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**THE QUESTION CONCERNING THE INACTIVATION
OF VIRUSES IN THE AIR**

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**Foreign Technology Division
Wright-Patterson Air Force Base, Ohio**

20 November 1974

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U. S. BOARD ON GEOGRAPHIC NAMES TRANSLITERATION SYSTEM

| Block | Italic | Transliteration | Block | Italic | Transliteration |
|-------|------------|-----------------|-------|------------|-----------------|
| А а | <i>А а</i> | A, a | Р р | <i>Р р</i> | R, r |
| Б б | <i>Б б</i> | B, b | С с | <i>С с</i> | S, s |
| В в | <i>В в</i> | V, v | Т т | <i>Т т</i> | T, t |
| Г г | <i>Г г</i> | G, g | У у | <i>У у</i> | U, u |
| Д д | <i>Д д</i> | D, d | Ф ф | <i>Ф ф</i> | F, f |
| Е е | <i>Е е</i> | Ye, ye; E, e* | Х х | <i>Х х</i> | Kh, kh |
| Ж ж | <i>Ж ж</i> | Zh, zh | Ц ц | <i>Ц ц</i> | Ts, ts |
| З з | <i>З з</i> | Z, z | Ч ч | <i>Ч ч</i> | Ch, ch |
| И и | <i>И и</i> | I, i | Ш ш | <i>Ш ш</i> | Sh, sh |
| Я я | <i>Я я</i> | Y, y | Щ щ | <i>Щ щ</i> | Shch, shch |
| К к | <i>К к</i> | K, k | Ъ ъ | <i>Ъ ъ</i> | " |
| Л л | <i>Л л</i> | L, l | Ы ы | <i>Ы ы</i> | Y, y |
| М м | <i>М м</i> | M, m | Ь ь | <i>Ь ь</i> | ' |
| Н н | <i>Н н</i> | N, n | Э э | <i>Э э</i> | E, e |
| О о | <i>О о</i> | O, o | Ю ю | <i>Ю ю</i> | Yu, yu |
| П п | <i>П п</i> | P, p | Я я | <i>Я я</i> | Ya, ya |

- * ye initially, after vowels, and after ъ, Ё; e elsewhere. When written as Ѣ in Russian, transliterate as y^Ѣ or Ѣ. The use of diacritical marks is preferred, but such marks may be omitted when expediency dictates.

**FOLLOWING ARE THE CORRESPONDING RUSSIAN AND ENGLISH
DESIGNATIONS OF THE TRIGONOMETRIC FUNCTIONS**

| Russian | English |
|-----------|--------------------|
| sin | sin |
| cos | cos |
| tg | tan |
| ctg | cot |
| sec | sec |
| cosec | csc |
| sh | sinh |
| ch | cosh |
| th | tanh |
| cth | coth |
| sch | sech |
| csch | csch |
| arc sin | sin ⁻¹ |
| arc cos | cos ⁻¹ |
| arc tg | tan ⁻¹ |
| arc ctg | cot ⁻¹ |
| arc sec | sec ⁻¹ |
| arc cosec | csc ⁻¹ |
| arc sh | sinh ⁻¹ |
| arc ch | cosh ⁻¹ |
| arc th | tanh ⁻¹ |
| arc cth | coth ⁻¹ |
| arc sch | sech ⁻¹ |
| arc csch | csch ⁻¹ |
| <hr/> | |
| ret | curl |
| lg | log |

GREEK ALPHABET

| | | | | | | |
|---------|---|---|---|---------|---|---|
| Alpha | A | α | • | Nu | Ν | ν |
| Beta | B | β | | Xi | Ξ | ξ |
| Gamma | Γ | γ | | Omicron | Ο | ο |
| Delta | Δ | δ | | Pi | Π | π |
| Epsilon | Ε | ε | • | Rho | Ρ | ρ |
| Zeta | Ζ | ζ | | Sigma | Σ | σ |
| Eta | Η | η | | Tau | Τ | τ |
| Theta | Θ | θ | • | Upsilon | Υ | υ |
| Iota | Ι | ι | | Phi | Φ | φ |
| Kappa | Κ | κ | • | Chi | Χ | χ |
| Lambda | Λ | λ | | Psi | Ψ | ψ |
| Mu | Μ | μ | | Omega | Ω | ω |

