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Expression of oncostatin M and its receptors in normal and cirrhotic human liver

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***Background/Aims*: In the cirrhotic liver, gene expression of the multifunctional cytokine oncostatin M (OSM) is up-regulated, but its cellular origin is unknown. Therefore, we investigated the expression of OSM protein and its specific receptor subunits, OSMRβ and LIFRβ in normal and cirrhotic human liver using immunohistochemical and Western blot analysis.**

***Results*: OSM protein was expressed in Kupffer cells, variably in normal liver but consistently in cirrhosis. OSMRβ was expressed at low level in hepatocytes of all normal livers examined, but in no cirrhotic sample. In contrast, LIFRβ receptor was expressed weakly in normal livers, but much more intensely in cirrhosis, in reactive ductules, bile duct epithelial cells and perisinusoidal areas. Double immunostaining showed co-localization of LIFRβ with cytokeratin 7, proliferating cell nuclear antigen (PCNA) and leukemia inhibitory factor (LIF), in bile duct epithelial cells, but not with α-smooth muscle actin, a myofibroblast marker.**

***Conclusions*: In human liver, OSM protein is expressed in Kupffer cells, variably in normals but universally in cirrhosis. The differential expression pattern of OSM and its receptors could allow for differential OSM signaling by alternative utilization of receptors to promote hepatocyte proliferation in acute injury and, with its homologue LIF, for the bile ductular reaction in cirrhosis.**

***Keywords*: Liver disease; Differential expression; Oncostatin M and oncostatin M receptors**

1. Introduction

Oncostatin M (OSM), a pleiotropic glycoprotein cytokine that was originally purified from the conditioned media of phorbol ester stimulated U937 (histocytic lymphoma) cells, is related structurally and functionally to the subfamily of hematopoietic and neurotrophic cytokines known as the interleukin-6 (IL-6) cytokine family. IL-6 cytokines include leukemia inhibitory factor (LIF), cardiotrophin-1, ciliary neurotrophic factor and IL-11 [1,2]. OSM signals through 2 receptors each of which consists of the gp130 transducing receptor subunit that is common to all members of the IL-6 family [3], coupled either to the LIF receptor β subunit (LIFRβ) to generate the type I receptor, which is shared by LIF and OSM, or to the OSM receptor β subunit (OSMRβ) to generate the OSM type II receptor [4,5]. It is highly likely that among its many other activities OSM is fibrogenic in the injured liver [6].

We have reported that hepatic stellate cells, key players in the pathogenesis of liver fibrosis and the major source of the fibrotic extracellular matrix (ECM) in liver disease, respond to stimulation by OSM in vitro by increasing their secretion of collagen and tissue inhibitor of metalloproteinase-1 (TIMP-1) [6,7]. Others have shown that OSM, but not other IL-6-type cytokines (namely LIF, G-CSF, IL-6, or IL-11), binds specifically to collagen types I, III, IV, and VI that have been immobilized on polystyrene

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or nitrocellulose [8]. The notion that OSM participates in liver fibrosis is in keeping with its fibrogenic role in other organs, specifically in tubulointerstitial fibrosis in the kidney [9], fibrosis of the lung and skin in scleroderma [10–12] and in an OSM-transgenic mouse model of pancreatic fibrosis [13].

Activated T lymphocytes and monocytes are considered to be the main sources of OSM in adults [14], but it is unknown if there are other cellular sources, especially in the injured liver. In the embryonic murine liver, OSM mRNA localizes predominantly in hematopoietic cells from 12 days post-coitum to the neonatal stage, but it is not detected in adult mouse liver tissue [15]. According to our recently published observations [16], OSM is not expressed in the liver of three adult rodent species, namely the mouse, rat and gerbil. However, there are no published data on the expression of OSM protein or its receptors in adult human liver, either normal or cirrhotic. Therefore, using immunohistochemistry we localized the expression of OSM and the specific subunits of its Type I and Type II receptors, LIFRβ and OSMRβ respectively, in normal and cirrhotic human liver.

2. Materials and methods

2.1. Tissue samples

Human liver samples were obtained from donor livers that were procured but not used for liver transplantation and from cirrhotic liver explants removed at the time of liver transplantation, according to ethical guidelines approved by the Institutional Review Board.

2.2. Protein purification and Western blot analysis

Fragments of normal and cirrhotic liver tissue were homogenized in RIPA buffer (50 mM Tris–HCl, pH 8, 150 mM sodium chloride, 0.02% sodium azide, 0.1% SDS, 1% Igepal, 0.5% sodium deoxyholate, 50 mM sodium fluoride, 10 mM sodium pyrophosphate), spun at 32,000 rpm at 4 °C for 1 h; supernatant was collected and its protein concentration was measured. Equal amounts of protein were loaded onto 7 or 10% polyacrylamide gels, transferred to nitrocellulose membrane and hybridized with rabbit polyclonal anti-human LIFRβ antibody (Santa Cruz, La Jolla, CA) in dilution 1:300, goat polyclonal anti-human OSMRβ antibody (Santa Cruz) in dilution 1:250, or mouse monoclonal anti-human β actin antibody (Sigma, St. Louis, MI) in dilution 1:2000 as the loading control. Signals were visualized with ECL Western Blotting Detection Reagents (Amersham Biosciences, Little Chalfont, UK). Western analysis of each sample was performed at least three times. For quantitative analysis gel blots were probed in turn for LIFRβ, OSMRβ and β-actin immunoreactivity. Band intensities were normalized to the band intensities of the loading control (β-actin). Since more than one gel slab was needed for the electrophoresis of all of the samples in the study, one or more samples were run repeatedly on subsequent gels, as internal standards to permit correction for differences in band intensities between gels.

