Peer reviewed article

Control of immune responses by scavenger liver endothelial cells

Percy A. Knolle, Andreas Limmer

Institute for Molecular Medicine and Experimental Immunology, University of Bonn, Germany

Summary

The liver appears to be an organ favoring the induction of immune tolerance rather than immunity. Among the hepatic cell populations possibly involved in regulation of immune responses, liver sinusoidal endothelial cells (LSEC) are well suited to fulfill this role. LSEC are resident cells lining the hepatic sinusoidal wall and therefore are in intimate contact with leukocytes passing through the liver. They are equipped with numerous scavenger receptors rendering antigen-uptake in these cells extremely efficient. Antigen processing and MHC-restricted presentation of exogenous antigens for CD4 as well as CD8 T cells occurs equally with high efficiency. Importantly, CD4 and CD8 T cells that engaged in cognate interaction with LSEC have a tolerant phenotype. Thus LSEC contribute an important immune function to the liver: control of the immune response against circulating soluble antigens.

Key words: immune response; liver sinusoidal endothelial cells; tolerance

Introduction

The "decision" whether to mount immunity or tolerance towards an antigen is an important issue for the immune system. While immunity needs to be generated against infectious pathogens, immune responses against innocuous or self-antigens are not desirable and need to be prevented. Deletion of auto-reactive T cells in the thymus is one mechanism assuring absence of auto-aggression. However, not all auto-reactive cells are eliminated in the thymus, thus requiring mechanisms of induction of immune tolerance in the periphery. Dendritic cells are the key-players in the induction of immunity. They perform their immuno-stimulatory function in lymphatic tissue, which provides an optimised anatomical platform for interaction between antigen-presenting dendritic cells and lymphocytes. Antigen-presenting cell populations involved in induction of immune tolerance appear to be more diverse. Moreover, different organs have different requirements with regard to control of immune responses which may be reflected by different, perhaps even organ-resident cell types that influence the immune response locally.

The liver seems to favour the induction of immune tolerance rather than immunity. A number of observations demonstrate that antigen specific immune tolerance is the result of presentation of antigen within the liver. Firstly, allogeneic liver organ transplants are often well accepted by the recipient [1, 2] and lead to tolerance to further organ transplants from the same donor but not to third party grafts ("split tolerance") [3]. Secondly, portal venous drainage of an allogeneic organ transplant [4–6] as well as pre-transplant portal venous injection of donor leukocytes lead to increased graft acceptance [7, 8]. Further evidence for tolerance induction after application of antigen via the portal route comes from the observation that porto-systemic shunting results in loss of tolerance towards orally ingested antigens [9]. It is important to note, that clonal elimination of antigen-reactive T cells or immune ignorance are not the mechanisms leading to immune tolerance in these situations, because adoptive transfer of lymphocytes from animals treated as described above again leads to development of antigen specific immune tolerance in the recipient [10]. Still, contribution of clonal elimination by apoptosis in the liver to hepatic tolerance induction can not be entirely excluded.

This work was supported by grants from the Volkswagenstiftung and Deutsche Forschungsgemeinschaft.

Contact of liver cells with passenger leukocytes

The liver has a dual blood supply with portal venous blood draining together with hepatic arterial blood into the hepatic sinusoids. Leukocytes entering the liver with the bloodstream pass through the hepatic sinusoids and are obviously in direct contact with sinusoidal cells, the liver sinusoidal endothelial cells (LSEC) and the Kupffer cells. Intensive contact between sinusoidal cells and passenger leukocytes can occur for a number of reasons. The sinusoids are on average 5–7 μm wide forcing leukocytes to contact sinusoidal cells [11]; leukocyte margination occurs in hepatic sinusoids and together with intermittent blood flow and low perfusion pressure, contact of leukocytes with sinusoidal cells is enforced [12, 13]; leukocyte adhesion to LSEC does not require expression of selectins [14] but depends on constitutively expressed cellular adhesion molecules, such as CD54, CD106 and others.

