Protection Against Gastrointestinal Effects of Whole-Body X-Irradiation by a Bile Acid Sequestrant in Rats

Loss of body fluids and electrolytes is an important secondary consequence of whole-body exposure to doses of ionizing radiation which produce the gastrointestinal (GI) syndrome in experimental animals and in man[1]. JACKSON et al.[2] demonstrated in rats that during the post-irradiation period loss of body sodium is sufficient to account for death. These workers proposed that bile was the major source of excreted sodium in irradiated animals and showed that bile duct ligation decreased the rate of body sodium loss, postponed onset of diarrhea and prolonged survival[3]. SULLIVAN[4] found that diarrhea did not develop in irradiated bile duct-cannulated rats when isotonic saline was introduced into the duodenum, but that it did occur when bile or bile salts were injected intraduodenally. He concluded from this limited study that the bile salt component of bile is specifically involved in development of radiation-induced diarrhea. SULLIVAN also proposed that bile and bile salts remove mucous from the intestinal epithelium of irradiated rats and demonstrated that bile duct cannulation prevented mucous loss and diarrhea, even though the basic radiation damage to the intestinal epithelium was unaltered[5]. These findings were confirmed by GORIZONTOV et al.[6], who also found that bile duct cannulation prior to irradiation decreased injury to the intestinal epithelium, including the mucous-secreting goblet cells, and prevented diarrhea. X-irradiation inhibits ileal bile salt absorption in rats and causes their accumulation in the small and large intestine during the post-irradiation period[7]. Bile salts inhibit sodium and water absorption from perfused rat colon[8], induce secretion of water and electrolytes into the colon in man[9] and possibly influence colonic motility[10,11]. These actions have been proposed as causes of the diarrhea seen clinically following ileal resection, termed cholereic enteropathy by HOFMANN[12] and also may contribute to the diarrhea of the GI syndrome in addition to the direct effects of bile salts on irradiated intestinal mucosa described above.

These observations suggested that oral administration of a bile acid sequestrant DEAE Sephadex[13] might increase survival times of X-irradiated rats dying with the GI syndrome, thereby simulating surgical diversion of bile from the intestine[3].

Male Sprague-Dawley rats, 250–350 g, immobilized by placing them in individual polyethylene bags with air holes, were exposed to whole-body X-radiation generated by a 2 mev Van de Graaf electrostatic accelerator with a hollow, water-cooled copper target. General operating conditions were: electron voltage 1.9 mev, beam current approximately 110 μamp, beam scan 12.5 cm, belt speed 0.91 m/min. The target was 2 cm thick, 8 cm wide and 25 cm long and was constructed of copper approximately 1 mm thick. The lower side of the target was covered with a piece of methylmethacrylate 1.25 cm thick to absorb secondary electrons. Beam current was varied slightly during irradiation so that the total final dose remained at desired levels. Radiation exposure was measured in roentgens (R) by placing the probe of a Victoreen Radocon R-meter on the conveyor belt with the rats. Animals were placed prone on an aluminium tray in groups of 10 and were passed under the target on a conveyor belt; multiple passes were used to deliver the desired total radiation dose. After irradiation animals were placed in individual cages and were allowed free access to food and water. In Study 1, 10 animals exposed to 2100 R were given via stomach tube 4 ml of a suspension containing 570 mg DEAE Sephadex A-25 (Pharmacia Fine Chemicals, Inc.) in distilled water and 10 animals were given distilled water only. Dosing was continued at 24-h intervals for the next 3 days. All rats died by 9 days post-irradiation. DEAE Sephadex increased mean survival time (MST) by 1.2 day (Table). In Study 2, irradiated rats were dosed with DEAE Sephadex daily during the entire survival period following irradiation (5–7 day); controls were dosed with distilled water. MST of animals which were exposed to 1540 or 1250 R was increased by 0.7 and 1.4 day, respectively. In addition to prolonging survival, DEAE Sephadex delayed the onset of diarrhea and decreased its severity in both Studies 1 and 2.

Increases in MST which we have observed in rats dosed with the bile acid sequestrant are comparable to those reported by JACKSON and ENTENMAN in bile duct-ligated animals[3]. In other studies (unpublished) we have also found a protective effect of DEAE Sephadex in dogs exposed to 1000–1500 R whole-body ^{60}Co radiation. Our findings support the hypothesis that bile salts are involved in development of radiation-induced diarrhea, since DEAE Sephadex specifically reduces the effective concentration of this component of bile, in contrast to the multiple effects of bile duct ligation or cannulation. In addition they suggest that bile acid sequestrant therapy might

Effect of DEAE-Sephadex A-25 on survival of X-irradiated rats

Study	Radiation exposure (R)	Mean survival time (days)	
		Controls	DEAE-Sephadex
1	2100	5.2 ± 0.13	6.4 ± 0.43
2	1540	4.2 ± 0.13	4.9 ± 0.23
	1250	4.7 ± 0.21	6.1 ± 0.28

Data represent means ± standard error of the means; 10 animals per group. Statistical significance ($p < 0.05$) of the differences between mean survival times of control and treated groups was determined by DUNNETT's t-test[14].