2.3. Immunohistochemistry

Single antigen immunostaining. Liver samples were fixed in 4% zinc-formalin, dehydrated and embedded in paraffin, sectioned and, when needed, pretreated for 5 min with proteinase K (DakoCytomation, Carpinteria, CA) (cytokeratin 7 staining) or for 10 min at 96 °C in Antigen Unmasking Solution (Vector Laboratories, Burlingame, CA) (OSMRβ). Samples were immunostained using a horseradish peroxidase-based Vectastain ABC kit (Vector Laboratories), as described previously [17]. The following primary antibodies were used: polyclonal rabbit anti-human OSM antibody diluted 1:1000 without or with preincubation with corresponding blocking peptide (clone N-1, Santa Cruz); mouse monoclonal anti-αSMA, 1:1000 (Sigma); goat polyclonal anti-CD68, 1:500 (C-18, Santa Cruz); goat polyclonal anti-OSMRβ, 1:150 (C-20, Santa Cruz); rabbit polyclonal anti-LIFRβ, 1:400 (C-19, Santa Cruz); goat polyclonal anti-LIF, 1:250 (N-18, Santa Cruz); mouse monoclonal cytokeratin 7, 1:500 (DakoCytomation, Carpinteria, CA). The corresponding secondary horse anti-mouse and goat anti-rabbit antibodies were from the Vectastain ABC kit.

Double antigen immunostaining. Sections were incubated with rabbit anti-human LIFRβ antibodies. Secondary sheep anti-rabbit antiserum conjugated to horse raddish peroxidase (Amersham Biosciences) diluted 1:200 was applied at room temperature for 1 h. Immunoreactivity was developed by incubation with DAB substrate (Vector Laboratories). After subsequent washes, slides were immediately taken to the second round of immunostaining using the alkaline phosphatase based ELF^{R} 97 Immunohistochemistry Kit (Molecular Probes, Eugene, OR) according to the manufacturer's protocol using corresponding fluorescent substrate, or Vector Blue Alkaline Phosphatase Substrate Kit III (Vector Laboratories). Primary mouse monoclonal antiserum to α smooth muscle actin (αSMA) (Sigma) in dilution 1:500 or mouse monoclonal anti-proliferating cell nuclear antigen (PCNA) (PC-10, Santa Cruz) antibody 1:50 was employed followed by secondary biotin conjugated hourse anitiserum to mouse immunoglobulin (Vector Laboratories) diluted 1:50,000.

2.4. Microscopy

Immunostained slides were viewed with a Zeiss microscope (Analytical Imaging Facility, Bronx, NY) in bright field for single staining, and using both light and fluorescent microscopy for double staining. Substrate for alkaline phosphatase, utilized in ELF^{R}97 kit after enzymatic activation, formed a fluorescent yellow-green precipitate (excitation 350 nm), which was visualized with a standard Hoechst/DAPI filter. Slides were photographed using a SPOT digital microscope camera and corresponding software (Diagnostic Instruments Incorporated, Sterling Heights, MI).

3. Results

3.1. Expression of OSM in normal and cirrhotic human liver

Ten samples of human liver, four normal and six cirrhotic, were examined for OSM protein expression. The absence or presence of cirrhosis in the liver samples was first determined by viewing hematoxylin-eosin and trichrome stained slides (data not shown). Cirrhosis was further corroborated in sections immunostained for αSMA, a marker for activated stellate cells in fibrotic livers [18]. Two of four normal livers showed no OSM staining (Fig. 1A) and two were immunopositive (Fig. 1B and C). In contrast, all six of the cirrhotic livers analyzed showed strong expression of OSM (Fig. 1D–F). The specificity for OSM was confirmed by complete loss of staining with specific antiserum preabsorbed with OSM peptide (data not shown). OSM-positive cells, which were located in the lumen of the hepatic sinusoids, had characteristic macrophage morphology. OSM-positive cells also showed positive staining for the macrophage marker CD68 (Fig. 1G), but not for αSMA (a marker of activated hepatic stellate cells) (Fig. 1H and I), or for CD31, a marker for endothelial cells (data not shown). Thus the OSM-positive

