CLINICAL SECTION

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Summary of Proceedings of the Bone Marrow Transplantation and Chemical Radiation Protection Conference

(Conference Organized by Members of the Biology Division of Oak Ridge National Laboratory and Held in Oak Ridge, Tennessee, on January 9 and 10, 1959; Co-Chairmen, Alexander Hollaender and C. C. Congdon)

Transplantation of Bone Marrow

Radiation accidents.—Flynn outlined in detail all the available information on the Los Alamos radiation accident of December 1958. The accident occurred in a plutonium salvage operation. Supportive therapy probably prevented the exposed person from dying almost immediately from a shock-like syndrome. He died 36 hours after the accident, from central nervous system damage. Vos reviewed newspaper reports on the Yugoslavian reactor accident. Several of the victims received bone marrow transplantation and are recovering. One man died shortly after bone marrow therapy. According to a personal communication received by Dameshek, there was transplantation of blood-forming cells. Bone marrow therapy was delayed for several weeks in these accident victims. The problem of delay in bone marrow injection was discussed later in the session. Andrews reviewed the health status of the persons exposed in the June 16, 1958, Y-12 (Oak Ridge) radiation accident. Hematologic recovery was excellent, but some individuals still complain of aches, pains and general muscle weakness. Fliedner described the effect of the accidental exposure on the mitotic index in the bone marrow of these patients. He thinks this index may help in evaluating the degree of exposure.

Bone marrow procurement in man.—Repplinger described a technic for removing marrow cells from the vertebral bodies of cadavers. As many as 130 x 10^9 cells per vertebral column could be obtained. Schwartz and Dameshek pointed out that they were now able to obtain large amounts of marrow by aspiration from living donors. Other discussion of tissue culture and preservation followed. Bender felt that tissue culture viability was the best method to test for living cells. Rinfret discussed engineering research at Linde Company on apparatus for low-temperature preservation.

Bone marrow transplantation in man.—Wilson and Dummin reported the clinical and pathologic findings in two patients given total-body irradiation and homologous bone marrow injection. Kidney homografting was performed in one patient but could not be carried out in the other. The homografted kidney did not show the expected graft rejection reaction during the 28 days the one patient lived. In neither case could bone marrow transplantation be
demonstrated. Multiple marrow donors were used in these two cases. The radiation exposures were 600 and 700 r.

Dameshek and Schwartz reviewed case histories of aplastic anemia patients that received bone marrow injection. In about one-third of the cases there was evidence of recovery in peripheral blood elements; in occasional instances, a possible myelostimulatory effect of the marrow injection was noted.

Delayed injection of bone marrow in animals.—Vos said that the Dutch research group was trying to decide, on a cell dose requirement basis, whether a one day injection was superior to a 0 day treatment. The one day delay seemed superior, but the results were not entirely clear cut. Congdon pointed out that, in isologous experiments, good 30 day survival with optimal bone marrow doses could be obtained for four days after exposure and that the work of Dejessari suggested that delay of seven days still allowed some survival with supraoptimal doses of bone marrow. In Lorenz’s work with chronic gamma irradiation of guinea pigs, the bone marrow injection was delayed for about three months and still promoted greater survival in the treated group. All this work deals with experiments in which the donor and recipient had no genetic incompatibility. In homologous and heterologous experiments, it is still not clear whether delay helps to prevent the foreign bone marrow reaction. In general, it seems inevitable that some short delay will occur in getting bone marrow therapy for accident cases where this treatment might be lifesaving. Overman said that at 550 r, an LD10 in his monkeys, a delay of 2 to 3 days in injection of homologous bone marrow gave better survival than injection 24 to 30 hours after irradiation. Fliedner thought that his anatomical studies in irradiated bone marrow pointed to the desirability of quickly replacing the bone marrow mass to preserve the integrity of the thin, unsupported vascular wall.

Radiation dose needed for homologous bone marrow transplantation.—Congdon pointed out that there was now very good evidence from experiments on several species to show that the radiation dose for successful homologous bone marrow transplantation must be very high—in the range 800-1000 r. The LD50/30 day figure for a species does not help us to decide what exposure to use for foreign marrow transplantation. At this very high radiation dose, the homologous bone marrow dose must be adequate to prevent radiation death.

Bone marrow transplantation in large animals.—McAlpine summarized the marrow transplantation research program at Parke, Davis & Company. Monkeys given autologous marrow survived after exposures of 600 r; none injected with homologous marrow survived. Rat marrow was not effective in irradiated monkeys. Overman reported evidence of a take of Rhesus bone marrow in an irradiated cynomologous monkey after 700 r. A hemoglobin marker was used.

Immunologic problems.—Wilson presented evidence from bone marrow homotransplantation and skin homografting in irradiated rabbits indicating that multiple bone marrow donors were not absolutely contraindicated, as had been postulated by others. He found a second small dose of radiation, 100 to 300 r, to be helpful in stopping “secondary disease” in rabbits. Short courses of amethopterin, according to Uphoff, prevented the “secondary disease” in a few
F₁ hybrid mice given parent-type bone marrow after total-body irradiation. Schwartz reported the suppression by 6-mercaptopurine of precipitin antibody formation in rabbits. He further showed that this was not related to an effect on the reticuloendothelial system. Cosgrove showed survival data on sensitizing the parental donor to host-type tissues before injection of bone marrow into an irradiated F₁ hybrid mouse. If the sensitizing tissues were related to the irradiated host, the marrow had a very detrimental effect on survival. The effect was not detrimental, however, if the sensitizing tissues were unrelated to the host.

