

The Role of Collagen I, Magnesium and Phosphorus in Iraqi Meningioma Patients

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Abstract

Meningiomas are the most common primary intracellular slow growing benign tumor that is mostly arisen from meningeothelial (arachnoid) cells which surround the brain and the spinal cord. The aim of the current study is to innovate a biochemical relationship between collagen I and meningiomas in Iraqi patients and also to examine the biochemical role of magnesium and phosphorus in this complex nervous disease. Thirty (30) diagnosed meningioma patients were participated in the current study and were categorized into two (2) groups : G₃ composed of (15) newly diagnosed meningioma males and G₄ composed of (15) newly diagnosed meningioma females , both G₃ and G₄ subjects did not take any treatment linked with meningioma or any chronic disease. Patients groups were compared with healthy subjects as two control groups that did not suffer from any chronic disease , the first G₁ composed of (15) healthy males and the second G₂ composed of (15) healthy females in the range of age matched with patients groups. Blood samples were collected from each subject participated in this study and three biochemical parameters (collagen I, magnesium, phosphorus) were determined in sera of all groups. The results of the current study have confirmed that levels of collagen I , magnesium and phosphorus were highly significantly decreased in blood sera of G₃ and G₄ compared to G₁ and G₂ respectively, while the difference between G₃ and G₄ was non significant for the three biochemical parameters. The present study has contributed to collagen I novelty regarding Iraqi patients with meningiomas deficiency and also highlighted the biochemical role of both magnesium and phosphorus in accordance with meningiomas. The present study recommends checking periodically the levels of collagen I, magnesium and phosphorus by laboratory tests and treatment with collagen supplements Tablets and metabolic (magnesium phosphate) capsules if a deficiency has been notice

Keywords : Meningiomas . Collagen I . Magnesium . Phosphorus.

دور الكولاجين I و المغنيسيوم و الفوسفور لمرضى عراقيين مصابين بالسحايا

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الخلاصة

يمثل السحايا أو الورم السحائي أحد الأورام الحميدة السائدة الأكثر شيوعاً و الذي ينشأ من الخلايا السحائية المحيطة بالدماغ و الحبل الشوكي . الهدف من الدراسة الحالية هو إبتكار علاقة بايو كيميائية مابين الكولاجين I و مرضى عراقيين مصابين بالسحايا و كذلك لإختبار الدور الكيموحيوي للمغنيسيوم و الفوسفور بما يخص هذا المرض العصبي المعقد. تم اشتراك ثلاثون (30) مريضاً في الدراسة الحالية و تم تقسيمهم إلى مجموعتين : G₃ تضمنت (15) ذكراً و G₄ تضمنت (15) أنثى مصابين و مصابات بالسحايا تم تشخيصهم في بداية الدراسة و لم يتعاطوا أي علاج مرتبط بالسحايا أو غيره من الأمراض المزمنة . تم مقارنة مجموعتي المرضى مع مجموعتين ضابطة (أصحاء) بدون أمراض مزمنة ، الأولى G₁ تتألف من (15) ذكر و الثانية G₂ تتألف من (15) أنثى ضمن نفس الفئة العمرية للمصابين . تم جمع نماذج الدم لكل شخص إنضم لهذه الدراسة و تم قياس ثلاث متغيرات كيموحيوية و هي الكولاجين I و المغنيسيوم و الفوسفور في مصل دم جميع المجاميع . أثبتت نتائج الدراسة الحالية أن مستويات الكولاجين I و المغنيسيوم و الفوسفور إنخفضت بشكل معنوي عالي في مصل دم G₃ و G₄ مقارنة مع G₁ و G₂ على التوالي بينما كان الإختلاف غير معنوياً مابين G₃ و G₄ للمتغيرات الكيموحيوية الثلاث. إن الدراسة الحالية قدمت مفاهيم جديدة بما يخص نقص الكولاجين لمرضى السحايا العراقيين و أيضاً سلطت الضوء على الدور الكيموحيوي لكل من المغنيسيوم و الفوسفور بما يخص مرض السحايا. الدراسة الحالية توصي بإجراء فحص دوري لمستويات الكولاجين I و المغنيسيوم و الفوسفور عن طريق القياسات المختبرية و العلاج ببدائل الكولاجين (على شكل أقراص دوائية) و كبسول فوسفات المغنيسيوم في حالة وجود نقص.

