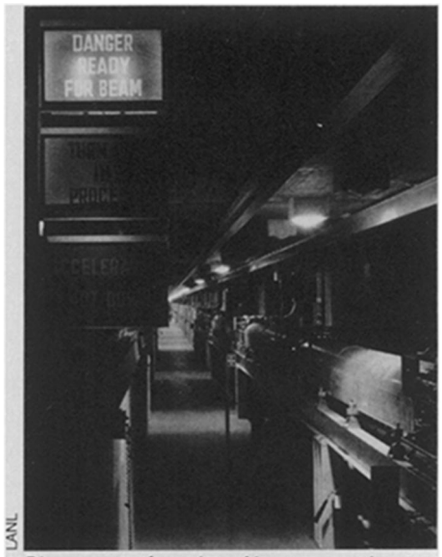
HEAVY RADIATION AND MAMMALIAN CELLS

To use radiation heavier than X-rays in cancer treatment, scientists must know what it does to cells



Pions come from Los Alamos accelerator.

First of two articles

By DIETRICK E. THOMSEN

The use of ionizing radiation in biology and medicine has always cut two ways. In her later years Marie Curie was a famous advocate of the medical use of radium, which she and her husband had discovered, but she also died of the effects of handling it. Ionizing radiation can destroy tumors; it can also create them.

Today physicians are beginning to experiment with artificially generated forms of radiation that were not available when the Curies flourished. These include subatomic particles such as neutrons and pions, and most recently ions — that is, atomic nuclei of various elements. Yet the double effect is still there. At the recent Symposium on Heavy Charged Particles in Research and Medicine, held at the Lawrence Berkeley Laboratory (LBL) in Berkeley, Calif., speakers dealt with both aspects. For instance, irradiation with helium or hydrogen ions has been successful against certain eye tumors known as choroidal melanomas (SN: 9/24/83, p. 204), but the same radiation can also cause cataracts.

So far the newer kinds of radiation have been found effective mainly against the same classes of tumors for which therapists have been using X-rays and gamma rays since the days of the Curies. Several speakers expressed disappointment at this, as therapists had hoped for something to use against the varieties of tumor known as "radiation resistant," which include some of the most common ones. Experimentation with neutrons, pions, hydrogen ions and helium ions so far has not found such a treatment. Experimentation with ions heavier than helium is just beginning.

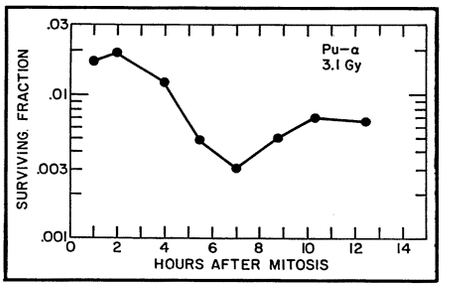
Disappointments notwithstanding, these heavy-particle radiations — heavy compared with X-rays and gamma rays, which have no rest mass — are finding a niche in the treatment of cancers in certain difficult locations. X-rays and gamma rays deliver energy more or less evenly as they traverse flesh, from their entry into the body to their exit. The charged particles, pions and ions, deliver most of their energy at the ends of their trajectories. Protons and ions are energized in accelerators, and pions and neutrons are made when energetic proton beams strike appropriate targets. Varying the energy of the accelerator varies the energy of the outcoming particles and so can determine how far into the body they penetrate. Magnetic fields can guide the charged particles.

All this means that the charged particles have shown themselves useful in treating tumors in locations near vital organs where X-rays or gamma rays might do unacceptable damage to those organs. Tumors in the eye, brain, liver and pancreas and those wrapped around the spinal cord are among the ones that have been experimentally treated with the heavy particles.

To design treatment plans, researchers must first know something about the action of these radiations at the cellular level. So must the planners of space flights, since astronauts are subject to bombardment by heavy ions in the cosmic radiation. "Iron dominates above the magnetosphere," says Paul W. Todd of the Pennsylvania State University in University Park. At the symposium he showed tracks of iron nuclei superimposed on groups of cells.

