8. B. B. Siegel, personal communication.
9. J. F. A. McManus and R. W. Mowry, *Staining Methods, Histologic and Histochemical* (Paul B. Hoeber, New York, 1960).
10. All pollen used in this study was purchased from C. G. Blatt and Co., Independence, Missouri.
11. M. F. Gridley, *Am. J. Clin. Pathol.* **23**, 303 (1953).
12. This work was supported in part by the National Aeronautics and Space Administration.

29 March 1963

Muscle-Equivalent Environmental Radiation Meter of Extreme Sensitivity

Abstract. *A 16.5 liter spherical ion chamber was constructed of conducting muscle-equivalent plastic and filled to 760 mm-Hg with a muscle-equivalent gas. The use of the chamber for measurements of natural environmental backgrounds was made quite feasible with the aid of the recently developed Shonka vibrating quartz fiber electrometer. This instrument is routinely operable at the extremely high sensitivity of better than 5000 divisions per volt. This system, therefore, has made possible reproducible measurements of absorbed dose-rates of fractions of a micro-rad per hour without any need for the usual corrections for wall-effect, stopping power, and so forth.*

The recent development by one of us (*1*) of a vibrating quartz fiber electrometer has made possible the construction of an uncomplicated, unpressurized, portable environmental radiation meter of extreme sensitivity.

The electrometer is routinely operable at the high sensitivity of better than 5000 divisions per volt. The sensitivity and balance adjustments may be made in the presence of a d-c signal on the fiber. The electrometer has an inherent capacitance of 1 to 2 picofarads and a rapid response with no detectable anistropy. Thus the instrument is an ideal detector for null measurements. A 3-lb transistorized power supply capable of operating the system continuously for 150 hours is in use for field measurements.

A 16.5-liter pseudosphere with walls 6 mm thick was welded from six molded sections of conducting muscle-equivalent plastic (*2*). The entire assembly contains no metal, and has a polyethylene guard-ring insulator and a molded polystyrene insulator supporting a thin central collecting rod, usually run at plus 80 volts and terminated by a thin-walled, hollow, tissue-equivalent sphere 4 cm in diameter. This was filled to 760 mm-Hg at 15°C with a stable, nonexplosive gas, muscle equivalent for photons and fast neutrons, which was formulated by Shonka, and consists of 41.11 percent neon, 39.59 percent ethylene, 16.17 percent ethane, and 3.13 percent nitrogen by volume. In this way the Bragg-Gray cavity principle applies without the usual stopping power corrections, and, furthermore, the cavity size restrictions were essentially removed.

Calibration of the chamber was carried out with a 1.11-mg radium needle (certified by the National Research Council of Canada) in 0.5 mm of platinum, with usual checks for saturation, scattering, and inverse-square law behaviors. An extremely precise and accurate determination of the ratio of muscle gas to air (designated "W")—the energy to create an ion pair—was made for us by W. P. Jesse of St. Procopius College, Lisle, Illinois. The volume and capacitance of the chamber were measured to better than 0.5 percent. The ionization rate calculated for our muscle chamber was compared with the ionization actually obtained. The agreement was good enough to assure that the ratio of electron stopping powers for our wall and gas is essentially unity.

The portability of the system and its extraordinary sensitivity of 0.33 mv/sec for 1 μrad/hour, enabled us to make measurements of environmental background in a skiff on Lake Michigan, on top of a ranger-type tower 40 m high, and at various land sites and in buildings within the Chicago area.

A value of 4.2 μrad/hour for the cosmic ray ionization at sea level was derived from the measurements made on Lake Michigan; the lake presumably shielded the device from terrestrial radiation and was itself assumed free of appreciable radioactivity. Corrections were made for the height of Lake Michigan (about 580 feet) and for radon content in the air, but not for geomagnetic latitude. When one considers the greater response of our chamber to low-energy radiation, the value is in reasonable agreement with those determined by other investigators. Solon (*3*), Burch (*4*), Neber (*5*), and Hess (*6*) obtained values of 3.8, 3.3, 4.7, and 3.4 μrad/hour, respectively.

