

A Case-Control Study of Comparison of Plasma Levels of Matrix Metalloproteinase-9 in Inhaled Opium Addicts and Clinically Healthy Persons

Maryam Amizadeh MD¹, Shahryar Dabiri MD², Armin Javedan MD³,
Ali Eftekharian MD⁴, Aliasghar Arabi-Mianroodi MD¹

Original Article

Abstract

Background: Metalloproteinase-9 exists in the basement membrane of normal tissues and is a major factor in cancer invasion and lymphatic metastasis. Smoking has been reported to increase the metalloproteinase level, but the role of opium consumption in metalloproteinase level has not yet been examined. The current research intended to examine the impacts of opium consumption on the serum levels of metalloproteinase.

Methods: This case-control research was conducted in Kerman (in the southeast of Iran), after getting medical approve by the ethics committee. Case group of 33 non-smokers with no active inflammatory diseases who had the experience of inhaled opium and its derivatives were compared with a control group of 40 non-smokers with no active inflammatory disease and no experience of inhaled opium and its derivatives. Student's t-test, mean, and chi-square test were employed to determine the correlation between the variables.

Findings: No statistically meaningful variation was detected in plasma metalloproteinase concentration between the case and control groups ($P = 0.160$). Also, there was no significant relation between the plasma metalloproteinase concentration and urinary morphine in case groups ($P = 0.410$), but a statistically significant correlation was found between gender and metalloproteinase in both the case and control groups ($P = 0.003$).

Conclusion: According to the possible role of opioid drugs in cancer and its prognosis both directly and through their impact on serum levels of metalloproteinase, further studies are suggested.

Keywords: Metalloproteinase; Opium; Case-control studies; Iran

Citation: Amizadeh M, Dabiri S, Javedan A, Eftekharian A, Arabi-Mianroodi A. A Case-Control Study of Comparison of Plasma Levels of Matrix Metalloproteinase-9 in Inhaled Opium Addicts and Clinically Healthy Persons. *Addict Health* 2017; 9(1): 11-6.

Received: 28.08.2016

Accepted: 03.11.2016

1- Assistant Professor, Clinical Research Unit, Shafa Hospital, School of Medicine, Kerman University of Medical Sciences, Kerman, Iran

2- Professor, Department of Pathology AND Stem Cell Research Center, School of Medicine, Kerman University of Medical Sciences, Kerman, Iran

3- Student of Medicine, Clinical Research Unit, Shafa Hospital, School of Medicine, Kerman University of Medical Sciences, Kerman, Iran

4- Professor, Department of Otolaryngology, School of Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran

Correspondence to: Maryam Amizadeh MD, Email: ma74ir@yahoo.com

Introduction

Extracellular matrix metalloproteinases (MMPs) are metal-dependent end peptidases involved in proteolysis' activities for which they need zinc ions. MMPs are a major class of enzymes related to the degradation of extracellular matrix elements. The MMP family is classified into six subgroups, including stromelysins, matrilysin, gelatinases, collagenases, membrane and other MMPs.^{1,2} Twenty-four types of MMPs have been so far identified, 23 of which are found in humans.

MMPs are involved in different physiological and pathophysiological processes, such as growth and differentiation of the embryo and fetus, ovulation, wound healing, heart failure, arthritis, atherosclerosis, diseases of the teeth, bone remodeling, cancers, etc. Evidence suggests that MMPs are involved in all stages of cancer progression, including proliferation, apoptosis, and angiogenesis.³ However, some studies have reported a relationship between metalloproteinase and diagnosis and poor prognosis of some cancers, such as lung, breast, gastric cancers, and melanoma. In some other studies, MMPs have been recognized to be effective in developing new treatments for cancers.⁴

So far, some researches were carried out to evaluate the impacts of tobacco products on cancer.

Methods

This case-control research was conducted in Kerman (in the southeast of Iran), after obtaining medical approval by the ethics committee. A case group including 33 non-smokers with no active inflammatory diseases who had the experience of inhaled opium and its derivatives were compared

with a control group of 40 non-smokers with no active inflammatory disease and no experience of inhaled opium and its derivatives.

