

## THE LETHAL EFFECT OF RELATIVE HUMIDITY ON AIR-BORNE BACTERIA\*

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An understanding of the influence of atmospheric conditions on the viability of air-borne microorganisms is essential to any attempt to formulate the epidemiology of air-borne infections. The influence of the relative humidity of the atmosphere has received inadequate quantitative study and conflicting effects have been attributed to it in promoting survival or destruction of air-borne pathogenic agents. Thus Williamson and Gotaas (1) concluded from their observations on *Serratia marcescens*, *Escherichia coli*, *Staphylococcus albus*, *Staphylococcus aureus*, and *Streptococcus salivarius*, that low relative humidities are more favorable than high ones for the viability of air-suspended microorganisms. Similarly, Edwards *et al.*, and Loosli and his associates (2) reported that influenza virus dispersed into the air is killed much more swiftly in humid than in dry air, and DeOme (3) found that the death rate of *Salmonella pullorum* atomized from aqueous suspension increases steadily with increase in the relative humidity from 15 to 80 per cent. On the other hand, Wells and Zappasodi (4) stated that hemolytic streptococci sprayed into the air are rapidly killed in dry atmospheres but are protected in the presence of moisture. In few of these investigations were many points on the relative humidity scale examined and in some cases, the exact values of the atmospheric moisture content were not determined. The marked influence of water vapor on the action of aerial germicides (5, 6) also makes desirable more careful study of its effects on microorganisms.

In the present study bacterial suspensions were atomized into an experimental chamber under controlled conditions of temperature and relative humidity, and the concentration of viable microorganisms remaining in the air after various time intervals was measured.

### Methods

The experiments were carried out in the 640 cubic foot chambers previously described (7) which allow precise control of temperature and relative humidity over fairly wide ranges.

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Seven and a half hour cultures of microorganisms were grown in heart infusion broth (Difco) containing added serum and dextrose, and suspensions prepared from these cultures were atomized into the experimental chambers for 1 minute by the standard technique already described (7). The bacterial suspensions were dispersed by reflux type atomizers (8) whose particle size distribution was measured by means of a cascade impactor<sup>1</sup> (9).

By careful standardization of the spraying procedure it was found possible to introduce a fairly constant number of microorganisms into the 640 cubic foot chamber in each experiment. This number was fixed at about 5 million. Daily checks on the total number of microorganisms discharged by the atomizer were carried out by repeating the spraying procedure with the atomizer connected directly to a bubbler sampler (10), so that the entire spray was absorbed in the bubbler fluid, aliquots of which were then plated in nutrient agar. The bacterial content of the air was determined over a 2 hour period after atomization of the microorganisms, by means of successive 2 minute bubbler samples and by exposure of agar settling plates (7).

TABLE I

*Survival of Pneumococci, Type I, Sprayed from Broth Suspension into Air at a Temperature of 22.2°C. and a Relative Humidity of 19 Per Cent*

Mass median diameter of the bacterial spray emergent from the atomizer was 3.2  $\mu$ .

Time of sample (interval between end of spray and start of sample)	No. of pneumococci per cubic ft. of air recovered by bubbler sampler	No. of pneumococci collected on a 5 min. settling plate
<i>min.</i>		
0	5200	332
2	4760	
5	4580	290
10	4000	247
20	3800	193
30	3240	177
45	2700	152
60	2420	106
75	2240	96
90	1940	90
105	1800	74
120	1620	66

The survival curves obtained by these two sampling methods paralleled each other very closely. By and large, the bubbler sampler values were somewhat more uniform, and so these have been used for calculations of the survival constants which will be reported here. A typical experimental protocol is shown in Table I.

The relative humidity was varied over a range extending from 3 to 80 per cent at three different temperatures, 14.4, 22.2 and 33.3°C. For those regions of temperature and humidity where very high bacterial death rates were encountered, the results were checked by many repetitions of each experiment.

#### EXPERIMENTAL RESULTS

The most striking fact revealed by these tests was the demonstration of the existence of a narrow range of relative humidity in the vicinity of 50 per cent

<sup>1</sup> The same atomizer was employed for all the experiments here described, except for those in which the effect of changing particle size was studied.

which is rapidly lethal for microorganisms freshly sprayed into the atmosphere from a broth suspension. The pneumococcus, a relatively delicate microorganism, exhibited the greatest sensitivity to this killing action of the atmosphere, while that of the streptococcus group C and staphylococcus was much less pronounced. In Fig. 1 are presented typical results of experiments with the pneumococcus at three different relative humidities, which illustrate the capacity of the microorganism to survive for long periods at both very low and very high relative humidities, but not at intermediate values.