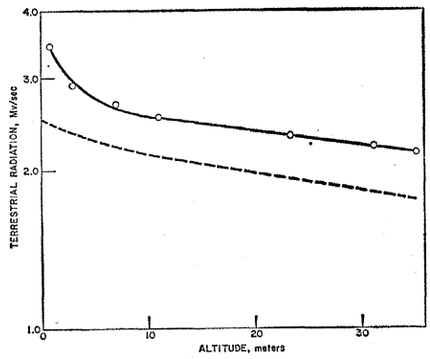

Fig. 1. Air attenuation of terrestrial radiation. Solid line, experimental data; broken line, theoretical, after Hultquist (*8*) and O'Brien (*9*).

Daily measurements in a first-floor laboratory over a period of months yielded a mean of 6.8 μrad/hour with a coefficient of variation of better than 0.5 percent.

The total radiation at points outside the building corresponded to about 14 to 15 μrad/hour, which is a shade higher than the values of 12 to 13 μrad/hour reported for this area by the Health and Safety Laboratory of the Atomic Energy Commission (*3*). The material in our building walls evidently attenuates the outdoor radiation considerably.

Measurements were carried out over a period of months in an Iron Room with an 8-inch wall. The mean of very reproducible readings was 1mv/sec, corresponding to about 3 μrad/hr. Thus the cosmic ray attentuation by the Iron Room is about 67 percent, which agrees very well with measurements made by F. W. Spiers (*7*) in Leeds, England.

Figure 1 displays the data obtained on the tower. Each point is the mean of about 15 measurements made at intervals which were weeks apart. The coefficient of variation of the data at each altitude is less than 4 percent. The ordinate has been reduced to net terrestrial radiation by subtraction of the cosmic ray contributions. The attenuation by air of terrestrial radiation has been calculated both by Hultquist (*8*) and by O'Brien *et al.* (*9*) on the basis of assumed concentrations of U, Th, and K. After filtration of the soft components by the first 10 m of air there seems to be substantial agreement between the observed and calculated attenuation factors.

The Shonka electrometer with its extreme sensitivity has made possible a system for the reproducible

measurements of absorbed dose rates of fractions of a microrad per hour. The apparatus is quite rugged, it exhibits no geotropism, is easily portable, and is battery operated. The electrometer and the muscle-equivalent, atmospheric-pressure, ion chamber seem to provide the simplest system (requiring the fewest corrections) for studying the dose to man from external environmental radiation, both natural and man-made.

JACOB KASTNER
JOHN E. ROSE
Radiological Physics Division, Argonne National Laboratory, Argonne, Illinois

FRANCIS R. SHONKA
Physical Sciences Laboratory, St. Procopius College, Lisle, Illinois

References and Notes

1. F. R. Shonka, *Radiology* **78**, 112 (1962).
2. ———, J. E. Rose, G. Failla, "Geneva conference paper," in *Progr. Nucl. Energy, Ser. XII*, **1** 753 (1958).
3. L. R. Solon *et al., U.S. At. Energy Comm., Rept., HASL-73* (1959).
4. P. R. J. Burch, *Proc. Phys. Soc. London* **67**, 421 (1954).
5. H. V. Neher, *Progress in Cosmic Ray Physics* (North-Holland, Amsterdam, 1952), vol. 1.
6. V. F. Hess and G. A. O'Donnell, *J. Geophys. Res.* **56**, 4 (1951).
7. F. W. Spiers, *Strahlentherapie* **111**, 65 (1960).
8. B. Hultquist, *Tellus* **1**, 54 (1952).
9. K. O'Brien *et al., Radiation Res.* **9**, 216 (1958).

10. Work performed under the auspices of the U.S. Atomic Energy Commission. Much of the initial impetus for this work was provided by the late Dr. G. Failla. We are grateful to William Prepejchal for helping both in the accumulation of data in odd places and in making the auxiliary apparatus more compact. We are grateful also to Richard York for his valuable contributions in the design and construction of the electrometer and chamber.

4 April 1963

Antimicrobial Substances from Aspen Tissue Grown in vitro

Abstract. *Isolated aspen tissue, when grown in vitro for 3 weeks on agar medium, yielded antimicrobial substances which produced inhibitory zones when the culture plates were inoculated with* Fusarium roseum, Saccharomyces cervisiae, Bacillus subtilis, Bacillus cereus, Penicillium roqueforti, Torula utilis, Sarcina lutea, Flavobacterium aquatile, Pullularia pullulans, *and* Staphylococcus aureus.