Due to the fenestrated hepatic endothelium it was believed that hepatocytes are easily accessible for leukocytes passing through the liver and that this leads to direct and continuous immune surveillance of hepatocytes [15]. However, a recent publication has clearly shown that antigen presented exclusively by hepatocytes is not recognised by T cells specific for the antigen during their passage through the liver [16]. Moreover, it was demonstrated that blood borne particles ≥12 nm in diameter do not diffuse freely through the endothelial fenestrae (approx. diameter 100 nm) and do not have direct contact with receptors on the hepatocyte surface [17]. These studies reveal LSEC as a barrier that protects hepatocytes from direct contact with passenger leukocytes. Thus, Kupffer cells and especially LSEC, through repeated contact with passenger leukocytes, encounter all the prerequisites for influencing the immunological function of T cells. This becomes even more important as the entire blood volume is passed through the liver more than 300 times per day.

Immune phenotype of liver sinusoidal endothelial cells

Table 1

Phenotype of liver sinusoidal endothelial cells.

Molecule	LSEC	EC
CD54	++	++
CD102	+	+
CD106	+	+
CD62P	(+)	+
CD31	++	++
L-SIGN	+	–
Mannose receptor	++	–
Scavenger receptor	++	–
Toll like receptor 4	+	–
CD14	+	n.d.
CD32	+	n.d.
CD36	+	n.d.
MHC class I	++	+
MHC class II	+	–
CD80	+	–
CD86	+	–
CD40	++	+
CD4	+	–
CD11c	+	–
CD95	+	+
CD95L	+	n.d.
TRAIL	+	n.d.
Membrane TNFα	+	n.d.

++ strong expression, + expression, (+) faint expression, ø absent expression)

LSEC are microvascular endothelial cells and display a unique phenotype compared to macrovascular endothelial cells and microvascular endothelial cells from other organs. Table I summarises the molecules known to be expressed on LSEC as detected by flow cytometry, by functional *in vitro* assays or by immuno-histochemistry in liver sections. LSEC constitutively express molecules necessary for establishment of interaction with leukocytes, giving supportive evidence for the observation of constitutive leukocyte adhesion to LSEC *in vivo*. LSEC further express several pattern recognition receptors that enable them to act as scavenger cells (see below). Moreover, LSEC constitutively express co-stimulatory molecules (CD80, CD86, CD40) as well as MHC class I and II molecules necessary for presentation of antigen to T cells. Finally, LSEC express a number of molecules that are typically found on cells of myeloid origin such as CD4 and CD11c. It can be concluded from these observations that LSEC are endowed with a set of surface molecules that renders them competent for both, recruitment of T cells and antigen presentation to T cells.

However, LSEC in adult mice are not derived from the bone marrow (A. Limmer and P. Knolle, unpublished observation) whereas Kupffer cells continuously repopulate from the bone marrow [18]. It rather seems that LSEC regenerate from cells resident to the liver, which is not surprising given the haematopoietic function of the liver early in life and the capacity of transplanted livers to establish micro-chimerism [19, 20].

Scavenger function of liver LSEC

Receptor-mediated endocytosis of macromolecules is well studied in LSEC and known to occur with very high efficiency. The expression of pattern recognition receptors by LSEC, such as the mannose and scavenger receptors, points to a contribution of these cells to clearing sinusoidal blood of macromolecules that are recognised as "unwanted", eg, by their glycosylation pattern or even low affinity receptor binding. LSEC are further endowed with the capacity to phagocytose particles up to 200 nm in diameter [21]. These findings imply that LSEC are part of the non-specific arm of the immune system.

However, there appears to be a conceptual problem with highly efficient antigen uptake by LSEC. What is the fate of ingested material? Are antigens only processed and presented by LSEC or are endocytosed antigens transported through the LSEC and handed over to hepatocytes? It has been shown that delivery of endocytosed antigens to the lysosomal compartment occurs comparatively slowly and in an inefficient way in LSEC [22]. How can LSEC combine efficient uptake with slow removal of ingested material via the lysosomal pathway? Tavassoli et al. have described that LSEC have the ability to transport endocytosed molecules in a vector fashion from the luminal side towards the hepatocytes and thereby accomplish transcytosis [23, 24]. This implies that clearance function of the liver is supported by efficient uptake of macromolecules by LSEC from the blood and shuttling of these molecules to the hepatocytes for excretion via the bile or metabolisation. Although difficult to prove, transcytotic transport in LSEC has been assumed by independent groups to play a role in physiological clearance of macromolecules [17] as well as during the early steps of infection with hepatotropic viruses. However, we have evidence that antigen uptake by LSEC *in vivo* is accompanied by antigen presentation to T cells *in vivo* suggesting that at least part of the endocytosed antigen is used for presentation (see below).