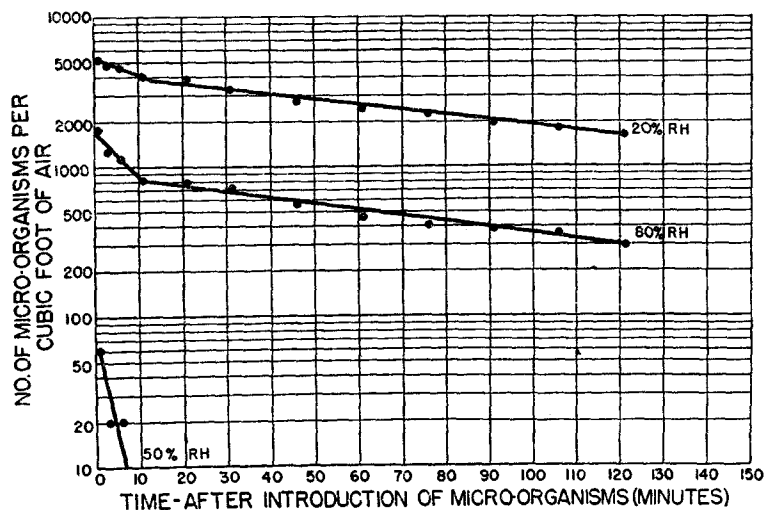


FIG. 1. Logarithmic plot of the survival of pneumococci sprayed from broth culture into atmospheres of various relative humidities, at 22.2°C. The cloud leaving atomizer had a mass median diameter of 3.2  $\mu$ . Approximately equal numbers of microorganisms were introduced into the chamber in each experiment.

Analysis of these survival curves reveals that at least two separate rate-determining lethal processes are involved. The initial one is always more rapid, and produces fairly extensive killing throughout the first 5 to 20 minutes after the droplets containing the microorganisms have been introduced into the air. The second decay process proceeds more slowly and generally lasts throughout the 2 hour observation period (Fig. 1). Both killing processes give linear logarithmic survival curves so that when the number of survivors is plotted as a logarithmic function of the time, the resulting curve consists of two straight lines meeting at a point somewhere between 5 and 20 minutes after the introduction of the microorganisms into the chamber. The slopes of these two parts of the curves have been found to be influenced by the relative humidity and temperature of the air, the nature of the microorganisms employed, the composition of the liquid medium in which they are suspended, and the

particle size of the droplets in which they are contained when sprayed into the atmosphere. Experimental effects demonstrating the operation of these factors have been studied in greatest detail for pneumococci.

Experiments similar to those presented in Fig. 1 were carried out at many different relative humidities, using a serum-dextrose broth suspension of a

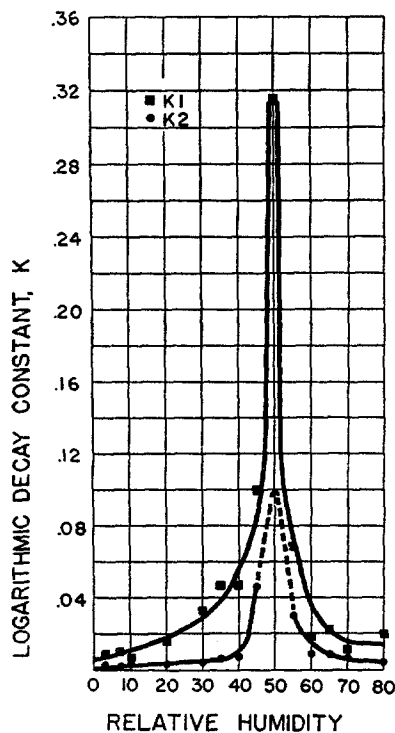


FIG. 2. Slopes of the logarithmic survival curves for pneumococci sprayed from broth suspension into atmospheres of various relative humidities.  $K_1$  is the slope of initial part of the survival curve, and  $K_2$  is the logarithmic decay constant for the final portion of the curve. The value shown for  $K_1$  at 50 per cent relative humidity,  $0.32 \pm 0.12$ , is the averaged result of more than a dozen experiments. Accurate values for  $K_2$  in the central humidity range could not be determined, because so few microorganisms survived beyond 20 minutes.