The medicinal properties of various plant materials and extracts have been recognized since the beginning of the 5th century B.C. This recorded information, the results of investigations performed in the late 19th and early 20th centuries, and the advent of penicillin and other antibiotics provided the impetus for the investigation of vast numbers of higher plants for antimicrobial substances. In 1943, 2300 species of plants were systematically tested for activity against *Staphylococcus aureus* and *Escherichia coli* (*1*). Numerous investigations dealing with the screening of plants for inhibitory substances have been conducted as a result of increased interest in the search for antimicrobial materials and they have been reviewed in detail (*2*).

The occurrence of antimicrobial materials in various members of the genus *Populus* have been reported by a number of workers. Boiling-water extracts of the bark have shown fungistatic and fungicidal activity against *Stereum purpureum* (*3*). *Populus candicans* and *P. trichocarpa* produced extracts with strong activity while *P. monilifera, P. eugenii, P. robusta, P. fremontii,* and *P. wislizenii* produced extracts with very little such activity. Extracts from *P. regenerata, P. serotina erecta,* and *P. marilandica* also showed some activity. A number of compounds with low fungistatic activity were extracted from the bark of *P. candicans* (*4*). These included pyrocatechol, salicin, saligenin, and salicylic acid derivatives as well as two highly active substances which inhibited the growth of *Botrytis cinerea, Penicillium italicum, Aspergillus niger,* and *Rhizopus nigricans*. Extracts from *P. balsamifera* and *P. gileadensis* were active against *Staphylococcus aureus* and in some cases *Escherichia coli* (*5*). Extracts from *P. alba* did not inhibit the growth of these organisms. Extracts of *P. fremontii* have also shown antimicrobial activity (*6*). The antibacterial properties of the sesquiterpenes in *P. tacamahaca* were investigated in 1957 (*7*). Antimicrobial activity has also been detected in extracts from *P. alba* (*8*), *P. tacamahaca* (*9*), and *P. trichocarpa* (*10*).

Pyrocatechol has recently been extracted and isolated (*11*) from the bark of aspen (*P. tremuloides*). It inhibited the growth of *Hypoxylon pruinatum*. Isolated tissues are a source of antifungal materials (*12*). Lilac phloem contains antimicrobial material which is inhibitory at naturally occurring concentrations, producing zones several millimeters in width when explants were grown for 3 to 15 days and then inoculated with *Cytospora* sp. Activity was also detected in extracts of phloem from *Fraxinus americana, Ligustrum vulgare, Ailanthus glandulosa,* and *Catalpa bignoniodes*.

Table 1. Inhibition of various microorganisms by isolated aspen tissue grown in vitro.

Organism	Fresh weight of tissue growth (mg)	Diameter of inhibitory zone (mm)
Fusarium roseum	204	27
*Saccharomyces cervisiae**	246	19
Aspergillus niger	91	0
Bacillus subtilis	77	23
Bacillus cereus	121	18
Proteus vulgaris	121	0
*Penicillium expansum**	178	17
Penicillium roqueforti	155	20
*Torula utilis**	107	15
Escherichia coli	257	0
Aerobacter aerogenes	91	0
Sarcina lutea	100	30
Serratia indica	98	0
Flavobacterium aquatile	110	18
Pseudomonas fluorescens	109	0
Pullularia pullulans	185	25
*Staphylococcus aureus**	208	23
Chaetomium globosum	181	0
Salmonella gallinarum	220	0
Hypoxylon pruinatum	171	0
Bacillus sp.	72	23

* Indicates variable results.

Much of the early work with antimicrobial substances from higher plants centered around the use of extracts and juices from intact plants. The growth of cultured isolated tissue from various plants may also be used in the detection of antimicrobial materials. This isolated tissue is of interest because it may be grown in large quantities (*13*) and may produce substances which differ from those in the intact parent plant (*14*).

The results of an investigation of the diversity of the antimicrobial activity from isolated aspen tissue grown in vitro are reported here.

Aspen tissue, originally isolated from the approximate cambial region of triploid stem sections on 26 December 1961, was grown on an agar medium containing major nutrients (*15*), trace elements (*16*), 3 ppm glycine, 0.1 ppm thiamine, 2 percent sucrose, 10 percent coconut milk, and 0.5 ppm naphthaleneacetic acid. Occasionally, cultures became contaminated with various organisms. One of these contaminants (*Bacillus* sp.) did not grow in the region surrounding tissue of either diploid or triploid origin which had been growing for 2 to 3 weeks. Measurements of the *p*H showed that a change in the acidity of the medium was not