LSEC present antigen on MHC II molecules to $CD4^+$ T cells

Given the constitutive expression of costimulatory and MHC class II molecules together with the efficient uptake of antigen it is not surprising that LSEC present soluble antigens to $CD4^+$ T cells. It is important to note that LSEC do not require a maturation step to induce antigen specific proliferation and cytokine release by antigen-specific $CD4^+$ T cells. LSEC are almost as efficient as Kupffer and bone marrow derived "professional" antigen presenting cells with regard to stimulation of $CD4^+$ T cells [25]. This finding raises the question of how hepatic immune tolerance relates to the unique function of LSEC to present antigen efficiently to $CD4^+$ T cells. Antigen presentation by LSEC to $CD4^+$ T cells is stringently controlled by mediators present in the local microenvironment such as PGE_2 and IL-10 [29], which are expressed by other hepatic cell populations, eg, Kupffer cells [26–28].

However, not only soluble mediators released by neighbouring Kupffer cells but also portal blood constituents directly influence antigen presentation by LSEC. Endotoxin is physiologically present in portal venous blood at 100 pg/ml to 1 ng/ml [30] and is cleared by Kupffer cells as well as LSEC from the blood [31]. Pre-treatment of LSEC with endotoxin (in physiological concentrations) reduced antigen presentation to $CD4^+$ T cells considerably [32].

Moreover, LSEC like dendritic cells can prime naïve $CD4^+$ T cells, but unlike dendritic cells LSEC induce immune tolerance in $CD4^+$ T cells upon cognate interaction. $CD4^+$ T cells stimulated by antigen presenting LSEC show a regulatory phenotype characterised by the expression of IL-4 and IL-10 upon antigen-specific restimulation [33]. Again, LSEC do not require maturation in order to perform their antigen presenting function for naïve CD4 T cells and thus are clearly different from microvascular endothelial cells from other organs [33–38]. We conclude that LSEC do not promote differentiation of naive $CD4^+$ T cells towards T_{H1} but rather induce regulatory T cells and thereby contribute to induction of hepatic immune tolerance.

Presentation of exogenous antigens on MHC class I molecules to $CD8^+$ T cells

Functioning as scavenger cells for blood borne molecules LSEC are in a strategic position to present systemically distributed antigens to T cells. While MHC class II restricted presentation of exogenous antigens to CD4 T cells is well accepted, MHC class I restricted presentation of endocytosed antigens to CD8 T cells was believed to be restricted to dendritic cells and to occur only in

certain conditions. Presentation of peptides on MHC class I molecules was thought to be restricted to endogenous proteins expressed in the same cell that presented the antigen to $CD8^+$ T cells. $CD8^+$ cytotoxic T cells are of crucial importance for immunity to infection with intracellular pathogens and tumours. $CD8^+$ T cells continuously patrol the body to screen the cell surface for presentation of foreign antigens and eliminate parenchymal cells presenting cognate antigens. Thus, induction of hepatic immune tolerance needs to include tolerance induction in $CD8^+$ T cells.

Initially identified by Bevan [39] during the last few years it has become increasingly evident that the presentation of exogenous antigen to $CD8^+$ T cells can occur in specialised subpopulations of myeloid antigen presenting cells such as macrophages and dendritic cells [40]. These cells take up exogenous antigens by phagocytosis or receptor-mediated endocytosis, respectively, and process exogenous protein for MHC class I presentation mainly via the same pathway used for presentation of endogenous proteins, critically depending on the proteasome and TAP (transporter associated with antigen processing) [40]. This process is termed cross-presentation and allows priming of $CD8^+$ T cells and induction of a protective cell mediated immune response against pathogens even in the absence of productive infection of the antigen presenting cell [41].

