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Dendritic Cell Apoptosis: Regulation of Tolerance versus Immunity

Rahul Kushwah and Jim Hu

Dendritic cell (DC) apoptosis is an important event that regulates the balance between tolerance and immunity through multiple pathways, and defects in DC apoptosis can trigger autoimmunity. DC apoptosis is also associated with immunosuppression and has been observed under several pathologies and infections. Recent studies indicate that apoptotic DCs can also play an active role in induction of tolerance. This review discusses the regulatory pathways of DC apoptosis, stimuli inducing DC apoptosis, and the implications of DC apoptosis in the induction of immunosuppression and/or tolerance. The Journal of Immunology, 2010, 185: 795–802.

Dendritic cells (DCs) are professional APCs that are critical for induction of adaptive immunity and tolerance (1). The subtypes of DCs under steady state conditions include conventional DCs (cDCs) and plasmacytoid DCs (pDCs). pDCs live longer than cDCs and are characterized by massive production of type I IFNs in response to viral infections. Both cDCs and pDCs can be found in the thymus, spleen, lymph node, and peyer’s patch, with only cDCs observed in the skin and liver (2). DCs are commonly classified as type I IFNs producing pDCs or myeloid DCs (mDCs). The DC life cycle starts with differentiation of progenitor cells into DCs to induction of apoptosis after Ag presentation. Myeloid progenitors in the bone marrow give rise to macrophage-DC progenitors, which further differentiate into monocytes and common DC progenitors (3). Common DC progenitors give rise to cDCs and pDCs, and monocytes can also differentiate into inflammatory DCs during inflammation. Moreover, lymphoid progenitors can also give rise to all subsets of splenic and thymic DC subsets (4).

DC apoptosis regulates the magnitude of immune response by limiting the Ag availability to T cells and is regulated both by extrinsic and T cell-mediated signals. Environments with significant DC apoptosis are immunosuppressive, promote regulatory T cell (Treg) generation, and display functional impairment of remaining DCs, indicating that DC apoptosis might contribute to tolerance. The importance of DC apoptosis is further highlighted by studies identifying defects in DC apoptosis as triggers of autoimmune diseases (5–8). It is only recently that a few studies have investigated the effects of DC apoptosis on the immune response and have identified that apoptotic DCs can be taken up by viable DCs and result in tolerance induction via generation of Ag-specific Tregs (9, 10).

Regulation of DC apoptosis

The DC life span is fairly limited; under steady state conditions, DCs have a rapid rate of turnover, with $t_{1/2}$ ranging between 1.5 and 2.9 d (11). Upon activation, DCs regulate genes that allow for induction of maximal immune response, with a consequence of initiating cell death. LPS-induced DC maturation initiates DC apoptosis through CD14 mediated NFAT activation (12). Although specific genes that regulate DC lifespan have not been completely identified, studies have shown involvement of multiple pathways that regulate DC apoptosis (Fig. 1). The similarities and/or differences in apoptosis of different DC subsets and their susceptibility to apoptosis inducing stimuli remain to be elucidated.

Apoptosis can be regulated by extrinsic and intrinsic pathways. The extrinsic pathway involves binding of a death-inducing ligand to a receptor, resulting in formation of the death-inducing signaling complex, which cleaves and activates caspases 8 or 10 (13). The intrinsic apoptosis pathway involves signals within the cell, inducing permeabilization of the mitochondrial outer membrane and activating caspases for apoptosis (14). Anti-apoptotic members of the Bcl-2 superfamily normally suppress mitochondrial membrane permeabilization by inhibiting proapoptotic proteins Bax and Bak, which induce mitochondrial membrane depolarization. Bcl-2 is normally highly expressed in immature DCs, but is downregulated in mature DCs (15).

