Histol Histopathol (2009) 24: 651-660

http://www.hh.um.es

Histology and Histopathology

Cellular and Molecular Biology

Review

Lysyl oxidases in mammalian development and certain pathological conditions

Joni M. Mäki

Oulu Centre for Cell-Matrix Research, Biocenter Oulu and Department of Medical Biochemistry and Molecular Biology, University of Oulu, Oulu, Finland

Summary. Lysyl oxidase (LOX) catalyzes the oxidation of the side chain of a peptidyl lysine converting specific lysine and hydroxylysine residues of α–aminoadipic-δ-semialdehydes, which form covalent crosslinks in collagens and elastin. Five different but closely related lysyl oxidase isoenzymes have been identified to date, and they seem to have overlapping functions in many tissues. Modification of the extracellular matrix by lysyl oxidases has been shown to be a critical contributor to the development of various organs and certain pathological conditions.

Key words: LOX, Copper enzyme, Branching morphogenesis, Aneurysm

Introduction

Lysyl oxidase (LOX) has traditionally been described as an extracellular cuproenzyme that initiates lysine-derived crosslink formation in collagens and elastin. Its role in physiology, development and disease states has proved to be much more complicated, however, and cannot be entirely explained by its function in the extracellular matrix (ECM). LOX has also been identified inside cells, and even in the nucleus, and researchers have identified new functions and substrates for it in recent years. Current findings have shown that LOX is required for the hypoxia-mediated metastasis of certain cancer cells and for the development of major organs, such as the cardiovascular and respiratory systems, and have assigned it highly important roles in development and diseases in terms of migration, invasion, epithelial-mesenchymal transition (EMT) and even intracellular signalling (Fig. 1). In addition, other members of the lysyl oxidase gene family (LOXL1-4) seem to have a wide range of effects on the behaviour of different cell types.

Offprint requests to: Joni M. Mäki, Oulu Centre for Cell-Matrix Research, Biocenter Oulu and Department of Medical Biochemistry and Molecular Biology, University of Oulu, P.O. Box 5000, 90014 Oulu, Finland. e-mail: joni.maki@oulu.fi

LOX in mammalian development

The role of LOX in cardiovascular development and diseases has been studied mainly using animal models and tissues. Inactivation of the LOX gene in the mouse leads to perinatal death caused by major dysfunction and developmental defects in the cardiovascular and respiratory systems, and the mice have generalized elastolysis and abnormal collagen content in various tissues (Mäki et al., 2002, 2005; Hornstra et al., 2003). Mouse LOX mRNA is expressed prominently in the cardiovascular system during embryonic development, and its expression level is upregulated significantly between E11.5-E13.5 (Tsuda et al., 2003). Lysyl oxidation, reflecting the activity of LOX, can be detected in rat embryos starting from E9.5, which corresponds to a period of transition from the post-blastocystic stage to the pre-embryonic state and follows expression of the ATP7A gene, which provides a copper ion for the

Abbreviations: AD, Alzheimer disease; ALS, amyotrophic lateral sclerosis; ßapn, ß-aminopropionitrile; BMP, bone morphogenic protein; CNS, central nervous system; CRL, cytokine receptor-like domain; FGF, fibroblast growth factor; FN, fibronectin; FAK, focal adhesion kinase; ECM, extracellular matrix; EMT, epithelial-mesenchymal transition; ERK, extracellular signal-regulated kinase; HIF, hypoxia-inducible factor; HNSCC, neck squamous carcinoma cell; LOX, lysyl oxidase; LOXL, lysyl oxidase-like protein; LOX-PP, LOX propeptide; LTQ, lysine tyrosylquinone cofactor; MEK, mitogen-activated protein kinase; NF-κB, nuclear factor-κB; OHS, occipital horn syndrome; PCP, procollagen C-proteinase; PI3K, phosphoinositide-3 kinase; PXS, pseudoexfoliation syndrome; SRCR, scavenger receptor cysteine-rich region; TGF-ß, transforming growth factor-ß; TNF-α, tumor necrosis factor-α; Spry, Sprouty proteins.

reaction catalyzed by LOX (Tchaparian et al., 2000). The temporal expression pattern of LOX overlaps with those of the main collagen types and elastin, and the roles of its substrates, especially those of collagen types I and IV, are crucial for normal tissue growth in the early development of mouse embryos, as observed in studies of corresponding mouse models (Löhler et al., 1984; Pöschl et al., 2004). It is therefore surprising that depletion in LOX activity causes major organ defects only at the later stages in embryonic development, suggesting that the other LOX isoenzymes probably have compensatory effects. Development of the vasculature in elastin-knockout mice is indistinguishable from that in the wild type until E17.5, clearly demonstrating the restricted role of elastin in the maturation of the cardiovascular system during embryonic development (Li et al., 1998), and also pointing to the fact that the defects observed in $Lox^{-/-}$ mice are not purely caused by elastinopathy. LOX seems to have a major role in all the cell types that contribute to the normal structure and function of vascular walls. Endothelial cells in the vascular system of $Lox^{-/-}$ E18.5 embryos showed degenerative changes, such as blebbing, vacuolization and detachment from the internal elastic lamina (Mäki et al., 2002). This observation supports the finding that LOX is needed for maintenance of the endothelial barrier. LOX thus seems to have a role in atherogenesis and endothelial dysfunction triggered by atherosclerotic risk factors and proinflammatory cytokines such as TNF-α (Alcudia et al., 2008). The most obvious affect of LOX during embryogenesis seems to be its ability to enhance the physical properties of the ECM of vascular walls and other tissues. Improperly crosslinked elastin and collagens not only affect the function of specific cell types, such as vascular smooth muscle cells, but have also wide systemic effects, as shown by ultrasonographic studies of $Lox^{-/-}$ embryos with increased pulsatility indices in the major arteries and veins (Mäki et al., 2002). LOX seems to be the major crosslinking enzyme before parturition, as it is responsible for over 80% of total lysyl oxidase activity in skin fibroblasts and aortic smooth muscle cells isolated from E18.5 mouse embryos (Mäki et al., 2005). Lack of its activity reduces the amount of desmosine crosslinks in embryonic lungs and aorta by approximately 60%, and immature collagen crosslinks in the whole embryonic body by 40% (Hornstra et al., 2003). These findings also reveal the partially compensatory mechanisms provided by the other lysyl oxidase family members. LOX activity seems to enhance the durability of elastic fibres and the ECM in the vascular system, since the prevalence of aneurysms and the destruction of elastic fibres are more prominent in the large arteries near the heart that have a higher blood pressure.