الكلمات المفتاحية : السحايا ، الكولاجين I ، المغنيسيوم ، الفوسفور

Introduction

Meningiomas are the most prevalent major benign tumor related to the central nervous system (intracranial tumor) which approximately constitutes a third of central nervous system tumors⁽¹⁾. The basic pathognomonic histologic symptom of meningioma is spherical formations of meningeothelial cells (whorls) that finally

mineralized into small lesions called (psammoma bodies) ,⁽²⁾ or arachnoid cells which are defined as a cellular component of pia mater , arachnoid mater , trabeculae and septae of the subarachnoid space.¹ Meningioma patients are classified into three grades or classes : class I (typical) 81.12% , class II (atypical) 16.9% and class III (very rare) 1.7%⁽²⁾ .

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Regarding epidemiology, meningioma incidence increases with age. Also exposure to ionizing radiation is considered as the only environmental risk factor. Moreover, meningioma etiology affected by pesticide / herbicide, allergies and diet. Meningiomas is diagnosed by magnetic resonance imaging (MRI) or constant enhanced computed tomography (CT) in patients with contraindications to MRI such as pacemaker⁽¹⁾. Collagen is the unique fibrous protein making up the major part of the extracellular matrix⁽³⁾. Type I collagen (collagen I) is the most popular and studied⁽⁴⁾. It makes up the primary component of many vital tissues, it has a great role in maintenance the structure, the integrity and the functions of the body⁽³⁾. Magnesium is the fourth prevalent mineral ion in the human body, it is included in more than 300 kind of enzymatic reactions, regulation of many transporters and ion channels and also plasma membrane integrity. High magnesium diet increase magnesium level in serum or plasma and nerve tissues which support neuro-behavioral and electrophysical functions, inhibit the deposition of the major inflammatory cells (like macrophages) and the expression of inflammatory cytokines⁽⁵⁾. Phosphorus is regarded as a one of the pivotal elements of the human body which required for a wide range of processes such as ATP biosynthesis, signal transduction and bone mineralization. The most abundance (85%) of phosphorus in human body found as a biochemical component called hydroxyapatite [$\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$] in the extracellular matrix of bones and teeth. Conversely, the intracellular phosphorus constitutes 14% of the total body phosphorus and only 1% found as inorganic phosphate (Pi)⁽⁶⁾. The aim of the current study is to innovate a biochemical relation between collagen I and meningioma in Iraqi patients and also to examine the biochemical role of magnesium and phosphorus in the Pathophysiology of meningioma for the same patients.

Materials and Methods

Patients Selection

Thirty (30) meningioma patients participated in the present study who attended Saad Al – Witry Hospital for nervous diseases in Iraq / Baghdad from July 1st 2021 to September 1st 2021, they were diagnosed by both CT scan and MRI as meningioma cases. Anyway, the study protocol involved four groups: Two control groups (healthy subjects) G₁ and G₂ who did not suffer from any chronic disease and two meningioma patient groups G₃ and G₄. The first G₁ was composed of (15) males (24-55) years and the second G₂ was composed of (15) females (45-70) years. Patients were classified into two groups: G₃ was composed of (15) newly diagnosed males (20-56) years with

meningioma without any treatment related to meningioma or another chronic disease and G₄ was composed of (15) newly diagnosed meningioma females (45-70) years also without any treatment just like G₃. Remarkably, all females who enrolled in this study (patients and control) groups were menopausal.

Blood Sampling

Five (5) milliliters of venous blood were withdrawn from each patient and healthy subjects enrolled in this study, sera were isolated from blood cells by centrifugation (3-5) min at 4000 r.p.m, then blood sera were divided into three small divisions and kept frozen (-20°C) until biochemical analysis which were performed in the International Center for research and Development in Baghdad / Iraq.

Biochemical Analysis

According to collagen I, the quantitative sandwich enzyme - linked immunosorbent assay ELISA technique was used, using a kit (human Collagen Type I) supplied from Cusabio – USA. A specific antibody for collagen I was precoated on a microplate, standard and the samples were pipetted into the wells. Avidin conjugated horse radish peroxidase was used as an antibody. The optical density (OD) was recorded at 450 nm and the concentration of collagen I was calculated according to it. On the other hand, a reaction occurred between (magnesium and chromogen) and (phosphorus and chromogen) within a hand reaction (an ion bases chromogenic method or chromogenic detection) resulting in a complex intensity proportional to the concentration of magnesium and phosphorus within a sample exposed to test, the absorbance was recorded at 578nm.

