Szydełko Joanna, Litwińczuk Michał, Szydełko Magdalena. Matrix metalloproteinases and their tissue inhibitors as novel markers in invasive pituitary adenomas – a review. Journal of Education, Health and Sport. 2019;9(9):828-851. eISNN 2391-8306. DOI http://dx.doi.org/10.5281/zenodo.3460403
http://ojs.ukw.edu.pl/index.php/johs/article/view/7503

The journal has had 5 points in Ministry of Science and Higher Education parametric evaluation. § 8. 2) and § 12. 1. 2) 22.02.2019.

The authors declare that there is no conflict of interests regarding the publication of this paper.

Received: 25.08.2019. Revised: 31.08.2019. Accepted: 22.09.2019.

Matrix metalloproteinases and their tissue inhibitors as novel markers in invasive pituitary adenomas – a review

Joanna Szydełko[1a], Michał Litwińczuk[1b], Magdalena Szydełko[2c]

[1]Department of Endocrinology, Medical University of Lublin, Poland

[2]Medical Student, I Faculty of Medicine with Dentistry Division, Medical University of Lublin, Poland

[a] jszydelko@interia.pl, ORCID ID: https://orcid.org/0000-0003-3744-9058

[b] mlitwinczuk405@gmail.com, ORCID ID: https://orcid.org/0000-0002-4086-6779

[c] mszydelko@interia.pl, ORCID ID: https://orcid.org/0000-0001-6216-9934

Corresponding author:

Joanna Szydełko

Department of Endocrinology

Jaczewskiego 8 Street

20-954 Lublin, Poland

phone: +48 81 72 44 668

e-mail: jszydelko@interia.pl

Abstract

Introduction: Pituitary adenomas are generally benign central nervous system neoplasms with still increasing prevalence, especially in younger people. According to the recent data, approximately 30-45% of them invade the local structures, which make their total resection

impossible and result in high recurrence rate. Despite the huge advances in management with pituitary tumors, there are no universal biomarkers predicting their course. That is why, the two-way relationship between matrix metalloproteinases (MMPs) activities and the microenvironment of pituitary adenomas is an important object of extensive studies.

Aim of the study: This article summarizes the current knowledge about selected MMPs and their tissue inhibitors (TIMPs) in tumorigenesis of pituitary adenomas as well as their role in local invasion in different hormonally active and inactive pituitary tumors.

Description of knowledge: MMPs are a family of zinc-dependent proteolytic enzymes, which are engaged in various physiological and pathological conditions. So far, their role in different malignancies have been known for many years. Nevertheless, the possible effects of MMPs and TIMPs in pituitary adenomas are not fully understood. Recent studies suggested that MMPs' expression is significantly higher in invasive pituitary tumors as compared to non-invasive pituitary adenomas, while the expression of TIMPs is decreased, which may prove their involving in tumorigenesis.

Conclusions: MMPs may be predictive factors of the invasiveness and the higher recurrence rate in the group of patients with pituitary adenomas. Understanding the changes in the MMPs-TIMPs system and discovering of its exact mechanisms may result in applying novel screening options as well as modifying diagnostic process and treatment scheme. Therefore, further researches are required to determine the effects of MMPs and TIMPs and their role in the pathogenesis of invasive pituitary tumors.

Key words: matrix metalloproteinase, tissue inhibitor of matrix metalloproteinase, pituitary adenoma

Introduction

Pituitary adenomas, also known as pituitary neuroendocrine tumors, are generally benign central nervous system neoplasms originating from adenohypohyseal cells [1]. According to different data, they are detected in about 15-20% of patients with primary brain tumors with still increasing prevalence in recent years, especially in younger people [2-7]. Despite their non-malignant nature, slow growing, and the absence of metastases to distant organs, approximately 30-45% of them invade the local structures, such as cavernous sinus, sphenoid sinus, orbit, and clivus. What is more, they may also cause damage to optic chiasm or

even other cranial nerves as well as can destroy venous structures, internal carotid artery, and very rarely brain tissues [2-10]. Therefore, complete resection of the tumor in such cases is impossible, which in turn is related to high risk of recurrence and at the same time worsens postoperative therapeutic efficacy. The current guidelines for monitoring the pituitary tumors' course and predicting the risk of invasion, recurrence, response to adjuvant treatment are limited to a few diagnostic methods, which include hormone analysis in blood serum, functional laboratory tests, radiological techniques, usually magnetic resonance imaging (MRI), intra-operative evaluation, and histopathology examination [3-4]. It is worth to underline that there are no universal criteria for diagnosis of invasive pituitary tumors, which make it difficult to compare the results of various studies [10].

More than two-thirds of pituitary adenomas are characterized by hormonal over secretion, which lead to wide spectrum of endocrine disorders from slight symptoms to syndromes significantly debilitating the quality of life and shortening patients' life expectancy [11]. Among them the most common pathologies are prolactin-secreting adenomas (PRL-omas) (40%-60% of all pituitary tumors and about 80% of functioning adenomas), following by somatotropinomas (GH-omas) (20%), which is frequently associated with prolactin co-secretion occurring in 30-50% of patients, corticotropinomas (ACTH-omas) related with Cushing's disease (10%-15%), gonadotropinomas secreting LH or FSH (LH-omas or FSH-omas) (20%), thyreotropinomas (TSH-omas) secreting βTSH or αSU (<1%) as well as mixed tumors secreting GH and PRL (8%) [11-12]. The incidence of recurrence rate in pituitary tumors is accounted for 36% in noninvasive prolactinomas and even 100% in the course of invasive prolactinomas, about 8% in GH-secreting adenomas, and 12-45% in ACTH-secreting pituitary tumors [10,12-14]. On the other hand, the overwhelming majority of neoplasms arising in the pituitary gland are non-functioning tumors (NFPAs), which constitute about 14-54% of all pituitary adenomas [15]. These clinically silent adenomas are characterized by more aggressive course than functioning tumors, mainly due to asymptomatic nature, which usually result in delayed diagnosis and too late implementation of adequate treatment when irreversible changes have developed [15-16].