As LSEC are so efficient in antigen uptake we wondered whether they were capable of cross-presenting exogenous antigen to $CD8^+$ T cells. Using a monoclonal antibody that recognises a specific peptide (ova 257–64 SIINFEKL) after processing of antigen (ovalbumin) on a specific MHC class I molecule (H2–K^b), we have shown that LSEC as an homogenous cell population processed and presented SIINFEKL on K^b molecules after receptor-mediated uptake of ovalbumin. Moreover, LSEC cross-presented exogenous antigen to SIINFEKL-specific $CD8^+$ T cells and induced cytokine release (IL-2) in ova-specific $CD8^+$ T cells. Antigen uptake is a prerequisite but alone is not sufficient to endow a cell with the ability to cross-present antigen to $CD8^+$ T cells as B cells do not cross-present albeit efficient uptake of ovalbumin and K^b surface expression [42].

Our findings imply that cross-presentation is not restricted to myeloid cells but can occur outside lymphatic tissue in the liver by organ resident LSEC. To prove this finding we established a new experimental system where LSEC are adoptively transferred into syngeneic littermates and orthotopically implanted in the hepatic sinusoids. When ovalbumin loaded K^b LSEC are implanted into mutant K^{bm1} mice unable to present SIINFEKL on K^b, the transferred LSEC are the only cell population able to cross-present ovalbumin to SIINFEKL-specific K^b-restricted $CD8^+$ T cells. In this system, we were able to show that LSEC cross-presented ovalbumin to ovalbumin-specific $CD8^+$ T cells *in vivo*. T cells re-isolated from liver and from peripheral blood but to a lesser degree from spleen and lymph nodes showed signs of activation suggesting that cross-presentation occurred in the liver but not in other organs [42].

Induction of CD8 T cells tolerance by cross-priming LSEC

LSEC not only cross-present antigen to a T cell hybridoma, but are also able to induce cytokine expression and proliferation in naive $CD8^+$ T cell receptor transgenic T cells. The outcome of T cell priming by cross-presenting LSEC is clearly distinct from T cell priming by splenocytes or bone marrow derived antigen-presenting cells. T cells primed by LSEC lose their ability to express cytokines such as IFNγ and IL-2 and do not exhibit specific cytotoxicity any more compared to T cells primed by conventional antigen presenting cells. *In vivo*, LSEC induce antigen-specific (ovalbumin) $CD8^+$ T cell tolerance as shown by inability of $CD8^+$ T cells to reject an s.c. implanted syngeneic tumour expressing ovalbumin [42].

Several laboratories have shown that myeloid cross-presenting cells are required for induction of a protective $CD8^+$ immune response to circulating soluble antigens. Our experiments add a new perspective to the way the organism co-ordinates the immune response to soluble antigens. $CD8^+$ T cell immunity has not been achieved to soluble antigen unless antigens were added together with an adjuvant or were applied as particulate antigen. It has further remained unknown why intravenous injection of antigens results in specific immune tolerance. Efficient uptake and cross-presentation of soluble antigen by LSEC accompanied by induction of immune tolerance in $CD4^+$ as well as $CD8^+$ T cells may provide an explanation to these questions. Several of our experimental findings further support this view: (i) LSEC are 1.00 to 10.000 fold more efficient in cross-presentation of soluble antigen than has been reported for dendritic cells and macrophages, (ii) antigen uptake and subsequent cross-presentation is not enhanced by incorporation of antigen into immune complexes, whereas this process increases cross-presentation in dendritic by a factor of 1000, (iii) cross-presentation by LSEC occurs within 30 minutes after antigen contact.

LSEC-mediated tolerance in $CD4^+$ and $CD8^+$ T cells may protect the organism from an unwanted immune reaction towards antigens derived from the gastrointestinal tract and confine at the same time immune reactions once antigen circulates in the systemic blood pool. Antigenaemia is dangerous for the integrity of the organism as

widespread activation of the immune system, as observed during sepsis, leads to breakdown of immunological function and can cause severe damage to vital organs by non-specific immune amplification mechanisms. It is therefore mandatory for immune homeostasis to restrict immune responses to antigen, which is present systemically. This scenario has been observed *in vivo* when functionally inactive virus specific T cells were detected exclusively in the liver of mice three days after onset of symptomatic influenza-induced pneumonia and after viraemia had occurred [43]. In addition to rendering T cells tolerant, clonal eliminiation by apoptosis may be operative in the liver as has been suggested by N. Crispe et al. Also here, sinusoidal cells appear to participate in this process as LSEC express death-inducing receptors (see Table I) and both LSEC and Kupffer cells induce apoptosis in susceptible T cells [45, 46]. It is intriguing to speculate that so far aetiologically undefined conditions with immune mediated hepatic damage, such as certain forms of autoimmune hepatitis, may be related to inefficient induction of immune tolerance through LSEC.

