This dissertation has been microfilmed exactly as received

SMITH, Richard Furnald, 1922–
BIOLOGICAL EFFECTS OF IONIZED AIR,

University of Arizona, Ph.D., 1962
Chemistry, biological

University Microfilms, Inc., Ann Arbor, Michigan
Copyright by
Richard Furnald Smith
1962
I hereby recommend that this dissertation prepared under my direction by Richard Furnald Smith entitled BIOLOGICAL EFFECTS OF IONIZED AIR be accepted as fulfilling the dissertation requirement of the degree of Doctor of Philosophy.

Dissertation Director 5-1-62

After inspection of the dissertation, the following members of the Final Examination Committee concur in its approval and recommend its acceptance:

Warren H. Furrer 5-1-62
GR. Kerner 5-13-62
Henry W. Kingle 5-13-62
R. L. Hammadeh 5-13-62
A. Mair 5-13-62

*This approval and acceptance is contingent on the candidate's adequate performance and defense of this dissertation at the final oral examination. The inclusion of this sheet bound into the library copy of the dissertation is evidence of satisfactory performance at the final examination.
STATEMENT BY AUTHOR

This thesis has been submitted in partial fulfillment of requirements for an advanced degree at The University of Arizona and is deposited in the University Library to be made available to borrowers under rules of the Library.

Brief quotations from this thesis are allowable without special permission, provided that accurate acknowledgment of source is made. Requests for permission for extended quotation from or reproduction of this manuscript in whole or in part may be granted by the head of the major department or the Dean of the Graduate College when in their judgement the proposed use of the material is in the interests of scholarship. In all other instances, however, permission must be obtained from the author.

SIGNED: Richard E. Smith

APPROVAL BY THESIS DIRECTOR

This thesis has been approved on the date shown below:

Warren M. Freese 5-1-62
ACKNOWLEDGMENT

Sincere thanks are given to Dr. Wallace Fuller who made this investigation possible, to Dr. Arthur Kemmerer who gave sound advice and endured with fortitude my predatory raids on his supplies, to Dr. Mitchell Vavich, Shirley Bosma, Gordon Johnson, Ray Cattani, Henry Mayland, and all the others who gave the basement of the Aggie Building its special character and made it such a congenial place in which to work.
# Table of Contents

## Introduction

- A. Historical Background ........................................ 1
- B. Definition, Occurrence, and Measurement of Air Ions ........... 7

## Experimental Methods and Results

- A. The Action of Air Ions on Plant Tissue ................. 12
  - a. Selection of the Initial Test Organism ............ 12
  - b. Effect of Air Ions on Microcoleus vaginatus ........ 13
  - c. Identification of the Growth-stimulating Component of Positively-ionized Air .................. 16
  - d. Effect of Air Ions on Avena sativa ...... 19

- B. The Action of Air Ions on Mammalian Tissue .......... 23
  - a. Effect of Air Ions on Ciliated Epithelium ........ 23
  - b. Identification of the Biologically Active Gaseous Ion Species .......... 24
  - c. Effect of Air Ions on the Cytochrome System .................. 25
  - d. 5-hydroxytryptamine as the Endogenous Mediator of Air Ion Effects .......... 25

- C. The Mechanism of Air Ion Action on Plant Tissue ........ 32
  - a. Indole-3-acetic Acid as the Endogenous Mediator of Ion-induced Growth Stimulation ........ 32
  - b. Mathematical Feasibility of the Indole-3-acetic Acid Hypothesis .......... 33
  - c. Effect of Added Indole-3-acetic Acid on the Growth of Microcoleus vaginatus ........ 36
  - d. Effect of Positively-ionized Air on the Release of Bound Indole-3-acetic Acid .......... 37
<table>
<thead>
<tr>
<th>DISCUSSION</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. The Perennial Appeal of Pseudoscientific Dualisms</td>
<td>41</td>
</tr>
<tr>
<td>B. Air Ions and Human Health</td>
<td>42</td>
</tr>
<tr>
<td>C. Air Ions and Agriculture</td>
<td>46</td>
</tr>
<tr>
<td>D. The Direction of Future Air Ion Research</td>
<td>47</td>
</tr>
</tbody>
</table>

| SUMMARY                          | 51   |
|==================================|------|
| BIBLIOGRAPHY                     | 52   |
LIST OF TABLES

Table No.                                                                 Page

1. Classification of Air Ions by Size and Electrical Mobility ......................... 8

2. The Major Forces Responsible for Air Ion Formation in Nature ...................... 8

3. Effect of Positively-ionized Air on the Release by Germinating Corn Kernels of Added Indole-3-acetic Acid ....................... 39

