Influence of shear stress on the anti-inflammatory response to low-dose ionizing radiation

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RATIONALE

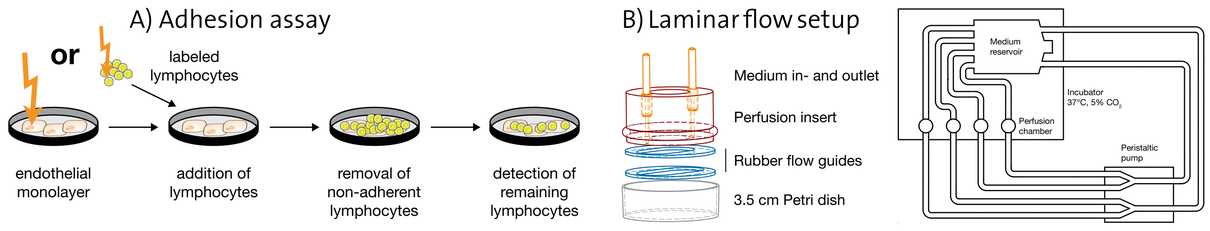
The therapeutic benefits of low-dose radiation are well established for painful degenerative joint diseases and other inflammatory processes.[1,2] However, there is little known about the underlying cellular mechanisms.[3] One hallmark of inflammation is the adhesion of peripheral blood lymphocytes (PBL) to endothelial cells (EC) and can therefore be used as an indication for the onset of inflammation or the inhibition of an ongoing inflammatory process. We investigated the effect of X-ray and carbon ion irradiation (the latter as a surrogate for radon-emitted alpha-particles) on the adhesion of PBL to EC.[4] Because PBL are more radiosensitive than EC and thus apoptosis could play a role in the development of inflammation, we also examined the occurrence ofapoptosis in irradiated PBL.

METHODS

Human endothelial cells (HMVEC, microvasculature; HPAEC, pulmonary aorta) or peripheral blood lymphocytes (PBL) were irradiated with X-rays or carbon ions (150 MeV/u; 30 keV/µm).

Following irradiation, cultivation of endothelial cells in the presence of TNF-a (1 ng/ml; R&D systems) under static (A) or laminar flow (B) conditions. After 24h PBL were added and the adhesion assay was performed.

Annexin apoptosis assays were performed with irradiated PBL to assess influence of cell death on adhesion under static or laminar flow conditions.



X-IRRADIATED ENDOTHELIAL CELLS

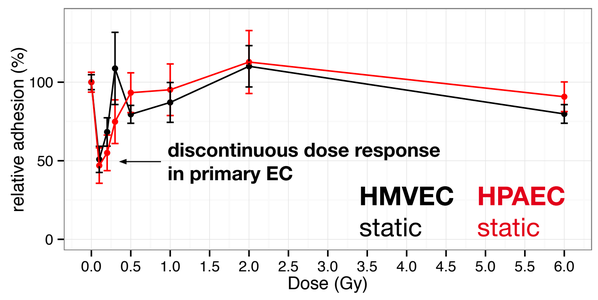


Fig 1 Inflammatory stimulation of EC with TNF-apha enhances the adhesion of PBL (here 100%), whereas stimulation combined with exposure to low doses of photons (significantly for 0.1–0.5 Gy) of EC reduces the PBL adhesion, which was not the case for higher doses. (N=2, n=2)

