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## The role of Herpes simplex virus type 1 and 2 in patients with neurodegenerative diseases

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### Abstract

With the growing number of patient's being diagnosed with Parkinson's disease and Multiple Sclerosis each year it is becoming ever more important to find the cause for these neurological disorders. The present study attempts to shed light on one of the factors that may play a role as a causative agent in these neurological diseases by finding a correlation between the Herpes simplex virus type 1 and 2 in patients with Parkinson's disease and multiple sclerosis by detecting the virus in these patients using immunological techniques. Sixty patients with neurological diseases (40 patients with Multiple sclerosis and 20 patients with Parkinson's disease) who's ages ranged from (17-76) years have been investigated. Samples were collected during the time period between November 2017 and April 2018 and compared to twenty five apparently healthy individuals as a control group. All the studied groups were measured for herpes simplex virus type-1 IgM and herpes simplex virus type-2 IgM by using the enzyme linked immuno sorbent assay. The results of the present study showed that there was a highly significant difference ( $p < 0.01$ ) in the concentration of IgM HSV-1 and HSV-2 in the sera of patients with MS and PD compared to the control group, While there was a none-significant difference ( $p > 0.05$ ) in the concentration of IgM HSV-1 and HSV-2 according to the gender. Thus, there is a likely possibility the HSV could be a contributing factor in the activation of some neurological diseases by the means of initiating an autoimmune reaction against the host's nerve cells. The nerve cells have proteins that resemble portions of the virus from a structural and genetic stand point.

**Keywords:** Multiple Sclerosis, Parkinson's disease, herpes simplex virus, ELISA.

### دور فايروس القوباء البسيط النمط الاول والثاني في المرضى المصابين ببعض الامراض العصبية

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### الخلاصة

مع تزايد اعداد المرضى المصابين بالاضطرابات الحركية وتصلب الاعصاب المتعدد كل سنة ، اصبح من المهم ايجاد سبب هذه الاعتلالات العصبية وقد تم في هذه الدراسة محاولة تسليط الضوء على احدى العوامل التي تلعب دوركعامل مسبب في هذه الامراض العصبية وذلك بوساطة التحري عن فايروس القوباء البسيط النمط 1 او 2 في المرضى المصابين بالاضطرابات الحركية وتصلب الاعصاب المتعدد باستخدام التقنيات المناعية ، تم التحري عن ستين مريض يعانون من الاعتلالات العصبية (40) مريض بتصلب الاعصاب

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المتعدد و (20) مريض بالاضطرابات الحركية باعمار تتراوح من (17-76) سنة للفترة من تشرين الثاني 2017 ولغاية نيسان 2018 ، وتمت المقارنة مع 25 شخص من الاصحاء ظاهريا كمجموعة سيطرة. خضعت جميع عينات الدراسة لقياس مستوى اضرار IgM لفيروس القوباء البسيط النمط الأول والثاني باستخدام تقنية الامتزاز المناعي المرتبط بالانظيم، اظهرت نتائج الدراسة فرقا معنويا عاليا ( $p < 0.001$ ) في تركيز الاضرار IgM لفيروس القوباء البسيط النمط 1 و 2 في مصول المرضى المصابين بتصلب الاعصاب المتعدد والاضطرابات الحركية بالمقارنة مع مجموعة السيطرة، بينما لم يلاحظ هناك فروقا معنوية ( $p > 0.05$ ) في تركيز اضرار IgM لفيروس القوباء البسيط النمط 1 و 2 بالنسبة للعمر. استنتجت هذه الدراسة ان الفايروس يحتمل ان يكون احد العوامل المسببة لظهور الاعراض للامراض التي اجريت التجربة عليها كون الفايروس قادر علي بدء مرض مناعة ذاتية داخل المضيف مما يؤدي الى مهاجمة الخلايا العصبية في الانسان التي تمتلك بروتينات تشبه بروتينات موجودة بالفايروس من ناحية وراثية و تركيبية.

## Introduction

The herpes simplex virus type 1 is a virus that has a worldwide distribution estimated to be between 60 to 90% throughout the human population[1, 2]. In the majority of cases the virus is transmitted maternally from mother to child [3, 4]. As for the second strain known as type 2 has a far less distribution due to its route of transmission via sexual contact[5]. These two strains of the herpes virus are not considered life threatening but symptoms in both cases manifest in the appearance of ulcers in the oral and genital areas of the body[6]. Both viruses are unique in that they can evade the host's immune system by going into a dormant phase inside of the host's nerve cells[7, 8]. It is because of this dormancy that researchers believe that viruses may play a role in the activation of a number of neurological diseases like multiple sclerosis (MS) and Parkinson's disease (PD). MS is a chronic autoimmune, inflammatory neurological disease of the central nervous system[9]. MS attacks the myelinated axons in the central nervous system, destroying the myelin sheath and the axons to a degree[10]. PD is a chronic, neurodegenerative disease mainly caused by the degeneration of dopamine-producing cells in the substantia nigra, located in the midbrain which presents idiopathically[11]. To this day there is no known cure for PD or MS although there are certain medications that may alleviate some of the symptoms providing a better quality of life for the patient[12]. By frequent reactivation of this virus the host's immune system acquires a state of autoimmunity to proteins in the virus that are similar in structure to proteins found in the nerve cell[13, 14]. The aim of this study is to find a relationship between the herpes simplex virus type 1 and type 2 and the neurological diseases MS and PD.