The standard deviation (SD), mean, and frequency were employed in both groups to express the qualitative and quantitative parameters. The chi-square test and Student's t-test were employed to evaluate the qualitative parameters among the groups and to compare the quantitative variables, respectively. The data were analyzed using Stata Statistical Software (Release 11.0, Stata Corporation, College Station, TX, USA). The P-value less than 0.05 was considered statistically significant.

Results

In the current research, there were 25 males (56.8%) and eight females (27.6%) in the case group and 19 men (43.1%) and 21 women (72.4%) in the control group. The average age was 47.2 and 44.6 years in the case and control groups respectively. Fourteen people (42.4%) in the case group and 16 people (40.0%) in the control group were ≤ 40 years of age. In the 41-60 age groups, there were 14 and 19 people (42.4% and 47.5%) in the case and control groups, respectively. In the age group over 60 years, five people (15.2%) were in the case group and 27 (67.5%) in the control group. The average serum levels of MMP-9 were 905 in the case group and 1481 in the control group. Six subjects of the case group (18.2%) and 13 of the control group (32.5%) were positive for serum levels of MMP-9. Twenty-seven subjects of the control group (81.8%) and 27 of the case group (67.5%) were negative for serum levels of MMP-9.

Statistical analysis indicated no difference in serum levels of MMP-9 among the case and the control groups ($P = 0.140$) (Table 1).

Table 1. Demographic data of the case and control groups

Variables	Control group	Case group	P*
Gender [n (%)]			0.010
Male	19 (43.2)	25 (56.8)	
Female	21 (72.4)	8 (27.6)	
Age (year) (mean $\pm$ SD)	44.60 $\pm$ 1.93	47.20 $\pm$ 2.47	0.890
< 40 [n (%)]	16 (40.0)	14 (42.4)	
41-60 [n (%)]	19 (47.5)	14 (42.4)	
> 60 [n (%)]	5 (12.5)	5 (15.2)	
MMP-9 (ng/ml) (mean $\pm$ SD)	1482 $\pm$ 396	905 $\pm$ 237	0.160
Positive [n (%)]	13 (32.5)	6 (18.2)	
Negative [n (%)]	27 (67.5)	27 (81.8)	

*Level of significance < 0.05

MMP-9: Matrix metalloproteinase-9; SD: Standard deviation

Although a positive correlation was found between serum level of morphine and MMP-9 (Pearson $\chi^2 = 0.670$), this association was not statistically significant ($P = 0.410$). There was a positive correlation between the gender and level of MMP-9 ($P = 0.040$), and the positive probability of the serum levels of MMP-9 was six times higher in female (Pearson $\chi^2 = 0.603$) (Table 2).

Table 2. The relationship of metalloproteinase with urinary morphine and gender

Variables	Pearson χ^2	P*
Morphine	0.670	0.410
Gender	0.603	0.003

*Level of significance < 0.05

Discussion

The current research was conducted to evaluate the impacts of opium consumption on the serum levels of MMP-9. The study of risk factors, early diagnosis, and cancer treatment methods have led researchers worldwide to apply appropriate strategies for reducing mortality caused by this disease and enhancing patient's quality of life by reducing risk factors, early diagnosis, and implementing an appropriate treatment method.

Lymphatic metastasis is a factor involved in the determination of the type of treatment and prognosis of cancer patients that reduces the patient survival by 50%.⁵ The MMP-9 that exists in the basement membrane in normal tissues⁶ is a major factor in cancer invasion and lymph node metastasis. Changes in the extracellular matrix and destruction of intermolecular barriers and increased secretion of the enzyme which leads to destruction of cell barriers can increase cancer metastases.

