International Journal of Radiation Biology

ISSN: 0955-3002 (Print) 1362-3095 (Online) Journal homepage: https://www.tandfonline.com/loi/irab20

Radiation victim management and the haematologist in the future: Time to revisit therapeutic guidelines?

Michel Drouet & Francis Hérodin

To cite this article: Michel Drouet & Francis Hérodin (2010) Radiation victim management and the haematologist in the future: Time to revisit therapeutic guidelines?, International Journal of Radiation Biology, 86:8, 636-648, DOI: 10.3109/09553001003789604

To link to this article: https://doi.org/10.3109/09553001003789604

Published online: 05 Jul 2010.

Int. J. Radiat. Biol., Vol. 86, No. 8, August 2010, pp. 636–648

informa
healthcare

Radiation victim management and the haematologist in the future: Time to revisit therapeutic guidelines?

MICHEL DROUET & FRANCIS HÉRODIN

IRBA-antenne La Tronche-CRSSA, La Tronche, France

(Received 17 July 2009; Revised 11 March 2010; Accepted 16 March 2010)

Abstract
Purpose: The use of nuclear/radiation devices against the civilian population is now a realistic scenario. Haematopoietic syndrome is the primary therapeutic challenge in the case of whole body acute exposure over 2 Grays (Gy) whereas burns and combined injuries would be frequently observed in myelo-suppressed patients. Optimisation of scoring and treatments are important goals to achieve.
Conclusion: The European Response Category (RC) concept represents an attempt to integratively assess haematological/extrahematological radiation-induced lesions. Based on the frequently observed heterogeneity of bone marrow damage in accidental/intentional irradiations, the stimulation of residual stem cells using granulocyte Colony-stimulating factor remains the therapeutic standard after exposure to less than the lethal dose 50 % (Haematopoietic[H] score 3-H3). Allogeneic stem cell transplantation is indicated in case of medullary eradication (Haematopoietic score 4-H4) whereas extramedullary toxicity may determine the outcome. Especially in case of numerous casualties exhibiting acute radiation sydrome, the administration of survival factor combinations remains questionable, at least as a palliative treatment. In addition pleiotropic cytokines injection such as erythropoietin and keratinocyte growth factor and grafting multipotent mesenchymal stem cells – from underexposed bone marrow areas or fat tissues – could be proposed to prevent multiple organ failure syndrome development. Multi-disciplinary teams should be prepared to manage such patients.

Keywords: *irradiation, cytokine, METREPOL, mesenchymal stem cells*

Introduction

In addition to accidental exposure from a nuclear industrial source or similar, the use of nuclear/radiation devices against the civilian population is now a threatening possibility (Dainiak et al. 2003, Waselenko et al. 2004, Weisdorf et al. 2006). The explosion of Radiological Dispersal Devices (i.e., 'dirty bombs') represents a realistic scenario which could produce numerous casualties if the bomb is set off in a very densely populated area (difficult to estimate depending on environmental conditions, chemical and physical form of the radioisotope, type and amounts of explosives; say some hundreds of victims), mainly mechanical (penetrating trauma, crush injuries) but also including Radiological contamination – external and internal. Detonation of an Improvised Nuclear Device/weapon could lead in certain cases to casualties similar to classical nuclear weapons, including numerous Acute Radiation Syndromes and combined injuries. Intentional use of hidden high activity sources (Radiation Exposure Device) could be another pertinent strategy to panic populations. For example, a Cobalt 60 (^{60}Co) source of 1000 Curie hidden in a subway (0.5–2 m distance, i.e., 50-3 Gy/hours dose rate and 25-1 Gy absorbed dose) has been estimated to lead to 4–16 victims every 30 min of exposure by the experts of the French Institute of Radioprotection and Nuclear Safety (Vaux de Cernay meeting, France 2003). Here numerous victims would be exposed to a single acute high dose (classical Acute Radiation Syndrome) or to a more protracted exposure (whole body, partial or local exposures to high dose rates). Thus in future attacks against civilian populations, medical teams have to consider the treatment of numerous Acute Radiation Syndromes combining medullary and extra-medullary

Correspondence: Dr Michel Drouet, Department of Radiobiology, IRBA-antenne La Tronche-CRSSA, 24 Avenue des Maquis du Grésivaudan, 38702, La Tronche, France. Tel: +33 4766 36931. Fax: +33 4766 36922. E-mail: micheldrouet@crssa.net

ISSN 0955-3002 print/ISSN 1362-3095 online
DOI: 10.3109/09553001003789604

damages. In this context the haematopoietic syndrome would still remain the first therapeutic challenge in numerous cases. Today allogeneic haematopoietic stem cell grafting (bone marrow, peripheral blood or cord blood) is still the gold standard in case of medullary eradication. However, mainly based on animal models, the benefit of stimulating residual haematopoiesis using haematopoietic growth factors has been demonstrated in a limited cohort of victims.

In this review we first intend to present the debate on the currently used radiation dose categorisation system. Then using data from the most recent animal studies and progresses in clinic we would like to discuss the interest of revisiting the therapeutic guidelines in the near future. In particular, in the context of numerous victims to be managed, the adaptation of cytokine treatment based on optimised triage/categorisation should be considered. Importantly the spreading development of new stem cell therapy strategies mainly focusing on adult multipotent mesenchymal stem cell grafting, requires the establishment of clear recommendations. Importantly, the treatment of highly irradiated victims – from global to significant partial exposure – would involve multidisciplinary teams.

Acute radiation syndrome: Damage versus dose assessment as a new paradigm?

The dose problem

Acute Radiation Syndrome following total body irradiation (TBI) or significant partial body irradiation has been classically described as dose-dependent with progressive involvement of tissues according to their respective radiosensitivity, i.e., haematopoietic/H(>2 Gy TBI) then gastrointestinal G(>10 Gy) and then neurovascular syndrome/N(>30 Gy) (Young 1987). Acute radiation syndrome occurs in three successive waves: Initial prodromal phase within the first hours following exposure, latent period and then manifest illness. The onset, duration and gravity (score) of each phase depend on total dose received, instantaneous or protracted delivery and dose distribution in the body. In addition the clinical and biological signs would be modified in the case of delivery of therapeutic agents (especially in early treatment schedule).

However, radiation dose is a physical measurement which alone, in spite of its widespread use and recognised interest (described correlation between dose and lethality in human: >10–12 Gy = 100% lethality; 3–4 Gy = lethal dose 50%), physical dosimetry (dose/dose rate), appears insufficient to reflect radiation-induced biological effects as well as to predict the clinical evolution of a patient, because of inter-individual response variability. Indeed dose distribution is a critical parameter. For example, in the case of haematopoiesis, animal data have clearly shown that even unilateral total body irradiation does not result in uniform damage due to body thickness attenuation which for example reaches 50 % in baboons with ^{60}Co gamma. In fact, most accidental irradiations have proved to be heterogeneous. An evaluation of the average dose is still required and remains a useful guide to the physician. Complete blood cell count (CBC) (every 4–6 h during the first 24 h then twice a day for two days) is the classical indicator of an average dose of more than 2 Gy when the decrease in lymphocyte count is over 50 %. Evaluation by bio-dosimetry is still based on chromosome aberration scoring following a one to three-day lymphocyte culture and ^{24}Na measurement in case of neutron exposure (Fliedner et al. 2001, Gorin et al. 2006). Obviously none of these parameters would help a physician in an emergency. Clinical parameters to be checked include early vomiting, diarrhoea, asthenia, abdominal pain, headache and cutaneous erythema whose level and earliness (30 min to 5 h) are graded according to dose. Unfortunately, such parameters are frequently transient. Later on, physical dosimetry (from tooth enamel etc.) and dose reconstruction would become available. Bio-indicators such as Fetal Liver Tyrosine Kinase 3(Flt3)-ligand serum level and citrullinemia are proposed to assess bone marrow and gastrointestinal damage during the critical illness phase (Bertho et al. 2008). For some years there has been a great deal of work to identify radiation responsive genes that could potentially be used as biomarkers and microarray analysis has been proposed as a new triage strategy. Interlaboratory reproducibility, individual susceptibility and confounding factors still remain important concerns in this domain. Multiparametric approaches to be validated may be of interest – even in emergency context – as suggested by the studies conducted by the Armed Forces Radiobiology Research Institute which recommend the integration of changes in blood cell counts, multiple blood-proteins and gene expression and encoded target proteins and develop software applications (Chaudhry 2008, Straume et al. 2008, Ossetrova and Blakely 2009).

