Reticuloendothelial System Blockade as an Effective Method of Radioprotection

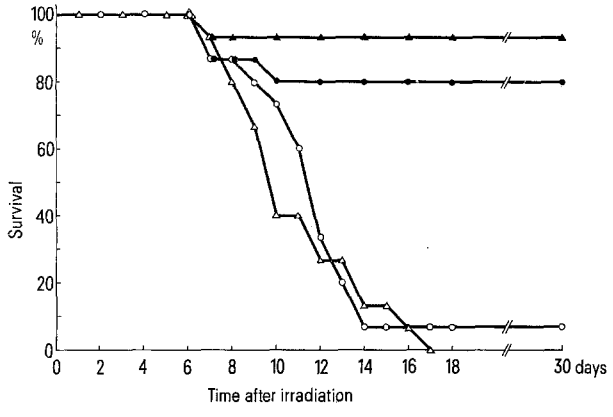
'Blockade' of reticuloendothelial system (RES) has been demonstrated to result in an increased survival of sublethally irradiated mice[1,2]. It has also been shown that such a favorable effect of RES-blockade may be at least partly attributable to the functional blockade of RES as well as the subsequent stimulation of hemopoiesis[3,4]. In carrying out these studies, the question occurred whether such an effect is solely due to RES-blockade and not due to some action characteristic to carbon particles which were used to blockade RES in the earlier studies.

In the present experiments, polystyrene latex particles and glycogen were employed as other chemically inert foreign particles in order to obtain further evidence that any non-toxic particulate materials can protect animals from radiation lethality through non-specific RES-blockade.

Materials and methods. Male mice of ICR-JCL strain, 7 weeks old at the start of the experiments, were used.

RES-blockade. Polystyrene latex particles of diametre of 0.109 ± 0.0027 μm were purchased from Dow Chemical Co., USA and 10 mg of the particles were injected into mice i.v. 24 h prior to irradiation. Glycogen was dissolved in physiological saline and aliquots of 10 mg were injected into each mouse of another group 24 h before irradiation.

Irradiation. Mice were exposed to 600 R to 700 R of whole-body X-irradiation, using 200 KVP — 20 mA unit, with a filter of 0.5 mm Al + 0.5 mm Cu, and at a dose rate of 100 R/min.



Survival of X-irradiated mice which have received 10 mg of polystyrene latex particles 24 h before irradiation. Open circles, 650 R-irradiated control; open triangles, 700 R-irradiated control; solid circles, 650 R-irradiated after latex injection; solid triangles, 700 R-irradiated after latex injection.

Endogenous spleen colony. As previously described[3], mice were irradiated with appropriate doses of X-rays. 24 h before irradiation, they were injected either with 10 mg latex particles or with 10 mg glycogen. Control mice were injected with the same amount (0.5 ml) of saline. All mice were given 275 mg/l chlortetracycline (Takeda Co., Japan) dissolved in drinking water from 7 days before the start of the experiment until sacrifice. Survivors were killed on day 10, and their spleens were scored for colonies after being fixed in Bouin's solution.

Results. The Figure shows the survival of irradiated mice after treatment with latex particles. All control mice died within 17 days after the exposure to 700 R of X-rays and 14 mice out of 15 died after irradiation with 650 R. Pretreatment of mice with 10 mg latex particles protected the animals from death. 12 and 14 mice survived the dose of 650 R and 700 R, respectively.

Endogenous spleen colony-formation was also examined to investigate the effect of RES-blockade on the survival of hemopoietic stem cells. As summarized in Table I, approximately 2 colonies were observed at 10 days after the exposure of mice to 650 R of X-rays. Survival of hemopoietic stem cells was remarkably increased by the pretreatment of mice with latex particles, the survival being 10 times as high as that of the control mice.

On the other hand, survival of irradiated mice was scarcely affected by the previous treatment with glycogen (Table II). In the mice irradiated with 600 R of X-rays, the survival seems slightly increased by the treatment, whereas those irradiated with more than 650 R showed no difference at all in their survival. As is also shown in Table II, number of spleen colonies was increased by the treatment from 1.4 in the control mice to 4.8 in the mice exposed to 600 R, indicating a slightly favorable effect on hemopoietic recovery.

Discussion. The present experiments demonstrate that administration of polystyrene latex particles to mice strongly protect the animals from radiation-induced death. It is also shown that the 'blockade' of RES with latex particles results in a favorable microenvironment for the hemopoietic stem cells to recover after irradiation. These results are similar to the data obtained with the carbon particles[1-3].

