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Dominant role of monocytes in control of tissue function and aging

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Summary We propose that monocyte-derived cells regulate expression of epitopes of specific tissue cells, and in that way control recognition of tissue cells by autoreactive T lymphocytes and autoantibodies. Such T cells and antibodies are suggested to participate in stimulation of tissue cell differentiation. This may ultimately result in the aging and degeneration of tissue cells. By the end of their adaptation in early ontogeny, the monocyte-derived cells are supposed to encounter the most differentiated tissue cells in a tissue specific manner, and then prevent tissue cells to differentiate beyond the encoded state. Retardation or acceleration of certain tissue differentiation during adaptation results in a rigid and permanent alteration of this tissue function. The ability of monocytes to preserve tissue cells in the functional state declines with age, and this is accompanied by functional decline of various tissues within the body, and an increased incidence of degenerative diseases. © 2000 Harcourt Publishers Ltd

INTRODUCTION

Tissue aging and degeneration not only accompany advanced age, but can affect some tissues and organs from early periods of life. A typical example is insulin-dependent diabetes mellitus, where monocyte-derived cells (MDCs), such as tissue macrophages and/or tissue dendritic cells (DCs), are activated and initiate homing of T cells from the blood. Activated T lymphocytes and autoantibodies then cause apoptosis of pancreatic β cells (1,2). However, natural autoantibodies and autoreactive T cells are present in the blood of normal healthy individuals, and accompany and regulate regeneration and function of certain tissues. For example, natural IgM and IgG autoantibodies in normal human sera specifically bind to epidermal keratins (3) and other normal tissues (4), and intestinal intraepithelial T cells influence proliferation, differentiation, and function of epithelial cells (5,6).

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MONOCYTES AND FUNCTION OF TISSUES

We postulate that monocytes play a dominant role in the preservation of tissues in the normal functional state. Monocyte-derived cells secrete cytokines and growth factors that regulate differentiation and function of specific tissue cells (7,8). The MDCs are essential for tissue regeneration (7,9–11). They produce cytokines and chemotactic factors attracting fibroblasts and endothelial cells, and often activate them to produce additional mediators stimulating angiogenesis and maturation of specific tissue cells (7,9,12–18). Various phenotypes of MDCs, e.g. histiocytes, microglia, Kupffer cells, and Langerhans DCs, are found in virtually all tissues within the body. In many epithelial tissues, the MDCs also interact with intraepithelial T lymphocytes which secrete additional cytokines (5,6). In light of emerging knowledge concerning the dominant role of MDCs in regulation of immune responses to foreign antigens (19,20), and presence of MDCs in virtually all tissues in the body, one must also consider their role in regulation of autologous tissue function.

MONOCYTES AND THE TISSUE CONTROL SYSTEM

Recent developments in the understanding of the role of mesenchymal cells and their products in regulation of

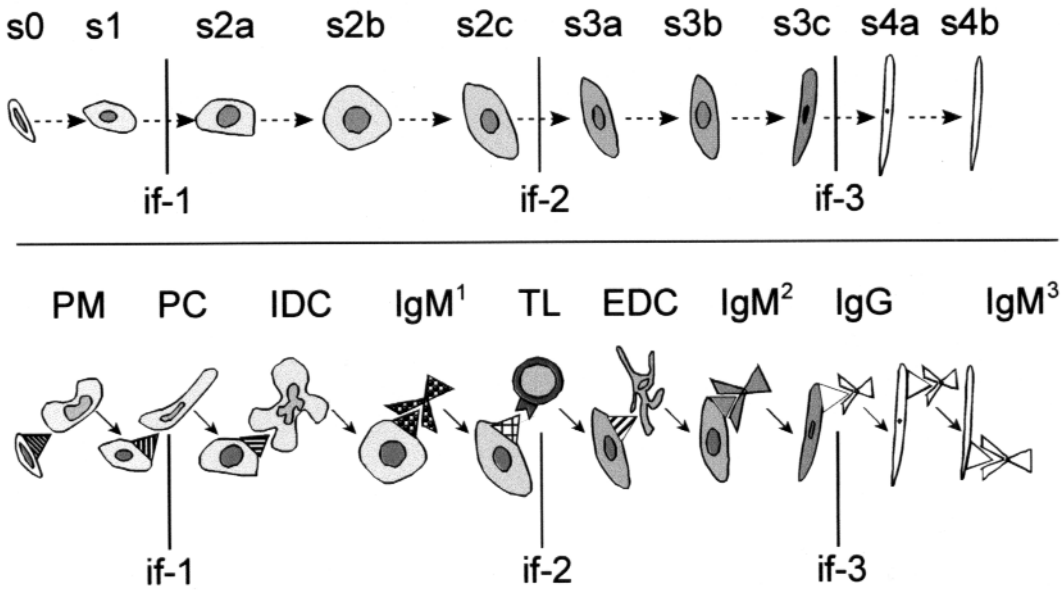

Fig. 1 Schematic drawing of stages of differentiation of tissue cells (*top*), and complete TCS pathway (*bottom*). s, stages (s0, s1, s2, s3, s4) and substages (a, b, c) of tissue cell differentiation (see text); if 1, if 2 and if 3, interfaces between distinct cell layers (squamous epithelium of the ectocervix); PM, perivascular MDCs; PCs, pericytes; IDC, immature dendritic cells (DC precursors); TLs, T lymphocytes; EDC, mature epithelial dendritic cells.