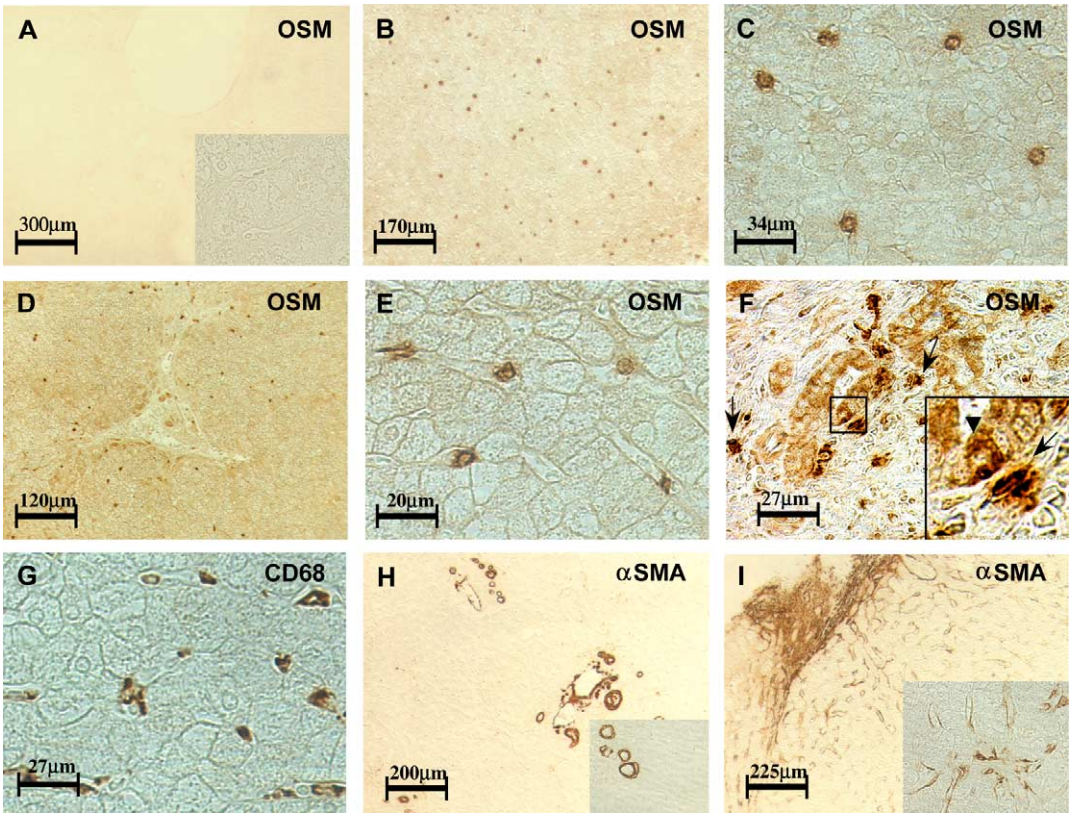

Fig. 1. Immunohistochemistry of OSM expression in the liver. (A) OSM expression in normal human liver (OSM negative), magnification 100×, inset 400×, respectively. (B, C) OSM expression in normal human liver (OSM positive). Strong expression was observed in liver macrophages, magnification 100×, 400×. (*D–F*) OSM expression in cirrhotic human liver (magnification 100×, 400×). In cirrhotic human livers, besides strong OSM immunoreactivity in macrophages in the sinusoids (D,E) and portal areas (F, arrows), there is a weak staining in bile duct cells too, which was slightly greater in the vicinity of OSM positive macrophages (Fig. 1I, inset, arrowhead). Magnification 400×: boxed area is enlarged 2.5 fold and shown in inset. (G) Expression of CD68, a macrophage marker, by Kupffer cells in human cirrhotic liver, magnification 400×. (H) Expression of αsmooth muscle actin, in the normal human liver, magnification 100×; inset (magnification 400×) shows strong expression in smooth muscle cells around blood vessels without detectable perisinusoidal αSMA staining. (I) Expression of smooth muscle α-actin, in the cirrhotic human liver, magnification 100×; inset (magnification 400×) shows perisinusoidal αSMA staining, typical of fibrosis.

cells were interpreted as Kupffer cells (liver macrophages). OSM- positive cells accounted for about a quarter (28 ± 4%) of all Kupffer cells identified. Three out of six cirrhotic samples, besides showing very strong staining in macrophages (Fig. 1F, arrows) also demonstrated a weak signal in bile duct cells, that was slightly greater in the vicinity of OSM positive macrophages (Fig. 1F, inset, arrowhead).

3.2. Expression level of OSM receptor subunits OSMRβ and LIFRβ in normal and cirrhotic human liver, by Western analysis

Using Western blot analysis, we studied the expression level of OSMRβ and LIFRβ receptor subunits in protein extracts from 5 normal and 11 cirrhotic samples (Fig. 2A and B). Low level expression of OSMRβ receptor was found in all the normal livers (5/5) but in none of the 11 cirrhotic samples. In contrast, cirrhotic samples demonstrated a higher expression level of LIFRβ than did the majority of normal livers.

3.3. Immunohistochemical localization of OSM receptor subunits expression in normal and cirrhotic human liver

Immunostaining showed OSMRβ expression exclusively in normal liver (Fig. 3A), which was localized to hepatocytes mostly in the region of hepatocyte–hepatocyte junctions (Fig. 3A, inset, arrows). Cirrhotic livers showed no detectable OSMRβ staining (Fig. 3B). In contrast, the level of LIFRβ expression was lower in most normal samples (Fig. 3C) than in cirrhotic livers (Fig. 3D–F). In the normal liver, LIFRβ showed background staining in hepatocytes and bile duct epithelium (Fig. 3C). In the cirrhotic liver, LIFRβ was localized mostly in bile duct epithelial cells and proliferating bile ductules (Fig. 3D and E), and in the perisinusoidal regions (Fig. 3F, arrowheads).

3.4. Analysis of LIFRβ expression in cirrhotic human liver

Staining for LIFRβ (Fig. 4A and E) and the bile duct marker cytokeratin 7 (Fig. 4B, C, F and G) demonstrated that most but not all reactive bile ductules (positive for

