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ADAMTS-18: A metalloproteinase with multiple functions

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1. ABSTRACT

ADAMTS-18 is a member of a disintegrin and metalloproteinase with thrombospondin motifs (ADAMTS) family of proteases, which are known to play important roles in development, angiogenesis and coagulation; dysregulation and mutation of these enzymes have been implicated in many disease processes, such as inflammation, cancer, arthritis and atherosclerosis. Mutations of ADAMTS-18 have been linked to abnormal early eye development and reduced bone mineral density. In this review, we briefly summarize the structural organization and the expression of ADAMTS-18. We will also focus on the emerging role of ADAMTS-18 in several pathophysiological conditions which include: hematological diseases, tumorgenesis, osteogenesis, eye-related diseases, central nervous system disorders, and last but not least a research perspective of ADAMTS-18 and its potential as a promising diagnostic and therapeutic target.

2. INTRODUCTION

The ADAMTS family was first identified in 1997 (1). Since that time, the ADAMTS family has grown to include 19 members. In detail, the first subgroup consists of ADAMTS-1, -4, -5, -8 and -15, while another consists of ADAMTS-9 and -20. Both divisions combine to form a larger subgroup. Another division includes ADAMTS-2, -3, and -14. ADAMTS-13 alone forms a unique group. Additionally, ADAMTS-17 and -19, ADAMTS-16 and -18, ADAMTS-7 and -12, and ADAMTS-6 and -10 consists the structurally-related ADAMTS pairs division (Figure 1) (2-5).

The ADAMTS family of enzymes is proteolytically active and is involved in normal physiological processes and in the pathogenesis of various diseases. For example ADAMTS-1 is associated with follicular rupture and ovulation (6), ADAMTS-2,

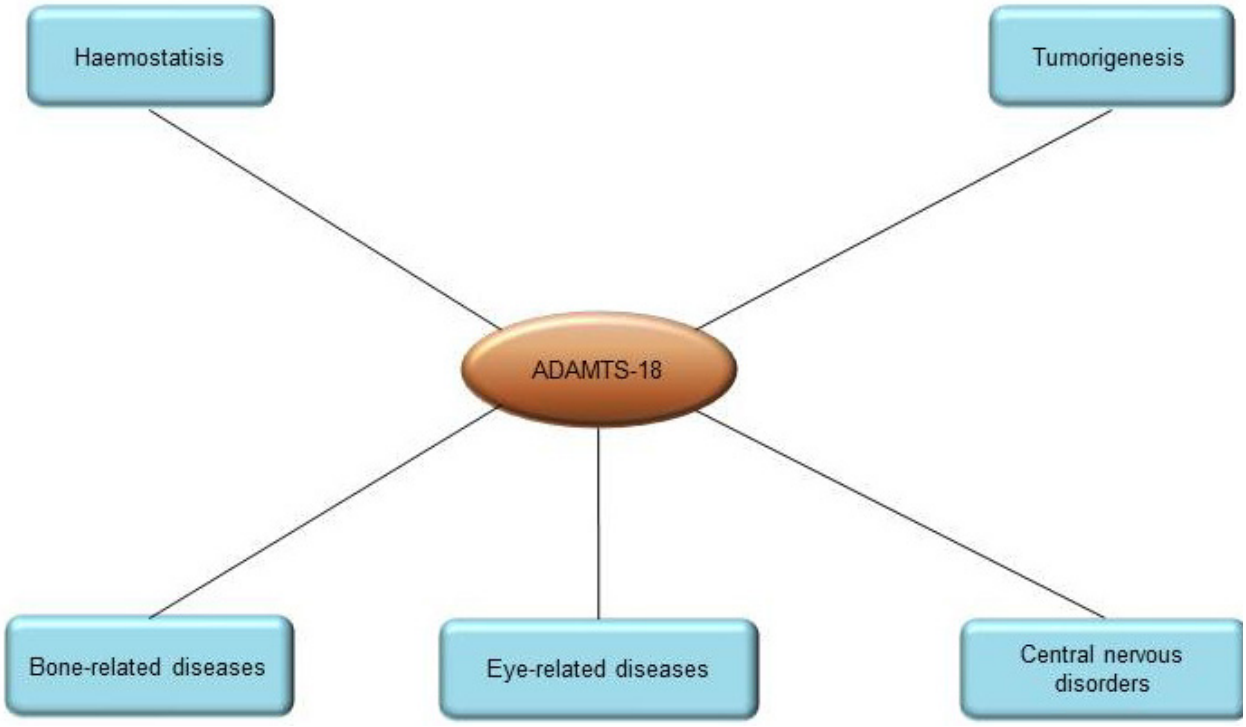

Figure 1. A schematic representation of divisions of ADAMTS family. The ADAMTS family can be categorized by the structural similarities. Group A consists of two subgroups, in detail, ADAMTS-1, -4, -5, -8 and -15 form one subgroup, while another subgroup consists of ADAMTS-9 and -20. Group B includes ADAMTS-2, -3, and -14. ADAMTS-13 alone forms a dependent group C. ADAMTS-17 and -19, ADAMTS-16 and -18, ADAMTS-7 and -12, and ADAMTS-6 and -10 consist of the group D.