In addition to the cardiovascular system, LOX deficiency disturbs development of the respiratory system and some other tissues. Lung parenchymal and pleural urea-extractable lysyl oxidase activity is relatively high in rabbits in the first 3 weeks, but starts to decrease by approximately 50% during weeks 4-10. By contrast, lysyl oxidase activity in the airways remains high for the first 10 weeks and then decreases by 50%. Pneumonectomy, and the subsequently decreased oxygen content of tissues (by 12-13%) resulted in a prompt and sustained increase in lung, but not pleural or airway enzyme activity (Kagan, 1986), thus suggesting differential roles and/or differential tissue distributions among the lysyl oxidases. Postnatal inhibition of LOX by ß-aminopropionitrile (ßapn) in rats during the first 4 weeks of life results in a 40-56% reduction in alveoli. Moreover, the alveoli are significantly enlarged, with fragmented elastic fibres and loosely arranged collagen fibres of widely varying diameter (Kida and Thurlbeck, 1980). Lysyl oxidation thus seems to play a role in the regulation of postnatal lung development. It nevertheless

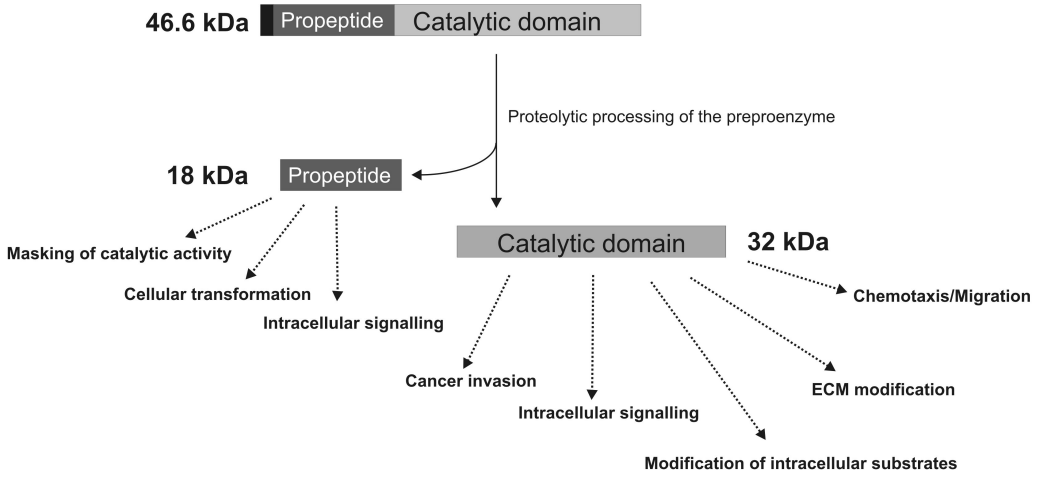

Fig. 1. Proposed functions of LOX in cell biology. Multifunctional LOX modifies ECM proteins and intracellular substrates, contributes to the motility and migration of cells and promotes cancer invasion and cellular transformation.

has an even more crucial role in the development of the embryonic lungs and other respiratory organs, such as the diaphragmatic muscle (Mäki et al., 2005). The development and branching of the distal and proximal airways of $Lox^{-/-}$ mouse embryos are severely impaired, with characteristic abnormalities in elastic and collagen fibres. Lung morphogenesis in mice starts with the formation of two lung buds at E9.5, and thereafter branching is continued until the proper structures of the lung lobes are achieved (Ten Have-Opbroek, 1991). Branching morphogenesis itself is strictly regulated by various factors, such as the FGFs, Spry's, BMP-4 and TGF-ß1 (Cardoso, 2001). At the pseudoglandular stage of lung maturation in the rat (E9.5 to E14.2 in the mouse embryo), collagen fibres and fibronectin (FN) are predominantly concentrated at the sites of bifurcations in the primitive bronchial primordia, giving rise to further branching of the primitive bronchi and bronchioles at later stages of development (Wasowicz et al., 1996; Roman, 1997). FN has been found to bind LOX with high affinity, and to play a role in the regulation of its proteolytic activation (Fogelgren et al., 2005), so that it may enhance the LOX-mediated crosslinking of collagens at sites of bifurcation and subsequently promote branching. This may be one of the regulatory steps that control the ECM remodelling that is needed for proper branching of the lungs. Other known regulators of branching morphogenesis in lung tissue are hypoxia (Gebb and Jones, 2003) and various effectors, such as retinoic acid and TGF-ß, all of which also regulate LOX transcription and activity (Smith-Mungo and Kagan, 1998; Cardoso, 2001; Csiszar, 2001). In addition to its contribution to inhibiting the proliferation and differentiation of epithelial cells in the lungs, TGF-ß is known to promote the synthesis of the ECM, which, when deposited at the epithelial-mesenchymal interface, is thought to prevent local branching (Cardoso, 2001). TGF has also been found to upregulate the mRNA expression and activity of LOX (Csiszar, 2001) and may thus contribute to the remodelling of the ECM at branching sites via LOX itself.

The elastin and collagen defects caused by the decreased lysyl oxidation activity in $Lox^{-/-}$ animals is not limited to the respiratory and cardiovascular systems but is also manifested in other tissues. The total quantity of collagen crosslinks is reduced by 40% in the body of mutant embryos (Hornstra et al., 2003), and both elastic and collagen fibres have abnormal appearance in their skin (Mäki et al., 2005). LOX clearly makes a major contribution to ECM components, which not only regulate embryonic development but affect various pathological conditions.

LOX in pathological conditions

The role of LOX in cardiovascular diseases has been recently reviewed thoroughly by Alcudia and co-workers (2008), and will not be discussed further here. A great number of reports have suggested a strong association between organ fibrosis and increased lysyl oxidase activity in hepatic fibrosis in the rat, in models of lung, arterial and dermal fibrosis and in chronic human liver fibrosis, adriamycin-induced kidney fibrosis and other pathological conditions leading to fibrosis (see Kagan 1986, 1994; Smith-Mungo and Kagan, 1998). It has been suggested that chemotherapeutic inhibition of lysyl oxidase activity may prove to be a useful tool for the control of fibrosis, since it leads to the accumulation of insufficiently crosslinked and thus more soluble collagen molecules that are significantly more susceptible to proteolytic degradation.

Reduced lysyl oxidase activity has been detected in two X-linked recessively inherited disorders, Menkes disease and its milder variant occipital horn syndrome (OHS) (Horn and Tümer, 2002; OMIM 309400 and 304150). Both diseases are caused by mutations in the ATP7A gene that encodes a copper-transporting P-type ATPase, leading to inadequate availability of copper (Horn and Tümer, 2002). Due to the inefficient functioning of many cuproenzymes, including lysyl oxidase family members, the manifestations of these diseases are widespread and affect several tissues. Menkes disease patients have severe abnormalities in neural development and certain connective tissue abnormalities, and often die prematurely (Horn and Tümer, 2002), whereas OHS patients exhibit only mild neurological changes and mainly connective tissue defects, suggesting that the lysyl oxidases may be the most sensitive enzymes to copper availability in cells (Horn and Tümer, 2002). Mottled blotchy mice, an animal model for OHS, have a significantly higher incidence of aortic aneurysms, exhibit disrupted elastic fibres in the vascular walls and lungs, and have structurally and functionally abnormal lungs (Andrews et al., 1975; Fisk and Kuhn, 1976; Brophy et al., 1988), thus bearing a striking resemblance to lathyric, $Lox^{-/-}$ and $Lox11^{-/-}$ animal models (Mäki et al., 2002; Steinmann et al., 2002; Hornstra et al., 2003; Liu et al., 2004; Mäki et al., 2005) and animals exposed to copper deficiency (Kagan, 1986). Although the lysyl oxidases are implicated in the connective tissue problems associated with Menkes disease and OHS, no mutations in the LOX or LOXL genes are currently known that cause a predisposition to these diseases. Both upregulation and downregulation of LOX have been observed in Wilson disease, another condition which results from abnormal copper metabolism, attributed to mutations in the ATP7B gene (OMIM 277900), also encoding a copper-transporting P-type ATPase (Linz and Lutsenko, 2007). The fact that the change in LOX activity is clearly milder and more restricted to certain tissues in Wilson patients can be explained by differential expression of the ATP7A and ATP7B genes (Vadasz et al., 2005; Linz and Lutsenko, 2007).