The 'Medical Treatment Protocols' (METREPOL) – Response Category concept

In an attempt to quantify organism impairment integratively without relying on dose, the Response Category (RC) concept has been proposed (Fliedner et al. 2001). This elegant dynamic clinical (semi-quantitative) scoring combines the damage evaluation for each of the first tissues to

respond: Haematopoietic/H, Gastrointestinal/G, Neurovascular/N and cutaneous radiation syndrome/C. Thus Response Categories 1 to 4 can be distinguished. Interestingly, these Response Categories evolve over time as a function of organ damage and depending on the treatment. According to METREPOL, Haematopoietic score 3-H3 corresponds to possible autologous recovery and Haematopoietic score 4-H4 to most unlikely autologous recovery. A simpler 3-graduation score, at least useful in emergency, has been proposed with grade 2 corresponding to curable patients and grade 3 to patients of poor prognosis. The METREPOL concept proposes a consistent approach to assess the complex interplay of irradiation with tissues as well as cross talks between organs distant or not. Indeed, evidence that radiation syndrome has to be considered as a global systemic illness for a broad dose range has (re)emerged since some years. Complex abscopal effects and functional disorders of different tissues following irradiation at doses below their recognised threshold have been reported in acute or more protracted exposure schedule. For example central nervous system is now recognised to be radiosensitive in terms of functional criteria (brain electrical activity and neurochemical metabolism for doses of a few Grays) (Tofilon and Fike 2000, Gourmelon et al. 2002). Recent studies have also highlighted the role of functional damage in gastro-intestinal pathology following irradiation (Linard et al. 1997, Otterson 2007). In fact, one of the main targets of these triage methods is to identify victims who will develop multiple organ failure syndrome (Fliedner et al. 2005). Albeit non-specific (Powles et al. 1983), the latter is likely to occur following high dose total or even partial body irradiation especially in case of associated cutaneous radiation syndrome or extensive gastrointestinal damage. The pathophysiology of multiple organ failure is complex and numerous hypotheses have been proposed (Barton and Cerra 1989, Gando et al. 1995, Godin and Buchman 1996, Gordon et al. 2000, Hassoun et al. 2001, Aird WC 2003, Araki et al. 2005, Monti et al. 2005). Sepsis represents a good model of systemic inflammatory response and multiple organ failure syndrome. Interestingly, according to the model of immunologic dissonance, it has been pointed out that the pathology may result from a lack of balance between inflammatory and anti-inflammatory responses (Bone 1996). Recently the chronology of the events has been documented with an early activation of both pro and anti-inflammatory responses followed by a delayed phase of anti-inflammatory response (Hotchkiss et al. 2009). This offers numerous similarities with irradiation context whereas the onset of iterative inflammatory waves remains frequent in the latter. Thus the systemic inflammatory response may represent the core of radiation induced multiple organ failure syndrome (Figure 1; Van der Meeren et al. 2003, 2005); pathological disorders include endothelial cell activation/damage, parenchymal cell damage and bone marrow failure/sepsis (Akashi 2005, Gaugler 2005), leukocyte activation, proinflammatory cytokine and reactive oxygen species production, neurogenic inflammation/neuropeptide release and metabolic disorders (Parke et al. 2002, Singer et al. 2004, Gourmelon et al. 2005). Primary and secondary multiple organ failure resulting from the direct insult itself and as a consequence of host response (i.e., systemic inflammatory response) should be distinguished. The precise role of endothelial apoptosis in this process remains debated, as intestinal crypt death descriptions illustrate (Paris et al. 2001, Schuller et al. 2006).

Altogether, any patient categorisation (both in the early stages and as the disease progresses) remains a difficult task. Based on multidisciplinary team involvement (Blakely et al. 2007, Dainiak et al. 2007), a combined approach including clinical signs and symptoms, biomarkers and physical dosimetry would be the most pertinent way to help physicians. One must keep in mind that each parameter displays a proper kinetics of evolution following total body irradiation. Patients' time of presentation can be a factor of uncertainty in diagnosis/progress assessment. For instance, inflammatory markers such as fever or C-reactive protein level evolve in waves with a first peak during acute phase response, followed by erratic phases of increase due to neutropenia/signs of infection whereas, on the contrary, haematopoietic recovery is also associated with a benign febrile pattern (Figure 2). An early drop of citrulline level

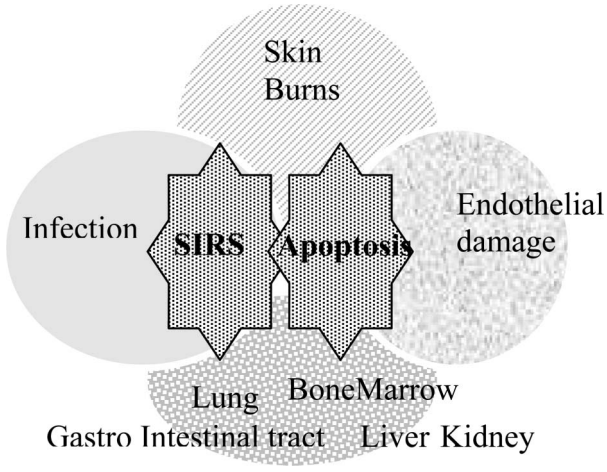

Figure 1. Multiple Organ Failure (MOF) following high dose irradiation represents a radiation induced (RI) dysfunction in two or more organs. Radiation syndrome has to be considered as a global systemic illness for a broad dose range. Systemic inflammatory radiation syndrome (SIRS) is believed to represent the core of RI-MOF.

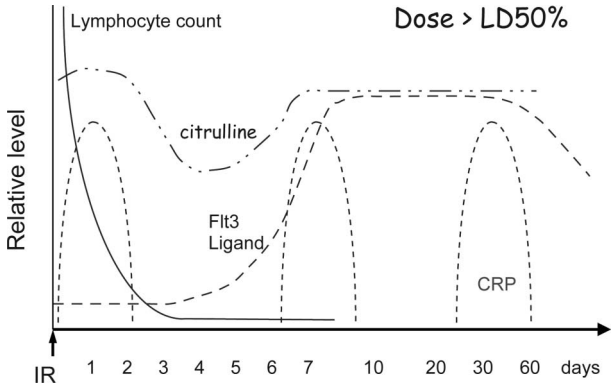

Figure 2. Relative levels of a few biomarkers of damage following high dose whole body exposure. Inflammatory markers such as C reactive protein evolve in waves with an initial peak during acute phase response; early lymphocyte count decrease and delayed Fetal Liver Tyrosine Kinase 3(Flt3)-ligand increase reflect bone marrow damage; citrullinemia reflects gastro-intestinal damage.

followed by a slow recovery to basal level is typically observed in the case of extended gastro-intestinal damage. Obviously, mass casualty scenarios make such a categorisation more difficult: Medical management would be delayed with lack of indication regarding early phase symptoms and large-scale cytogenetic analysis would require important support.

Facing haematopoietic syndrome

Current treatment: Cytokine and stem cell transplantation

Following whole body exposure, haematopoietic syndrome is mainly the consequence of the early radiation-induced cell death (apoptosis and necrosis) of cycling haematopoietic stem cells and progenitors (De Revel et al. 2005). Medical management consists in haematopoietic growth factor administration in the case of intermediate bone marrow damage (Haematopoietic score 3-H3 or from 3 to 7 Gy) and allogeneic stem cell transplantation if bone marrow has been severely depleted (Haematopoietic score 4-H4 or >7 Gy). This review does not intend to emphasise this point which has already been outlined in other reviews (Baranov et al. 1989, International Atomic Energy Agency 1993, Dainiak and Ricks 2005). Haematopoietic stem cells from bone marrow, peripheral blood or cord blood have already been transplanted and previous consensus recommends the delay of grafting in order to give autologous recovery a chance. Putative residual haematopoiesis should be stimulated using cytokines during the first weeks. This strategy also allows the minimisation of risks of immune conflicts due to early inflammatory burst. Transplanted victims frequently exhibited a transient recovery (see Chernobyl and Tokai Mura accidents). However, in numerous cases, extrahaematological radiation damage was severe and determined the outcome, patients dying from multiple organ failure syndrome (Maekawa 2001, Nagayama et al. 2002). In the Soreq and Tokai Mura accidents, cytokines were also given as a co-treatment but with no clear additional benefit.

Apoptosis and the Emergency Antiapoptotic Cytokine Therapy concept

The rationale of cytokine treatment is the heterogeneity of bone marrow damage as discussed above. Based on animal studies and experience from oncology, granulocyte colony-stimulating factor (G-CSF) with granulocyte-monocyte colony-stimulating factor (GM-CSF) as an alternative is currently the therapeutic standard after accidental exposure to less than lethal dose 50 % (estimated about 4.5 Gy gamma) in order to shorten neutropenia (Butturini et al. 1988, Schuening et al. 1989, MacVittie et al. 1990, Waselenko et al. 2004; for a review see Hérodin and Drouet 2005). The rationale is to enhance cell proliferation from residual haematopoiesis. However G-CSF/GM-CSF did not stimulate multilineage recovery and their efficacy is well-known to decrease with dose escalation and combined injury. Moreover, crisis following terrorist attack would result in delayed administration, i.e., reduced efficacy. It has been shown that synergistic effects result from combining cytokines (Schuening et al. 1993, Baranov et al. 1994, Farese et al. 1996, Neelis et al. 1997a, 1997b). This is why the stem cell factor (SCF) + erythropoietin (Epo) + pegylated G-CSF (pegG-CSF) combination was recently given to two patients in a short-term administration schedule. Patients recovered on day 31 and 37, respectively. However estimated doses were 3.5 and 4.2 Gy TBI only and administration was delayed (21 days); spontaneous recovery may have occurred (Fagot et al. 2006). For some years, our group has been developing a strategy based on counteracting radiation-induced apoptosis at the level of haematopoietic stem cells and progenitors (Hérodin et al. 2007). Indeed, as shown in Figure 3, the combination of SCF, Flt-3 ligand, thrombopoietin (Tpo) and interleukin-3 [SFT3] mitigated the induction of caspase 1, caspase 6, caspase 3 and caspase 9 in cluster of differentiation (CD)34^+ hematopoietic stem and progenitor cells following in vitro irradiation (Drouet et al. 1999, 2008a).