ADAMTS-3, and ADAMTS-14 are procollagen N-propeptidases (7-9), ADAMTS-1, -8, and -9 participate in the inhibitory process of angiogenesis (10-14). Mutations in the ADAMTS-2 gene have been implicated in Ehlers–Danlos syndrome type 7C, which is also named "Ehlers-Danlos syndrome, dermatosparactic type" according to the new nosology, a genetic condition characterized by procollagen processing (9, 15). Both ADAMTS-4 and ADAMTS-5 have been associated with the breakdown of cartilage, through Aggrecan degradation (16-21). ADAMTS-7 and ADAMTS-12 are associated with the pathological process of arthritis (5, 22-25). Mutations in ADAMTS-13 are associated with the development of thrombotic thrombocytopenic purpura, a disease characterized by decreased numbers of circulating platelets (26).

ADAMTS-18 was first identified in 2002 by bioinformatics screening of the human genome through sequence similarity to the metalloproteinase signature of previously described ADAMTS (27). It is reported that there is a significant percentage of identity of domain architecture between ADAMTS-16 and ADAMTS-18 (overall identities: 57%, catalytic domains: 85%) by amino acid sequence alignments analysis (27). ADAMTS-18 and ADAMTS-16 are assigned to a subgroup of ADAMTS family according to their similar structure. Although the potential roles of ADAMTS-18 in different tissues have been studied over past decade, the mechanisms underlying are still not clear. Particularly the substrate of ADAMTS-18 is still unknown. In this review, we will discuss our current understanding of ADAMTS-18 by focused on its potential role in different tissues and associated diseases (Figure 2).

3. GENE, STRUCTURE, AND EXPRESSION OF ADAMTS-18

ADAMTS-18 gene is located on 16q23 in human genome (27, 28). The structure of ADAMTS-18 includes a signal peptide, a pro-domain, a metalloprotease domain, a disintegrin domain, a central TS-1 domain, a cys-rich domain, a spacer domain and a TS-1 like repeat domain (Figure 3) (27). In pro-domain, ADAMTS-18 contains a subtilisin-like pro-protein convertase cleavage site, which is the furin recognition sequences. ADAMTS-18 can be cleaved at the N terminal by furin or related pro-protein convertases leading to secretion of mature ADAMTS-18 (27). Expression of ADAMTS-18 has been found in a variety of tissues in both human and mouse. In human fetal tissues, ADAMTS-18 is expressed in lung, liver, and kidney while in adult tissues ADAMTS-18 is detected in brain, prostate, submaxillary gland, endothelium, and retina (27, 29). In addition, ADAMTS-18 is also expressed in bone according to a genome-wide bone mass candidate genes analysis (30). Recently, ADAMTS-18 is also detected in heart, skeletal muscle, spleen, pancreas, esophagus, stomach, colon, larynx, breast, cervix, placenta,

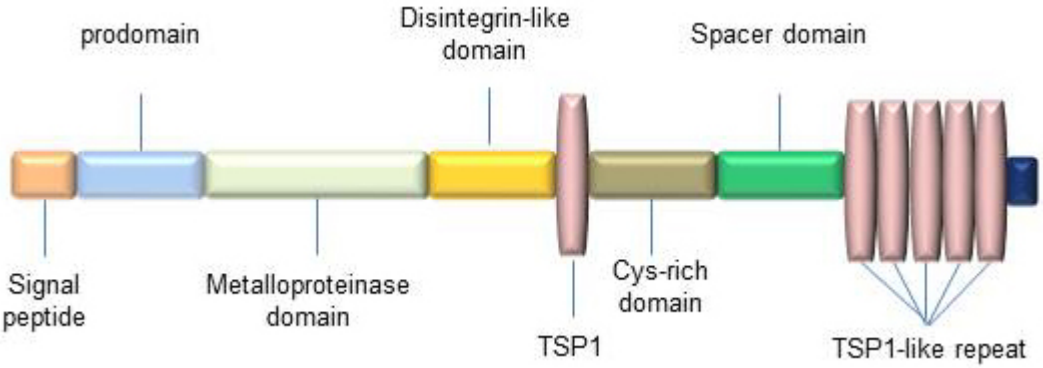

Figure 2. A schematic representation of ADAMTS-18's multi-systemic functions.To date, the potential role of ADAMTS-18 in different tissues and associated diseases which have been discovered includes haemostatic balance maintain and tumor suppression. In addition, ADAMTS-18 may play an important role in bone-, eye- and CNS-related diseases due to the significant change of its expression. However, the mechanism is still not clear.

