medium was demonstrated in the vessels of the tube by x-ray.

A second method was based on the premise that if good anastomoses were present between the vessels of the flap and those of the heart, they might carry sufficient blood to support the tube with its pedicle divided. Accordingly, in an animal with a flap implanted for one month, the base of the pedicle was completely transected so that its only source of blood was through the surface of the flap attached to the heart. Arterial blood spurted from the distal cut surface of the tube, and one week later, when the animal was sacrificed, the tube appeared viable, with healthy granulations on its cut surface.

In another animal we observed direct evidences of anastomoses during a secondary operation, 1 month after a flap had been applied. The descending ramus of the left coronary artery was dissected out, ligated, and divided distal to the ligature. The distal cut end of the vessel spurted arterial blood which must have come either through the pedicle or from one of the other coronary arteries with the flap acting as a bridge.

In order to demonstrate the anastomoses visually, a method was developed by which radiopaque material could be injected through the vessels of the tube. The injection medium used was a 20% aqueous suspension of bismuth oxychloride, which has a particle size too large to pass through the capillaries, and will not pass through arterioles smaller than 35 or 40 μ in diameter. The technique consisted of isolating the coronary vessels by ligating the aorta immediately distal to their origin. The segment of aorta whose branches supplied the left chest wall and the vessels of the flap was then isolated by ligating the descending thoracic aorta above the diaphragm, and the injection was carried out through this segment under a pressure of 220 mm Hg. The injection mass flowed through the vessels of the chest wall into the vessels of the tube and then passed through numerous anastomoses to fill the coronary vessels. None of the injection medium was found in the veins because the bismuth could not pass the capillary barrier. Since the injection medium found in the coronary vessels was derived from the flap, the anastomoses were demonstrated to be at least arteriolar in caliber (Fig. 1).

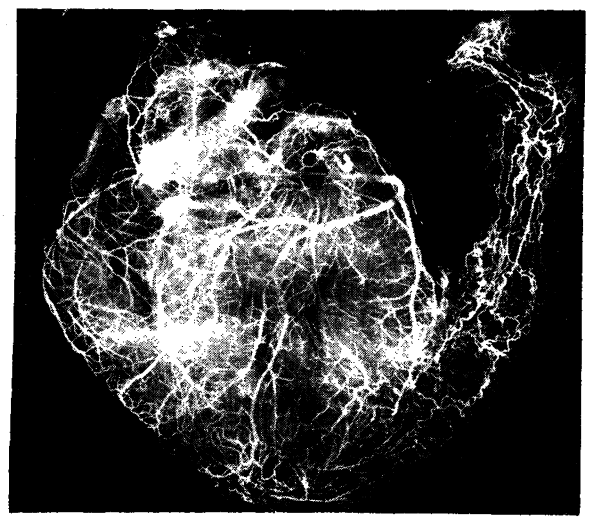

FIG. 1. X-ray of a heart to which a flap had been attached for 1 month, injected by method described in text. All the bismuth seen in the coronary arteries passed through the pedicle.

The experiments carried out so far have demonstrated that a skin flap may be attached to the heart without harmful sequelae, and that macroscopic anastomoses occur between the vessels of the flap and the coronary arterial circulation of the heart. Although three animals protected by a flap have had the descending ramus of the left coronary artery ligated without mortality, the series is too small to be significant, and further studies are under way to establish whether the flap carries a sufficient volume of blood to protect the heart against coronary occlusion.

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Abnormal Growth and Fructification of the Cultivated Mushroom

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Since the seventeenth century it has been customary to induce the development of the mushroom fruiting bodies, or sporophores, by covering the surface of mycelium-impregnated compost with a layer of loam about 1 in. deep. The function of this layer of soil in initiating fructification of the cultivated mushroom *Agaricus campestris* L. has never been proved experimentally. A study of abnormal growth may provide the entering wedge for understanding this function.

The stems of mushrooms growing on beds sometimes elongate abnormally, and at the same time, the growth of the caps is retarded. Lambert (*1*) claims to have demonstrated experimentally that this abnormal growth is due to the injurious effects of carbon dioxide released during respiration of sporophores. An evaluation of his results reveals, however, that there is slight, if any, elongation of the stem when the CO_2 introduced under the bell jars is derived from commercial cylinders; whereas severe abnormal growth occurs if the gases accumulate from respiration. It appears that in the gases accumulating naturally there may be another volatile substance which is responsible for this abnormal growth.

In this paper, an attempt is made to demonstrate that an elongation of the stem and retardation of the cap may occur even in the complete absence of carbon dioxide. The device employed to investigate this point (*2*) was a bell jar with two openings, one in the top and the other in the side at the bottom (Fig. 1, No. 3). When this kind of bell jar was placed over mushrooms

[1] The writer is indebted to J. R. Stauffer, University of Wisconsin, for facilities and aid.

