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Contacts between mast cells and dendritic cells in the human skin

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Summary

Langerhans cells are a dendritic cell type characteristic of the epidermis. Since mast cells secrete molecules potentially influent on dendritic cell differentiation, we have addressed the degree of proximity between these two cell types in biopsies of skin diseases characterized by massive influx of dendritic cell precursors. By fluorescence microscopy, avidin labeled mast cells were found in contact with CD1a+ dendritic cells. By electron microscopy, contacts between mast cells and blunty dendritic cells were found in areas corresponding to those where CD1a+ cells were localized by immunohistochemistry. We propose that mast cells can induce the differentiation of precursors into Langerhans cells through both the release of short range acting soluble factors and contact-mediating plasma membrane molecules.

Key words

Skin, Langerhans cells, Mast cells

Introduction

Langerhans cells (LC) play a crucial role in the defense against viral pathogens and cancer and the induction of contact hypersensitivity (Pieri et al., 2001; Merad et al., 2008). Dermal dendritic cells (DC), including perivascular dendritic macrophages, also take up antigens, mature and present these antigens to T cells and B cells. Blockade of dermal DC migration from skin to lymph node may prevent effective cytotoxic T cell activation (Gerlini et al., 2005; Zaba et al., 2009).

Mast cell (MC) secretory products play a role in regulating microcirculation and in the recruitment, differentiation and function of cells involved in inflammatory processes (Hofmann and Abraham, 2009) and wound healing (Bacci et al., 2009).

Mast cells and immature DC co-localize at antigen entry sites, i.e. skin and mucosa. Both human and mouse activated MC express high levels of chemokines that attract immature DC (Sallusto et al., 1998, Nakajima et al. 2002). Reciprocally, DC produce CCL5 and CCL8 (Sallusto et al., 1999) that can interact with CCR3 on MC (Ochi et al., 2000).

Mast cells can affect DC differentiation and function by different mechanisms. Histamine induces human monocyte-derived dendritic cells (MoDC) to transiently express CD86 (Caron et al., 2001b), to produce more IL-10 and less IL-12 and to differentiate into Th2-promoting DC (Caron et al., 2001a). Prostaglandin G2 reduces IL-12 production by MoDC and hence promotes Th2 responses (Gosset et al., 2003; Gosset et al., 2005). Thymic stromal lymphopoietin, which is also produced by MC

(Reche et al., 2001), promotes the maturation of CD11c+ circulating leukocytes to DC and induce them to secrete chemokines such as thymus and activation-regulated chemokine (TARC) and macrophage derived chemokine (MDC) (Soumelis et al., 2002). MC derived exosomes have been shown to induce DC maturation (Skokos et al., 2003). Contact in co-cultures is needed to allow activated MC to efficiently stimulate DC to produce IL-10 and Th2 cytokines (Kitawaki et al., 2006).

In this study, the possible occurrence of MC-DC contacts related to DC differentiation has been addressed in skin biopsies derived from immunomediated inflammatory skin diseases, where DC precursor are recruited and differentiate into LC and dermal DC (Breathnack et al., 1986; Mori et al., 1994; Rho et al., 2004).

Material and methods

Skin biopsies from 5 patients (3 men and 2 women, aged between 18 and 75 years, mean 46,6) affected by acute graft-versus-host-disease (aGVDH; 2 cases), atopic dermatitis (2 cases), and lupus erythematosus (1 case), were obtained for diagnostic purpose and used in part for this study, in the respect of Helsinki declaration and the Italian law, and in part for diagnostic purposes.

The specimens were snap frozen and cryosections were fixed in cold acetone. MC were stained by tetramethylrhodamine isothyocianate-labeled avidin (Immunotech, Marseille, France) (Tharp et al. 1985). This method is based on the same principle of basophilia (Ehrlich, 1878) but is much more sensitive and allows for MC staining in autoptical (Bacci et al., 2006) as well as bioptical specimens (Krishnaswamy and Chi, 2006). LC were stained with a monoclonal antibody against CD1a (Ortho, Milan, Italy), revealed with fluorescein isothiocyanate conjugated goat anti mouse serum (Sigma, Milan, Italy), The specificity of the immunostaining was tested by omitting the first antibody or substituting it with an irrelevant one.