That is why, pituitary adenomas are a great challenge for clinicians of different specialties, such as endocrinologists, neurologists, gynaecologists, ophthalmologists, neurosurgeons, and pathologists.

So far, many studies were conducted in order to explain the invasive and non-invasive phenomenon of pituitary adenomas as well as to assess the differences in tumors' behavior, including their growth, local invasion, and recurrence, but the pathogenesis is high complex

and still not completely understood. The cross-talk between genetic and epigenetic factors, hormonal stimulation, and growth factors is taken into consideration in the development of various types of pituitary adenomas [6-7]. The recent studies suggested the role of matrix metalloproteinases (MMPs) and tissue inhibitors of matrix metalloproteinases (TIMPs) in the tumorigenesis of pituitary neoplasms [17-22]. The understanding of the two-way relationship between the MMPs-TIMPs system and the microenvironment of pituitary adenomas may improve both diagnostic process and therapeutic outcomes.

Aim of the study

The aim of this review was to present the role of matrix metalloproteinases and their tissue inhibitors' activities in tumorigenesis of pituitary adenomas. Moreover, we also discussed the current and future perspectives of the usefulness of these enzymes, especially gelatinases MMP-2 and MMP-9, as possible markers for invasion and recurrence in hormonally active and inactive pituitary tumors.

Materials and methods

The available literature was subjectively selected due to its usefulness in showing clinical approach to the role of matrix metalloproteinases and their tissue inhibitors in the pathogenesis of pituitary adenomas. We also presented their value as markers of invasiveness of pituitary tumors. Furthermore, data which reveals inconsistency in results was shown as well. Eligible articles in English obtained from the EBSCO and the PubMed database have been analyzed using key words in various combinations: matrix metalloproteinase, tissue inhibitor of matrix metalloproteinase, MMP-2, MMP-9, pituitary adenoma.

MMPs-TIMPs system and microenvironment of pituitary adenomas

MMPs, also called matrixins are a group of 23 zinc-dependent endopeptidases exerting pleiotropic role in physiological processes, such as embryogenesis, angiogenesis, normal tissue remodeling or wound healing as well as they are key players in many pathological conditions, inter alia rheumatoid arthritis, fibrotic disorders, pulmonary emphysema, Alzheimer's disease, disorders of cardiovascular system, diabetes mellitus, sepsis, and neoplasms [23-24]. They are involving in multi-step carcinogenesis process, that is the development, differentiation and progression of the tumors. So far, the importance of MMPs and their gene polymorphisms was determined in neoplasms of different organs, such as breast, lung, colorectal, gastric, and prostate cancers [7]. Although, their role in several tumors of central nervous system like

astrocytoma, glioblastoma, meningioma, medulloblastoma or primitive neuroectodermal tumors, has been known for many years, MMPs' value as biomarkers of invasion and migration of tumor cells, mechanisms of apoptosis and immune escape as well as angiogenesis in pituitary adenomas is still not completely clarify [10,21,25-28]. The reports regarding the MMPs' involvement in the process of invasion, their impact on recurrence rate and resistance for the treatment in pituitary adenomas are lacking and the results of current studies are not consistent [2-3,6,8-10,12,14,22,28-35]. Moreover, MMPs were proposed as markers in other pituitary disorders, such as pituitary adenoma fibrosis and pituitary apoplexy related to hemorrhagic transformation in pituitary tumors, but there is only a few research on this theme [36-37].

It also worth to underline, that multivariate analysis of selected MMPs, other serum or tissue tumor markers and radiological examination may be a useful tool in the diagnostic process and predicting the further development of the disease [38-39].

The crucial role of MMPs, the enzymes with proteolytic activities, is destabilization and degradation in regards to almost all extracellular matrix (ECM) components and basement membrane (BM), which constitute significant elements of tumor microenvironment [19,23,31,40]. It is worth mentioning that although MMPs are important for the angiogenesis, survival and expansion of tumor cells, they are only slightly produced by them. Tumor cells secrete interleukins, interferons, growth factors, and inductors of MMPs, which act at paracrine level and at the same time initiate surrounding host cells to produce required MMPs.

According, to our knowledge, the expression and activity of MMPs in the context of pituitary adenomas were described, probably for the first time, in rat anterior pituitary homogenates in 1984, but data about their exact mechanisms are still scant [41]. It is suggested that possible signaling pathways, that may be involved in the ivasiveness of pituitary adenomas, regulate the levels of MMP-2 and MMP-9 through interactions with some proteins, such as β–catenin, kazal motifs, and FRα-targeted liposomal doxorubicin, but pituitary tumor transforming gene or increased expression of the discoidin domain receptor-1 also may be related to higher pituitary tumors' invasion [10,42].

The activity of MMPs is mainly regulated by endogenous inhibitors, called tissue inhibitors of matrix metalloproteinases (TIMPs), that can play roles both MMPs' inhibitors and activators, but with different specificities. TIMPs may promote neoplasm progression by participating in the inhibition of tumor cell apoptosis, stimulation of tumor cell growth, and participation in the initiation of tumor angiogenesis [12,20,23].

So far, only a few MMPs, especially gelatinases: MMP-2 and MMP-9, MMP-1 and MMP-8 belonging to collagenases, MMP-14 (MT-1/MMP) and MMP-15 (MT-2/MMP) from

membrane type MMPs group as well as their natural, endogenous TIMPs, that is TIMP-1, TIMP-2, TIMP-3 and TIMP-4, were assessed in the context of pituitary tumors' behavior.

