effect may be obtained by hanging a white cardboard on a wire so that it may be drawn back and forth as needed.

Occasional subjects require a gray background. Any shade of gray may be obtained by varying the time for white and black. Obviously the longer the time with black, the shorter with white, the darker the background produced.

This method will eliminate the tedium of opaquing and blocking out of negative where black backgrounds have been found desirable in making the negative but undesirable in the print.

The writer intends to use a modification of the method for making photomicrographs. For this purpose it will be necessary to prepare a substage, the illumination of which may be alternated by white and black disks such as are provided on binocular microscopes.

FRANK P. MCWHORTER
OREGON STATE AGRICULTURAL COLLEGE

SPECIAL ARTICLES

COMPONENTS OF AIR IN RELATION TO ANIMAL LIFE

OXYGEN, since the time of Lavoisier, has been considered the vital component of the air. The 79 per cent. inert part has had little use assigned to it. Popular opinion had stated that animal life would be more efficient if these inert gases were replaced by oxygen.

Carefully conducted experiments, covering a period of eight years, have been carried out by the writer and his assistants.

A glass jar of five-gallon capacity was used as an air-tight container for the larger animals and a gallon bottle for the smaller ones. The gas was introduced through holes in a stopper fitting the larger mouth of the container, entering at the top and passing out near the bottom. The natural feed for the different animals and water were kept before them constantly. A small container of sodium hydroxide was placed at the bottom of the jar to take up any excess of carbon dioxide. The outflow of gas from the glass jar was passed through lime water to show that carbon dioxide was given off and the speed of the escaping gas.

The gas in the jar was frequently tested quantitatively, and the analysis showed that it never had an excess of carbon dioxide over that of ordinary atmosphere. A very accurate record was kept of the rate of breathing and the general appearance of the animals. Chemically pure oxygen was used.

Animals can not live in an atmosphere alone of oxygen, nitrogen, carbon dioxide, helium or argon. In a series of thirty experiments it was found that small animals such as mice, rats, pigeons, cats, guinea-pigs, snakes, monkeys, etc., can live in a medium of air under control, but in pure oxygen under the same conditions they will die within from two to five days. In only one case did any of the animals live over a week in oxygen—the snake lived four weeks—while in a current of air we had the different animals under control from one to three weeks without any signs of ailments. With the representative varieties of animal life it was found that in an atmosphere of pure oxygen, with other conditions normal, without a single exception every one would die in oxygen and none in air.

The breathing of the animals was usually found to increase gradually at first in the oxygen atmosphere, but later they appeared to breathe with more difficulty and more slowly until they died. From the appearance of their actions they did not seem to suffer much from pain.

AUTOPSY SHOWS HEMORRHAGE

An examination made by Dr. G. S. Terry of the lung tissue from a guinea-pig which had died in an atmosphere of pure oxygen showed marked evidence of inflammation and interstitial hemorrhage. Cultures made from the lung tissue showed a heavy infection of *Bacillus coli* associated with a few Staphylococci. The conclusion drawn from the autopsy was that an atmosphere of oxygen would not only rupture the lung tissue but also accelerate the growth of certain micro-organisms.

CARBON DIOXIDE AND OXYGEN

Animals were placed in an atmosphere of 99.97 per cent. oxygen and the normal .03 per cent. of carbon dioxide by volume. The animals used for these experiments were guinea-pigs.

With these experiments of carbon dioxide and oxygen the condition of the animals was found to be about the same as with pure oxygen. For this series of four experiments some one was watching them and taking observations day and night.

The general belief that animals could live with the normal amount of carbon dioxide that we have in the atmosphere added to the oxygen tested out to the contrary. Death followed in every case within two to five days as in the oxygen experiments.

THE EFFECT OF PURE OXYGEN UPON WATER ANIMALS

The experiments were continued with water animals in pure oxygen which was passed continuously through the water. The animals for these experiments were different kinds of fish, tadpoles, snails, newts and water turtles.

Some distilled water was boiled so that the mineral matter and most of the air would be removed to see if there could be any difference from that of tap water. The general results were found to be the same. These water animals tested out just the opposite from the land animals. In this case the pure oxygen could not have had such a burning effect when diluted with water which also soon became saturated with carbon dioxide.

AN ATMOSPHERE OF HELIUM AND OXYGEN

Seventy-nine per cent. helium and 21 per cent. oxygen form an atmosphere under which animal life will exist normally, or in some cases apparently even better. Mice were used for these experiments. Observations were taken every fifteen minutes day and night.

