Cell. Mol. Life Sci. 64 (2007) 1591 – 1596
1420-682X/07/131591-6
DOI 10.1007/s00018-007-7032-z
© Birkhäuser Verlag, Basel, 2007

Cellular and Molecular Life Sciences

# Visions & Reflections (Minireview)

# Hyaluronan

**A. Almond**

Faculty of Life Sciences, University of Manchester, Manchester Interdisciplinary Biocentre, Princess Street, Manchester, M1 7DN (United Kingdom), Fax: +44 161 30 68918, e-mail: andrew.almond@manchester.ac.uk

Received 23 January 2007; received after revision 25 February 2007; accepted 22 March 2007
Online First 14 May 2007

**Abstract.** The polysaccharide hyaluronan is an essential component of the vertebrate extracellular matrix and also produced by viruses, bacteria and fungi. Although the hyaluronan polymer is simply a disaccharide that repeats many thousands of times, it has an amazing array of biological functions and medical uses. For example, it is an efficient space filler that maintains hydration, serves as a substrate for assembly of proteoglycans and cellular locomotion, regulates cellular function and development, and is involved in tumor progression, inflammation and wound healing. Its physical properties and biocompatibility also make it of considerable importance in the development of engineered tissue, biomaterials and in clinical applications.

**Keywords.** Polysaccharide, glycosaminoglycan, proteoglycan, extracellular matrix, CD44, tissue engineering, tumour, arthritis.

## Background

Vertebrate tissue is held together by an elaborate network of macromolecules that entrap water and ions, while permitting the flow of important nutrients. The component molecules of this extracellular matrix (ECM) are not only superstructural but also regulators of cellular function, and thus they have unique biotechnological and medical applications. A case in point is the ECM polysaccharide, the glycosaminoglycan hyaluronan, which together with heparan sulphate comprises the major fraction of the carbohydrate-rich vertebrate pericellular matrix [1]. In some cases hyaluronan can make a simple ECM in the virtual absence of other macromolecules, e.g., around the ovum and developing embryo.

Meyer and Palmer first described the vertebrate polysaccharide now known as hyaluronan (usually referred to as simply 'HA') in 1934 [2]. The sugar repeat was determined to be GlcNAcβ(1→4)GlcUAβ(1→3) throughout a molecule that can have a molecular mass of several million Daltons [3], and HA is unusual among the glycosaminoglycan family because it is not sulphated or modified in any other way throughout its length. The total amount of HA in an adult human has been estimated to be 20 g, and it is abundant in the eye vitreous humor (where it was first found) and in soft connective tissues (synovial fluid of articular joints and the intercellular space of the epidermis). There is also a literature suggesting that HA may be found inside the cell [4, 5], but its function here is not yet understood.

## Structural and physical studies

It is not surprising that significant work has been directed towards understanding HA's three-dimensional organisation in solution, based on its key structural role in the ECM. However, its dynamic nature and short and repeated chemical unit often make this a frustrating exercise. The three-dimen-

