The Pathogenesis of the Hemorrhagic State in Radiation Sickness: A Review

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The importance of hemorrhage as a part of the acute radiation syndrome is well recognized. Warren and LeRoy cite bleeding as a major cause of death in radiation sickness among the Japanese exposed to atomic bomb radiation. The frequency of hemorrhagic manifestations has been emphasized by Cronkite.

Although bleeding is a more or less constant feature of the acute radiation syndrome, its presence and degree are dependent on species differences, dose, and time. The influence of these factors has often been overlooked, as a result of which confusion exists in the literature. Hemorrhage is more widespread and severe in man, the dog, and the guinea pig than in the mouse, rat, or rabbit.

The early clinical and experimental reports have been cited elsewhere.

In the present review emphasis will be placed on recent studies of the pathogenesis of radiation-induced hemorrhage.

Pathogenesis

Although the hemorrhagic aspect of acute radiation injury has been familiar for many years, the pathogenesis of the bleeding is still not entirely clear. Three major etiologic factors have been postulated. These are (1) thrombocytopenia, (2) capillary fragility, and (3) clotting defects unrelated to platelets.

Although it is now evident, from the studies of Cronkite and Brecher, that deficiency of platelets plays a crucial role in the development of the purpura, the relative importance of vascular fragility and of clotting disturbances as such is not yet established.

Vascular Factors

Little is known about the actual process of irradiation hemorrhage itself. The nature and site of the vascular defect are unknown. The characteristically positive tourniquet test suggests the existence of vascular fragility; however, what this denotes in terms of alteration of the endothelial cells, the interendothelial cement, or the pericapillary sheath is not clear.

Diffuse hemorrhages in the peripheral sinus of lymph nodes are typical of this disorder and have been correlated by Furth and co-workers with the occurrence of blood cells in the lymph. These investigators inferred that the capillaries were permeable to erythrocytes, as well as to macromolecular substances,
and thought that blood cells leaked into the tissues, were then collected in the lymphatics, and subsequently returned, at least in part, to the blood stream via the thoracic duct. This process was associated with a significant reduction in the circulating red cell mass and was envisioned as affecting the capillary bed throughout the body, a view consistent with the diffuse distribution of gross hemorrhages observed in dogs by Allen.10

Direct microscopy of the blood vessels has yielded thus far little information of help in explaining the bleeding. Haley et al.11 observed in rats no effect of irradiation on the terminal arterioles or precapillary sphincters of the mesentery but noted release of a vasodepressor material from the liver during the first week after total-body exposure to 600 r of x-rays. This substance had the characteristics of ferritin. Conversely, during the second week a vasoexcitor material (VEM) made its appearance in the blood, having the properties of the VEM observed during the reversible phase of shock. Mason et al.12 reported abnormal reactivity of isolated segments of the carotid arteries of dogs irradiated with 250–550 r, associated with histologic changes suggestive of muscle cell degeneration in the tunica media. These observations do not, however, elucidate the mechanism of bleeding.

Reduction in the severity of bleeding by the postirradiation administration of flavonoids has been reported13-18 and disputed15-19. Field and Rekers20 observed a synergistic effect of ascorbic acid when it was added to rutin. These findings, although inconsistent, point toward the possible importance of abnormalities of the interendothelial cement or pericapillary sheath in causing the observed vascular fragility.

The great importance in hemostasis of the vascular mechanism per se is indicated by the microscopic observations of Spaet21: in rats rendered thrombocytopenic by antiplatelet serum, vessels were seen to seal themselves at the sites of petechiae by direct reestablishment of vascular continuity, without the intervention of platelet or fibrin thrombi.

**Platelets**

The period of bleeding characteristically follows the onset of thrombocytopenia; hemorrhage is maximal in laboratory animals during the second week after median lethal doses of x-radiation4,6 and in man during the third and fourth weeks after exposure.5 Following supralethal doses, death usually supervenes before the onset of purpura; however, in certain instances bleeding has been observed shortly after whole-body irradiation, preceding the onset of thrombocytopenia, i.e., in the rat mesentery22 within the first few hours and in the cheek pouch of the hamster23 within the first several days after exposure.