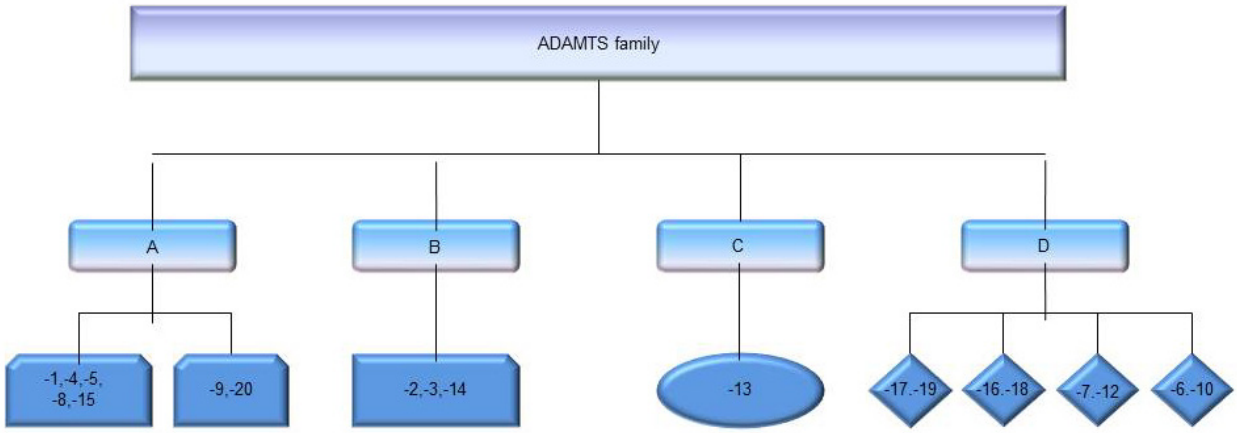

Figure 3. Domain structure and organization of ADAMTS-18. ADAMTS-18 consists of a signal peptide, prodomain, metalloprotease domain, disintegrin domain, the first thrombospondin type 1 repeat (TSP1), Cys-rich domain, spacer domain and 5 additional TSP1 repeats. Notably, the intact molecular weight (WM) of ADAMTS-18 is 135KD.

ovary, bone marrow and lymph node (28). Moreover, ADAMTS-18 truncation is detected in brain, kidney, lung, and testicle of C57BL/6 mice embryo by western blot assay (31). ADAMTS-18 is also found in the eyes of developing mice especially in the lens and retina (32).

4. ALTERNATIVE FORM AND THE TRUNCATED FRAGMENTS OF ADAMTS-18

It has been shown that thrombin cleaved ADAMTS-18 (33) between Arg775 and Ser776 (31). Analysis of putative glycosylation sites in catalytic domain indicates that there is one consensus sequence (which is called NVT) for N-glycosylation in ADAMTS-18. ADAMTS-18 protein contains three repeats in the second thrombospondin module. In the process of transcription or translation, un-spliced intron or spliced exon may produce a stop codon, which lies in the region of coding the middle of ADAMTS-18's third TS-1 like repeat domain, thus easily resulting in a truncated ADAMTS-18 motif (27). Analysis of the EST databank indicated that there was at least one EST (AV730422) corresponding to this region of ADAMTS-18, showing a sequence compatible with that reported herein (27). It was reported that there were always two bands (one is 135KDa while the other one is 75KDa) in the Western blot assay in the *in vitro* translation of ADAMTS-18 experiment (31). The size of intact ADAMTS-18 is 135KDa and the 75KDa band may indicate one of the isoforms of ADAMTS-18 (31). However, genetic codon optimization has no effect on production of ADAMTS-18 short form (31). In addition, both protease inhibitors and mutations in catalytic domain have no effect on the generation of ADAMTS-18 short form (31). The potential function of the alternative forms of ADAMTS-18 has been studied (34-36). It was reported that trADAMTS-18^{F650} (Met^{1}–Phe^{650}, cDNA constructs terminating at the corresponding conserved Phe residue of ADAMTS-18 as mentioned before (27)) was generated and purified to test its GAG-binding affinity and Aggrecanase activity comparing with ADAMTS-5, 9 and 16 (36). trADAMTS-18^{F650} had no effective GAG-binding affinity or Aggrecanase activity. In addition, trADAMTS-18^{F650}

bound heparin poorly possibly due to lacking of a consensus heparin-binding sequence (XBBXBX) (36).