4. Effect of Positively-ionized Air on the Release of Added Indole-3-acetic Acid by Corn Coleoptiles as Shown by Their Appearance After Treatment with the Gordon-Weber Colorimetric Reagent .......... 39
<table>
<thead>
<tr>
<th>Figure No.</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>A Polonium air ion generator</td>
<td>10</td>
</tr>
<tr>
<td>2.</td>
<td>An air ion current probe</td>
<td>10</td>
</tr>
<tr>
<td>3.</td>
<td>Exposure chamber for studying the effects of unipolar air ions on algae</td>
<td>15</td>
</tr>
<tr>
<td>4.</td>
<td>Colonies of Microcoleus vaginatus grown in the above exposure chamber</td>
<td>15</td>
</tr>
<tr>
<td>5.</td>
<td>The effect of positively-ionized air on the growth and migration of single M. vaginatus hormogones</td>
<td>17</td>
</tr>
<tr>
<td>6.</td>
<td>Comparison of the effects of positively-ionized nitrogen and positively-ionized air on M. vaginatus</td>
<td>20</td>
</tr>
<tr>
<td>7.</td>
<td>Abolition of the growth-stimulating effect by removal of carbon dioxide from positively-ionized air</td>
<td>20</td>
</tr>
<tr>
<td>8.</td>
<td>Effect of positively- and negatively-ionized air on the growth of 4.0 mm Avena coleoptiles</td>
<td>22</td>
</tr>
<tr>
<td>9.</td>
<td>Effect of positive and negative air ions on the survival of staphylococci suspended in 50 lambda droplets of distilled water</td>
<td>28</td>
</tr>
<tr>
<td>10.</td>
<td>Arrangement for observing the effect of air ions on the isolated rabbit trachea</td>
<td>28</td>
</tr>
<tr>
<td>11.</td>
<td>Effect of air ions on the ciliary activity of the isolated rabbit trachea</td>
<td>29</td>
</tr>
<tr>
<td>12.</td>
<td>Effect of air ions on the rate of mucus flow on the surface of the rabbit trachea</td>
<td>29</td>
</tr>
<tr>
<td>13.</td>
<td>Capacity of negative air ions to reverse the inhibition of ciliary activity caused by positive air ions on the rabbit trachea</td>
<td>29</td>
</tr>
<tr>
<td>Figure No.</td>
<td>Description</td>
<td>Page</td>
</tr>
<tr>
<td>-----------</td>
<td>------------------------------------------------------------------------------</td>
<td>------</td>
</tr>
<tr>
<td>14.</td>
<td>Effect of positive and negative air ions on mucus flow rate and ciliary rate</td>
<td>29</td>
</tr>
<tr>
<td></td>
<td>in a living, tracheotomized rabbit</td>
<td></td>
</tr>
<tr>
<td>15.</td>
<td>Test for unipolar negative and positive gaseous ion activity on the rabbit</td>
<td>30</td>
</tr>
<tr>
<td></td>
<td>trachea</td>
<td></td>
</tr>
<tr>
<td>16.</td>
<td>Rate of conversion of succinate to fumarate</td>
<td>30</td>
</tr>
<tr>
<td></td>
<td>by a Keilin-Hartree pig-heart homogenate</td>
<td></td>
</tr>
<tr>
<td></td>
<td>in normal and negatively-ionized atmospheres</td>
<td></td>
</tr>
<tr>
<td>17.</td>
<td>Re-oxidation of reduced cytochrome c by cytochrome oxidase in positively- and</td>
<td>30</td>
</tr>
<tr>
<td></td>
<td>negatively-ionized atmospheres</td>
<td></td>
</tr>
<tr>
<td>18.</td>
<td>The effect of intravenous injections of 5-hydroxytryptamine, iproniazid, and</td>
<td>31</td>
</tr>
<tr>
<td></td>
<td>reserpine on the ciliary activity of a living, tracheotomized rabbit under</td>
<td></td>
</tr>
<tr>
<td></td>
<td>nembutal anesthesia</td>
<td></td>
</tr>
<tr>
<td>19.</td>
<td>The effect of negative air ions on the rate of recovery in the extirpated</td>
<td>31</td>
</tr>
<tr>
<td></td>
<td>trachea of a rabbit pre-treated with 5-hydroxytryptamine</td>
<td></td>
</tr>
<tr>
<td>20.</td>
<td>The structural formulae of 5-hydroxytryptamine and indole-3-acetic acid</td>
<td>34</td>
</tr>
</tbody>
</table>
INTRODUCTION

A. Historical Background

The existence of atmospheric electricity was established independently in the middle of the eighteenth century by Benjamin Franklin with his famous kite experiment (19) and by the French scientist, T. F. d'Alibard (1). A few decades later, the Abbé Bertholon wrote a book describing the alleged therapeutic properties of atmospheric electricity (4).

In 1795, Coulomb showed clearly that ordinary air can conduct electricity (12). More than one hundred years passed, however, before Elster and Geitel demonstrated that the ability of air to conduct electricity resides in positively and negatively charged gaseous particles which they called "ions" (18).

A persistent folklore soon developed to the effect that negative air ions are physiologically and psychologically "good", while positive air ions are physiologically and psychologically "bad". For example: the malaise induced by the Fohn, Sirocco, Mistral, Zonda, Khamsin, Chinook, and other seasonal winds has been attributed by many authors to the excess of positive air ions they often contain (13, 15, 24). On the other hand,
C. W. Hansell of RCA (22) recently suggested that Moses was inspired to lead his people out of Egypt because of the "sudden and powerful dose of negatively ionized air" produced by the burning bush (5); he also suggests that negative ions from a corona discharge so exhilarated the Apostles as to excite the comment, "these men are full of new wine" (5).

In the 1920s, Friedrich Dessauer made an elaborate clinical study of the subject. He concluded that positive air ions increase respiration, basal metabolic rate, and blood pressure, cause headaches dizziness, nausea, and fatigue; while negative air ions decrease respiration, basal metabolic rate, blood pressure, and produce a sense of well-being (16).

In most of Dessauer's experiments air ions were generated artificially by means of high voltage brush discharge, a method which also disseminates ozone and various oxides of nitrogen. In other experiments finely-divided MgO was used to carry the electrical charge. Thus Dessauer's patients were subjected to a number of other factors besides air ions, and the reported results cannot be attributed exclusively to ionized air.

At Harvard University during the 1930s Yaglou and his co-workers were consistently unable to repeat Dessauer's work. The Harvard group generated air ions
by an invisible point discharge from "a multiplicity of phonograph needles", supplied with a high tension rectified current. Neither diseased nor normal human subjects showed any significant physiological responses or subjective sensations when exposed to unipolar air ions of either charge (61, 62). Shortly before his death in 1960, Dr. Yaglou reported at a national symposium on air pollution that from all his investigations "nothing definite was found to justify the use of artificial ionization in ventilation or air conditioning" (63).

These sobering observations, however, have been all but drowned in a flood of papers claiming that air ions influence blood pressure (46), tumor growth (40), the course of tuberculosis (39), hay fever (25), whooping cough (47), the egg production of chickens (10), the performance of Russian weightlifters (59), and the attendance of Oregon schoolchildren (57). Almost invariably, negative ions are described as beneficial in these papers, while positive ions are described as harmful.

A conspicuous dissenter in this tide of unanimity has been Dr. G. Schorer of Switzerland. Since 1928 Schorer reported results almost exactly the opposite of those of his fellow-workers. In 1953 he was induced to test the polarity of his air-ionizer, whereupon he
discovered his terminals to be reversed (22).

Since then, every major electrical company (RCA, Philco, Westinghouse, General Electric) and a number of minor ones (Puritron, Granco, Wesix, Tubin) have designed and patented air-ionizers. Typical of the claims being made for these devices is a full-page advertisement appearing in the August 4, 1961, issue of *Life* magazine: "NEWS FOR HAY-FEVER AND AIR-BORNE-ALLERGY SUFFERERS! Clinical test records show negative ion treatment brings relief to 63% of cases.....The portable Philco "Ionitron 5000" is sold in pharmacies, or consult your physician. The price: one hundred dollars....."

The Wesix Electric Heater Company of San Francisco has supported research on air ion effects for many years and claims to deplore commercial exploitation of air-ionizing devices. Yet it regularly runs advertisements in *California Medicine* and the *American Journal of Physical Medicine* picturing its own air-ionizer with the headline: "RELIEVES HAY FEVER".

A broader range of benefits is attributed to the "Ion-o-matic Air Improver" of Tubin Electronics, Los Angeles. The manufacturers recommend their device:

"If you are tense, nervous, depressed... (picture of woman chewing handkerchief),

"If you have asthma, hay fever, suffer from
allergies... (picture of man sneezing),

"If you seek relief from pain... (picture of man wincing),

"Even if you are healthy and normal... (no picture).