The role of LOX in certain neurodegenerative diseases has also been studied. Upregulation and altered distribution of LOX has been observed in the central nervous system of a mouse model (mSOD1) for

amyotrophic lateral sclerosis (ALS) (Li et al., 2004). Interestingly, LOX is expressed not only in the fibrogenic cells, including the vascular walls of the central nervous system (CNS), but also in the brain matrix and neurons of normal mice and rats. In mSOD1 mice LOX expression and immunoreactivity is observed in sites where it is not normally observed, and the amount of LOX protein is increased in the neurons of the spinal cord, brain stem and cortex and the Purkinje cells of the cerebellum. In addition, the expression level is enhanced in the neurons of various parts of the CNS and the enzyme activity increases coincidentally with late-stage ALS (Li et al., 2004). LOX has also been characterized as an electroconvulsive shock-inducible gene of the CNS, and thus shows conditional and tissue-specific expression, which supports its potential function in the CNS (Sun et al., 2005). Lysyl oxidase activity is increased by approximately 30% in Alzheimer disease (AD) and in non-Alzheimer dementia, and is localized in the blood vessel walls and in plaque-like structures typical of these diseases (Gilad et al., 2005). Moreover, the number of LOX-positive plaque-like structures is more than two-fold higher in AD than in patients with non-AD dementia (Gilad et al., 2005). These findings suggest that LOX itself not only contributes to connective tissue manifestations of diseases, but may also affect the normal functioning and development of the CNS.

LOX is capable of suppressing cellular transformation, but paradoxically, also enhances invasion by certain cancer cells. The first evidence of the tumor suppressor activity of LOX was the observation that it is able to suppress the activation of *c-H-ras* in mouse NIH 3T3 cells, and thus subsequently inhibits transformation of these cells (Contente et al., 1990). Numerous studies have since supported this finding, demonstrating that the levels of expression and activity of LOX are reduced in many types of cancer cell (Csiszar, 2001; Payne et al., 2007). There is also a considerable amount of evidence that the tumor suppressor activity of LOX is caused by its intracellular actions, through its effects on intracellular signalling (Payne et al., 2007). The reduced expression of LOX mRNA in tumor cells may at least partly be the result of autocrine growth factor pathways, such as bFGF, or signalling cascades related to ras or other oncogenic processes (Palamakumbura et al., 2003). Ras itself mediates cellular transformation partly, and indirectly, by activation of the transcription factor NF-κB. Ectopic expression of LOX in ras-transformed NIH-3T3 cells resulted in decreased NF-κB transcriptional activity, inhibiting the nuclear intake of NF-κB. This effect was mediated via strong downregulation of both PI3K and Akt kinases and partial inhibition of MEK. Thus some of the anti-oncogenic effects of LOX on ras-mediated cellular transformation may result from a LOX-dependent inhibition of signalling pathways leading to the activation of NF-κB (Jeay et al., 2003). The exact role of the catalytic activity of LOX in transformation is currently unclear, but there is some evidence to suggest that the LOX propeptide (LOX-PP) may be mainly responsible for induction of the reversion of ras-transformed NIH-3T3 cells to the non-oncogenic phenotype (Palamakumbura et al., 2003, 2004; Min et al., 2007; Wu et al., 2007a). The LOX-PP was found to inhibit transformation of breast cancer cells driven by Her-2/neu, an upstream activator of ras, by suppressing activation of the extracellular signal-regulated kinase Akt and NF-κB. In addition, it was found to inhibit signalling cascades induced by Her-2/neu that promote a more invasive phenotype (Min et al., 2007). In addition, ectopic expression of LOX-PP in pancreatic cancer cells has been shown to reduce ERK and Akt activities, inhibit growth in soft agar and migration, and reduce levels of NF-κB and its target BCL2 (Wu et al., 2007a).

Both the catalytic activity of LOX and its mRNA expression are dramatically downregulated in most cancer cells. Recent studies have demonstrated, however, that the LOX mRNA level is elevated in highly invasive cancer cells, for example, and affects their invasive properties (Payne et al., 2007). LOX mRNA was found to be upregulated in invasive breast cancer cells, enhancing their invasive capability, and the mediator of this was shown to be the catalytically active form of LOX. The LOX-dependent chemotactic response of these cells was elicited by H_2O_2 (a by-product of the LOX-catalysed reaction) produced by LOX acting on one or more unidentified substrates (Payne et al., 2007). The role of H_2O_2 produced by the LOX-catalyzed reaction in chemotaxis has also been pinpointed in other studies and cell lines (Lazarus et al., 1995; Li et al., 2000; Lucero et al., 2008), but the actual mechanism is still under investigation. Inhibition of LOX activity increases actin stress fibre formation and Rho activity in breast cancer cells through the p130(Cas)/Crk/DOCK180 signalling complex (Payne et al., 2006) and thus inhibition of the LOX catalytic activity could limit the invasiveness of cancer cells. Erler et al. (2006) observed that the elevation of LOX expression under hypoxic conditions appears to be essential for the hypoxia-induced metastatic response of breast cancer and head and neck squamous carcinoma cells (HNSCC), and that inhibition of LOX reduced the invasiveness of cancer cells. Mechanistically, the secreted and active form of LOX seemed to be responsible for the invasive properties of the hypoxic cancer cells through its effects on focal adhesion kinase (FAK) activity and cell-to-matrix adhesion. Postovit and co-workers (2008) confirmed that hypoxia greatly enhances LOX expression, but at the same time reduces its catalytic activity. The LOX-dependent activation of FAK/Src and migration of poorly invasive breast cancer cells is markedly increased during re-oxygenation following the hypoxia, but not in hypoxia alone. Furthermore, LOX expression seems to be only partially dependent on the hypoxia-inducible transcription factor 1 (HIF-1) and to be independent of a hypoxic environment at the later stages of tumor progression. An

association between LOX and EMT, the initial step in metastasis, has also been established (Higgins et al., 2007; Sahlgren et al., 2008). Recent findings have shown that LOX may be a good therapeutic target for preventing and treating tumor metastases.

Lysyl oxidase-like proteins (LOXL1-4)

The lysyl oxidase gene family consists of 5 members, coding for LOX itself and lysyl oxidase-like (LOXL) proteins 1-4. LOX and LOXL1 differ drastically from the other family members within their N-terminal regions, but all the members have high similarity in their catalytic domain (Fig. 2). The spectrum of the substrates and biological functions of LOXL1, 2, 3 and 4 remain to be established.