TNF superfamily and DC apoptosis. CD40, a member of the TNF receptor superfamily, is a costimulatory molecule required for DC activation, which interacts with CD154 (CD40L) expressed on activated T cells. CD40–CD154 interaction induces anti-apoptotic signaling in DCs via Akt1 activation (16). TNF-related activation-induced cytokine (TRANCE) is another
member of the TNF superfamily, expressed by activated T cells, which binds to receptor activator of NFκB (RANK) on DCs, resulting in activation of NFκB and JNK pathways and Bcl-xl upregulation, initiating antiapoptotic signaling (17). Another receptor to which TRANCE binds is osteoprotegerin (OPG), which functions as a decoy receptor. OPG−/− DCs have better survival than wild type DCs, which is likely due to the absence of OPG in limiting TRANCE–RANK interactions (18).

DC maturation also results in upregulation of CD95 (Fas), a member of the TNF receptor superfamily and an inducer of apoptotic signaling in immature DCs. However, ligation of CD95 on mature DC surface with its ligand CD95L (FasL) results in upregulation of c-FLIPL, which inhibits apoptosis (19).

TNF-related apoptosis-inducing ligand (TRAIL) is a transmembrane protein expressed on effector cells and induces apoptosis of leukemic pDCs upon binding to DR4 and DR5 receptors, which are not expressed on normal DCs (20). TRAIL-dependent apoptosis involves the extrinsic pathway with caspase 8 and 10 acting as initiator caspases.

**Nur77 family and DC apoptosis.** The Nur77 family is a group of zinc finger transcription factors belonging to the steroid nuclear receptor superfamily, consisting of three receptors: Nur77, Nur1, and MINOR. Nur77 and MINOR have been implicated in the regulation of T and B cell apoptosis (21). MINOR is upregulated in DCs upon activation; forced expression of MINOR leads to induction of apoptosis and its suppression results in the inhibition of DC apoptosis (22).

**Other factors and DC apoptosis.** Chemokine receptor CCR7 is upregulated on the DC surface upon maturation, and binding of its ligands CCL19 and CCL21 results in activation of the Akt1 pathway, which inhibits proapoptotic GSK3β and FOXO1 and also promotes translocation of prosurvival NFκB to the nucleus (23). The consequence of this signaling cascade is inhibition of DC apoptosis. Leptin, an adipocyte-derived hormone, also promotes DC survival by inducing Akt1 and NFκB activation.
along with upregulation of Bcl-2 and Bcl-xl gene expression (26). Type I IFNs have also been shown to regulate survival of mDCs by promoting DC apoptosis through downregulation of Bcl-2 and Bcl-xl expression (27). It remains to be identified whether the proapoptotic role of type I IFNs are dependent on its activity in promoting DC maturation. Amyloid peptides have also been shown to induce DC apoptosis, via activation of acid sphingomyelinase resulting in the production of ceramide, which results in autocatalysis of caspase 8 resulting in activation of apoptotic signaling (28).

**Extrinsic triggers of DC apoptosis**

**DC apoptosis triggered by infections.** Viruses, parasites, and bacteria are known to induce DC apoptosis, which results in a bystander effect of induced immunosuppression. Measles virus (MV) infection is usually associated with secondary infections resulting from immunosuppression. MV infects resting and mature DCs, and it induces human mDC apoptosis via the Fas pathway and likely also via the TRAIL pathway (29). MV-pulsed human mDC–T cell cultures show enhanced rates of DC apoptosis along with an impairment of remaining viable DCs to induce T cell proliferation and undergo maturation (30). Other viruses, such as foot and mouth disease virus, have also been shown to induce apoptosis of murine immature mDCs (31). *Brugia malayi*, a nematode and causative agent of lymphatic filariasis (elephantiasis), promotes T cell hyporesponsiveness and induces human mDC apoptosis through TRAIL pathway, raising the possibility that T cell hyporesponsiveness may be a consequence of DC apoptosis (32). Gram-positive bacteria can induce DC apoptosis via bacteria-encoded virulence factors, and many Gram-negative bacteria can induce DC apoptosis by caspase 3 or caspase 8 activation (33, 34). Studies need to be conducted to address whether there is impairment of DC function upon bacterial infections known to induce DC apoptosis.

**DC apoptosis during pathologic conditions.** DC apoptosis has been observed in several pathologies, such as breast cancer, sepsis, and trauma, which parallel induction of immunosuppression. Patients with advanced breast cancer have defective cellular immunity in mounting immune response to different pathogens and have high levels of apoptotic DCs (35). Studies have identified that DCs from breast cancer patients are defective in inducing T cell proliferation and in undergoing maturation in response to an inflammatory stimuli (36).

