Ultraviolet B Light Inactivates Bone Marrow T Lymphocytes but Spurs Hematopoietic Precursor Cells

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Bone marrow cells from ten normal donors were exposed to ultraviolet (UV) light for total exposures of 0.1 to 100 mJ/cm², and assayed for granulocyte-macrophage colony-forming units (CFU-GM), erythroid burst-forming units (BFU-E), and phytohemagglutinin (PHA)-stimulated proliferative responses. After exposure to UVC CFU-GM, BFU-E and PHA responses showed a UV dose-dependent sharp decrease to levels <1% of controls with 0.5, 2.0, and 10 mJ/cm², respectively. With UVB, PHA responses were most sensitive, declining to <1% at 5 mJ/cm². BFU-E decreased to <1% of control with 15 mJ/cm² UVB. CFU-GM, at UVB doses of 0.1 to 2.0 mJ/cm², increased to 125% to 130% of control and decreased to <1% only at exposures >20 mJ/cm². Thus, these studies show that UVB, but not UVC light, can be used to inactivate bone marrow T lymphocytes selectively while sparing hematopoietic precursor cells. The data suggest that UVB irradiation can be used for T-lymphocyte purging for allogeneic marrow transplantation.

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CFU-GM. Colony numbers are expressed per 10^5 cells. Nonirradiated marrow cells served as controls.

Presentation of results. In evaluating the effect of UV light on the response of marrow cells to PHA, responses of nonirradiated marrow mononuclear cells were considered as 100%. The responses of UV-treated cells were corrected for background proliferation in cells cultured in medium alone and the delta value was expressed as percent of control. Similarly, for BFU-E and CFU-GM, the numbers of colonies per 10^5 cells obtained with nonirradiated cells served as the 100% reference values. The numbers of colonies obtained with cells exposed to various doses of UV light were expressed as percentage of control.

RESULTS

Results are summarized in Fig 1A and B. After exposure to UVC light, marrow cells showed a rapid decrease in functional ability of marrow cells. PHA responses (shown are results with 0.2% PHA), CFU-GM and BFU-E decreased to 50% of control with UVC doses of 0.2 to 0.4 mJ/cm^2. These functions were reduced to <1% with ~0.5 mJ/cm^2 for BFU-E, 2 mJ/cm^2 for CFU-GM and 10 mJ/cm^2 for PHA stimulation.

With UVB light, longer exposure (ie, higher energy) was necessary for cell inactivation. Cell viability following UV exposure was ≥98%. The most UV-sensitive function was the response to PHA (shown are results with 0.2%), which declined to 50% after an exposure of 1 to 2 mJ/cm^2, and was reduced to <1% at 5 mJ/cm^2. These cells were also unable to generate cytotoxic effector cells against allogeneic targets (data not shown). BFU-E formation was reduced to 50% with an exposure of 3 mJ/cm^2 and to <1% with 15 mJ/cm^2. Most strikingly, CFU-GMs increased by 25% to 30% relative to control with UVB doses of 0.1 to 2 mJ/cm^2. More than 20 mJ/cm^2 was necessary to reduce the number of CFU-GM to <1%.

Thus, the amount of energy required overall for cell inactivation with UVB was about tenfold higher than with UVC. With UVC exposure, BFU-Es and CFU-GMs were as sensitive as or more sensitive than PHA responses. However, UVB exposure rapidly eliminated proliferative responses to PHA, whereas hematopoietic precursors (BFU-E, CFU-GM) maintained their colony-forming ability.

DISCUSSION

We showed in this study that UV light in the intermediate (UVB) but not short-wave (UVC) range can be used to selectively inactivate bone marrow T lymphocytes while preserving the function of hematopoietic precursors as determined by in vitro colony-forming assays. These data are consistent with work by other investigators showing that UVC treatment is mostly cytocidal, whereas UVB modulates cell function. The results indicate furthermore that different cell populations have differential sensitivity to UVB light. This could be due to a relative resistance to UV exposure, possibly mediated by differences in cell membrane or cytoplasmic components, or it could result from different repair processes in the cells; hematopoietic precursor cells but not T lymphocytes may be equipped to bypass UV-induced damage or repair it quickly.

We previously showed that among peripheral blood leukocytes T cells are more sensitive than B cells, and among T lymphocytes, CD8^+ cells are more sensitive than CD4^+ cells. UV sensitivity in peripheral blood leukocytes is manifested by an increase in intracellular calcium in unstimulated cells and an unresponsiveness of calcium mobilization to mitogenic stimuli. Similar studies in bone marrow cells may shed additional light on the mechanisms involved.
Our results are of interest in the context of murine data showing that UV treatment of lymphocytes added to hematopoietic precursor cells prevented graft-versus-host (GVH) reactivity. The present study indicates that human marrow can be directly exposed to UV light, thus inactivating T cells contained in the marrow inoculum used for transplantation. Ongoing investigations in an in vivo model show that murine spleen cells can be treated with UV so that lymphocyte reactivity is abrogated while ability for hematopoietic reconstitution is spared (J. Deeg et al, unpublished observations).

REFERENCES

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