The Food and Drug Administration recently seized two types of "air purifiers" as not delivering the advertised benefits. More suits appear imminent (11).

Undismayed, the makers of the "Vornado Vionizer" ("The ONLY Auto Air Conditioner with the Amazing NEGATIVE ION GENERATOR!") advertised in the April 12, 1962 Tucson Daily Citizen that their apparatus relieves:

"Respiratory Troubles! Allergies! Headaches! Prevents road accidents... keeps you alert and refreshed! Stops tobacco odors...!"

Studies of the action of electricity on plants also date back to the time of Benjamin Franklin. In 1746, the Scotch scientist Maimbray reported he had "electrified" two myrtle bushes and that "they put forth small branches and blossomed sooner than other shrubs which had not been electrified" (41). This work was confirmed by the Court Physician of King Louis XV, the Abbé Nollet, who stated that electrical discharges stimulated the germination and growth of mustard plants (44).

Ever since these experiments, investigators have been subjecting plants to all manner of electrical shocks.
In the 1920s, there was a widespread "electroculture" movement in England; wires were embedded in the ground and growing crops were given jolts of electricity. Apparently the results were disappointing, for the movement soon died out. Typical experiments have been described by J. C. Bose (8) and K. Stern (55). When E. C. Barton-Wright surveyed the entire field in 1933, he concluded that in general the results had been "remarkably poor" (2).

No doubt many of these experiments, especially those employing high voltage discharges (7), resulted in variable numbers and kinds of air ions being present in the vicinity of the growing plant. But the extent to which this may have influenced the reported results is now impossible to determine.

In 1953 Breazeale and McGeorge found that direct applications of electrical currents altered the cation uptake of plants (9). And over the years Shrank has obtained various curvature and growth inhibition effects in electrically treated Avena coleoptiles (50).

It is important to distinguish between these direct electrical effects and effects due to air ions per se. The former are dependent upon the electrical current itself, while the latter are dependent upon the gaseous molecular species carrying the current. Up until 1960, there had been no studies of the effects of air ions on plants comparable in scope to those made on mammalian
tissues. In 1957, Krueger, Smith, and Gan showed that air ions could be mildly lethal to bacteria under certain rather special conditions (27). And more recently air ions have been reported to slow the growth of fungi (48). Yet the influence of air ions on photosynthesizing organisms remained unexplored until the investigation begun at this University in September, 1960.

B. Definition, Occurrence, and Measurement of Air Ions

An air ion is a molecule or molecular complex of an atmospheric constituent which has lost or gained an electron and thus acquired an electrical charge. Physicists characterize air ions according to their size and mobility (Table 1).

The earth's atmosphere normally contains from 500 to 2000 small and intermediate positive air ions per milliliter and a somewhat smaller number of negative air ions. In nature they are formed continually as positive and negative ion pairs by the forces listed in Table 2.

In the laboratory, air ions are usually generated by means of radio-isotopes. The beta radiations of Tritium ($\text{H}^3$) or the alpha radiations of Polonium ($\text{P}^{210}$) form ion pairs in the adjacent atmosphere. A rectifying circuit removes the ions of unwanted charge, and the ions of desired charge move in the electrical field to the object.
Table 1. Classification of Air Ions by Size and Electrical Mobility (32).

<table>
<thead>
<tr>
<th></th>
<th>Diameter in μ</th>
<th>Average mobility cm/sec in field of 1 volt/cm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Large (Langevin) ions</td>
<td>.03 — .10</td>
<td>.0005</td>
</tr>
<tr>
<td>Intermediate ions</td>
<td>.003 — .03</td>
<td>.05</td>
</tr>
<tr>
<td>Small ions</td>
<td>.001 — .003</td>
<td>1.0</td>
</tr>
</tbody>
</table>

Table 2. The Major Forces Responsible for Air Ion Formation in Nature (52).

1. Radiation:
   (a) from radioactive substances in the soil
   (b) from radioactive gases produced by the decay of (a)
2. Cosmic radiation
3. Electrical discharges
4. Short wave ultraviolet light
5. Frictional electricity generated by:
   (a) rain or waterfall droplets
   (b) blowing sand, dust, snow
undergoing exposure (Fig. 1). In this manner air ion concentrations several thousand times higher than levels found in nature can be produced.

Naturally occurring air ion concentrations can be measured by drawing air through a duct which contains a series of insulated polarizing plates. Deposition of ions on these plates results in a small electric current which is measured with a micro-microammeter (31). Artificially generated air ions moving in an electrical field can be measured more conveniently by means of a specially designed probe (Fig. 2). The probe is placed in the same position as the object undergoing exposure. As the ions impinge on the probe, a minute current is produced which is measured with a micro-microammeter or an ultrameter. The current produced is converted into ions/cm²/sec by using the formula:

\[ N = \frac{I}{qA} \]

in which I equals the current produced, q equals \(1.6 \times 10^{-19}\) coulombs, and A equals the area of the probe surface (27).

Air contains \(27 \times 10^{18}\) molecules per milliliter. Under optimum laboratory conditions, it is only possible to produce \(1 \times 10^6\) unipolar ions per milliliter. Thus "ionized air" contains just one ionized molecule per 27 trillion molecules of un-ionized air.
Fig. 1. A Polonium air ion generator. Negative ions are moving out in the electric field, while positive ions are being removed. (32)

Fig. 2. An air ion current probe (27).
In addition to being highly dilute, air ions move very slowly in comparison, for example, with various types of radiation. Air ions appear to have no distinctive attributes other than the possession of an electric charge. Yet they are capable of exerting a number of highly specific, albeit minor, effects on biological systems.
A. The Action of Air Ions on Plant Tissue

a. Selection of the Initial Test Organism - It was not definitely known at the start of this investigation whether air ions had any biological effects on plants or not. If they did, common sense suggested that such effects would be most readily detected in some relatively simple photosynthetic organism; a species of algae, particularly one growing in a subaerial habitat, seemed to offer the greatest promise.

Accordingly, a number of green and blue-green algal species were screened: Chlamydomonas reinhardi, Protococcus viridis, Chlorella pyrenoidosa, Nostoc muscorum, Microcoleus vaginatus, Scytonema hofmannii, and Anabaena spiroides. It soon became evident that the common blue-green alga, Microcoleus vaginatus, displayed an easily detectable response to positively-ionized air.