The mechanism of radioprotection by RES-blockade with carbon particles has been extensively studied[1-7]. The results point to the fact that the protection is attribut-

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Table I. Effect of RES-blockade on endogenous spleen colony-formation in irradiated mice

	Treatment of mice			
	580 R-irradiated	650 R-irradiated	Latex[a] + 650 R	Latex[a] + 700 R
No. of colonies per spleen	4.5 ± 1.6	1.8 ± 0.8	Numerous (30)	24.7 ± 3.2

[a] Mice were injected i.v. with 10 mg polystyrene latex particles 24 h prior to irradiation.

Table II. Effect of glycogen-treatment on survival and endogenous spleen colony-formation in irradiated mice

Treatment	Survival dose of X-irradiation			No. of colonies per spleen (600 R-irradiated)
	600 R	650 R	700 R	
Control	9/25 (36%)	2/15 (13%)	2/15 (13%)	1.4 ± 0.8
Glycogen-treated[a]	13/25 (54%)	2/15 (13%)	1/15 (7%)	4.8 ± 1.3

[a] Mice were injected with 10 mg glycogen i.v. 24 h prior to X-irradiation.

able to a functional blockade of RES in the first step and to the subsequent enhancement of hemopoietic recovery. The latter has also been shown to be the result of the enhancement of the recovery as well as of the survival of hemopoietic stem cells possibly through humoral factor(s) and direct control by fortified RES[5,6]. Again, it is suggested that reinforcement of RES by blockading with particulate materials plays an important role in controling the parenchymal cells to maintain their normal function[8].

The data presented herein provides evidence which supports the hypothesis that any foreign particles which will be phagocytized by RE cells, such as macrophages, can be used as an effective radioprotectant only if they are not toxic to the animals.

One finding to be noted is that although glycogen did not protect mice effectively from radiation lethality, it also had a slightly but significantly favorable effect on the hemopoietic recovery as manifested by an increase in the number of endogenous spleen colonies. It seems, therefore, that any substance can stimulate RES non-specifically, when phagocytized, to release some factor(s) such as interferon[9] and colony-stimulating factor[10] as well as to provide a favorable environment. The degree of such effects is likely to depend on the size and dose of the particulate materials and on the persistence of them in the RES[11].

Zusammenfassung. Die i.v. Injection von Latexpartikeln an Mäusen, 24 h vor einer Bestrahlung mit 650 oder 700 R, bietet einen bemerkenswerten Strahlenschutz. Es wird mit Hilfe der endogenen Koloniebildungsmethode auch nachgewiesen, dass die Strahlenresistenz und/oder die Regeneration der hämatopoetischen Stammzellen durch die reticulo-histiocytäre Systemblockade gesteigert worden ist.

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Interaction of Thrombasthenic Platelets with Subendothelium: Normal Adhesion, Absent Aggregation

Adhesion of blood platelets to extracellular connective tissue represents the first observable step in hemostasis and thrombosis. Subsequent to adhesion, platelet aggregation results in the formation of a hemostatic plug or a mural platelet thrombus. Baumgartner[1] has recently developed a perfusion chamber for studying the interaction of platelets in anticoagulated whole blood with the subendothelial surface of rabbit aorta under controlled flow conditions that are similar to those in arteries. Platelet adhesion to subendothelium was significantly reduced, compared with control values in von Willebrand's disease[2] and the Bernard-Soulier syndrome[3]. A defect in platelet adhesion probably accounts for the prolonged bleeding time in patients with these disorders.

In this communication we report our findings on platelet interaction with subendothelium in the perfusion chamber using blood from 2 subjects with classical Glanzmann's thrombasthenia, a hemorrhagic diathesis characterized by prolonged bleeding time, absent platelet aggregation and markedly impaired clot retraction[4]. Previous investigations suggested that the adhesion of platelets in platelet-rich plasma to connective tissue is normal in thrombasthenia[5–7].

Materials and methods. Clinical and laboratory findings of the 2 patients M.C. and M.M. (the latter studied through the courtesy of Dr. Marjorie B. Zucker) have been described previously[8,9]. All 9 control subjects (ages 25–44) showed second phase aggregation with epinephrine in platelet-rich plasma on the day studied. Patients and controls were asked not to take any medication for 1 week prior to the experiment.

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