In animals, the SFT3 combination significantly protects stem cells and progenitors from radiation-induced cell death (massive during the first 24 h) if administered early after irradiation. Early and short-term administration of SFT3 prevents lethally irradiated mice from death and enhances multilineage

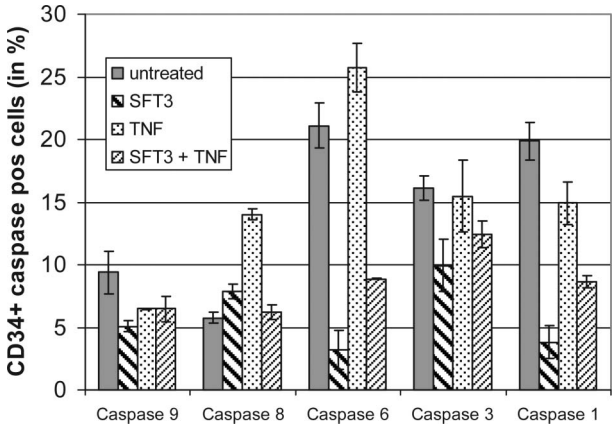

Figure 3. Activated caspases and radiation-induced (RI) apoptosis at hematopoietic stem and progenitor $CD34^+$ cell level. To assess caspase involvement in RI apoptosis at haematopoietic stem and progenitor cells level, non-human primate $CD34^+$ cells were irradiated in vitro using a ^{60}Co gamma source at a dose leading to 95 % cell death 24 h after irradiation. Immediately after irradiation, $CD34^+$ cells were incubated with or without anti-apoptotic cytokines (Stem cell factor + Flt3-ligand + Thrombopoietin + interleukine-3 i.e. SFT3; each cytokine: 50 ng/ml) ± Tumour Necrosis Factor alpha (TNF-α; 800 IU/ml) in fetal bovine serum-free medium at 5 % CO_2 in air. Ten hours later, which was defined as the optimal time, the cells were collected and caspase activation assessed using the Smolewski et al. (2001) method based on the detection of caspase activation by flow cytometry using fluorochrome-labeled inhibitors. Results are expressed as the percentage of caspase positive $CD34^+$ cells (Drouet 2008b) and error bars indicate the standard error of the mean (SEM) for $n = 4$ experiments).

recovery in 5 Gy-irradiated monkeys, with a notable protective effect on megakaryoblasts and megakaryocytes (Hérodin et al. 2003, Drouet et al. 2004). A significant thrombopoietic effect was observed in 7 Gy-irradiated monkeys and the addition of pegG-CSF significantly stimulates neutrophil recovery. These studies were not consistent with any exhaustion of long-term hematopoiesis. Long-term cytogenetic analysis did not suggest an impact of emergency antiapoptotic cytokine therapy on radiation-induced carcinogenesis (Hérodin et al. 2007).

New thrombopoietic agents

As thrombopoietin (and Pegylated Megakaryocyte Growth and Development factor) are immunogenic, new thrombopoietic agents have been developed such as AKR501, LGD-4665, AMG531/Romiplostim/Nplate and Eltrombopag (Kuter 2007, Bussel et al. 2007, Kuter et al. 2008). Nplate is a peptide mimetic made from the fusion of human immunoglobulin G1 heavy chain and kappa light chain constant regions (Fragment C carrier domains) and thrombopoietin agonist peptide domains without sequence homology with thrombopoietin. Eltrombopag, an orally active platelet growth factor, is a small non-peptide thrombopoietin receptor agonist which does not compete with thrombopoietin for binding to physiologic receptors. It has been demonstrated that a single injection of Nplate induce a dose-dependent increase in platelet count in healthy subjects (up to a six-fold increase between days 13–17). Both agents stimulated platelet production in idiopathic thrombocytopenic purpura (ITP) patients (prolonged administration scheme) and have been approved by the Federal Drug Agency in the United-States. To our knowledge, no data have been yet published regarding chemotherapy-induced thrombocytopenia. The capacity of currently available thrombopoietin substitutes – especially in combination – to mitigate apoptosis and to enhance haematopietic recovery following high dose irradiation remains to be established in ad hoc animal models.

Non haematopoietic stem cell support

Osteoblastic niche, including the fibroblastic/multipotent mesenchymal stem cells (MSC), and vascular niche are radiosensitive compartments (Kiel et al. 2005, Taichman 2005, Adams and Scadden 2008, Li et al. 2008). This is why grafting non-haematopoietic supportive cells has been proposed to repair irradiated stroma and sustain residual haematopoiesis or allogeneic transplanted stem cells. Among the candidates are bone marrow mesenchymal stem cells, first described by Friedenstein, and adipocyte derived stem cells. Not entirely similar, adult multipotent stem cells from these tissues and others (cord blood, skin ...) may be members of the large pericyte family (Crisan et al. 2009). They may act via local delivery of trophic factors and/or (trans)differentiation. Today, most of the authors consider mesenchymal stem cells as a safe cell product (Tarte et al. 2009). The intra-osseous injection of large amounts of mesenchymal stem cells has been evaluated in highly irradiated monkeys by our group and others without leading to any evidence of hematopoietic support (Drouet et al. 2005). However the Chute group recently reported the pro-survival and/or pro-haematopoietic activity of endothelial cells in mice (Chute et al. 2007, Salter et al. 2009). Ex vivo manipulation could enhance mesenchymal stem cell homing capacity (Karp and Leng Teo 2009) or allow the overexpression of selected factors – transient gene therapy based on the use of non-viral tools (Drouet et al. 2009a).

Facing extrahaematological toxicity

Radiation-induced multiple organ failure syndrome

Radiation-induced multiple organ failure, as a main consequence of extra-haematological damage, has

become a major therapeutic challenge. According to the American College of Chest Physicians/Society of Critical Care Medicine consensus in 1991, it can be defined as 'a progressive and potentially reversible pathophysiological dysfunction in two or more organs, induced by a variety of acute insults, including sepsis'. The two grafted patients in the Tokai-Mura accident represent good examples of this pathophysiologic process, i.e., patient named S (2.5 Gy neutron + 4.58 Gy gamma; receiving unrelated cord blood cells) and patient named O (5.5 Gy neutron + 8.5 Gy gamma; receiving peripheral blood from human leukocyte antigen [HLA] identical family donor [Maekawa 2001, Nagayama et al. 2002]). The first one died on day 211 from respiratory failure in spite of stable mixed chimerism. He presented infectious complications, pneumonia and delayed (day 90) significant gastrointestinal bleeding. The second one died on day 82 (renal shutdown, liver failure, hemodynamic instability, bone marrow hypoplasia etc.). The current treatment of multiple organ failure mainly consists of non-specific supportive care (Jackson et al. 2005). Systemic administration of autologous multipotent mesenchymal stem cells has been proposed as a new approach of cell therapy (Chapel et al. 2003, François et al. 2006, Mouiseddine et al. 2007). The rationale is the documented preferential homing of mesenchymal stem cells towards damaged areas. In animals, mesenchymal stem cells were shown to restore villus height (Semont et al. 2006), to improve intestinal absorption and protect the liver. Recently a transient pain reduction following autologous mesenchymal stem cells injection has been reported in some patients presenting proctracted gastro-intestinal irradiations (Gorin et al., oral communication European Group for Blood and Marrow Transplantation, annual meeting 2008). However the feasibility of autologous mesenchymal stem cells grafting in mass radiation accident is questionable; allogeneic grafting based on stem cell banking allowing an early treatment has been proposed but the therapeutic efficacy of theses latter cells remains to be established. Interestingly, adipose-derived (ADSC) stem cell transplantation has also been demonstrated to repair Crohn's fistula (Garcia-Olmo et al. 2005).