In contrast, presence of a "danger" signal such as an adjuvant, antigen in particulate form (resembling a pathogenic microorganism for phagocytosing cells) and coating of antigen with antibodies will result in improved antigen uptake and maturation of conventional myeloid antigen presenting cells shifting the immune response towards induction of $CD8^+$ T cell immunity. Therefore, antigen formulation itself may, through antigen presentation on different antigen presenting cells, contribute to the "decision" whether immunity or tolerance is induced.

Conclusions

Two immune tolerance phenomena have been observed, but have not been sufficiently explained so far: hepatic tolerogenicity and induction of immune tolerance towards soluble systemically circulating antigens. Experimental data suggest that liver sinusoidal endothelial cells (LSEC) may represent a link between both phenomena for the following reasons: (i) LSEC are resident cells, lining liver sinusoids and have therefore direct contact to blood borne antigens, (ii) LSEC express various scavenging receptors rendering them very efficient in antigen uptake, (iii) LSEC express all known costimulatory molecules needed to stimulate T cells, (iv) LSEC can prime naive $CD4^+$ T cells to become regulatory T cells, (v) LSEC cross-present exogenous antigens and render $CD8^+$ T cells tolerant.

According to our model, the liver is "in charge" of tolerance induction towards antigens, which circulate in the blood or are released from hepatocytes. Among these antigens are self- and food antigens, against which an immune response would be deleterious. In contrast, immunity is observed when antigens are present locally in the periphery (ie, skin), when present only for a short time or when presented in the context of a danger signal (with adjuvant). The "decision" whether immunity or tolerance is induced, seems to require the participation of two different antigen presenting cell populations and different microenvironments. While induction of immunity requires activated dendritic cells, which are the best known immune stimulatory cells, and the unique well structured microenvironment of secondary lymphoid tissue, induction of immune tolerance in the liver appears to involve LSEC combined with the unique hepatic microenvironment.

Although in many aspects LSEC and dendritic cells share a common phenotype and the capacity to present exogenous antigen via MHC class I and II, there are important differences between LSEC and dendritic cells that may account for their different functional activity with regard to immune regulation. However, the molecular mechanisms underlying the "decision" whether immunity or immune tolerance is induced, still remain unknown. We conclude that LSEC represent a new type of antigen presenting cell that is organ-resident, does not require functional maturation and mediates down regulation rather than induction of immunity in the context of the local microenvironment. Our concept of local hepatic immune regulation explains a so far poorly recognised function of the liver: control of immunity by local induction of immune tolerance through tolerogenic organ-resident antigen-presenting cells.

Correspondence:
Institute for Molecular Medicine
and Experimental Immunology
University of Bonn
Sigmund-Freud-Str. 25
D-53105 Bonn
Germany
E-Mail: Percy.Knolle@ukb.uni-bonn.de,
Andreas.Limmer@ukb.uni-bonn.de

References

1 Cantor H, Dumont A. Hepatic suppression of sensitization to antigen absorbed into the portal system. Nature 1967;215:744.