Part of the biopsies were fixed in Karnovsky's solution in 0,1 mol/L osmicated and embedded in epoxy resin. Ultrathin sections were stained with uranyl acetate followed by bismuth tartrate or lead acetate and observed in Siemens Elmiskop II electron microscopes, at 80kV.

Results

At fluorescence microscopy, MC were found in contact with CD1a positive LC (see inset of Fig. 1).

By electron microscopy, contacts were seen in the dermis between MC and cells with blunty dendritic shape, containing smooth endoplasmic reticulum, primary but not secondary lysosomes and no Birbeck granules (Fig. 1). Contacts were sometimes on tip of MC microvilli, more often on wide, flat MC surfaces; specialized junctions were not detected between MC and DC.

Discussion

This study showed intercellular contacts between immature DC and MC in human skin, in conditions characterized by intense influx of DC precursor and their

final differentiation in the skin. The cells of dendritic lineage could be interpreted as elements of the Langerhans cell lineage by the abundance of smooth endoplasmic reticulum, the presence of primary but not secondary lysosomes and the localization in sites where immunofluorescence demonstrated the presence of CD1a positive cells; they were interpreted as immature elements because of the blunty dendritic shape and the absence of Birbeck granules.

Acute GVHD and lupus erythematosus, as well as atopic dermatitis, are characterized by intense recruitment and differentiation of LC precursors (Pimpinelli et al., 1993; Mori et al., 1994), which may explain why contacts between MC and immature LC are more easy to find in these conditions than in normal skin or other diseases. In this respect this study expands the results of Kitawaki et al (2006), who showed close proximity between DC and MC in atopic dermatitis but could not ascertain by their technique whether the two cell types were in contact with each other; these authors did not address the expression of CD1a by the DC close to MC.

Langerhans cells are considered to differentiate in the epidermis (for review: Romagnoli et al., 1991; Pieri et al., 2001), but can start this process already in the dermis. Since MC are a source of many cytokines and other signal molecules (Bacci et al., 2009), the close relationship between MC and DC allows to prospect that MC can deliver stimulating signals for the differentiation of precursors into LC through the release of short range acting soluble factors and through direct contact mediated by plasma membrane molecules. Therefore MC may influence adaptive T cell-mediated immune responses by modulating the differentiation of DC, and in particular of LC, in inflamed skin and perhaps in other tissues.

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References

Bacci S., Romagnoli P., Norelli G.A., Forestieri A.L., Bonelli A. (2006) Early increase in TNF-alpha containing mast cells in skin lesions. Int. J. Leg. Med. 120: 38-42.

Bacci S., Bonelli A., Romagnoli P. (2009) Mast cells in injury response. In: Abreu T., Silva G.: Cell movement: New Research Trends; Nova Science Publishers, Hauppage, NY. Pp. 81-121.

Breathnach S.M., Shimada S., Kovac Z., Katz S.I. (1986) Immunologic aspects of acute cutaneous graft versus host disease: decreased density and antigen presenting function of Ia+ Langerhans cells and absent-presenting capacity of Ia+ keratinocytes. J. Invest. Dermatol. 86: 226-234.

Caron G., Delneste Y., Roelandts E., Duez C., Bonnefoy J.Y., Pestel J., Jeannin P. (2001a) Histamine polarizes human dendritic cells into Th2 cell-promoting effector dendritic cells. J. Immunol. 167: 3682-3686.

Caron G., Delneste Y., Roelandts E., Duez C., Herbault N., Magistrelli G., Bonnefoy J.Y., Pestel J., Jeannin P. (2001b) Histamine induces CD86 expression and chemokine production by human immature dendritic cells. J. Immunol. 166: 6000-6006.

Ehrlich P. (1878) Beiträge zur theorie und praxis der Histologischen Färbung. Thesis, Leipzig.

Gerlini G., Romagnoli P., Pimpinelli N. (2005) Skin cancer and immunosuppression. Crit. Rev. Oncol. Hematol. 56: 127-136.