Administration of pleiotropic cytokines may represent a more realistic strategy in the context of mass casualties. Interestingly, the addition of erythropoietin to SFT3 (early/short term injection) enhances in place of the early and short term injection of SFT3 + erythropoietin enhances the survival rate of lethally-irradiated mice (Drouet et al. 2008b). No improvement of blood cell count and hemoglobin level was observed in this model which suggests prevalent extrahaematological targets. The origin of this pro-survival activity remains to be established – apoptosis mitigation, endothelial progenitor recruitment, etc ... (Agnello et al. 2002, Nathan 2003, Heeschen et al. 2003, Villa et al. 2003). Importantly, erythropoietin only acts in the presence of SFT3 which emphasises the importance of combining cytokines. As another candidate, keratinocyte growth factor (KGF) has demonstrated for some years a radioprotective potential in rodents (lung, etc.) when administered before irradiation. Interestingly, early post-irradiation treatment expands salivary gland progenitors (Dörr et al. 2005, Lombaert et al. 2008). In clinic, KGF reduces the incidence and severity of oral and intestinal mucositis in high-dose chemotherapy protocols (treatment schedule day-3/day + 3) (Spielberger et al. 2004) but the European Medical Agency has recently limited its use. Finally, it stimulates immune reconstitution in transplanted monkeys (Seggewiss et al. 2007). In accordance with these data, the SFT3 + KGF combination acts in a similar way to SFT3 + Epo in our mouse model. However, KGF as a single treatment (3 i.p. injections, day 1–3; 250 μg/kg) KGF was unable to protect gastro-intestinal tract – citrullinemia evaluation – in highly irradiated monkeys (8 Gy frontal TBI; Figure 4). In this model, lymphocyte counts did not significantly differ between treated and control animals.

Last but not least, counteracting exacerbated inflammatory response in irradiated patients would be crucial but today there is a lack of specific drugs. Novel strategies such as silencing inflammatory or pro-apoptotic genes may be useful in the future (Song et al. 2003, Wesche-soldato et al. 2005) but are still in infancy. Thus, in vivo delivery of small interfering RNA (siRNA) directed towards CD95-Fas antigen and caspase 8 has been demonstrated to improve the survival of septic mice and antiapoptotic peptides have been also tested in mice following irradiation (McConnell et al. 2007, McDunn et al.

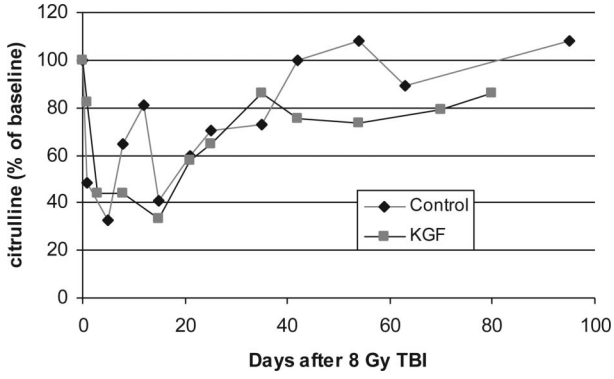

Figure 4. Effect of Keratinocyte Growth Factor (KGF) administration (day 1–3; 250 μg/kg) on citrullinemia evaluated using high performance liquid chromatography in a 8 Gy (total body irradiation) monkey model (each group $n = 2$; Hérodin and Drouet, unpublished data). KGF as a single treatment was unable to protect gastro-intestinal tract and lymphocyte counts did not significantly differ in treated and control animals.

2009). In fact, further studies are required to define targets (Figure 5) and optimise specific cell delivery. As an example, our group recently checked tumour necrosis alpha (TNF-α) and chemokine (C-X-C motif) ligand 1(CXCL-1) genes. In spite of a tendency to delay lethality if early injected (2 h after irradiation) which corresponded to a delayed medullary gene induction, no statistical difference in survival was observed between siRNA-TNF-α and controls. However it has to be pointed out that the TNF-α response is complex as this Janus factor is also involved in the reduction of $CD34^{+}$ hematopoietic stem cell apoptosis (Drouet et al. 2009b).

Cutaneous radiation syndrome

Cutaneous radiation syndrome is the well-described delayed consequence of skin exposure to high doses of ionising radiation (superior to 20 Gy). Radiation burns are characterised by extensive inflammation and clinical evolution made of iterative inflammatory necrotic waves. An early erythema reaction (superior to 4 Gy) is observed followed by hair loss, dry then moist desquamation after several weeks. Radiation ulcer damage represents the classical complication. Specifying the scope of the highly exposed area is a difficult task. Current treatment includes surgery, transient xenografts, artificial derma and then autografting. In historic cases, skin damages and myelosuppression were frequently associated. Thus, the patient named O in Tokai-Mura accident exhibited severe radiation burns with general skin loss (after three weeks) and massive effusion. He experienced repeated allogeneic skin grafts.

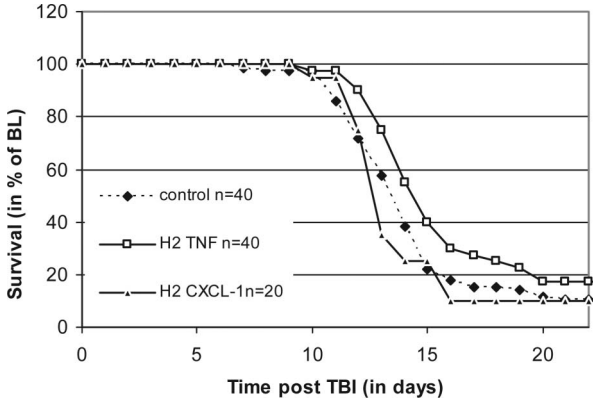

Figure 5. Effect of small interfering RNA (siRNA) injection on the survival of mice irradiated at lethal dose 90 % over 30 days. Mice were irradiated using a ^{60}Co gamma source (9 Gy total body irradiation; dose rate 25 cGy/min). Two hours after irradiation, mice were given Mock siRNA, siRNA targeted towards TNF-α or CXCL-1 (Dharmacon, Lafayette CO, USA; Drouet 2009b). No statistical difference in survival was observed between treated animals and controls in spite of a delayed medullary gene induction with respect to TNF-α.

Based on experimental mouse studies (François et al. 2007), local injections of autologous bone marrow multipotent mesenchymal stem cells to favour wound healing have been proposed. Today four patients suffering from radiation burns have been grafted (Lataillade et al. 2007). In these cases, surgery was associated with skin autografting and cell injection; clinical outcome was considered as good with no recurrence of inflammatory waves – estimated dose at the centre of the skin surface lesion 20 Gy or more, excision up to 10 cm in diameter. However, these results remain controversial due to the limited size of the cohort, the frequent combining of skin and mesenchymal stem cells grafting and obviously the absence of controls. Regarding alternative source of multipotent stem cells to cure radiation burns, Ebrahimian et al. (2009) recently reported in a mouse model (20 Gy, locally) that grafted adipocyte derived stem cells participate in dermal wound healing in physiological and pathological conditions via reepithelialisation and proangiogenic activity. This was also assessed by other groups (Figure 6; Agay et al. 2009a).

Thus further studies are required to validate this strategy and to propose guidelines: Recommended time to cell injection, minimal amounts of cells to be grafted etc. This is the reason why we have set up a new minipig model close to human (vascularisation morphology) for the improvement of cell therapy strategies applied to radiation burns. Such models will allow us to definitely state on the topic: Migratory capacity of mesenchymal stem cells as

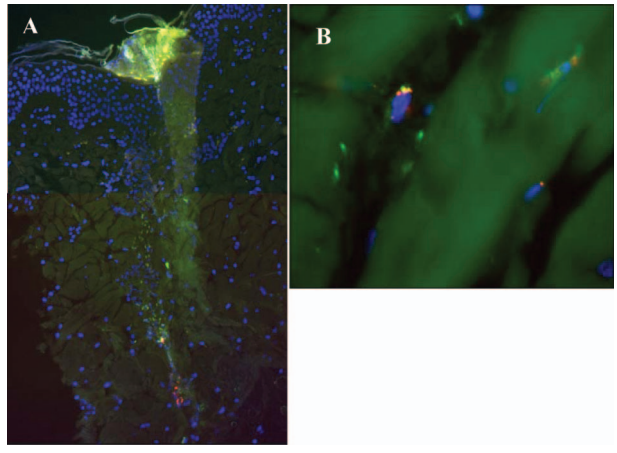

Figure 6. Mesenchymal stem cells, intradermally injected, migrated between collagen fibre bundles. Three days following injection of pig autologous mesenchymal stem cells, specifically labelled stem cells (arrows) are visible in the dermis. (A) Blood clot and needle channel stain brightly green. Nuclei in blue (4′,6-diamidino-2-phenylindole/DAPI), collagen fibres in green, specifically-labelled mesenchymal stem cells contain red signal dots (B). Magnification in the original 100× (A) and 630× (B) Unpublished data from H. Scherthan, Bundeswehr Institute of Radiobiology, Munich, FRG (manuscript in preparation).

illustrated by the Figure 6 (Agay et al. 2009b), trophic factor secretion versus transdifferentiation, and eventually therapeutic potential of allogeneic mesenchymal stem cells. Finally, the safety of the mesenchymal stem cells has to be clearly established, especially regarding their potential to undergo malignant transformation which has only been reported in mice (Miura et al. 2006, Tarte et al. 2009).