One of the hallmarks of septic syndrome is induced immunosuppression, which is responsible for the mortality. Sepsis-induced immunosuppression is associated with rapid and extensive caspase 3-mediated apoptosis of >50% of DCs, observed both in humans and mouse models of sepsis (37). Concomitant to DC apoptosis, there is an increase in the levels of circulating Tregs and an increase in their suppressive function (38). However, the mechanism of the increase in Tregs is not well understood, although in mice, postseptic splenic DCs are good inducers of Foxp3+ Tregs in vitro (39). Studies have shown that suppressing DC apoptosis in mice results in resistance to endotoxin-induced sepsis and immunosuppression, highlighting the role of DC apoptosis in mediating septic pathology (40).

Severe acute trauma is often followed by complications involving organ dysfunction and septicemia. DCs isolated from patients with multiple trauma show reduced expression of antiapoptotic genes and higher expression of proapoptotic genes. Three to 5 d after trauma, there is significant depletion of mDCs with an approximately 3-fold decrease in the ratio of mDCs to pDCs and a parallel increase in the plasma levels of IL-10 (41, 42). An elevated mDC-to-pDC ratio is associated with an exacerbated immune response and is a prognostic marker for acute cellular rejection in pediatric small bowel transplantation and pediatric liver rejection (43). A several-fold decrease in myeloid-to-plasmacytoid ratio in trauma patients is likely an indicator of attenuated immune response, probably owing to increased mDC apoptosis. Studies have also shown that in posttraumatic hemorrhage in mice there is an increase in apoptosis of splenic DCs (mDCs and pDCs), and surviving DCs show impaired cytokine production and have a significant reduction in their Ag-presentation ability and MHC class II expression (44).

**Glucocorticoid-induced DC apoptosis.** Studies indicate that after liver, heart, and kidney transplantation, there is a strong decline in the circulating numbers of DCs. As the dose of glucocorticoids is tapered, the DC numbers gradually recover. Patients and healthy volunteers treated with corticosteroids show a decrease in the levels of circulating pDCs (45). It has been shown that glucocorticoid treatment of human pDCs can induce apoptosis via the extrinsic pathway (46).

**Tumor-induced DC apoptosis.** Tumors secrete factors that impair the function of both circulating and tumor-infiltrating DCs by inducing their apoptosis (47). Lymphatic drainage from tumors in multiple cancers occurs in the sentinel lymph node, where massive reduction of DC numbers is observed compared with other lymph nodes (48). In cancers, such as hepatocellular carcinoma, patients show a functional impairment of DCs and high levels of mDC-apoptosis–inducing α-fetoprotein, indicating that DC apoptosis can somehow regulate functional impairment of remaining viable DCs (49). Human melanoma tumors have also been shown to secrete factors, such as gangliosides, that induce mDC apoptosis (50).

**UV-induced DC apoptosis.** Langerhans cells, which are mDCs found in the skin, form an extensive network in the epidermis to capture Ags and subsequently potentiate adaptive immune reactions. However, UV-induced apoptosis of these Langerhans cells has not been studied in detail.

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**Table 1. Effects of apoptotic cell uptake on DCs and macrophages**

<table>
<thead>
<tr>
<th>Live Cells</th>
<th>Apoptotic Cells</th>
<th>Uptake</th>
<th>Suppression of Live Cell Activation</th>
<th>Phosphatidylserine-Dependent Suppression</th>
<th>TGF-β1 Production</th>
<th>Treg Induction</th>
<th>References</th>
</tr>
</thead>
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<td>Splenocytes</td>
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<td>Yes</td>
<td>Yes/No</td>
<td>No</td>
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<td>DCs</td>
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<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>9, 10</td>
</tr>
<tr>
<td>DCs</td>
<td>Macrophages</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>61</td>
</tr>
<tr>
<td>Macrophages</td>
<td>Splenocytes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>68</td>
</tr>
</tbody>
</table>

