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REVIEW

Proteomics in aortic aneurysm – What have we learnt so far?

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Aortic aneurysm is a deceptively indolent disease that can cause severe complications such as aortic rupture and dissection. In the normal aorta, vascular smooth muscle cells within the medial layer produce and sustain the extracellular matrix (ECM) that provides structural support but also retains soluble growth factors and regulates their distribution. Although the ECM is an obvious target to identify molecular processes leading to structural failure within the vessel wall, an in-depth proteomics analysis of this important sub-proteome has not been performed. Most proteomics analyses of the vasculature to date used homogenized tissue devoid of spatial information. In such homogenates, quantitative proteomics comparisons are hampered by the heterogeneity of clinical samples (i.e. cellular composition) and the dynamic range limitations stemming from highly abundant cellular proteins. An unbiased proteomics discovery approach targeting the ECM instead of the cellular proteome may decipher the complex, multivalent signals that are presented to cells during aortic remodelling. A better understanding of the ECM in healthy and diseased vessels will provide important pathogenic insights and has potential to reveal novel biomarkers.

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1 Pathophysiology of aortic aneurysms

Aortic aneurysms (AAs) are defined as an increase in the aortic diameter exceeding 1.5-fold over what would be expected based on age, gender and body mass index of the patient [1]. AA can occur at any location along the aorta from the aortic valve to the aortic bifurcation in the pelvis and are a herald of aortic dissection and rupture; both conditions are associated with high mortality [2]. The progression of AA is usually slower in the ascending aorta (0.1 cm/year) compared to the descending aorta (0.3 cm/year) [3]. Ruptured AA account for 30 000–60 000 deaths/year in the United States [4] and the mortality increases exponentially when the diameter exceeds 6 cm and 7 cm in the thoracic and abdominal aorta, respectively [1]. The principal causes are as follows:

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Abbreviations: **AA**, aortic aneurysm; **AAA**, abdominal aortic aneurysm; **BAV**, bicuspid aortic valve; **ECM**, extracellular matrix; **MMP**, matrix metalloproteinase; **TAV**, tricuspid aortic valve

1.1 Degenerative and inflammatory aortic dilatation

Degenerative and inflammatory aortic dilatation mainly affects the ascending aorta. The histological appearance of the aortic medial layer is characterized by extracellular matrix (ECM) degradation, smooth muscle cell apoptosis and destruction of medial elastic fibres [5]. Genetic conditions or infections are common causes of AA in younger patients [5, 6]. Diseases such as atherosclerosis are associated with AA in the elderly [7]. Genetic mutations associated with AA are listed in Table 1 (for details, see Lindsay et al. [8]). For several genes, knock-out mice are available to provide causal links to AA formation. Major genetic diseases with aortic involvement are Marfan syndrome [9], familial and non-familial thoracic AA and dissection [10], Ehlers-Danlos and Loeys-Dietz syndromes [11, 12]. Congenital malformations of the aortic valve are also associated with AA. Bicuspid aortic valves (BAVs) occur in 0.5–2% of the general population [13]. The aetiology has not been clearly defined, but abnormal valvulogenesis appears a major contributing factor that is genetically determined [14]. Both genetic and haemodynamic factors contribute to aortic dilatation. More rare congenital aortic malformations associated with AA are unicuspid, quadricuspid and quinticuspid aortic valves [15].