$7\frac{1}{2}$  hour pneumococcus culture. For each experiment, the numbers of survivors were plotted as a logarithmic function of the time (as shown in Fig. 1) and values for the two constants,  $K_1$  and  $K_2$  (which respectively represent the logarithmic rates of disappearance of viable microorganisms from the air during the first 10 to 15 minutes ( $K_1$ ) and for the interval thereafter ( $K_2$ )) were determined at each relative humidity at  $22.2^\circ\text{C}$ . The atomizer employed in these tests produces a cloud of bacteria suspended in broth droplets, whose mass

median diameter<sup>2</sup> is  $3.2 \mu$ , on issuing from the atomizer. A plot of the slopes of these survival curves is presented in Fig. 2. It shows that within the narrow range of relative humidities between 40 and 55 per cent, the death rate of these microorganisms sprayed in the air from a broth suspension is enormously accelerated.

*Effect of the Type of Microorganism.*—*Staphylococcus albus* and *Streptococcus hemolyticus* group C exhibited the same kind of logarithmic survival curves as that shown for the pneumococcus in Fig. 1. The effect of the relative

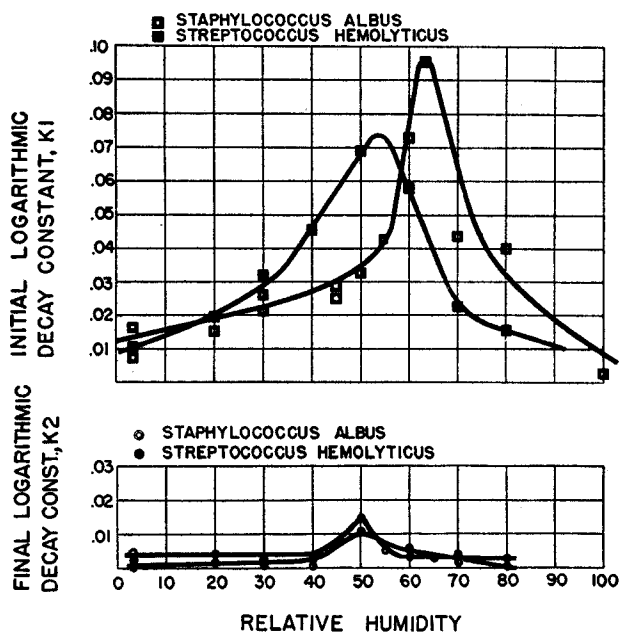


FIG. 3. Logarithmic decay constants,  $K_1$  and  $K_2$  of hemolytic streptococcus group C, and *Staphylococcus albus*, as a function of the relative humidity of the atmosphere. Microorganisms were sprayed from broth suspension.

humidity on the death rates of these microorganisms also followed the same pattern. The only observed difference lay in the relative magnitudes of the effect and in slight displacement of the value of the relative humidity producing the maximum death rate. The order of increasing susceptibility to this killing action of intermediate relative humidity was: hemolytic streptococcus group C, staphylococcus, and pneumococcus Type I. The effect of various relative humidities on the survival constants of the streptococcus and staphylococcus is presented in Fig. 3.

<sup>2</sup> *I.e.* 50 per cent of the mass of the cloud was contained in droplets whose diameter was less than  $3.2 \mu$ .

*Effect of Settling.*—It is necessary to know to what extent this disappearance of the microorganisms from the air represents a real lethal process, rather than simply a removal of air-suspended particles by settling or coalescence. In order to test this point the series of experiments represented by the points of Fig. 2 was repeated with a broth culture of microorganisms to which methylene blue had been added. Samples of the chamber air were bubbled through a solution of 70 per cent alcohol in  $H_2O$ , at the same time intervals as those used in the previous experiments. Then the total amount of dye collected in each of these samples was determined photometrically. In this way the rate of removal of these particles from the air by inelastic collisions with the walls and