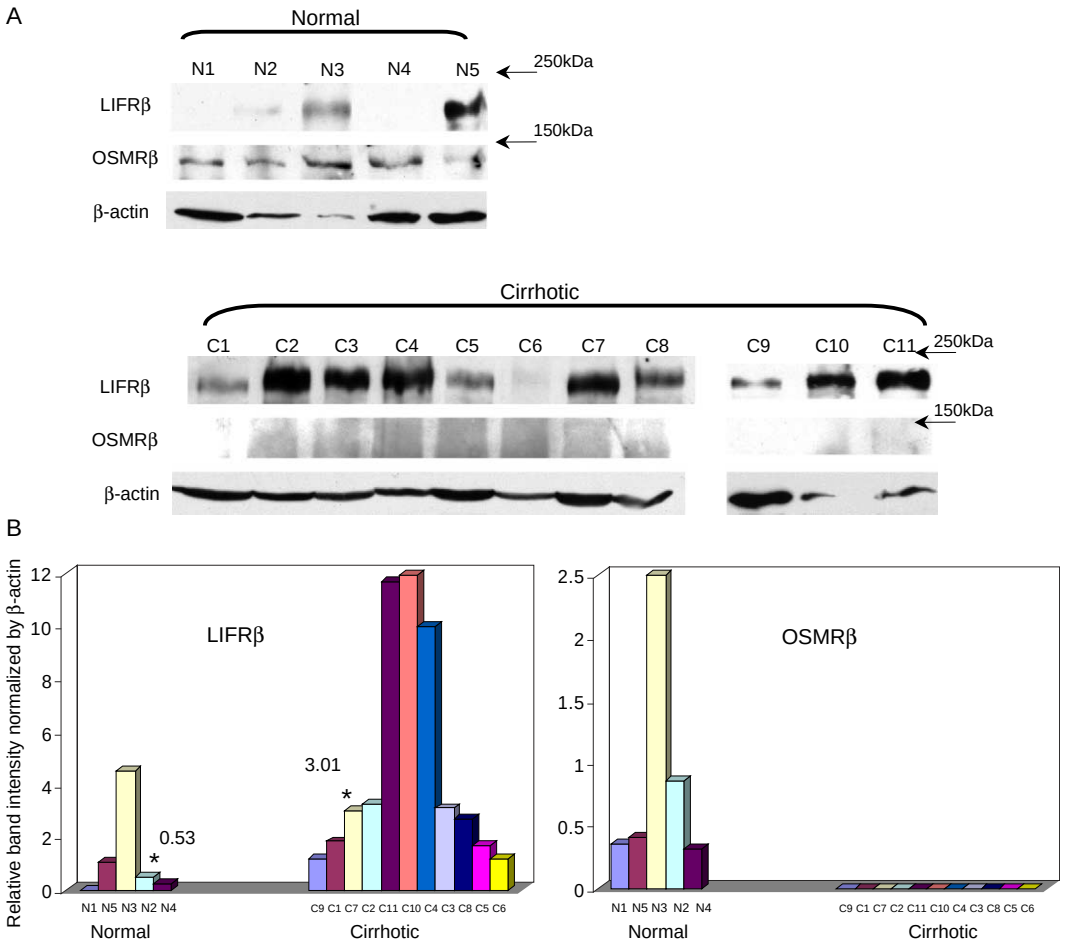

Fig. 2. Expression level of OSM receptor subunits OSMRβ and LIFRβ in normal and cirrhotic human liver, by Western analysis. (A) A representative Western blot of protein samples purified from normal (N1–N5) and cirrhotic (C1–C11) human liver hybridized with anti-LIFRβ OSMRβ, and β-actin (loading control) antibodies. Arrows indicate location of molecular weight marker bands. (B) Quantitative analysis of LIFRβand OSMRβ Western blot results. LIFRβ and OSMRβ band intensities are normalized to the corresponding band intensities of the loading control and internal control (see Section 2). Sample identification numbers are shown above each lane (A) and below each bar in (B). Asterisk indicates the median value for LIFRβ.

cytokeratin 7) express LIFRβ (Fig. 4, arrows). Double staining for LIFRβ and PCNA showed that in cirrhotic liver LIFRβ was localized to the proliferating bile duct cells (Fig. 5).

To test if the perisinusoidal localization of LIFRβ was accounted for by its expression in hepatic stellate cells, double immunostaining for LIFRβ and αSMA was performed (Fig. 6). Sections of normal liver showed low background LIFRβ immunoreactivity (Fig. 6A) and only tiny cords of αSMA positive smooth muscle cells that were seen around blood vessels (Fig. 6C). Sections of livers with cirrhosis (Fig. 6B and D) showed an extensive network of activated hepatic stellate cells reactive for αSMA. However, LIFRβ immunostaining (Fig. 6B and D) did not co-localize with these αSMA positive areas.

Since LIFRβ/gp130 receptor is shared by OSM and LIF, we examined the expression of LIF (Fig. 7). None of the four normal livers showed any bile duct LIF staining (Fig. 7A, arrows), whereas 5/6 cirrhotic samples demonstrated specific LIF expression in reactive bile ductules (Fig. 7C) which co-localized with the expression of LIFRβ (Fig. 7D and E).

4. Discussion

OSM was originally characterized by its ability to inhibit the proliferation of tumor cells. Accumulating evidence now indicates that OSM plays a role in many other biological processes, i.e. inflammation, hematopoiesis, embryonic development and tissue remodeling. That OSM has a role in liver development [19] and regeneration has already been shown [20], and it may be involved in human liver fibrosis too [6]. However, to date, there is no information on the expression of OSM protein and its receptors in either normal or cirrhotic human liver. In the current paper we report that Kupffer cells represent an important cellular source of OSM expression in the human liver. These results are consistent with the findings of Nakamura et al., [20], who performed RT-PCR on fractionated rat liver tissue and found OSM

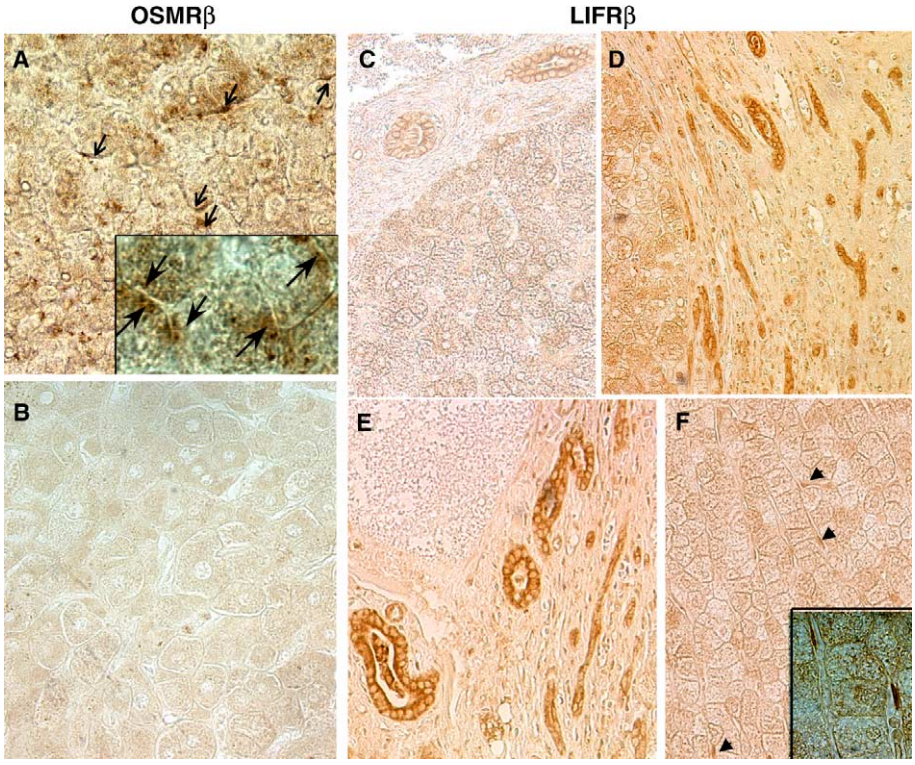

Fig. 3. Immunostaining of OSMRβ and LIFRβ receptor subunits in human liver. (A) Expression of OSMRβ was observed in hepatocytes of normal liver in the region of hepatocyte–hepatocyte junctions (inset, arrows). (B) No staining was observed in cirrhotic livers. (C) LIFRβ subunit staining in normal liver shows background expression that is evenly distributed between hepatocytes and bile duct cells. (D–F) LIFRβ immunoreactivity in cirrhotic liver. LIFRβ expression is intense in cirrhotic liver (D), being concentrated in the latter in regions of ductular reaction (E) and perisinusoidal areas (F arrowheads). Magnification 400× (A and B); 200× (C–F); 1000× (insets in A and F).