differentiation of tissue cells were initiated more than seventy years ago, when Alexis Carrel demonstrated that leukocyte extracts, like embryonic tissue extracts, stimulate multiplication of fibroblasts in vitro and suggested that leukocytes can bring growth-activating substances to tissue cells (21). Later, in the 1960s and 1970s, lymphocytes were shown to promote tissue growth and regeneration [reviewed in Ref (22)]. In the early 1980s, we suggested that the immune system is also a component of the more complex 'tissue control system' (TCS) (23), and then refined the role of the TCS (24), and postulated its involvement in the regulation of ovarian function [reviewed in Ref (25)] and placental aging and regeneration (11). Here we expand the TCS concept, in line with recent proposals on the dominant role of MDC in regulation of lymphoid cell function (19,20).

Differentiation and aging of tissue cells

The complete pathway of differentiation and aging of tissue cells can be observed during corpus luteum (CL) development and regression, and in squamous epithelia (25). Squamous epithelium of the vagina and ectocervix shows four morphologically distinct layers: basal, parabasal, intermediate and superficial, divided by three morphologically well defined interfaces (interface 1, 2, and 3; top, Fig. 1). A single cell layer of basal cells consists of resident stem cells (s0) and their daughter cells entering differentiation (s1). Parabasal (s2) and intermediate (s3) layers can be subdivided into lower (a), mid (b), and upper regions (c), and superficial cells (s4) into lower (a) and upper regions (b). A hierarchy of differentiating tissue cells (s0–s4b, top Fig. 1) is, in principle, regulated by a hierarchy of TCS elements according to their appearance during phytogency and ontogeny. The bottom of Figure 1 demonstrates a complex relationship of the MDC and other TCS components to tissue cells, during differentiation of tissue cells into the immature (s1/s2a) and mature cells (s2c/s3a), and degeneration into the aged (s3c) and apoptotic tissue cells (s4a/s4b).

Monocytes and complete TCS pathway

The TCS components, including Thy-1 pericytes, MDC subsets, T cells, and immunoglobulins IgM and IgG, appear to recognize particular steps in differentiation of tissue cells for which they have been committed. Figure 2A shows young postmitotic epithelial cells (G0) committed to differentiation. These Ki-*67*$^+$ cells represent s2a stage of differentiation (Fig. 1), because they already left the basal layer (s1↘s2a). Fig. 2B demonstrates that Thy-1$^+$ "granules" [subcellular vesicles (25)] derived from vascular pericytes migrate through the basement membrane, among basal cells and toward the lower region of parabasal cells (arrow, inset 2B). This suggests that Thy-1$^+$ granules may participate in the stimulation of s1↘s2a transition (Fig. 1).

While the role of pericytes and some other TCS components (T cells and IgGs – see below) appears to be restricted to the stimulation of a certain single step in

