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MINI REVIEW

The thymus is relevant in the migration of mature lymphocytes

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Abstract

The thymus is a primary lymphoid organ where T lymphocyte proliferation and selection takes place. The different subsets of lymphocytes leave the thymus as recent thymic emigrants. Peripheral dendritic cells migrate to the thymus. In addition to the homing of hematopoietic progenitor cells to the thymus, there is evidence for lymphocyte entry from peripheral lymphoid tissues mainly into the medulla. The entry sites are the venules in the medullary part near to the cortex with a higher endothelium. Furthermore, there are also B lymphocytes in the thymus. The thymus is not only a primary lymphoid organ but is well integrated in lymphocyte traffic as shown in several different species.

Keywords Thymus · Medulla · Lymphocyte entry · Peripheral lymphoid organs · B lymphocytes

Introduction

The thymus is normally taken as a typical primary lymphoid organ thus independent of the influx of mature lymphocytes from the periphery (see textbooks of immunology, e.g., Cowan et al. 2016, or of anatomy, see Standring 2016). However, there are many data documenting the entry of lymphocytes from the blood and secondary lymphoid organs into the thymus, which have mostly been neglected so far.

The other primary lymphoid organ—the bone marrow—is also part of T lymphocyte migration and a site of long-term memory plasma cells as recently summarized (Pabst 2018).

Ontogeny and basic structure

The epithelium of the thymus is derived from the third and/or fourth pharyngeal endoderm and ectoderm from the third brachial cleft and neural crest (for detailed references see von Gaudecker 1986; Boyd et al. 1993). For the development of subsets of T cells and their expansion, a complex microenvironment is essential (Boyd et al. 1993 and the detailed review by Petri and Zúñiga-Pflücker 2007). Rodewald (2007) included a cervical thymus in the mouse, which produces T cells only after birth.

Stem cells from the bone marrow enter this epithelial structure and differentiate into different subsets and form the structural backbone. There is a multistep cascade of adhesion molecules essential for bone marrow–derived progenitor cells to home to the thymus (Scimone et al. 2006; Krueger et al. 2010). The influx of a precursor is necessary throughout postnatal life as shown by parabiotic experiments in mice by Donskoy and Goldschneider (1992) and Zlotoff and Bhandoola (2011). There are two compartments: the cortex and medulla (Fig. 1). In the cortex, the clonal expansion in producing specific T cell responses and at the same time acquiring immune tolerance for its own tissue components is essential. The homing of dendritic cells to the thymus is critical for the development of central tolerance (Hadeiba and Butcher 2013), because these plasmacytoid dendritic cells transport peripheral antigens to the thymus (Hadeiba et al. 2012). The unique microenvironment of the thymus has recently been summarized (Cowan et al. 2016). In children without a thymus (di George Syndrome), there is a severe immune defect. These right and left anlage move downwards during development and therefore, in several species, there is a cervical thymus on both sides of the trachea and a retrosternal thymus, e.g., in humans. This migration into the thoracic cavity is the explanation why benign