sional organisation of HA was first probed by x-ray diffraction on orientated fibres around 1970 by Sheehan and Atkins [6]. It was noted that intramolecular hydrogen bonds could be made between adjacent sugar residues, which could be the basis of local secondary structure (as found in proteins). One of the most interesting observations was the exquisite sensitivity of HA to the hydration of the fibres and the cation present [7, 8]. In particular, the doubly-charged calcium ion is dominant in the solid state, inducing a contracted threefold helical form. Only when calcium is completely removed from the fibres is the true sodium form observed, which corresponds to a contracted fourfold helix [9]. Furthermore, under certain more exotic conditions, an antiparallel double helix (as found for DNA) was observed [10]; no biological role has yet been ascribed to this structure. While the x-ray studies were revealing, the solution organisation of HA remained a mystery. Based on the anomalously slow rate of periodate oxidation kinetics for HA, Scott suggested that hydrogen bonds may be present in solution [11]. Further work, including nuclear magnetic resonance (NMR) in dimethyl sulphoxide, led to a conclusion that HA is a twofold helix in solution [12], which was hypothesised to be the basis of tertiary organisation in solution [13]. However, it should be noted that such a structure was only seen in x-ray diffraction patterns at non-physiological pH, and the organisation of the chains in those fibres has never been fully elucidated. Recent structural work in aqueous solution has suggested that an exchange between hydrogen bonds and water molecules is the preferred explanation. This leads to a locally dynamic structure that is on average a contracted fourfold helix in solution [14], which is similar to the sodium hyaluronate x-ray diffraction refinement performed under physiological conditions [9]. Rheological and scattering studies have concluded that HA chains are semiflexible in solution. HA is therefore an excellent space-filling molecule that can undergo deformation as required during rapid growth and tissue remodelling, while entrapping water and ions to maintain tissue hydration and buffer the local environment. Its viscoelasticity allows it to move unhindered into vacant spaces where it can keep cells partially localised and give them a substrate on which to move. The high viscosity of HA at low shear rates and non-ideality at high shear rates, although often interpreted as evidence for the existence of stable networks [13], is simply due to chain entanglement. There is little evidence for specific interactions between chains in saline solution [15, 16]. Therefore, whether HA itself has a role in organising collagen fibrils, as has been suggested, remains an enigma.

## Aggregates and effects on cellular activity

While the unique viscoelastic properties of HA are undoubtedly central to its biological functions, a further modification to this story occurred around 1970, when the work of Hascall, Heinegård, Hardingham and Muir showed that HA chains were a substrate onto which proteoglycans could assemble [17]. This aggregation is a mechanism for the assembly of very large extracellular molecules that immobilize polyanionic sulphated polysaccharides (e.g., chondroitin sulphate and keratan sulphate) within the network of collagen fibrils, which determines the physical form of tissue as diverse as cartilage and brain [18, 19]. Not only do these macromolecules maintain the biomechanical properties of tissue, but they also define the form of the tissue during initial development, e.g., when chondrocytes are first establishing and expanding the cartilage matrix. Attachment to HA chains is via the proteoglycan G1 domain, which contains specialised lectin-like HA-binding domains (Link modules), and may involve the formation of an irreversible ternary complex with one of the link proteins [20]. Our understanding of disease and ability to perform regenerative medicine would be improved by determining the exact role of these HA-binding proteoglycans in different tissue types.

A further twist to the HA story came to light in 1972 when Pessac and Defendi showed that HA promoted the aggregation of cells [21]. Furthermore, Underhill and Toole showed in 1979 that HA could bind to the surface of cultured cells [22]. These observations have led to HA being the focus of intense study by the cell and molecular biology community. The original membrane receptor responsible for the binding of cells to HA turned out to be the lymphocyte homing receptor, CD44 [23], which remains the most widely expressed and extensively studied of the HA receptors. The CD44 protein is a cell-surface glycoprotein involved in physiological cell-cell interactions, cell adhesion, migration and pathological tumor metastasis. CD44 is thought to be the primary receptor for HA, providing cells a mechanism for sensing and attachment to HA, mediating cell-cell and cell-matrix interactions. In particular, assembly and retention of the pericellular matrix of many cells involves interactions between CD44 and HA. Although the protein occurs as several spliced variants that can interact with various ECM molecules, all contain an extracellular Link module HA-binding domain. Furthermore, most isoforms include a 70-amino acid cytoplasmic domain that has the potential for intracellular signalling [24]. Several other membrane-bound HA receptors have been identified, including LYVE-1, the stabilins and RHAMM [25]. LYVE-1 is similar to CD44, having a

single transmembrane domain and a single N-terminal HA-binding link module. It is implicated in the trafficking of cells within lymphatic vessels and lymph nodes and may be a HA transporter [26]. However, it is likely that one of the two stabilins (stabilin-2) is the primary receptor responsible for HA internalisation in liver. Interestingly, while both stabilins have a high sequence homology and are predicted to share N-terminal HA-binding link module domains and C-terminal signalling motifs, they appear to have quite separate functions; stabilin-1 is thought to regulate cell secretory processes by providing signals from the extracellular environment [27].