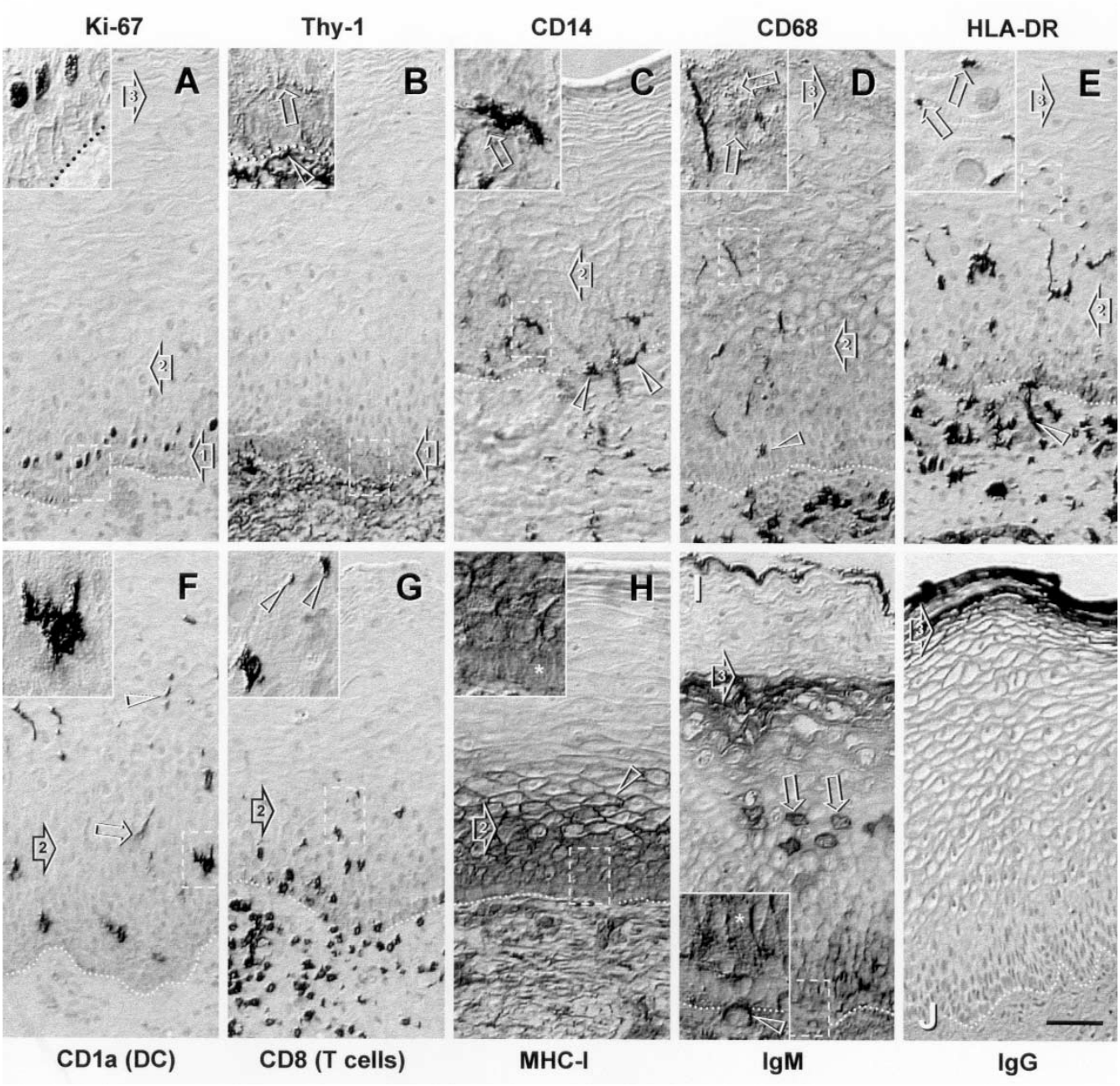

Fig. 2 Staining for the Ki-67 and TCS components (as indicated at the top and bottom rows) in the uterine ectocervix. Areas shown in insets are indicated by dashed boxes. Dotted line = basement membrane. Wide arrows assigned 1, 2 and 3, indicate interfaces if-1, if-2 and if-3 (see Fig. 1). (**A**) Majority of Ki-67$^+$ cells are in the lower parabasal region (s2a stage, see Fig. 1); (**B**) Thy-1$^+$ material from pericytes (inset, arrowhead) migrates toward s2a cells (arrow; s1▲s2a, Fig. 1); (**C**) CD14$^+$ primitive MDC (arrowheads) associate with basal cells (s0▲s1, Fig. 1), and some migrate (asterisk) into the epithelium. Immature DC (see also **F**) release CD14$^+$ granules (inset, arrow) among parabasal cells (s2a▲s2b, Fig. 1); (**D**) Some immature DC may express CD68 (arrowhead). Mature epithelial DC release CD68 granules which associate with epithelial cells (inset, arrows; s3a▲s3b, Fig. 1) and accompany them till early stage of apoptosis (above the interface 3); (**E**) HLA-DR is expressed by perivascular MDC and endothelial cells (arrowhead). Mature epithelial DC entering the mid intermediate region show apoptotic fragmentation (inset, arrows);(**F**) Immature DC show veiled-like character (inset) and differentiate into mature epithelial DC (arrow), which undergo apoptotic fragmentation (arrowhead) in the mid intermediate region (see also **E**); (**G**) MHC class I restricted T cells migrate through the parabasal layers, and exhibit apoptotic fragmentation (arrowheads, inset) in the lower intermediate region (s2c▲s3a, Fig. 1); (**H**) MHC class I heavy chain (mAb W6/32) is weakly expressed in basal cells (inset, asterisk). Parabasal cells exhibit moderate cytoplasmic (heavy chain synthesis) and surface expression. Cells in lower intermediate region, where T cells degenerate (above interface 2, see **G**) exhibit lack of cytoplasmic but strong surface staining (arrowhead); (**I**) Some parabasal cells (inset, asterisk; s2b▲s2c., Fig. 1), isolated cells in the mid intermediate region (arrows), and all cells in the upper intermediate region (s3b▲s3c) show surface binding and cytoplasmic staining for IgM. The cells at the surface are also stained (s4a▲b4b). Note IgM capping (arrowhead) of the lymphocyte entering epithelium, and sharp interface 3 between the IgM$^+$ and IgM$^-$ epithelial cells; (**J**) IgG binds to the cells entering (s3c▲s4a) and undergoing apoptosis (s4a▲s4b). Bar = 60 μm; for insets the bar = 20 μm.

differentiation of tissue cells, the role of MDCs is much more complex. The MDCs differentiate along with tissue cells, interact with less and more mature tissue cells, and secrete stage-specific 'granules' among differentiating tissue cells. During differentiation pathway, the CD14 is expressed by less differentiated MDCs, which associate with the basal epithelial cells (arrowheads, Fig. 2C). This suggest that primitive MDC may stimulate s0▲s1 transition (Fig. 1). The MDC eventually migrate into the epithelium (asterisk, Fig. 2C) and secrete CD14 [known as a receptor for lipopolysaccharide (26)] granules among parabasal cells (arrow, inset Fig. 2C). This may stimulate s2a▲s2b transition (Fig. 1). The mature epithelial DC secrete CD68 [the lysosome-associated glycoprotein (27)] granules, which associate with lower intermediate cells and accompany epithelial cells till the early stage of apoptosis (Fig. 2D). This suggests that mature DC may stimulate s3a▲s3b transition (Fig. 1). Next, the mature epithelial DC undergo apoptotic fragmentation within the mid region of intermediate cells (Fig. 2, E and F).