In this study, no significant variation was observed in the serum levels of MMP-9 among the opium users and the control group. This finding, at least in this study, suggests that the use of opioid drugs has no impact on the serum levels of MMP-9. As far as we know, no similar study has been performed to evaluate the impact of opium on the MMP-9. Torii et al.⁷ studied the relationship between MMP-9 and gastric cancer and reported that MMP-9 in human plasma has 96 percent specificity and 56 percent sensitivity in gastric cancer evaluation. So far, some studies have been conducted on the relationship between opioid drugs and its derivatives and the incidence of cancers, such as esophageal, bladder, laryngeal,

and recently gastric cancers, but in most of them, the effects of age, gender, and other main confounding variables have not been well controlled. This study had a small sample size, and it may be the reason why the correlation between MMP and the use of opioid drugs was not statistically significant.

Smoking is one of the main factors that can increase MMP-9.⁸ In a research performed by Lim et al.,⁹ the balance among MMP-9 and tissue inhibitor of metalloproteinase-1 was examined in smokers, and it was revealed that their levels were higher in smokers that may lead to the destruction of lung tissue and eventually, emphysema in these people.

In a research carried out by Xu et al., the impacts of nicotine on differentiation, oxidation, bacterial killing, and secretion of MMP-9 by neutrophils was investigated. The results showed that nicotine increased the number of cells in the last phases of differentiation (metamyelocytes and segmented neutrophils) and in the neutrophils that were exposed to nicotine, oxidation and bacterial killing was reduced and the secretion of MMP-9 was increased.¹⁰

No studies have carried out so far to evaluate the impact of opium as an independent factor on the markers associated with cancer. However, according to a study conducted on 50000 Iranians in Golestan Province, Iran,¹¹ to examine the effect of opium on mortality, it was shown that the opium addicts had a higher risk of mortality compared with the control group (hazard ratio: 1.81).

Masjedi et al.¹² also examined the effect of opium as an independent parameter for lung cancer and found that opium increases the danger of lung cancer to an odds ratio of 3.1. Naghibzadeh Tahami et al.¹³ conducted research on the upper gastrointestinal cancers (stomach, esophagus, liver, and pancreas) in Kerman. They indicated that the use of opium and its derivatives increased the risk of these cancers by four times, and the increased use of opioid drugs had a direct correlation with the risk of this cancers.¹³

All studies have shown that the growing use of opioid drugs increase the risk of cancers, and the repeatability of the results confirms causality. Therefore, according to studies, it seems that opium consumption can increase mortality rate and the risk of cancers, especially lung cancer. In the present research, a positive correlation was

detected among the consumption of opioid drugs and the increased level of MMP-9, as a predictive factor for diagnosing cancers and the lymphatic metastases. However, probably due to the small sample size, this correlation was not statistically significant. Studies on the effect of opium on the markers associated with cancer are not available, but some studies have been conducted on markers that are related to cancer. For example, a survey carried out on 101 patients to examine the effect of focal adhesion kinase (FAK) and E-cadherins in cervical metastases in supraglottic cancer, and it was found that the decreased E-cadherin and increased FAK were associated with a higher risk of metastasis.¹⁴

Schneider et al.¹⁵ conducted a study to evaluate the impact of FAK on the invasion of tumor cells of the oral cavity, and they concluded that FAK expression was higher in squamous cell carcinoma (SCC25) cells which are more invasive than SCC15 cells.

Field et al.¹⁶ conducted a study to examine the effect of p53 on cases with head and neck SCC who had the history of smoking. They found that p53 was increased in 67% of 73 patients studied. Also, p53 was more positive in those who were heavy smokers.

Christopoulos et al.¹⁷ found that in laryngeal SCC, as the stage of cancer advances, the CD44 hyaluronin receptor is expressed more than the hyaluronidase.

Hashimoto et al.¹⁸ investigated the link

between MMP-1 and MMP-3 and the head and neck SCC. There was a significant positive relation between the serum levels of metalloproteinase and prognosis of cancer and all have indicated that the serum level of metalloproteinase is a good predictive factor for the course of the cancer. As mentioned, some studies have recently reported the relationship between the use of opium and its derivatives and the diffusion of cancer.

Conclusion

In our study, no significant difference was observed in plasma concentrations of MMP-9 between addicts and healthy controls. Also, no correlation was found between the urinary morphine measured and MMP-9. Further studies are required in this regard with a larger sample size and stronger methodology such as prospective studies, and also other markers are recommended to be examined, including FAK, E-cadherin, CD44, hyaluronidase, Test lamp-Multipurpose (TL-MP), membrane type-1 MMP, MMP-1, etc.