This table summarizes the current knowledge on uptake of various apoptotic cells (splenocytes, DCs, or macrophages) by DCs or macrophages. Cells that uptake apoptotic cells are indicated as live, and the effects of apoptotic cell uptake on live cells are classified as suppression of live cell activation (macrophage uptake/DC maturation), along with whether it is dependent on phosphatidylserine, and whether the uptake results in secretion of TGF-β1 and induction of Tregs.
FIGURE 2. Proposed model for tolerance induction by apoptotic DCs. A and B, Exposure to UV radiation induces apoptosis of both lymphocytes and DCs, leading to exposure of phosphatidylserine on the cell surface of both DCs and lymphocytes along with another protein specifically on apoptotic DCs that could mediate preferential tolerance induction by apoptotic DCs. In addition, there could also be an exposure of a trigger for αvβ8 integrins. C, Scavenger receptors expressed on the DC surface play a role in the uptake of apoptotic cells, and interaction of phosphatidylserine on apoptotic cells with its receptor on DCs likely leads to prevention of subsequent maturation in response to inflammatory stimuli, such as LPS, with no evidence for secretion of TGF-β1. The suppression of NFκB pathway could be mediated by signaling through the phosphatidylserine receptor, the role of which is controversial in DCs. D, In contrast to apoptotic lymphocyte uptake, apoptotic DC uptake by viable DCs results in secretion of TGF-β1, suppression of maturation, and induction of Ag-specific Foxp3+ Tregs. Secretion of TGF-β1 is dependent on the mTOR pathway, although the signal that results in mTOR activation upon apoptotic cell uptake is not known. mTOR
responses. Langerhans cells isolated from UV-treated mice fail to prime an immune response upon transfer to naive mice, but induce tolerance via Treg generation. UV-induced immunosuppression is absent in transgenic mice that overexpress antiapoptotic molecules (51). UV radiation induces DC apoptosis by activation of caspases 3, 8, and 9, loss of mitochondrial membrane potential, and cellular and nuclear degradation. Immature DCs (both human and mouse) are highly susceptible to UVB radiation, whereas mature DCs upregulate c-FLIP and Bcl-2 and are less susceptible to UV-induced apoptosis (52). Prevention of apoptosis of Langerhans cells blocks UV-induced immunosuppression, suggesting that UV-induced DC apoptosis makes viable DCs tolerogenic; however, the mechanism remains unclear (53).

UV radiation is used in extracorporeal photophoresis (ECP) immunosuppression therapy, in which PBMCs are treated with photoactive 8-methoxypsoralen and then exposed to UV light, which activates 8-methoxypsoralen to covalently bind to DNA and induce apoptosis. In addition to lymphocyte apoptosis, ECP also results in modulation of APC function and induction of human DC apoptosis, which plays a key role in ECP-induced immunosuppression (54). Immature human DCs have a higher susceptibility to ECP-induced apoptosis, with up to 50% of DCs undergoing apoptosis within 24 h of treatment and >90% undergoing apoptosis within 72 h of ECP treatment (55). Thereafter, the remaining viable DCs become tolerogenic with a severely diminished ability to undergo maturation in response to LPS, with a defect in inducing T cell proliferation (56). Concomitantly, there is also an increase in the levels of Tregs (57). These findings suggest a role for UV-induced DC apoptosis in mediating ECP-induced immunosuppression by affecting the function of the remaining viable DCs.

**DC apoptosis by T cells.** NK T cells are a distinct population of T cells, stimulated by α-glycosylceramides and α-glucosylsylceramide in a CD1d and TCR-dependent manner. Among the human subpopulations of Vo24NKT cells, there are CD4+CD8–Vo24NKT and CD4+CD8–Vo24NKT cells, with both having cytotoxic activity against mDCs from normal donors (58). Activated CD4+Vo24NKT cells upregulate CD40L and induce DC apoptosis via CD40/CD40L signaling through the extrinsic pathway. Induction of DC apoptosis could be one of the mechanisms used by NK T cells to prevent autoimmunity.