message in the cell fraction that was positive for a macrophage marker. There is a perception in the literature that OSM is not expressed extensively in adult tissues except for cases in which there is either inflammation or active tissue remodeling, when it is mostly produced by activated T lymphocytes and monocytes [14]. Our observation of strong OSM expression in liver macrophages (Kupffer cells) suggests that this cytokine may have an important constitutive function in the liver that is distinct from the general inflammatory response involving OSM delivered by infiltrating inflammatory cells from the circulation.

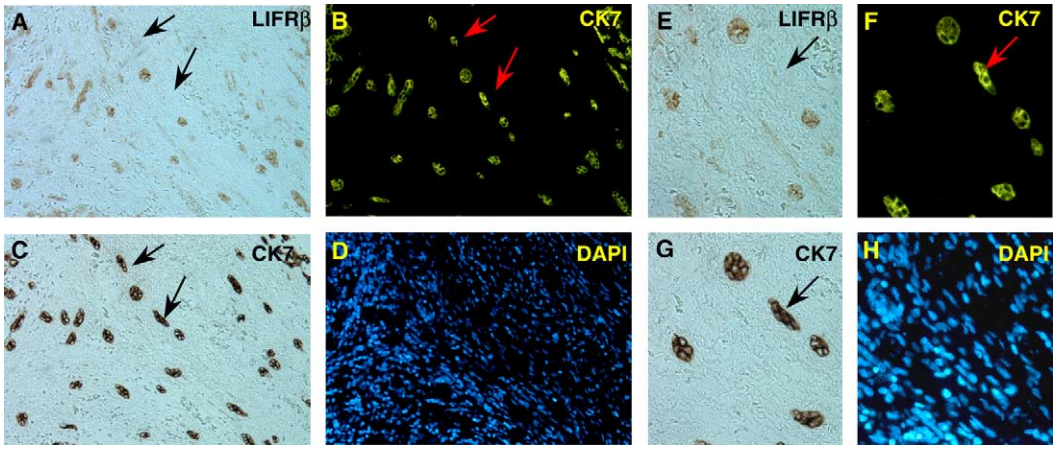

Fig. 4. Co-localization of LIFRβ and cytokeratin 7 in cirrhotic human liver. Co-localization of LIFRβ (A and E) and cytokeratin 7 (C and G) shown by light microscopy on sequential serial sections (LIFRβ and cytokeratin 7 staining using DAB) and double immunostaining using light microscopy for LIFRβ visualization (A, E) and immunofluorescence for cytokeratin 7 on the same section (B and F, respectively). Arrows indicate the positions of bile ductules that are cytokeratin-positive but LIFRβ negative. Nuclei were visualized by DAPI counterstaining (D and H, corresponding to C and G, respectively). Magnification 100× (A–D); 400× (E–H).

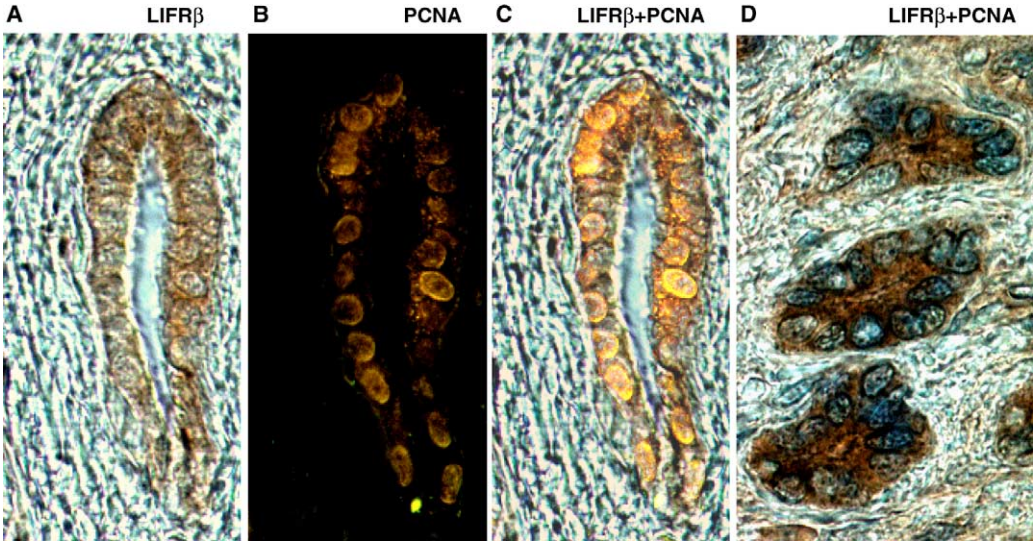

Fig. 5. Double immunostaining of cirrhotic liver, for LIFRβ assessed by light microscopy (A), for PCNA by immunofluorescence (B), and by overlay (C)–all on the same section. Light microscopy for both LIFRβ (horse radish peroxidase/DAB) and PCNA (alkaline phosphatase/blue substrate) on the same section (D). Magnification 1000×.

In a previous publication, we reported that OSM mRNA was up-regulated in all seven cirrhotic human livers analyzed but in only one of seven normal livers [6]. Up-regulation of OSM expression has been documented during fibrosis of other organs [10,12,21,13]. It is possible that increased OSM expression in half (2/4) of the normal liver samples analyzed in the present study could have been caused by procurement injury before transplantation, since a role for OSM in inflammatory reactions is well documented [22,23].