The inhibition assay using antibody against C-terminal of ADAMTS-18 provides evidence that ADAMTS-18 C terminal has function. A polyclonal antibody (pAb) against active C-terminal ADAMTS-18 fragment (ADAMTS-18C) was generated from rabbits by immunizing ADAMTS-18C recombinant protein (rADAMTS-18C) (35). The binding specificity was confirmed by ELISA and Western blot assay with rADAMTS-18C and natural ADAMTS-18 protein. And the bioactivity of pAb was tested *in vivo* and it has been shown that pAb shortened the mouse tail bleeding time in a dose-dependent manner indicating the role of ADAMTS-18 C-terminal fragment in regulating thrombus stability.

5. ROLES OF ADAMTS-18 IN VARIOUS PATHPHYSIOLOGICAL CONDITIONS

5.1. Hemostasis

Endothelial cell play a central role in regulating the coagulation process (33, 37-40). Both reverse-transcription (RT)-PCR assay in human umbilical vein endothelial cells (HUVEC) and immunocytochemistry assay in human tissue confirmed that endothelial cells could express and secrete ADAMTS-18 (33). It has been shown that thrombin and TNF-α as the HUVECs activators could enhance ADAMTS-18 secretion subsequently induced platelet fragmentation, platelet disaggregation, and thrombus dissolution after thrombin cleavage of ADAMTS-18 (33, 41, 42).

It has been reported that anti-GPIIIa49-66 antibodies commonly found in HIV-1 ITP patients induce destruction of platelets through sequential activation of 12-lipoxygenase and NADPH oxidase, which suggests another mechanism of platelet activation and death (33, 43-45). In order to find the physiological ligand interacts with GPIIIa49-66, the peptide reacted with GPIIIa49-66 was identified through phage surface display screening. It has been shown that the peptide interacts with GPIIIa49-66 had 70% homology with C-terminal sequence of ADAMTS-18. Both ADAMTS-18 and anti–GPIIIa49-66 Ab did not fragment platelets in GPIIIa$^{-/-}$ knockout mice and C-terminal truncated ADAMTS-18 had no binding affinity to platelets (33). In addition, a second rabbit antibody against the N-terminal domain of ADAMTS-18 was inactive while antibody against C-terminal domain significantly inhibited ADAMTS-18 induced platelet fragmentation (33). Since C-terminal portion of ADAMTS-18 might contain its functional properties interacting with GPIIIa, three truncated rADAMTS-18 were generated to determine the C-terminal function. They were rADAMTS-385-amino acid (AA) structure (contain the GPIIIa binding site), rADAMTS-188–amino acid structure (partial active) and rADAMTS-66–amino acid structure (do not contain the GPIIIa binding site) (33). Only rADAMTS-385-AA was potent with high platelet fragmentation. Furthermore, LDH release was measured to confirm that ADAMTS-18 C-terminal could induce platelet fragmentation. Accordingly, ADAMTS-18 C-terminal peptide is likely to be the physiological ligand interacting with GPIIIa49-66 to induce platelet oxidative fragmentation (33).

Thrombin is able to cleavage full length ADAMTS-18 into 85KDa and 45KDa molecular weight (MW) while the process could be inhibited by hirudin, the specific inhibitor of thrombin(33). The thrombin cleavage site was identified through mass spectrum assay and it is between Arg775 and Ser776 (31). Since thrombin was generated during the formation of thrombus formation and ADAMTS-18 was detected in plasma after thrombin stimulation (33, 46, 47), thrombin and ADAMTS-18 might interplay in the hemostatic process. These provocative findings highlight the physiologic processes regulating thrombus dissolution. However, the impairment of thrombin/ADAMTS-18–dependent platelet dissolution could conceivably produce a prothrombotic phenotype. It is also possible that thrombin/ADAMTS-18 could act as a safeguard in thrombin compromised situations (48, 49).

However, it seems that there is no difference in platelet aggregation trace and activation and adhesion on immobilized ligand by *in vitro* experiments when comparing wild type and ADAMTS-18-deficient platelet mice (50). In addition, the expression profile data has not shown any ADAMTS-18 expression in megakaryocyte so far. Therefore, we can assume that ADAMTS-18 is not present in thrombocytes, neither WT nor ADAMTS-18-deficient mice. In that case, the function of platelets from both WT and ADAMTS-18-deficient should be the same. Furthermore, *in vitro* assays cannot simultaneously reproduce the interactions of all of the components of the hemostatic process that occur *in vivo* nor do they reflect the importance of hemodynamic factors resulting from blood flow (51).