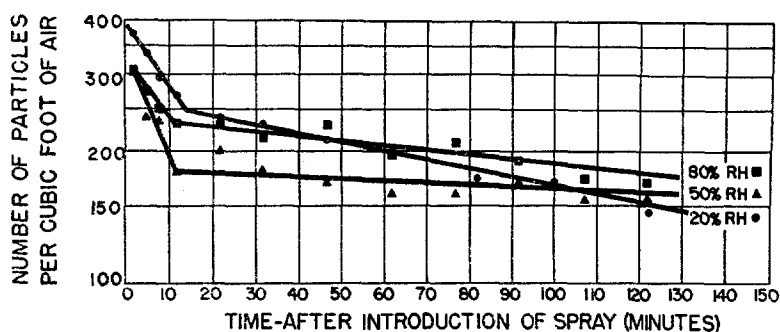


FIG. 4. Determination of settling rates by measurement of the persistence in the air of sprayed droplets containing microorganisms and a dye, at various relative humidities. The suspending medium was broth, and the instantaneous mass median diameter of the cloud was  $3.2 \mu$ . (The ordinate units are arbitrary, the figures simply indicating the relative amounts of dye present in a cubic foot of air, as a function of the time.)

floor of the chamber was determined. Sample plots of these data are shown in Fig. 4. The logarithmic curves so obtained also consist of two straight lines with the break occurring at a point usually between 10 and 20 minutes. However, when as in Fig. 5, the slopes of these curves are plotted for the entire range of relative humidity, no maximum occurs in either  $K_1$  or  $K_2$  as was found for the recovery of viable microorganisms from the air. The rate of settling remains low at all relative humidities,<sup>3</sup> whereas the rate of disappearance of viable pneumococci, for example, may be increased by 50- to a 100-fold at intermediate relative humidities. Since the rapid disappearance of microorganisms sprayed from a broth suspension into atmospheres of intermediate relative humidity cannot be accounted for by settling or collision processes, a true lethal

<sup>3</sup> It is especially low below 12 per cent relative humidity. The sharp break in the curve at this point would indicate that complete dehumidification of these particles occurs at this relative humidity.

action must be operating at relative humidities in the neighborhood of 50 per cent.<sup>4</sup>

An explanation for this rapid killing of air-borne microorganisms which occurs in atmospheres of intermediate relative humidities must be sought in an analysis of the processes which occur when bacteria-laden droplets are introduced into unsaturated air. Evaporation of water from such droplets first results in an increase in the concentration of any soluble substances present in the fluid surrounding the microorganism. If sufficient water is lost, one or more of these solutes may attain a concentration which is toxic to the cell. However, at the same time, water is also lost from the bacterial cell itself. If the cell becomes highly desiccated it becomes resistant to many kinds of physical

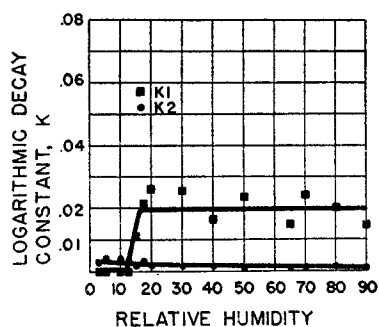


FIG. 5

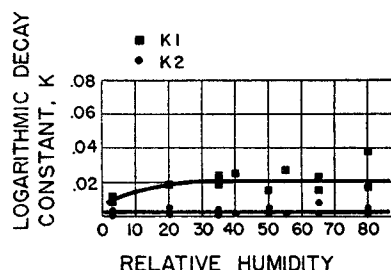


FIG. 6

FIG. 5. Slopes of the settling curves of droplets of a  $3.2 \mu$  cloud introduced into atmospheres of varying relative humidities.  $K_1$  is the slope of the initial part of the curve, and  $K_2$  the logarithmic decay constant for the final period. The difference between this curve and that of Fig. 2 indicates that the increased rate of disappearance of viable microorganisms from the air at intermediate relative humidities cannot be due to a settling process.

FIG. 6. Logarithmic survival constants of pneumococci sprayed from a distilled  $H_2O$  suspension as a function of the atmospheric relative humidity.

and chemical stresses (11) and so acquires a degree of immunity to the destructive action of the high solute concentration.