Taking into consideration the well-established pleiotropic potential of OSM, it seems very likely that its different

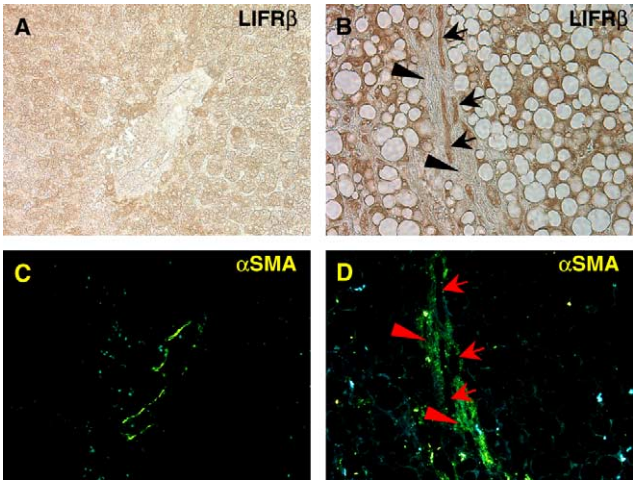

Fig. 6. Double immunostaining of normal human liver in health (A, C), and cirrhosis (B, D). LIFRβ staining assessed by light microscopy (upper panels, A, B) and for αSMA assessed by immunofluorescence (lower panels, C, D) on the same sections. In normal liver, there is LIFRβ background staining (A) and αSMA is localized exclusively in smooth muscle cells around blood vessels (C). In cirrhotic liver there is strong expression of LIFRβ in ductular (B) and intense expression of αSMA in periductular (D) regions. However, LIFRβ and αSMA expression are not co-localized (B and D). Arrows on the right side of each figure indicate position of LIFRβ-positive areas, arrowheads on the left show αSMA positive areas. Magnification 100×.

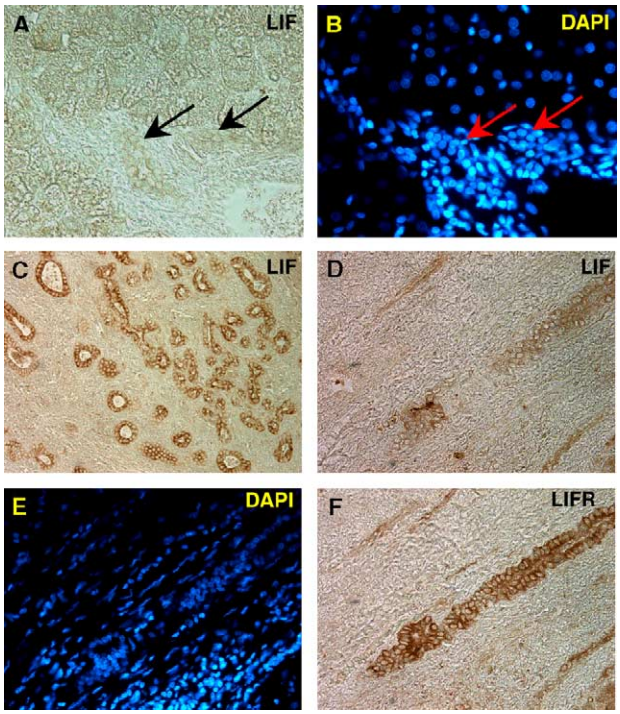

Fig. 7. LIF expression in human liver and its co localization with LIFRβ. (A, B) LIF is not detected in normal livers; arrows indicate location of bile ducts; nuclei, visualized by DAPI counterstain (B). (C) The majority of cirrhotic livers (5/6) showed marked expression of LIF in the area of reactive ductules. (D–*F*) Co-localization of LIF and LIFRβ assessed by light microscopy on serial sections (D and F, respectively). Nuclei visualization by DAPI counterstain (E) corresponding to (F). Magnification 400× (A, B, D–F); 200× (C).

functions are enabled by using different signaling pathways, rather than solely as a result of an increase in expression of OSM per se. Thus, when we looked for OSM receptor subunits we found expression of OSMRβ only in normal liver but not in cirrhosis, whereas LIFRβ was expressed in normal and more so in cirrhotic liver. Results of recent animal studies [20] have shown that mice deficient in the OSMRβ receptor show delayed liver regeneration, lower proliferation, and higher apoptosis rates of hepatocytes in acute liver injury, compared to wild type mice. Furthermore, administration of OSM facilitated the healing process after liver injury in wild type mice by preventing both hepatocyte apoptosis and tissue destruction [20]. Possibly the constant low level of OSMRβ expression that we found in normal human liver, which has also been documented for adult mice [24], reflects the very low proliferation level of hepatocytes in the normal liver, yet it may serve as liver protection mechanism by providing a threshold level of OSMRβ type receptors that can be rapidly up-regulated in acute liver injury [20]. The absence of OSMRβ expression in cirrhotic liver can be explained by the fact that these samples were taken from the liver explants of patients with end-stage liver disease who were undergoing liver transplantation, and hepatocyte proliferation is impaired in the late stages of cirrhosis [25]. The localization of OSMRβ to the hepatocytes, which we observed in the normal liver mostly in the regions of hepatocyte–hepatocyte junctions, is consistent with the fact that OSM enhances E-cadherin-based adherens junction formation between hepatocytes, and induces morphological maturation, at least in the mouse [26].