It was reported in an ABSTRACT that ADAMTS-18 functioned as a pro-vasculature gene using ADAMTS-18-deficient mice model (50). It was confirmed that adventitial collagen deposition was increased in ADAMTS-18 knock-out mice by immunohistochemistry staining. In addition, ADAMTS-18-deficient mice had lower blood vessel density compared to wild type mice. Without ADAMTS-18, the vessels show a premature phenotype resulting in a lower blood flow. The hemodynamic change leads to a shorter time of thrombus formation. It was proven in a transient middle cerebral artery occlusion (tMCAO) model that knock-out mice got a bigger infarction size (50). Perhaps the vascular change finally affects thrombus formation in some extent.

ADAMTS-18 had been reported to be down regulated in the rapid atrial pacing (RAP) model in pigs (52). RAP model was established to mimic the atrial fibrillation (AF). AF is the most commonly sustained cardiac arrhythmia disease. AF has been characterized as an independent and important risk factor of stroke (53-56). Since ADAMTS-18 was proven to have a protective role in several thrombosis models including stroke (33), the down-regulation of ADAMTS-18 might contribute to aggravating the hemostatic imbalance in the endocardium resulting in thrombus formation (52).

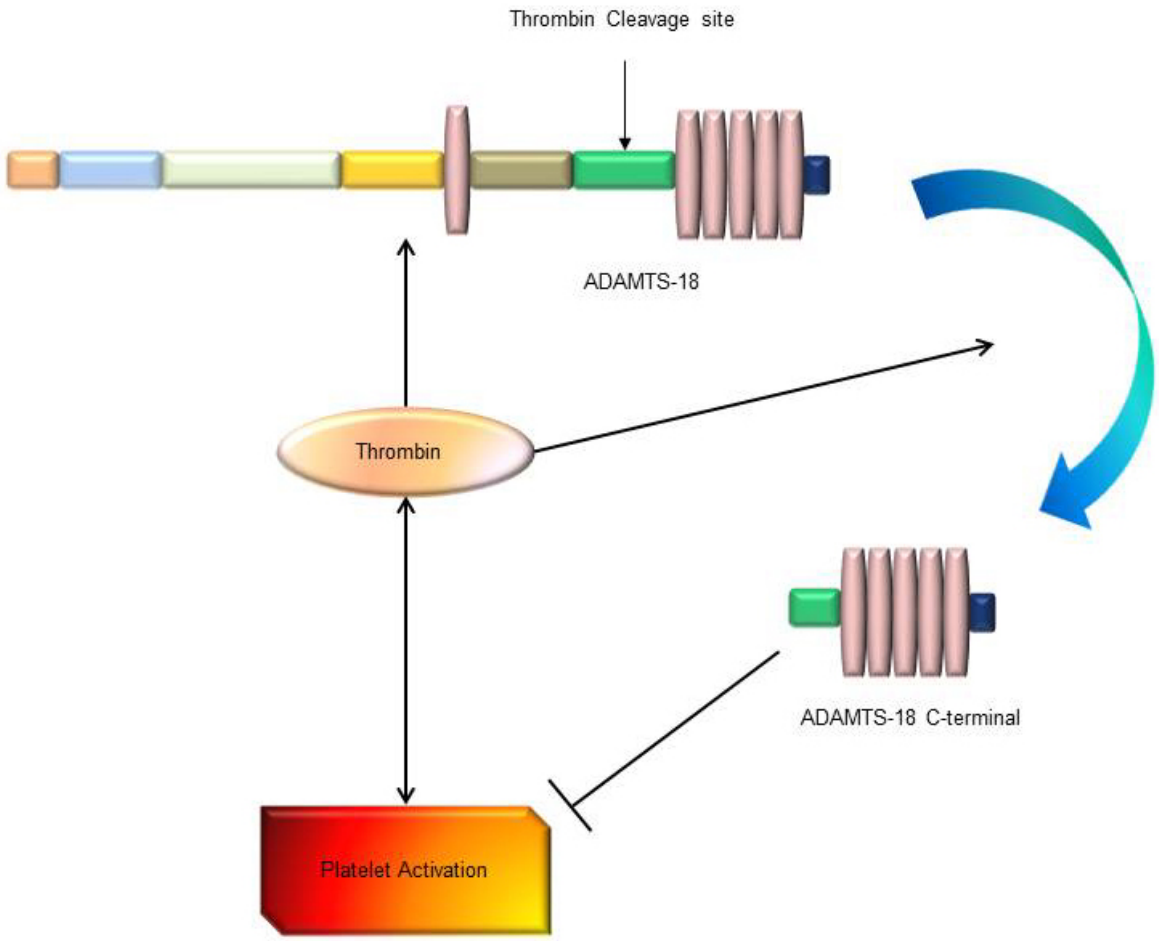

Figure 4. Interaction between thrombin and ADAMTS-18 in haemostatic balance. Endothelial cell can express ADAMTS-18. Thrombin increases the expression of ADAMTS-18 while have the ability to cleave it into 45KD active C-terminal. ADAMTS-18 C-terminal inhibits while thrombin promotes the platelet aggregation. Notably, thrombin cleavage site is in the spacer domain between Arg775 and Ser776. Abbreviations: arrows indicate a stimulatory effect; perpendicular lines indicate an inhibitory effect.

Together the current data apparently supports the concept that the interaction of ADAMTS-18 with thrombin may play an important role in maintaining the systemic hemostatic balance (Figure 4).

5.2. Tumor

The potential role of ADAMTS-18 in tumor mestastasis and tumor genesis was first suggested by the genetic linkage analysis (28). It has been shown that the loss of heterozygosity assay of 16q23 region is strongly associated with a variety of cancers. Since ADAMTS-18 is one of these genes located around 16q23 region, it has been studied as a potential oncogene. The fact that the mutations and high methylated promoter of ADAMTS-18 gene are strongly associated with a variety of tumors suggests that ADMATS-18 could be a tumor suppressor gene. (28, 57-62).