Table 1. Genes associated with human aortic aneurysm (AA) disease

Gene	Gene name	Chromosome	Human disease	Animal models
45, X	Unidentified	XO	Turner syndrome	Not associated with AA
ACTA2	α-Smooth muscle actin	10	Familial aortic aneurysm, type 6	Schildmeyer et al. (2000) [87]
ACVRL1	Activin receptor-like kinase-1	12	Hereditary haemorrhagic telangiectasia, type 2	Oh S. P. (2000) [88]
COL1A1	Collagen α-1 (I)	17	Osteogenesis imperfecta, type 1; EDS, type 7A	Rahkonen et al. (2004) [89]
COL1A2	Collagen α-1 (II)	7	Osteogenesis imperfecta, type 2,3,and 4; EDS, type 7B	Not associated with AA
COL3A1	Collagen α-1 (III)	2	Ehlers-Danlos syndrome, type 4	Liu et al. (1997) [90]
COL4A5	Collagen α-5 (IV)	X	Alport syndrome	Kashtan et al. (2010) [91]
EFEMP2	EGF containing fibulin-like extracellular matrix protein 2	11	Cutis laxa	Huang et al. (2010) [92]
ELN	Elastin	7	Cutis laxa	Li et al. (1998) [93]
ENG	Endoglin	9	Hereditary haemorrhagic telangiectasia	Arthur et al. (2000) [94]
FBN1	Fibrillin-1	15	Marfan syndrome	Carta (2006) [95]
FLNA	Filamin-A	X	Periventricular nodular heterotopia, Ehlers–Danlos variant	Feng et al. (2006) [96]
JAG1	JAGGED1	20	Alagille syndrome; Tetralogy of Fallot	Xue et al. (1999) [97]
MYH11	Smooth muscle myosin, heavy chain 11	16	Familial aortic aneurysm, type 4	Morano et al. (2000) [98]
NKX2–5	NKX2–5	5	Hypoplastic left heart syndrome 2	Zhou et al. (2012) [99]
NOTCH1	NOTCH1	9	Bicuspid aortic valve disease	Garg et al. (2005) [100]
PLOD1	Lysyl hydroxylase 1	1	EDS, type 6	Takaluoma et al. (2007) [101]
PTPN11	Protein-tyrosine phosphatase 2C	12	Noonan syndrome; LEOPARD syndrome	Araki et al. (2004) [102]
SLC2A10	Glucose transporter type 10	20	Arterial tortuosity syndrome	Cheng et al. (2009) [103]
SMAD3	SMAD family member 3	15	Loeys-Dietz syndrome, type 1C	Lu et al. (2012) [104]
TGFBR1	TGF-β receptor type 1	9	Loeys–Dietz syndrome, type 1A and 1B	Larsson et al. (2001) [105]
TGFBR2	TGF-β receptor type 2	3	Loeys–Dietz syndrome, type 2A and 2B	Langlois et al. (2010) [106]
TSC2	Tuberin	16	Tuberous sclerosis	Cao et al. (2010) [107]

1.2 Atherosclerotic aortic dilatation

Atherosclerotic aortic dilatation mainly affects the abdominal aorta. It is related to cardiovascular risk factors, in particular high blood pressure (hypertension), and thus more common in the elderly [16]. Lopes-Candales and colleagues observed a decrease in the density of vascular smooth muscle cells and increased cell death in the elastic medial layer of abdominal aortic aneurysm (AAA) when compared to normal aortas [17].

1.3 Other causes

Other rare causes for the development and rupture of AA are drug abuse, such as cocaine for the ascending aorta [18], Behcet's disease and Takayasu arteritis for the abdominal aorta [19, 20]. Takayasu arteritis can also lead to the development of AA in the ascending aorta and the aortic arch [21]. Infectious diseases, such as Syphilis and Q fever, can cause aortitis [22, 23] and contribute to the development of both ascending AA and AAA, however, to a much lesser extent in the antibiotic era.

2 Clinical management

AA is usually a slowly progressing disease that can go undetected for decades and might only be discovered accidentally during imaging for other pathologies such as spinal problems. However, fatal complications may occur, namely aneurysm rupture and dissection [24]. The interplay of several factors can expedite the progression of AA such as initial aortic size, the presence of family history and cardiovascular risk factors, in particular age, male gender and smoking [25, 26]. AA can be managed conservatively with medical treatment such as β-blockers to decrease the haemodynamic stress on the aortic wall [27]. Losartan, an angiotensin converting

enzyme inhibitor, has been proven to slow down the progression of aortic dilatation by antagonising TGF-β signalling [28, 29] and is currently tested in a multi-centre trial [30, 31]. Surgical excision of the affected area and replacement by interposition Dacron grafts is the gold standard for treatment if the dilatation of the aorta exceeds 0.5 cm/year or the ascending aorta is greater than 5 cm in diameter [1]. The conventional surgical approach carries the risks of bleeding, infection, stroke and graft failure [32]. While minimally invasive interventions are getting more frequently used in the treatment of AAA [33], the use of expanding endovascular stent-grafts is more limited in ascending AAs due to the presence of branching vessels in the aortic arch [34]. Finally, restricting physical activity and limiting practice to moderate-mild exercise is advised as a prophylactic measure [35].