Prospective guidelines

The establishment of a definite consensus on cytokine use in radiation accidents remains a difficult task (Fliedner et al. 2009). Haematopoietic score 3-H3 METREPOL patients could be identified by the onset of an early 'abortive' wave of recovery. In such patients it has been postulated that spontaneous recovery will occur – even if delayed – from under-exposed bone marrow area whether or not cytokines are administered. However, most of the experts have recommended the administering of G-CSF/peg-G-CSF 'as soon as possible' in this case. As dose assessment using complete blood cell count (lymphocyte decrease) and cytogenetics from lymphocyte culture is also recommended, treatment in practical would be delayed. From our point of view, the emergency antiapoptotic cytokine concept emphasises the importance of clinical dosimetry (even if limited: early vomiting etc.) in cases of high-dose exposure and early diagnosis/hospitalisation to benefit from the crucial therapeutic window of the first 24 h.

In fact, the two main points to consider are the delay of diagnosis/hospitalisation and the estimated damage. Tables I–IV provide information on the different diagnoses, triage of victims and guidelines for suggested cytokine treatments. In some cases, hospitalisation would occur rapidly and diagnosis of high-dose exposure would be obvious (see Tokaï Mura for example). According to the anti-apoptotic strategy, such victims (Haematopoietic score 4-H4) should be considered as emergencies regarding bone marrow protection and the more efficient cytokine combination should be discussed. In contrast, delayed diagnosis (panic, delayed consultation) would be frequent in hidden sources scenario and not everyone would benefit from an emergency treatment. However, enlarged combination including G-CSF/peg-G-CSF, SCF and thrombopoietin would give a better chance to stimulate residual hematopoiesis in the case of medullary eradication, provided that thrombopoietin-mimetics demonstrate sustained activity in this context. Moreover, in the context of mass casualties, albeit reactive hospital networks exist, there are still concerns about the feasibility of massive hematopoietic stem cells grafting. In addition, there will be cases of combined pathology with no reasonable prospect of recovery (Haematopoietic score 3-H3 or Haematopoietic score 4-H4). In such cases, cytokine injection could be proposed as a palliative approach. On the other hand, one must keep in mind that, in a crisis context, supportive care – antibiotics – would at least give a chance to Haematopoietic score 3-H3 patients. Fortunately, in mass radiation accidents, the highest number of victims could be in the lowest response categories.

In conclusion, from our point of view, the METREPOL categorisation system represents an interesting concept to complete the dose evaluation

Table I. Prospective cytokine guideline. In the case of early diagnosis/hospitalisation, victims should be considered as an emergency to allow optimised cytokine treatment including the reduction of radiation-induced cell death.

Diagnosis	Victims to be treated/Scoring	Cytokine treatment
Acute radiation syndrome ± localised exposure (C)	Scores 2/3 transitory scoring or delayed H3/H4 ± C3/C4 score	To prevent apoptosis and/or stimulate residual haematopoiesis
Early diagnosis and hospitalisation (within 24 h)	Mainly immediate or early vomiting (<6 h) ± headache, fatigue, hypotension, early cutaneous erythema, H3/H4 as per Response Category (METREPOL) as positive criteria for starting cytokine treatment then biological/physical dosimetry for confirmation/modification ± consider HSC transplant (HLA typing, search donor)	Victims should be considered as an emergency Tpo* substitutes to be especially considered if H3/H4 + C3/C4, i.e., rebuttal for transplantation
Delayed diagnosis and hospitalisation	CBC and biological parameters to evaluate bone marrow damage and start cytokine treatment Then biological/physical dosimetry for confirmation/modification ± consider HSC transplant (HLA typing, search donor)	Not urgent but the sooner the better Tpo* substitutes see above

METREPOL categorisation: H = haematopoiesis scoring (Haematopoietic score 3-H3, Haematopoietic score 4-H4), C = cutaneous scoring (Cutaneous score 3-C3, Cutaneous score 4-C4); CBC = complete blood cell counts; HSC = haematopoietic stem cells; Epo = erythropoietin, Tpo = thrombopoietin, KGF = Keratinocyte Growth Factor, SCF = Stem Cell Factor, PegG-CSF = pegylated granulocyte growth factor. *Efficacy of Thrombopoietin substitutes validated for Idiopathic Thrombocytopenic Purpura; to be validated in ad hoc whole body irradiated animal model.

Table II. Prospective cytokine guideline according to Response Category (METREPOL) and classical dose categorisation: Combining cytokine to enhance their efficacy.

PegG-CSF/G-CSF + SCF + KGF/Epo	H3 Response Category (METREPOL)	3–7 Gy
PegG-CSF/G-CSF + SCF + Tpo substitutes* + KGF/Epo	H4 Response Category (METREPOL)	>7 Gy

METREPOL categorisation: H = haematopoiesis scoring, C = cutaneous scoring; CBC = complete blood cell counts; HSC = haematopoietic stem cells; Epo = erythropoietin, Tpo = thrombopoietin, KGF = Keratinocyte Growth Factor, SCF = Stem Cell Factor, PegG-CSF = pegylated granulocyte growth factor. *Efficacy of Thrombopoietin substitutes validated for Idiopathic Thrombocytopenic Purpura; to be validated in ad hoc whole body irradiated animal model. H3: Haematopoietic score 3; H4: Haematopoietic score according to Response Category (METREPOL) concept.

Table III. Prospective cytokine guideline: treatment schedule.

Cytokine/drug	Dose for an adult victim; to be discussed and adapted for children and the elderly	Treatment schedule, duration
G-CSF (filgrastim, lenograstim)	5 μg/kg/day	day 0, then daily until recovery
Erythropoietin (e.g., Darbepoietin alpha)	500 μg	Delayed: once every 21 days
SCF	20 μg/kg/day	Early: single delayed: 7 days
KGF (palifermin)	60 μg/kg/day	day 0, day 1 and day 2
Tpo substitute: Romiplostim*	10 μg/kg 3 μg/kg/day	early H4: single delayed H4: 3 weeks
Tpo substitute: Eltrombopag*	50 mg/day	delayed H4: 21 days

*Efficacy of Thrombopoietin substitutes validated for Idiopathic Thrombocytopenic Purpura; to be validated in ad hoc whole body irradiated animal model. H3: Haematopoietic score 3; H4: Haematopoietic score according to Response Category (METREPOL) concept.

Table IV. Prospective cytokine guideline: Cytokine and their targets.

Cytokine/drug	Target(s)
G-CSF	Neutrophil recovery ± possible stem cell mobilisation (especially vascular)
SCF	Synergistic effects demonstrated in vitro with other cytokines; anti-apoptotic activity; neutrophil and platelet recovery
Thrombopoietin substitute	Anti-apoptotic activity of Tpo on bone marrow progenitors, in addition to thrombopoietic activity; Following early, single injection of Tpo and G-CSF in irradiated monkeys, no competitive effect observed, but neutrophil and platelet recovery improved.
Erythropoietin	Pleiotropic factor and anaemia treatment
KGF	Epithelium protection and regeneration ± possible immune protection

to medically score irradiated victims. The optimisation of current cytokine therapy – haematopoietic growth factors and other pleiotropic or tissue-specific cytokines – in order to raise the transplantation threshold remains an important goal in view of managing future radio-nuclear crisis (European Group for Blood and Marrow Transplantation, Nuclear Accident Committee recommendations; website in preparation). The size crisis would obviously impacts the treatment as, due to time accessibility and price, it is unlikely that sufficient amounts of cytokines would be available in the context of mass casualties. However cytokines could be stockpiled in selected centres, comparable to war emergency treatment at the battlefield level. Such pre-positioning may allow their early and useful use in limited cohorts of highly irradiated victims. Regarding the adult mesenchymal stem cells debate, further studies are required to definitely state but the first results in clinic are encouraging. In addition, the first results obtained from the minipig model of Cutaneous Radiation Syndrome we have developed suggests/underlines the capacity of mesenchymal stem cell, as a single treatment, to significantly favour wound healing and especially dermis recovery (manuscript in preparation). Finally, identifying and training networks of complementary specialists – including those from cell therapy units, led by haematologists or other specialists – to be activated in this context would be of interest.

Declaration of interest: The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

References

Adams GB, Scadden DT. 2008. A niche opportunity for stem cell therapeutics. Gene Therapy 15:96–99.

Agay D, Garrigou P, Forcheron F, Grenier N, Touvard L, Chancerelle Y, Meineke V, Hérodin F, Drouet M. 2009a. Adipocyte tissue-derived stem cells to treat severe cutaneous radiation syndrome: Preliminary study in a rat model. Bone Marrow Transplantation 43 (Suppl. 1):S205.

Agay D, Forcheron F, Scherthan H, Grenier N, Meineke V, Hérodin F and Drouet M. 2009b. Mesenchymal stem cell grafting to treat cutaneous radiation syndrome: Development of a new minipig model. Blood (ASH Annual Meeting Abstracts) 114:4574.

Agnello D, Bigini P, Villa P, Mennini T, Cerami A, Brines ML, Ghezzi P. 2002. Erythropoietin exerts an anti-inflammatory effect on the CNS in a model of experimental autoimmune encephalomyelitis. Brain Research 952:128–134.

Aird WC. 2003. The role of the endothelim in severe sepsis and in multiple organ dysfunction syndrome. Blood 101:3765–3777.