Experiments of this sort use colonies of cells grown *in vitro* as targets for radiation. Favorite varieties include yeast spores, mouse embryo cells, Chinese hamster cells and human T1 cells. As Todd's pictures show, a single iron nucleus can hit several cells. If it goes through a cell nucleus, it may damage the cell's DNA. If it misses the nucleus, it can do other damage. These are ionizing radiations — that is, they easily knock electrons out of atoms and molecules. Some of those electrons are hit hard enough to go off laterally (making tracks called delta rays) and penetrate cells not hit by the primary ion beam, where they may do some damage. Ionization of water, the most prevalent fluid in biological structures, can produce negatively charged hydroxyl ions. Hydroxyl ions are very reactive chemically, and some of the reactions they cause can also damage DNA.

A popular question, says Todd, is what happens to a cell that is struck by a heavy particle and doesn't die. He emphasizes the importance of the question by pointing out that in the space environment, the radiation running through our galaxy bombards a surface of 1 square centimeter in 1 hour with 8 particles that deliver more than 100,000 electron-volts of energy per micrometer they traverse. Of



Todd

Survival of cells struck by helium nuclei depends on their stage of life.

these, 2 in each hour on the average will deliver more than 200,000 electron-volts per micrometer. (The energy delivered per unit length of path, electron-volts per micrometer, is called the linear energy transfer, or LET, of the radiation.)

Experiment first had to determine whether cells can survive a hit through the nucleus. If a hit is always a kill, there's no question of what happens to survivors. In fact, as more than one speaker reported to the meeting, up to 80 percent of cells do survive direct hits. There appear to be two populations of cells, Todd says: the sensitive and the nonsensitive. For a sensitive cell, one hit is enough to cause a biological effect.

"A single hit by a charged particle can cause a single cell effect," agrees Victor P. Bond of Brookhaven National Laboratory in Upton, N.Y. The result for a cell is that "it dies or does not die." If it lives, "it is mutated or not." If it is mutated, "it is carcinogenic or not." There is no middle ground for any of these choices, Bond says.

"The response is a statistical phenomenon," he adds, and from those statistics he is engaged in the complicated task of trying to find a "hit size efficiency function," which will yield a single number to represent the amount of physical disturbing agent to which the cells are exposed in any single instance and the total probable damage to the cells involved and to the corresponding organ. One possible quantity, he suggests, is the primary particle fluence, the number of particles striking a given area of surface in a given time.

The damage of greatest interest is injury to the cell's DNA. This comes in two forms: breakage of one or both DNA strands or chemical alteration of the base pairs out of which the DNA is constructed. Both kinds of damage can be caused by direct ionization by the radiation or by hydroxyl radicals generated in the cell's water by the passage of the radiation, says John F. Ward of the University of California, San Diego, School of Medicine.

Cells do not take such damage passively; they have their repair mechanisms. However, repair does not always mean return to the status quo ante. "It is clear that damage in both members of a base pair leads to loss of information and can only be repaired correctly by accident," Ward says.

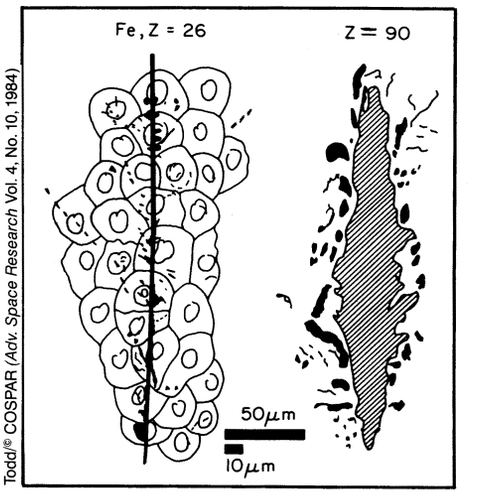
Misrepair can cause mutation, and mutants may be the start of a tumor. This can be a problem as a side effect of therapy and in space medicine. An interest in the potential cancer-causing effects of cosmic rays on space travelers prompted Tracy C. Yang of the Lawrence Berkeley Laboratory and collaborators to study cell transformation by charged heavy particles in cultured mammalian cells. They bombarded mouse embryo cells known as C3H10T½ with ions of carbon, neon, silicon, iron and uranium. They also bombarded the cells with X-rays by way of comparison.