2 Calne RY. Induction of immunological tolerance by porcine liver allografts. Nature 1969;223:472–6.

3 Dahmen U, Qian S, Rao AS, Demetris AJ, Fu F, Sun H, et al. Split tolerance induced by orthotopic liver transplantation in mice. Transplantation 1994;58:1–8.
4 Barker CF, Corriere JN, Jr. Canine renal homotransplantation with venous drainage via the portal vein. Ann Surg 1967;165: 279–82.
5 Boeckx W, Sobis H, Lacquet A, Gruwez J, Vandeputte M. Prolongation of allogeneic heart graft survival in the rat after implantation on portal vein. Transplantation 1975;19:145–9.
6 Gorczynski RM, Chan Z, Chung S, Cohen Z, Levy G, Sullivan B, et al. Prolongation of rat small bowel or renal allograft survival by pretransplant transfusion and/or by varying the route of allograft venous drainage. Transplantation 1994;58:816–20.
7 May AG, Bauer S, Leddy JP, Panner B, Vaughan J, Russell PS. Survival of allografts after hepatic portal venous administration of specific transplantation antigen. Ann Surg 1969;170:824–32.
8 Gorczynski RM. Immunosuppression induced by hepatic portal venous immunization spares reactivity in IL-4 producing T lymphocytes. Immunol Lett 1992;33:67–77.
9 Yang R, Liu Q, Grosfeld JL, Pescovitz MD. Intestinal venous drainage through the liver is a prerequisite for oral tolerance induction. J Pediatr Surg 1994;29:1145–8.
10 Gorczynski RM. Adoptive transfer of unresponsiveness to allogeneic skin grafts with hepatic gamma delta + T cells. Immunology 1994;81:27–35.
11 Wisse E, De Zanger RB, Charels K, Van Der Smissen P, McCuskey RS. The liver sieve: considerations concerning the structure and function of endothelial fenestrae, the sinusoidal wall and the space of Disse. Hepatology 1985;5:683–92.
12 Granger DN. Cell adhesion and migration. II. Leukocyte-endothelial cell adhesion in the digestive system. Am J Physiol 1997;273(5 Pt 1):G982–6.
13 MacPhee PJ, Schmidt EE, Groom AC. Intermittence of blood flow in liver sinusoids, studied by high- resolution in vivo microscopy. Am J Physiol 1995:G692–8.
14 Wong J, Johnston B, Lee SS, Bullard DC, Smith CW, Beaudet AL, et al. A minimal role for selectins in the recruitment of leukocytes into the inflamed liver microvasculature. J Clin Invest 1997;99:2782–90.
15 Ando K, Guidotti LG, Cerny A, Ishikawa T, Chisari FV. CTL access to tissue antigen is restricted in vivo. J Immunol 1994; 153:482–8.
16 Limmer A, Sacher T, Alferink J, Kretschmar M, Schonrich G, Nichterlein T, et al. Failure to induce organ-specific autoimmunity by breaking of tolerance: importance of the microenvironment. Eur J Immunol 1998;28:2395–406.
17 Kempka G, Kolb-Bachofen V. Binding, uptake, and transcytosis of ligands for mannose-specific receptors in rat liver: an electron microscopic study. Exp Cell Res 1988;176(1):38–48.
18 Yamamoto T, Naito M, Moriyama H, Umezu H, Matsuo H, Kiwada H, et al. Repopulation of murine Kupffer cells after intravenous administration of liposome-encapsulated dichloromethylene diphosphonate. Am J Pathol 1996;149:1271–86.
19 Starzl TE, Demetris AJ, Trucco M, Ramos H, Zeevi A, Rudert WA, et al. Systemic chimerism in human female recipients of male livers. Lancet 1992;340(8824):876–7.
20 Lu L, Rudert WA, Qian S, McCaslin D, Fu F, Rao AS, et al. Growth of donor-derived dendritic cells from the bone marrow of murine liver allograft recipients in response to granulocyte/macrophage colony-stimulating factor. J Exp Med 1995; 182:379–87.
21 Steffan AM, Gendrault JL, McCuskey RS, McCuskey PA, Kirn A. Phagocytosis, an unrecognized property of murine endothelial liver cells. Hepatology 1986;6:830–6.
22 Hellevik T, Martinez I, Olsen R, Toh B, Webster P, Smedsrod B. Transport of residual endocytosed products into terminal lysosomes occurs slowly in rat liver endothelial cells. Hepatology 1998;28:1378–89.
23 Tavassoli M, Kishimoto T, Soda R, Kataoka M, Harjes K. Liver endothelium mediates the uptake of iron-transferrin complex by hepatocytes. Exp Cell Res 1986;165:369–79.
24 Tavassoli M, Kishimoto T, Kataoka M. Liver endothelium mediates the hepatocyte's uptake of ceruloplasmin. J Cell Biol 1986;102:1298–303.
25 Lohse AW, Knolle PA, Bilo K, Uhrig A, Waldmann C, Ibe M, et al. Antigen-presenting function and B7 expression of murine sinusoidal endothelial cells and Kupffer cells. Gastroenterology 1996;110:1175–81.