The MHC class I restricted T cells accumulate among basal epithelial cells (Fig. 2G), which show weak staining for MHC class I heavy chain (asterisk, inset Fig. 2H). Some of the T cells migrate among parabasal cells, which show enhanced cytoplasmic and surface MHC class I expression (Fig. 2H). After reaching interface 2, the T cells undergo apoptotic fragmentation among the lower intermediate cells (Fig. 2G). This suggests that T cells may stimulate the s2c???s3a transition (Fig. 1). This transition of epithelial cells is accompanied by a diminution of MHC class I heavy chain synthesis (Fig. 2H).

Distribution of TCS elements within the layers of the squamous epithelium of ectocervis (Fig. 2, B–J), mirrors the development of the TCS and immune system components throughout evolution (phylogeny), and during ontogeny of an individual. The immunoglobulins M, which evolve earlier in phylogeny than immunoglobulins G, and represent a class of most naturally occurring autoantibodies (4), show specific binding to the parabasal, upper intermediate, and upper superficial regions of epithelial cells (Fig. 2I). This suggests that similarly to MDC, the autoreactive IgMs may also the involved in the regulation of several steps in differentiation of tissue cells.

Based on Fig. 2I, and studies of other samples and tissues, e.g. developing and regressing CL (data not shown), possible sites for IgM interaction with tissue cell differentiation (s2b???s2c, s3b???s3c, and s4a???s4b) are proposed in Figure 1. In addition to the surface binding and cytoplasmic occurrence of IgM in epithelial cells (Fig. 2I), the binding to the front of lymphocytes entering the epithelium (IgM capping) is apparent in the inset of Figure 2I (arrowhead). On the other hand, immunoglobulins G, the class evolving later during phylogeny, i.e. in higher vertebrates, bind to the surface layers of epithelial cells only (Fig. 2I). This suggests that autoreactive IgGs may stimulate apoptosis of aging (s3c) tissue cells (Fig. 1).

No population of somatic cells that multiplies in vitro expresses the full panoply of differentiated traits of which the cell type is capable (28). Normal cells show proliferation and certain degree of differentiation in vitro only if supplied by serum and/or growth factors and cytokines, i.e. substances infected by immune cells (MDCs and lymphocytes). During immune reaction, the target cells (e.g. virus-infected cells) are subjected to cytolysis. In contrast, during regeneration of tissue cells, the immune cells degenerate while the tissue cells differentiate. Hence, the apoptotic fragmentation of T cells in the lower intermediate region (s2c▲s3a), and mature epithelial DC in the mid intermediate region (s3a▲s3b), suggest that a suicide of specialized mesenchymal cells may be required for the stimulation of certain steps in differentiation of tissue cells.

However, the continuation of the complete TCS pathway in vivo requires that tissue cells not only proceed to the higher level of differentiation, but that they also express epitopes which can be recognized by a subsequent element in the TCS hierarchy (bottom, Fig. 1). A lack of expression of new epitopes may result in an interruption of the TCS pathway (stop-effect, see below).

Involvement of autonomic innervation and lymph nodes

Regeneration of tissue cells requires regulation of the quality, but also of the quantity of differentiating tissue cells. Autonomic innervation has been suggested to regulate the quantity of specific cells within the tissues (24, 25). Autonomic innervation is associated with the basic 'tissue control unit' accompanying microcirculation (top left, Fig. 3), where it may control an activation of vascular pericytes (25,29). In this way, the autonomic innervation can exert a local control over the TCS pathway, from its very beginning.

We suggest that the differentiation of tissue cells is initiated by disinhibited stem cells (s0*, Fig. 3) requesting support (arrow 1) from perivascular MDCs, which exhibit physical contacts with immature epithelial cells through the basement membrane (30). The request is extended toward the pericytes (arrow 2), and followed by the request toward autonomic innervation (arrow 3). If not inhibited (step 4, ai^+), the pericytes activate monocyte-like cells (arrow 5), which promote proliferation and early differentiation of tissue cells (arrow 6), including expression of new markers (step 7). Subsequently, perivascular MDCs differentiate into the immature DC activating pericytes (arrow 8), which stimulate the next step of differentiation of tissue cells (arrow 9, and step 10) (30).