The amount of water lost by such a cell depends upon the atmospheric

<sup>4</sup> These data on settling also suggest that the more rapid rate of disappearance of a bacterial suspension during the first 10 to 20 minutes after its atomization (i.e. the difference between  $K_1$  and  $K_2$ ) is due to processes which selectively affect the larger particles more than the smaller ones. Settling of the larger particles of the cloud during the first 10 to 15 minutes undoubtedly accounts for the increased initial rate of disappearance of the dye aerosol. The same processes plus any influence which the larger particle size *per se* may exert in hastening the killing of bacteria contained within the droplets would account for the existence of the two separate death rates,  $K_1$  and  $K_2$ , which are characteristically observed when a cloud of bacteria-containing droplets is introduced into the atmosphere under the conditions described here (see below, section on particle size).

relative humidity and the binding energy between water and the various cellular constituents. At very low relative humidities dehydration is so extensive that a stable state may be achieved wherein the microorganism is not affected by the high concentration of solutes which remains. Conversely, at high relative humidities, enough water is bound to and about the cell so that lethal conditions are not achieved. The experiments here described suggest that at intermediate humidities, the amount of water retained in intimate association with the microorganism produces a critical state in the cell. In this condition the bacterium becomes highly susceptible to the toxic action of the unphysiologic concentration of solutes remaining from the original medium in which it was suspended.

*Effect of Varying the Composition of the Suspending Fluid.*—In the light of the foregoing analysis, it may be predicted that if the microorganisms are sprayed into air from a suspension in distilled water rather than broth, the rise in death rate at the intermediate relative humidities should be greatly reduced. This conclusion was verified experimentally. Seven and one-half hour cultures of pneumococcus, Type I, were centrifuged, washed once in distilled H<sub>2</sub>O, and then resuspended in water before spraying into the experimental chamber. The survival rate was then determined in the standard manner. The characteristic linear logarithmic survival curves exhibiting a sharp break at the end of 10 or 15 minutes were obtained, exactly like those observed in the broth experiments. However, in contrast to these latter tests, substitution of distilled water for broth completely eliminated the great increase in death rate at humidities around 50 per cent. The variation with relative humidity of the logarithmic survival constants of pneumococci sprayed into the air from a distilled water suspension is shown in Fig. 6. Comparison of the curves of Figs. 6 and 5 reveals that the survival pattern of these air-borne pneumococci is practically identical with that obtained for the persistence of an aerosol of dye droplets, indicating the absence of any lethal process.

It became of interest to ascertain which of the components of the broth solution was exerting the toxic effects observed at relative humidities in the vicinity of 50 per cent. Separate solutions of the various constituents of the nutrient broth were prepared and a solution of each component in turn, in the same concentration in which it occurs in the broth, was employed as a suspending medium from which pneumococci were sprayed into the air. Such experiments revealed that NaCl present in the original solution in a concentration of 0.5 per cent, was the agent largely responsible for the lethal action. All the other constituents of the nutrient broth, when tested either singly or together failed to produce a marked lethal effect at intermediate relative humidities. Thus determination of the survival constants of a pneumococcus culture sprayed into the air from a suspension containing all the components of



nutrient broth except NaCl,<sup>5</sup> resulted in a pair of curves like that of Fig. 6. Conversely, when a solution containing only 0.5 per cent NaCl was employed survival curves superposable on those of Fig. 2 resulted. Thus the characteristic sharp rise in mortality rate at intermediate relative humidities occurs only in the presence of the NaCl.

In the light of these results, it was important to test the effect of relative humidity on bacteria sprayed into the air from a saliva suspension. This fluid constitutes the natural medium from which respiratory bacteria are introduced into the atmosphere, so that the survival pattern of microorganisms suspended in it would have direct bearing on problems of natural air-borne infection. Saliva was collected from human volunteers who chewed paraffin for several minutes to induce a rapid flow. No attempt was made to free the fluid from its normal bacterial population, since in the final suspension which was prepared, these microorganisms were so far outnumbered by the pneumococci as to represent a negligibly small degree of contamination. A 7½ hour culture of pneumococci Type I was spun down and resuspended in an equal volume of saliva. The resulting suspension was immediately transferred to the atomizer and sprayed into the chamber by the standard procedure used in the preceding experiments. The resulting survival pattern was identical with that obtained when broth or saline (Fig. 2) was used as the suspending medium, rather than the distilled water type of curve (Fig. 6).