ADAMTS-18 was down-regulated in multiple samples of carcinoma cell lines such as esophageal, nasopharyngeal (28). Research has shown that there was a homozygous 16q23.1. deletion in some types of carcinoma. However the decreased expression of ADAMTS-18 was not due to this deletion. In fact, ADAMTS-18 is not in this deletion in all these described cell lines (28). The silenced or reduced expression of ADAMTS-18 is likely due to the methylation of ADAMTS-18 CGI (CpG Island, (63-65)). The tumor-specific methylation of ADAMTS-18 CGI was further supported by the fact that there is little methylation of CGI of ADAMTS-18 in non-tumor cell lines. Both pharmacological inhibitors and genetic mutation were used for the demethylation indicating that ADAMTS-18 silence was directly mediated by CGI methylation (28). High-resolution melting (HRM) analysis was used to determine the ADAMTS-18 methylation status in gastric, colorectal and pancreatic cancers as well as their adjacent normal tissues (58, 66-69). Data showed that ADAMTS-18 was highly methylated in tissues with tumor than in normal tissues. ADAMTS-18 expression level was inversely correlated with methylation status level and no evidence was identified between ADAMTS-18 methylation status and TNM staging in tumors (58, 70-72). Since ADAMTS-

18 was silenced or down-regulated in tumor, re-express ADAMTS-18 in different tumor cell lines resulted in significantly reduced colony number both in monolayer and soft agar culture indicating ADAMTS-18 suppresses both anchorage-dependent and anchorage-independent tumor growth (28). Moreover, data showed that ADAMTS-18 was methylated in a large collection of primary carcinoma samples (28). Thus, ADAMTS-18 could be used as a biomarker for both the diagnosis and prognosis of cancer.

Although ADAMTS-18 gene was not deleted in many carcinomas as described previously, it is reported that ADAMTS-18 gene was deleted in the breast cancer (57). Both single nucleotide polymorphism-comparative genomic hybridization (SNP-CGH) and the LOH analysis indicated ADAMTS-18 functioned as a tumor suppressor gene (TSG). The 16q deletion was associated with better prognosis (57).

Despite the fact that ADAMTS-18 was methylated or deleted in some carcinomas, it was reported that ADAMTS-18 mutant was involved in the melanoma (59). It has been shown that ADAMTS-18 mutant promoted growth, migration, and metastasis of melanoma.(59). Melanoma is one of the skin diseases accompanied by a series of genetic changes (73-75). 408 ADAMTS exons genes were extracted from genomic databases of 31 melanoma patients and was analyzed. Data showed that ADAMTS-18 was highly mutated and the mutant was positively selected during tumorigenesis. Analysis of cell adhesion to the extracellular matrix components indicated that mutant ADAMTS-18-expressing cells had lower adhesion ability to laminin-I compared with wild type (WT) cells resulting in facilitation of cell migration. Mutant ADAMTS-18 was identified as essential for the migration of melanoma by analysis of migration after ADAMTS-18 knock-down. This concept was further supported by *in vivo* experiment (59).