According to our Western blot analysis, 100% of cirrhotic samples demonstrated intense LIFRβ expression at an approximately 6-fold higher level than in 80% of normal livers. The intra-hepatic localization of LIFRβ expression in cirrhotic samples in the ductular reaction (Fig. 3D and E) and in perisinusoidal regions (Fig. 3F, arrowheads) is noteworthy too. According to our previous in vitro results [6,7,27], hepatic stellate cells, key players in the pathogenesis of liver fibrosis and the major source of abnormal ECM in liver disease, increase their collagen secretion and production of tissue inhibitor of metalloproteinase-1 (TIMP-1) in response to OSM stimulation. Thus it was logical to expect that perisinusoidal localization of LIFRβ would correspond to hepatic stellate cells in vivo. However based on co-staining with αSMA, we did not observe LIFRβ expression in activated stellate cells (myofibroblasts). That OSM receptors are present on hepatic stellate cells in culture is evident from the vigorous response of these cells to exogenous OSM stimulation [6,7,27]. Either the sensitivity of the current immunostaining method is seemingly inadequate for demonstrating OSM receptors on stellate cells in liver sections, or it is conceivable that LIFRβ is expressed by hepatic stellate cells that do not express αSMA and therefore cannot be readily distinguished in our liver sections. This is in keeping with the results demonstrated recently by Magness et al., of gene expression heterogeneity in mouse hepatic stellate cells, in which the expression of αSMA or lack thereof is dissociated from the expression of other genes, e.g. collagen α1(I) [28].

The novel observation that LIFRβ is expressed intensely in reactive bile ductules in cirrhotic liver is of interest in the context of both cirrhosis and biliary tract disease, in which bile duct proliferation occurs frequently. We found that LIFRβ is expressed in most of proliferating ductules, though there are LIFRβ negative ductules as well (Fig. 4). Our results on co-localized expression of LIFRβ and LIF in ductular reaction in cirrhosis is in agreement with the data of in situ hybridization in a rat model of chronic liver disease [29]. Omori et al. showed previously that LIF and LIFRβ messages were markedly up-regulated in bile duct epithelial cells in a rat model of persistent liver injury compared to a model of acute liver disease [29]. The strong immunoreactivity of LIF that we observed in reactive ductules of cirrhotic livers together with the fact that LIF mRNA is synthesized by bile duct cells in a rat model of chronic injury [29], leads us to hypothesize the existence of an autocrine mechanism of LIF synthesis and signaling in bile ducts. In any case, it suggests an important biological role for LIF signaling in ductular reaction, which is worthy of study. In the contrast, the low level of OSM immunoreactivity that we observed in cirrhotic liver, most likely reflects OSM from macrophages, acting on bile duct cells.

When the expression pattern of the OSM receptor subunits is taken together with data from animal studies in acute [20] and persistent [29] liver injury, it suggests that OSM is multifunctional in the human liver. A role for OSM in liver injury is proposed schematically in Fig. 8. Following acute liver injury and inflammation, OSM is secreted by activated

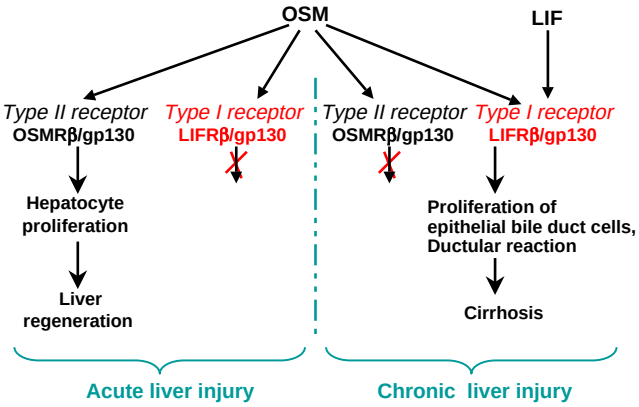

Fig. 8. Hypothetical model for the role of OSM and its interplay with LIF in acute and chronic liver injury. In acute liver inflammation, OSM secreted by activated Kupffer cells, interacts with its type II receptor (gp130/OSMRβ) to participate in liver regeneration, by promoting hepatocyte proliferation, inhibiting apoptosis, and enhancing intracellular adherens junction formation and maturation of hepatocytes. In chronic liver injury, especially when hepatocyte proliferation is delayed or inhibited as in the later stages of cirrhosis, there is reduced OSMRβ expression. The pathway involving utilization of the type I (gp130/LIFRβ) receptor enables OSM together with LIF to participate in proliferation of epithelial bile duct cells and the bile ductular reaction.

Kupffer cells and interacts with its type II receptor (gp130/OSMRβ) which is normally constitutively expressed on hepatocytes at a basal level and which is rapidly induced after acute liver injury [20]. Via this pathway, OSM is able to participate in liver regeneration by promoting hepatocyte proliferation, inhibiting apoptosis [20], and enhancing intracellular adherens junction formation and maturation of hepatocytes [26]. In chronic liver injury, especially when hepatocyte proliferation is delayed or inhibited, as in the late stages of cirrhosis [30], there is reduced OSMRβ expression. However, the pathway that utilizes the type I gp130/LIFRβ receptor enables OSM, together with LIF, to participate in proliferation of epithelial bile duct cells and the bile ductular reaction. This intriguing hypothesis remains to be tested.