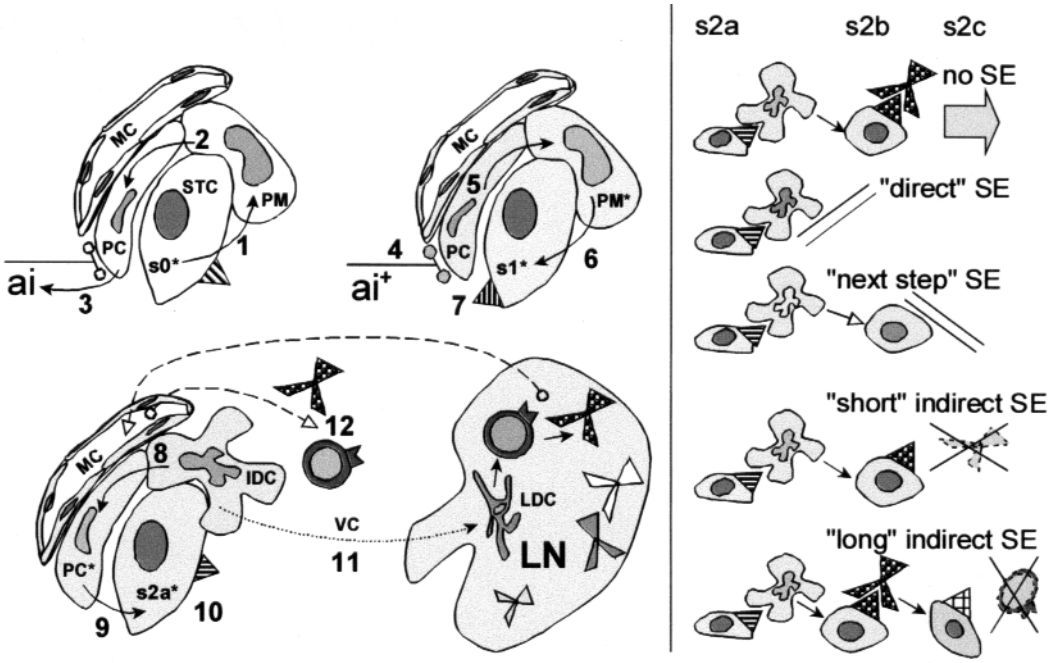

Fig. 3 Involvement of innervation and lymph nodes in TCS function (*left*) and stop-effect (SE) of monocyte-derived cells (*right*). STC, specific tissue cells; ai, autonomic innervation; MC, microcirculation; VC, veiled cells; LDC, lymphoid dendritic cells; LN, lymph node; asterisks, activated cells; crosses (X), systemic or local defects. Other abbreviations are explained in Figure 1. For details see text.

The involvement of autonomic innervation does not apply for tissues where the innervation is known to be absent, i.e. CL, placenta and cancer. In these situations, the TCS either stimulates maturation of entire structure toward aging, e.g. CL that is unable to regenerate (29), or the activity of the TCS within placenta is controlled by the maturity of fetal MDCs (11). However, in malignant diseases, the alteration of autonomic innervation at the tumor–host interface causes permanent activation of basic TCS units, which contributes to the acceleration of tumor growth progression (in preparation).

In epithelial tissues, the immature DCs migrate among epithelial cells and differentiate into the epithelial DCs (31). The immature intraepithelial DCs can also return into the subepithelial stroma, where they are transformed into veiled cells, which reach the regional lymph nodes via afferent lymphatics (dotted arrow 11, Fig. 3) (20,31, 32). In lymph nodes, the veiled cells differentiate into the lymphoid DCs instructing T cells and influencing production of antibodies (20,31). In this way, the lymphoid DC may facilitate production of tissue-committed autoreactive T cells and natural autoantibodies, which reach the peripheral tissues via efferent lymphatics and the blood (dashed arrows, step 12). The pericytes and differentiating perivascular MDCs were also proposed to regulate differentiation and properties of the endothelial cells in microvasculature (29), which influence homing of monocytes and T cells (33–35).

'STOP-EFFECT' OF MONOCYTE-DERIVED CELLS

We have earlier proposed that tissues can be prevented from aging by a stop-effect interrupting continuation of the TCS pathway (24,25), but we have not addressed involvement of MDCs and possible mechanisms of the stop-effect in detail. The MDCs may play a dominant role in tissue preservation via various stop-effect mechanisms. The 'direct' stop-effect (right, Fig. 3) represents a situation when MDCs by themselves do not promote continuation of differentiation of dependent tissue cells. The 'next step' stop-effect (Fig. 3) consists of the promotion of tissue cells into the next stage of differentiation, but expression of new epitopes is not stimulated. The MDCs may also act indirectly, by prevention of production of certain tissue-specific natural autoantibodies ('short' indirect stop-effect, Fig. 3) or by inhibition of production, homing, and activation of tissue-committed autoreactive T cells ('long' indirect stop-effect, Fig. 3). The last two events may contribute to the preservation of the functional CL during pregnancy (29).

Stop-effect independent and dependent tissues

The complete TCS pathway is required for the complete differentiation and aging of epithelial cells in the epidermis and ectocervix, and development and regression of the CL of menstruation (29) (stop-effect independent