*Effect of the Size of the Droplet.*—If death of bacteria sprayed into the air from a broth or saliva suspension is indeed due to a process connected with dehydration then an atmospheric relative humidity which is ordinarily lethal should be much less so when the bacteria are suspended in small droplets rather than in large ones. This follows because the smaller droplet, containing a smaller total amount of the injurious agents exposes the microorganism to a lower concentration of them as dehydration proceeds. This effect was tested by comparing the survival rates of pneumococci sprayed into the air from a serum-dextrose broth suspension, by two different atomizers, delivering clouds whose instantaneous particle size distributions corresponded to mass median diameters of 1.6  $\mu$  and 3.2  $\mu$  respectively. A typical set of experiments at 22.2°C. and 50 per cent relative humidity is presented in Fig. 7, demonstrating the greater rate of killing for the pneumococci suspended in the larger droplets. In a series of over fifteen experiments at 22.2°C. the average value of  $K_1$ , the decay constant observed during the first 10 to 20 minutes after the end of the spray, was  $0.012 \pm 0.005$  per minute for the 1.6  $\mu$  particles, and  $0.32 \pm 0.12$  per minute for those 3.2  $\mu$  in diameter. The average value of  $K_2$ , the killing rate during the second part of the survival curve was 0.045 per minute for the 1.6  $\mu$  particles. (So few bacteria remained alive at the end of 20 minutes that

<sup>5</sup> *I.e.* this solution contained 1 per cent peptone, 1 per cent tryptose, 0.05 per cent dextrose, and 2.0 per cent serum.

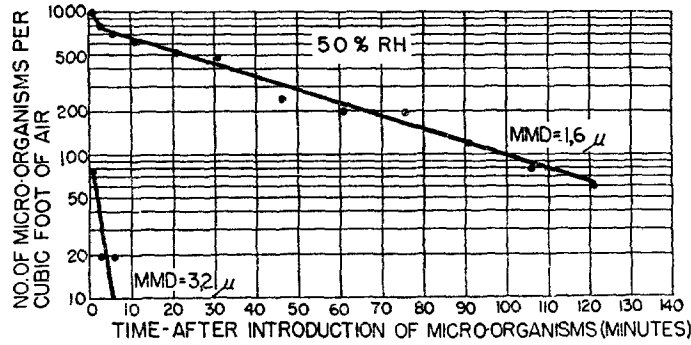


FIG. 7. Effect of cloud particle size on the survival of pneumococci sprayed from broth suspension into an atmosphere of 50 per cent relative humidity at 22.2°C.

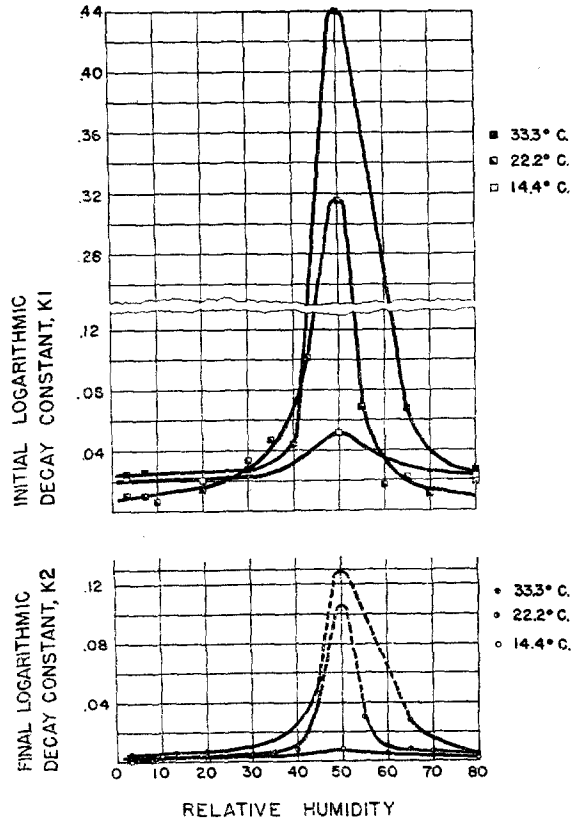


FIG. 8. Effect of temperature on the survival constants of pneumococci sprayed from broth suspension into atmospheres of varying relative humidity.

it was impossible to obtain a satisfactory value of  $K_2$  for the  $3.2 \mu$  particles at 50 per cent relative humidity.)