Gosset P., Bureau F., Angeli V., Pichavant M., Faveeuw C., Tonnel A.B., Trottein F. (2003) Prostaglandin D2 affects the maturation of human monocyte-derived dendritic cells: consequence on the polarization of naive Th cells. J. Immunol. 170: 4943-4952.

Gosset P., Pichavant M., Faveeuw C., Bureau F., Tonnel A.B., Trottein F. (2005) Prostaglandin D2 affects the differentiation and functions of human dendritic cells: impact on the T cell response. Eur. J. Immunol. 35: 1491-1500.

Hofmann M.A., Abraham N.S. (2009) New roles for mastcells in modulating allergic reactions and immunity against pathogens. Curr. Opin. Immunol. 21: 679–686.

Kitawaki T., Kadowaki N., Sugimoto N., Kambe N., Hori T., Miyachi Y., Nakahata T., Uchiyama T. (2006) IgE activated mast cells in combination with pro-inflammatory factors induce Th2-promoting dendritic cells. Int. Immunol. 18: 1789-1799.

Krishnaswamy G., Chi D.S. (2006) Mast cells: methods and protocols. Humana Press, Totowa.

Merad M., Ginhoux F., Collin M. (2008) Origin, homeostasis and function of Langerhans cells and other langerin-expressing dendritic cells. Nat. Rev. Immunol. 8: 935-947.

Mori M., Pimpinelli N., Romagnoli P., Bernacchi E., Fabbri P., Giannotti B. (1994) Dendritic cells in cutaneous lupus erythematosus: a clue to the pathogenesis of lesions. Histopathology 24: 311-321.

Nakajima T., Inagaki N., Tanaka H., Tanaka A., Yoshikawa M., Tamari M., Hasegawa K., Matsumoto K., Tachimoto H., Ebisawa M., Tsujimoto, G., Matsuda, H., Nagai, H., Saito, H. (2002) Marked increase in CC chemokine gene expression in both human and mouse mast cell transcriptomes following Fcepsilon receptor I crosslinking: an interspecies comparison. Blood 100: 3861-3868.

Ochi H., De Jesus N.H., Hsieh F.H., Austen K.F., Boyce J.A. (2000) IL-4 and IL-5 prime human mast cells for different profiles of IgE-dependent cytokine production. Proc. Natl. Acad. Sci. USA 97: 10509-10513.

Pimpinelli N., Romagnoli P., Bosi A., Santucci M., Mori M., Guidi S., Giannotti B. (1993) Dendritic cells in the skin after allogeneic bone marrow transplantation: immunohistochemical and electron microscopic monitoring. Eur. J. Dermatol. 3: 310-317.

Pieri L., Domenici L., Romagnoli P. (2001) Langerhans cells differentiation: a three-act play. It. J. Anat. Embryol. 106: 47-69.

Reche P.A., Soumelis V., Gorman D.M., Clifford T., Liu M.R., Travis M., Zurawski S.M., Johnston J., Liu Y.J., Spits H., de Waal Malefyt R., Kastelein R.A., Bazan J.F. (2001) Human thymic stromal lymphopoietin preferentially stimulates myeloid cells. J. Immunol. 167: 336-343.

Rho N.K., Kim W.S., Lee D.Y., Lee J.H., Lee E.S., Yang, J.M. (2004) Immunophenotyping of inflammatory cells in lesional skin of the extrinsic and intrinsic types of atopic dermatitis. Br. J. Dermatol. 151: 119-125.

Romagnoli P., Pimpinelli N., Lotti T. (1991) Dendritic cells in the skin and oral mucosa. It. Gen. Rev. Dermatol. 24: 49-79.

Sallusto F., Schaerli P., Loetscher P., Schaniel C., Lenig D., Mackay C.R., Qin S., Lanzavecchia A. (1998) Rapid and coordinated switch in chemokine receptor expression during dendritic cell maturation. Eur. J. Immunol. 28: 2760-2769.

Sallusto F., Palermo B., Lenig D., Miettinen M., Matikainen S., Julkunen I., Forster R., Burgstahler R., Lipp M., Lanzavecchia A. (1999) Distinct patterns and kinetics of chemokine production regulate dendritic cell function. Eur. J. Immunol. 29: 1617-1625.