Lysyl oxidase-like 1 (LOXL1)

LOXL1 cDNA was originally characterized by Kenyon et al. (1993), and it has been identified as a secreted protein that is expressed in the ECM in active fibrotic diseases and in the early stromal reaction in breast cancer (Decitre et al., 1998). An inactive LOXL1 precursor has been isolated from the bovine aorta, and can be activated by BMP-1 *in vitro* on collagen and elastin (Borel et al., 2001). Liu and co-workers (2004) showed that LOXL1 is essential for elastic fibre homeostasis. Loxl1$^{-/-}$ mice do not deposit entirely normal elastic fibres in the uterine tract post partum and they develop pelvic organ prolapse, enlarged airspaces in the lungs, loose skin, intestinal diverticula and vascular abnormalities with concomitant tropoelastin accumulation. The LOXL1 protein was observed to co-localize with fibulin-5 at sites of elastogenesis. The study clearly showed the significance of elastic fibre renewal for tissues that are exposed to physical forces and therefore need the ability to deform repetitively and reversibly. In a continuation to this work, Liu and coworkers analysed the role of LOXL1 in elastic fibre formation and renewal in pelvic floor disorders using the Loxl1$^{-/-}$ mouse line as an animal model. LOXL1 was found to be highly expressed in the reproductive tract and downregulated during ageing. Loxl1$^{-/-}$ animals were unable to replenish their elastic fibres after parturition, which led to pelvic organ collapse, weakening of the vaginal wall, paraurethral pathology and bladder and lower urinary tract dysfunction (Liu et al., 2006, 2007). These findings are very similar to the phenotype of fibulin-5 knockout animals, thus strongly supporting an essential interaction between fibulin-5 and LOXL1 in elastic fibre homeostasis (Nakamura et al., 2002). LOXL1 is assumed to act via fibulin-5, a component of elastic fibres which has also been shown to bind tropoelastin, the uncrosslinked and soluble form of elastin (Liu et al., 2004). The role of fibulin-5 seems to be highly critical for elastogenesis, as it serves as an organizer molecule for some of the components of elastic fibres (Hirai et al., 2007). It deposits microfibrils, promotes aggregation of tropoelastin molecules through coacervation and also interacts not only with LOXL1, but also with LOXL2 and LOXL4, and may therefore tether these enzymes to microfibrils and subsequently facilitate aggregation and crosslinking of elastin itself to microfibrils (Hirai et al., 2007). The pro-region of LOXL1 is probably needed for its efficient secretion from cells and mediates its association in the ECM (Thomassin et al., 2005). This may be feasible, since the pro-regions of these enzymes do have a positive effect on their solubility and a negative effect on their activation, and therefore will change their physical properties, keeping the actual enzymatic activity latent until it is needed and then induced by the processing enzymes (Smith-Mungo and Kagan, 1998; Kagan and Li, 2003; Lucero and Kagan, 2006).

LOXL1 gene mutations (R141L, G153D) have recently been linked to the pseudoexfoliation syndrome (PXS) (Thorleifsson et al., 2007), which is described as

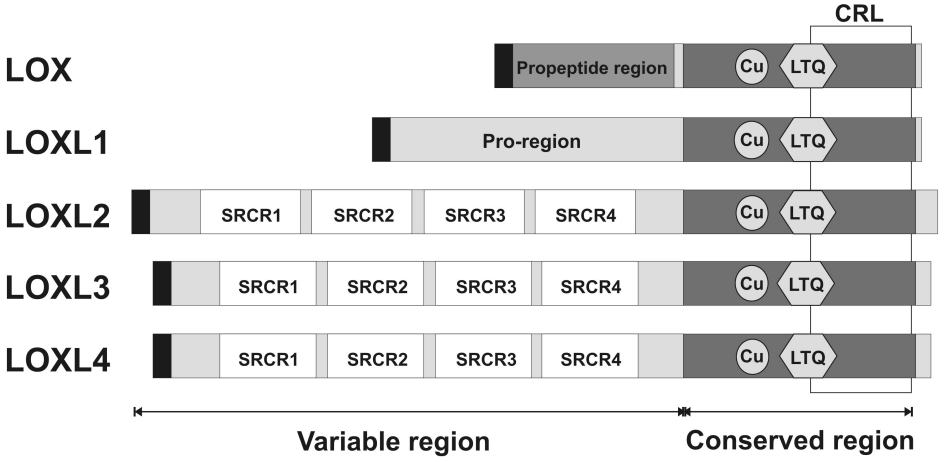

Fig. 2. The lysyl oxidase protein family. The predicted signal peptides are represented by black boxes and the four scavenger receptor cysteine-rich regions (SRCR) in LOXL2, 3 and 4, the propeptide region of LOX and the proline rich-region of LOXL1 are indicated. The putative sites of copper binding (Cu), lysine tyrosylquinone cofactor formation (LTQ) and the cytokine receptor-like domain (CRL) are highly conserved between all family members.

an age-related, systemic elastic microfibrillopathy (Ritch, 2008). PXS is also associated with an increased risk of cardiovascular and cerebrovascular diseases (Ritch and Schötzer-Schrehardt,2001). Interestingly, the same G153D mutation of the LOXL1 gene was found to be associated with spontaneous cervical artery dissection (Kuhlenbäumer et al., 2007). Both detected mutations are located in the pro-region of LOXL1, which is thought to be involved on account of its association with the ECM. The effects of these mutations are not limited to the eye, however, since abnormal elastic fibres can also be found in the heart, lung, liver, kidney and other tissues of patients with PXS (Tarkkanen et al., 2008). Overlapping manifestations can be seen in patients suffering from cutis laxa, a disease caused by mutations in the fibulin-5 and elastin genes (Zhang et al., 1999; Loeys et al., 2002), and in OHS (Horn and Tümer, 2002). The latter is also characterized by defects in collagen crosslinking and extractability (Horn and Tümer 2002), for which no direct association with LOXL1 defects has been found in humans or in animal models.

In cancer, LOXL1 has been localized to the stromal reaction in bronco-alveolar carcinomas and ductal breast tumors (Payne et al., 2007). LOXL1 expression can be detected in highly invasive/metastatic MDA-MB-231 and Hs578T breast cancer cell lines, but is absent from the poorly invasive cell lines MCF-7 and T47D (Kirschmann et al., 2002). The LOXL1 gene has been shown to be silenced by methylation in bladder cancer cells and primary tumors, and reintroduction of LOXL1 genes into bladder cancer cells leads to decreased colony formation ability (Wu et al., 2007b). In the same study, overexpression of LOXL1 was found to antagonize ras via the ERK signalling pathways. LOXL1 has also been shown to interact and cooperate with the Snail transcription factor, a regulator of EMT (Peinado et al., 2005).

Lysyl oxidase-like proteins with scavenger receptor cysteine-rich repeats (LOXL2-4)

In contrast to the C-terminal regions, the N-terminal regions of the LOX family members show only minor sequence homology, except that full-length LOXL2, LOXL3, and LOXL4 contain four repeated copies of scavenger receptor cysteine-rich (SRCR) domains in their N termini (Fig. 2). These domains, which are known to mediate protein-protein interactions in cell adhesion and cell signalling, are found on either cell surface proteins or secreted proteins (Csiszar, 2001). The actual function of the SRCR domains with respect to LOXL2-4 remains to be resolved.