The experiments of Cronkite and Brecher6 indicate conclusively the therapeutic efficacy of transfused platelets in correcting or preventing bleeding; however, questions about the platelet mechanism remain to be answered. Some of the many functions ascribed to normal platelets by Stefanini24 involve virtually every stage in the clotting process. The first reaction, platelet agglutination, is presumably initiated by a change in the vascular wall; however, it is dependent on factors within the platelet itself and probably also within the plasma,25 as well as by the number of platelets per volume of blood. Savitsky and Sherry26
observed a decline in platelet adhesiveness and clot retraction in dogs after whole-body irradiation, which preceded the onset of thrombocytopenia; these changes were associated with the presence of a nondialyzable clot retraction inhibitor in the plasma of the irradiated animals. Subsequently, Savitsky\textsuperscript{27} isolated a cell-free fraction from the spleen which, when injected daily into heavily irradiated animals, delayed the onset of thrombocytopenia, maintained platelet adhesiveness, clot retraction, and clotting time within normal limits, and prevented the hemorrhagic state. Material extracted from bovine spleen was highly effective in dogs and guinea pigs, indicating an absence of species specificity. Although the administration of such extracts did not enhance the survival of his irradiated animals, the relation of Savitsky’s agent to the splenic protective factor of Jacobson,\textsuperscript{28} and to Moolten’s thrombocytosin,\textsuperscript{25} deserves investigation.

Other evidence pointing toward qualitative changes of platelets postirradiation has been obtained by Cronkite and Brecher,\textsuperscript{6} and by Ferguson et al.,\textsuperscript{29} who have observed deficiency in prothrombin utilization preceding by several days the onset of thrombocytopenia; similarly, when the platelet count begins to rise in dogs which are starting to recover from irradiation hemorrhage, during the third or fourth week after exposure, the utilization of prothrombin rapidly returns to and stays in the normal range even though the platelet count may still be less than 100,000 per cu. mm.\textsuperscript{6} Cronkite and Tocantins\textsuperscript{6} have also observed that the platelets newly formed in the period of recovery are larger than normal.

These findings may possibly be correlated with the observation that hemorrhage is less severe in animals protected against acute radiation death by procedures such as partial-body shielding\textsuperscript{28} and injection of bone marrow\textsuperscript{26} than in animals not similarly protected; with such protective measures, however, there is, in addition to an earlier resumption of platelet formation, earlier regeneration of all hemopoietic precursors, more rapid recovery of the immunologic mechanism, and less predisposition to infection, factors believed to influence the tendency to bleed.\textsuperscript{6}

There is need for more knowledge about the rate of delivery and utilization of platelets in relation to hemostatic function in the irradiated animal. Studies of this problem by Odell et al.\textsuperscript{31} with platelets labeled by C\textsuperscript{14} and S\textsuperscript{35} have indicated a maximal circulation time of approximately 6 days for normal rat platelets transfused into normal homologous hosts. Odell has not yet determined conclusively the survival time of platelets transfused into irradiated hosts, but his preliminary observations suggest that platelet production from megakaryocytes continues for at least a short time postirradiation.\textsuperscript{32} It is desirable to know whether such platelets, formed from irradiated precursors, are functionally intact. There is basis in the clinical disease thrombocytasthenia\textsuperscript{33} for postulating qualitative as well as quantitative abnormalities of platelets.

The discovery that irradiation hemorrhage can be largely prevented or mitigated by platelet transfusion suggests the importance of platelets in the pathogenesis of this condition. Cronkite and Brecher\textsuperscript{34, 35} first demonstrated this in irradiated dogs by prophylactic daily transfusions of freshly separated platelets. Later, the same investigators aided Woods and associates\textsuperscript{36} to confirm and extend these observations in dogs and rats, with the use of the lymph erythrocyte count as a quantitative index. It was noted that within a few hours after a plate-
let transfusion a previously bloody lymph would be temporarily cleared of erythrocytes. The precise mechanism of this effect is still unknown.

Although these findings offer the encouraging prospect of platelet banks for transfusing thrombocytopenic people, Cronkite and Brecher observed that after storage for only one day at 5 C. platelets transfused into normal or irradiated dogs failed to circulate, and in some instances actually precipitated thrombocytopenia. Although this may be correlated with histo-incompatibility, other mechanisms appear more likely, since the complication was not observed with fresh platelet preparations.

**Thromboplastin**

Following the discovery by Quick and by Brinkhous that the interaction of platelet and plasma factors is needed for thromboplastin formation, the advances in this field have been rapid. Although no attempt will be made here to review the problem in detail, the plasma of irradiated dogs has been observed to be deficient in thromboplastin. It appears from the studies of Penick and associates that irradiation does not significantly reduce the amount or activity of antihemophilic globulin, since the plasma of irradiated animals readily coagulates upon the addition of platelet-rich hemophilic plasma. However, it is possible that thromboplastin formation may be inhibited by an increased activity of antithromboplastin. This has been demonstrated by Tocantins, who found the lipidsoluble plasma antithromboplastin significantly elevated in dogs within 5 days after exposure to only 150 r of x-rays. The relative importance of this observation remains to be determined.