Additionally, some HA-binding proteins are not membrane-associated and serve to change the physical form and properties of HA within the ECM, e.g, during inflammation [28]. For instance, it was found that smooth muscle cells respond to virus infection by elaborating cell surface-bound HA in a form that is highly adhesive for mononuclear leukocytes, and it is postulated that CD44 is primarily responsible for this cell-cell interaction [29]. In separate studies inflamed colon revealed a pronounced protein-associated and filamentous-appearing HA matrix in the interstitial connective tissue between the crypts that is normally not present [30]. There is strong evidence that these fibrous structures have direct relevance for inflammatory processes and that they are associated with intracellular HA. Furthermore, within inflamed tissue and in the cumulus cell-oocyte complex, HA can be covalently decorated by transfer of heavy chain from inter-α-trypsin inhibitor by a trans-esterification reaction [31]. These derivates are thought to foster bridges between HA molecules, which modifies the local physical properties of the matrix. Specifically, ultracentrifugation experiments have shown that the bridges increase the resultant gel density.

Not all protein interactions with HA are by means of Link module domains. For example, RHAMM and related proteins bind HA via a short amino acid sequence containing multiple basic amino acids. This domain is the sole HA-binding site and is thought to generate a motility signal from exogenous HA. In spite of this, the amino acid sequence of the cytoplasmic domain does not resemble that of intracellular or cell-surface proteins known to be involved in regulating signalling of cell motility or cell cycle. The precise molecular basis by which cellular behaviour is regulated by HA remains to be unravelled.

There are numerous studies that demonstrate the critical role of HA in biological stimuli. Such observations are not unexpected, considering that feedback from specific cell-matrix interactions via surface receptors is required for coordinated cellular activity within the ECM, which is essential for multicellular tissue development and remodelling. For example, HA is implicated in neural crest cell migration, cardiac development and prostate ductal formation. It has also been found to be an adhesion substrate during wound healing and is required for cellular transformation of epithelial to mesenchymal phenotype during development [32], and production of HA appears to be increased following ischaemic stroke [33]. Furthermore, inhibition of HA-receptor function has been demonstrated to decrease cell migration and tumor growth. HA-rich ECM facilitates tumour cell migration and invasion into surrounding tissues and is also a facile route for the development of a blood supply into a tumour [34]. Not only do carcinomas frequently produce HA, but growth factors and cytokines produced by them may stimulate fibroblasts in surrounding tissue to increase production. In general, therefore, HA stimulates the growth, survival and invasiveness of carcinomas.

## Production, degradation and the differential effects of polymer size

In vertebrates, HA is produced on the inner face of the plasma membrane of fibroblasts and directly extruded (unlike other extracellular polysaccharides that are synthesised in the Golgi). In particular, synthesis occurs by hyaluronan synthase (HAS) sequentially adding sugars to the reducing terminus. Currently, three HAS genes have been identified in mammals. The differences between them are not clear, but it is a distinct possibility that their gene products each make chains with different lengths [35]. Furthermore, agreement on the point at which organisms started to produce HA during biological evolution is far from resolved; one hypothesis is that HA appeared rather late in evolution, since a HAS gene has been identified in chordates, but no corresponding gene exists in *Drosophila* [36].

One complicating factor in understanding the heritage of HA production is that HAS genes are not restricted to higher organisms; they are also found in streptococcal bacteria and viruses [37]. It is therefore possible that HA served as a primitive glycocalyx for single-celled organisms and that HAS genes have been transferred horizontally between pathogens and hosts; HA continues to be important in host-pathogen interactions and is regarded as a virulence factor when produced by pathogenic organisms that infect mammals, e.g., *Pasteurella multocida.* Furthermore, the presence of HAS in bacteria has permitted the possibility of pure HA production by fermentation methods [38]. This is a significant advance because previous extractions from biological tissue (e.g.,