Akashi M. Role of infection and bleeding in multiple organ involvement and failure. 2005. British Journal of Radiology (Suppl. 27):69–74.

Araki S, Israel S, Leskov KS, Criswell TL, Beman M, Klokov DY, Sampalth L, Reinicke KE, Cataldo E, Mayo LD, Boothman DA. 2005. Clusterin proteins: Stress-inducible polypeptides with proposed functions in multiple organ dysfunction. British Journal of Radiology (Suppl. 27):106–113.

Baranov A, Gale R, Guskova A, Piatkin E, Selidovkin G, Muravyova L, Champlin RE, Danilovna N, Yevseeva L, Petrosyan L. 1989. Bone marrow transplantation after the Chernobyl nuclear accident. New England Journal of Medicine 321:205–212.

Baranov AE, Selidovkin GD, Butturini A, Gale RP. 1994. Hematopoietic recovery after 10-Gy acute total body radiation. Blood 83:596–599.

Barton R, Cerra FB. 1989. The hypermetabolism multiple organ failure syndrome. Chest 96:1153–1160.

Bertho JM, Roy L, Souidi M, Benderitter M, Guegen Y, Lataillade JJ, Prat M, Fagot T, De Revel T, Gourmelon P. 2008. New biological indicators to evaluate and monitor radiation-induced damage: An accident case report. Radiation Research 169:543–550.

Blakely WF, Ossetrova NI, Manglapus GL, Salter CA, Levine IH, Jackson WE, Grace MB, Prasanna PGS, Sandgren DJ, Ledney GD. 2007. Amylase and blood cell-count haematological radiation-injury biomarkers in a rhesus monkey radiation model-use of multiparameter and integrated biological dosimetry. Radiation Measurement 42:1164–1170.

Bone RC. 1996. Immunologic dissonance: A continuing evolution in our understanding of the systemic inflammatory response syndrome (SIRS) and the multiple organ dysfunction syndrome (MODS). Annals of Internal Medicine 125:680–687.

Bussel JB, Cheng G, Saleh MN, Psaila B, Kovaleva L, Meddeb B, Kloczsko J, Hassani H, Mayer B, Stone NL, Arning M, Provan D, Jenkins JM. 2007. Eltrombopag for the treatment of chronic idiopathic thrombocytopenic purpura. New England Journal of Medicine 357:2237–2247.

Butturini A, De Souza PC, Gale RP, Cordiero JM, Lopes DM, Neto C, Cunha CB, De Souza CE, Ho WG, Tabak DG, Burla A and The Navy Hospital Radiation Team. 1988. Use of recombinant granulocyte-macrophage colony stimulating factor in the Brazil radiation accident. The Lancet 2:471–475.

Chapel A, Bertho JM, Bensidhoum M, Fouillard L, Young RG, Frick J, Demarquay C, Cuvelier F, Mathieu E, Trompier F, Dudoignon N, Germain C, Mazurier C, Aigueperse J, Borneman J, Gorin NC, Gourmelon P, Thierry D. 2003. Mesenchymal stem cells home to injured tissues when co-infused with hematopoietic cells to treat a radiation-induced multi-organ failure syndrome. The Journal of Gene Medicine 5:1028–1038.

Chaudhry MA. 2008. Biomarkers for human radiation exposure. Journal of Biomedical Science 15:557–563.

Chute JP, Muramoto GG, Salter AB, Meadows SK, Rickman DW, Chen B, Himburg HA, Chao NJ. 2007. Transplantation of vascular endothelial cells mediates the hematopoietic recovery and survival of lethally irradiated mice. Blood 109:2365–2372.

Crisan M, Yap S, Casteilla L, Chen CW, Corselli M, Park TS, Andriolo G, Sun B, Zheng B, Zhang L, Norotte C, Teng PN, Traas J, Schugar R, Deasy BM, Badylak S, Buhring HJ, Giacobino JP, Lazzari L, Huard J, Péault B. 2009. A perivascular origin for mesenchymal stem cells in multiple human organs. Cell Stem Cell 3:229–230.

Dainiak N, Waselenko JK, Armitage JO, MacVittie TJ, Farese AM. 2003. The hematologist and radiation casualties. Hematology American Society of Hematology Education Program. pp 473–496.

Dainiak N, Ricks RC. 2005. The evolving role of haematopoietic cell transplantation in radiation injury: Potentials and limitations. British Journal of Radiology (Suppl. 27):169–74.

Dainiak N, Berger P, Albanese J. 2007. Relevance and feasibility of multi-parameter assessment for management of mass casualties from a radiological event. Experimental Hematology 35(Suppl. 1):17–23.

De Revel T, Fagot T, Malfuson JV, Souleau B, Nedellec G. 2005. Le traitement de l'aplasie médullaire radio-induite. In: De Revel T, Gourmelon P, Vidal D, Renaudeau, editors. The terrorist threat nuclear, radiological, biological, chemical – a medical approach. Montrouge: John Libbey Eurotext. pp 105–111.

Dörr W, Heider K, Spekl K. 2005. Reduction of oral mucositis by palifermin (rHuKGF): Dose-effect of rHuKGF. International Journal of Radiation Biology 81:557–565.

Drouet M, Mathieu J, Grenier N, Multon E, Sotto JJ, Hérodin F. 1999. The reduction of in vitro radiation-induced Fas-related apoptosis in $CD34^+$ progenitor cells by SCF, Flt-3 Ligand, TPO and IL-3 in combination resulted in $CD34^+$ cell proliferation and differentiation. Stem Cells 17:273–285.

Drouet M, Mourcin F, Grenier N, Leroux V, Denis J, Mayol JF, Lataillade JJ, Hérodin F. 2004. Single administration of stem cell factor, flt-3 ligand, megakaryocyte growth and development factor and interleukin-3 in combination soon after irradiation prevents nonhuman primates from myelosuppression: Long-term follow-up of hematopoïesis. Blood 103:878–885.

Drouet M, Mourcin F, Grenier N, Delaunay C, Mayol JF, Lataillade JJ, Peinnequin A, Hérodin F. 2005. Mesenchymal stem cells rescue $CD34^+$ cells from radiation-induced apoptosis and sustain hematopoietic reconstitution after co-culture and co-grafting in lethally irradiated baboons: Is autologous stem cell therapy in nuclear accident settings hype or reality? Bone Marrow Transplantation 35:1201–1209.

Drouet M, Garrigou P, Mayol JF, Delaunay C, Peinnequin A, Hérodin F. 2008a. In vivo injection of synthetic small interfering RNA to lethally irradiated mouse as a new model to study apoptotic pathways. Blood 112 (ASH Annual Meeting Abstracts):1336.

Drouet M, Garrigou P, Collombet JM, Grenier N, Mayol JF, Hérodin F. 2008b. Erythropoietin in combination with SCF, FLT3-ligand, TPO and IL-3 promotes survival of lethally irradiated mice: A pleiotropic cytokine to mitigate radiation-induced damages? Experimental Hematology 36 (Suppl. 1):S24.

Drouet M, Garrigou P, Grenier N, Delaunay C, Mayol JF, Hérodin F. 2009a. Hedgehog signaling and hematopoiesis regulation: Preliminary study in myelosuppressed adult non human primate model. Blood (ASH Annual Meeting Abstracts) 114:4603.

Drouet M, Mayol JF, Delaunay C, Mathieu J, Peinnequin A, Hérodin F. 2009b. In vivo injection of synthetic small interfering RNA to lethally irradiated mouse as a new model to study the systemic inflammatory response syndrome. Experimental Hematology 37(Suppl. 1):S78.

Ebrahimian TG, Pouzoulet F, Squiban C, Buard V, André M, Cousin B, Goumelon P, Benderitter M, Casteilla L, Tamarat R. 2009. Cell therapy based on adipose tissue-derived stromal cells promotes physiological and pathological wound healing. Arteriosclerosis, Thrombosis, and Vascular Biology 29:503–510.

Fagot T, Bottolier JF, Roy L, Samson T, Desangles F, Lataillade JJ, Bertho JM, Cailliot C, Gourmelon P, De Revel T. 2006. A pure haematological syndrome secondary to an accidental radiation exposure treated with G-CSF, Epo, and SCF. Experimental Hematology 34(Suppl. 1):76.

Farese AM, Hunt P, Grab LB, MacVittie TJ. 1996. Combined administration of recombinant human megakaryocyte growth and development factor and granulocyte colony-stimulating factor enhances multilineage hematopoietic reconstitution in nonhuman primates after radiation-induced marrow aplasia. Journal of Clinical Investigation 97:2145–2151.

Fliedner TM, Friesecke I, Beyrer K, editors. 2001. Medical management of radiation accidents. Manual on the acute radiation syndrome. British Journal of Radiology.

Fliedner TM, Dörr HD, Meineke V. 2005. Multi-organ involvement as pathogenetic principle of the radiation syndromes: A study involving 110 case histories documented in SEARCH and classified as the bases of haematopoietic indicators of effect. British Journal of Radiology (Suppl. 27):1–8.