Later it was found that positively-ionized air produces analogous responses in other algal species and in higher plants. However, considerably more elaborate techniques are required for their demonstration. The remarkable hardiness of Microcoleus and its capacity for rapid, aggressive growth under favorable conditions made
it ideal for these early experiments.

b. **Effect of Air Ions on Microcoleus vaginatus** - A growth chamber was adapted from the glass manifold of a micro-Kjeldahl apparatus. It consisted of a horizontal glass cylinder, 22 inches long and 2 inches in diameter, through which water-saturated air was continuously drawn. The Tritium head of the ion source was placed inside the cylinder, just under the roof, in about the center of the chamber. (Fig. 3)

The Microcoleus samples were arranged lengthwise in the growth chamber. Those samples located upstream of the ion source were the control samples; those samples downstream of the ion source were the test samples. No sample was located closer than 5.0 cm to the ion source.

Stock cultures of a specimen of *Microcoleus vaginatus* (Vaucher) Gomont, isolated from a local soil crust, were maintained on the Medium I of Sager and Granick (49). Although this medium was developed for *Chlamydomonas reinhardi*, it proved upon addition of 2% agar to be especially favorable for *Microcoleus* growth. Cultures were incubated at room temperature in water-saturated atmospheres and were exposed to 1,000 foot-candles for 12 hours each day.

A Wesix "Ionaire" consisting of Tritium in a
sealed foil with a reversible rectifying circuit that permitted selection of either positive or negative ions was used as the air ion source (3). Unipolar ion densities of $1 \times 10^8$ ions/cm$^2$/sec were produced by this apparatus, as measured with a Beckman micro-microammeter used in conjunction with a target probe. The monitoring technique and the calculations have already been described above.

Single Microcoleus hormogones or 1.0 mm discs excised from the stock culture were placed on the center of fresh 5 ml plates of Sager and Granick agar, and these were then placed inside the growth chamber. Every 12 hours the plates were removed and examined under low power. The amount of growth occurring was estimated by counting the number of hormogones migrating off the inoculum agar, and measuring the migration distance of each hormogone.

After one day's exposure to positively-ionized air, the 1.0 mm test discs could be seen under low power magnification (20X) to have put forth numerous filaments in all directions to a radius of 5 mm. Control discs during the same period put forth few or no filaments.

After 2-3 days' exposure to positively-ionized air, the difference between control and test samples was readily apparent to the naked eye (Fig. 4). The positively-ionized samples were invariably heavier,
Fig. 3. Exposure chamber for studying the effects of unipolar air ions on algae (52).

Fig. 4. Colonies of *Microcoleus vaginatus* grown in the above exposure chamber. Samples 2, 3, and 4 received positively-ionized air for three days (54).
greener, and more extensive than the corresponding control samples. These results were observed in over 50 ionized and control pairs.

Similar experiments with negatively-ionized air failed to produce any significant differences between control and test samples in over a dozen attempts.

In order to put the increased Microcoleus growth rate in quantitative terms, single hormogones were exposed to ionized air under the same conditions. As shown in Fig. 5, positively-ionized air caused large increases both in the numbers of new hormogones produced and in the distances these hormogones migrated. Viewed under 100X magnification, the positively-ionized hormogones displayed continuous "creeping" movements, while the control hormogones remained relatively stationary.

These results were obtained in a total of 34 experiments with positively-ionized air. Once again, however, similar experiments with negatively-ionized air failed to produce any significant differences between control and test samples in more than a dozen attempts.

c. Identification of the Growth-stimulating Component of Positively-ionized Air - The first step in elucidating the mechanism by which positively-ionized air enhances algal growth is to determine which of the various
Fig. 5. The effect of positively-ionized air on the growth and migration of single \textit{M. vaginatus} hormogones (53).
gaseous ions present in positively-ionized air is responsible for the observed effects.

When this problem arose earlier in connection with the mammalian trachea, it was solved very simply (see section B., b.). Since air ions cause an alteration in the tracheal ciliary rate within twenty minutes, it was possible to expose extirpated pieces of trachea to ionized pure gases and determine which ones raised or lowered the normal ciliary rate of 850 beats per minute.

This technique obviously will not serve for algae. Growth effects appear much more slowly than ciliary rate changes. Placing a photosynthesizing organism in atmospheres of 100% oxygen or carbon dioxide for extended periods will produce metabolic changes so profound as to completely overshadow any ion-induced effects. Therefore it was necessary to devise a new technique.

First of all the exposure period was shortened as much as was compatible with unequivocal growth effects. Test conditions were alternated with normal growing conditions each hour, in order to insure continued viability of the Microcoleus. Arbitrary growth classifications were defined, based on the numbers and distances of the migrating filaments. Distilled water that had been freshly boiled and cooled was used to saturate the air in the growth chamber. When pure gases were
alternated within the chamber with air, the water was changed with each change in the ambient atmosphere. A conventional CO$_2$-absorption train filled with Ascarite was used to remove carbon dioxide when required.

The treatments used and the results are summarized in Figures 6 and 7.

The increased growth effect was strictly dependent on the presence of positively-ionized carbon dioxide. No other ionized gas had any detectable effect upon algal growth under these experimental conditions.

d. Effect of Air Ions on Avena sativa - The action of ionized air on higher plants was investigated next. The Avena straight-growth coleoptile test was selected, because of its widespread use and the high degree of standardization it has undergone.

Experiments were performed in light-proof growth chambers consisting of enameled metal bowls, 7.0 cm high and 15.0 cm in diameter, covered with canopies of opaque polyethylene film (containing 25% carbon black). The canopies were fastened tightly around the bowls by means of rubber bands. The test growth chambers were identical with the control growth chambers except for the ion source which protruded through the center of the polyethylene canopy.
Fig. 6. Comparison of the effects of positively-ionized nitrogen and positively-ionized air on M. vaginatus growth (53).

Fig. 7. Abolition of the growth-stimulating effect by the removal of carbon dioxide from positively-ionized air (53).
The Nitsch and Nitsch modification of Bentley's Avena section straight-growth test was used (43). The 4.0 mm oat coleoptile sections were floated in watch glasses or 5 ml beakers, which were placed inside the light-proof growth chambers. Atmospheres within both test and control growth chambers were water-saturated.

The results of two typical experiments with their associated sample statistics are shown in Table 3. Similar results were obtained in 22 experiments involving more than 500 coleoptile sections.

The positively-ionized coleoptiles showed an increased elongation that corresponded to $5 \times 10^{-3}$ ug of indole-3-acetic acid in the 1.0 ml suspending medium used, or 5.0 ug per liter.