rooster comb) required extensive and costly separation procedures to remove protein contaminants. Ready access to pure polysaccharide will allow the effects of HA on cellular function to be studied in a more controlled fashion, and medical preparations to be made on a larger scale and more cost effectively. The turnover of HA in mammalian tissue is astonishing, being around 15 g per day in humans [39]. This rapid turnover is due, in part, to lymphatic removal of HA from the tissues and subsequent degradation in lymph nodes and liver. It is proposed that scavenger receptors expressed on the surface of liver endothelial cells are responsible for part of this degradative process [40]. At the same time, hyaluronidase enzymes function to break down HA polymer into smaller fragments, and six homologous hyaluronidase genes have been found in the human genome. Recent work has been focussed on elucidating the function of these hyaluronidase proteins. The two primary mammalian hyaluronidases, Hyal-1 and Hyal-2, have different activities and degrade HA to either oligosaccharides or larger fragments [41]. Of the other homologues, only PH-20 has been studied in detail, which is an important sperm surface hyaluronidase with at least three functions in mammalian fertilization. In particular, it facilitates penetration of sperm through the cumulus mass to the ovum. This enzyme is often called testicular hyaluronidase and can be purchased as relatively inexpensive preparations and is therefore commonly used in experiments. Several HA-degrading enzymes can be found in bacteria, fungi and other organisms, such as leeches.

Another emerging area of HA research relates to the specific biological functions of HA fragments. While high molecular mass HA promotes tissue integrity, HA fragments signal that injury may have occurred; therefore their presence can engender changes in tissue morphology. For example, the first report of HA oligomer involvement in angiogenesis appeared in 1985, in which HA fragments limited to the 6–20 size range were shown to be angiogenic [42]. It was later shown that similar HA fragments could stimulate fibroblast proliferation and synthesis of collagen [43]. Furthermore, they appear to have important signalling roles in response to ECM degradation that takes place during inflection and wounding. For instance, it has been shown that HA fragments can deliver maturational signals to dendritic cells and stimulate cytokine production. It is hypothesised that fragments are used to identify areas of wounding through Toll-like receptors (TLRs) on endothelial cells [44]; this response has been localised to specific receptors (TLR-2 and -4) [45].

## Tissue engineering and clinical applications

The intrinsic biocompatibility of HA makes it an excellent basis for the development of engineered tissues and biomaterials for a variety of biomedical needs, including orthopedic, cardiovascular, pharmacologic and oncologic applications [46]. Chemical modifications (e.g., crosslinking) can produce biomaterials and engineered tissues that can be degraded controllably and can facilitate angiogenesis, osteointegration and cell phenotype preservation. Carefully chosen chemical conjugation can introduce new properties while maintaining the unique properties of HA. For example, it has been shown that fluorescently labelled HA retains its biological activity *in vitro* [47]. Such derivatives can be the basis of novel biochemical markers or drug-delivery methods.

Various clinical applications of HA exist, which take advantage of the viscoelastic properties of HA and its non-immunogeneity. It was initially shown by Balazs that pure preparations of HA injected into arthritic joints of horses could have a very positive effect on the clinical symptoms [48]. Today, sodium hyaluronate preparations are used for symptomatic relief from osteoarthritis. It has been reported that this preparation suppresses cartilage degeneration and release of proteoglycans from the extracellular matrix in cartilage tissues, protects the surface of articular cartilage, normalises the properties of synovial fluids, and reduces pain. The other major application is in eye surgery. In this case, a 1% solution is used during cataract eye surgery to facilitate manipulation of tissues without the risk of damaging ocular cells. It is therefore possible to use HA in many surgical procedures where manipulation and hydration of delicate tissue is necessary and adhesion to surrounding tissue needs to be minimised. Furthermore, these properties and the tissue-integrity-promoting behaviour of HA also give it many possibilities in the area of wound repair [49]. It is envisioned that many more clinical uses for HA will be found in the future.