"Transformation is not as easy as killing," Yang says, but nevertheless they found significant cell transformation by all these species of radiation. Argon ions at an energy of 330 million electron-volts (330 MeV) for each neutron and proton deliver an LET of 140,000 electron-volts per micrometer. They proved much more effective at causing cell transformations than X-rays. On the other hand, uranium at 400 MeV per neutron and proton, which delivers an LET of 1,900,000 electron-volts per micrometer, was only 0.7 times as effective as X-rays. Effectiveness seems to peak at about 200,000 electron-volts per micrometer LET.

However, the big surprise came when they mixed X-rays and ions. Transformation was enhanced in all cases. This leads them to conclude that the lesions caused in cells by ions and those caused by X-rays interact. "Transformation is a multistep process," says Yang, "and it can be treated with chemicals." The chemical was a 0.5 percent solution of dimethylsulfoxide (DMSO). It brought about a significant suppression of transformation.

DNA may not be the only thing in the cell that heavy particle radiation damages. Heidi Fritz-Niggli of the Radiobiological Institute of the University of Zurich says: "For years we [have] suggested that besides the DNA also the repair systems and (or) membranic systems could be injured by radiation...." She described experiments involving repair-proficient and repair-deficient strains of cells from the fruit fly *Drosophila melanogaster* that she and colleagues irradiated with pions from the piotron accelerator at the Swiss Institute for Nuclear Research in Villigen. The results bear out the statement, she says.

Eleanor Blakely and her colleagues at LBL studied cells' responses to bombardment by X-rays and neon ions at different times in their life cycles. They found that cells were capable of a "significant" amount of repair after irradiation with X-rays. After irradiation with neon the younger cells, in early and middle stages of the part of their life cycle called G1, could manage only a negligible amount of repair. "Only late G1-phase cells repaired neon damage," they conclude. Introduction of the DNA polymerase inhibitor β-ara-A decreased repair in all cases.



Todd/© COSPAR (*Adv. Space Research* Vol. 4, No. 10, 1984)

Track of iron nucleus superimposed on colony of cells and track of heavy (z = 90) nucleus at same magnification. Lateral blobs and dashes are delta rays.

Finally, it seems that repair is also dependent on the size of the radiation dosage. Dudley T. Goodhead of the Medical Research Council Radiobiology Unit in Harwell, England, remarks that investigators "have usually assumed that cell repair is dose independent. I want to contest that." He proposes instead a model in which repair capability declines as dosage increases. In support, he cites experimental evidence that appears to show "a rate of repair that decreases as dose goes up."

All this makes, as Blakely puts it, "a problem for clinicians." To use these radiations for medical therapeutics, clinicians must design treatment programs that stand a good chance of killing tumors without inducing too many complications. "Which is the best plan to treat a patient?" asks John T. Lyman of LBL. To figure it out the therapist must pay attention to healthy tissue as well as to the tumor, keeping track of the dose delivered to a given volume.

One has to introduce the concept of a tolerance dose for the tissue in question, Lyman says. The probability of complications then depends on the relation between the tolerance dose and the dose actually delivered to a given volume.

"If you have a treatment plan with a 5 percent complication probability," he says, "a 5 percent error in dosimetry doubles the complication probability. If your tolerance dose is 5 percent lower than you think, it doubles the complication probability." Finally, "If you have not estimated the volume of an organ properly, a 10 percent change of volume treated doubles the complication probability." And he concludes that "the consequences of being a little bit off can make a significant difference in the outcome of treatment." □

NEXT: A survey of experimental treatment programs at various accelerators around the world