Human LOXL2 was originally cloned and characterized by Saito et al. (1997), who named it WS9-14 due to its possible association with Werner syndrome, a disease characterized by premature ageing. LOXL2 is abundantly expressed in senescent fibroblasts and several adherent tumor cell lines, but is down-regulated in non-adherent cell lines, suggesting that LOXL2 may be involved in cell adhesion and may thus contribute to the metastasis of cancer cells (Saito et al., 1997). LOXL2 expression is increased in a variety of cancer cells, including colon adenocarcinomas, esophageal squamous cell carcinomas and invasive breast cancer cell lines (Kirschman et al., 2002; Fong et al., 2007). On the other hand, it is downregulated in some cancer cell lines such as ras-transformed rat fibroblasts, head and neck squamous cell carcinomas and ovarian tumors (Hough et al., 2000; Ono et al., 2000; Zuber et al., 2000; Rost et al., 2003), which suggests that it is regulated differentially in various cancer cell types. Akiri and co-workers (2003) found that LOXL2 expression in periductal carcinomas is significantly correlated with tumor malignancy. It is highly expressed in invasive/metastatic breast cancer cells, but not in the non-metastatic estrogen-dependent MCF-7 cells. Overexpression of recombinant LOXL2 in the MCF-7 cells of nude mice produces estrogen-dependent tumors which develop more rapidly than tumors originating from control cell lines, are surrounded by a high concentration of dense collagen fibres and contain many fibrotic foci. MCF-7 cells overexpressing LOXL2 invade the pseudocapsules that surround the tumors, and proceed to the adjacent blood vessels, nerves and muscles. In highly invasive cancer cell lines LOXL2, 3 and 4 expression was found to correlate with the production of Snail, one of the mediators of EMT. Further analysis using a two-hybrid yeast screen showed that Snail could interact with all the lysyl oxidases, the catalytic domains of LOXL2 and LOXL3 being the most potent interacting partners, and could collaboratively repress E-cadherin transcription (Peinado et al., 2005). Interaction in the Snail/LOXL2/LOXL3 protein complex is likely to be dependent on the Snail N-terminal (SNAG) domain and the lysine residues K98 and K137 within it, which seem to be important for Snail stability and function. Knockdown studies employing RNA interference in Snail-expressing metastatic carcinoma cells showed that the downregulation of LOXL2 leads to decreased tumor growth associated with an increase in apoptosis and reduced expression of mesenchymal and invasive/angiogenic markers.

LOXL2 (and LOX) have been found to be upregulated in the hepatocytes of patients with Wilson's disease, primary biliary cirrhosis and some other conditions characterized by liver fibrosis, with an accompanying increase in collagen deposition (Vadasz et al., 2005). Moreover, recombinant LOXL2 seems to be able to oxidize type I collagen. This was inhibited by D-penicillamine, a copper-chelator, but not by ,ßapn at a concentration that completely abolishes the activity of LOX. In addition, LOXL2 inhibits the proliferation of HepG2 hepatoblastoma cells and catalyzes the oxidation of cell surface proteins (Vadasz et al., 2005). LOXL2 has also been identified as a gene conferring susceptibility to intracranial aneurysms (Akagawa et al., 2007). Lelièvre et al. (2008) suggested that LOXL2 is able to interact

with VE-statin/egfl7, which co-localizes with elastic fibres in the vascular walls, thus suggesting a possible role for its regulation of elastogenesis. Moreover, VE-statin/egfl7 may also regulate the activity of other lysyl oxidase forms, since it binds directly to the catalytic domains of enzymes and its repression with siRNA enhances lysyl oxidation.

LOXL3 mRNA is expressed in a variety of tissues (Huang et al., 2001; Jourdan Le Saux et al., 2001; Mäki and Kivirikko, 2001), but its function is largely unknown. LOXL3 has a tissue-specific variant lacking exons 1-3 and 5, and thus lacking the SRCR domains 1-3, and it has also been suggested that this variant may be enzymatically active and oxidize collagens and elastin (Lee and Kim, 2006). LOXL3 is expressed in highly invasive/metastatic breast cancer cells but not in poorly invasive/metastatic ones (Kirschmann et al., 2002). Overexpression of LOXL3 in epithelial cells induces EMT, and is thought to co-operatively regulate Snail transcription factor in cancer cells (see previous chapter and Peinado et al., 2005). In mouse models of cardiac remodelling, LOXL3 expression and activity have also been associated with ventricular stiffness and incongruence with lymphocyte function (Yu et al., 2008).

LOXL4 was originally described as a cartilage-specific protein in mice with the ability to oxidize fibrillar collagens (Ito et al., 2001), but its mRNA expression pattern in human tissues was found to be much wider (Asuncion et al., 2001; Mäki et al., 2001). Like the other members of the lysyl oxidase gene family, it is expressed in highly invasive/metastatic breast cancer cells but not in poorly invasive/metastatic ones (Kirschmann et al., 2002). It is also upregulated in HNSCC cells relative to normal epithelial cells (Holtmeier et al., 2003), and in invasive HNSCC tumors and primary/metastatic HNSCC cell lines (Görögh et al., 2007; Weise et al., 2008). On the other hand, both LOXL4 and LOXL1 have been commonly observed to be silenced by methylation in human bladder cancer cells and to lose their expression in primary bladder tumors. Overexpression of the LOXL4 gene in bladder cancer cells leads to a decrease in colony formation ability and seems to antagonize ras via the ERK signalling pathways. Somatic mutations in LOXL4 have been found in bladder cancer, all of them in exon 8, which encodes for a SRCR domain (Wu et al., 2007b).

Concluding remarks and future prospects

Current results indicate that the lysyl oxidase family members have various roles, both intracellular and extracellular, in development and in pathological conditions, the most topical among these functions clearly being those connected with cancer biology. Regulation of these enzymes, especially LOX and LOXL2, clearly differs with the stage of cancer progression, being driven mainly by microenvironmental factors such as hypoxia. From a developmental biology viewpoint, the effects of the lysyl oxidases on cellular adhesion, migration and proliferation in particular may be a driving force during the development of various organs and should be investigated in more detail.

Acknowledgements. I apologize to all the authors whose work could not be cited due to space limitations. I also regret that I have had to cite some reviews instead of the original reports on account of the broad nature of this article. I would like to thank Professor Johanna Myllyharju (University of Oulu) for valuable comments that have led to improvements in the manuscript.

References

Akagawa H., Narita A., Yamada H., Tajima A., Krischek B., Kasuya H., Hori T., Kubota M., Saeki N., Hata A., Mizutani T. and Inoue I. (2007). Systematic screening of lysyl oxidase-like (LOXL) family genes demonstrates that LOXL2 is a susceptibility gene to intracranial aneurysms. Hum. Genet. 121, 377-387.

Akiri G., Sabo E., Dafni H., Vadasz Z., Kartvelishvily Y., Gan N., Kessler O., Cohen T., Resnick M., Neeman M. and Neufeld G. (2003). Lysyl oxidase-related protein-1 promotes tumor fibrosis and tumor progression in vivo. Cancer Res. 63, 1657-1666.

Alcudia J.F., Martinez-Gonzalez J., Guadall A., Gonzalez-Diez M., Badimon L. and Rodriguez C. (2008). Lysyl oxidase and endothelial dysfunction: mechanisms of lysyl oxidase down-regulation by pro-inflammatory cytokines. Front. Biosci. 13, 2721-2727.