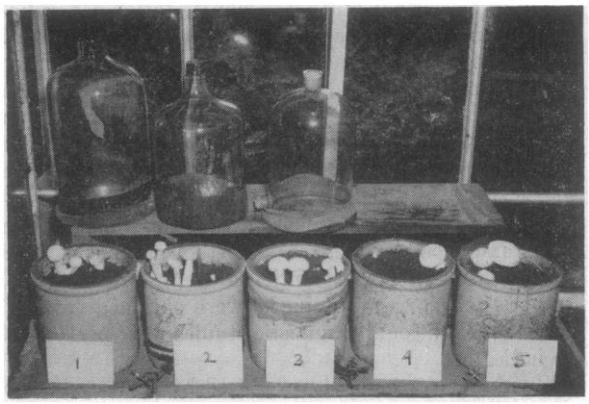

FIG. 1. Effect of volatile substances on growth of sporophores when covered with bell jars which have openings at top only, Nos. 1 and 2; at top and side, No. 3. Pots Nos. 4 and 5 are controls. (Jars were fastened to pots with rubber bands cut from inner tubes.)

growing in 2-gal earthenware pots, it was found that there was no accumulation of CO_2 in the bell jar as a result of respiration, whereas in bell jars where there was an opening only in the top (Fig. 1, Nos. 1 and 2), there was a considerable accumulation of CO_2. The openings in the jars were left *open* throughout the experiment, except momentarily when drawing samples for analysis. The atmosphere in the bell jars was analyzed for CO_2 with an Orsat apparatus (accuracy $\pm 0.1\%$). To withdraw 100-ml samples, a glass tube was inserted through a rubber stopper placed in the opening at the top of the bell jar and lowered to about an inch above the soil. The side opening in the bell jar with two outlets was closed with a rubber stopper during analysis. Prior to fastening the jars onto the pots, the surface soil was sprinkled with 50 ml water in order to enhance the release of CO_2 microbiologically. The CO_2 used was the gas naturally evolved from respiration rather than that obtained from commercial cylinders and injected under the jars, as in Lambert's experiments. Mushrooms were growing in the pin-size stage on pots selected for the experiments.

It is evident from Fig. 1 that abnormal growth was very severe at high concentrations of CO_2; however, the effect on growth was much more marked at 0.8–1.2% CO_2 in these experiments than in Lambert's, where the CO_2 was delivered from commercial cylinders. It may be observed from Pot No. 3, Fig. 1, and from Table 1 that mushrooms growing in the bell jar with two outlets have distinctly elongated stems and retarded caps, even though no CO_2 was detected in the jar. That CO_2 was actively evolving from the soil (even when no sporophores were growing) could be proved by closing the outlets of the bell jars with a rubber stopper. Under these conditions the CO_2 in the jars increased to 18% in 5 days. It may be inferred, therefore, that the CO_2 diffused out completely from the bell jar with two outlets, but only partially from the bell jars with one outlet; the amount of CO_2 accumulating in the latter depended on the diameter of the opening.

TABLE 1

ACCUMULATION OF CO_2 IN BELL JARS WHEN FASTENED ON POTS SHOWN IN FIG. 1. EXPERIMENT REPEATED FOUR TIMES

No.	Diam of opening at top of jar (cm)	No. of days (percentage CO_2)			
		1	3	4	6
1	5.5	0.8	1.1	1.2	1.2
2	3.0	1.6	2.8	3.4	4.0
3	3.0*	0	0	0	0

* Diam of opening at side, 1.5 cm.

From the fact that the mushrooms grew abnormally in the absence of CO_2, it may be hypothesized that another volatile substance affected the growth. Carbon dioxide is a small molecule, with a small relative density or molar weight, with a very low boiling point, and a gas at ordinary temperature. Accordingly, the rate of diffusion of CO_2, considering Graham's law of diffusion, would be expected to be rapid, especially through the "chimney" effect of the bell jar with a lower and upper opening. Reasoning similarly for the hypothetical volatile substance, it may be considered to have a large molecular weight, with a high boiling point and low volatility, so that its rate of diffusion would be much slower than that of CO_2. Since the latter diffused slowly, a sufficient concentration was present in the bell jar, especially surrounding the sporophores, to affect the nature of their growth. Thus, by means of a bell jar with two outlets, it was possible to arrange for a differential rate of diffusion.

A hypothesis may be proposed that some volatile substance emitted from the mycelium in the compost and in the casing soil accumulates when there is insufficient ventilation or when the mushrooms are growing thickly on the beds. Mader (*3*) also found that "wherever mushrooms are grown, substances of a volatile nature accumulate and become detrimental to their growth and fructification," although the writer is at a loss to understand his experimental procedure. A volatile substance or substances emitted from the mycelium have been detected by absorbing solutions of guaiacum resin or benzidine in pure filter paper, and then suspending the papers in bell jars over a pot of mycelium-impregnated compost (*4*). The papers become colored blue and brown, respectively, indicating that the substance volatilizing is of an oxidizing nature. Adequate controls were carried out simultaneously by suspending papers containing the reagents in jars over pots of water. Similar color changes were produced in agar slants as "halos" advancing ahead of the growing mycelium, with these reagents as well as with many others.