Conflict of Interests

The Authors have no conflict of interest.

Acknowledgements

We would like to thank the Neurology Research Center of Kerman University of Medical Sciences, which participated in this research.

References

1. Scurry WC Jr, Stack BC Jr. Role of metalloproteins in the clinical management of head and neck squamous cell carcinoma. *Head Neck* 2007; 29(12): 1144-55.
2. Rautelin HI, Oksanen AM, Veijola LI, Sipponen PI, Tervahartiala TI, Sorsa TA, et al. Enhanced systemic matrix metalloproteinase response in *Helicobacter pylori* gastritis. *Ann Med* 2009; 41(3): 208-15.
3. Nagase H, Woessner JF Jr. Matrix metalloproteinases. *J Biol Chem* 1999; 274(31): 21491-4.
4. Fink K, Boratynski J. The role of metalloproteinases in modification of extracellular matrix in invasive tumor growth, metastasis and angiogenesis. *Postepy Hig Med Dosw (Online)* 2012; 66: 609-28.
5. Stamenkovic I. Extracellular matrix remodelling: The role of matrix metalloproteinases. *J Pathol* 2003; 200(4): 448-64.
6. Egeblad M, Werb Z. New functions for the matrix metalloproteinases in cancer progression. *Nat Rev Cancer* 2002; 2(3): 161-74.
7. Torii A, Kodera Y, Uesaka K, Hirai T, Yasui K, Morimoto T, et al. Matrix metalloproteinase 9 in human plasma has 96% specificity and 56% sensitivity for gastric cancer screening. *Int J Clin Oncol* 1996; 1(1): 39-43.
8. Nakamura T, Ebihara I, Shimada N, Koide H. Effect of cigarette smoking on plasma metalloproteinase-9 concentration. *Clin Chim Acta* 1998; 276(2): 173-7.
9. Lim S, Roche N, Oliver BG, Mattos W, Barnes PJ, Chung KF. Balance of matrix metalloproteinase-9 and tissue inhibitor of metalloproteinase-1 from alveolar macrophages in cigarette smokers. Regulation by interleukin-10. *Am J Respir Crit Care Med* 2000; 162(4 Pt 1): 1355-60.
10. Xu M, Scott JE, Liu KZ, Bishop HR, Renaud DE,

- Palmer RM, et al. The influence of nicotine on granulocytic differentiation-inhibition of the oxidative burst and bacterial killing and increased matrix metalloproteinase-9 release. *BMC Cell Biol* 2008; 9: 19.
11. Khademi H, Malekzadeh R, Pourshams A, Jafari E, Salahi R, Semnani S, et al. Opium use and mortality in Golestan Cohort Study: Prospective cohort study of 50,000 adults in Iran. *BMJ* 2012; 344: e2502.
12. Masjedi MR, Naghan PA, Taslimi S, Yousefifard M, Ebrahimi SM, Khosravi A, et al. Opium could be considered an independent risk factor for lung cancer: A case-control study. *Respiration* 2013; 85(2): 112-8.
13. Naghibzadeh Tahami A, Khanjani N, Yazdi Feyzabadi V, Varzandeh M, Haghdoost AA. Opium as a risk factor for upper gastrointestinal cancers: A population-based case-control study in Iran. *Arch Iran Med* 2014; 17(1): 2-6.
14. Rodrigo JP, Dominguez F, Suarez V, Canel M, Secades P, Chiara MD. Focal adhesion kinase and E-cadherin as markers for nodal metastasis in laryngeal cancer. *Arch Otolaryngol Head Neck Surg* 2007; 133(2): 145-50.
15. Schneider GB, Kurago Z, Zaharias R, Gruman LM, Schaller MD, Hendrix MJ. Elevated focal adhesion kinase expression facilitates oral tumor cell invasion. *Cancer* 2002; 95(12): 2508-15.
16. Field JK, Spandidos DA, Malliri A, Gosney JR, Yiagnosis M, Stell PM. Elevated P53 expression correlates with a history of heavy smoking in squamous cell carcinoma of the head and neck. *Br J Cancer* 1991; 64(3): 573-7.
17. Christopoulos TA, Papageorgakopoulou N, Theocharis DA, Mastronikolis NS, Papadas TA, Vynios DH. Hyaluronidase and CD44 hyaluronan receptor expression in squamous cell laryngeal carcinoma. *Biochim Biophys Acta* 2006; 1760(7): 1039-45.
18. Hashimoto T, Uchida K, Okayama N, Imae Y, Suehiro Y, Hamanaka Y, et al. Association of matrix metalloproteinase (MMP)-1 promoter polymorphism with head and neck squamous cell carcinoma. *Cancer Lett* 2004; 211(1): 19-24.