In tumor lymph nodes, Foxp3+ Tregs can directly interact with tumor Ag-bearing DCs and induce their apoptosis through perforin-dependent pathways (59). CD8+ T cells can also play a role in inducing DC apoptosis. Ag-loaded mDCs rapidly undergo depletion in mice with an ongoing immune response, and the rate of DC depletion is accelerated in transgenic animals with increased levels of CD8+ T cells (60).

**Apoptosis and cross-priming by DCs**

DCs are able to take up apoptotic cells and present Ags derived from apoptotic bodies to naive CD8+ T cells. Studies have demonstrated that DCs can take up apoptotic macrophages infected by influenza or *Salmonella* and cross-present Ags to CD8+ T cells to induce a potent CTL response against the virus proteins (61). CD8α+TCRβ+ T cells, which play a role in controlling experimental autoimmune encephalitis by killing pathogenic CD4+ T cells and also in controlling colitis disease, are activated by DCs that capture apoptotic CD4+ T cells and cross-present self-peptides via MHC class I to the CD8α+TCRβ+ T cells (62). DCs express scavenger receptors, such as αvβ5 integrin, CD36, and Lox-1, making DCs highly efficient at taking up apoptotic cells and cross-presenting apoptotic cell-derived peptides to CD8+ T cells (63, 64).

DCs undergo apoptosis rapidly upon interaction with Ag-specific T cells. However, in vivo Ag presentation persists beyond 7 d, which is the peak time point for experimental CTL response (65). Because DCs have a limited life span, apoptotic DCs likely interact with viable DCs, leading to cross-presentation. However, the effects of this interaction are not well understood. The role of DC apoptosis in cross-presentation by DCs in mediating immunological tolerance warrants further investigation.

**Defects in DC apoptosis trigger autoimmune disease**

Mice with genetic defects in FasL and Fas develop autoimmune diseases, highlighting the importance of apoptosis in the maintenance of immunological tolerance (7). However, in mice with selective ablation of Fas in T and/or B cells, no lymphoproliferative disease is observed (66). Furthermore, selective expression of apoptosis inhibitory enzymes, such as cytokine response modifier A, a serpin-like protease inhibitor encoded by cowpox virus, in T cells failed to induce autoimmunity in mice (67). These findings suggest that apoptosis defects in non-lymphoid cells result in induction of autoimmunity.

Transgenic mice expressing baculovirus p35, an inhibitor of apoptosis that functions by inhibiting caspase 8, in DCs, displayed DC-specific apoptosis defects and hyper activation of T and B cells (5). Upon adoptive transfer, transgenic DCs were able to induce autoantibody formation, indicating a direct role of DC apoptosis in the maintenance of peripheral tolerance.

Bim is a proapoptotic BH3-only protein in the Bcl-2 family. Bim–/– mice develop autoimmunity (8), which is mainly due to defects in the negative selection of T and B lymphocytes. However, studies indicate that Bim–/– DCs are defective in apoptosis and are highly potent in induction of T cell activation and autoantibody formation, indicating that defects in DC apoptosis could be responsible for the autoimmunity observed in Bim–/– mice (6).