*Effect of Temperature.*—The influence of temperature changes on this lethal process was also studied. The survival of suspensions of pneumococci in broth sprayed into the air at various relative humidities, was measured at  $14.4^\circ\text{C}$ . and  $33.3^\circ\text{C}$ ., as well as at  $22.2^\circ\text{C}$ . As might be expected a pronounced temperature coefficient was observed,  $K_1$  varying by a factor of fifteenfold between these temperature extremes. Values for  $K_1$  and  $K_2$  as a function of the relative humidity at the three temperatures are contained in Fig. 8. As before, the values for  $K_2$  in the intermediate humidity zone are uncertain at the two higher temperatures, since the extremely high initial death rates under these conditions leave too few survivors to enable the subsequent lethal process to be studied.

#### DISCUSSION

When a suspension of a given composition is atomized into the atmosphere the ultimate extent of dehydration depends on the relative humidity, since the droplet eventually comes into a condition of equilibrium with the atmosphere. When this occurs, the water content of the particle will be such that the aqueous tension within it is equal to the partial pressure of water in the air. At low relative humidities, then, a particle containing a microorganism will have lost almost all its water—even some which may have been in fairly tight chemical combination with certain cellular constituents. At high relative humidities, however, even very loosely bound water will be retained. At intermediate relative humidities, partial dehydration of the various cellular systems will occur. Calculation shows that even at a relative humidity of 50 per cent, spherical droplets of distilled water can evaporate from an initial diameter of  $3 \mu$  to one-tenth that size within 0.004 second.<sup>6</sup> This time is much faster than any of the death rates observed in the experiments reported here so that the evaporation process would be finished before the lethal process was well under way. Hence, the rates of killing observed in these experiments must be interpreted as constituting the natural mortality of the microorganisms in a given chemical environment, when in a state of hydration which is in equilibrium with the prevailing relative humidity.

If the theoretical formulation here proposed is correct, then the lethal action

<sup>6</sup> The rate of evaporation of small droplets in unsaturated air has been shown to proceed according to the equation (12):  $\frac{dA}{dt} = \frac{8\pi DM}{RTd} (p_s - p'_s)$  where  $A$  = area of the droplet,  $D$  = diffusion coefficient of the vapor, equal to  $0.24 \text{ cm}^2$  per second for water vapor in air at room temperature;  $M$  = molecular weight of the vapor, 18.0 for  $\text{H}_2\text{O}$ ;  $R$  = gas law constant, 62,400 cc.  $\times$  mm. Hg. mole<sup>-1</sup> deg.<sup>-1</sup>;  $T$  = absolute temperature,  $296^\circ \text{A}$ ;  $d$  = density of the lique-

observed at intermediate relative humidities is not due simply to the salt activity by itself. Droplets of pure sodium chloride solution sprayed into the air become saturated with this solute at a relative humidity of 75 per cent (13) and at lower humidities would dry out completely. However, in our experiments microorganisms suspended in pure saline showed a maximum lethal effect at relative humidities near 50 per cent rather than at 75 per cent. This apparent discrepancy can be explained if one considers that in a bacterial suspension the salt enters into solution and association with some of the cellular constituents. The maximum lethal action of the salt which is bound within the cell, is exercised at the relative humidity which dehydrates the microorganism to the point where it becomes most vulnerable, rather than at one which is in equilibrium with a saturated solution of pure NaCl.

The present experimental results have been explained by postulating the existence of a critical degree of moisture content at which a bacterial cell becomes much more susceptible to toxic agents than when it contains either more or less water. Other lines of evidence indicate that this is a phenomenon characteristic of bacterial systems. This principle underlies the lyophilization process for preparing dried microorganisms in a viable condition. If water is removed from such cells at room temperature, the culture dies with great rapidity. Hence, in order to avoid destruction the bacteria are first cooled to a temperature so low that the rate of this lethal process is greatly diminished. Then the water is removed as rapidly as possible. Once dehydrated beyond the critical region, the cells can again be warmed to room temperature without extensive mortality. Application of these considerations has made it possible to devise a simple method for drying microorganisms without resorting to the low temperatures utilized in the lyophilization process. It has been found that cells may be dehydrated at room temperature without loss of viability provided that the desiccation is so rapid that the stable state is achieved before the lethal processes have had time to operate. These studies will be described in a forthcoming publication (14).