Andrews E.J., White W.J. and Bullock L.P. (1975). Spontaneous aortic aneurysms in blotchy mice. Am. J. Pathol. 78, 199-210.

Asuncion L., Fogelgren B., Fong K.S., Fong S.F., Kim Y. and Csiszar K. (2001). A novel human lysyl oxidase-like gene (LOXL4) on chromosome 10q24 has an altered scavenger receptor cysteine rich domain. Matrix Biol. 20, 487-491.

Borel A., Eichenberger D., Farjanel J., Kessler E., Gleyzal C., Hulmes D.J., Sommer P. and Font B. (2001). Lysyl oxidase-like protein from bovine aorta. Isolation and maturation to an active form by bone morphogenetic protein-1. J. Biol. Chem. 276, 48944-48949.

Brophy C.M., Tilson J.E., Braverman I.M. and Tilson M.D. (1988). Age of onset, pattern of distribution, and histology of aneurysm development in a genetically predisposed mouse model. J. Vasc. Surg. 8, 45-48.

Cardoso W.V. (2001). Molecular regulation of lung development. Annu. Rev. Physiol. 63, 471-494.

Contente S., Kenyon K., Rimoldi D. and Friedman R.M. (1990). Expression of gene rrg is associated with reversion of NIH 3T3 transformed by LTR-c-H-ras. Science 249, 796-798.

Csiszar K. (2001). Lysyl oxidases: a novel multifunctional amine oxidase family. Prog Nucleic Acid Res. Mol. Biol. 70, 1-32.

Decitre M., Gleyzal C., Raccurt M., Peyrol S., Aubert-Foucher E., Csiszar K. and Sommer P. (1998). Lysyl oxidase-like protein localizes to sites of de novo fibrinogenesis in fibrosis and in the early stromal reaction of ductal breast carcinomas. Lab. Invest. 78, 143-151.

Erler J.T., Bennewith K.L., Nicolau M., Dornhöfer N., Kong C., Le Q.T., Chi J.T., Jeffrey S.S. and Giaccia A.J. (2006). Lysyl oxidase is essential for hypoxia-induced metastasis. Nature 440, 1222-1226.

Fisk D.E. and Kuhn C. (1976). Emphysema-like changes in the lungs of the blotchy mouse. Am. Rev. Respir. Dis. 113, 787-797.

Fogelgren B., Polgár N., Szauter K.M., Ujfaludi Z., Laczkó R., Fong K.S. and Csiszar K. (2005). Cellular fibronectin binds to lysyl oxidase with high affinity and is critical for its proteolytic activation. J. Biol. Chem. 280, 24690-24697.

Fong S.F., Dietzsch E., Fong K.S., Hollosi P., Asuncion L., He Q., Parker M.I. and Csiszar K. (2007). Lysyl oxidase-like 2 expression is increased in colon and esophageal tumors and associated with less differentiated colon tumors. Genes Chromosomes Cancer 46, 644-655.

Gebb S.A. and Jones P.L. (2003). Hypoxia and lung branching morphogenesis. Adv. Exp. Med. Biol. 543, 117-125.

Gilad G.M., Kagan H.M. and Gilad V.H. (2005). Evidence for increased lysyl oxidase, the extracellular matrix-forming enzyme, in Alzheimer's disease brain. Neurosci. Lett. 376, 210-214.

Görögh T., Weise J.B., Holtmeier C., Rudolph P., Hedderich J., Gottschlich S., Hoffmann M., Ambrosch P., Csiszar K. (2007). Selective upregulation and amplification of the lysyl oxidase like-4 (LOXL4) gene in head and neck squamous cell carcinoma. J. Pathol. 212, 74-82.

Guarino M., Rubino B. and Ballabio G. (2007). The role of epithelial-mesenchymal transition in cancer pathology. Pathology 39, 305-318.

Higgins D.F., Kimura K., Bernhardt W.M., Shrimanker N., Akai Y., Hohenstein B., Saito Y., Johnson R.S., Kretzler M., Cohen C.D., Eckardt K.U., Iwano M. and Haase V.H. (2007). Hypoxia promotes fibrogenesis in vivo via HIF-1 stimulation of epithelial-to-mesenchymal transition. J. Clin. Invest.117, 3810-3820.

Hirai M., Ohbayashi T., Horiguchi M., Okawa K., Hagiwara A., Chien K.R., Kita T. and Nakamura T. (2007). Fibulin-5/DANCE has an elastogenic organizer activity that is abrogated by proteolytic cleavage in vivo. J. Cell Biol. 176, 1061-1071.

Holtmeier C., Görögh T., Beier U., Meyer J., Hoffmann M., Gottschlich S., Heidorn K., Ambrosch P. and Maune S. (2003). Overexpression of a novel lysyl oxidase-like gene in human head and neck squamous cell carcinomas. Anticancer Res. 23, 2585-2591.

Horn N. and Tümer Z. (2002). Menkes disease and the Occipital horn syndrome. In: Connective tissue and its heritable disorders: Molecular, genetic and medical aspects. Royce P.M. and Steinmann B. (eds). Wiley-Liss Inc., New York. pp 431-523..

Hornstra I.K, Birge S., Starcher B., Bailey A.J., Mecham R.P. and Shapiro S.D. (2003). Lysyl oxidase is required for vascular and diaphragmatic development in mice. J. Biol. Chem. 278,14387-14393.

Hough C.D., Sherman-Baust C.A., Pizer E.S., Montz F.J., Im D.D., Rosenshein N.B., Cho K.R., Riggins G.J. and Morin P.J. (2000). Large-scale serial analysis of gene expression reveals genes differentially expressed in ovarian cancer. Cancer Res. 60, 6281–6287.

Huang Y., Dai J., Tang R., Zhao W., Zhou Z., Wang W., Ying K., Xie Y. and Mao Y. (2001). Cloning and characterization of a human lysyl oxidase-like 3 gene (hLOXL3). Matrix Biol. 20, 153-157.

Ito H., Akiyama H., Iguchi H., Iyama K., Miyamoto M., Ohsawa K. and Nakamura T. (2001). Molecular cloning and biological activity of a novel lysyl oxidase-related gene expressed in cartilage. J. Biol. Chem. 276, 24023-24029.

Jeay S., Pianetti S., Kagan H.M. and Sonenshein G.E. (2003). Lysyl oxidase inhibits ras-mediated transformation by preventing activation of NF-kappa B. Mol. Cell. Biol. 23, 2251-2263.

Jourdan-Le Saux C., Tomsche A., Ujfalusi A., Jia L. and Csiszar K. (2001). Central nervous system, uterus, heart, and leukocyte expression of the LOXL3 gene, encoding a novel lysyl oxidase-like protein. Genomics 74, 211-218.

Kagan H.M. (1986). Characterization and regulation of lysyl oxidase. In Biology of extracellular matrix. Mecham. R.P. (ed). Academic Press. Orlando. pp 321-389.

Kagan H.M. (1994). Lysyl oxidase: mechanism, regulation and relationship to liver fibrosis. Pathol. Res. Pract. 190, 910-919.

Kagan H.M. and Li W. (2003). Lysyl oxidase: properties, specificity, and biological roles inside and outside of the cell. J. Cell. Biochem. 88, 660-672.