In no case did negatively-ionized air cause an increased elongation over the control samples beyond that attributable to chance.
Fig. 8. Effect of positively- and negatively-ionized air on the growth of 4.0 mm Avena coleoptiles. Two experiments selected at random. The semicircles represent the range of the Standard Error of the Mean at the 99.8% level (53).
B. The Action of Air Ions on Mammalian Tissue

a. Effect of Air Ions on Ciliated Epithelium - In order to understand fully the reasoning behind subsequent experiments on plants, it will be necessary to summarize work done by the author and Dr. A. P. Krueger from 1956 to 1960.

Encouraged by our discovery that ionized air has a measurable effect on the survival rates of staphylococci, albeit under rather specialized conditions (Fig. 9), we turned our attention to the respiratory tract. If air ions had any physiological effects at all, this seemed like the most likely area for them to appear.

Most of the mammalian respiratory tract is lined with ciliated epithelial cells which beat at a characteristic rate, continually propelling mucus up from the lungs (17). This rate of ciliary beating can be accurately determined by a stroboscopic method developed by T. E. Dalhamn of Sweden (14). We placed isolated strips of rabbit trachea in a glass and plastic chamber containing air of high relative humidity. By means of a microscope and a strobotachometer, it was possible to observe both the rate of ciliary beating and the rate of mucus flow (Fig. 10).

We found that positively-ionized air caused the ciliary rate to decrease sharply, in some cases to cease
altogether; negatively-ionized air increased the ciliary rate (Fig. 11). The mucus flow rates roughly paralleled the ciliary rates (Fig. 12). Negatively-ionized air reversed the effects of positively-ionized air, and vice versa (Fig. 13).

We extended our observations to include the tracheas of living rabbits, guinea pigs, rats, mice, and cynomolgus monkeys (29, 31, 34). The same general responses were found as in the tracheal strip (Fig. 14). In addition, positively-ionized air caused blanching, smooth muscle contraction, and an increased vulnerability of the mucosa to mechanical trauma; negatively-ionized air increased the vascularity of the tracheal mucosa.

b. Identification of the Biologically Active Gaseous Ion Species - The specific components of ionized air responsible for the observed effects were identified by exposing tracheal strips to pure gases, both ionized and un-ionized. Negatively-ionized carbon dioxide and negatively-ionized nitrogen had no effect on the ciliary rate; but negatively-ionized oxygen produced the characteristic rise. Un-ionized oxygen did not give this effect under these conditions. On the other hand, positively-ionized oxygen and positively-ionized nitrogen had no effect on the ciliary rate; but positively-ionized carbon dioxide produced a rapid decline. Under these
conditions, un-ionized carbon dioxide did not give this effect (Fig. 15).

c. **Effect of Air Ions on the Cytochrome System**

The discovery that negatively-ionized oxygen mediates negative air ion effects suggested the possible involvement of the respiratory enzymes. Accordingly, the effect of negative air ions on the rate of conversion of succinate to fumarate by a Keilin-Hartree pig-heart homogenate was studied. Fumarate formation was more rapid in the negatively-ionized sample (Fig. 16).

The next step was to determine exactly where in the respiratory chain of enzymes negative ions operate. Negative ions were found to have no effect on cytochrome c alone, but they have a distinctly accelerating effect on the rate at which cytochrome oxidase re-oxidizes reduced cytochrome c (Fig. 17).

Thus it appears that negative air ions act directly on cytochrome oxidase, or else form a radical upon contact with water which acts directly on cytochrome oxidase (33).

d. **5-hydroxytryptamine as the Endogenous Mediator of Air Ion Effects** - While the above experiments furnish a plausible explanation for the effect of negative ions on ciliary activity, they fail to account for the other
negative ion effects, or for any of the positive ion effects. At this point, it was noted that positive ion effects bear a strong resemblance to some of the effects attributed to 5-hydroxytryptamine (5-HT, serotonin). In particular, the ability of this hormone to cause smooth muscle contraction, vasoconstriction, and increased respiratory rates.

When 10 milligrams of 5-HT are injected intravenously into an anesthetized tracheotomized rabbit, a condition indistinguishable from that caused by positive air ions is produced in the trachea. The ciliary rate drops, the posterior tracheal wall contracts, the mucosa becomes blanched, and its vulnerability to trauma is greatly increased. Iproniazid, a drug which causes 5-HT levels to rise by blocking the action of monamine oxidase, has a similar but more lasting effect. Reserpine, which is thought to accelerate the oxidation of 5-HT, produces a ciliary rate rise and other negative-ion-like effects. (Fig. 18)

The rate at which an animal recovers from 5-HT effects is greatly accelerated by negative ions (Fig. 19).

These and a number of other experiments, including direct analyses of 5-HT levels in the trachea, strongly suggest that positive air ions exert their physiological effects by causing a local release of 5-hydroxytryptamine in the trachea. Negative ions counter this by increasing
the rate at which 5-hydroxytryptamine is oxidized, presumably through a cytochrome-linked reaction (36, 37).
Fig. 9. Effect of positive and negative air ions on the survival of staphylococci suspended in 50 lambda drops of distilled water. The ordinate represents the logarithm of the numbers of surviving bacteria per drop. Significant effects were only obtained in samples which were stirred throughout the exposure period. (27)

Fig. 10. Arrangement for observing the effect of air ions on the isolated rabbit trachea (28).
Fig. 11. Effect of air ions on the ciliary activity of the isolated rabbit trachea (28).

Fig. 12. Effect of air ions on the rate of mucus flow on the surface of the rabbit trachea (28).

Fig. 13. Capacity of negative air ions to reverse the inhibition of ciliary activity caused by positive air ions on the rabbit trachea (28).

Fig. 14. Effect of positive and negative air ions on mucus flow rate and ciliary rate in a living, tracheotomized rabbit (29).
Fig. 15. Test for unipolar negative and positive gaseous ion activity on the rabbit trachea. Only negatively-ionized oxygen and positively-ionized carbon dioxide have any effect on the rate of ciliary beating. (30)

Fig. 16. Rate of conversion of succinate to fumarate by a Keilin-Hartree pig-heart homogenate in normal and negatively-ionized atmospheres (33).

Fig. 17. Re-oxidation of reduced cytochrome c by cytochrome oxidase in positively- and negatively-ionized atmospheres (33).
Fig. 18. The effect of intravenous injections of 5-hydroxytryptamine, iproniazid, and reserpine on the ciliary activity of a living tracheotomized rabbit under nembutal anesthesia (36).

Fig. 19. The effect of negative air ions on the rate of recovery in the extirpated trachea of a rabbit pre-treated with 5-hydroxytryptamine (36).
C. The Mechanism of Air Ion Action on Plant Tissue

a. Indole-3-acetic Acid as the Endogenous Mediator of Ion-induced Growth Stimulation - It was shown earlier that positively-ionized carbon dioxide is the constituent of ionized air responsible for the enhanced growth of Microcoleus and Avena. On theoretical grounds there is no reason for supposing carbon-dioxide-minus-an-electron to be any more efficient in photosynthesis than un-ionized carbon dioxide. Quite the reverse, in fact, since the transformation of carbon dioxide into sugars is a reductive process.