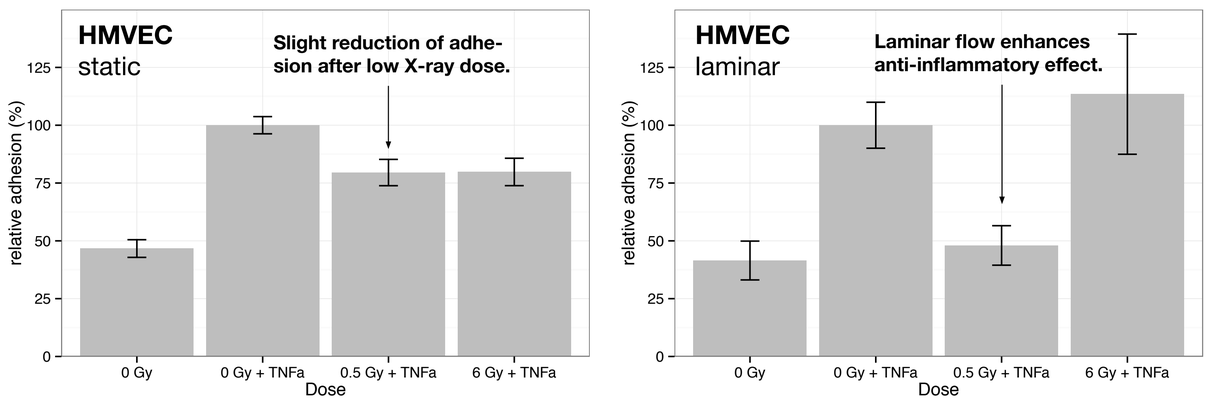


Fig 3 Comparing static and laminar conditions for selected doses in HMVEC reveals that the reduction of adhesion of PBL is more pronounced for laminar (right panel) compared to static conditions (left panel).

X-IRRADIATED LYMPHOCYTES

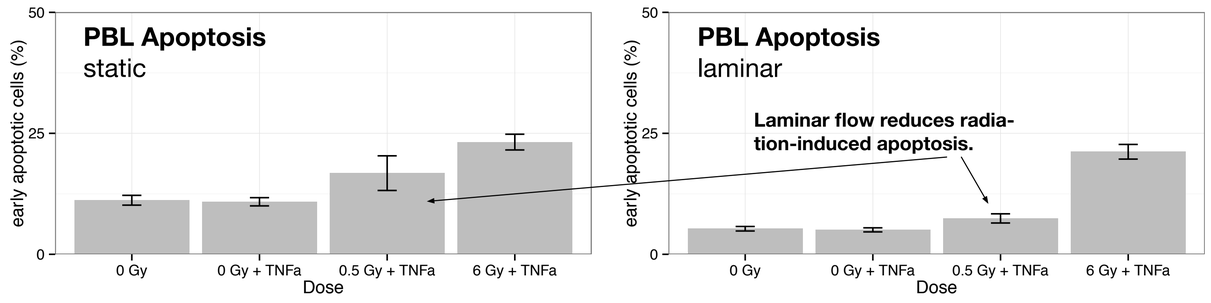


Fig 3 Laminar conditions (right) compared to static conditions (left) reduce apoptosis in lymphocytes irradiated with a low (0.5 Gy), but not with a high photon dose (6 Gy) (stimulated HMVEC, irradiated PBL; N=2, n=4).