Fliedner TM, Chao NJ, Bader JL, Boettger A, Case C Jr, Chute J, Confer DL, Ganser A, Gorin NC, Gourmelon P, Graessle DH, Krawisz R, Meineke V, Niederwieser D, Port M, Powles R, Sirohi B, Weinstock DM, Wiley A, Coleman CN. 2009. Stem cells, multiorgan failure in radiation emergency medical preparedness: A US/European consultation workshop. Stem Cells 27:1205–1211.

François S, Bensidhoum M, Mouiseddine M, Mazurier C, Allenet B, Semont A, Frick J, Saché A, Bouchet S, Thierry D, Gourmelon P, Gorin NC, Chapel A. 2006. Local irradiation induces not only homing of human mesenchymal stem cells (huMSC) at exposed sites but promotes their widespread engraftment to multiple organs: A study of their quantitative distribution following irradiation damages. Stem Cells 24:1020–1029.

François S, Mouissedine M, Mathieu N, Semont A, Monti P, Dudoignon N, Saché A, Boutarfa A, Thierry D, Gourmelon P, Chapel A. 2007. Human mesenchymal stem cells favour healing of the cutaneous radiation syndrome in a xenogenic transplant model. Annals of Hematology 86:1–8.

Gando S, Nakanishi Y, Tedo I. 1995. Cytokines and plasminogen activator inhibitor-1 in posttrauma disseminated intravascular coagulation: Relationship to multiple organ dysfunction syndrome. Critical Care Medicine 23:1835–1842.

Garcia-Olmo D, Garcia-Arranz M, Herreros D, Pascual I, Peiro C, Rodriguez-Montes JA. 2005. A phase I clinical trial of the treatment of Crohn's fistula by adipose mesenchymal stem cell transplantation. Diseases of the Colon & Rectum 48:1416–1423.

Gaugler MH. 2005. A unifying system: Does the vascular endothelium have a role to play in multi-organ failure following radiation exposure? British Journal of Radiology (Suppl. 27):100–105.

Godin PJ, Buchman TG. 1996. Uncoupling of biological oscillators: A complementary hypothesis concerning the pathogenesis of multiple organ dysfunction syndrome. Critical Care Medicine 24:1107–1116.

Gordon B, Lyden E, Lynch J, Tarantolo S, Pavletic ZS, Bishop M, Reed E, Kessinger A, Haire W. 2000. Central nervous system dysfunction as the first manifestation of multiple organ dysfunction in stem cell transplant patients. Bone Marrow Transplantation 25:79–83.

Gorin NC, Fliedner TM, Gourmelon P, Ganser A, Meineke V, Sirohi B, Powles R, Apperly J. 2006. Consensus conference on European preparedness for haematological and other medical management of mass radiation accidents. Annals of Hematology 85:671–679.

Gourmelon P, Lebaron-Jacob L, Marquette C, Clarencon D. 2002. Radiation-induced neurovascular injuries. In: Fliedner TM, Feinendegen LE, Hopewel JW, editors. Chronic irradiation: Tolerance and failure in complex biological systems. British Journal of Radiology (Suppl. 26):210–216.

Gourmelon P, Marquette C, Agay D, Mathieu J, Clarençon D. 2005. Involvement of the central nervous system in radiation-induced multi-organ dysfunction and/or failure. British Journal of Radiology (Suppl. 27):62–68.

Hassoun HT, Kone BC, Mercer DW, Moody FG, Weisbrodt NW, Moore FA. 2001. Post-injury multiple organ failure: The role of the gut. Shock 15:1–10.

Heeschen C, Aicher A, Lehmann R, Fichtlscherer S, Vasa M, Urbich C, Mildner-Rihm C, Martin H, Zeiher AM, Dimmeler S. 2003. Erythropoietin is a potent physiological stimulus for endothelial progenitor mobilization. Blood 102:1340–1346.

Hérodin F, Bourin P, Mayol JF, Lataillade, Drouet M. 2003. Short-term injection of antiapoptotic cytokine combinations soon after lethal gamma irradiation promotes survival. Blood 101:2609–2616.

Hérodin F, Drouet M. 2005. Cytokine-based treatment of accidentally irradiated victims and new approaches. Experimental Hematology 33:1071–1080.

Hérodin F, Grenier N, Drouet M. 2007. Revisiting cytokine based-treatment. Experimental Hematology 35(Suppl. 1):28–33.

Hérodin F, Roy L, Vaurijoux A, Gregoire E, Martin C, Grenier N, Alonso A, Mayol JF, Drouet M. 2007. Single injection of Stem Cell Factor, Flt-3 ligand, Thrombopoietin, interleukin-3 combined with Pegylated G-CSF mitigates myelosuppression in highly irradiated nonhuman primates. Experimental Hematology 35:1172–1181.

Hotchkiss RS, Coopersmith CM, McDunn JE, Ferguson TA. 2009. The sepsis seesaw: Tilting toward immunosuppression. Nature Medicine 15:496–497.

International Atomic Energy Agency (IAEA). 1993. The radiological accident in Soreq. Vienna: IAEA.

Jackson WL, Gallagher C, Myhand RC, Waselenko JK. 2005. Medical management of patients with multiple organ dysfunction arising from acute radiation syndrome. British Journal of Radiology (Suppl. 27):161–168.

Kiel MJ, Yilmaz OH, Iwashita T, Terhorst C, Morrison SJ. 2005. SLAM family receptors distinguish hematopoietic stem and progenitor cells and reveal endothelial niches for stem cells. Cells 121:1109–1121.

Karp JM, Leng Teo GS. 2009. Mesenchymal stem cell homing: The devil is in the details. Cell Stem Cell 4:206–216.

Kuter DJ. 2007. New thrombopoietic growth factors. Blood 109:4607–4616.

Kuter DJ, Bussel JB, Lyons RM, Pullarkat V, Gernsheimer TB, Senecal FM, Alerdort LM, George JN, Kessler CM, Sanz MA, Liebmnan HA, Slovick ST, De Wolf JT, Bourgeois E, Guthrie TH, Newland A, Wasser JS, Hamburg SI, Grande C, Lefrère F, Lichtin AE, Tarantino MD, Terebelo HR, Viallard JF, Cuevas FJ, Go RS, Henry DH, Redner RL, Rice L, Schipperus MR, Guo DM, Nichol JL. 2008. Efficacy of romiplostim in patients with chronic immune thrombopoietic purpura: A double-blind randomised controlled trial. The Lancet 371:395–403.

Lataillade JJ, Doucet C, Bey E, Carsin H, Huet C, Clairand I, Bottollier-Depois JF, Chapel A, Ernou I, Gourven M, Boutin L, Hayden A, Carcamo C, Buglova E, Joussemet M, De Revel T, Gourmelon P. 2007. New approach to radiation burn treatment by dosimetry-guided surgery combined with autologous mesenchymal stem cell therapy. Regenerative Medicine 2:785–794.

Li XM, Hu Z, Jorgensen ML, Wingard JR, Slayton WB. 2008. Bone marrow sinusoidal endothelial cells undergo non apoptotic cell death and are replaced by proliferating sinusoidal cells in situ to maintain the vascular niche following lethal irradiation. Experimental Hematology 36:1143–1156.

Linard C, Griffiths NM, Esposito V, Aigueperse J, Gourmelon P. 1997. Changes in gut neurotensin and modified colonic motility following whole-body irradiation in rat. International Journal of Radiation Biology 71:581–585.

Lombaert IM, Brunsting JF, Wierenga PK, Kampinga HH, de Haan G, Coppes RP. 2008. Keratinocyte growth factor prevents radiation damage to salivary gland by expansion of the stem/progenitor pool. Stem Cells 26:2595–2601.

Maekawa K. 2001. Overview of medical care for highly exposed victims in the Tokaimura accidents. In: Ricks RC, Berger ME, O'Hara FM editors. The medical basis for radiation-accident preparedness. The clinical care of victims. London: The Parthenon Publishing Group. pp 313–318.

McConnell KW, Muenzer JT, Chang KC, Davis CG, McDunn JE, Coopersmith CM, Hilliard CA, Hotchkiss RS, Grigsby PW, Hunt CR. 2007. Anti-apoptotic peptides protect against radiation-induced cell death. Biochemical and Biophysical Research Communications 355:501–507.

McDunn JE, Muenzer JT, Dunne B, Zhou A, Yuan K, Hoekzema A, Hilliard C, Chang KC, Davis CG, McDonough J, Hunt C, Grigsby P, Piwnica-Worms D, Hotchkiss RS. 2009. An anti-apoptotic peptide improves survival in lethal total body irradiation. Biochemical and Biophysical Research Communications 382:657–662.

MacVittie TJ, Monroy RL, Patchen ML, Souza LM. 1990. Therapeutic use of recombinant human G-CSF in a canine model of sublethal and lethal whole-body irradiation. International Journal of Radiation Biology 57:723–736.

Miura M, Miura Y, Padilla-Nash HM, Molinolo AA, Fu B, Patel V, Seo BM, Sonoyama W, Zheng JJ, Baker CC, Chen W, Ried T, Shi S. 2006. Accumulated chromosomal instability in murine bone marrow mesenchymal stem cells leads to malignant transformation. Stem Cells 24:1095–1103.