Moreover, it was noted that positively-ionized air had no effect on the rate at which illuminated spinach chloroplasts reduce 2,6-dichlorophenol. Thus the Hill reaction is not involved in the mechanism.

In the preliminary screening experiments positively-ionized Chlamydomonas reinhardi suspensions showed no increased growth over control suspensions (as indicated by cell counts). However, the filtrates of the positively-ionized Chlamydomonas suspensions had a marked growth-stimulating effect on Microcoleus cultures. This suggested that positive air ions had caused the release of an auxin-like material which, although unable to increase Chlamydomonas growth under the given experimental
conditions, was nevertheless able to stimulate Microcoleus growth.

The role of hormones in algal physiology is still not completely understood. Yet many workers have reported that indole-3-acetic acid stimulates the growth of several Chlorophyte species. These investigations have been summarized by R. W. Krauss (26).

In animal cells, positive air ions apparently act by causing a local release of "bound" 5-hydroxytryptamine (36, 37). This compound is structurally related to the common plant growth hormone, indole-3-acetic acid (Fig. 20). Conceivably, in plant cells positive ions might cause a release of indole-3-acetic acid. The basic mechanism of positive air ion action would then be the same in both plants and animals; the different responses (enhanced growth in plants, vasoconstriction etc. in animals) would be due simply to the different properties of free indole-3-acetic acid and free 5-hydroxytryptamine within their respective tissues.

With a view towards testing this hypothesis, the following calculations and experiments were carried out.

b. Mathematical Feasibility of the Indole-3-acetic Acid Hypothesis - Is there enough bound indole-3-acetic acid naturally present in plant tissue to produce the observed growth increases if released? Before proceeding
Fig. 20. The structural formulae of 5-hydroxytryptamine and indole-3-acetic acid.
further it was necessary to answer this question.

According to Wildman and Bonner (60), the first 5.0 mm of an Avena coleoptile contains approximately $1 \times 10^{-4}$ ug free indole-3-acetic acid. The Nitsch and Nitsch assay uses a 4.0 mm section cut 3.0 mm from the tip. Interpolating from the concentration gradient of indole-3-acetic acid in the coleoptile as given by Thimann (56), we may estimate that at the time of cutting the 4.0 mm section contains $0.5 \times 10^{-4}$ ug free indole-3-acetic acid. According to Van Overbeek (58), this value represents only 8% of the bound indole-3-acetic acid present. Because the amount of bound indole-3-acetic acid varies with the type of extraction used, the age and growing conditions of the seedling, we shall use a range of 3% to 10% in this calculation. Thus, a single 4.0 mm Avena section may contain from $2 \times 10^{-3}$ to $5 \times 10^{-4}$ ug bound indole-3-acetic acid. Since ten sections are used in each assay sample, 0.02 to 0.005 ug bound indole-3-acetic acid are present in each sample.

The lower limit of sensitivity of the Avena assay is reported to be 0.005 ug indole-3-acetic acid (43). Thus, in the least favorable cases there is still enough bound indole-3-acetic acid present to cause a detectable growth response if released. In the most favorable cases only one-fourth of the bound indole-3-acetic acid present
would have to be released in order to cause a detectable
growth response.

These calculations cannot replace direct analyses, of course. But they do indicate that the hypothesis is well within the realm of possibility.

c. Effect of Added Indole-3-acetic Acid on the Growth of Microcoleus vaginatus - The hypothesis postulates that the observed growth increases are due to release of indole-3-acetic acid. However, it has not been established whether indole-3-acetic acid does in fact cause increased growth in the blue-green alga, Microcoleus vaginatus.

The effect of graded concentrations of indole-3-acetic acid on this organism was observed. 5.0 mm Microcoleus discs were excised from the stock cultures and placed on the surface of conventional Petri dishes containing 20 ml of Sager and Granick medium. Concentrations of indole-3-acetic acid (Fisher) ranging from 0.01 to 200.0 μg per plate (3 x 10⁻⁵ to 3 x 10⁻⁵ molar) were either incorporated directly into the medium, or placed on filter paper discs (Difco sterile blanks, 6.5 mm) on the agar surface and allowed to diffuse into the medium.

Enhanced growth of the treated discs was clearly visible to the naked eye within 24 hours. After three
days, the diameter of the treated discs was from 5 to 25 times greater than that of the control discs. A maximum effect was obtained with 1.0 to 2.0 ug indole-3-acetic acid per plate (3 to 6 x 10^{-7} molar). However, concentrations as low as 0.01 ug (3 x 10^{-9} molar) produced unequivocal (i.e., 2X) growth increases.

d. Effect of Positively-ionized Air on the Release of Bound Indole-3-acetic Acid - If the hypothesis is correct, it should be possible to couple pure indole-3-acetic acid with suitable binding materials and then show that positive air ions cause the indole-3-acetic acid to be released in greater amounts than under control conditions. With such a technique, indole-3-acetic acid may be assayed by a colorimetric method rather than by the time-consuming Avena test.

Indole-3-acetic acid in aqueous solution was combined with a variety of materials: Dowex 1-X8, Amberlite IR-4B, soil, carbon, and several proteins. In no case could it be demonstrated that positive air ions had the slightest effect on the release of indole-3-acetic acid from these materials. Eventually germinating corn kernels ("Golden Victory" variety) were found to be convenient and satisfactory binding materials. The kernels were exposed in the same growth chambers and under
the same conditions as the Avena coleoptiles in an earlier section. Indole-3-acetic acid was assayed by means of the colorimetric method of Gordon and Weber (21).

At first, the germinating corn kernels were soaked overnight in $1 \times 10^{-3}$ molar indole-3-acetic acid solutions, washed thoroughly, then exposed in distilled water on watch glasses to positive air ions. As shown in Table 3, the amount of indole-3-acetic acid in the distilled water increased more rapidly in the positively-ionized samples. However, in roughly one-third of these experiments there was no significant difference between the control and the positively-ionized supernatants.

A more reproducible demonstration of ion-induced release of indole-3-acetic acid was obtained as follows: corn seedlings were submerged in $1 \times 10^{-3}$ molar indole-3-acetic acid overnight, the first 3.0 mm of the coleoptiles removed, washed, and exposed to positive air ions while floating on 4.0 ml distilled water. At intervals, coleoptile tips were removed, placed on spotting plates, and treated with 0.5 ml of the Gordon-Weber colorimetric reagent.