مطالعه مورد - شاهی سطح پلاسمایی ماتریکس متالوپروتئیناز-۹ (MMP-9) در افراد معتاد به تریاک استنشاقی و افراد سالم

دکتر مریم عامی زاده^۱، دکتر شهریار دبیری^۲، دکتر آرمین جاودان^۳، دکتر علی افتخاریان^۴،
دکتر علی اصغر عربی میاندوابی^۱

مقاله پژوهشی

چکیده

مقدمه: متالوپروتئیناز-۹ در غشای پایه بافت‌های طبیعی وجود دارد و عامل اصلی در تهاجم سرطان و متاستاز لنفاوی به شمار می‌رود. تاکنون نقش مصرف سیگار در افزایش میزان متالوپروتئیناز مشخص شده، اما نقش مصرف اپیوم بر روی میزان متالوپروتئیناز بررسی نشده است. مطالعه حاضر جهت بررسی تأثیر مصرف اپیوم بر روی میزان متالوپروتئیناز سرمی صورت گرفت.

روش‌ها: در این مطالعه مورد-شاهدی که در شهر کرمان (واقع در جنوب شرقی ایران) انجام گردید، گروه مورد شامل ۳۳ نفر فرد غیر سیگاری و بدون بیماری فعال التهابی و دارای سابقه مصرف استنشاقی اپیوم و مشتقات آن، با ۴۰ نفر گروه شاهد غیر سیگاری و بدون بیماری فعال التهابی که سابقه مصرف استنشاقی اپیوم و مشتقات آن را نداشتند، مقایسه شدند. با استفاده از آزمون‌های t و χ^2 ، میانگین و نسبت بین متغیرها مورد بررسی قرار گرفت.

یافته‌ها: بین نسبت غلظت پلاسمایی متالوپروتئیناز در دو گروه مورد و شاهد هیچ‌گونه تفاوت معنی‌داری وجود نداشت ($P = 0/160$). همچنین، ارتباط معنی‌داری بین نسبت غلظت پلاسمایی متالوپروتئیناز و مورفین ادرار در دو گروه مورد و شاهد مشاهده نشد ($P = 0/410$). از طرف دیگر، ارتباط بین جنسیت و متالوپروتئیناز در دو گروه مورد و شاهد معنی‌دار بود ($P = 0/003$).

نتیجه‌گیری: با توجه به نقش احتمالی مواد اپیوئیدی بر روی سرطان و ارتباط مستقیم متالوپروتئیناز با پیش‌آگهی و پیشرفت سرطان‌ها، انجام مطالعات بیشتر پیشنهاد می‌شود.

واژگان کلیدی: متالوپروتئیناز، اپیوم، مطالعات مورد-شاهدی، ایران

ارجاع: عامی‌زاده مریم، دبیری شهریار، جاودان آرمین، افتخاریان علی، عربی علی اصغر. مطالعه مورد-شاهدی سطح پلاسمایی ماتریکس متالوپروتئیناز-۹ (MMP-9) در افراد معتاد به تریاک استنشاقی و افراد سالم. مجله اعتیاد و سلامت ۱۳۹۵؛ ۹ (۱): ۱۱-۱۶.

تاریخ پذیرش: ۹۵/۸/۱۳

تاریخ دریافت: ۹۵/۶/۷