Besides, there are also other proteinases, a broad family of adamalysines called ADAMs that is a-disintegrin-and-metalloproteases, which are close related to MMPs and it is suggested they may also be involved in pituitary adenomas' tumorigenesis. Among them only ADAM10 and ADAM12 were evaluated in pituitary adenomas [5,43-44]. ADAM12 probably acts by inducing epithelial to mesenchymal transition and promoting cell proliferation, migration, and invasion in pituitary adenomas via EGFR/ERK signaling pathway [5]. In turn, ADAM10 may exert its effects in tumors of pituitary gland by regulating cleavage of CD44 and L1, and in this way promoting cell migration [43]. Recent study showed significantly increased ADAM12 both at mRNA and protein level in patients with invasive pituitary adenomas compared to non-invasive ones [44]. This indicated its potential as marker of cavernous sinus invasion and shed a new light on the possibilities for the treatment of pituitary tumors. Therefore, further researches are required to understand ADAMs' mechanisms involving in the invasiveness of pituitary tumors as well as their usefulness in diagnosis and therapy.

Matrix metalloproteinases and invasiveness of pituitary adenomas

So far, there are only few studies on the changes in the expression and activity of MMPs in pituitary tumors' behavior. What is interesting, but not widely known, pituitary capsule, medial wall of the cavernous sinus, and reticular fiber roof of the hypophysis are mainly composed of type IV collagen, which is the target of gelatinase activity [45]. It is also worth noted that Kawamoto et al. proved the presence of type IV collagen as a key component of the dura mater, although its main component is type I collagen [3,29]. Therefore, activity of gelatinases: MMP-2 and MMP-9, also known as type IV collagenases, is an object of extensive studies in pituitary adenoma development as well as progression and they are currently the best understood among all MMPs [2-3,8-9,12,14,22,26,29-35,39,46-48].

The first report about the expression of MMP-9 in human pituitary adenomas was described in the case-control study by Kawamoto et al. [29]. The authors revealed significantly higher activity of MMP-9 in tissue samples from adenomas cells arising from pituitary glands, which proved their ability to invade the surrounding structures [29]. Besides, the immune positive cells for MMP-9 were also found in the dura mater of the cavernous sinus in patients with invasive pituitary adenomas [29]. The positive correlation between MMP-2 and/or MMP-9 expression and invasiveness of pituitary tumors was demonstrated by many following researches [2-3,8,12,22,29-31,33-34,47]. One meta-analysis of twenty-four case control trials

involving 1320 patients indicated similarly that both MMP-2 and MMP-9 expression may be correlated with the invasiveness of pituitary tumors [10]. On the other hand, some publications showed no correlation between the expression of type IV collagenases and the invasion of adjacent structures in the course of pituitary adenomas [32,35,39]. The important results were obtained by Yokoyama S et al., who did not show any differences in the expression of MMP-9 between invasive and non-invasive non-functioning pituitary adenomas, but they suggested that increased incidence of tumor extension into the cavernous sinus is probably associated with the weakness of its medial wall [35]. It is worth to underline that the invasion of surrounding structures by pituitary adenomas was assessed by one or more methods, such as Knosp's classification, modified Hardy's classification, intraoperative observation, preoperative MRI or CT scans, and pathological examination in most analyzed case-control studies. That is why in the context of above-mentioned data, it is important from the clinical point of view that the diversity of methods used to evaluating the invasiveness of tumors may be one of the cause of divergent results.

What is more, the invasiveness of pituitary tumors is strictly connected with angiogenesis [18]. Turner HE et al. showed that angiogenesis assessed by microvascular density was related to MMP-9 expression, but it requires further research [31].

The value of the expression and activity of MMP-2 and MMP-9 as biomarkers of recurrence rate in pituitary adenomas is not proved well [3,12,14,31-32,47].

Moreover, many scientists wonder if higher levels of MMPs and their increased activity may contribute not only the regulation of tumor cells proliferation, but also pituitary hormone synthesis and secretion. It is well known established that microadanomas (tumor size <10 mm) are hormonally active in most cases and due to characteristic clinical symptoms recognized in younger age groups, whereas the diagnosis of macroadenomas (tumor size ≥10 mm), usually hormonally inactive, is connected with the mass effects. Data evaluating the correlation between MMP-2 and/or MMP-9 expression and tumor size [3,8,14,31-33,44,47-48] as well as their hormonal status are still scant and inconsistent [3,8,31-33,46-47].

Furthermore, it is worth to noted that the relationship between the expression of MMP-9 and drug treatment may exist [12]. Gültekin GD et al. suggested that dopamine agonists, that is bromocriptine and cabergoline, are able to reduce the level of MMP-9 expression from strong to moderate intensity in patients with prolactinomas, who were treated before operation [12]. However, the extension of the studied group is required and these observations should be also evaluated in other hormonally active pituitary tumors.

Moreover, there are single reports about MMP-8 and MMP-14 as potential biomarkers of invasiveness in pituitary tumors, but further studies in a larger scale are necessary [6,9,44].

It is also of high interest in recent years if MMPs gene polymorphisms may play a role in the development of pituitary adenomas, their invasion of local structures, recurrence rate, and tumor activity, but researches on this theme are still lacking [4,7,16]. The recent studies revealed that MMP-1 gene polymorphisms are associated with the invasiveness of pituitary adenomas, while MMP-9 gene polymorphisms may play a pivotal role in nonrecurrent, inactive as well as both invasive and non-invasive pituitary tumors' development [4,7,16]. What is more, the methylation status of the MMP-14 promoter was studied in pituitary adenomas' behavior, but any correlations were observed in tumors' functioning and risk of recurrence, only positive association with the male gender was noted in case of methylated MMP-14, whereas unmethylated one correlated with the female gender [49].

Taking into consideration the above-mentioned data, MMPs have a great potential as targets of anti-cancer drugs, especially high hope rises using natural or synthetic inhibitors of MMPs in this group of neoplasms in the future.

Tissue inhibitors of matrix metalloproteinases and invasiveness of pituitary adenomas

While MMPs are widely studied in pituitary adenomas development and invasiveness, the role of TIMPs and its value in the MMPs-TIMPs system homeostasis in tumors arising from pituitary gland is poorly understood. So far, all TIMPs, such as TIMP-1, TIMP-2, TIMP-3, and TIMP-4 were discovered in human pituitary adenomas, but the extent of their expression differed [20]. It is known that TIMPs and MMPs are multifunctional molecules and exert contradictory effects in progression of most tumors. Therefore, when MMPs expression and activity decrease, TIMPs ones increase and these interactions should provide the balance between them. On the other hand, TIMPs may exert both stimulatory or inhibitory effects on MMPs, which make their relationship highly complicated.