Statistical Analysis

The results of the present study were explained as mean \pm SE (standard error). Student's T. test was used to compare the difference between the studied groups, values $p \leq 0.05$ were statistically considered as statistically significant and when p value ≤ 0.001 , the difference is considered as highly significant while when p value > 0.05 , the difference is considered as non-significant.

Results and Discussion

Table 1. Collagen I levels (ng/mL) in blood sera of patients and control groups

Group	Mean±SE	Group	Mean±SE
G ₁	19.062±3.212	G ₂	8.032±0.567
G ₃	3.502±0.426	G ₄	3.266±0.460
p G ₃ /G ₁ : H.S* (0.00028) p G ₄ /G ₂ : H.S* (1.32 × 10 ⁻¹⁰) p G ₄ /G ₃ : N.S (0.448)			

Table 2. Magnesium levels (mg/dL) in blood sera of patients and control groups

Group	Mean±SE	Group	Mean±SE
G ₁	1.973±0.214	G ₂	1.8±0.279
G ₃	1.314±0.456	G ₄	1.128±0.395
p G ₃ /G ₁ : H.S* (0.00032) p G ₄ /G ₂ : H.S* (0.000112) p G ₄ /G ₃ : N.S (0.3082)			

Table 3. Phosphorus levels (mg/dL) in blood sera of patients and control groups

Group	Mean±SE	Group	Mean±SE
G ₁	3.413±0.492	G ₂	3.021±0.560
G ₃	0.443±0.146	G ₄	0.465±0.103
p G ₃ /G ₁ : H.S* (1.099 E ⁻¹²) p G ₄ /G ₂ : H.S* (1.534 E ⁻¹⁰) p G ₄ /G ₃ : N.S (0.495)			

G₁ : healthy control group (male subjects) , G₂ : healthy control group (female subjects)

G₃ : Meningioma patients (male subjects) , G₄ : Meningioma patients (female subjects)

SE : standard error , P value : statistical probability

The results of the current study have reported that collagen I level was highly significantl decreased in sera of G₃ (3.502±0.426) ng/mL and G₄ (3.266±0.460) ng/mL compared with G₁(19.062±3.212) ng/mL and G₂(8.032±0.567) ng/mL respectively , the difference between G₃ (3.502±0.426) ng/mL and G₄ (3.266±0.460) was non significant (Table (1)). Regarding magnesium , its level was highly significant decreased in blood sera of G₃ (1.314±0.456) mg/dL and G₄ (1.128±0.395) mg/dL compared with G₁ (1.973±0.214) mg/dL and G₂ (1.8±0.279) mg/dL respectively, while the difference between G₃ (1.314±0.456) mg/dL and G₄ (1.128±0.395) mg/dL was non-significant (Table (2)). Moreover, Phosphorus level was highly significantly decreased in G₃ (0.443±0.146) mg/dL and G₄ (0.465±0.103) mg/dL compared with G₁(3.413±0.492) mg/dL and G₂ (3.021±0.560) mg/dL respectively, the difference between G₃ (0.443±0.146) mg/dL and G₄ (0.465±0.103) mg/dL was non-significant (Table (3)).

Collagen has a crucial effect on the immune system because it is detected in peripheral blood after ingestion. Also , collagen derived peptides are involved in inflammatory diseases. Collagen derived oligopeptides including polyhydroxy or proxy hydroxyl proline have been noticed to do chemotactic activities on fibroblasts , neutrophils and monocytes , all of these play vital roles in inflammatory responses and wound healing. ⁷Anyway , a number of studies have

