In genetically different mice, termed UVB-resistant (UVB-R),[12,13] CH develops readily when hapten is painted on UVR-exposed skin. However, if the hapten is painted at cutaneous sites distant from the site of UVR and immediately after the last UVR exposure, CH develops normally in UVB-S mice, indicating that the primary immune defect created by UVR in this protocol is at the irradiated site.[14] Moreover, when sensitizing doses of the hapten are painted simultaneously on UVR-exposed and unexposed skin, shortly after the last dose of UVR, normal levels of CH are induced.[14] This result considerably strengthens the conclusion that acute, low-dose UVR impairs CH induction by a strictly local action. It has been proposed that the "strictly local" effects of acute, low-dose UVR on CH induction are mediated primarily by tumor necrosis factor (TNF)-α,[13,14] although other factors, such as *cis*-urocanic acid, reactive oxygen intermediates, and alpha-melanocyte stimulating hormone have also been implicated.

Although the evidence just summarized indicates that the acute, low-dose protocol of UVR has a strictly local effect on CH induction at the site, evidence exists that this same UVR protocol also perturbs the immune system systemically. If DNFB is painted directly on UVR-exposed skin immediately after the last exposure, a second application of the same hapten applied two weeks later to a non-irradiated site fails to generate CH.[2,14] This form of unresponsiveness, which has been called tolerance, is hapten-specific.[2] Recent experiments have revealed that tolerance also occurs when DNFB is painted (immediately or after a 3-day delay) on unirradiated skin at a distance from the UVR-exposed site.[14] Unlike the local effect of low dose UVR on CH induction, the ability of acute low-dose UVR to induce tolerance when hapten is painted on a distant skin site is not reversed by systemic administration of neutralizing anti-TNF-α antibodies (Ab).[14,15] Moreover, simultaneous application of hapten to UVR-exposed and unexposed skin generates intense CH as well as tolerance.[14] Based on these results, we have speculated that failed CH induction and tolerance induction are related, but separate and mechanistically distinct, immunologic consequences of acute, low-dose exposure to UVR.

In the recent past, Niizeki *et al.*[16] reported that these paradoxical results could be explained by testing the hypothesis that the deleterious effects of acute, low-dose UVR on cutaneous immunity are mediated primarily by TNF-α and interleukin (IL)-10, the former of which accounts for failed CH induction, whereas the latter accounts for tolerance induction. Niizeki *et al.* showed that anti-IL-10 Ab reverses tolerance induction after acute, low-dose of UVR exposures. Furthermore, anti-IL-10 Ab partially reverses tolerance induction by urocanic acid (UCA). Based on these findings, the following speculation was entertained for the pathogenesis of CH failure and tolerance after acute low dose UVR: UVR converts *trans*-UCA to *cis*-UCA; *cis*-UCA in turn induces keratinocytes (and perhaps other skin cells) to secrete IL-10; IL-10 then induces the formation of a tolerance-promoting signal within the irradiated skin. Yarosh *et al.*[17] have reported that *cis*-UCA is incapable of activating keratinocytes to secrete IL-10, yet our data indicate that anti-IL-10 Ab partially reverses *cis*-UCA-dependent tolerance. This has led us to suspect that cells other than keratinocytes are involved in the genesis of UVR-dependent tolerance (discuss below).

A Cutaneous Source of TNF-α after UVR: The Roles of Mast Cells in Impaired Cutaneous Immunity

We have previously demonstrated that neutralizing anti-TNF-α Ab can restore the capacity of UVB-treated skin to support the induction of CH in UVB-S mice.[13,14] Furthermore, in preliminary experiments conducted in our laboratory, we have determined that keratinocytes from normal murine skin that were exposed to UVR *in vitro* contained mRNA for TNF-α. However, we have been unable to distinguish a quantitative difference in TNF-α production between UVR-exposed keratinocytes prepared from UVB-S and UVB-R mice (Alard *et al.*, unpublished data). Thus, we have sought other intracutaneous sources of this cytokine that might help explain the UVB-R and UVB-S phenotypes. To address this issue, we tested the hypothesis that mast cells are the immediate intracutaneous source of TNF-α following UVR. First, using immunohistochemistry we were able to show that mast cells in the dermis released TNF-α following exposure to UVB.[18] Second, we loaded Fc receptors of mast cells of UVB-S (C3H/HeN), UVB-R (C3H/HeJ), and mast cell-deficient (Sl/Sl^d) mice with intradermal injections of anti-dinitrophenyl (DNP)-IgE Ab. Twenty-four hours later, DNP was injected intravenously, and within 30 minutes oxazolone was painted on injected skin sites. CH induction was impaired in UVB-S mice, but not in UVB-R or Sl/Sl^d mice, and treatment with anti-TNF-α Ab was able to reverse this impairment of CH.[18] Third, we have found that UVR did not impair CH induction when DNFB was painted on irradiated skin of mast cell-deficient mice, even though CH induction was impaired by UVR in wild-type littermates of the deficient mice.[18,19] Since UVR impairs CH induction through a TNF-α-dependent mechanism,[13] we concluded that UVR triggers the prompt release of TNF-α from dermal mast cells, and that mast cell-derived TNF-α interferes with generation of the hapten-specific signal required for CH induction. In addition, using isolated mast cells from bone marrow, we were able to