There are only a few studies about TIMPs in the context of pituitary adenomas and their association with invasiveness. Most of them evaluated TIMP-1 and/or TIMP-2 expression and activity [6,12,22,30,32,46], but there is also a single research, which assessed the role of TIMP-3 [50]. However, data are conflicting and further researches in a larger scale are needed. It is also worth mentioning that one of the latest studies proved the role of TIMP-1 in postoperative prognosis of survival rate [22]. The authors showed that postoperative survival rate of patients was connected with low expression of MMP-9 and high expression of TIMP-1 [20].

TIMPs and their exogenous synthetic ones seem to be promising therapeutic targets in many neoplasms. The discovering of TIMPs exact mechanisms will likely result in innovative approach to diagnostic process and rationalizing treatment methods of these highly prevalent disorders as pituitary adenomas are.

Alterations in the expression of selected MMPs and TIMPs as well as their role in the tumorigenesis of pituitary adenomas in case-control studies from 1996 up to August 2019 were summarized in table 1 [Table 1].

Conclusions

To conclude, it is worth to emphasize that MMPs, particularly MMP-2 and MMP-9, may be predictive factors for invasion in the group of patients with incidentally detected pituitary adenomas. Moreover, the changes in the expression of MMPs and TIMPs may be valuable for the evaluation of postoperative tumor recurrence rate. The discovering of their exact mechanisms will likely result in modifying novel screening options as well as innovative approach to diagnostic process and treatment scheme. Therefore, further researches are required to determine the effects of the MMPs-TIMPs system and its role in the pathogenesis of pituitary tumors.

Table 1. Analysis the association between the expression of selected matrix metalloproteinases, their tissue inhibitors and local invasion, recurrence rate, tumor size, and hormonal status of pituitary adenomas in case-control studies from 1996 to 2019.

Study (Reference)	Type of pituitary tumor	Criteria of invasiveness	Invasive pituitary adenomas vs non-invasive pituitary adenomas	Matrix metalloproteinase	Tissue inhibitor of matrix metalloproteinase	Level of expression (mRNA/protein)	Correlation with invasiveness	Correlation with recurrence rate	Correlation with tumor size	Correlation with hormonal status	Comments
Kawamoto H et al., 1996 [29]	PRL: 2 GH: 1 Mixed: 1 NFPA: 3	intraoperative observation	3 vs 4	MMP-9 (+)	not examined	protein	correlation	not examined	not examined	not examined	high level activity of MMP-9 was observed in all 3 invasive PAs
Kawamoto H et al., 1996 [30]	PRL: 4 GH: 5 Mixed: 1 NFPA: 12	intraoperative observation	9 vs 13	MMP-9 (+)	TIMP-1 (-)	protein	correlation (MMP-9)	not examined	not examined	not examined	MMP-9 was significantly higher in invasive PAs as compared to non-invasive ones, TIMP-1 secretion was undetectable in all PAs
Tomita T., 1997 [46]	PRL: 17 GH: 5 ACTH: 3 NFPA: 6	not examined	not examined (normal vs adenoma tissue)	MMP-2, MMP-9 (+/-)	TIMP-1, TIMP-2 (+/-)	protein	not examined	not examined	not examined	no correlation	normal anterior-pituitary cells all contained MMPs and lesser amount of TIMPs, significantly fewer MMPs and TIMPs were observed in PAs tissues
Turner HE et al., 2000 [31]	PRL: 24 NFPA: 31	modified Hardy's classification	11 vs 8	MMP-9 (+)	not examined	protein	correlation (PRLoma)	correlation (NFPA)	no correlation	no correlation	the correlation between MMP-9 expression and angiogenesis assessed

											by microvascular density was noted
Yokoyama S et al., 2001 [35]	NFPA: 20	Knosp's classification	10 vs 10	MMP-9 (+)	not examined	protein	no correlation	not examined	not examined	not examined	no statistical difference in MMP-9 immunostaining between invasive and non-invasive groups was noted
Knappe UJ et al., 2003 [32]	PRL: 14 GH: 17 ACTH: 21 TSH: 1 NFPA: 22	invasion of surrounding structures according to preoperative MRI scans, intraoperative observation	50 vs 34	MMP-2, MMP-9 (+/-)	TIMP-2 (+/-) (overexpressed in non-invasive n=31 as compared to invasive n=44)	protein	no correlation (MMP-2, MMP-9)	no correlation (none of the tumors showed progression of any MMPs or TIMP expression)	no correlation	correlation (increased expression of MMP-2, MMP-9, TIMP-2 in ACTH PAs as compared to other adenomas)	positive reactions (diffuse expression) (invasive vs non-invasive): MMP-2 74% of 50 cases (20/27 vs 17/23) MMP-9 49% of 75 cases (21/44 vs 16/31) TIMP-2 88% of 75 cases (36/44 vs 30/31) MMP-2/MMP-9/TIMP-2 were positive in: PRL: 6 of 9/8 of 14/11 of 14 GH: 8 of 10/7 of 17/14 of 17 ACTH: 18 of 20/15 of 21/21 of 21 TSH: excluded