THE QUESTION CONCERNING THE INACTIVATION OF VIRUSES IN THE AIR

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Although the role of the air medium in the propagation of respiratory virus infections is commonly known, the mechanism of their transmission has still not been studied completely. Respiratory viruses enter the air during the coughing and sneezing of sick people. In this case the large drops of aerosol settle rapidly and the fine are found suspended for a long time and are moved by air currents and cause diseases in susceptible contingents. In connection with this, the question concerning the influence of the different factors of the air medium on the inactivation of viruses in an aerosol is of considerable interest. According to the literature, one of the factors which affect the inactivation of viruses in the air is the relative humidity. Thus a number of investigators noted the rapid inactivation of the virus of influenza at moderate and high indices of relative humidity [1, 2, 5, 6, 10, 11, 13]. Furthermore, there are reports concerning the effect of relative humidity on the virus of parainfluenza [12], adenoviruses [2, 12], the viruses of poliomyelitis [5, 6], vesicular stomatitis, smallpox, Venezuelan encephalomyelitis [5], and the virus of measles [7, 8].

We studied the effect of the different indices of relative humidity on the processes of inactivation of the viruses of parainfluenza, respiratory-syncytial virus, adenoviruses, enteroviruses, and the virus of Newcastle disease in the droplet phase of an aerosol.

Experimental investigations were conducted on the following models of viruses, the infectious activity of which was expressed in negative logarithms: the virus of parainfluenza (type 3) 6.5-7.0 lg CPE_{50/0.2 ml}; respiratory-syncytial (Randall strain) 3.0-3.6 lg CPE_{50/0.2 ml}; the virus of the Newcastle disease (strain B₁) 7.0-7.5 lg IED_{50/0.2 ml}; adenoviruses of type 5 (standard and 22 f, isolated from the feces of people afflicted with infectious hepatitis) 6.5-7.0 lg CPE_{50/0.2 ml}; ECHO 7.0 lg CPE_{50/0.2 ml}. [MBA - CPE cytopathic effect; MЭД - IED expansion unknown].

The investigations were conducted in a 500 l experimental chamber, in which with the help of the glass sprayer from a Barby system for 2 min a virus-containing liquid was dispersed (flow-rate of the sprayer was 0.37 ml/min). Samples of air in the amount of 20 l were taken with the help of a Rechmenskiy bacteria trap after 5 and 30 min, 1, 3, 5, 7 and 24 hours after the dispersion of the virus suspension. As the trapping liquid in the bacteria trap 3 ml of medium No. 199 with antibiotics (penicillin and streptomycin) was used.

The separation and titration of viruses in the samples was done by the standard methods: the ECHO-7 virus - on the initially trypsinized culture of the kidneys of monkeys, the viruses of parainfluenza, respiratory-syncytial virus and adenoviruses - on the transplanted line of Hela cells, the virus of Newcastle disease - on 9-day chick embryos. For each dilution of sample 2 test tubes of tissue culture or 4 chick embryos were used. Final results were considered based on the cytopathic effect

with adenoviruses in 21-28th days, with the ECHO-7 virus in 10 days, with respiratory-syncytial virus for 10-12 days, whereupon for the acceleration of the appearance of the cytopathic effect the replacement of the medium in the infected test tubes was done every 5 days [3]. For the determination of the parainfluenza virus on the 7th day after infection the reaction of hemadsorption was set up, for the determination of the virus of Newcastle disease with the allantoic fluid of the infected embryos after 2 days of cultivation the hemagglutination reaction (HGA) was set up. The inactivation of viruses in air was judged by the loss of infectious activity by them.

The investigations were conducted with 3 indices of relative humidity: 20-25, 50-55 and 80-85%. The air temperature varied from 19 to 22°.

As the results of the investigations showed, the degree of inactivation of different viruses was dissimilar. Most rapidly in the air of the chamber the viruses of parainfluenza and respiratory-syncytial virus were inactivated, for a more prolonged period of time it was possible to detect adenoviruses, enteroviruses, and the virus of Newcastle disease. The degree of inactivation of viruses in the aerosol was influenced significantly by the relative humidity. Thus adenoviruses (both strains) and the ECHO-7 virus were inactivated most rapidly with low indices of relative humidity. With humidity within the limit of 20-25% these viruses could be determined in the air of the chamber for only an hour. On the contrary, the average and high indices of relative humidity contributed to the most prolonged preservation of the infectious activity of these viruses in the air. Thus at an atmospheric humidity of 50-55% both viruses were detected in the chamber after 7 hours, and with a humidity of 80-85% - in certain cases even in 24 hours after the creation of the aerosol (see the table).