concerned collagen I in meningioma tissues with results poorly understood but no one link collagen I in blood sera of meningioma patients ^(8,9) .Chondrocytes (cells responsible for cartilage formation) interact with extracellular matrix and growth factors. Extracellular matrix provides important signals for chondrocytes behavior , this complex interaction between chondrocytes and extracellular matrix is partially mediated by a family of extracellular matrix molecules receptors called integrin ⁽¹⁰⁾ .Chondrocytes have a specific biochemical relationship with inflammation and cytokines ⁽¹¹⁾ .Chondrocytes effectively regulate the immune responses by affecting T cells proliferation , impairing monocytes differentiation , inhibiting T cells response to antigen- dependent and independent proliferative stimuli ⁽¹²⁾ .Interestingly , collagen I signaling at the central nervous system injury sites and astrogliosis. Moreover, collagen types are bound with integrin. Hence, integrin subtypes are all existed in astrocytes ⁽¹³⁾ .Collagen I is degraded by a specific enzyme called collagenase 3 , it is a sub-type of collagenases which compromise a large family of reactive proteolytic enzymes ⁽³⁾ .Collagenase 3 is mainly expressed , biosynthesized by human chondrocytes, it has been suggested that collagenase 3 is not present in normal tissues but identified in some carcinomas tissues such as breast. Moreover , it has been reported that collagenase 3 gene in normal tissues is expressed at very minimal levels while its expression may be stimulated under physiological

conditions⁽¹⁴⁾. In this regard, meningotheial (arachnoid) cells play a crucial role in immunological responses via secretion of pro and anti-inflammatory cytokines signaling as immunomodulator factors. [By linking immunological responses related to meningotheial cells and chondrocytes as the secretors of collagen 3, the present study has reported and concluded that collagen I level is decreased in Iraqi meningioma patients (Table (1)) caused by defects in immune system reflected by meningotheial cells and mediated by the activity of chondrocytes which responsible for collagenase 3 biosynthesis, Figure (1). The biochemical role of collagen I in astrogliosis is pivotal, by protecting astrocytes from an expression of cell adhesion molecules and also by activating and driving astrocytes toward neuronal regeneration after injury⁽¹³⁾. According to astrocytes, a recent study has shown that microglia/astrocyte tumor interactions are highlighted in brain tumors and mediated by a number of factors but the exact mechanism was not understood⁽¹⁵⁾. Regarding magnesium, a recent study has shown that magnesium sulfate was given as a drug (treatment) to meningioma patients but the details were not highlighted. Remarkably, this study may give a hint to magnesium deficiency in meningioma patients without any absolute indicator⁽¹⁶⁾. In contrast, the present results disagree with a very old study which has found that magnesium level was significantly increased in sera of eight meningioma patients compared with normal subjects⁽¹⁷⁾. Additionally, a previous study has indicated a cellular response of chondrocytes to magnesium alloys⁽¹⁸⁾. Interestingly, a recent study agrees with the present study by revealing that magnesium deficiency can be linked with a number of neurodegenerative diseases such as Parkinson's and Alzheimer's diseases, this is why magnesium plays a pivotal role in maintaining health of the central nervous system. Magnesium promotes axon growth and neural stem cells proliferation, regulates inflammatory responses and inhibits apoptosis. In particular, magnesium regulates the transmission of neurotransmitters such as serotonin and dopamine.⁵ Collectively, all literature review cited above regarding magnesium supports the highly significantly decrease in magnesium levels compared with healthy subjects, Table (2). According to phosphorus, a previous study has indicated that a decrease in Alkaline phosphatase activity predicts the recurrence of meningiomas but the physiological role of this enzyme is poorly understood⁽¹⁹⁾. Alkaline phosphatase (ALP) is the enzyme responsible for hydrolysis of phosphate monoesters and generating a free phosphate group. Interestingly, ALP is a magnesium metalloenzyme⁽²⁰⁾. As a result, ALP deficiency may indicate lower levels of both magnesium and phosphorus and serves as a biochemical link between meningioma