Under these conditions, the positively-ionized coleoptile tips rapidly lost their ability to form a color with the Gordon-Weber reagent. This effect was not obtained when the tips were suspended in 0.01 molar KCN during ionization. Moreover, no binding of indole-3-
Table 3. Effect of Positively-ionized Air on the Release by Germinating Corn Kernels of Added Indole-3-acetic Acid (53).

<table>
<thead>
<tr>
<th>Hr EXPOSED</th>
<th>OD&lt;sub&gt;560&lt;/sub&gt; OF SUPERNATANT&lt;sup&gt;a&lt;/sup&gt;</th>
<th>mg IAA/1 IN SUPERNATANT&lt;sup&gt;b&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(C) (+)</td>
<td>(C) (+)</td>
</tr>
<tr>
<td>0</td>
<td>0.00 0.00</td>
<td>0.0 0.0</td>
</tr>
<tr>
<td>4</td>
<td>0.12 0.13</td>
<td>4.0 4.0</td>
</tr>
<tr>
<td>12</td>
<td>0.20 0.32</td>
<td>6.0 9.0</td>
</tr>
<tr>
<td>24</td>
<td>0.33 0.45</td>
<td>9.0 12.5</td>
</tr>
</tbody>
</table>

<sup>a</sup> After treatment with the Gordon-Weber colorimetric reagent.
<sup>b</sup> Read from a freshly prepared standard curve.

Table 4. The Effect of Positively-ionized Air on the Release of Added Indole-3-acetic Acid by Corn Coleoptiles, as Shown by Their Appearance After Treatment with the Gordon-Weber Colorimetric Reagent (53).

<table>
<thead>
<tr>
<th>Hr EXPOSED</th>
<th>COLOR OF COLEOPTILES</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>UNTREATED</td>
</tr>
<tr>
<td></td>
<td>(C)  (+)</td>
</tr>
<tr>
<td>0</td>
<td>Purple</td>
</tr>
<tr>
<td>2</td>
<td>Purple</td>
</tr>
<tr>
<td>10</td>
<td>Mottled</td>
</tr>
</tbody>
</table>

Purple Purple Purple Purple Purple
Mottled Purple Purple Purple Purple
White Purple Purple Purple Purple
acetic acid occurred when the seedlings were pre-treated with 0.01 molar KCN. (Table 4)
A. The Perennial Appeal of Pseudoscientific Dualisms

"The real charm of the intellectual life," Aldous Huxley wrote in 1928, "the life devoted to erudition, to scientific research, to philosophy, to esthetics, to criticism—is its easiness. It's the substitution of simple intellectual schemata for the complexities of reality; of still and formal death for the bewildering movements of life." (23)

To what extent this mordant observation applies to genuine science is debatable. But it may explain a recurrent pattern of popular scientific thinking, a pattern which we may call the Pseudoscientific Dualism. Of universal occurrence, the Pseudoscientific Dualism can be defined as a theory which purports to explain some complex natural phenomenon in terms of a naive dichotomy.

How much easier it is to consider people as either "introverts" or "extroverts", instead of attempting to deal with all the multiplicities of human character. The factors making up "good nutrition" are discouragingly numerous and complex; but anyone can eat "alkaline" foods and avoid "acid" foods, or eat "organic" vegetables and
avoid "inorganic" vegetables. Surely the Germans under Hitler must have found—once they accepted the notion that "Aryans" were noble and "non-Aryans" degenerate—that social relations, the interpretation of history, and life in general were enormously simplified.

Needless to say, this kind of thinking is not science. It is romance. Through it, an individual's life is transformed into a perilous adventure played out between Good and Evil Agents.

Air ion folklore, as epitomized in the Reader's Digest article "Air Ions Can Do Strange Things To You" (45), fits into this pattern of Pseudoscientific Dualism with disturbing ease. Negative ions bring health and happiness; they stimulate, yet somehow, at the same time, they tranquilize. Positive ions, on the other hand, cause headaches, pessimism, and the sniffles. A rather paranoid note is sounded by the insistence that, although unseen, air ions are constantly around us, exercising their baleful (or salubrious) influences.

A genuine understanding of the biological significance of air ions will only be possible when this simple-minded dualism is abandoned once and for all.

B. Air Ions and Human Health

The obvious need in this area is for increasing numbers of objective clinical studies. Whenever such
studies have been made, as for example last year's elaborately controlled arthritis study in the University of Pennsylvania's "Climatron" (42), the therapeutic benefits of ionized air seem to vanish.

Meanwhile, a number of facts suggest that air ions, however fascinating to play with in the laboratory, are something less than environmental agents of the first magnitude.

Air ions have never been shown to produce any effect, mental or physical, that cannot be more efficiently and reliably produced by other agents. No human being has ever been found who can consistently tell whether he is receiving positive ions, negative ions, or no ions at all. The growth, behavior, and longevity of mice is not affected to the slightest degree by air ions (38).

Air ions have no penetrating ability. The cytochrome oxidase effects described earlier are only obtained when the homogenate is continuously stirred (33). Bacterial suspensions in 50 lambda droplets of distilled water must also be stirred constantly in order to show altered survival curves; even then, the presence of a trace of nutrient will abolish the effect (27).

Air ions do affect the 5-hydroxytryptamine levels in the respiratory tract. But the only mammal in which 5-hydroxytryptamine plays a part in foreign body response is the mouse. Repeated experiments involving at least
500 mice and 500 guinea pigs failed to show that air ions have the slightest influence on the anaphylactic response of these animals (38). Even an asthmatic mouse is unlikely to derive any benefit from ionized air.

There is some evidence that air ions are effective only once in an individual's life, at least as far as the effect on 5-HT levels is concerned. When guinea pigs are first exposed to unipolar air ions, their excretion of 5-hydroxy-indoleacetic acid (the chief metabolite of 5-HT) rises for a day or two. Thereafter, for the remainder of the animal's life, further exposures to air ions of either charge have no effect on the level of 5-hydroxy-indoleacetic acid excretion (37). Should the alteration of respiratory 5-HT levels confer some hidden benefit, it might be a serious therapeutic limitation if one had to be "ionically virgin" in order to receive it.

Negative air ions will undeniably increase the rate of ciliary beating in the respiratory tract. Much of the current air-ionizer advertising is based on this fact.

But is it desirable to have one's cilia flap more rapidly than they normally do? If one could count on inhaling just one or two pathogens a day, there might be some comfort in knowing that the inhaled bacterium or pollen grain would be flushed up to the larynx and expectorated in 4 minutes instead of the usual 5. But ordinarily one inhales pathogens and irritants almost
continuously, and the fact that one's cilia are beating at 1100/minute rather than 900/minute can make very little difference.