The greatly increased efficacy of steam sterilization over dry heating doubtless stems from the same phenomenon. Microorganisms heated in the presence

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fied vapor, which is unity for water;  $p_o$  = partial pressure of water vapor in the atmosphere, which would be equal to the product of the atmospheric relative humidity and the vapor pressure of water at the temperature of the experiment; and  $p'_o$  = pressure of water vapor in equilibrium with the droplet. For droplets of pure water,  $p'$  would be equal to the pure vapor pressure, which is 20.1 mm. Hg. at 22.2°C. If the droplet contains other substances dissolved in the aqueous phase,  $p'$  would be lowered by an amount corresponding to the vapor pressure lowering produced by the combined solutes. The presence of salts and proteins within the droplets would decrease the rate of evaporation somewhat but not even a saturated salt solution would lower the vapor pressure sufficiently to decrease this evaporation time by a significant amount.

of steam are maintained in a state of intermediate hydration which makes them more susceptible to the killing action of the high temperatures than would be the case if they were allowed to dry out completely. Still another example of the operation of this same effect may be found in the diminished effectiveness with which aerial disinfectants such as the glycols, kill air-borne microorganisms at low relative humidities (5, 6). This effect is partly due to failure of the germicidal vapor to condense as readily on the desiccated particles which are produced when a bacterial cloud is dispersed in a very dry atmosphere. However, recent experiments *in vitro* have also demonstrated that completely dehydrated bacteria are more resistant to the killing action of the glycols in the liquid state (15).

The values for  $K_1$  and  $K_2$  as here calculated represent the logarithmic rates of removal of viable bacteria from the atmosphere, and so include both the settling rates and the lethal process due to the presence of NaCl at the critical atmospheric humidity. When the appropriate settling rates obtained from Fig. 5 are subtracted from these, extremely small values are obtained for the mortality rates at both very low and very high relative humidities for the three microorganisms tested here. In some cases, the result is negative, which simply means the death rate is too small to be evaluated accurately by this technique.

The nature of the lethal action exercised by NaCl at the critical levels of cellular dehydration probably involves denaturation of one or more essential enzyme systems. Further study of the mechanism of this killing process and of the behavior of bacterial cells in varying states of hydration is being carried out.

Evaluation of the significance of the results reported here for epidemiological problems must await further study. It will be necessary to test in more detail the behavior of a larger number of pathogenic agents, particularly those of the virus group. It is possible that these effects may help to explain certain features of the seasonal pattern of spread of some respiratory diseases. The possibility also exists that humidity regulation by itself might prove to have some effect as a prophylactic measure. Preliminary experiments have shown that by the introduction of a carefully regulated amount of steam into the air of a chamber it is possible to rehumidify and produce rapid and extensive killing of an aerial dispersion of pneumococci which have been allowed to become highly desiccated by an hour's sojourn in an extremely dry atmosphere. While the raising of the relative humidity to the 50 per cent point may not reduce aerial contamination to the same extent as can be achieved by glycol vapors or ultraviolet irradiation, the simplicity of this measure would commend its use in a large variety of situations if it could be shown to exercise even a partially beneficial effect.

## SUMMARY

The viability of pneumococcus, Type I, sprayed into the atmosphere from a liquid suspension was measured as a function of the relative humidity. When broth, saliva, or 0.5 per cent saline solution is employed as the suspending medium, a very high mortality rate is observed at relative humidities in the vicinity of 50 per cent. However, at humidities above or below this value the microorganisms survive for long periods.

Measurement of the rate of settling of droplets employed in these experiments demonstrated that the disappearance of microorganisms from the air is a true lethal process, rather than a manifestation of aerosol collision processes.

When a saline-free fluid was used, the sharp peak in death rate at intermediate relative humidities disappears.

The lethal effect of intermediate relative humidities on pneumococci atomized from a saline-containing suspension is increased when the particle size of the atomized droplets is increased or when the temperature is raised.

Cultures of hemolytic streptococcus group C and staphylococcus sprayed from a broth medium exhibit the same general survival pattern as a function of relative humidity although the mortality rates are smaller than that of the pneumococcus.

These effects can be explained by assuming the existence of a critical degree of cellular dehydration at which microorganisms become much more sensitive to toxic agents than in states where either more or less water is bound to the cell.

The results presented here may be significant in elucidating certain aspects of the epidemiology of air-borne infections.

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