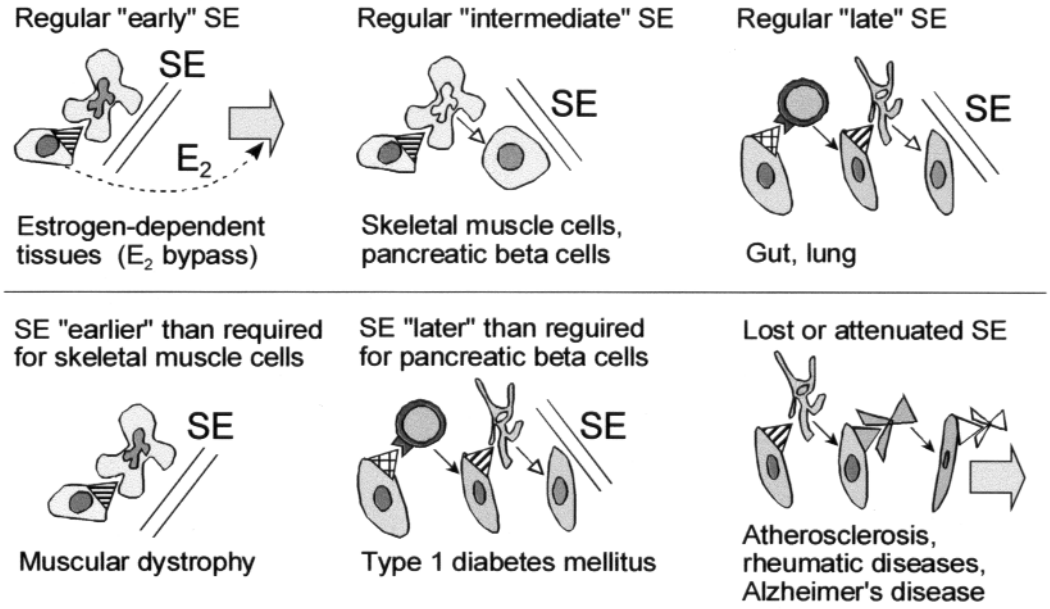

Fig. 4 Role of the stop-effect in tissue physiology (*top*) and pathology (*bottom*). Thin open arrows indicate 'next step' stop-effect (see Fig. 3). Note 'direct' stop-effect in the estrogen-dependent tissues and muscular dystrophy. Large wide arrows = continuation of tissue cell differentiation toward aging and apoptosis.

tissues). The specific cells of these tissues should be enabled to enter apoptosis; an existence of the stop-effect will cause insufficient maturation of stratified epithelia, and a lack or prolongation of the CL regression.

In contrast, the specific cells of most other tissues should be prevented from aging and degeneration by the stop-effect of the TCS (stop-effect dependent tissues). Moreover, the parenchymal and epithelial tissues usually differ in their needs for the optimal stage of differentiation. Some examples are given at the top of Figure 4. An 'early' stop-effect (stage s2a) is required for the regular function of certain estrogen-sensitive tissues, particularly in the female reproductive tract (endometrium, vagina). This 'early' stop-effect in these tissues can be bypassed by estrogens (25). After estrogen withdrawal, the 'early' stop-effect enables the estrogen-sensitive tissues to return to the early stage of differentiation. The 'intermediate' stop-effect (stage s2b) is characteristic for parenchymal tissues lacking T cells, whereas a 'late' stop-effect (stage s3b) is characteristic for epithelial tissues with intraepithelial T lymphocytes. The 'late' stop-effect enables homing of intraepithelial T cells and their interaction with epithelial cells required for the function of the gut and lung (*6*), yet it still prevents epithelial cells in these tissues from undergoing apoptosis.

Determination of the stop-effect

Function of the TCS is proposed to be established in early ontogeny, during the TCS development and adaptation, which encompasses the period of immune adaptation (24,25,*29*). By the end of the TCS adaptation, circa at the end of the second trimester of intrauterine life in man and during second postnatal week in the rat and mouse, the most differentiated tissue cells are encoded within the TCS in a tissue-specific manner. Thereafter, the tissues are preserved in this encoded state by the stop-effect of MDCs.

Retardation or acceleration of certain tissue differentiation during adaptation results in a rigid and permanent alteration of this tissue function, such as muscular dystrophy associated with permanent immaturity of muscle fibers (*36*–*38*), or type 1 diabetes mellitus associated with permanent aging of pancreatic beta cells (2), respectively (bottom, Fig. 4). When the stop-effect of MDCs is 'later' than required, the alteration of tissue may resemble an autoimmune condition. However, in contrast to virus-infected cells, the specific cells of such tissues are not directly affected by infiltrating T cells, but they are stimulated to age and undergo apoptosis (*39*), a process that physiologically occurs in the epidermis. In estrogen-dependent tissues, the 'later' stop-effect than required, induced by estrogenic stimulation during TCS adaptation, is proposed to be a cause of the estrogen-independent proliferation and hyperplasia of the vagina (vaginal adenosis) found in young 'diethylstilbestrol females' (40).

Aging of the TCS and functional decline of tissues

Continuation of normal tissue function during childhood and adulthood may depend on the capacity of the TCS to produce MDCs exhibiting adequate stop-effect (24). The

TCS function declines with advancing age, and this is accompanied by a functional decline (aging) of other tissues within the body, including diminution of the female reproductive function (25). Simultaneously, the incidence of degenerative diseases increases, e.g. multiple sclerosis, rheumatic diseases, atherosclerosis, and Alzheimer's disease, possibly due to the attenuation or loss of the stop-effect toward certain tissues in some individuals (bottom right, Fig. 4).