Due to the life-threatening complications associated with AA [4], early diagnosis is important. Clinical assessment can reveal a pulsatile abdominal mass, aortic regurgitation and hence a diastolic murmur, pericardial involvement, signs of shock, fatigue, shortness of breath, loss of consciousness, hypotension, ischemic paresis, ischemic gut and tearing chest pain [36, 37]. All of these signs and symptoms can be vague or can mimic other pathologies such as pulmonary embolism and therefore, diagnosing aortic rupture is not straightforward. Imaging is imperative to assess the location and extent of the aneurysm to guide the correct intervention. Echocardiography is highly reproducible in diagnosing ascending AA but it has limited visual access to the distal ascending, arch and descending aorta [38]. Therefore, computed tomography and magnetic resonance angiography are the best diagnostic imaging techniques [39]. However, the high costs make them unsuitable for population-wide screening. Instead, ultrasound scanning is cheap [40]. Ideally, soluble biomarkers could prioritize patients for vascular imaging [41].

3 Proteomics of AA

Matrix metalloproteinases (MMPs) have been implicated in the development and progression of AAA [42]. The balance of expression of MMPs and their tissue inhibitors (TIMP) regulate the net degeneration of ECM and can lead to formation and rupture of AAA [43, 44]. Studies correlated MMP9 plasma level to disease severity [45]. However, it is not clear whether the rise in MMPs is causal in AA [46]. Attempts to use MMPs as a biomarker have so far not been successful [47]. Better diagnostic tools are urgently needed to identify patients at risk, monitor disease progression, promptly diagnose acute aortic complications and implement novel therapeutic and pre-emptive strategies. Although several proteomics studies have been conducted to identify biomarkers for aneurysms [41, 48–50], none of them has yet revealed biomarker candidates that are in clinical use [51, 52]. Table 2 summarises the proteomics literature on aneurysm to date.

3.1 Circulating biomarkers

Blood, plasma or serum samples from patients have been used to identify potential circulating biomarkers in aneurysms [48, 50, 53–55], but apparent discrepancies between different proteomics datasets demonstrate that candidate biomarkers will have to be carefully reassessed. For instance, Gamberi et al. observed a negative correlation between AAA and the abundance of vitamin D binding protein in plasma whereas Spadaccio et al. described a positive correlation for the same protein in serum [53, 54]. In general, plasma proteomics has three major limitations: (i) Highly abundant plasma proteins such as vitamin D binding protein are difficult to reliably quantify by proteomics. (ii) The classical plasma proteins lack vascular specificity, for example, plasma levels of vitamin D binding protein are also related to hepatic function [56]. (iii) Proteomic analysis of serum or plasma samples cannot determine whether the biomarker candidates originate from the diseased tissue. As pointed out by Anderson et al. [57], the major pitfall of proteomics is the very high dynamic range of protein abundances in plasma or serum. Low-abundance proteins including growth factors, cytokines, proteases and protease inhibitors are difficult to quantify by conventional untargeted proteomic methods. Antibody arrays are an alternative for reliably quantifying scarce proteins [58] and have already been employed for the detection of various cytokines in AAA [59–61]. Furthermore, Lamblin et al. have generated a customised antibody array for the detection of a panel of proteins of interest in macrophages and plasma from AAA patients, thus facilitating the quantification of proteases and protease inhibitors [62]. Another possibility to overcome the limitations of plasma proteomics is targeted measurements by using multiple reactions monitoring MS (MRM-MS) [63]. This approach allows the specific quantification of low-abundance biomarker candidates, but setting up and validating the transitions can be time-consuming. Nevertheless, the adaptation of such approaches will be an important step towards defining aneurysm-specific biomarkers directly in plasma or serum.