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References

[1] Zarling JM, Shoyab M, Marquardt H, Hanson MB, Lioubin MN, Todaro GJ. Oncostatin M: a growth regulator produced by differentiated histiocytic lymphoma cells. Proc Natl Acad Sci USA 1986;83:9739–9743.
[2] Heinrich PC, Behrmann I, Muller-Newen G, Schaper F, Graeve L. Interleukin-6-type cytokine signalling through the gp130/Jak/STAT pathway. Biochem J 1998;334(Pt 2):297–314.
[3] Mosley B, De Imus C, Friend D, Boiani N, Thoma B, Park LS, et al. Dual oncostatin M (OSM) receptors. Cloning and characterization of an alternative signaling subunit conferring OSM-specific receptor activation. J Biol Chem 1996;271:32635–32643.
[4] Gearing DP, Bruce AG. Oncostatin M binds the high-affinity leukemia inhibitory factor receptor. New Biol 1992;4:61–65.
[5] Kuropatwinski KK, De Imus C, Gearing D, Baumann H, Mosley B. Influence of subunit combinations on signaling by receptors for oncostatin M, leukemia inhibitory factor, and interleukin-6. J Biol Chem 1997;272:15135–15144.
[6] Levy MT, Trojanowska M, Reuben A. Oncostatin M: a cytokine upregulated in human cirrhosis, increases collagen production by human hepatic stellate cells. J Hepatol 2000;32:218–226.
[7] Sohara N, Trojanowska M, Reuben A. Oncostatin M stimulates tissue inhibitor of metalloproteinase-1 via a MEK-sensitive mechanism in human myofibroblasts. J Hepatol 2002;36:191–199.
[8] Somasundaram R, Ruehl M, Schaefer B, Schmid M, Ackermann R, Riecken EO, et al. Interstitial collagens I, III, and VI sequester and modulate the multifunctional cytokine oncostatin M. J Biol Chem 2002;277:3242–3246.
[9] Nightingale J, Patel S, Suzuki N, Buxton R, Takagi KI, Suzuki J, et al. Oncostatin M, a cytokine released by activated mononuclear cells, induces epithelial cell-myofibroblast transdifferentiation via Jak/Stat pathway activation. J Am Soc Nephrol 2004;15:21–32.
[10] Atamas SP, White B. Cytokine regulation of pulmonary fibrosis in scleroderma. Cytokine Growth Factor Rev 2003;14:537–550.
[11] Luzina IG, Atamas SP, Wise R, Wigley FM, Choi J, Xiao HQ, et al. Occurrence of an activated, profibrotic pattern of gene expression in lung CD8+T cells from scleroderma patients. Arthritis Rheum 2003;48:2262–2274.
[12] Scaffidi AK, Mutsaers SE, Moodley YP, McAnulty RJ, Laurent GJ, Thompson PJ, et al. Oncostatin M stimulates proliferation, induces collagen production and inhibits apoptosis of human lung fibroblasts. Br J Pharmacol 2002;136:793–801.
[13] Bamber B, Reife RA, Haugen HS, Clegg CH. Oncostatin M stimulates excessive extracellular matrix accumulation in a transgenic mouse model of connective tissue disease. J Mol Med 1998;76:61–69.
[14] Malik N, Haugen HS, Modrell B, Shoyab M, Clegg CH. Developmental abnormalities in mice transgenic for bovine oncostatin M. Mol Cell Biol 1995;15:2349–2358.
[15] Kamiya A, Kinoshita T, Ito Y, Matsui T, Morikawa Y, Senba E, et al. Fetal liver development requires a paracrine action of oncostatin M through the gp130 signal transducer. Eur Mol Biol Org J 1999;18:2127–2136.
[16] Znoyko I, Sohara N, Spicer SS, Trojanowska M, Reuben A. Comparative studies of oncostatin M expression in the tissues of adult rodents. Anat Rec A Discov Mol Cell Evol Biol 2005;283A:182–186.
[17] Nakazawa K, Spicer SS, Schulte BA. Postnatal expression of the facilitated glucose transporter, GLUT 5, in gerbil outer hair cells. Hear Res 1995;82:93–99.
[18] Schmitt-Graff A, Kruger S, Bochard F, Gabbiani G, Denk H. Modulation of alpha smooth muscle actin and desmin expression in perisinusoidal cells of normal and diseased human livers. Am J Pathol 1991;138:1233–1242.
[19] Kinoshita T, Sekiguchi T, Xu MJ, Ito Y, Kamiya A, Tsuji K, et al. Hepatic differentiation induced by oncostatin M attenuates fetal liver hematopoiesis. Proc Natl Acad Sci USA 1999;96:7265–7270.
[20] Nakamura K, Nonaka H, Saito H, Tanaka M, Miyajima A. Hepatocyte proliferation and tissue remodeling is impaired after liver injury in oncostatin M receptor knockout mice. Hepatology 2004;39:635–644.
[21] Ihn H, LeRoy EC, Trojanowska M. Oncostatin M stimulates transcription of the human alpha2(I) collagen gene via the Sp1/Sp3-binding site. J Biol Chem 1997;272:24666–24672.
[22] Richards CD, Brown TJ, Shoyab M, Baumann H, Gauldie J. Recombinant oncostatin M stimulates the production of acute phase proteins in HepG2 cells and rat primary hepatocytes in vitro. J Immunol 1992;148:1731–1736.
[23] Langdon C, Kerr C, Hassen M, Hara T, Arsenault AL, Richards CD. Murine oncostatin M stimulates mouse synovial fibroblasts in vitro and induces inflammation and destruction in mouse joints in vivo. Am J Pathol 2000;157:1187–1196.
[24] Tamura S, Morikawa Y, Tanaka M, Miyajima A, Senba E. Developmental expression pattern of oncostatin M receptor beta in mice. Mech Dev 2002;115:127–131.
[25] Wilfredo Canchis P, Gonzalez SA, Isabel Fiel M, Chiriboga L, Yee H, Edlin BR, et al. Hepatocyte proliferation in chronic hepatitis C: correlation with degree of liver disease and serum alpha-fetoprotein. Liver Int 2004;24:198–203.
[26] Matsui T, Kinoshita T, Morikawa Y, Tohya K, Katsuki M, Ito Y, et al. Ras mediates cytokine-induced formation of E-cadherin-based adherens junctions during liver development. Eur Mol Biol Org J 2002;21:1021–1030.
[27] Sohara N, Znoyko I, Levy MT, Trojanowska M, Reuben A. Reversal of activation of human myofibroblast-like cells by culture on a basement membrane-like substrate. J Hepatol 2002;37:214–221.
[28] Magness ST, Bataller R, Yang L, Brenner DA. A dual reporter gene transgenic mouse demonstrates heterogeneity in hepatic fibrogenic cell populations. Hepatology 2004;40:1151–1159.
[29] Omori N, Evarts RP, Omori M, Hu Z, Marsden ER, Thorgeirsson SS. Expression of leukemia inhibitory factor and its receptor during liver regeneration in the adult rat. Lab Invest 1996;75:15–24.
[30] Falkowski O, An HJ, Ianus IA, Chiriboga L, Yee H, West AB, et al. Regeneration of hepatocyte 'buds' in cirrhosis from intrabiliary stem cells. J Hepatol 2003;39:357–364.