IMMUNODEFICIENCY AND HOMEOSTASIS OF TISSUES

Tissue function in osteopetrotic mice

Osteopetrotic (op/op) mice are deficient in macrophage colony stimulating factor (M-CSF) because of the recessive osteopetrotic (op/op) mutation. Osteopetrosis is associated with a lack of osteoclasts, the phagocytic cells required for remodeling in bone. Some additional macrophage populations have also been proved to be very M-CSF dependent. The op/op mice have few and sometimes no peritoneal cavity phagocytes, splenic marginal zone metallophils, and lymph node subcapsular sinus macrophages. Other populations, however, reach substantial levels in the absence of M-CSF, including MDCs in the thymic cortex, splenic red pulp, lymph node medulla, intestinal lamina propria, liver (Kupffer cells), lung (alveolar macrophages), and brain (microglia).

Dendritic cells, which are specialized accessory cells for T-dependent immune responses and tolerance, were readily identified in the skin, and in T-dependent regions of spleen, lymph nodes and Peyer's patches (41). In addition, only a few MDC populations are critically dependent upon M-CSF in vivo. With respect to dendritic cells, it was noted that granulocyte/macrophage (GM)-CSF but not M-CSF supports dendritic cell viability, function, and growth (41). The phenotype of gross deficiency in the macrophage and osteoclast lineages corrects significantly with age, suggesting that other factors can substitute for M-CSF. It appears that op/op mouse is not completely M-CSF deficient and alternative splicing within the M-CSF gene might bypass the mutation, yielding an incompletely penetrant phenotype (42). Hence, the lack of M-CSF in op/op mice affects those tissues where the M-CSF macrophages are required, but does not affect the function of macrophages and dendritic cells in other tissues.

Role of the TCS in regulation of self

The TCS concept has also to deal with the thymus-dependent reactions toward non-self, such as cytotoxic reaction toward virus-infected cells and toward allo- and xenografts. These thymus-dependent reactions are severely altered in the congenitally athymic (nude) mice and mice with severe combined immunodeficiency (SCID) (43,44). However, except for a lack of thymus-dependent reactions, these rodents do not exhibit marked alteration of their tissue function, suggesting that thymus-derived T cells (carrying αβ T cell receptor) are not necessarily required for the self tissue homeostasis. There is one interesting exception, characterized by the persistence of aging corpora lutea (CL) and accumulation of secondary interstitial tissue in the ovaries (45,46). Morphological regression of the CL and ovarian interstitial cells requires intervention of T cells (*29*) (unpublished data), and a beginning of the immune senescence is associated with the retardation of the CL regression in normal individuals (47). These data indicate that thymus-derived T cells are involved in the regression of the CL, i.e. self structure absent during immune adaptation.

Although the nude and SCID mice are deficient in thymus-derived αβ T cells, they have a large number of $CD3^+$ and $CD8^+$ extrathymic γδ T cells in the spleen, mesenteric lymph nodes, peritoneal cavity, lamina propria and epithelial layer of the small and large intestine. Low numbers of CD3+ T cells can only be detected in the inguinal, popliteal, and axillary lymph nodes (48–51). Hence, the congenital lack of αβ thymic T cells is not critical for the normal function of epithelial tissues, where αβ T cells are substituted with extrathymic γδ T cells, but it affects the immune surveillance.

EPIGENETIC PROGRAMMING OF TISSUE FUNCTION AND AGING

Data in the literature (45,46,52–*62*) and our recent observations indicate that function of tissues during adulthood is programmed during immune adaptation. Figure 5 summarizes studies on the role of tissue differentiation (rows 1–5) and immune adaptation (rows *6*–10) within the critical window of rat and mouse development. If the ovarian development is inhibited by multiple doses of synthetic estrogens, the maturation of follicles in adult ovaries is severely inhibited (row 2). Single injection of testosterone propionate (TP) causes delayed (row 3) or early anovulation (row 4) associated with the premature aging of the ovary. The time at which the manifestation of an ovulation after postnatal TP occurs is dose-dependent (row 3 vs row 4). However, the anovulation with premature aging of the ovary can be prevented by an adoptive transfer of the suspension of thymic cells from prepubertal females (row 5). Although the ovarian development is also severely accelerated toward the aging phenotype by injection of TP after termination of immune adaptation (Day 10), morphology and function of adult ovaries is not affected (not shown). These observations indicate that the stage of differentiation of ovaries prior to the termination of immune adaptation determines stage of differentiation

of ovarian structures in adult rat females. However, the functional failure can be prevented by adoptive transfer of immune cells obtained from normal immunocompetent rats.

Congenitally athymic and SCID females lack functional thymus-derived T cells (no immune adaptation toward self/non-self), and their ovaries show an occurrence of normally developed antral follicles (row *6*, Fig. 5). If normal mice are subjected to neonatal thymectomy (prior to the critical window), ovarian function is not affected (row *7*). The same applies for female mice thymectomized after the end of immune adaptation (row 8). However, thymectomy performed within the critical window results in the severe inhibition of follicular development in adult ovaries (row *9*). If, however, the last experiment is preceded by adoptive transfer of splenic or thymic cells from adult mice, adult ovaries are not affected (row 10).