**Tolerance induction by apoptotic DCs**

Kushwah et al. (9, 10) explored the effects of UV-induced apoptotic DCs on viable DCs both in vitro and in vivo. In vitro studies indicate that apoptotic DCs are rapidly taken up by immature DCs, which prevents subsequent maturation of activation could be mediated by a protein specifically present on apoptotic DCs, which could also result in suppression of DC maturation via suppression of NFκB activation. Phosphatidylserine signaling may also mediate suppression of DC maturation, though its role in suppressing DC maturation is highly controversial. Furthermore, activation of inactive TGF-β1 to the active form could be mediated through αvβ8 signaling triggered by an yet unidentified protein present specifically on the surface of apoptotic DCs, although the mechanism of how apoptotic DCs could trigger this signaling is not known. Presentation of exogenous Ags via MHC II pathway to naive CD4+ T cells along with secretion of TGF-β1 drives differentiation of naive CD4+ T cells into Ag specific Foxp3+ Tregs.
immature DCs in response to LPS (10). In contrast to uptake of apoptotic DCs, necrotic DC uptake by viable DCs had no effect on the ability of viable DCs to undergo maturation and was recognized as an immunologically null event. Uptake of apoptotic DC induces immature DCs to secrete TGF-β1, which induces differentiation of naïve T cells into Foxp3+ regulatory T cells. At the same time, the ability of DCs that take up apoptotic DCs to induce Th17 differentiation is also reduced, likely because of the resistance of these tolerogenic DCs to undergo maturation, resulting in reduced levels of IL-6 production. Previous studies have shown that as cells undergo apoptosis, phosphatidylserine, an anionic aminophospholipid exposed to cell surface, plays an important role in recognition and clearance of apoptotic cells by macrophages and also results in induction of TGF-β1 secretion (68). Although it is tempting to speculate that the inhibitory effects of apoptotic DCs on viable DCs is phosphatidylserine dependent, previous studies have indicated that exposure of DCs to apoptotic non-DCs does not induce TGF-β1 secretion (69). In addition, exposure of DCs to phosphatidylserine-containing liposomes does not inhibit DC maturation or the ability to induce T cell activation, although it does inhibit macrophage activation (70). These studies indicate that there could be differences between macrophages and DCs in processing apoptotic cells, with macrophages relying on phosphatidylserine-dependent signaling for apoptotic cell-induced suppression. The phosphatidylserine-independent mechanism could regulate uptake of apoptotic cells, such as apoptotic splenocytes and apoptotic DCs, by viable DCs. It is plausible that uptake of apoptotic DCs by viable DCs could result in upregulation of programmed death ligand-1 and programmed death ligand-2, which are known to interact with the programmed death 1 receptor found on the surface of activated T and B cells, resulting in the attenuation of T cell function including IFN-γ production, proliferation, and increased T cell apoptosis (71, 72). IFN-γ can also play an immunoregulatory role via induction of IDO, an enzyme for tryptophan catabolism also involved in the induction of T cell anergy and apoptosis of activated T cells (73). Studies have shown that exposure of DCs to apoptotic T cells can result in IFN-γ mediated IDO induction, which can suppress T cell proliferation (74). However, uptake of apoptotic DCs by viable DCs does not result in IDO induction, indicating that IDO is likely not responsible for the observed immunosuppression (10). However, the secretion of TGF-β1 by immature DCs upon uptake of apoptotic DCs appears to occur through a highly specific mechanism dependent on apoptotic DC uptake, because uptake of apoptotic splenocytes failed to induce TGF-β1 secretion by DCs. Further study is required to identify the receptors for uptake of apoptotic DCs. It is possible that as DC are undergoing apoptosis, certain molecules are upregulated on their surface that can possibly interact with viable DCs. These molecules could be highly specific to apoptotic DCs and not present on other cell types, which could account for preferential TGF-β1 release upon uptake of apoptotic DCs by viable DCs. It appears that TGF-β1 secretion upon uptake of apoptotic DCs by viable DCs depends on the mTOR pathway, which could enhance rates of TGF-β1 mRNA translation (10). Table 1 provides an overview of our current knowledge on the uptake of different apoptotic cells by DCs and macrophages.

TGF-β1 activation is dependent on αvβ8 integrins, which are largely expressed on DCs, and DCs lacking this particular integrin fail to induce Tregs (75). Studies are needed to identify whether αv integrins play a role in the uptake of apoptotic DCs by viable DCs and whether uptake of apoptotic DC acts as a trigger for αvβ8 integrins to activate TGF-β1.

Delivery of apoptotic DCs to mice via i.v. injection resulted in uptake mostly by immature DCs in the lymph nodes and the spleen and promoted induction of tolerance (9). The authors showed that delivery of apoptotic DCs followed by delivery of Ag resulted in induction of Ag-specific Foxp3+ Tregs. Overall, the findings establish a model whereby selective uptake of apoptotic DC induces immunologic tolerance via suppression of DC function and induction of Tregs (Fig. 2). These findings could explain how, in immunosuppressive environments with significant DC apoptosis, such as sepsis, severe trauma, or breast cancer, and even during certain infections, the remaining viable DCs are poorly responsive to antigenic stimulation. Furthermore, these findings could also indicate yet another mechanism of how DC apoptosis can maintain immunologic homeostasis—by maintaining peripheral tolerance.

