infective titers of the pooled hearts' bloods determined in mice were as follows: controls, $10^{-2.5}$; group treated with injections of 2.5 mg of drug, $10^{-1.3}$; group fed 5 mg of drug, less than 10^{-1} (1 of 5 mice receiving a 10^{-1} dilution of blood succumbed).

Discussion. Penicillin and drugs of the sulfonamide series have a therapeutic effect on experimental infections caused by a number of agents of the psittacosis-lymphogranuloma group. Furthermore, these substances have been employed in the treatment of patients with lymphogranuloma venereum or psittacosis (see [5]). The present data obtained in infected embryonated eggs and mice treated with Chloromycetin suggest that the new antibiotic possesses about the same degree of activity as that displayed by penicillin and sulfadiazine in similar types of studies.[5,12] It is of interest that none of the drugs are really effective in treatment of mice infected by the intracerebral route with these agents.

Conclusion. Chloromycetin possesses considerable therapeutic activity in embryonated eggs and mice infected with the viruses of psittacosis or lymphogranuloma venereum. This activity is comparable in amount to that demonstrated by others for sulfadiazine and penicillin tested under similar conditions.

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Inhibition of Multiplication of Influenza Virus by Tannic Acid.*

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Recently it has been reported[1-8] that various substances inhibit multiplication of one or more viruses of the mumps-influenza group as well as the pneumonia virus of mice. In some instances these substances inhibit both virus multiplication and virus hemagglutination. Moreover, of considerable interest is the fact that some of these substances are themselves capable of agglutinating erythrocytes. For the most part, when tested *in vitro* they exert little or no direct effect upon the viruses against which they are active *in vivo*. With the exception of the enzyme described by Burnet,[8] which apparently acts upon the receptor substance of the cell, the mode of inhibitory action and the relationship of inhibition of hemagglutination to that of multiplication are obscure.

The fact that tannin agglutinates erythrocytes has been known for some time.[9] During the course of studies on hemagglutination it was observed that tannic acid in concentrations as low as 45 μg per cc agglutinates chicken erythrocytes. Subsequent investigation showed that smaller amounts of tannic acid actually inhibit the agglutination of chicken erythrocytes by influenza A virus. Furthermore, tannic acid inhibits the multiplication of influenza A virus *in vivo* and inactivates the virus *in vitro*.

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Methods. The tannic acid employed was a commercial sample. The PR8 strain of influenza A virus was used exclusively. Allantoic fluid stored in a CO_2 chest was used as a source of virus. With minor differences, tests for inhibition were carried out as previously described.[4]

Experimental. Tannic acid was dissolved in water, usually in a concentration of 2 mg per cc, neutralized with sodium hydroxide, autoclaved and again adjusted to pH 7. 0.25 cc amounts of serial 2-fold dilutions of such solutions were mixed with 0.25 cc amounts of saline containing 8 to 16 hemagglutinating units of influenza A virus. After an interval of 5 to 15 minutes 0.25 cc of a 1% suspension of chicken erythrocytes was added to each tube. Tannic acid, in final concentration of from 5 to 20 μg per cc, inhibits hemagglutination.

Varying amounts of tannic acid were injected into the chorio-allantoic sacs of 10-day-old embryonated hens' eggs and after an interval of one-half hour 10 to 100 ID_{50} of influenza A virus were inoculated by the same route. As a rule 0.5 cc containing one mg of tannic acid was injected. This amount of tannic acid inhibits the multiplication of virus to such an extent that the allantoic fluids from eggs so treated do not contain sufficient virus to produce hemagglutination. One mg of tannic acid injected into the allantoic sac at intervals up to 6 hours preceding the inoculation of virus as well as at intervals up to one hour following the inoculation of virus also causes inhibition of multiplication.

Allantoic fluid having an ID_{50} titer of 10^{-7} was diluted 1-40 with saline and a portion of this was mixed with an equal volume of saline containing 0.2 mg of tannic acid per cc; as a control, another portion of the same allantoic fluid was mixed with an equal volume of saline. After standing at 25°C for 15 minutes tests for infectivity were done by making serial 10-fold dilutions of each mixture and injecting aliquots of the various dilutions into the allantoic sacs of 10-day eggs. In such experiments tannic acid reduces the titer of virus by at least 3 logs.

Comment. The chemical effects of tannic acid on tissues are well known and have been summarized by Olitsky.[10] However, there is little information available concerning its effect on viruses. Olitsky *et al.* showed[10,11] that tannic acid instilled intranasally was able to prevent infection with the viruses of equine encephalomyelitis and poliomyelitis subsequently administered by the intranasal route. In this instance the action of tannic acid was said to be exerted on the nasal mucosa of the host rather than on the virus itself. The fact that tannic acid combines with proteins to form a material which shows a high degree of resistance to the destructive action of enzymes[10] provides interesting ground for speculation as to the mechanism of its inhibitory action. In the experiments reported, however, it would appear that at least part of the inhibitory effect of tannic acid is dependent on a direct action upon the virus.

Summary. Tannic acid inhibits both hemagglutination and multiplication of influenza A virus. *In vitro,* the virus is inactivated by tannic acid.

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