Kenyon K., Modi W.S., Contente S. and Friedman R.M. (1993). A novel human cDNA with a predicted protein similar to lysyl oxidase maps to chromosome 15q24-q25. J. Biol. Chem. 268, 18435-18437.

Kida K. and Thurlbeck W.M. (1980). The effects of beta-aminopropionitrile on the growing rat lung. Am. J. Pathol. 101, 693-710.

Kirschmann D.A., Seftor E.A., Fong S.F., Nieva D.R., Sullivan C.M., Edwards E.M., Sommer P., Csiszar K. and Hendrix M.J. (2002). A molecular role for lysyl oxidase in breast cancer invasion. Cancer Res. 62, 4478-4483.

Kuhlenbäumer G., Friedrichs F., Kis B., Berlit P., Maintz D., Nassenstein I., Nabavi D., Dittrich R., Stoll M. and Ringelstein B. (2007). Association between single nucleotide polymorphisms in the lysyl oxidase-like 1 gene and spontaneous cervical artery dissection. Cerebrovasc. Dis. 24, 343-348.

Lazarus H.M., Cruikshank W.W., Narasimhan N., Kagan H.M. and Center D.M. (1995). Induction of human monocyte motility by lysyl oxidase. Matrix Biol. 14, 727-731.

Lee J.E. and Kim Y. (2006). A tissue-specific variant of the human lysyl oxidase-like protein 3 (LOXL3) functions as an amine oxidase with substrate specificity. J. Biol. Chem. 281, 37282-37290.

Lelièvre E., Hinek A., Lupu F., Buquet C., Soncin F. and Mattot V. (2008). VE-statin/egfl7 regulates vascular elastogenesis by interacting with lysyl oxidases. EMBO J. 27, 1658-1670.

Li D.Y, Brooke B., Davis E.C., Mecham R.P., Sorensen L.K., Boak B.B., Eichwald E. and Keating M.T. (1998). Elastin is an essential determinant of arterial morphogenesis. Nature 393, 276-280.

Li W., Liu G., Chou I.N. and Kagan H.M. (2000). Hydrogen peroxide-mediated, lysyl oxidase-dependent chemotaxis of vascular smooth muscle cells. J. Cell. Biochem. 78, 550-557.

Li P.A., He Q., Cao T., Yong G., Szauter K.M., Fong K.S., Karlsson J., Keep M.F. and Csiszar K. (2004). Up-regulation and altered distribution of lysyl oxidase in the central nervous system of mutant SOD1 transgenic mouse model of amyotrophic lateral sclerosis. Brain Res. Mol. Brain Res. 120, 115-122.

Linz R. and Lutsenko S. (2007). Copper-transporting ATPases ATP7A and ATP7B: cousins, not twins. J. Bioenerg. Biomembr. 39, 403-407.

Liu X., Zhao Y., Gao J., Pawlyk B., Starcher B., Spencer J.A., Yanagisawa H., Zuo J. and Li T. (2004). Elastic fiber homeostasis requires lysyl oxidase-like 1 protein. Nat. Genet. 36, 178-182.

Liu X., Zhao Y., Pawlyk B., Damaser M. and Li T. (2006). Failure of elastic fiber homeostasis leads to pelvic floor disorders. Am. J. Pathol. 168, 519-528.

Liu G., Daneshgari F., Li M., Lin D., Lee U., Li T. and Damaser M.S. (2007). Bladder and urethral function in pelvic organ prolapsed lysyl oxidase like-1 knockout mice. BJU Int. 100, 414-418.

Loeys B., Van Maldergem L., Mortier G., Coucke P., Gerniers S., Naeyaert J.M. and De Paepe A. (2002). Homozygosity for a

missense mutation in fibulin-5 (FBLN5) results in a severe form of cutis laxa. Hum. Mol. Genet. 11, 2113-2118.

Lucero H.A. and Kagan H.M. (2006). Lysyl oxidase: an oxidative enzyme and effector of cell function. Cell. Mol. Life. Sci. 63, 2304-2316.

Lucero H.A., Ravid K., Grimsby J.L., Rich C.B., Dicamillo S.J., Mäki J.M., Myllyharju J. and Kagan H.M. (2008). Lysyl oxidase oxidizes cell membrane proteins and enhances the chemotactic response of vascular smooth muscle cells. J. Biol. Chem. 283, 24103-24117.

Löhler J., Timpl R. and Jaenisch R. (1984).Embryonic lethal mutation in mouse collagen I gene causes rupture of blood vessels and is associated with erythropoietic and mesenchymal cell death. Cell 38, 597-607.

Mäki J.M. and Kivirikko K.I. (2001). Cloning and characterization of a fourth human lysyl oxidase isoenzyme. Biochem. J. 355, 381-387.

Mäki J.M., Tikkanen H. and Kivirikko K.I. (2001). Cloning and characterization of a fifth human lysyl oxidase isoenzyme: the third member of the lysyl oxidase-related subfamily with four scavenger receptor cysteine-rich domains. Matrix Biol. 20, 493-496.

Mäki J.M., Räsänen J., Tikkanen H., Sormunen R., Mäkikallio K., Kivirikko K.I. and Soininen R. (2002). Inactivation of the lysyl oxidase gene Lox leads to aortic aneurysms, cardiovascular dysfunction, and perinatal death in mice. Circulation 106, 2503-2509.

Mäki J.M., Sormunen R., Lippo S., Kaarteenaho-Wiik R., Soininen R and Myllyharju J. (2005). Lysyl oxidase is essential for normal development and function of the respiratory system and for the integrity of elastic and collagen fibers in various tissues. Am. J. Pathol. 167, 927-936.

Min C., Kirsch K.H., Zhao Y., Jeay S., Palamakumbura A.H., Trackman P.C. and Sonenshein G.E. (2007). The tumor suppressor activity of the lysyl oxidase propeptide reverses the invasive phenotype of Her-2/neu-driven breast cancer. Cancer Res. 67, 1105-1112.

Nakamura T., Lozano P.R., Ikeda Y., Iwanaga Y., Hinek A., Minamisawa S., Cheng C.F., Kobuke K., Dalton N., Takada Y., Tashiro K., Ross Jr J., Honjo T. and Chien K.R. (2002). Fibulin-5/DANCE is essential for elastogenesis in vivo. Nature 415, 171-175.

Ono K., Tanaka T., Tsunoda T., Kitahara O., Kihara C., Okamoto A., Ochiai K., Takagi T. and Nakamura Y. (2000). Identification by cDNA microarray of genes involved in ovarian carcinogenesis. Cancer Res. 60, 5007–5011.

Palamakumbura A.H., Sommer P. and Trackman P.C. (2003). Autocrine growth factor regulation of lysyl oxidase expression in transformed fibroblasts. J. Biol. Chem. 278, 30781-30787.

Palamakumbura A.H., Jeay S., Guo Y., Pischon N., Sommer P., Sonenshein G.E. and Trackman P.C. (2004). The propeptide domain of lysyl oxidase induces phenotypic reversion of ras-transformed cells. J. Biol. Chem. 279, 40593-40600.

Payne S.L., Hendrix M.J. and Kirschmann D.A. (2006). Lysyl oxidase regulates actin filament formation through the p130(Cas)/Crk/DOCK180 signaling complex. J. Cell. Biochem. 98, 827-837.