AN ATMOSPHERE OF ARGON AND OXYGEN

By using argon instead of helium and with the same percentage mixture the animals (mice) would not survive as they did with helium. The argon mixture would diffuse through the living cells less rapidly than the natural air and the helium more rapidly. The density of nitrogen compared to air is .967, that of argon 1.379 and helium .138, which might account for this difference.

By using 87 per cent. argon and 13 per cent. oxygen the mice would live forty-two hours. The respiration of the animals decreased slowly until death but without any apparent suffering. An atmosphere of 80 per cent. argon and 20 per cent. oxygen permitted life for ninety-two hours. Using 75 per cent. argon and 25 per cent. oxygen supported life normally, if not better than normal air, so far as we could observe.

An atmosphere made up of 66 2/3 per cent. argon and 33 1/3 per cent. oxygen supported life but not normally as with a higher per cent. of argon. The mice after six or seven days' confinement in such a mixture would be in poor health. The point of highest efficiency had apparently been passed.

AN ATMOSPHERE OF NITROGEN AND OXYGEN

Natural air contains 21 per cent. oxygen, 78 per cent. nitrogen and 1 per cent. mixture of argon, neon, krypton, xenon, helium and carbon dioxide. An atmosphere which contained 21 per cent. of oxygen and 79 per cent. of nitrogen by volume was prepared, leaving out the rare gases. The experiment was repeated six times. Blank tests with normal atmospheres were under simultaneous observation. White mice were used as experimental subjects. No difference in the physiological effects was noted because of insufficient atmosphere supply.

This series of experiments led to the conclusion that the rare air gases such as argon, helium, neon and carbon dioxide are vital for normal respiration. Without these rare gases included with the normal amount of nitrogen and oxygen, life would not exist longer than ten days. The rare gases seem to play a part in normal life equally as important as oxygen.

By increasing the mixture of oxygen to 25 per cent. and decreasing the nitrogen to 75 per cent. the animals were under control for three weeks without any signs of ailment. The experiments were continued by using mixtures as follows: oxygen 30 per cent. and nitrogen 70 per cent.; oxygen 40 per cent. and nitrogen 60 per cent.; oxygen 50 per cent. and nitrogen 50 per cent.; oxygen 60 per cent. and nitrogen 40 per cent., when the animals appeared to be normal and in a few cases better than in the normal air. By making a mixture of oxygen 70 per cent. and nitrogen 30 per cent. they seemed abnormal after being in this atmosphere for about a week. In an atmosphere of 80 per cent. oxygen and 20 per cent. nitrogen they would die in nine days, and they would live but seven days in an atmosphere of 90 per cent. oxygen and 10 per cent. nitrogen.

In an atmosphere of pure nitrogen alone the animals would live but six minutes; in argon alone, three minutes; in pure oxygen, from two to six days, and in hydrogen thirty-seven minutes.

From these experiments with pure oxygen and with the different mixtures of oxygen and other gases it seems that one might be able to find a mathematical curve between the mixture of gases and the time of living with animal life.

In a number of cases it was found that artificial atmospheres could be prepared that supported life in white mice more effectively than the normal air we breathe every day.

In the field of practical applications of prepared atmospheres there is a wide range of commercial uses and values. In deep-sea diving, mines and submarines foul air is encountered and there is often a lack of sufficient amount of air to sustain life. A prepared atmosphere for such activities would broaden their respective ranges of usefulness. An artificial atmosphere in a submarine that sustained life even more effectively than normal air would bring about a safer and more efficient submarine. A prepared atmosphere would be a great advantage to the high altitude fliers.

Medical men have a fair knowledge of the action of oxygen in the air, but little is understood by them concerning the other gases, especially the rare gases.

It is quite possible that a knowledge of atmosphere may aid in the control of diseases. The widest field probably will be in the pathological application.

J. WILLARD HERSHEY

MCPHERSON COLLEGE

SOME CONSTITUENTS OF DERRIS AND "CUBE"[1] ROOTS OTHER THAN ROTENONE

SINCE the introduction of derris root extracts as insecticides, rotenone has been considered mainly responsible for their activity. However, reports from entomologists and others interested in insect control indicate that derris extracts may contain little or no rotenone and still be effective sprays, and also that extracts from which rotenone has been removed as completely as possible are still effective. It may be assumed, then, that such preparations contain materials other than rotenone possessing insecticidal properties. In this connection it is significant that Greshoff,[2] Tattersfield and Roach,[3] Sillevoldt[4] and Durham[5] have isolated various products from derris root, some of which were toxic to fish or insects while others were not. None of these products, however, has been examined chemically.