3.2 Immune cells

Immune and inflammatory processes play important roles in the pathogenesis of AAA [64]. Analysing the proteome of immune cells offers the advantages that abundant plasma proteins are omitted from the analysis and using only a specific cell type directly reveals the origin of a protein. Lamblin et al. isolated circulating monocytes from AAA patients and peripheral aortic occlusion patients (controls). The cells were differentiated into macrophages in vitro and macrophage extracts were subjected to 2D-DIGE analysis. Peroxiredoxin-1 was shown to be up-regulated in macrophages from AAA patients, reflecting the oxidative stress associated with AAA [62]. It is, however, not clear to what extent these macrophages cultured in vitro resemble those residing in the aneurysmal

Table 2. Biomarker candidates of aortic aneurysm (AA) identified by proteomics

Disease	Sample type	Method(s)	Identified potential biomarker(s)	Differential expression	Validation	Reference
AAA versus controls	Plasma	2D-DIGE, MALDI-TOF MS, LC-MS/MS	Vitamin D-binding protein	–	None	[53]
AAA versus controls	Plasma	LC-MS/MS (PAcIFIC MS)	Extracellular superoxide dismutase	+	WB	[55]
			Protein AMBP	+	WB	
			Adiponectin	+	WB	
			Carboxypeptidase B2	+	WB	
AAA, small (AD $\leq$ 4.2 cm)	Plasma	2D-DIGE, LC-MS/MS	Glycosylphosphatidylinositol-specific phospholipase D	+	none	[48]
AAA (mean diameter 5.6 cm) versus controls (diameter <2.5 cm)	Plasma	SELDI-TOF-MS	Signature ion profile	+	None	[108]
AAA (mean diameter 7.8 cm) versus controls (mean diameter 2.4 cm)	Serum	2D-DIGE, MALDI-TOF MS	Vitamin D-binding protein	+	None	[54]
			Apolipoprotein A1	–	WB	[54]
			Haptoglobin	+	WB	
AAA	ILT-conditioned medium	2D-DIGE, MALDI-MS/MS	Peroxiredoxin-1	+	WB, ELISA	[66]
AAA	ILT-conditioned medium	1D SDS-PAGE, LC-MS/MS	Clusterin	–	ELISA	[67]
			Thrombospondin-1	–	ELISA	
AAA vs peripheral aortic occlusion	Primary macrophage culture	2D-DIGE, MALDI-TOF MS	Peroxiredoxin-1	+	None	[62]
AAA, large (AD >5 cm) and small (AD 3–5 cm)	Circulating PMNs	2D-DIGE, MALDI-MS/MS	Catalase	–	WB, IHC	[65]
Ascending AA (BAV/TAV) and controls (BAV/TAV)	Aortic wall	2D-DIGE (no MS)	-	N/A	N/A	[80]
Ascending AA (BAV/TAV)	Aortic wall	2D-DIGE, MALDI-TOF MS	Hsp27	AA TAV > AA BAV	WB, ELISA, IHC	[75]
Ruptured AAA versus non-ruptured AAA	Aortic wall	2D-DIGE, LC-MS/MS	Vitronectin	(Ruptured < non-ruptured)	WB	[71]
			Peroxiredoxin-2	(Ruptured > non-ruptured)	WB	
Ascending AA (Marfan/BAV) and controls (non-BAV, non-Marfan)	Aortic wall	2D-DIGE, MALDI-TOF MS/MS, LC-MS/MS	Filamin A (C-terminal fragment)	Control < AA (BAV/Marfan)	WB	[76]

Table 2. Continued

Disease	Sample type	Method(s)	Identified potential biomarker(s)	Differential expression	Validation	Reference
Ascending aortic dissection versus ascending AA and non-aneurysmal control	Ascending aortic wall	2D-DIGE, LC-MS/MS	Alpha-1-Antitrypsin	Control > aneurysm > dissection	WB	[78]
Ascending AA/medial degeneration, no control	Aortic media	2D-DIGE, MALDI-TOF MS	Vimentin	N/A	None	[109]
			Apolipoprotein A1	N/A	None	
AAA, no control	ILT	SELDI-TOF-MS	Hemorphin-7	(Control < small < large AAA)	IHC, ELISA	[68]
AAA (3.5–8.6 cm)	Aortic wall	2D-DIGE, LC-MS/MS	Vitronectin	(small > large AAA)	WB, IHC	[69]
AAA versus controls	Inferior mesenteric veins	2D-DIGE, MALDI-TOF MS/MS	Annexin A1	–	WB	[70]
			Prohibitin	–	WB	
			Vimentin (cleaved)	+	WB	
AAA (4.8–5.8 cm), aneurysmal versus non-aneurysmal regions	Aortic wall	2D-DIGE, MALDI-TOF MS/MS	Fibrinogen	+	IHC	[72]
			Complement C4 beta	+	IHC	
AAA (AD >5 cm) versus controls (organ donors)	Aortic wall	2D-DIGE, MALDI-MS/MS	Triosephosphate isomerase	–	WB, enzyme activity assay	[74]
			Pyruvate dehydrogenase	–	WB, enzyme activity assay	
AAA versus controls	Aortic wall	1D SDS-PAGE, LC-MS/MS	Collagen XII	+	WB	[73]
			Tenascin	+	WB	
			Fibronectin	+	WB	
			Extracellular superoxide dismutase	–	WB	
			ACLP	+	WB, IHC	
			MMP-12	+	WB, IHC	
			Thrombospondin-2	+	WB	
			Periostin	+	WB	
			TIMP-1	+	WB	
Ascending AA (BAV/TAV) and non-aneurysmal controls (BAV/TAV)	Aortic wall (intima-media)	2D-DIGE/MALDI-TOF, LC-MS/MS	Lactate dehydrogenase A	+ (AA TAV > TAV)	WB, IHC	[79]
			Transglutaminase 2	+ (AA TAV > TAV)	IHC	
			Vimentin	+ (AA TAV > TAV)	IHC	
			Transthyretin	– (AA BAV < BAV)	IHC	