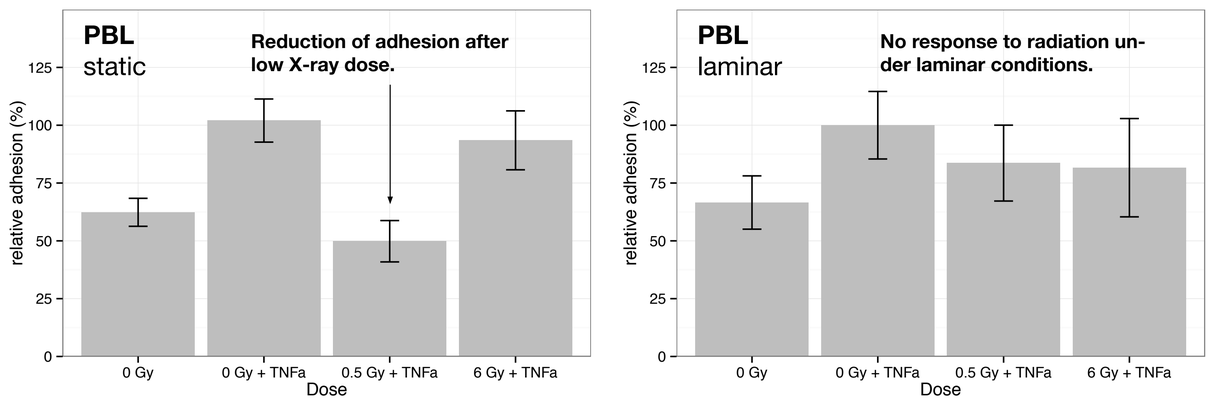


Fig 4 Irradiation of PBL leads to the opposite effect as irradiation of HMVEC: when performed under static conditions, irradiation with 0.5 Gy X-rays reduces the adhesion of PBL to HMVEC, whereas under laminar flow conditions no effect is visible at the same dose.

CARBON ION-IRRADIATED HMVEC

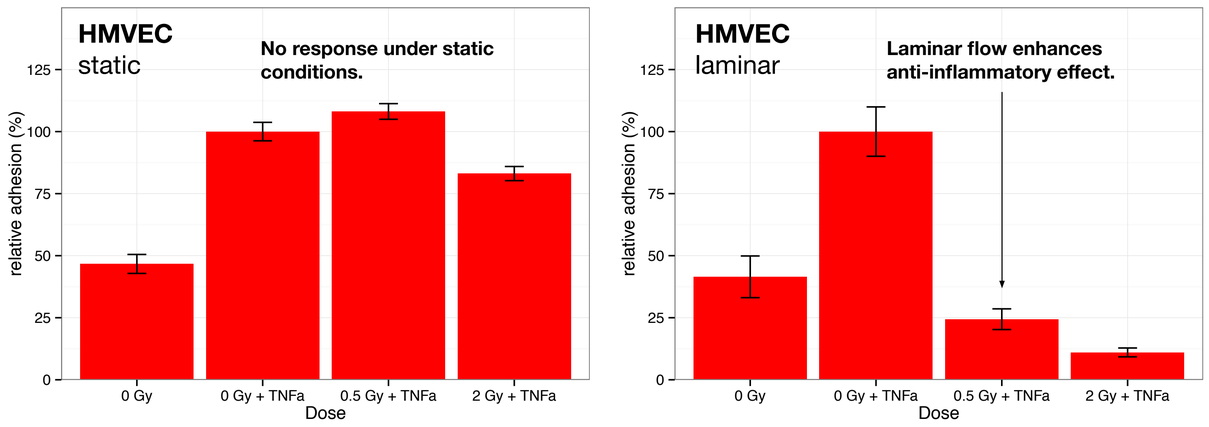


Fig 5 Irradiation with carbon ions does not lead to a reduction of PBL adhesion under static conditions (0.5 Gy). However, under laminar conditions, carbon ions enhance the reduction of adhesion at low and moderate doses (N=2, n=2).

CONCLUSIONS

Primary human endothelial cells originating from different tissues demonstrate a discontinuous dose response for PBL adhesion. Shear stress enhances this effect.

At least under laminar conditions, carbon ions show a more pronounced reduction of adhesion compared to X-irradiation. This supports the idea that low doses of charged particles, e.g., originating from radon decay, can inhibit the onset of an inflammatory response.

PBL apoptosis does not show an apparent link to PBL adhesion. The reduction of adhesion we observed in the static adhesion assay with irradiated PBL might be related to the assay conditions, given that under laminar conditions PBL do not adhere less after radiation exposure.

In sum, we confirm that low doses (0.1–0.5 Gy) of X-rays and carbon ions have anti-inflammatory effects in *primary* cells, especially under shear stress.

Our ongoing research focuses on the underlying molecular mechanism, in particular the impact of TNF-alpha stimulation and the role of TGF-beta and other cytokines released after irradiation.

References
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