to pass over the upper part of the body and when tightly fastened to the hook it restrains any motion of this part of the body.

When the animal is properly stretched out before securing the three chains in position the animal is completely restrained and very little, if any, movement is permitted except by the fore paws.

The advantages of this holder appear to us to lie in its compactness, simplicity and utility with a minimum number of adjustments or attachments.

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SPECIAL ARTICLES

THE VIRUCIDAL ACTION OF HIGH FREQUENCY SOUND RADIATION[1]

THE effect of high frequency sound radiation on biological material was first studied by Wood and Loomis.[2] Since the publication of their investigations in 1927 considerable work dealing with this phenomenon has followed. Much of the literature on this subject has recently been reviewed by Chambers and Gaines.[3] In our experiments we have determined the effect of such high frequency sound on tobacco mosaic virus.

The apparatus used by us was similar to that described by Harvey, Harvey and Loomis.[4] In brief, the sound radiation originated in the vibration of a one-inch-square quartz crystal immersed in a water-cooled, circulating oil bath and excited by means of a 75-watt vacuum tube oscillator in connection with a step-up voltage arrangement. The natural frequency of the crystal was 450,000 cycles per second.

Leaves from tobacco plants affected with the virus of typical tobacco mosaic were crushed, frozen overnight and thawed, and the juice was pressed from the tissue and centrifuged. Three cubic centimeters of juice were then pipetted into a small test-tube, the end of which had been blown into a thin-walled bulb about one inch in diameter. The bulb was immersed in the oil bath directly above the quartz crystal. Separate samples from the same lot of juice were irradiated for 30, 60 and 120 minutes. The temperature of the liquid within the bulb immediately after each experiment was found to have risen from 24° C. to approximately 35° C.

It was shown by a test that this rise in temperature during the course of the experiment was not a factor in the inactivation of the virus. The total number of local lesions formed on 20 half leaves of *Nicotiana glutinosa* inoculated as described below was 1,052 for juice held at 35° C. and 1,058 for unheated juice.

After each experiment the juice treated with sound radiation and the untreated controls were diluted with distilled water, as indicated in Table 1, and were used to inoculate opposite halves of leaves of *Nicotiana glutinosa,* according to the method devised by Holmes[5] and Samuel and Bald.[6] The number of local lesions produced on the leaf halves gives an indication of the relative concentration of active virus present in the suspensions.

TABLE 1

THE INACTIVATION OF TOBACCO MOSAIC VIRUS BY HIGH FREQUENCY SOUND RADIATION

Number of experiment	Duration of experiment, minutes	Number of local lesions per 20 half leaves of *Nicotiana glutinosa* inoculated with juice diluted 1: 500, except as noted	
		Untreated juice	Juice treated with sound radiation
1	30	980	50
	60	1,446	9
	120	872	0
2	30	1,301	584*
	60	1,218	52*
	120	1,116	0*

* Diluted 1: 50.

The results obtained indicate that the tobacco mosaic virus is inactivated by high frequency sound radiation. A progressive inactivation of virus with time of exposure was found to take place. After exposing the juice for two hours the presence of active virus could not be detected by inoculating

[1] A joint contribution from the Division of Plant Pathology and the Fruit Products Laboratory, University of California.

[2] R. W. Wood and A. L. Loomis. "The Physical and Biological Effects of High Frequency Sound Waves of Great Intensity," *Phil. Mag.,* 4: 417–436, 1927.

[3] Leslie A. Chambers and Newton Gaines. "Some Effects of Intense Audible Sound on Living Organisms and Cells," *Jour. Cell. and Comp. Physiol.,* 1: 451–471, 1932.

[4] E. N. Harvey, E. B. Harvey and A. L. Loomis. "Further Observations on the Effect of High Frequency Sound Waves on Living Matter," *Biol. Bull.,* 55: 459, 1928.

[5] F. O. Holmes, "Local Lesion in Tobacco Mosaic," *Bot. Gaz.,* 87: 39–55, 1929.

[6] Geoffrey Samuel and J. G. Bald. "On the Use of the Primary Lesions in Quantitative Work with Two Plant Viruses," *The Annals of Applied Biology,* 20: 70–99, 1933.

leaves of *N. glutinosa*. The results of five other experiments showed a similar inactivation.