AAA, abdominal aortic aneurysm; BAV, bicuspid aortic valve; ILT, intraluminal thrombus; MMP, matrix metalloproteinase; PMN, polymorphonuclear neutrophils; TAV, tricuspid aortic valve; TIMP, tissue inhibitor of matrix metalloproteinase; (–), down-regulated in AA; (+), up-regulated in AA.

aortic wall. Proteomics analyses of circulating neutrophils revealed a down-regulation of catalase in neutrophils, also indicating oxidative stress associated with AAA [65].

3.3 Thrombus material

Intraluminal thrombus size correlates with the risk of aneurysm rupture but it is debated whether intraluminal thrombi contribute to AAA pathogenesis. In two studies, intraluminal thrombi were collected during surgical AAA repair and cultured ex vivo. The intraluminal thrombus-conditioned medium was subjected to proteomic analysis. Martinez-Pinna et al. detected peroxiredoxin-1 as biomarker candidate. A positive correlation of peroxiredoxin-1 serum levels and AAA could subsequently be confirmed in a cohort of 83 AAA patients and 33 controls [66]. In a similar approach by Moxon et al., thrombospondin-1 and clusterin were shown to be negatively correlated with AAA in the serum of 313 men with AAA and 690 non-aneurysmal controls [67].

3.4 Aortic tissue from aneurysm patients

Aortic tissue from aneurysm patients has been subjected to proteomics screening in order to investigate cellular and extracellular events that contribute to, or are affected by the development of AAA [68–74], ascending AA [75–79] and ascending AA associated with Marfan syndrome [76]. These publications identified several proteins with an altered abundance in AAs: Vitronectin, for example, was found to be down-regulated in ruptured compared to non-ruptured AAA and its abundance negatively correlated with aneurysm diameter [69, 71]. Other studies indicate a de-regulation of proteases and protease inhibitors: Filamin A, haemoglobin and vimentin cleavage products were more abundant in aneurysm samples [68, 70, 76], whereas levels of the protease inhibitor alpha-1-antitrypsin were decreased [78]. Modrego et al. identified metabolic changes in AA, implying alterations in glucose metabolism and anaerobic conditions in AAA [74]. Kjellqvist et al. have recently published a comprehensive proteomic analysis of changes in the aortic wall of ascending AA from patients with tricuspid aortic valve (TAVs) and BAVs compared to non-aneurysmal TAV and BAV controls, respectively (44 samples in total). Intima-media tissue was subjected to 2D-DIGE and differentially expressed spots were analysed by MALDI-TOF MS. As a validation of the DIGE analysis, most protein changes could also be identified by a gel-free LC-MS/MS analysis. Interestingly, proteins differentially expressed in ascending AAs from patients with TAVs compared to non-dilated TAVs indicate the involvement of inflammatory processes whereas BAV-specific changes were mostly observed for proteins related to wound healing. A gene expression profiling was carried out in parallel revealing very few overlaps between changes on the transcriptomic and proteomic levels in ascending AA from patients with both BAVs and TAVs. Nevertheless, this study provides proteomic evidence that the development of ascending aneurysms in patients with TAVs is related to inflammation whereas ascending aneurysms in patients with BAVs are linked to increased endothelial permeability and reduced endothelial regeneration, which might be causally related to altered haemodynamic parameters [79].