Monti P, Wysocki J, Van der Meeren A, Griffiths NM. 2005. The contribution of radiation-induced injury to the gastrointestinal tract in the development of multi-organ dysfunction syndrome or failure. British Journal of Radiology (Suppl. 27):89–94.

Mouiseddine M, Francois S, Semont A, Saché A, Allenet B, Mathieu N, Thierry D, Chapel A. 2007. Human mesenchymal stem cells home specifically to radiation-injured tissues in a non-obese diabetes/severe combined immunodeficiency mouse model. British Journal of Radiology 2007;80(Spec. No. 1):S49–55.

Nagayama H, Misawa K, Tanaka H, Ooi J, Iseki T, Tojo A, Tani K, Yamada Y, Kodo H, Takahashi TA, Yamashita N, Shimazaki S, Asano S. 2002. Transient haematopoietic stem cell rescue using umbilical cord blood for a lethally irradiated nuclear accident victims. Bone Marrow Transplantation 29:197–204.

Nathan C. 2003. Immunology: Oxygen and the inflammatory cell. Nature 422:675–676.

Neelis KJ, Dubbelman YD, Qingliang L, Thomas GR, Eaton DL, Wagemaker G. 1997a. Simultaneous administration of TPO and G-CSF after cytoreductive treatment of rhesus monkeys prevents thrombocytopenia, accelerates platelet and red cell reconstitutuion, alleviates neutropenia and promotes the recovery of immature bone marrow cells. Experimental Hematology 25:1084–1093.

Neelis KJ, Hartong SC, Egeland T, Thomas GR, Eaton DL, Wagemaker G. 1997b. The efficacy of single-dose administration of thrombopoietin with coadministration of either granulocyte/macrophage or granulocyte colony-stimulating factor in myelosuppressed rhesus monkeys. Blood 90:2565–2573.

Ossetrova NI, Blakely WF. 2009. Multiple blood-proteins approach for early-response exposure assessment using an in vivo murine radiation model. International Journal of Radiation Biology 85:837–850.

Otterson MF. 2007. Effects of radiation upon gastrointestinal motility. World Journal of Gastroenterology 13:2684–2692.

Paris F, Fuks Z, Kang A, Capodieci P, Juan G, Ehleiter D, Haimovitz-Friedman A, Cordon-Cardo C, Kolesnick R. 2001. Endothelial apoptosis as the primary lesion initiating intestinal radiation damage in mice. Science 293:293–297.

Parke AL, Liu PT, Parke DV. 2002. Multiple organ dysfunction syndrome. Inflammopharmacology 11:87–95.

Powles RL, Morgenstern GR, Kay HE, McElwain TJ, Clink HM, Dady PJ, Barrett A, Jameson B, Depledge MH, Watson JG, Sloane J, Leigh M, Lumley H, Hedley, Lawler SD, Filshie J, Robinson D. 1983. Mismatched family donors for bone-marrow transplantation and treatment for acute leukaemia. The Lancet 1:612–615.

Salter AB, Meadows SK, Muramoto GG, Himburg H, Doan P, Daher P, Russell IL, Chen B, Chao NJ, Chute JP. 2009. Endothelial progenitor cell infusion induces haematopoietic stem cell reconstitution in vivo. Blood 113:2104–2107.

Schuening FG, Storb R, Goehle S, Graham TC, Appelbaum FR, Hackman R, Sandmaier BM, Urdal DL. 1989. Effect of recombinant human granulocyte colony-stimulating factor on haematopoiesis of normal dogs and on haematopoietic recovery after otherwise lethal total body irradiation. Blood 74:1308–1313.

Schuening FG, Appelbaum FR, Deeg HJ, Sullivan-Pepe M, Graham TC, Hackman R, Szebo KM, Storb R. 1993. Effects of recombinant canine stem cell factor, a c-kit ligand, and recombinant granulocyte colony-stimulating factor on haematopoietic recovery after otherwise lethal total body irradiation. Blood 81:20–26.

Schuller BW, Binns PJ, Riley KJ, Ma L, Hawthorne MF, Coderre JA. 2006. Selective irradiation of the vascular endothelium has no effect on the survival of murine intestinal crypt stem cells. Proceedings of the National Academy of Science of the USA 103:3787–3792.

Seggewiss R, Loré K, Guenaga FJ, Pittaluga S, Mattapallil J, Chow CK, Koup RA, Camphausen K, Nason MC, Meier-Schellersheim M, Donahue E, Blazar BR, Dunbar CE, Douek DC. 2007. Keratinocyte growth factor augments immune reconstitution after autologous hematopoietic progenitor cell transplantation in rhesus macaques. Blood 110:441–449.

Semont A, François S, Mouiseddine M, François A, Saché A, Frick J, Thierry D, Chapel A. 2006. Mesenchymal stem cells increase self-renewal of small intestinal epithelium and accelerate structural recovery after radiation injury. Advanced Experimental Medical Biology 585:19–30.

Singer M, DeSantis V, Vitale D, Jeffcoate W. 2004. Multiorgan failure is an adaptative, endocrine-mediated, metabolic response to overwhelming systemic inflammation. The Lancet 364:545–548.

Smolewski P, Bedner E, Du L, Hsieh TC, Wu JM, Phelps DJ, Darzynkiewicz Z. 2001. Detection of caspases activation by fluorochrome-labeled inhibitors: Multiparameter analysis by laser scanning cytometry. Cytometry 44:73–82.

Song E, Lee SK, Wang J, Ince N, Ouyang N, Min J, Chen J, Shankar P, Lieberman J. 2003. RNA interference targeting Fas protects mice from fulminant hepatitis. Nature Medicine 9:347–351.

Spielberger R, Stiff P, Bensiger W, Gentile T, Weisdorf D, Kewalramani T, Shea T, Yanovitch S, Hansen K, Noga S, McCarty J, LeMaistre CF, Sung EC, Blazard BR, Elhardt D, Chen MG, Emmanouilides C. 2004. Palifermin for oral mucositis after intensive therapy for hematologic cancers. New England Journal of Medicine 351:2590–2598.

Straume T, Amundson SA, Blakely WF, Burns FJ, Chen A, Dainiak N, Franklin S, Leary JA, Loftus DJ, Morgan WF, Pellmar TC, Stolc V, Turteltaub KW, Vaughan AT, Vijayakumar S, Wyrobek AJ. 2008. NASA radiation biomarkers workshop, 27–28 September 2007. Radiation Research 170:393–405.

Taichman RS. 2005. Blood and bone: Two tissues whose fates are intertwined to create the hematopoietic stem-cell niche. Blood 105:2631–2639.

Tarte K, Gaillard J, Lataillade JJ, Fouillard L, Becker M, Mossafa H, Tchirkov A, Rouard H, Henry C, Spinglard M, Dulong J, Monnier D, Gourmelon P, Gorin NG, Sensebe L. 2010. Clinical-grade production of human mesenchymal stromal cells: Occurrence of aneuploidy without transformation. Blood 115:1549–1553.

Tofilon P, Fike J. 2000. The radioresponse of the central nervous system: A dynamic process. Radiation Research 153:357–370.

Van der Meeren A, Vandame M, Squiban C, Gaugler MH, Mouthon MA. 2003. Inflammatory reaction and changes in expression of coagulation proteins on lung endothelial cells after total-body irradiation in mice. Radiation Research 160:637–646.

Van der Meeren A, Monti P, Vandamme M, Squiban C, Wysocki J, Griffiths N. 2005. Abdominal radiation exposure elicits inflammatory responses and abscopal effects in the lungs of mice. Radiation Research 163:144–152.

Villa P, Bigini T, Mennini D, Agnello D, Laragione A, Cagnotto B, Viviani B, Marinovich M, Cerami A, Coleman TR, Brines M, Ghezzi P. 2003. Erythropoietin selectively attenuates cytokine production and inflammation in cerebral ischemia by targeting neuronal apoptosis. Journal of Experimental Medicine 198:971–975.

Waselenko JK, MacVittie TJ, Blakely WF, Pesik N, Wiley AL, Dickerson WE, Tsu H, Confer DL, Coleman CN, Seed T, Lowry P, Armitage JO, Dainiak N. 2004. Medical management of the acute radiation syndrome: Recommendations of the strategic national stockpile radiation working group. Annals of Internal Medicine 140:1037–1051.

Weisdorf D, Chao N, Waselenko JK, Dainiak N, Armitage JO, McNiece I, Confer D. 2006. Acute radiation injury: Contingency planning for triage, supportive care, and transplantation. Biology of Blood and Marrow Transplantation 12:672–682.

Wesche-soldato DE, Chung CS, Lomas-Neira J, Doughty LA, Gregory SH, Ayala A. 2005. In vivo delivery of caspase-8 or fas siRNA improves the survival of septic mice. Blood 106:2295–2301.

Young RW. 1987. Acute radiation syndrome. In: Conklin JJ, Walker IW, editors. Military radiobiology. London: Academic Press. pp 165–190.