											NFPA: 5 of 12/6 of 21/19 of 22 primary and recurrent tumors were investigated in 6 cases (ACTH: 4, PRL: 1, NFPA: 1)
Liu W et al., 2005 [8]	PRL: 11 GH: 12 ACTH: 4 TSH: 1 FSH:1 NFPA: 25	Knosp's classification, invasion of surrounding structures according to preoperative MRI scans, intraoperative observation	12 vs 42	MMP-2, MMP-9 (+)	not examined	mRNA, protein	correlation (MMP-2, MMP-9)	not examined	no correlation	no correlation	the expression of MMP-2 and MMP-9 at mRNA level was assessed in 16 cases (4 invasive and 12 non-invasive), the significantly higher percentage of MMP-2 mRNA/β-actin mRNA and MMP-9 mRNA/β-actin in invasive vs non-invasive group, there were significant correlations between mRNA and immunohistochemistry score in MMP-2 and MMP-9
Liu W et al., 2005 [33]	PRL: 12 GH: 11 ACTH: 4 TSH: 1 FSH: 1 NFPA: 25	invasion of surrounding structures according to preoperative MRI or CT scans,	12 vs 42	MMP-2 (+/-)	not examined	mRNA, protein	correlation	not examined	no correlation	no correlation	MMP-2 was assessed at protein level in 54 tissue samples from PAs and at mRNA level in 16 ones, 11 microadenomas and 43

		intraoperative observation									macroadenomas were evaluated
Hussaini IM et al., 2007 [2]	NFPA: 8	confirmation of invasion of surrounding structures pathologically	3 vs 5	MMP-9 (+)	not examined	mRNA, protein	correlation	not examined	not examined	not examined	very low activity of MMP-2 in both invasive and non-invasive PAs was observed in the study
Yamada S et al., 2007 [39]	NFPA: 40	confirmation of invasion of surrounding structures pathologically	20 vs 20	MMP-9 (+)	not examined	protein	no correlation	not examined	not examined	not examined	there was no significant difference in MMP-9 expression in the clinically NFPAs with and without cavernous sinus invasion, cavernous sinus invasion occurred most frequently in silent corticotroph adenomas (85%), Subtype 3 adenomas (67%), null cell adenomas (38%), and silent gonadotroph adenomas (11%) according to histology examination
Gong J et al., 2008 [3]	PRL: 3 GH: 11 ACTH: 9 NFPA: 50	intraoperative observation, confirmation of invasion of surrounding structures pathologically	46 vs 27	MMP-9 (+)	not examined	mRNA, protein	correlation (MMP-9 expression both at mRNA and protein levels was significantly	correlation (MMP-9 expression both at mRNA and protein levels was	correlation (MMP-9 expression both at mRNA and protein levels was positively correlated with	no correlation (regarding MMP-9 expression, no difference in	histological subtypes of PAs (non-invasive vs invasive): null cell adenomas (7/16), gonadotrophic adenomas (8/9), silent ACTH adenomas

							increased in the majority of invasive PA)	increased in all redo-surgery tumors vs primary ones and significantly increased at mRNA level in 7 re-operated NFPAs)	increase of tumor size in NFPAs as well as MMP-9 expression was significantly higher in extending adenomas as compared to microadenomas, but in FPAs, its expression in microadenomas was higher than in macroadenomas and was not statistically significantly different from extending adenomas, in all adenomas – no significant changes between micro- and macroadenomas were observed, but significant increase of MMP-9	invasiveness between FPAs and NFPAs was observed, but MMP-9 expression in invasive silent-ACTH adenomas, active ACTH-omas, PRL-omas was slightly increased at mRNA level and significantly higher at protein level as compared to other invasive subtypes)	(1/7), silent GH adenomas (1/1), ACTH-oma (6/3), GH-oma (4/7), PRL-oma (0/3), 60 primary and 13 recurrent (n=1) or persistent (n=12) tumors, NPFAs/FPAs: microadenoma (8/13), macroadenoma (4/3), suprasellar extension (21/2), cavernous sinus extension (7/3), sphenoid sinus extension (10/2)

									expression was noted in extending tumors)		
Qu X et al., 2010 [47]	PRL: 25 GH: 10 ACTH: 2 Mixed: 17 NFPA: 20	modified Hardy's classification, invasion of surrounding structures according to preoperative MRI/CT scans, intraoperative observation, confirmation of invasion of surrounding structures pathologically	40 vs 34	MMP-2 (+/-)	not examined	protein	correlation (MMP-2 expression was significantly higher in invasive PAs (32/40, 80%) as compared to non-invasive PAs (14/34, 41.2%)	correlation (positive relation between MMP-2 expression and tumor size in invasive PAs)	correlation	no correlation	4 recurrent adenomas (3 inasive and 1 non-invasive), MMP-2 was expressed in 15 of 31 microadenomas and in 31 of 43 macroadenomas
Qiu L et al., 2011 [34]	PRL: 28 GH: 18 ACTH: 3 TSH: 1 Mixed: 1 NFPA: 24	modified Hardy's classification, Knosp's classification, invasion of surrounding structures according to preoperative MRI scans,	40 vs 35	MMP-9 (+/-)	not examined	mRNA, protein	correlation (MMP-9 expression both at mRNA and protein levels was statistically higher in patients with invasive PAs than in non-invasive ones)	not examined	not examined	not examined	MMP-9 was expressed at protein level in 37 of 40 invasive PAs and in 20 of 35 non-invasive PAs

		confirmation of invasion of surrounding structures pathologically									
Mao JH et al., 2015 [6]	not examined	invasion of surrounding structures according to preoperative imaging, intraoperative observation, confirmation of invasion of surrounding structures pathologically	30 vs 30	MMP-8 (+)	TIMP-1 (+)	mRNA, protein	correlation (significantly overexpressed MMP-8 and decreased TIMP-1 expression in invasive PAs)	not examined	not examined	not examined	MMP-8 expression in the serum was increased and serum TIMP-1 expression was significantly decreased in invasive PAs patients compared to non-invasive ones
Hui P et al., 2015 [9]	PRL: 18 GH: 13 ACTH: 6 TSH: 4 LH or FSH: 4 Mixed: 16 NFPA: 21	Knosp's classification	37 vs 45	MMP-1, MMP-2, MMP-9, MMP-14, MMP-15 (+)	not examined	mRNA (all), protein (MMP-2, MMP-14)	correlation (MMP-14 both at mRNA and protein level)	not examined	not examined	not examined	all assessed MMPs were expressed at higher levels in invasive PAs as compared to non-invasive PAs or normal pituitary gland tissue, MMP-14 expression compared to other MMPs was elevated only in invasive PAs