This volatile substance may also give some indication of a defensive mechanism (*4*). For example, when lead acetate test papers, darkened (showing evolution of a sulfide) by suspending them over cultures of *Pseudobalsamia microspora* (a pathogen of the mushroom mycelium), were suspended over mushroom cultures by the same procedure described above, the dark-

ened papers were decolorized. It was suggested this substance is in the nature of a quinone.

The significance of this volatile substance may be, then, that it acts like an "antibiotic" or defensive mechanism for mycelial growth; that it inhibits fructification unless it is modified in the casing soil; and that it affects normal growth when allowed to accumulate. It is of interest to note that pathogens which do not attack the mycelium in the compost, do so in the casing soil—that is, in the medium where the antibiotic value of this volatile substance is at the lowest ebb. The intensity or high potential of the substance diffusing from the mycelium was shown by embedding oxidation-reduction indicators in agar slants (*5*). It appears that the oxidizing intensity of this substance is so great that it prevents the thickening of the hyphae from which sporophores develop. The necessity for adjusting the soil at a relatively high pH for best fructification tends to show that the soil must be a medium suitable for rapid oxidation-reduction reactions. A movement of air or oxygen-ventilation is, of course, required for such reactions. Accordingly, at least one of the functions of the casing soil is to provide an alkaline-oxygenated medium for the destruction of this volatile substance. A fuller presentation will be published elsewhere.

References

1. LAMBERT, E. B. *J. Agr. Research,* **47,** 599 (1933).
2. STOLLER, B. B. Ph.D. Thesis, Univ. Wis. (1945).
3. MADER, E. O. *Phytopathology,* **33,** 1134 (1943).
4. STOLLER, B. B. *Ibid.,* 13.
5. ———. *Abstr. Am. Soc. Plant Physiol.* (1942).

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The Medical Examination of Hiroshima Patients with Radiation Cataracts[1]

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Radiation cataracts, the first late manifestation of exposure to atomic bomb radiation reported in man, have been described by Cogan, Martin, and Kimura (*1*), and Cogan, Martin, Kimura, and Ikui (*2*). Kimura (*3*) reported 98 Hiroshima patients with radiation cataracts. Eighty-five of the 98 were among 922 survivors 1000 m (1094 yards) or less from the hypocenter,[3] an incidence of 9.8%.

This group of 98 patients with radiation cataracts is a select group because all were unquestionably exposed and all show unmistakable delayed response to radiation injury. Seventy-eight of them have been examined by the medical department of the Atomic Bomb Casualty Commission Clinic in Hiroshima in an effort to discover if other late manifestations of radiation injury exist. The examinations were performed approximately five years after exposure.

The group comprised 45 men and 33 women between the ages of 12 and 69 years, and they had been from 150 to 1240 m from the hypocenter; 64 were between 700 and 1099 m (710–1202 yards). Detailed radiation and medical histories were obtained, and physical examinations were performed. Roentgenograms of the chest and blood, and urine and stool specimens were examined. Thirty-four patients were proctoscoped, and sternal marrow specimens were obtained from 27 patients. Additional diagnostic studies were performed as indicated.

Symptoms presumably produced by acute radiation were vomiting, fever, diarrhea, oropharyngeal lesions, bleeding gums, purpura, epilation, amenorrhea, and abnormal periods after amenorrhea. At least 50% of the patients experienced fever, purpura, epilation, amenorrhea, and vomiting on the day of exposure, suggesting that at least half of them received severe irradiation.

Forty-six of the 78 patients received mechanical injuries described as minor lacerations and penetrating wounds from flying glass and other small objects. Several patients were bruised by falling beams and roofs, but the only major injury was a depressed skull fracture. As far as could be determined, all injuries resulted from indirect blast effects. Small thermal flash burns occurred in 15 patients. Five of the burned patients were free from flash burn scars, suggesting partial shielding from thermal injury. One scar resembled a keloid.

Seventy-seven of the 78 patients experienced scalp epilation. This observation suggests that a cataractogenic dose will produce some degree of scalp epilation in the majority of patients.

Two males and one female above 39 years of age had diastolic blood pressures of 100 mm Hg or higher. The remaining individuals had blood pressure recordings within the usual range. Thirty-six patients had scars from burns and/or wounds sustained at the time of the explosion. The majority were minor and showed only small amounts of scar tissue. Radiation cataract was the only physical finding attributed to the late effects of the atomic bomb.

At the time of the study the hematological findings did not disclose any blood dyscrasias. Two patients with radiation cataracts have developed acute leukemia, one of them subsequent to her examination in this study and one child not included in this report. The aspiration sternal marrow specimens obtained on 27 patients were compatible with the peripheral hematological findings.

Chest films, stool and urine examinations, and serological tests for syphilis revealed no abnormalities that could be attributed to the atomic bomb. The histories did not reveal any information which suggested late effects of the atomic bomb other than visual complaints.

The shielding factor was not studied. The greatest

[1] Sponsored by the Atomic Bomb Casualty Commission, National Research Council, with funds supplied by the U. S. Atomic Energy Commission.

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[3] Point on the ground directly below the explosion.