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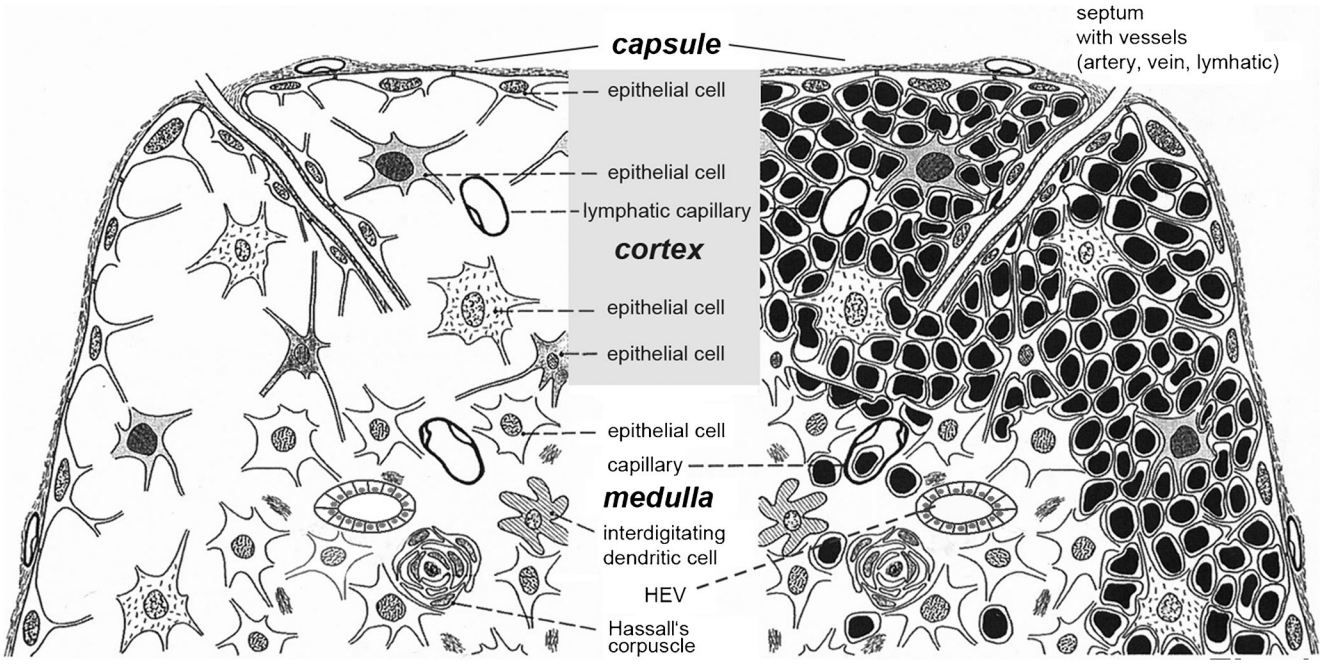

Fig. 1 Schematic drawing of the histological structure of the thymus (modified after Pabst 2004)

tumors (thymoma) or thymic carcinoma are localized in the anterior mediastinum (for review see Hess et al. 2016; Scorsetti et al. 2016). Within the thymus, developing T cells show a very heterogeneous migratory behavior (Bajoghli et al. 2015). With age, the thymus atrophies and many thymocytes are replaced by fat cells (Standring 2016). However, even in old age (> 80 years), functioning thymic tissue can be found in humans (Steinmann 1986).

In 1963, the group of Starzl tried to improve the results of kidney transplantation by a previous thymectomy in four adult patients without obvious effects (Starzl et al. 1963).

The thymus is very much stress-dependent, probably due to the rich innervations (reviewed by Boyd et al. 1993). It would be of interest, how the reconstitution of the thymus happens after stress atrophy. Is it a repetition of ontogeny? The size of the thymus in full-term newborn humans has been studied by ultrasound, e.g., the left lobe is longer and thicker and boys have a larger mass than girls (Varga et al. 2011). Immunosenescence is often related to thymus atrophy. However, it has been shown that the human thymus functions life-long, although to a progressively diminishing extent (Bertho et al. 1997; Marusić et al. 1998; Poulin et al. 1999).

The thymus can also have local infections and systemic infections have many effects on thymic function by inflammatory mediators (Nunes-Alves et al. 2013). The thymic function of T lymphocyte expansion depends on an effective barrier between the external and internal thymic environment—the blood-thymic barrier (Boyd et al. 1993).

In textbooks of anatomy and immunology, it is generally stated that there is an influx of stem cells into the thymic parenchyma and enormous numbers of T cells are exported to peripheral lymphoid organs. An often neglected compartment is the perivascular space, which has carefully been studied in detail by different EM techniques (Ushiki and Takeda 1997).