## Concluding remarks

The simple polysaccharide HA may have served as a primitive glycocalyx and pericellular matrix that predates multicellularity, or it may have appeared rather late in biological evolution. Its unique space-filling properties make it an important constituent of the vertebrate ECM today, which fills up space and organises other macromolecules, e.g., proteoglycans. Vertebrates produce HA at their cell surfaces via a family of HA synthases, and a consequence of the ubiquitous presence of HA in the ECM makes it an

important molecule to which cells can adhere and facilitate cellular locomotion via cell-surface binding proteins and receptors (e.g., CD44). Signals transduced from HA can actually regulate cellular function, e.g., growth, adhesion and secretion. Furthermore, the polysaccharide appears to be important in some inflammations, where HA-protein complexes result in non-constitutive fibrous structures that recruit and organise immune cells. HA is also associated with cancer and stimulates growth, survival and invasion of carcinomas. It also has important roles to play in host-pathogen interactions, and HA-synthases are found in viruses, bacteria and fungi.

High molecular mass HA is a constituent of an intact matrix and results in cellular signals that promote normal cell function and growth. Turnover of HA (via specific hyaluronidase enzymes) is high in mammals, but the presence of abnormally high levels of oligosaccharide fragments signal tissue damage or invasion, which can illicit defence mechanisms, cellular differentiation and tissue morphogenesis (e.g, angiogenesis). The intrinsic biocompatibility of HA and its unique physical properties make it an important basis for drug delivery, production of biomaterials, a surgical aid and also in specific clinical applications, such as the symptomatic relief of osteoarthritis. This is quite an astonishing set of functions for a molecule that is, after all, a simple repeating disaccharide.