26 Jacob AI, Goldberg PK, Bloom N, Degenshein GA, Kozinn PJ. Endotoxin and bacteria in portal blood. Gastroenterology 1977;72:1268–70.
27 Knolle P, Schlaak J, Uhrig A, Kempf P, Meyer zum Buschenfelde KH, Gerken G. Human Kupffer cells secrete IL-10 in response to lipopolysaccharide (LPS) challenge. J Hepatol 1995;22:226–9.
28 Knolle PA, Uhrig A, Protzer U, Trippler M, Duchmann R, Meyer zum Buschenfelde KH, et al. Interleukin-10 expression is autoregulated at the transcriptional level in human and murine Kupffer cells. Hepatology 1998;27:93–9.
29 Knolle PA, Uhrig A, Hegenbarth S, Loser E, Schmitt E, Gerken G, et al. IL-10 down-regulates T cell activation by antigen-presenting liver sinusoidal endothelial cells through decreased antigen uptake via the mannose receptor and lowered surface expression of accessory molecules. Clin Exp Immunol 1998; 114:427–33.
30 Lumsden AB, Henderson JM, Kutner MH. Endotoxin levels measured by a chromogenic assay in portal, hepatic and peripheral venous blood in patients with cirrhosis. Hepatology 1988;8:232–6.
31 Catala M, Anton A, Portoles MT. Characterization of the simultaneous binding of Escherichia coli endotoxin to Kupffer and endothelial liver cells by flow cytometry. Cytometry 1999; 36:123–30.
32 Knolle PA, Germann T, Treichel U, Uhrig A, Schmitt E, Hegenbarth S, et al. Endotoxin Down-Regulates T Cell Activation by Antigen-Presenting Liver Sinusoidal Endothelial Cells. J Immunol 1999;162:1401–7.
33 Knolle P, Schmitt E, Jin S, Germann T, Duchmann R, Hegenbarth S, et al. Liver sinusoidal endothelial cells can prime naive $CD4^+$ T cells in the absence of IL-12 and induce IL-4 production in primed $CD4^+$ T cells: Implications for tolerance induction in the liver. Gastroenterology 1999;116:1428–40.
34 Briscoe DM, Henault LE, Geehan C, Alexander SI, Lichtman AH. Human endothelial cell costimulation of T cell IFN-gamma production. J Immunol 1997;159:3247–56.
35 Epperson DE, Pober JS. Antigen-presenting function of human endothelial cells. Direct activation of resting CD8 T cells. J Immunol 1994;153:5402–12.
36 Cunningham AC, Zhang JG, Moy JV, Ali S, Kirby JA. A comparison of the antigen-presenting capabilities of class II MHC-expressing human lung epithelial and endothelial cells. Immunology 1997;91:458–63.
37 Marelli-Berg FM, Hargreaves RE, Carmichael P, Dorling A, Lombardi G, Lechler RI. Major histocompatibility complex class II-expressing endothelial cells induce allospecific nonresponsiveness in naive T cells. J Exp Med 1996;183:1603–12.
38 Perez VL, Henault L, Lichtman AH. Endothelial antigen presentation: stimulation of previously activated but not naive TCR-transgenic mouse T cells. Cell Immunol 1998;189:31–40.
39 Moore MW, Carbone FR, Bevan MJ. Introduction of soluble protein into the class I pathway of antigen processing and presentation. Cell 1988;54:777–85.
40 York IA, Rock KL. Antigen processing and presentation by the class I major histocompatibility complex. Annu Rev Immunol 1996;14:369–96.
41 Sigal LJ, Crotty S, Andino R, Rock KL. Cytotoxic T-cell immunity to virus-infected non-haematopoietic cells requires presentation of exogenous antigen. Nature 1999;398(6722): 77–80.
42 Limmer A, Ohl J, Ljunggren HG, Reiss Y, Groettrup M, Momburg F, Arnold B, Knolle PA. Efficient presentation of exogenous antigen by liver endothelial cells to $CD8^+$ T cells results in antigen-specific T-cell tolerance. Nature Medicine 2000;6: 1348–54.
43 Belz G, Altman J, Doherty P. Characteristics of virus-specific $CD8^+$ T cells in the liver during the control and resolution phases of influenza pneumonia. Proc Natl Acad Sci USA 1998; 95:13812–7.
44 Huang L, Soldevila G, Leeker M, Flavell R, Crispe IN. The liver eliminates T cells undergoing antigen-triggered apoptosis in vivo. Immunity 1994;1:741–9.
45 Müschen M, Warskulat U, Douillard P, Gilbert E, Häussinger D. Regulation of CD95 (APO-1/Fas) receptor and ligand expression by lipopolysaccharide and dexamethasone in parenchymal and nonparenchymal rat liver cells. Hepatology 1998;27: 200–8.
46 Müschen M, Warskulat U, Peters-Regehr T, Bode JG, Kubitz R, Häussinger D. Involvement of CD95 (Apo-1/Fas) ligand expressed by rat Kupffer cells in hepatic immunoregulation. Gastroenterology 1999;116:666–77.