Gültekin GD et al., 2015 [12]	PRL: 57	modified Hardy's classification, Knosp's classification, intraoperative observation	35 vs 22	MMP-9 (+)	TIMP-1 (+/-), TIMP-2 (+/-)	protein	correlation (significantly higher MMP-9 and TIMP-2 expression in invasive PAs)	correlation (TIMP-2)	not examined	not examined	surgical treatment was performed in all patients (n=22) in non-invasive group and in 31 of 35 patients in invasive group for the first time, 4 patients with invasive PAs were operated for the second time, there was no correlation between TIMP-1 expression (in 31 of 57 PAs) and invasion, TIMP-2 was expressed in 32 of 57 PAs
Chen Z et al., 2015 [26]	PRL: 40 NFPA: 30	data not available	data not available	MMP-9 (+)	not examined	mRNA, protein	data not available	data not available	data not available	data not available	MMP-9 expression both at mRNA and protein level was higher in PAs than in normal pituitary gland tissue
Wang J et al., 2016 [44]	PRL: 1 GH: 3 ACTH: 1 NFPA: 30	Knosp's classification, intraoperative observation	19 vs 16	MMP-14	not examined	mRNA, protein	correlation	not examined	no correlation	no correlation	expression of MMP-14 at mRNA and protein level was statistically higher in invasive group and correlated with cavernous sinus invasion
Babula D et al., 2017 [48]	not examined	not examined	not examined	MMP-2, MMP-9 (+)	not examined	protein	not examined directly	not examined	correlation (higher activity of MMP-2 in	not examined	18 macroadenomas and 3 microadenomas, serum activities of

			(microadenoma vs macroadenoma)				(lower MMP-9 activity in microadenomas suggested their low invasiveness in contrast to macroadenomas with high MMP-9 activity and higher risk of invasion)		microadenomas, increased activity of MMP-9 in macroadenomas)		MMP-2 and MMP-9 were not statistically different between patients with micro- and macroadenomas (venous blood samples were collected before the surgery)
Liu X et al., 2018 [14]	ACTH: 55	not examined	not examined (recurrent vs nonrecurrent)	MMP-9 (+/-)	not examined	protein	not examined	correlation (significantly higher MMP-9 expression in the recurrent group compared with the nonrecurrent group	no correlation	not examined	recurrence status according to 33-59 months (mean 41.8) follow-up: 28 nonrecurrent ACTH-oma individuals and 27 recurrent ones, MMP-9 expression strongly correlated with the recurrence-free interval, MMP-9 expression was significantly higher in both recurrent and nonrecurrent groups than in normal pituitary gland tissue (n=2)

Guo H et al., 2019 [22]	data not available	data not available	58 vs 50	MMP-9	TIMP-1	mRNA, protein	correlation (MMP-9 expression at both mRNA and protein levels was significantly increased in invasive PAs as compared to non-invasive ones, TIMP-1 was highly expressed in non-invasive PAs, and the differences were statistically significant	data not available	data not available	data not available	MMP-9 and TIMP-1 levels in serum were assessed before and after operation, MMP-9 level in serum from invasive PAs group and TIMP-1 level in non-invasive PAs group was relatively high before surgical removal of the tumor, the relatively high, postoperative survival rate of patients was connected with low expression of MMP-9 and high expression of TIMP-1

References:

1. Lopes MBS. The 2017 World Health Organization classification of tumors of the pituitary gland: a summary. Acta Neuropathol. 2017;134(4):521-535. doi: 10.1007/s00401-017-1769-8.

2. Hussaini IM, Trotter C, Zhao Y, Abdel-Fattah R, Amos S, Xiao A, et al. Matrix metalloproteinase-9 is differentially expressed in nonfunctioning invasive and noninvasive pituitary adenomas and increases invasion in human pituitary adenoma cell line. Am J Pathol. 2007; 170(1):356-365.

3. Gong J, Zhao Y, Abdel-Fattah R, Amos S, Xiao A, Lopes MB, et al. Matrix metalloproteinase-9, a potential biological marker in invasive pituitary adenomas. Pituitary. 2008;11(1):37-48.

4. Altaş M, Bayrak OF, Ayan E, Bolukbasi F, Silav G, Coskun KK, et al. The effect of polymorphisms in the promoter region of the MMP-1 gene on the occurrence and invasiveness of hypophyseal adenoma. Acta Neurochir (Wien). 2010; 152(9):1611-1617; discussion 1617. doi: 10.1007/s00701-010-0671-0.

5. Wang J, Zhang Z, Li R, Mao F, Sun W, Chen J, et al. ADAM12 induces EMT and promotes cell migration, invasion and proliferation in pituitary adenomas via EGFR/ERK signaling pathway. Biomed Pharmacother. 2018; 97:1066-1077. doi: 10.1016/j.biopha.2017.11.034.

6. Mao JH, Guo H, Si N, Qiu L, Guo LF, Sun ZS, et al. Regulating effect of MMP-9 and TIMP-1 in pituitary adenoma invasion. Genet Mol Res. 2015;14(4):17091-17098. doi: 10.4238/2015.

7. Glebauskiene B, Liutkeviciene R, Vilkeviciute A, Kriauciuniene L, Jakstiene S, Zlatkute E, et al. Does MMP-9 Gene Polymorphism Play a Role in Pituitary Adenoma Development? Dis Markers. 2017; 2017:5839528. doi: 10.1155/2017/5839528.

8. Liu W, Matsumoto Y, Okada M, Miyake K, Kunishio K, Kawai N, et al. Matrix metalloproteinase 2 and 9 expression correlated with cavernous sinus invasion of pituitary adenomas. J Med Invest. 2005; 52(3-4):151-158.