Transplantation of fetal blood-forming cells.—Simmons commented on the long-term survival of irradiated rabbits that were treated with suspensions of mouse or rabbit embryo. Some male survivors, after treatment with rabbit embryo material, showed female granulocytes. Congdon pointed out the advantages of fetal homologous liver over adult bone marrow in lethally irradiated mice. Owen described the identification of donor-type red blood cells in mice treated with homologous fetal liver.

Neutron exposure.—Vogel discussed the effect of bone marrow therapy on neutron-irradiated mice. Streptomycin was an important adjunct in his experiments, probably due to its protective effect on the damaged gastrointestinal tract. The best survival was obtained with chemical protection before neutron exposure and bone marrow-streptomycin therapy afterward. Neutron-irradiated mice given homologous marrow did not develop secondary disease.

Myleran injury to bone marrow.—McAlpine gave a detailed report of radiation versus Myleran effect on differential bone marrow counts in rats with and without marrow therapy.

Chemical Protection AET

Toxicity in man.—Schlosser reported his preliminary experiments with AET toxicity in human beings and the use of AET before x-irradiation or nitrogen mustard therapy. Thirteen persons received AET orally; the highest dose given was 1200 mg. Some patients were nauseated and had epigastric pain and diarrhea; but the symptoms did not correlate with the dose. Hypotension was not observed. The drop in white blood cell count was studied in cancer patients given AET before X irradiation or before mustard therapy. It was difficult to determine whether or not these doses of AET gave significant protection. Overman suggested that the oral AET tablets on an empty stomach might have contributed to the gastrointestinal toxicity. Dameshek thought that the reticulocyte response might be the most sensitive end point to follow for protective effects.

Zubrod said that intravenous AET had caused severe hypotension in one patient. He advised continuing the search for less toxic compounds than AET. It was apparent from the discussion that more information about toxicity in man is needed.

*The following abbreviations are employed: AET=S₂, Aminoethylisothioronium. APT=S₃, aminopropylisothioronium. PAPP=Paraaminopropyl phenone. APMT=a methylated derivative of APT=compound 62.
Pharmacology of active compounds.—Di Stefano described the basic pharmacology of AET as seen after intravenous administration in the cat. There were striking effects on blood pressure, respiration, heartbeat, gut contraction and ganglionic response. APT gave about the same effects. In contrast, APMT was nearly inert pharmacologically and hence is of great interest as a radioprotective chemical.

Protection in dogs and mice.—Overman reviewed his experiments with AET protection in dogs. In some experiments, he combined AET with PAPP and obtained good protection at 500 r. Kelly reviewed her studies on AET protection against nitrogen mustard toxicity in mice and dogs. In AET-pretreated mice there was marked histologic protection against gut damage from nitrogen mustard. The bone marrow also was protected. AET seemed to protect some tumors, as well as normal tissues, against nitrogen mustard or x-ray effects. Other tumors were not protected, and mice having these types showed improved survival from tumor death. Autoradiographs of tissues of mice injected with isotope-labeled AET showed poor localization in tumor tissue, whether primary or metastatic. White pulp of the spleen also showed poor uptake of labeled AET. Congdon pointed out that the spleen study correlated with the functional findings by Makinodan and associates that AET did not protect the immune mechanism from radiation as well as might be expected on a dose-reducing basis. Doherty also commented that there was some delay in skin homograft rejection in AET-protected irradiated mice as well as those given isologous bone marrow after exposure.

Doherty reviewed his current studies on chemical protection. AET and APT seemed to give about the same level of radiation protection. A new effective compound no. 62 (APMT) is not quite so potent as AET but is much less toxic. The bone marrow response of AET-treated irradiated mice (900 r) was directly proportional to the amount of AET used. At high AET levels the minimum count was $5 \times 10^6$ nucleated cells; the count returned to normal by the fifth or sixth day. In contrast, there was no apparent relation between the APMT dose and the bone marrow response, protective doses bringing about a rapid but delayed recovery. Maisin said that both gross and histologic studies showed that AET protected against gut damage at 2000 r. At this radiation level, however, mice did not survive. Smith discussed AET radiation protection of bone marrow suspensions in vitro. No direct toxicity of AET to the bone marrow cells was seen. Good protection was observed in vitro. Upton reported that AET did not seem to protect against radiation-induced nephrosclerosis. He also reported that, in preliminary experiments, AET in the drinking water was not influencing survival time during chronic irradiation at 80 rads per days. Further experiments on the influence of AET and APT on survival during chronic irradiation are planned. Doherty thought that the AET was probably undergoing too rapid decomposition in the drinking water. He plans to incorporate it into dry food pellets. Doherty also reported that AET and APT gave some protection against fast neutron lethality. Anderson pointed out that experiments on chemical protection and bone marrow treatment of irradiated monkeys are being started at the School of Aviation Medicine.