Traffic of mature lymphocytes into the thymus

Many years ago, Michie and Rouse (1989) transferred lymphocytes intravenously from peripheral lymph nodes in congenic mice and recovered 0.2–0.3% of the lymphocytes in the thymic medulla. In parabiotic mice, the immigration of T lymphocytes into the thymic medulla was also shown (Hirokawa et al. 1989). These cells frequently expressed adhesion molecules of the peripheral lymph node type. Hale and Fink (2009) reviewed the migration of peripheral T cells to the thymus. Sprent and Surth (2009) argued also about the reentry of mature lymphocytes into the thymus. The role of the adhesion molecule interaction and the entry of CD45RC CD4 T cells were documented. Other adhesion molecules or chemokines have not yet been studied, as far as I know. Mostly, cell suspensions of lymphocytes of peripheral organs were used in these experiments. In mice, peripheral T lymphocytes entering the thymus contributed to the selection (Kirberg et al. 2008; Bosco et al. 2009). Thoracic duct lymphocytes, which

are physiologically migrating cells, were collected in rats and transferred into allotype-marked donors and also T lymphocytes in spleen- or lung-transplanted animals were studied (Westermann et al. 1996). The thymic immigrants from splenic transplants consisted of 40% T lymphocytes and 55% B lymphocytes. In contrast, from lung transplants, 85% of the immigrants were T and in 10% B cells. Furthermore, in both situations, some immigrants showed markers of NK cells. All these thymic immigrants were found in the medulla, mostly in the corticomedullary junction and not in the cortex. When rats were injected with the DNA precursor (BrdU) for 12 weeks orally, more antigens experienced CD45R than naïve T lymphocytes migrated to the thymus and entered the medulla (Westermann et al. 1996). Obviously, the reentering peripheral T cells interfere in the thymus with central tolerance induction (Edelmann et al. 2011).

In young pigs, peripheral lymphoid organs such as the spleen (Pabst et al. 1980; Pabst and Binns 1989a,b) or mesenteric lymph nodes were selectively labeled with different markers by direct injection into the supplying artery (Pabst et al. 1977; Pabst et al. 1978). In all these experiments, the thymic immigrants were identified in the medulla near to the corticomedullary junction as shown in Fig. 2 (Binns and Pabst 1988; Pabst et al. 1989). Venules with atypical high endothelium have been detected before the medulla and perimedullary cortex as an entry site for hematopoietic stem cells in the mouse (reviewed in Petri and Zúñiga-Pflücker 2007). Despite the low labelling index of only 0.02%, the total number was considerable due to the large size of the thymus at that age of the piglets, e.g., 30×10^6 after labelling the pig spleen (Pabst et al. 1978). There is also a traffic of mature lymphocytes from the bone marrow to the thymus. In the thymus, only a low labelling index was documented but, due to the enormous total numbers of lymphocytes in the thymus, a total estimated number of mature lymphocyte migrating from the hind leg bone marrow to the thymus was 20×10^6 (Pabst et al. 1983). Comparable experiments in lambs resulted in 7×10^6 small- and medium-sized lymphocytes (Pabst et al. 1986). The latter data could only be obtained in a larger animal such as the pig after labelling the cells in their normal microenvironment.

Thus, the thymus is not only exporting lymphocytes (recent thymic emigrants) but is also included in the migratory route of lymphocytes. The physiological role of these immigrants in particular the B cells (Pabst et al. 1989) is still not known.