9. Hui P, Xu X, Xu L, Hui G, Wu S, Lan Q. Expression of MMP14 in invasive pituitary adenomas: relationship to invasion and angiogenesis. Int J Clin Exp Pathol. 2015; 8(4):3556-3567. eCollection 2015.

10. Liu HY, Gu WJ, Wang CZ, Ji XJ, Mu YM. Matrix metalloproteinase-9 and -2 and tissue inhibitor of matrix metalloproteinase-2 in invasive pituitary adenomas: A systematic review and meta-analysis of case-control trials. Medicine (Baltimore). 2016; 95(24):e3904. doi: 10.1097/MD.0000000000003904.

11. Tortosa F, Webb SM. Novel aspects in histopathology of the pituitary gland. Endocrinol Diabetes Nutr. 2017;64(3):152-161. doi: 10.1016/j.endinu.2016.10.004.

12. Gültekin GD, Çabuk B, Vural Ç, Ceylan S. Matrix metalloproteinase-9 and tissue inhibitor of matrix metalloproteinase-2: Prognostic biological markers in invasive prolactinomas. J Clin Neurosci. 2015; 22(8):1282-1287. doi: 10.1016/j.jocn.2015.02.021.

13. Dineen R, Stewart PM, Sherlock M. Acromegaly. QJM. 2017;110(7):411-420. doi: 10.1093/qjmed/hcw004.

14. Liu X, Feng M, Zhang Y, Dai C, Sun B, Bao X, et al. Expression of Matrix Metalloproteinase-9, Pituitary Tumor Transforming Gene, High Mobility Group A 2, and Ki-67 in Adrenocorticotropic Hormone-Secreting Pituitary Tumors and Their Association with Tumor Recurrence. World Neurosurg. 2018; 113: e213-e221. doi: 10.1016/j.wneu.2018.01.214.

15. Ntali G, Wass JA. Epidemiology, clinical presentation and diagnosis of non-functioning pituitary adenomas. Pituitary. 2018;21(2):111-118. doi: 10.1007/s11102-018-0869-3.

16. Zlatkutė E, Liutkevičienė R, Vilkevičiūtė A, Glebauskienė B, Kriaučiūnienė L, Jakštienė S, et al. The role of *MMP-1* and *FGFR4-R388* gene polymorphisms in pituitary adenoma. Acta Med Litu. 2017; 24(4):177-190. doi: 10.6001/actamedica.v24i4.3613.

17. Páez Pereda M, Ledda MF, Goldberg V, Chervín A, Carrizo G, Molina H, et al. High levels of matrix metalloproteinases regulate proliferation and hormone secretion in pituitary cells. J Clin Endocrinol Metab. 2000; 85(1):263-269.

18. Turner HE, Harris AL, Melmed S, Wass JA. Angiogenesis in endocrine tumors. Endocr Rev. 2003;24(5):600-632.

19. Mete O, Ezzat S, Asa SL. Biomarkers of aggressive pituitary adenomas. J Mol Endocrinol. 2012; 49(2):R69-78. doi: 10.1530/JME-12-0113.

20. Tofrizal A, Fujiwara K, Azuma M, Kikuchi M, Jindatip D, Yashiro T, et al. Tissue inhibitors of metalloproteinase-expressing cells in human anterior pituitary and pituitary adenoma. Med Mol Morphol. 2017; 50(3):145-154. doi: 10.1007/s00795-017-0155-x.

21. Han X, Geng X, Li Z, Chen Z, Liu Y, Liu P, at al. The Relationship Between Phospho-p38, Matrix Metalloproteinase 9, and Major Histocompatibility Complex Class I Chain-Related Molecule A Expression in Pituitary Adenomas Demonstrates a New Mechanism of Pituitary Adenoma Immune Escape. World Neurosurg. 2019;123:e116-e124. doi: 10.1016/j.wneu.2018.11.077.

22. Guo H, Sun Z, Wei J, Xiang Y, Qiu L, Guo L, at al. Expressions of Matrix Metalloproteinases-9 and Tissue Inhibitor of Metalloproteinase-1 in Pituitary Adenomas and Their Relationships with Prognosis. Cancer Biother Radiopharm. 2019;34(1):1-6. doi: 10.1089/cbr.2018.2589.

23. Yadav L, Puri N, Rastogi V, Satpute P, Ahmad R, Kaur G. Matrix metalloproteinases and cancer - roles in threat and therapy. Asian Pac J Cancer Prev. 2014;15(3):1085-1091. doi: 10.7314/apjcp.2014.15.3.1085.

24. Galliera E, Tacchini L, Corsi Romanelli MM. Matrix metalloproteinases as biomarkers of disease: updates and new insights. Clin Chem Lab Med. 2015;53(3):349-355. doi: 10.1515/cclm-2014-0520.

25. Koutroulis I, Zarros A, Theocharis S. The role of matrix metalloproteinases in the pathophysiology and progression of human nervous system malignancies: a chance for the development of targeted therapeutic approaches? Expert Opin Ther Targets. 2008; 12(12):1577-1586. doi: 10.1517/14728220802560307.

26. Chen Z, Li Z, Chang Y, Ma L, Xu W, Li M, et al. Relationship between NF-κB, MMP-9, and MICA expression in pituitary adenomas reveals a new mechanism of pituitary adenomas immune escape. Neurosci Lett. 2015; 597:77-83. doi: 10.1016/j.neulet.2015.04.025.

27. Paez-Pereda M, Kuchenbauer F, Arzt E, Stalla GK. Regulation of pituitary hormones and cell proliferation by components of the extracellular matrix. Braz J Med Biol Res. 2005; 38(10):1487-1494.

28. Beaulieu E, Kachra Z, Mousseau N, Delbecchi L, Hardy J, Béliveau R. Matrix metalloproteinases and their inhibitors in human pituitary tumors. Neurosurgery. 1999; 45(6):1432-1440; discussion 1440-1441.

29. Kawamoto H, Uozumi T, Kawamoto K, Arita K, Yano T, Hirohata T. Type IV collagenase activity and cavernous sinus invasion in human pituitary adenomas. Acta Neurochir (Wien). 1996; 138(4):390-395.