Skokos D., Botros H.G., Demeure C., Morin J., Peronet R., Birkenmeier G., Boudaly S., Mécheri S. (2003) Mast cell derived exosomes induce phenotypic and functional maturation of dendritic cells and elicit specific immune responses in vivo. J. Immunol. 170: 3037-3045.

Soumelis V., Reche P.A., Kanzler H., Yuan W., Edward G., Homey B., Gilliet M., Ho S., Antonenko S., Lauerma A., Smith K., Gorman D., Zurawski S., Abrams J., Menon S., McClanahan T., de Waal-Malefyt R.D., Bazan F., Kastelein R.A., Liu Y.J. (2002) Human epithelial cells trigger dendritic cell mediated allergic inflammation by producing TSLP. Nat. Immunol. 3: 673-680.

Tharp M.D.; Seelig L.L:, Tigelaar R.E:, Bergstresser P.R. (1985) Conjugated avidin binds to mast cell granules. J. Histochem. Cytochem. 33: 27-32.

Zaba L.C.; Krueger J.G., Lowes M.A. (2009) Resident and “inflammatory” dendritic cells in human skin. J. Invest. Dermatol. 129: 302-308.

Figures

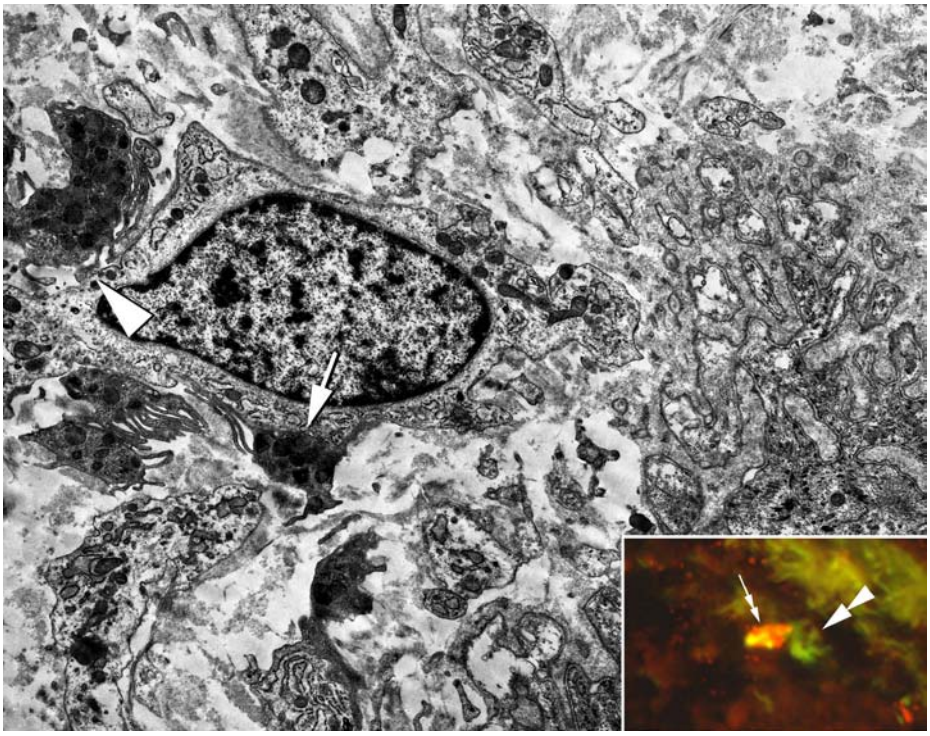

Figure 1 - An immature dendritic cell in contact with a mast cell in the dermis of a patient with aGVHD. One contact is through a wide, flat surface (arrow), another involves the tip of mast cell microvilli (arrowhead). Electron microscopy x 8000. Inset: Intercellular contacts between a dendritic cell (green labeled: double arrowhead) and a mast cell (red labeled: double arrow) in the dermis of a patient with atopic dermatitis. Fluorescence microscopy x 400.