446.

dered the stools negative for *Entamoeba histolytica* during the time of treatment, and for a three months' observation period afterwards. Other intestinal parasites were not consistently affected by the drug except that one animal was cleared of *Balantidium coli* and *Strongyloides stercorales* infestation. The remaining 5 monkeys were *Macacus cynomolgus;* 2 receiving 1 gm. per kilo of vioform, N.N.R., in 3 weeks time, 2 were given the same amount in

TABLE I.—Summary of Data on Vioform Treated Macaques.

Animal Species Sex	Total dosage mgm./kg.	Period of treatment weeks	Weight in gm.	Stool Examination									Character of stools	General condition after treatment
				E. histolytica	*E. coli*	*E. nana*	*Iod. butschlii*	Chilomastix	Giardia	Trichomonas	Oesophagostoma	Balantidium coli	Strongyloides	
M-342 *M. rhesus* Female	1200	6	a 2850 b 3250	+ o	++ o	+ o	+ o	++ o	o o	o o	+ o	o o	o o	formed "	excellent
M-335 *M. rhesus* Female	900	6	a 2550 b 2675	+ o	++ o	o o	o o	++ o	o o	o o	o o	o o	o o	" "	"
M-104 *M. rhesus* Male	1000	6	a 2450 b 3000	+ o	++ o	++ o	+ o	++ o	o o	++ o	++ o	+ o	+ o	" "	"
J-201 *M. cynomolgus* Male	1000	3	a 1300 b 1400	+[c] o	++ o	++ o	o o	+ o o	o o	o o	+ o o	o o	o o	" "	"
J-202 *M. cynomolgus* Male	1200	6	a 1250 b 1275	+*	+ o	+ o	++	+ o	o o	++ o	+ o	+ o	++	diarrhea formed	good
J-203 *M. cynomolgus* Female	1000	4	a 1650 b 1700	+ o	o o	o o	++	o o	o o	+ o	o o	o o	o o	" "	excellent
J-204 *M. cynomolgus* Female	1000	3	a 1500 b 1550	+ o	++ o	++	+ o	+ o	++	++	o o	o o	o o	dysentery formed	"
J-205 *M. cynomolgus* Male	1000	4	a 1200 b 1250	+ o	++ o	+ o	o o	++	+ o	++	+ o	++	o o	" "	"

a = before treatment. b = 3 months after cessation of treatment. c = Charcot-Leyden crystals.
+ = positive o = negative. * = motile forms.

divided doses over a 4-week period, while the fifth monkey, J-202, received 1200 mgm. per kilo in 6 weeks. All became and remained free of *Entamoeba histolytica* for the full period of study except J-202. This macaque was vomiting, suffered from dysentery, and was very emaciated before treatment, and showed motile amoebae in the stools. While his general condition improved markedly during observation we were unable completely to eradicate his amoebae. *Strongyloides stercoralis* was not affected by the drug in this monkey, but *Balantidium coli* was. In J-205, however, the *Balantidium coli* infestation was not cleared. Table I summarizes our results.

Summary. Seven of 8 macaques, naturally infested with *Entamoeba histolytica* have now remained free of these parasites for 3 months after receiving orally 900 to 1200 mgm. per kilo of iodochloroxyquinoline (vioform N.N.R.) given in divided doses over 3 to 6 weeks. The stools of 2 of 3 monkeys infested with *Balantidium coli* have been negative during this follow-up period. Of the 2 animals harboring *Strongyloides stercorales,* one was cleared of this infestation. We believe that important factors in our results were the hygienic conditions maintained and the precautions taken to prevent reinfection in animals under treatment. All treated animals gained weight, developed normally, and showed marked improvement in general physical condition following our therapeutic regime. No evidence of drug toxicity was noted in any animal.

5560

The Vaginal Smear of the Ewe.

H. H. COLE AND R. F. MILLER. (Introduced by C. S. Mudge.)

From the College of Agriculture, University of California, Davis.

As an introduction to the study of the physiology of reproduction in sheep we have investigated the vaginal smear of 15 ewes daily for variable periods of time, 6 of them for 11 months. Three of these have been followed through pregnancy and lactation. Data have also been accumulated regarding the lengths of the breeding periods, oestrous cycles and the length of oestrum. Oestrum was first evidenced about the first of September and 6 of the sheep that were maintained went into anoestrum about the first of March, a breeding period of about 6 months. Our data in regard to the