and lower levels of magnesium and phosphorus. This relationship absolutely agrees with the results of the present study regarding the lower levels of meningioma patients compared with healthy subjects, Table (2) and Table (3). Remarkably, chondrocytes might also regulate phosphorus homeostasis. Hence, phosphorus is pivotal for hypertrophic differentiation and stability of the chondrocyte cell line⁽⁶⁾. Regarding biochemical compounds containing phosphorus, a previous study has reported that total phospholipid content in meningioma tumors is the same as that from tissues of the normal leptomeninges.²¹ Indeed, biochemical compounds containing phosphorus have remarkable roles regarding functions and integrity of the central nervous system. A recent study has shown that the phospholipid phosphatidylinositol-3-phosphate is essential for the regulation of the levels of cell surface receptors for neurotransmitters, also phosphatidylinositol-4-phosphate has been suggested to inhibit the neural accumulation of amyloid beta 42 oligomers⁽²²⁾. Additionally, sphingosine-1-phosphate receptors modulators have a role in the treatment of multiple sclerosis as immune modulators by reducing new inflammatory lesions in the absence of global immune suppression, these reactive modulators are expressed by neurons, oligodendrocytes, astrocytes and microglia⁽²³⁾. However, the present study has strongly revealed that levels of collagen I, magnesium and phosphorus in Iraqi meningioma patients are not affected by the gender caused by the non significant difference between males and females patients, (Table (1)), (Table (2)) and (Table (3)) Clinically, male and female have the same hormones (estrogen, progesterone and testosterone), but their interactions with body organs is absolutely different. Predominantly, testosterone is secreted by males' tests while little amounts of estrogen and progesterone are secreted by tests, adrenal glands and peripheral tissues such as adipose tissue and liver via conversion of other precursors hormones. On the other hand, females' ovaries secrete estrogen and progesterone, and a small amount of testosterone is secreted by females' adrenal glands and the ovaries⁽²⁴⁾. Remarkably, the most number of subjects enrolled in the present study are above 45 years. At this point, a recent study has shown that the most common patterns of multimorbidity in women are different compared with men⁽²⁵⁾. Nevertheless, regarding the present study levels of collagen I, magnesium and phosphorus in meningioma patients are approximately the same in both males and females.

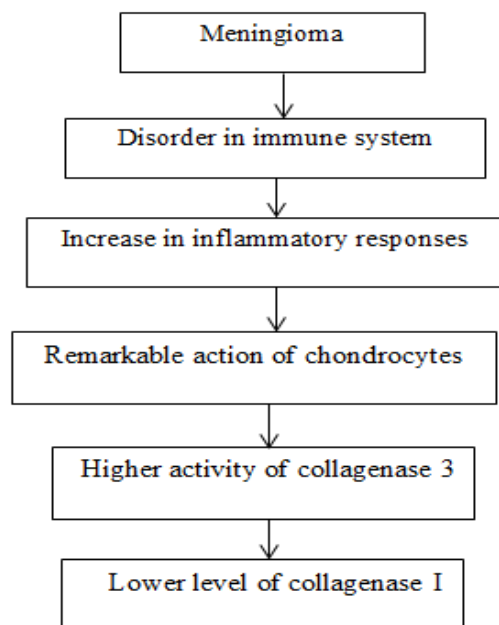


Figure 1. The suggested mechanism regarding lower levels of collagen I in Iraqi patients with meningiomas.

Conclusions

1. The present study is the first in Iraq reporting that a lower level of collagen I is a novel biochemical indicator for meningioma patients by highlighting inflammatory responses reflected by disorders in the immune system mediated by chondrocytes activity responsible for collagenase 3 biosynthesis which breakdown collagen I.

2. The present study has indicated the lower levels of magnesium and phosphorus in Iraqi meningioma patients by highlighting deficiency in ALP activity and the role of magnesium and phosphorus in the inflammatory responses and other neurodegenerative diseases.

3. Levels of collagen I, magnesium and phosphorus in Iraqi patients are not affected by the factors related to gender differences.

Recommendations

The present study recommends checking periodically the levels of collagen I, magnesium and phosphorus by laboratory tests and treatment with collagen supplements Tablets and metabolic (magnesium phosphate) capsules if a deficiency has been noticed.

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Ethics Statements

I (as an author) confirm that the published research article was performed according to the ethical considerations and I have received the ethical approval from my university and the Institute in which I did the blood sampling before recruiting patients. All clinical trials were conducted according to the [Helsinki Ethical Principles](#).

Conflict of Interest

Honestly I declare that there is no conflict of Interest regarding my research article.

Author Contribution

Rasha ZJ proposed the topic of this research article, designed the study protocol, collected blood samples and conducted the laboratory tests, performed the statistical analysis, discussed the results and wrote the manuscript and revised the manuscript draft, read and approved the final manuscript.

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