5.3. Bone-related diseases

ADAMTS-18 has been shown to be associated with several bone pathologies. Such discoveries were made through the use of genetic analysis and meta-analysis microarray (30, 76-78). It was reported that ADAMTS-18 was a bone mass candidate gene in different ethnic groups (30, 76). Bone mineral density (BMD) was a prominent osteoporosis risk factor (79-81). Masses of single nucleotide polymorphisms (SNPs) were genotyped in different ethnic groups indicating that both ADAMTS-18 and TGFBR3 were BMD candidate genes (30). In addition, Meta-analyses supported the significant associations of ADAMTS-18 and TGFBR3 with BMD (30). One SNP (rs16945612) was found to generate a binding site for the transcription factor TEL2. Furthermore, allele ‘‘C’’ of rs16945612 might enhance TEL2 factor binding ability and thus repressed the expression of ADAMTS-18 and subsequently enhanced the osteoporosis phenotype (30, 82). Moreover, a genome-wide association study of BMD in premenopausal women shows an association between femoral neck BMD and rs1826601 near the 5’ terminus of ADAMTS-18 (76). The NCBI GEO expression profiles showed that the ADAMTS-18 level was significantly lower in subjects with nonunion fractures than normal subjects (83). Thus, decreased ADAMTS-18 expression may contribute to the non-healing of skeletal fractures (30).

Evidence showed that ADAMTS-18 was correlated with kyphosis in swine (77). The human form of Kyphosis present a phenotypical over-curvature of the thoracic vertebrae , which could be either the result of degenerative diseases (such as arthritis), developmental problems (Scheuermann's disease), osteoporosis with compression fractures of the vertebrae, or trauma(84). However, Spine curvature defects had been reported in swine herds for the past three decades (85). Single nucleotide polymorphism (SNPs) associations were performed with 198 SNPs and microsatellite markers in Duroc-Landrace-Yorkshire resource (USMARC) population and F2 population. Data showed that ADAMTS-18 on SSC6 was significantly associated with kyphosis trait in the F2 population of swine ($P \leq 0.0.5$) while it seemed no association with USMARC resource herd. Thus, the association of ADAMTS-18 with kyphosis might be differed by population(77).

Studies have shown that ADAMTS-18 was approximately 2.6.4-fold up-regulated in permanent periodontal ligament (PDL) tissues compared with deciduous PDL by cDNA microarray analysis and quantitative reverse-transcription–polymerase chain reaction (qRT-PCR) analysis (78). Periodontal ligament (PDL) is known to be the most important tissue for the attenuation and conduction of masticatory forces and connection of the tooth to the alveolar jaw bone in the area surrounding the root surfaces (78, 86-89). It is reported that in permanent PDL cells IL-6 was up-regulated resulting in the increased expressions of ADAMTS-4 and ADAMTS-5 (78, 90). However, ADAMTS-4 and ADAMTS-5 were the key enzymes degrading extracellular matrix (91). Although it is unknown whether IL-6 and ADAMTS-18 were related and what their mechanisms are, it is intriguing to examine whether ADAMTS-18 and IL-6 play a part in the turnover of the extracellular matrix in permanent PDL tissues.

5.4. Eye-related diseases

It has reported that ADAMTS-18 is a novel gene associated with Knobloch syndrome (KS) by combining exome and homozygosity mapping in 13 patients of Saudi in origin and consanguineous (32). Knobloch syndrome is an autosomal recessive disease characterized by developing disorder of the eye and the occipital skull bone defect (92-96). After excluding COL18A1 (identified as the KS disease gene and 17 mutations have been reported to date (97-100)), analysis was undertaken to identify that ADAMTS-18 was associated with Knobloch syndrome (32). However the conclusion, was then corrected by the same group (92). It was reported that the KS case reported by this group was due to bi-allelic mutations in COL18A1 and ADAMTS-18 variant probably did not influence the phenotype of Knobloch syndrome disease (92).

Homozygosity mapping and whole exome sequencing were used to identify that ADAMTS-18 gene was responsible for autosomal recessive disease of retinal dystrophy (29). Inherited retinal dystrophies (IRD) were

heterogeneous disorders characterized by a progressive loss of visual acuity (VA) and deterioration of the visual field (VF) (101-103). Only a single homozygous missense variation named c.T3235 > C in the ADAMTS-18 gene was identified to be related to IRD by SNPs genotyping analysis. In addition, ADAMTS-18 was detected on human retina. Furthermore, ADAMTS-18 knockdown model was used to analyze its function. Data showed that there was a notable increase of light-induced rod photoreceptor damage in ADAMTS-18 knockdown compared with wild type fish by immunofluorescence analysis. In addition, there was a reduction of about 50% of the Rhodopsin-positive retinal areas in ADAMTS-18-deficient eyes as compared with wild type eyes. It is also important to mention that ADAMTS-18-deficient model had an aberrant central nervous system (CNS) phenotype which could be rescued by injecting the full-length coding human ADAMTS-18 synthetic mRNA (29).