The maturation status of DCs could also play a role in immunity versus tolerance upon apoptosis. Presumably, in a scenario in which there is a lot of apoptosis of immature DCs but the surrounding viable DCs are in a mature state, the result of the uptake could possibly be initiation of an immune response rather than tolerance. It is plausible that under such situations, apoptotic DCs have the signaling molecules on their surface to signal viable DCs to become tolerogenic. The maturation-induced signaling in mature DCs could counteract the tolerogenic effects of apoptotic DC interaction and instead result in priming of an immune response rather than tolerance induction.

Modulation of DC apoptosis and therapeutic implications

Apoptotic DCs are likely therapeutic agents for regulating the balance between tolerance and immunity. DC-based immunotherapy has been extensively explored for cancer, and clinical trials have reported limited success (76). It has been reported that 5–6% of injected DCs during DC therapy migrate to lymph nodes, with a questionable fate of the remaining 95% DCs (77, 78). There is a likelihood that a major proportion of DCs upon injection undergo apoptosis and that viable DCs in the immediate vicinity uptake these apoptotic DCs and promote tolerance, which could limit induction of protective antitumor immune response during DC immunotherapy (Fig. 2). Therefore, delivery of apoptosis-resistant DCs could limit tolerance induction by limiting DC apoptosis; it could also improve benefits of DC immunotherapy. Studies have shown that expression of siRNA targeting proapoptotic molecules in DCs presenting tumor Ags increases DC survival and potentiates a heightened antitumor CD8+ cytotoxic T cell response (79). In addition, treatment of DCs with TRANCE, a DC prosurvival factor, has been shown to result in better adjuvant properties upon delivery of DCs (78). Other strategies to prevent apoptotic DC-induced tolerance could involve triggering of antiapoptotic CD40 signaling on DCs, treatment of DCs with CCL19/CCL21 or leptin to prevent apoptosis, and using siRNA to suppress MINOR or other proapoptotic molecules, which would also result in suppression of DC apoptosis (25, 26, 80). These strategies can result in improvement of efficacy of DC-mediated immunotherapy by reducing the likelihood of apoptotic DC-induced tolerance.
In autoimmune disease and allergy, there is a need to induce tolerance to suppress ongoing immune response, and the efficacy of apoptotic DCs could be exploited in allergen-specific immunotherapy. Patient-derived DCs can be treated with UV radiation to undergo apoptosis and can subsequently be injected following delivery of the allergen to induce tolerance. Glucocorticoid-treated DCs have shown efficacy at inducing transplantation tolerance in animal models (81). Glucocorticoid treatment of DCs can result in DC apoptosis; therefore, the DCs delivered are a combination of apoptotic DCs with viable DCs, which can promote induction of transplant tolerance. Immature DCs have also shown efficacy at inducing tolerance (82). However, immature DCs have no effect on other DCs in vivo and can also undergo maturation. Delivery of apoptotic DCs can prevent maturation of DCs in vivo and can also suppress pre-existing inflammation (9, 10). Therefore, apoptotic DCs may likely be explored as therapeutic agents for induction of transplant tolerance.

Conclusions
DCs have a high rate of turnover, and their apoptosis is regulated by many different pathways. DC apoptosis is observed in several pathologies paralleled with an induced immunosuppression. Recent studies indicate that apoptotic DCs can be taken up by viable DCs, which can then induce tolerance. However, we believe that this field is still in a stage of infancy and further study is required to understand whether DC apoptosis is an active mechanism used by the immune system to maintain tolerance against self-Ags through generation or maintenance of the Treg population.