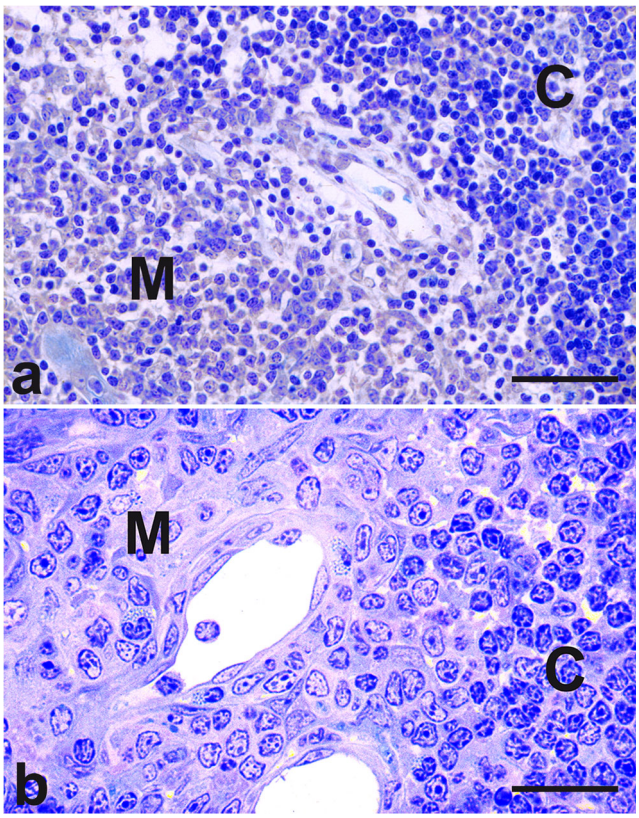

Fig. 2 Histological section of the thymus of a mini pig (~12 weeks of age) after perfusion fixation of the animal (Giemsa staining). C, cortex; M, medulla. **a** Overview (bar 40 μm). **b** Venules with lymphocytes in the lumen (bar 20 μm)

Emigration of T lymphocyte subsets from the thymus

The bone marrow was labeled selectively with fluorescein isothiocyanate (FITC) by an extra corporal perfusion in young pigs and the cells emigrating within 1 day localized other bones and secondary lymphoid organs (Pabst et al. 1983).

When T lymphocytes leave the thymus, they are not fully mature. The recent thymic emigrants are a distinct type of T cells, which also differ in glycolytic and mitochondrial metabolism (Cunningham et al. 2018a). The unique physiology of these recent thymic emigrants has not only been shown in mice but also in humans (Cunningham et al. 2018b).

After selective labelling of the pig, thymus lymphocyte subsets in the thymic vein were quantified (Binns et al. 1988). There were differences between lymphocytes in the peripheral blood, within the thymus and thymic emigrants. Kotani et al. described in 1966 the lymphatic vessels in Guinea pigs. In sheep, thymic emigration via the efferent lymphatics and thymus vein was quantified and characterized. The surface markers of the cells in the lymph differed from peripheral blood cells, e.g., by the expression of MHC antigens. The total number of emigrants was low in comparison with de novo thymocytes (Miyasaka et al. 1990a). Recently, Takeda et al. (2016) studied in detail the expression of receptors (sphingosine-I-phosphate) on T cells in the mouse