One general drawback is the lack of aortic tissue samples from healthy control patients. The non-aneurysmal aortic tissues used as controls include samples obtained during aortic valve replacement [73, 76, 80] or heart transplantation [78], non-aneurysmal regions of aortas from aneurysm patients [72] and aortas from organ donors [74]. These controls do not necessarily reflect the proteome of a healthy, normal aorta and therefore the results have to be interpreted with caution. Proteomic analyses in animal models of AAs could overcome this problem [81]. In contrast to patient material, defined and well-matched controls facilitate the identification of aneurysm-specific changes in the proteome. On the other hand, findings obtained in animal models are not necessarily applicable to human disease.

3.5 ECM in AA

The ECM serves as a scaffold; it transfers strain forces of blood flow and also regulates cell proliferation, adhesion, differentiation and migration. It plays an important role in regulating cellular events, cell–cell signalling pathways, binding, storage and modification of different components such as proteases, protease inhibitors, cytokines and growth factors. The abundant components of collagen, elastin, proteoglycans and glycoproteins provide tensile strength and flexibility. ECM remodelling is a crucial step in the development of aneurysm and a proteomics workflow tailored for the identification of extracellular proteins may yield important insights. Apart from its potential for biomarker discovery, proteomics could help to explore the underlying molecular mechanisms using an hypothesis-generating approach rather than focusing on selected genes, proteins or pathways of known importance [82]. While transcriptomic analyses can give an overview over differential gene expression at a specific time point, for example, in normal versus disease tissue, changes at the mRNA level do not necessarily correlate to the protein level. This is particularly relevant for structural components of the ECM. Some ECM proteins exhibit very long half-lives, for example, in the range of weeks to months for collagens or even decades for elastin [83]. Besides, ECM proteins are subject to pathological proteolytic degradation, which cannot be detected at the mRNA level.

We have recently developed a three-step extraction method for the enrichment of ECM components in aortic samples (Fig. 1A) [84, 85]. In a first step, the aortas are incubated with a high ionic strength buffer (0.5 M NaCl) in order to disrupt non-covalent protein–protein interactions, thereby extracting soluble proteins such as MMPs, growth factors and cytokines as well as newly synthesised matrix proteins and