**Thrombin**

There is no evidence that irradiation hemorrhage involves a deficiency of prothrombin or any of the accessory factors, with the possible exception of proaccelerin. A significant transient reduction of plasma proaccelerin has been reported in six of seven patients given P in the treatment of polycythemia. However, the mechanism of this change may conceivably be dependent on hepatic damage or other effects, rather than on whole-body irradiation per se.

Apart from thrombin formation, there are indications that thrombin antagonists may occur in increased amounts in acute radiation sickness: the presence of a circulating heparinoid anticoagulant in irradiated animals has been noted by Allen et al. and periodically by Cronkite and others. However, this factor is no longer considered to be of major importance as a cause of hemorrhage. On reevaluating his early data, Allen found that the heparinoid anticoagulant was prominent only after repeated blood transfusions; even under such conditions its occurrence is unexplained.

It is conceivable that enhanced sensitivity to heparin may result from an increase in “heparin cofactors,” or plasma antithrombin; this has been observed in one instance postirradiation. Likewise, the thrombocytopenia may contribute to heparin sensitivity. Similarly, although the mechanism of heparin clearance is disputed, if heparin is removed from the body by the reticuloendothelial system, as has been reported, it is possible that its clearance is suppressed.
in the acute radiation syndrome, since some of the phagocytic functions of the reticuloendothelial tissues may be impaired by irradiation. In pursuit of the heparin problem, Allen has reported a variable increase in the surface tension of the blood of irradiated dogs as well as in the protamine titration test; however, the biologic and clinical significance of these preliminary observations remain to be established.

**Fibrin**

Whereas deficiency of fibrinogen is not observed in the acute radiation syndrome, the lack of platelets, which inhibited earlier phases of clotting, may impair the gelification of fibrin; the major defect at this point, however, appears to be the shortage of thrombin, which has been discussed.

Another factor entering the fibrin mechanism is fibrinolysin, or plasmin, a proteolytic enzyme which destroys the fibrin clot. The studies of Kocholaty et al. and of Colgan and co-workers have demonstrated a significant increase of fibrinolysin activity after whole-body irradiation, coinciding with the period of prolonged clotting time. Although probably of only secondary importance in the pathogenesis of hemorrhage, this alteration deserves consideration and further evaluation.

**Clot Retraction**

The process of clot retraction has been found deficient by many observers. It is probable that quantitative, or possibly qualitative, deficiency of platelets contributes to this defect; in addition, however, a nondialyzable clot retraction inhibitor has been found in the plasma of irradiated dogs. Normal clot retraction may be preserved postirradiation by the administration of cell-free splenic extracts; the mechanism of this effect is not yet known. It is conceivable that a deficiency of the splenic hemostatic agent may result from irradiation, thus contributing to the clotting defect. Diminished "clot resistance" has been observed in rats within the first several days after whole-body exposure to 700 r of x-rays; the cause and significance of this abnormality remain to be established.

**Conclusion**

The hemorrhagic diathesis produced by whole-body irradiation is an important aspect of acute radiation sickness but varies in severity with species and dose. The pathogenesis of the bleeding is complex; disturbances of nearly all known major hemostatic mechanisms have been reported; however, the most important single etiologic factor appears to be thrombocytopenia, hemorrhage being prevented or arrested by transfusion of intact platelets. The precise role, or roles, of the platelet in this process are unknown. Likewise the nature of the vascular defect permitting escape of the erythrocytes and the mechanism of the various disturbances of clotting which contribute to radiation-induced hemorrhage remain to be determined.

**Conclusiones in Interlingua**

Le diathese hemorrhagic produce per irradiation del corpore integre es un aspecto importante del acute morbo de radiation, sed illo varia in severitate ab
un specie al altere e ab un dosage al altere. Le pathogenese del sanguination es complexe. Disturbationes de quasi omnne le cognoscite mechanismos hemostatic ha essite reportate, sed il pare que le plus importante factor individual del etiologia es thrombocytopenia, proque le hemorrhagia es prevenibile o arrestabile per transfusiones de placettas intacte. Le exacte rolo (o rolos) del placettas in iste processi non es cognoscente. Remane etiam a determinar le natura del defecto vascular que permette le extravasation of the erythrocytos e le mehcanismo del varie disturbationes de coagulation sanguine que contribue al hemorrhagias causate per irradiation.

REFERENCES

32 — (personal communication).


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