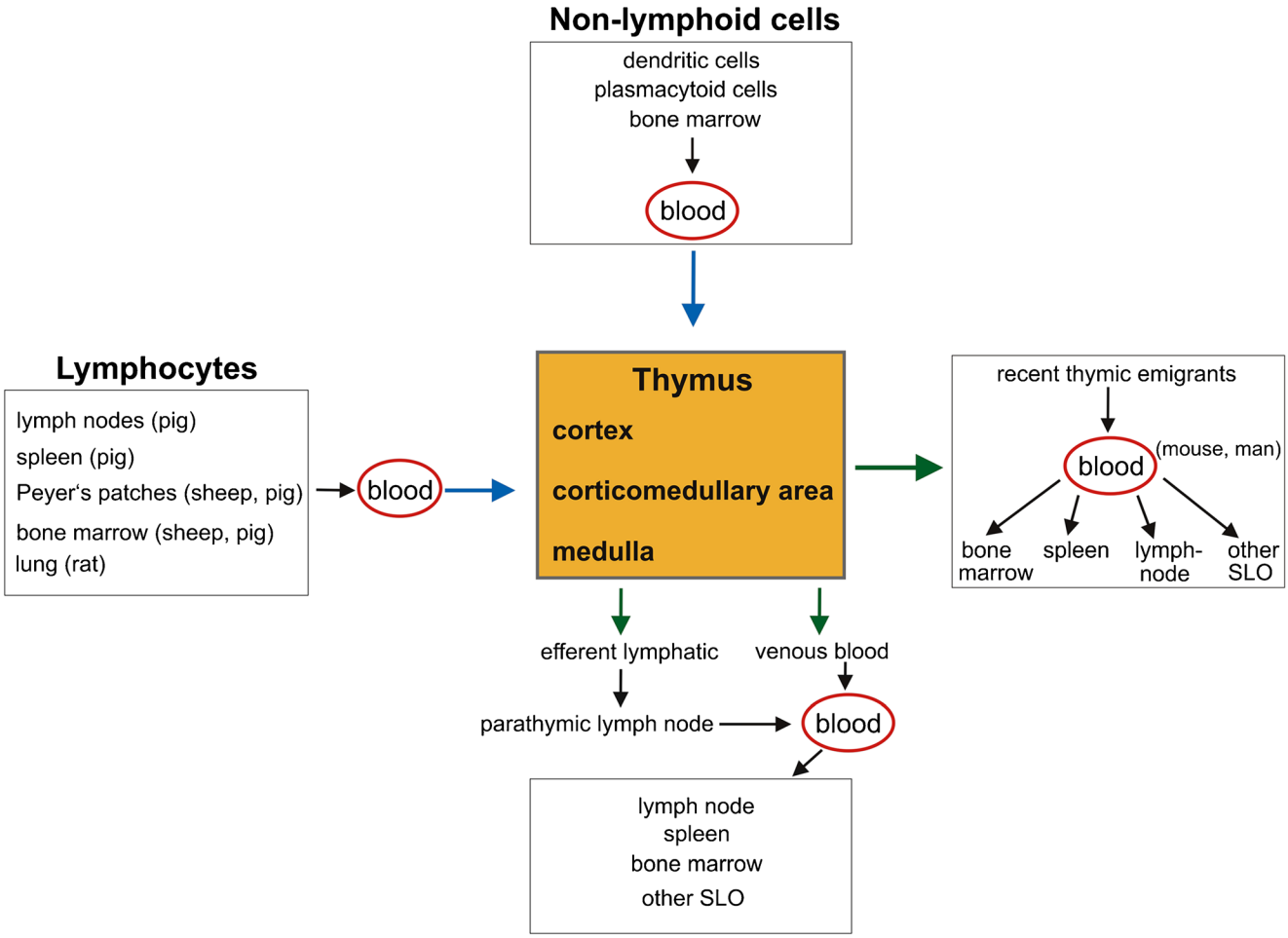

Fig. 3 Overview of the role of the thymus in lymphocyte migration. SLO, secondary lymphoid organs

thymus, which are essential for the thymic egression of lymphocytes (see also Aili et al 2018; Miyasaka et al. 2016).

B lymphocytes in the thymus

In contrast to many statements, the thymus is also the home of some developing B cells and also very few plasma cells in several species (Cukrowska et al. 1996). In pigs, mature B cells are rare in the thymus (less than 5%). These cells are also involved in Ig synthesis (Sinkora and Butler 2009). In the adult rat thymus, only 0.14% were B cells; however, they proliferated at a higher rate than in peripheral organs (Pabst et al. 1989). There are no data on B cells in the human thymus. In lambs, the cervical thymus of one side was labeled by an extracorporeal perfusion system using a fluorescent dye and lymphoid cells leaving the thymus via lymphatics and thymic vein were characterized by surface markers and quantified. Only a small proportion of all newly produced lymphocytes leaves the thymus in contrast to the data in the mouse (Miyasaka et al. 1990b).

Conclusion

The thymus is integrated in the migration of mature lymphocytes from peripheral lymphoid organs (Fig. 3). B lymphocytes are also included, as documented in larger animals like pigs and sheep. The functional and clinical relevance has to be studied in more detail.

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Compliance with ethical standards

Conflict of interest The author declares that there is no conflict of interest.

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