Recently it has been shown that rotenone is present abundantly in the Peruvian fish poison "cube,"[6] extracts of which are used to some extent as an insecticide in South America. Like derris root, "cube" yields a large quantity of non-crystallizable material, which remains in the rotenone mother liquors.

In the course of a survey of fish-poisoning plants as sources of insecticides which is now being made in this laboratory, the non-crystalline extractives of derris and "cube" roots were studied to determine whether it was possible to obtain any material of definite composition that might be responsible for their physiological action upon fish or insects.

Without entering into details of the methods employed, which will be published elsewhere, it may be stated briefly that, when the non-crystalline material from all samples of derris root thus far investigated was dissolved in methyl or ethyl alcohol and treated with a small quantity of dry sodium carbonate or dilute sodium hydroxide solution, a number of well-defined crystalline compounds were obtained and that invariably three substances predominated. One of these, toxicarol, is a greenish-yellow substance which crystallizes in thin hexagonal plates that melt at 218°–20°. It is a monohydroxy dimethoxy compound, $C_{23}H_{22}O_7$. Another substance, with a pale green color, crystallizes in rodlike plates, having a melting-point of 171°. It is a dimethoxy compound, $C_{23}H_{22}O_6$, and is thus isomeric with rotenone. Recently toxicarol and this second compound have also been found in *Cracca* (*Tephrosia*) *toxicaria*.[7] The third substance crystallizes in short, thick prisms which melt at 198°. It is a dimethoxy compound, $C_{23}H_{22}O_7$, and is possibly tephrosine, which Hanriot obtained from the leaves of *Cracca* (*Tephrosia*) *vogelii*.[8] Although Hanriot reported the melting-point of tephrosine at 187° and assigned to it the formula $C_{31}H_{26}O_{10}$, his preparation was likely impure, consisting in all probability of a mixture of tephrosine and the 171°-melting compound, since the writer found that the leaves of *Cracca* (*Tephrosia*) *vogelii* gave a mixture of these two materials which had a melting-point of about 187°.

Besides the three substances just described a variable number of yellow or orange compounds were found in small quantities in the crude crystalline mixture, but since they are extremely difficult to separate in an analytically pure condition they have not as yet been investigated.

When the rotenone mother liquors from "cube" roots are submitted to the same treatment as described for derris, they also yield a crystalline mixture, which has proved to be either tephrosine (melting-point 198°) and the 171°-melting compound found in derris and cracca or a mixture of these two compounds and a yellow crystalline dimethoxy compound having a melting-point of 217° and the formula $C_{22}H_{20}O_6$.

The yields of these products, both from derris and "cube," are remarkably high. Some specimens of derris, which yielded less than 1 per cent. rotenone, gave by the alkaline treatment as much as 4 to 5.5 per cent. of the crude crystalline mixture, and the samples of "cube" thus far available yielded uniformly about 5.5 per cent. of a 2 to 1 mixture of tephrosine and the 171°-melting compound.

Further work is in progress upon these substances, but from the analytical data now available it appears that all these compounds, including rotenone, are more or less related, and that this small group of chemical compounds may be responsible for the toxic properties of many widely distributed tropical fish-poisoning plants.

E. P. CLARK

BUREAU OF CHEMISTRY AND SOILS,
U. S. DEPARTMENT OF AGRICULTURE

1 Pronounced coo′ bay. Recently Killip and Smith, *Wash. Acad. Sci.*, 20: 73, 1930, identified the plant as *Lonchocarpus nicou* (Aubl.) D. C.

2 M. Greshoff, *Pharm. Journ. and Trans.* (3), 21: 559, 1890.

3 F. Tattersfield and W. A. Roach, *Ann. Appl. Biol.*, 10: 1, 1923.

4 H. E. Th. Sillevoldt, *Ned. Tijd. Pharm.*, 11: 246, 1899; *Arch. Pharm.*, 237: 595, 1899; *J. Chem. Soc.*, A., 1: 109, 1900.

5 Durham, reported in footnote 3.

6 E. P. Clark, SCIENCE, 70: 478–9, November 15, 1929.

7 E. P. Clark, in press.

8 M. Hanriot, *Compt. rend. Acad. Sci.*, 144: 150, 1907; *Compt. rend. Soc. Biol.*, 62: 384, 1907.