These data suggest that an alteration of tissue differentiation within the critical window of immune adaptation, or interruption of the immune adaptation during this period, cause severe and permanent alteration of tissue function in adult individuals. Hence, the immune adaptation plays an important role in the determination of adult tissue function. The data also show that T cells, MDCs, or other thymic or splenic components of immunocompetent females carry an information important for the determination of adult tissue function.

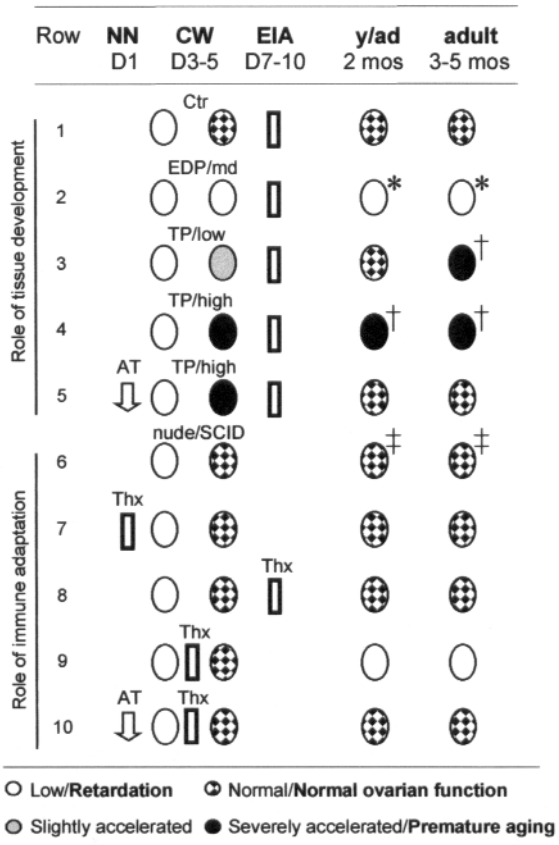

Fig. 5 Ovarian model on the role of early development and immune adaptation in determination of adult tissue function. Note prevention of ovarian failure by adoptive transfer (AT) of splenic or thymic cell suspension from normal rat and mouse adult females to neonatal (NN) recipients. Rounded symbols indicate stage of Postnatal differentiation/Resulting state of adult ovary (bold letters at the bottom). Boxes indicate end of immune adaptation (EIA) eventually induced by thymectomy (Thx). CW, critical window (postnatal Day 3–5); y/ad, young/adult; ctr, controls; EDC/md, estradiol dipropionate/multiple doses; TP/low, single injection of 5 μg testosterone propionate (TP); TP high, single injection of 100 or 500 μg TP.* = basal serum gonadotropin levels do not differ from controls (66,67); † = Normal function of hypothalamo-pituitary system in adult postnatally androgenized rats (68,69), and prematurely aging ovaries resume function when transplanted to normal adults recipients (55); ‡ = persisting CL and accumulation of steroidogenic interstitial cells in nude mice (45,46). Data for rows 1–5 from Refs (52–56) and unpublished observations, and for rows 6–10 from Refs (45,46,57–62).

TESTING THE HYPOTHESIS

Our hypothesis represents a novel interpretation of some observations and experiments that already have been done. For instance, in Duchenne muscular dystrophy, muscle cells do not differ from normal muscle cells in their ability to differentiate in vitro (*36*). However, the development of muscle fibers in congenital myotonic dystrophy is inhibited during the fetal period by maternal serum factors, and muscle immaturity persists thereafter (*37*). In mice, neonatal stimulation of estrogen-dependent tissues by injection of estrogens causes estrogen-independent cornification of the vagina in adult females (*63*,*64*).

More specifically for our proposals, small laboratory rodents can be used to test the effect of administration of antigens from certain adult tissues during the postnatal critical period (first week of postnatal life). For instance, introduction of the CL antigens could be used to demonstrate the importance of the lack of the stop-effect for normal regression of the CL in adult females. Another option is the adoptive transfer of MDCs from adult animals exhibiting morpho-functional diseases, e.g. experimentally induced polycystic kidney disease (*65*), to immunologically immature normal rodents. It has been shown that the adoptive transfer of immune cells from normal mature female rats prevents androgen induced anovulation (54). Hence, we propose to test the ability of MDCs from adult postnatally androgenized female rats to transfer anovulation when injected to normal neonatal recipients. Figure 5 (rows 1–5) also indicates other situations to be tested. Retardation of adult ovaries after postnatal estrogenization (row 2) might be prevented by injection of cells from normal prepubertal ovaries. Accordingly, premature aging of ovaries might

be induced by injection of ovarina cells from adult postnatally androgenized donors to neonatal rats.

CONCLUSION

In conclusion, we propose that the epigenetic or genetically programmed alteration of tissue development during immune adaptation predetermines alteration of adult tissue function. The functional failure may occur early or later during the life-span, depending on the extent of alteration of tissue development, e.g. early vs delayed anovulation (Fig. 5). A better understanding of the role of monocytes in regulation of tissue function will be helpful for novel approaches to the prevention and therapy of various idiopathic diseases, which are associated with an inability of tissue cells to attain a mature state, e.g. muscular dystrophy, or caused by premature aging of tissues, e.g. diabetes mellitus, multiple sclerosis, rheumatic diseases, atherosclerosis, and Alzheimer's disease.

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