1 Hedman, K., Kurkinen, M., Alitalo, K., Vaheri, A., Johansson, S. and Höök, M. (1979) Isolation of the pericellular matrix of human fibroblast cultures. J. Cell. Biol. 81, 83 – 91.
2 Meyer, K. and Palmer, J. (1934) The polysaccharide of the vitreous humor. J. Biol. Chem. 107, 629 – 634.
3 Linker, A. and Meyer, K. (1954) Production of unsaturated uronides by bacterial hyaluronidases. Nature 174, 1192 – 1193.
4 Stein, G. S., Roberts, R. M., Davis, J. L., Head, W. J., Stein, J. L., Thrall, C. L., Vanveen, J. and Welch, D. W. (1975) Are glycoproteins and glycosaminoglycans components of eukaryotic genome? Nature 258, 639 – 641.
5 Evanko, S. P. and Wight, T. N. (1999) Intracellular localization of hyaluronan in proliferating cells. J. Histochem. Cytochem. 47, 1331 – 1341.
6 Atkins, E. D. T., Phelps, C. F. and Sheehan, J. K. (1972) The conformation of the mucopolysaccharides. Hyaluronates. Biochem. J. 128, 1255 – 1263.
7 Atkins, E. D. T. and Sheehan, J. K. (1973) Hyaluronates – relation between molecular conformations. Science 179, 562 – 564.
8 Sheehan, J. K. and Atkins, E. D. T. (1983) X-ray fibre diffraction study of the conformational changes in hyaluronic acid induced in the presence of sodium, potassium and calcium cations. Int. J. Biol. Macromol. 5, 215 – 221.
9 Guss, J. M., Hukins, D. W. L., Smith, P. J. C., Moorhouse, R., Rees, D. A. and Arnott, S. (1975) Hyaluronic acid: molecular conformations and interactions in two sodium salts. J. Mol. Biol. 95, 359 – 384.
10 Sheehan, J. K., Gardner, K. H. and Atkins, E. D. T. (1977) Hyaluronic acid: a double-helical structure in the presence of potassium at low pH and found also with the cations ammonium, rubidium and caesium. J. Mol. Biol. 117, 113 – 135.
11 Scott, J. E. and Tigwell, M. J. (1978) Periodate oxidation and the shapes of glycosaminoglycans in solution. Biochem. J. 173, 103 – 114.
12 Atkins, E. D. T., Meader, D. and Scott, J. E. (1980) Model of hyaluronic acid incorporating four intra-molecular hydrogen-bonds. Int. J. Biol. Macromol. 2, 318 – 319.
13 Scott, J. E. and Heatley, F. (2002) Biological properties of hyaluronan in aqueous solution are controlled and sequestered by reversible tertiary structures, defined by NMR spectroscopy. Biomacromolecules 3, 547 – 553.
14 Almond, A., DeAngelis, P. L. and Blundell, C. D. (2006) Hyaluronan: the local solution conformation determined by NMR and computer modeling is close to a contracted left-handed 4-fold helix. J. Mol. Biol. 358, 1256 – 1269.
15 Cowman, M. K. and Matsuoka, S. (2005) Experimental approaches to hyaluronan structure. Carbohydr. Res. 340, 791 – 809.
16 Gribbon, P., Heng, B. C. and Hardingham, T. E. (2000) The analysis of intermolecular interactions in concentrated hyaluronan solutions suggest no evidence for chain-chain association. Biochem. J. 350, 329 – 335.
17 Heinegård, D. and Hascall, V. C. (1974) Aggregation of Cartilage Proteoglycans. 3. Characteristics of proteins isolated from trypsin digests of aggregates. J. Biol. Chem. 249, 4250 – 4256.
18 Hascall, V. C. and Sajdera, S. W. (1969) Proteinpolysaccharide complex from bovine nasal cartilage – function of glycoprotein in formation of aggregates. J. Biol. Chem. 244, 2384 – 2396.
19 Hardingham, T. E. and Muir, H. (1972) Specific interaction of hyaluronic acid with cartilage proteoglycans. Biochim. Biophys. Acta 279, 401 – 405.
20 Franzen, A., Bjornsson, S. and Heinegård, D. (1981) Cartilage proteoglycan aggregate formation – role of link protein. Biochem. J. 197, 669 – 674.
21 Pessac, B. and Defendi, V. (1972) Cell aggregation – role of acid mucopolysaccharides. Science 175, 898 – 900.
22 Underhill, C. B. and Toole, B. P. (1979) Binding of hyaluronate to the surface of cultured cells. J. Cell Biol. 82, 475 – 484.
23 Underhill, C. B., Green, S. J., Comoglio, P. M. and Tarone, G. (1987) The hyaluronate receptor is identical to a glycoprotein of Mr 85,000 (Gp85) as shown by a monoclonal antibody that interferes with binding activity. J. Biol. Chem. 262, 13142 – 13146.
24 Thorne, R. F., Legg, J. W. and Isacke, C. M. (2004) The role of the CD44 transmembrane and cytoplasmic domains in co-ordinating adhesive and signalling events. J. Cell Sci. 117, 373 – 380.
25 Turley, E. A., Noble, P. W. and Bourguignon, L. Y. W. (2002) Signaling properties of hyaluronan receptors. J. Biol. Chem. 277, 4589 – 4592.
26 Jackson, D. G. (2004) Biology of the lymphatic marker LYVE-1 and applications in research into lymphatic trafficking and lymphangiogenesis. Acta Path. Microb. Immun. Scand. 112, 526 – 538.
27 Kzhyshkowska, J., Gratchev, A. and Goerdt, S. (2006) Stabilin-1, a homeostatic scavenger receptor with multiple functions. J. Cell Mol. Med. 10, 635 – 649.
28 Day, A. J. and Prestwich, G. D. (2002) Hyaluronan-binding proteins: tying up the giant. J. Biol. Chem. 277, 4585 – 4588.
29 de la Motte, C. A., Hascall, V. C., Calabro, A., Yen-Lieberman, B. and Strong, S. A. (1999) Mononuclear leukocytes preferentially bind via CD44 to hyaluronan on human intestinal mucosal smooth muscle cells after virus infection or treatment with Poly(I·C). J. Biol. Chem. 274, 30747 – 30755.
30 Hascall, V. C., Majors, A. K., de la Motte, C. A., Evanko, S. P., Wang, A. M., Drazba, J. A., Strong, S. A. and Wight, T. N. (2004) Intracellular hyaluronan: a new frontier for inflammation? Biochim. Biophys. Acta 1673, 3 – 12.
31 Zhuo, L. S., Hascall, V. C. and Kimata, K. (2004) Inter-alpha-trypsin inhibitor, a covalent protein-glycosaminoglycan-protein complex. J. Biol. Chem. 279, 38079 – 38082.