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THE ETIOLOGY OF A RESPIRATORY DISEASE OF CHICKENS

THE occurrence of a severe type of respiratory infection in chickens, which was believed to be distinct and previously undifferentiated, was observed by Pistor, Hoffman, Beach and Schalm[1] in California early in 1933.

The disease is easily transmitted either by inoculation with exudative material or by contact exposure. In either case a nasal discharge develops which is followed by an edematous inflammation of the periorbital tissues, a purulent conjunctivitis and often a distention of the sinuses of the head with exudate. The infection may spread to the trachea, bronchii and lungs, causing rales, dyspnea, coughing, and sometimes death from suffocation. Infection by contact exposure may start as a tracheitis and bronchitis. The appetite diminishes or ceases with the onset of symptoms, resulting in a rapid and extreme emaciation. If the affected fowl does not die in the early stages of the disease, the edematous swellings of the head and the distention of the sinuses soon disappear, but a nasal discharge and a mild conjunctivitis and tracheitis may persist for many days, and finally the fowl may die in a greatly emaciated state or recovery may slowly occur.

Smears of the various exudates stained by Giemsa's or by Gram's method have quite consistently shown the presence of small bacilli, which vary in length and stain, in the majority of cases, only at the poles. This organism is not typical of the pasteurella group. Another organism, a vibrio, is also consistently found, sometimes in great profusion, in the nasal, conjunctival and tracheal exudates but not as yet in the edematous exudate.

The various exudates have been streaked on plates of the common culture media, including horse-blood agar plates which have been incubated aerobically, in an atmosphere containing 10 per cent. CO_2 and also sealed with modeling clay, the method which Nelson[2] found suitable for cultivating the causative organism of a coryza of chickens. Growth of an organism pathogenic for chickens has been obtained only on the blood agar plates incubated in an atmosphere at 10 per cent. CO_2. On such plates there has appeared, often in pure culture, very small (pinpoint) discrete colonies, which, when washed off and inoculated intranasally and intratracheally into susceptible chickens, from 6 weeks to six months old, have in 45 out of 48 cases produced symptoms varying from a slight nasal discharge to all those observed in fowls inoculated with exudate.

The incubation period of the culture-induced disease varies from 15 to 48 hours, which is the same as observed in the exudate-induced disease. Smears of the various exudates of the culture-induced disease have consistently shown the presence of the bacillus but never the vibrio. The duration of the disease induced by culture has varied between 2 and 22 days, a much less prolonged course than in exudate-induced infection.

The organism appears to belong to the class of hemophilic bacteria, for attempts to grow it in the absence of hemoglobin have been unsuccessful, and the influence of the "V" factor on its growth is readily observed on contaminated plates by the larger size and greater opacity (satellite phenomenon) of the colonies nearest the contaminant. In smears made after 24 hours incubation, the organism is found to be a small Gram negative rod, which has a tendency to form long filaments; after 48 hours' incubation, a few fragments and single rods, but rarely filaments, are observed and, after incubation for 60 or more hours, only fragments of indefinite shape, which stain faintly, are found. Transfers made after the organism has fragmented give rise to rods and filaments again in the transplant. The pathogenicity of the organism is not affected by the fragmentation. Polar staining of the organism in cultures has rarely been observed, but lightly stained areas are commonly seen in the filaments. The organism has grown aerobically in horse serum at the base of blood agar slants, and after prolonged incubation slight colonization has occurred on the surface of the slant.

The vibrio has grown in the horse serum at the base of blood agar slants, but up to the present time has not been isolated in pure culture. Until this is accomplished, it will be impossible to conclude whether it is concerned in the etiology of the disease.

Fowls after recovery from exudate-induced disease have reacted in a variable manner to subsequent inoculation with exudate; some have been refractory, while others have shown no resistance other than that the disease produced was less severe than that in the controls. Fowls that have recovered from culture-induced disease have, in 8 of 10 trials, proved refractory to a subsequent inoculation with cultures. Such fowls, however, have been susceptible to inoculation with exudate, although the resultant disease has been less severe than that in the controls, and, with one exception, edema of the periorbital region has not occurred.

The failure of infection with the organism to immunize against the exudate is not regarded as indi-

[1] W. J. Pistor, A. H. Hoffman, J. R. Beach and O. W. Schalm, *Nulaid News*, 11: 7, 1933.
[2] J. B. Nelson, *Jour. Exp. Med.*, 58: 289, 1933.