Payne S.L., Hendrix M.J. and Kirschmann D.A. (2007). Paradoxical roles for lysyl oxidases in cancer-a prospect. J. Cell. Biochem. 101, 1338-1354.

Peinado H., Del Carmen Iglesias-de la Cruz M., Olmeda D., Csiszar K., Fong K.S., Vega S., Nieto M.A., Cano A. and Portillo F. (2005). A molecular role for lysyl oxidase-like 2 enzyme in snail regulation and tumor progression. EMBO J. 24, 3446-3458.

Postovit L.M., Abbott D.E., Payne S.L., Wheaton W.W., Margaryan N.V., Sullivan R., Jansen M.K., Csiszar K., Hendrix M.J. and Kirschmann D.A. (2008). Hypoxia/reoxygenation: a dynamic regulator of lysyl oxidase-facilitated breast cancer migration. J. Cell. Biochem. 103, 1369-1378.

Pöschl E., Schlötzer-Schrehardt U., Brachvogel B., Saito K., Ninomiya Y. and Mayer U. (2004). Collagen IV is essential for basement membrane stability but dispensable for initiation of its assembly during early development. Development 131, 1619-1628.

Ritch R. (2008). Exfoliation syndrome: beyond glaucoma. Arch. Ophthalmol. 126, 859-861.

Ritch R. and Schlötzer-Schrehardt U. (2001). Exfoliation syndrome. Surv. Ophthalmol. 2001 45, 265-315.

Roman J. (1997). Fibronectin and fibronectin receptors in lung development. Exp. Lung Res. 23, 147-159.

Rost T., Pyritz V., Rathcke I.O., Gorogh T., Dunne A.A., Werner J.A. (2003). Reduction of LOX- and LOXL2-mRNA expression in head and neck squamous cell carcinomas. Anticancer Res 23, 1565–1574.

Saito H., Papaconstantinou J., Sato H., Goldstein S. (1997). Regulation of a novel gene encoding a lysyl oxidase-related protein in cellular adhesion and senescence. J. Biol. Chem. 272, 8157-8160.

Sahlgren C., Gustafsson M.V., Jin S., Poellinger L. and Lendahl U. (2008). Notch signaling mediates hypoxia-induced tumor cell migration and invasion. Proc. Natl. Acad. Sci. USA 105, 6392-6397.

Smith-Mungo L.I. and Kagan H.M. (1998). Lysyl oxidase: properties, regulation and multiple functions in biology. Matrix Biol. 16, 387-398.

Steinmann B., Royce P.M. and Superti-Furga A. (2002). The Ehlers-Danlos syndrome. Connective tissue and its heritable disorders. In: Molecular, genetic, and medical aspects. Royce P.M. and Steinmann B. (eds). Wiley-Liss Inc. New York. pp 431-523.

Sun W., Park K.W., Choe J., Rhyu I.J., Kim I.H., Park S.K., Choi B., Choi S.H., Park S.H. and Kim H. (2005). Identification of novel electroconvulsive shock-induced and activity-dependent genes in the rat brain. Biochem. Biophys. Res. Commun. 327, 848-856.

Tarkkanen A., Reunanen A. and Kivelä T. (2008). Frequency of systemic vascular diseases in patients with primary open-angle glaucoma and exfoliation glaucoma. Acta Ophthalmol. 86, 598-602.

Tchaparian E.H., Uriu-Adams J.Y., Keen C.L., Mitchell A.E. and Rucker R.B. (2000). Lysyl oxidase and P-ATPase-7A expression during embryonic development in the rat. Arch. Biochem. Biophys. 379, 71-77.

Ten Have-Opbroek A.A. (1991). Lung development in the mouse embryo. Exp. Lung. Res. 17, 111-130.

Thomassin L., Werneck C.C., Broekelmann T.J., Gleyzal C., Hornstra I.K., Mecham R.P. and Sommer P. (2005). The Pro-regions of lysyl oxidase and lysyl oxidase-like 1 are required for deposition onto elastic fibers. J. Biol. Chem. 280, 42848-42855.

Thorleifsson G., Magnusson K.P., Sulem P., Walters G.B., Gudbjartsson D.F., Stefansson H., Jonsson T., Jonasdottir A., Jonasdottir A., Stefansdottir G., Masson G., Hardarson G.A., Petursson H., Arnarsson A., Motallebipour M., Wallerman O., Wadelius C., Gulcher J.R., Thorsteinsdottir U., Kong A., Jonasson F. and Stefansson K. (2007). Common sequence variants in the LOXL1 gene confer susceptibility to exfoliation glaucoma. Science 317, 1397-1400.

Tsuda T., Pan T.C., Evangelisti L. and Chu M.L. (2003). Prominent expression of lysyl oxidase during mouse embryonic cardiovascular development. Anat. Rec. A. Discov. Mol. Cell. Evol. Biol. 270, 93-96.

Vadasz Z., Kessler O., Akiri G., Gengrinovitch S., Kagan H.M., Baruch Y., Izhak O.B. and Neufeld G. (2005). Abnormal deposition of collagen around hepatocytes in Wilson's disease is associated with hepatocyte specific expression of lysyl oxidase and lysyl oxidase like protein-2. J. Hepatol. 43, 499-507.

Wasowicz M., Yokoyama S., Kashima K. and Nakayama I. (1996). The connective tissue compartment in the terminal region of the developing rat lung. An ultrastructural study. Acta Anat. (Basel). 156, 268-282.

Weise J.B., Csiszar K., Gottschlich S., Hoffmann M., Schmidt A., Weingartz U., Adamzik I., Heiser A., Kabelitz D., Ambrosch P. and Görögh T. (2008). Vaccination strategy to target lysyl oxidase-like 4 in dendritic cell based immunotherapy for head and neck cancer. Int. J. Oncol. 32, 317-322.

Wu M., Min C., Wang X., Yu Z., Kirsch K.H., Trackman P.C. and Sonenshein G.E. (2007a). Repression of BCL2 by the tumor suppressor activity of the lysyl oxidase propeptide inhibits transformed phenotype of lung and pancreatic cancer cells. Cancer Res. 67, 6278-6285.

Wu G., Guo Z., Chang X., Kim M.S., Nagpal J.K., Liu J., Mäki J.M., Kivirikko K.I., Ethier S.P., Trink B. and Sidransky D. (2007b). LOXL1 and LOXL4 are epigenetically silenced and can inhibit ras/extracellular signal-regulated kinase signaling pathway in human bladder cancer. Cancer Res. 67, 4123-4129.

Zhang M.C., He L., Giro M., Yong S.L., Tiller G.E. and Davidson J.M. (1999). Cutis laxa arising from frameshift mutations in exon 30 of the elastin gene (ELN). J. Biol. Chem. 274, 981-986.

Zuber J., Tchernitsa O.I., Hinzmann B., Schmitz A.C., Grips M., Hellriegel M., Sers C., Rosenthal A. and Schafer R. (2000). A genome-wide survey of RAS transformation targets. Nat. Genet 24, 144–152.

Yu Q., Horak K. and Larson D.F. (2006). Role of T lymphocytes in hypertension-induced cardiac extracellular matrix remodeling. Hypertension 48, 98-104.

Accepted November 26, 2008