32 Camenisch, T. D., Spicer, A. P., Brehm-Gibson, T., Biesterfeldt, J., Augustine, M. L., Calabro, A., Kubalak, S., Klewer, S. E. and McDonald, J. A. (2000) Disruption of hyaluronan synthase-2 abrogates normal cardiac morphogenesis and hyaluronan-mediated transformation of epithelium to mesenchyme. J. Clin. Invest. 106, 349 – 360.
33 Al'Qteishat, A., Gaffney, J., Krupinski, J., Rubio, F., West, D., Kumar, S., Kumar, P., Mitsios, N. and Slevin, M. (2006) Changes in hyaluronan production and metabolism following ischaemic stroke in man. Brain 129, 2158 – 2176.
34 Toole, B. P., Wight, T. N. and Tammi, M. I. (2002) Hyaluronan-cell interactions in cancer and vascular disease. J. Biol. Chem. 277, 4593 – 4596.
35 Weigel, P. H., Hascall, V. C. and Tammi, M. (1997) Hyaluronan synthases. J. Biol. Chem. 272, 13997 – 14000.
36 Lee, J. Y. and Spicer, A. P. (2000) Hyaluronan: a multifunctional, megaDalton, stealth molecule. Curr. Opin. Cell Biol. 12, 581 – 586.
37 DeAngelis, P. L. (1999) Hyaluronan synthases: fascinating glycosyltransferases from vertebrates, bacterial pathogens, and algal viruses. Cell. Mol. Life Sci. 56, 670 – 682.
38 Yamada, T. and Kawasaki, T. (2005) Microbial synthesis of hyaluronan and chitin: new approaches. J. Biosci. Bioeng. 99, 521 – 528.
39 Laurent, U. B. G. and Reed, R. K. (1991) Turnover of hyaluronan in the tissues. Adv. Drug. Deliv. Rev. 7, 237 – 256.
40 McCourt, P. A. G. (1999) How does the hyaluronan scrap-yard operate? Matrix Biol. 18, 427 – 432.
41 Stern, R. and Jedrzejas, M. J. (2006) Hyaluronidases: their genomics, structures, and mechanisms of action. Chem. Rev. 106, 818 – 839.
42 West, D. C., Hampson, I. N., Arnold, F. and Kumar, S. (1985) Angiogenesis induced by degradation products of hyaluronic acid. Science 228, 1324 – 1326.
43 Rooney, P., Wang, M., Kumar, P. and Kumar, S. (1993) Angiogenic oligosaccharides of hyaluronan enhance the production of collagens by endothelial cells. J. Cell Sci. 105, 213 – 218.
44 Taylor, K. R., Trowbridge, J. M., Rudisill, J. A., Termeer, C. C., Simon, J. C. and Gallo, R. L. (2004) Hyaluronan fragments stimulate endothelial recognition of injury through TLR-4. J. Biol. Chem. 279, 17079 – 17084.
45 Jiang, D. H., Liang, J. R., Fan, J., Yu, S., Chen, S. P., Luo, Y., Prestwich, G. D., Mascarenhas, M. M., Garg, H. G., Quinn, D. A., Homer, R. J., Goldstein, D. R., Bucala, R., Lee, P. J., Medzhitov, R. and Noble, P. W. (2005) Regulation of lung injury and repair by Toll-like receptors and hyaluronan. Nat. Med. 11, 1173 – 1179.
46 Allison, D. D. and Grande-Allen, K. J. (2006) Hyaluronan: a powerful tissue engineering tool. Tissue Engin. 12, 2131 – 2140.
47 Seyfried, N. T., Blundell, C. D., Day, A. J. and Almond, A. (2005) Preparation and application of biologically active fluorescent hyaluronan oligosaccharides. Glycobiology 15, 303 – 312.
48 Balazs, E. A. and Denlinger, J. L. (1985) Sodium hyaluronate and joint function. J. Equine Vet. Sci. 5, 217 – 228.
49 Chen, W. Y. J. and Abatangelo, G. (1999) Functions of hyaluronan in wound repair. Wound Repair Regener. 7, 79 – 89.

To access this journal online:
http://www.birkhauser.ch/CMLS