30. Kawamoto H, Kawamoto K, Mizoue T, Uozumi T, Arita K, Kurisu K. Matrix metalloproteinase-9 secretion by human pituitary adenomas detected by cell immunoblot analysis. Acta Neurochir (Wien). 1996; 138(12):1442-1448.

31. Turner HE, Nagy Z, Esiri MM, Harris AL, Wass JA. Role of matrix metalloproteinase 9 in pituitary tumor behavior. J Clin Endocrinol Metab. 2000; 85(8):2931-2935.

32. Knappe UJ, Hagel C, Lisboa BW, Wilczak W, Lüdecke DK, Saeger W. Expression of serine proteases and metalloproteinases in human pituitary adenomas and anterior pituitary lobe tissue. Acta Neuropathol. 2003; 106(5):471-478.

33. Liu W, Kunishio K, Matsumoto Y, Okada M, Nagao S. Matrix metalloproteinase-2 expression correlates with cavernous sinus invasion in pituitary adenomas. J Clin Neurosci. 2005;12(7):791-794.

34. Qiu L, He D, Fan X, Li Z, Liao C, Zhu Y, et al. The expression of interleukin (IL)-17 and IL-17 receptor and MMP-9 in human pituitary adenomas. Pituitary. 2011; 14(3):266-275. doi: 10.1007/s11102-011-0292-5.

35. Yokoyama S, Hirano H, Moroki K, Goto M, Imamura S, Kuratsu JI. Are nonfunctioning pituitary adenomas extending into the cavernous sinus aggressive and/or invasive? Neurosurgery. 2001; 49(4):857-862; discussion 862-863.

36. Wang H, Li WS, Shi DJ, Ye ZP, Tai F, He HY, et al. Correlation of MMP(1) and TIMP (1) expression with pituitary adenoma fibrosis. J Neurooncol. 2008; 90(2):151-156. doi: 10.1007/s11060-008-9647-9.

37. Xiao Z, Liu Q, Mao F, Wu J, Lei T. TNF-α-induced VEGF and MMP-9 expression promotes hemorrhagic transformation in pituitary adenomas. Int J Mol Sci. 2011; 12(6):4165-4179. doi: 10.3390/ijms12064165.

38. Pan LX, Chen ZP, Liu YS, Zhao JH. Magnetic resonance imaging and biological markers in pituitary adenomas with invasion of the cavernous sinus space. J Neurooncol. 2005; 74(1):71-76.

39. Yamada S, Ohyama K, Taguchi M, Takeshita A, Morita K, Takano K, et al. A study of the correlation between morphological findings and biological activities in clinically nonfunctioning pituitary adenomas. Neurosurgery. 2007; 61(3):580-584; discussion 584-585.

40. Tofrizal A, Fujiwara K, Yashiro T, Yamada S. Alterations of collagen-producing cells in human pituitary adenomas. Med Mol Morphol. 2016; 49(4):224-232.

41. Friedman TC, Orlowski M, Wilk S. Peptide-degrading enzymatic activities in GH3 cells and rat anterior pituitary homogenates. Endocrinology. 1984; 114(4):1407-1412.

42. Zhao C, Zhang M, Liu W, Wang C, Zhang Q, Li W. β-Catenin knockdown inhibits pituitary adenoma cell proliferation and invasion via interfering with AKT and gelatinases expression. Int J Oncol. 2015; 46(4):1643-1650. doi: 10.3892/ijo.2015.2862.

43. Pan Y, Han C, Wang C, Hu G, Luo C, Gan X, Zhang F, Lu Y, Ding X. ADAM10 promotes pituitary adenoma cell migration by regulating cleavage of CD44 and L1. J Mol Endocrinol. 2012; 49(1):21-33. doi: 10.1530/JME-11-0174.

44. Wang J, Voellger B, Benzel J, Schlomann U, Nimsky C, Bartsch JW, Carl B. Metalloproteinases ADAM12 and MMP-14 are associated with cavernous sinus invasion in pituitary adenomas. Int J Cancer. 2016; 139(6):1327-1339. doi: 10.1002/ijc.30173.

45. Ceylan S, Anik I, Koc K, Kokturk S, Ceylan S, Cine N, et al. Microsurgical anatomy of membranous layers of the pituitary gland and the expression of extracellular matrix collagenous

proteins. Acta Neurochir (Wien). 2011;153(12):2435-2443; discussion 2443. doi: 10.1007/s00701-011-1182-3.

46. Tomita T. Matrix Metalloproteinases and Tissue Inhibitors of Metalloproteinases in Pituitary Adenomas: Possible Markers of Neuroendocrine Cells. Endocr Pathol. 1997;8(4):305-313.

47. Qu X, Yang W, Jiang M, Han T, Han L, Qu Y, et al. CD147 expression in pituitary adenomas and its significance for clinical outcome. Hum Pathol. 2010;41(8):1165-1171. doi: 10.1016/j.humpath.2009.10.023.

48. Babula D, Kocot J, Horecka A, Baran M, Kurzepa J. Different patterns of gelatinolytic activity in pituitary macro- and microadenomas. Clin Neurol Neurosurg. 2017; 158:90-92. doi: 10.1016/j.clineuro.2017.05.005.

49. Ruskyte K, Liutkevicienė R, Vilkeviciute A, Vaitkiene P, Valiulytė I, Glebauskiene B, Kriauciuniene L, Zaliuniene D. MMP-14 and TGFβ-1 methylation in pituitary adenomas. Oncol Lett. 2016;12(4):3013-3017.

50. Sun B, Liu X, Yang Y, Dai C, Li Y, Jiao Y, Wei Z, Yao Y, Feng M, Bao X, Deng K, Wang N, Wang R. The Clinical Utility of TIMP3 Expression in ACTH-Secreting Pituitary Tumor. J Mol Neurosci. 2016; 58(1):137-44. doi: 10.1007/s12031-015-0698-z.