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provide a practcal mieans of reducing the severity of the potentially lethal secondary effects of ionizing radiation in the human GI tract.

Zusammenfassung. Nachweis, dass die Gallensalze an der Entstehung des gastrointestinalen Strahlensyndroms mitverantwortlich sind und dass DEAE-Sephadex die Gallensalze bindet und die Überlebenszeit letal behandelter Ratten verlängert.

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Characteristics of the Nucleolini Observed Under the Photon Microscope

The term 'nucleolini' is an old one[1] and serves to designate tiny bodies, more or less spherical, that appear in the nucleolus, expecially in relation to a particular functional stage of it[2,3]. While some authors maintain that in the nucleolus there exists a skein-like filament that is to be termed 'nucleolonema'[4,5], other authors consider that the structures actually present in the nucleolus are the nucleolini[6,7] or, at any rate, nucleolar granules[8]. Unfortunately, up to a certain time ago, the use of silver nitrate as impregnation substance[9] often gave rise to super-impregnations that gave the mistaken idea that there actually existed in the nucleolus a filament wound like a skein: namely, the nucleolonema. However, when initially the electron microscope[10,11] showed the existence of a sort of reticulum, this was mistakenly identified with the nucleolonema[12]. And even if one of the authors – namely BERNHARD[13,14] – that contributed in creating this mistaken idea later stated how this reticulum should be considered – that is, not ascribable to the nucleolonema (though he proposed keeping this term) – many subsequent authors have continued to assign it, in fact, to the nucleolonema[15]. Fortunately, at present, in many works on the ultrastructure of the nucleolus, there is mention of its occasional reticulum-like appearance but no longer of 'nucleolonema'[16].

The object of this note is to make a further contribution to the knowledge of the nucleolini, as regards both certain typical aspects of them and their number in relation to dimensions of the nucleoli.

Materials and methods. The material chosen was the following: oocytes of molluscs, echinoderms, anuran and urodelan amphibians; cells of various tissues of vertebrates, particularly those of the nervous tissue; cells of Walker's tumour in the Rat and of human mammary carcinoma; and cells of the root apexes of the plants *Allium cepa* and *Vicia faba*. For the nucleolini, the method of impregnation especially used was that employing platinum chloride[9]. For the nucleolus, material fixed in

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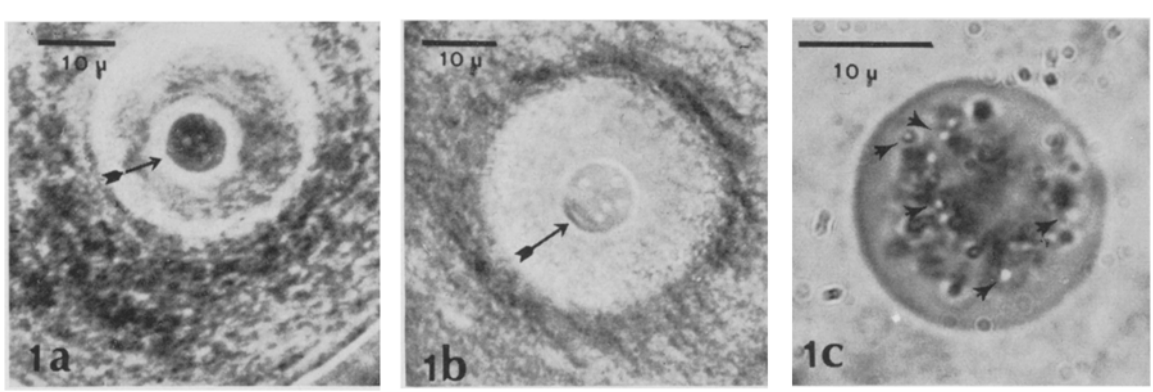

Fig. 1. Oocytes of *Paracentrotus lividus* observed in vivo under the phase-contrast microscope (a) and under the Nomarski's interference phase-contrast microscope (b): the arrows indicate the nucleoli with nucleolini. c) Nucleolus of *Echinus melo* oocyte in a preparation using platinum chloride; there are numerous nucleolini with a high refraction (arrows).