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References

44. Kawasaki, T., S. Fujimi, J. A. Lederer, W. J. Hubbard, M. A. Choudhry,

42. Maier, M., E. V. Geiger, D. Henrich, R. Ebrahimi, S. Wutzler, M. Lehnert, and

59. Boissonnas, A., A. Scholer-Dahirel, V. Simon-Blancal, L. Pace, F. Valet, A. Kissenpfennig,

58. Nieda, M., A. Kikuchi, A. nicol, Y. Koezuka, Y. Ando, S. Ishihara, N. Lapteva,

802 BRIEF REVIEWS: DC APOPTOSIS AND IMMUNOLOGIC HOMEOSTASIS


57. Xia, C. Q., K. A. Campbell, and M. J. Clare-Salzler. 2009. Extracorporeal pho-

266–278.

antitumor immunity.

T cell-dependent elimination of dendritic cells in vivo limits the induction of


2009. Apoptosis differs in dendritic cell subsets early after severe trauma.


Dendritic cells use endocytic pathway for cross-priming class IIb MHC-restricted

CD8alphaalpha+TCRalphabeta+ T cells with regulatory properties. J. Immunol.

182: 6959–6968.

63. Parlati, S., G. Romagnoli, F. Spadaro, I. Canini, P. Sibarella, P. Bonghi, C. Ramoni,

I. Filesi, S. Biocca, L. Gabriele, and F. Belardelli. 2009. LOX-1 as natural IFN-[alpha]-

mediated signal for apoptotic cell uptake and antigen presentation in dendritic cells.


64. Albert, M. L., S. F. Pearce, I. M. Francisco, B. Sauter, P. Roy, R. L. Silverstein, and

N. Bhardwaj. 1998. Immature dendritic cells phagocytose apoptotic cells via

alveolar macrophages and include the apoptotic cells in the phagosomes. J. Exp.


Fas leads to Fas ligand-mediated lymphocyte depletion and inflammatory pulmo-


67. Smith, K. G., A. Strasser, and D. L. Vaux. 1996. CrmA expression in T lympho-

cytes of transgenic mice inhibits CD95 (Fas/APO-1)-induced apoptosis, but does

not cause lymphopenumpathy or autoimmune disease. EMBO J. 15: 5167–5176.

68. Huynh, M. L., V. A. Fadok, and P. M. Henson. 2002. Phospholipidserine-

dependent ingression of apoptotic cells promotes TGF-[-beta]-secretion and the res-


69. Morelli, A. E., A. T. Larregina, W. J. Shufesky, A. F. Zahorchak, A. J. Logar,


dritic cells: dependence on complement receptors and effect on cytokine production.


70. Hoffmann, P. R., J. A. Kench, A. Vondraeck, E. Kruk, D. L. Daleke, M. Jordan,

P. Marrack, P. M. Henson, and V. A. Fadok. 2005. Interaction between phos-


71. Talamazi, T., H. Akiba, H. Iwai, H. Marsuda, M. Aoki, Y. Tanno, T. Shin,


72. Fibe, B. T., and J. A. Bluestone. 2008. Control of peripheral T-cell tolerance and

autoimmunity via the CTLA-4 and PD-1 pathways. Immunity Rev. 226: 166–182.


dendritic cell-mediated suppression via interferon-gamma-induced IDO. Immuno-


75. Lacy-Hullbert, A. M. A. Smith, H. Tissie, M. Barry, D. Crowley, R. T. Bronson,


76. Nencioni, A., C. Grunebach, S. M. Schmidt, M. R. Muller, D. Boy, F. Parrone,

A. Ballesterro, and P. Bossart. 2008. The use of dendritic cells in cancer immu-


78. Josien, R., H. L. Li, E. Ingulli, S. Sarna, B. R. Wong, M. Volodogokadia,


79. Kim, J. H., T. H. Kang, K. H. Noh, H. C. Bae, S. H. Kim, Y. D. Yoo, S. Y. Seong,


anti-apoptotic sRINAs targeting key pro-apoptotic proteins in cytotoxic CD8(alpha)+


80. Miga, A. J., S. R. Masters, B. G. Durell, M. Gonzalez, M. K. Jenkins,


member, enhances the longevity and adjuvant properties of dendritic cells in vivo.

71: 5167–5176.


strate that monocytes and dendritic cells are rendered apoptotic by extracorporeal


t-cell-dependent elimination of dendritic cells in vivo limits the induction of