No one, least of all the manufacturers of air-ionizers, has considered the implications of two other negative ion effects on the respiratory tract: relaxation of smooth muscle and increased mucosal vascularity.

The smooth muscle effect must be very slight, since it is completely overcome by anaphylactic shock. Yet presumably an unsensitized mammal exposed to negative air ions will have larger diameters than usual in the finer tubes (those unsupported by cartilage) of his respiratory tract. This will admit more air; it will also admit more pollen, virus, dust, or whatever else happens to be in the air. Inevitably a larger proportion of these agents will travel beyond the ciliary barrier than normally. The rate of ciliary beating will then be of no significance.

Increased mucosal vascularity might be beneficial if one's blood stream carries antibodies against an invading organism. If one's blood stream lacks the proper antibodies, then the invader will be nourished and disseminated more abundantly than under control conditions.

There is some evidence that this is exactly what happens under certain conditions. Ordinarily, ionized air does not affect the mortality rate of mice experimentally infected with respiratory pathogens (administered as
aerosols). However, when the pathogen concentration is greatly reduced, the mortality rates of both positively and negatively ionized mice increase well above that of the control mice. Autopsied lungs of the survivors show more extensive involvement in the ionized animals than in the control animals (38).

To sum up: the influence of air ions on human health is probably slight and usually outweighed by more powerful agents. In those few cases where air ions do affect health, it is wrong to assume that the traditional air ion folklore is correct.

C. Air Ions and Agriculture

From the point of view of comparative biochemistry, the analogous responses of plant and animal cells to positively-ionized air are highly interesting.

The role of 5-hydroxytryptamine--while providing countless graduate students with material for dissertations--remains dubious. Perhaps it is the biochemical equivalent of the vermiform appendix, a leftover from some earlier evolutionary stage, now functionless, but retaining the ability to cause trouble.

Indole-3-acetic acid, on the other hand, is essential to higher plants in many ways and gives no indication of becoming a biochemical has-been.
Is the response of plants to positive air ions merely a laboratory curiosity? Is it desirable to have a plant grow more rapidly at the expense of its own hormonal reserves?

Probably not, in most cases. Nor is this discovery likely to have any applications to commercial agriculture. The difficulty and expense of "ionizing" an acre of cotton would be enormous. And even if done, the results would not approach in extent and reliability those obtained with auxin sprays.

However, there may be special situations in which positively-ionized air would be of value. In small-scale greenhouse operations, for example, with plants that do not absorb or translocate externally applied auxin well. Several processes other than growth are regulated by indole-3-acetic acid. These include (1) the storage and dormancy characteristics of plant materials, (2) the rooting of cuttings, (3) the alteration of flowering behavior, and (4) the control of fruit-set.

The effectiveness of positive air ions in any of these applications would be limited, of course, by the amount of reserve indole-3-acetic acid within the plant.

D. The Direction of Future Air Ion Research

The above paragraphs were written as if the
hypothesized mechanism of air ion action on plants were true. Actually it is still far from proven.

Binding of indole-3-acetic acid to protein does occur in plant cells (20), although the exact nature of the bond or bonds has yet to be elucidated. Siegel and Galston have shown that conditions of active aerobic respiration are necessary for the binding of indole-3-acetic acid to pea roots (51). Such conditions are also necessary for the ion-induced release of indole-3-acetic acid, judging by its abolition by KCN and the repeated failure to obtain it from non-living systems (53).

While it seems reasonable to assume that added indole-3-acetic acid is bound to protein in the same way (or ways) as naturally occurring indole-3-acetic acid, it should be emphasized that this is, in fact, an assumption. The experiments described on pages 37-40 indicate that positive air ions will release added indole-3-acetic acid from plant tissue. This is not quite the same thing as showing that positive air ions will release naturally-occurring bound indole-3-acetic acid from plant tissue. Obviously this will have to be demonstrated before the hypothesis can be accepted.

The first objective, therefore, of future air ion research should be to make accurate analyses of free and
bound indole-3-acetic acid, their precursors and metabolites, in both ionized and un-ionized plant tissues. Paper chromatography and refined micro-analytical techniques would be needed for the purpose. Such techniques could also be used to advantage in enzymatic studies related to indole-3-acetic acid levels; in particular, evidence should be sought for ion-induced alterations of indole-3-acetic acid biosynthesis and of the activity of IAA-oxidase.

A special attempt should be made to determine whether negative air ions have any effect on the plant cytochrome system. Negative ions have been shown to act on mammalian cytochrome oxidase and to accelerate cytochrome-linked oxidations in tissue homogenates (33), and it is possible that—even though negative ions have no demonstrable effect on plant growth—negative ions may cause measurable metabolic changes in plants without causing any overall growth changes, at least under the experimental conditions thus far employed.

After the above experiments have been carried out, attempts can be made to identify the auxin-protein receptor (or receptors) in the plant cell and to characterize the type of bond (or bonds) formed with indole-3-acetic acid.

Only by carefully determining exactly what air
ions do and how they do it will their real uses and limitations become known.
SUMMARY

The action of positively and negatively ionized air on plants was investigated. Positively-ionized air was found to stimulate the growth and movement of the blue-green alga, *Microcoleus vaginatus*; techniques for the quantitating of these effects were developed. The gaseous ion component of positively-ionized air responsible for the observed effects was shown to be positively-ionized carbon dioxide. This component also caused statistically significant enhanced growth in *Avena coleoptiles*.

Preliminary investigations into the mechanism by which these effects are produced suggested that positive air ions act on plant tissues by causing the release of bound indole-3-acetic acid, just as they appear to act on mammalian tissue by causing the release of 5-hydroxytryptamine. This hypothesis was partially confirmed by showing (a) that all of the positive air ion effects could be reproduced by indole-3-acetic acid, (b) that the oat coleoptile contains more than enough bound indole-3-acetic acid to produce the observed effects, and (c) that when indole-3-acetic acid is experimentally bound to corn kernels and coleoptiles positive air ions cause it to be released more rapidly than in control samples.
BIBLIOGRAPHY


5. The Bible, Old Testament, Exodus, Chapter 3.


24. Kluberg, T., and D. Ashbel. Effect of the Khamsin on ulcer patients, reported in Scope Weekly, the Upjohn Company, Kalamazoo, Michigan, June 18, 1958.


47. Pisani, S. Treatment of whooping cough by negative artificial aeroionization. La Semana Medica (Argentina) 52: 905-908, 1945.


57. Todd, G. W. Electric heat in schools can improve attendance record—control of positive ions in electrically heated air has been found to reduce sinus and respiratory ills. Electrical West 115 No. 4, 1955.


*Starred references were not checked from a primary source, but from quotations and summaries contained in the unstarred references.