Swiss
Medical Weekly

Official journal of
the Swiss Society of Infectious disease
the Swiss Society of Internal Medicine
the Swiss Respiratory Society

The many reasons why you should choose SMW to publish your research

What Swiss Medical Weekly has to offer:

- SMW's impact factor has been steadily rising, to the current 1.537
- Open access to the publication via the Internet, therefore wide audience and impact
- Rapid listing in Medline
- LinkOut-button from PubMed with link to the full text website http://www.smw.ch (direct link from each SMW record in PubMed)

- No-nonsense submission – you submit a single copy of your manuscript by e-mail attachment
- Peer review based on a broad spectrum of international academic referees
- Assistance of our professional statistician for every article with statistical analyses

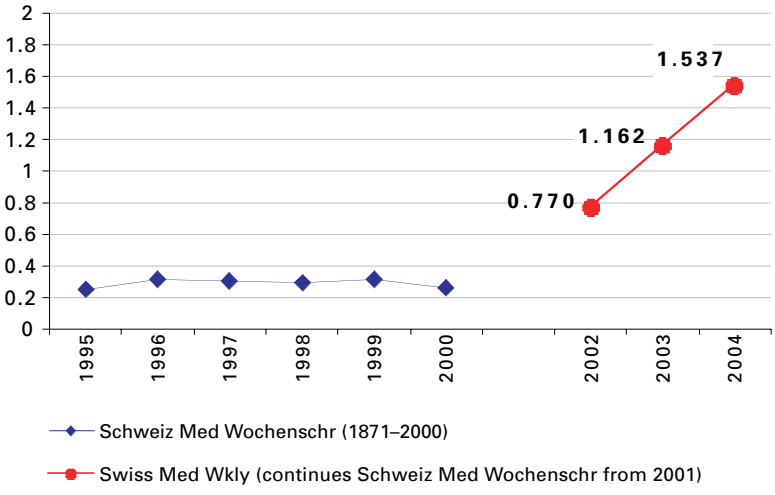
- Fast peer review, by e-mail exchange with the referees
- Prompt decisions based on weekly conferences of the Editorial Board
- Prompt notification on the status of your manuscript by e-mail
- Professional English copy editing
- No page charges and attractive colour offprints at no extra cost

We evaluate manuscripts of broad clinical interest from all specialities, including experimental medicine and clinical investigation.

We look forward to receiving your paper!

Guidelines for authors:
http://www.smw.ch/set_authors.html

Impact factor Swiss Medical Weekly



EMH FMH Schwabe
Editores Medicorum Helveticorum

All manuscripts should be sent in electronic form, to:

EMH Swiss Medical Publishers Ltd.
SMW Editorial Secretariat
Farnsburgerstrasse 8
CH-4132 Muttenz

Manuscripts: submission@smw.ch
Letters to the editor: letters@smw.ch
Editorial Board: red@smw.ch
Internet: http://www.smw.ch