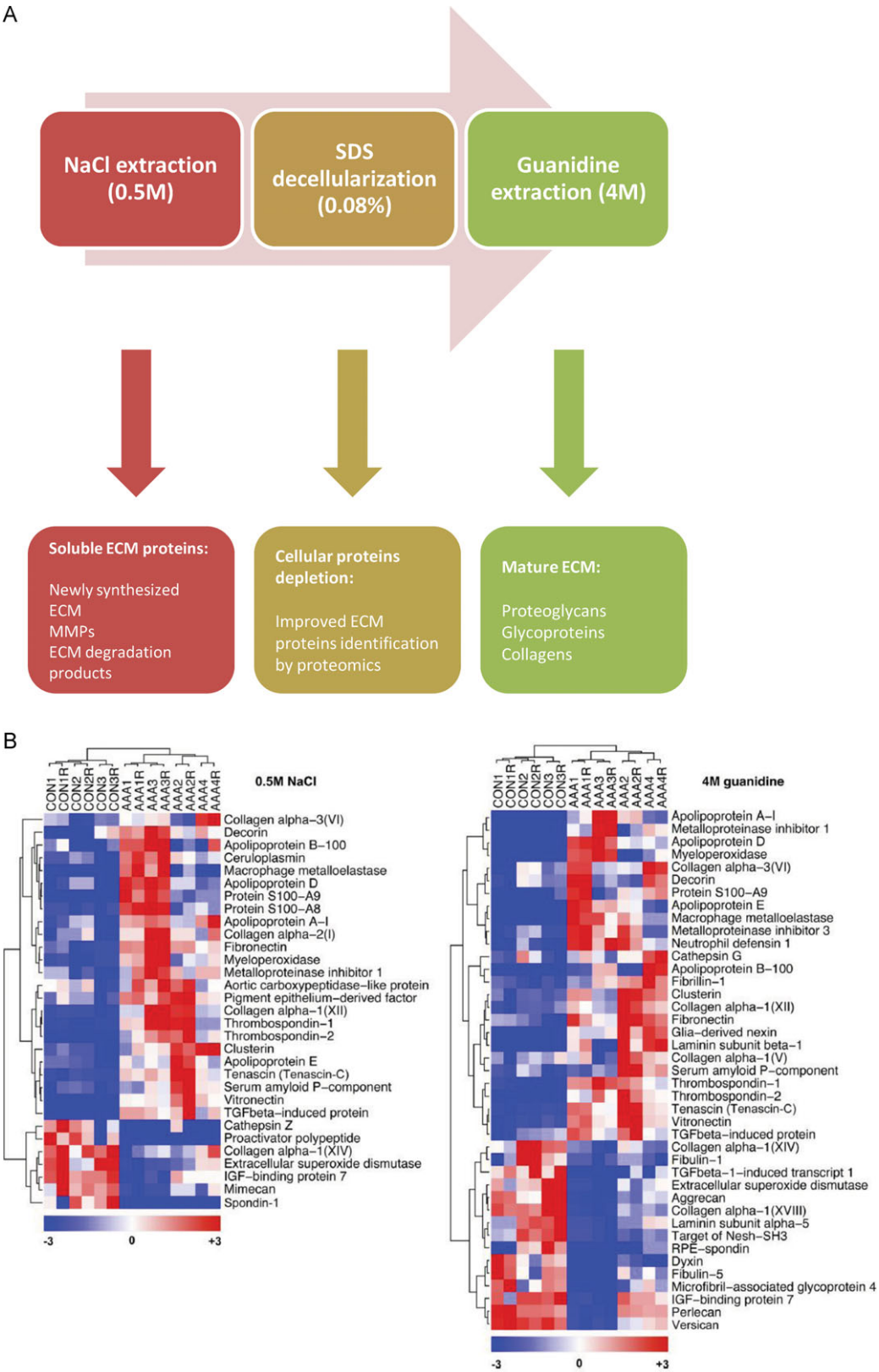

Figure 1. (A) Proteomic workflow of extracellular matrix (ECM) proteins extraction from cardiovascular tissues. (B) Hierarchical clustering of ECM proteins showing differentially expressed ECM proteins in 0.5 M NaCl and in 4 M guanidine (panel reproduced from Didangelos et al. [73]).

degradation products. The tissue is decellularised in a second step by incubation with 0.08% SDS. The remaining sample, consisting predominantly of the ECM, is then solubilised in a third step by denaturing the proteins using 4 M guanidine hydrochloride. After this extraction, the ECM proteins are deglycosylated to improve their electrophoretic properties and to facilitate their identification during MS analysis. Before the proteomics analysis, the samples are subjected to SDS-PAGE, followed by in-gel tryptic digestion and LC-MS/MS.

We have successfully applied this workflow to human AAAs (Fig. 1B), thus providing a more comprehensive view of the ECM composition and remodelling in AAA. Six glycoproteins, that is, collagen XII, thrombospondin-2, aortic carboxypeptidase-like protein (ACLP), periostin, fibronectin and tenascin were found to be up-regulated in AAA but at the same time shown to be degraded. This proteolysis could be attributed to MMP-12 activity [73]. While it is generally accepted that MMPs contribute to aneurysm formation, their vascular substrates are poorly characterised [86]. Proteomics will be the key to define the relationship between protease activity, ECM degradation and aneurysm formation.

4 Conclusion

Proteomics techniques will be invaluable tools for the elucidation of mechanisms of aneurysm formation and for the search of aneurysm biomarkers. Proteomics can reveal protein abundance, proteolysis as well as PTMs. A comprehensive proteomics analysis of the aortic ECM could be a better strategy for understanding the events leading to aortic dilation, rupture and dissection than analysing plasma, serum or homogenized tissue. The decellularisation reduces sample heterogeneity due to differences in cell composition, an inherent caveat of any omics anlaysis. Proteomics may allow insights into proteolytic activity by identifying proteases and their corresponding ECM degradation products within aneurysmal tissues. This will require optimised workflows with improved sample preparation/fractionation to enhance our understanding of the molecular mechanisms involved in aneurysm formation that may pave the way for novel diagnostic and therapeutic concepts.

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