The bi-allelic mutation in the gene on 16q23.1 encoding ADAMTS-18 caused MMCAT syndrome (104). MMCAT syndrome is a distinct syndrome featuring microcornea, myopic chorioretinal atrophy, telecanthus (105). The data showed that MMCAT syndrome was an autosomal recessive disease linked to chromosome of 16q23.1.

Taken together, since ADAMTS-18 was detected in eyes and ADAMTS-18 had genetic linkage to some hereditary ophthalmological diseases, ADAMTS-18 might participate in the pathological process of eye-related diseases.

5.5. Central nervous system

It was reported that ADAMTS-18 was related to the brain's white matter integrity (106). Genome-wide association analysis of 542,050 SNPs with g_{FA} was undertaken in 72-74 years old healthy people. g_{FA} is a global measure of white matter tract integrity which is derived from principal components analysis of tract-averaged fractional anisotropy measurements, accounting for 38.6.% of the individual differences across the white matter tract. Among the genome-wide association study, the strongest association was with rs7192208 whose SNP was located in an intron of the gene ADAMTS-18. Although, rs7192208 was nominally associated with almost all 12 white matter tracts by post hoc analysis, rs7192208 was most significantly linked to the left arcuate fasciculus. Moreover, the addition of the minor allele (G) in rs7192208 was associated with decreased g_{FA}. Furthermore, 30 exon probes that specific for ADAMTS-18 were tested by online resources indicated that 21 exon probes were associated with rs7192208 in brain tissue. However, rs7192208 was not associated with ADAMTS-18 in lymphoblastoid cell lines (SNPexp) or peripheral blood mononuclear cell tissue suggesting that rs7192208 influences ADAMTS-18 expression through cisacting genetic regulation in the brain (106, 107). In contrast, the association of ADAMTS-18 was not so strong by gene-based analysis possibly due to the underlying genetic architecture of the gene. It is important to mention that ADAMTS-18 was overexpressed in: the cerebellar vermis, cerebellum, cerebellar hemisphere, transverse colon, and the corpus callosum (NextBio Body Atlas; nextbio.com) (106). Further study the function of ADAMTS-18 in brain white matter would shed light on the role of ADAMTS-18 in central nervous system.

6. SUMMARY AND PERSPECTIVE

ADAMTS-18 has been shown to play a role in Osteoporosis and is genetically associated with some diseases such as: inherited retinal dystrophies (IRD), MMCAT syndrome and brain white matter integrity degeneration. In addition, ADAMTS-18 functions as a tumor suppressor gene, which is almost epigenetically silenced in all carcinoma cell lines resulting from methylation. It is deleted with some other genes in breast cancer. ADAMTS-18 C-terminal plays a unique role in maintaining hemostatic balance by inducing platelet fragmentation.

It has been reported that ADAMTS-18 can cleave Aggrecan. This was an interesting discovery because ADAMTS-18 has low efficiency at the aggrecanase site of Glu^{373}-Ala^{374}. More research must be done to identify the substrates of ADAMTS-18. This is the key to understanding the molecular mechanisms underlying ADAMTS-18 regulations of various kinds of diseases and conditions. It is a reasonable speculation that ADAMTS-18 interacts with its substrate around where it is present, such as endothelial cells. Growing evidence indicates that ADAMTS-18 is a metalloproteinase with multiple functions, the exact expression profiling, regulation and function of ADAMTS-18 in various pathophysiological processes, especially the signaling pathways and molecular events, remain to be delineated. Further research would help us to understand better of the function and regulation of ADAMTS-18.

7. ACKNOWLEDGEMENT

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Footnotes: ECM, Extracellular Matrix; ADAMTS, A Disintegrin and Metalloproteinase with Thrombospondin Motifs; MMP, matrix metalloproteinase; MW, molecular weight; AF, atrial fibrillation; RAP,rapid atrial pacing ; HRM, High-resolution melting; SNP-CGH, single nucleotide polymorphism-comparative genomic hybridization; TSG, tumor suppressor gene; BMD, Bone mineral density; PDL, permanent periodontal ligament; KS, Knobloch syndrome; VF, visual field; VA, visual acuity; CNS, central nervous system; pAb, polyclonal antibody

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