Other results were obtained during the study of dynamics of inactivation in air of the parainfluenza and respiratory-syncytial viruses. On the one hand, they turned out to be slightly stable under conditions of the air medium, on the other - are less subjected to the effect of different relative humidity. The virus of parainfluenza was inactivated most rapidly at moderate and high relative humidity (it was detected only for 1-1 1/2 hours), while with low relative humidity the degree of inactivation was considerably lower and the virus could be determined in the air of the chamber for 2-3 hours (see the table).

Dynamics of the inactivation of viruses in an aerosol at different atmospheric humidity.

| Virus | Relative humidity in % | Titer of virus | | | | | | | | |
|------------------------------------|------------------------|---------------------------|-------------------------------------|---------------------------------------|--------|---------|---------|---------|---------|---------|
| | | in the initial suspension | in the aerosol 5 min after spraying | in different periods after dispersion | | | | | | |
| | | | | 30 min | 1 hour | 2 hours | 3 hours | 5 hours | 7 hours | 9 hours |
| ECHO | 30-35 | 7.0 | 1.75 | 0.9 | 0.9 | — | 0 | 0 | 0 | 0 |
| | 50-55 | 7.0 | 0.85 | 2.25 | 1.35 | — | 0.35 | 0.3 | 0.15 | 0 |
| | 80-85 | 7.0 | 0.75 | 1.55 | 1.35 | — | 0.35 | 0.5 | 0.5 | 0.15 |
| Adenovirus type 4 reference strain | 30-35 | 7.0 | 2.15 | 1.95 | 0.65 | — | 0 | 0 | — | 0 |
| | 50-55 | 6.75 | 3.25 | 2.75 | 1.65 | — | 1.15 | 0.41 | 0.75 | 0.75 |
| | 80-85 | 6.9 | 3.8 | 3.0 | 2.4 | — | 1.3 | 1.0 | 0.35 | 0.35 |
| Strain 270 | 30-35 | 7.0 | 3.8 | 3.5 | 2.7 | — | 1.25 | 0.75 | 0.22 | 0 |
| | 50-55 | 7.0 | 3.75 | 3.25 | 2.2 | — | 1.75 | 1.0 | 0.5 | 0 |
| | 80-85 | 7.0 | 5.2 | 4.0 | 4.0 | — | 3.5 | 2.5 | 1.5 | — |
| Newcastle disease | 30-35 | 7.3 | 4.8 | 3.5 | 3.25 | — | 2.3 | 1.5 | 0.5 | — |
| | 50-55 | 7.25 | 5.0 | 3.25 | 3.2 | — | 2.45 | 0.95 | 0 | — |
| | 80-85 | 6.75 | 2.7 | 1.35 | 0.8 | 0.65 | 0.3 | 0 | 0 | — |
| Parainfluenza | 30-35 | 6.9 | 1.8 | 0.85 | 0.35 | 0 | 0 | 0 | 0 | — |
| | 50-55 | 7.0 | 2.05 | 1.7 | 0.7 | 0.2 | 0 | 0 | 0 | — |
| | 80-85 | 7.0 | 2.05 | 1.7 | 0.7 | 0.2 | 0 | 0 | 0 | — |
| Respiratory syncytial | 30-35 | 3.7 | 1.8 | Hp | Hp | 0 | 0 | 0 | 0 | — |
| | 50-55 | 3.8 | 1.3 | Hp | Hp | 0 | 0 | 0 | 0 | — |
| | 80-85 | 3.35 | 0.8 | Hp | — | 0 | 0 | 0 | 0 | — |

Note. The titer of viruses is expressed in the negative logarithms of the CPE_{50/0.2 ml} and is the geometric mean of 5-7 series of experiments on each index of relative humidity. Abbreviation: Hp=Hp - virus revealed only in undiluted samples; -titer was not determined.

Even less stability in the aerosol state is possessed by the respiratory-syncytial virus. In the air of a chamber it was possible to detect it basically after 5-30 min after the dispersion of the virus suspension. With low relative humidity the titers of the virus were somewhat higher than with moderate and high,

which testified to the favorable effect of low humidity. With humidity within the limits of 50-55 and 80-85% the respiratory-synovial virus was determined only in undiluted suspensions. The short period during which the respiratory-synovial virus could be detected in the air of the chamber is connected, on the one hand, with the high lability of this virus. On the other - with the low infectious titer of the initial virus-containing liquid utilized for the creation of the aerosol (see the table).

The virus of Newcastle disease, unlike the parainfluenza and respiratory-synovial viruses, possessed high stability in the droplet phase of the aerosol, and, just as they, was less subjected to the effect of different relative humidity. The differences in the inactivation of the virus of Newcastle disease in air under the action of relative humidity were insignificant and fluctuated, as a rule, within the limits of 1-2 hours. However, the findings testified to the clearly expressed tendency toward the greatest inactivation of virus at high and moderate relative humidity and greater stability at low (see the table). Thus with a humidity of 50-55% the virus of Newcastle disease was detected regularly in air for 5-6 hours, and with a humidity of 20-25% - for 7 hours. Moreover, with low humidity the titer of virus on the average in 7 hours after the dispersion of suspension was $1.6 \lg \text{ IED}_{50/0.1 \text{ ml}}$.

It should be noted that the greatest effect of relative humidity was exhibited in the first seconds and minutes of existence of the virus aerosol. This was testified to by the differences in the titers of virus in the sprayed virus-containing suspension and the sample of air, obtained in 5 min after the dispersion (see the table). Thus under the effect of low relative humidity the ECHO-7 virus was inactivated to the greatest degree - the difference in titers was more than 5 logarithms, for adenoviruses this difference was 4-4.5 logarithms. Under unfavorable conditions, i.e., with high and moderate relative humidity, the difference in titers for the virus of parainfluenza reached 3 logarithms, and for the virus of Newcastle disease - 2-3 logarithms.

The results obtained testified that many viruses can retain infectious activity for a long time under conditions of the air medium at room temperature, therefore, for a specific time they can constitute a threat to susceptible contingents. It is necessary to note that the viruses which retain infectious activity for a long time in air (adenoviruses, enteroviruses, and the virus of Newcastle disease) settle on the surface of surrounding objects, which in turn can be the secondary source of infection of people. Thus the dried drops of aerosol during dry cleaning [dusting] can be raised from the surfaces (dust phase of aerosol) into the air and be moved under the effect of its currents, supporting the constant circulation of viruses in enclosed premises. In the propagation of adenoviruses, enteroviruses and the virus of Newcastle disease both the droplet and dust phase of the aerosol and the infected surfaces can be important. The parainfluenza and respiratory-synctial viruses are unstable in the environment, and therefore their propagation is connected basically with the air-droplet and contact route of transmission.

The most important factor of the air medium which affects the process of the inactivation of viruses in the aerosol state is the relative humidity, especially in the first minutes of formation of the aerosol. During this period the greatest effect on a reduction in the infectious activity of the viruses is exerted by such factors as inactivation and the process of settling. However, as shown by Miller and Artenstein [12], who labeled the appropriate fractions of the virus of parainfluenza and then studied the changes in the content of dye and infectious activity of viruses in air, the reduction in the quantity of viruses occurred because of inactivation and to a lesser degree because of settling.

The mechanism of inactivation of viruses in air depending on moderate and high relative humidity is not clear. With low relative humidity the rapid dehydration of nucleic acids occurs, which in the final result leads to the inactivation of viruses in the aerosol state [13]. This hypothesis was confirmed by

the experiments of Jong and Winkler [3] on a model of the polio-virus and its nucleic acid, in which it was shown that the inactivation of virus in the air was connected first of all with a change in the structure of the nucleic acid. However, on the basis of these investigations it is not possible to explain the more rapid inactivation of a number of viruses with high relative humidity and considerable stability with low. Data from the literature make it possible to assume that the inactivation of viruses in the air depending on its relative humidity is connected largely with the structure of the virus particle. Thus the nucleoproteins of myxo- and paramyxoviruses (influenza, parainfluenza the virus of Newcastle disease, respiratory-syncytial virus) are surrounded by a secondary membrane which contains proteins, lipides, carbohydrates and other cell components. Apparently this membrane protects the viruses from the disastrous effect of low relative humidity. On the contrary, adeno- and enteroviruses do not have a similar membrane, as a result of which with low humidity they are rapidly inactivated. A certain confirmation of this hypothesis are the investigations of Benbough [4]: on a model of purified Semliki virus the author showed that the removal of protein and salts does not affect the stability of a virus at low relative humidity, but accelerates its inactivation at high, while carbohydrates, especially inositol, increase the stability of the virus at low humidity and do not affect infectious activity at high.

Thus the results of the investigations conducted testify that the prolonged preservation of infectious activity by viruses in the air has a specific epidemiological significance and it should be considered during the conducting of different hygienic and disinfecting measures.

Conclusions

1. In enclosed habitable and communal settings many viruses

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