### Methods for patient selection:

The study included a total of 85 samples, 40 samples with Multiple sclerosis, 20 samples with Parkinson's disease and 25 apparently healthy controls. The samples were collected from patients that attended the neurological consultancy at Baghdad Hospital and the Neurological Sciences Hospital in Baghdad between November 2017 and May 2018. The ages ranged between 17 and 76. All the samples were marked by a special number to identify the sample, the name of the patient, the date of sample collection and age of the patient along with the neurological disease affecting the patient.

### Blood sample collection:

Five milliliter of blood sample was collected from patients clinically diagnosed with Parkinson's disease and Multiple Sclerosis and healthy individuals. The blood sample was drawn into a yellow top gel vacuum tube and allowed to stand at room temperature for 10 minutes. The samples were then centrifuged at 4000 rpm for 10 minutes then the serum was transferred to plain white top tubes. All samples were stored at  $-20^{\circ}\text{C}$  until use in immunological tests.

### Immunological examination:

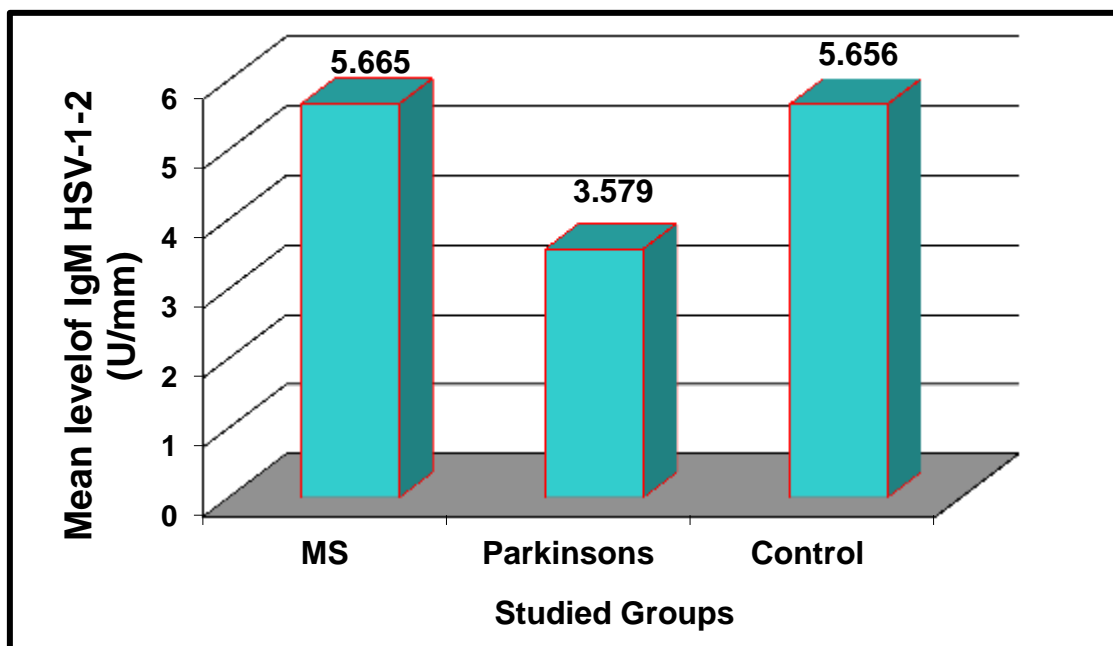
All the studied groups were tested by measuring anti -Herpes simplex virus type 1 and 2 IgM antibody by means of ELISA test (Clonit/Germany) according to the leaflet of kit[15].

### Statistical Analysis:

The Statistical Analysis System –SAS-program was used to find the effect of the different factors on the study parameters. ANOVA table was used to compare between means while Chi-square test was used to compare between percentages of this study [16].

### Results and discussion:

The results of this study revealed that there was a highly significant difference ( $p < 0.01$ ) in the concentration of IgM in the sera of MS and PD patients ( $5.66 \pm 0.36$ ) U/ml and ( $3.579 \pm 0.33$ ) U/ml compared to the control group ( $5.656 \pm 0.36$ ) U/ml as shown in Figure-1.



**Figure 1**-Mean level of anti HSV-1 and 2 IgM Ab (U/ml) in the sera of patients with neurological diseases (MS and PD) and control group.

Also the results of the present study show that there was a significant difference ( $p < 0.05$ ) in the concentration of IgM HSV-1 and 2 according to the age group as shown in Table-1.

**Table 1**-The effect of age on IgM mean concentrations

Age group (year)	Mean $\pm$ SE IgM (U/ml)
< 40	$5.67 \pm 0.46$
$\geq 40$	$4.43 \pm 0.35$
LSD value	1.139 *
P-value	0.033
* ( $P < 0.05$ )	

While there was a none significant differences ( $p > 0.05$ ) in the concentration of IgM HSV-1 and 2 according to the gender as shown in Table-2.

**Table 2**-The effect of gender on IgM mean concentrations

Gender	Mean $\pm$ SE IgM (U/ml)
Male	$4.67 \pm 0.41$
Female	$5.42 \pm 0.38$
LSD value	1.152 NS
P-value	0.194
NS: Non-Significant.	

The study analyzed the association between the Herpes simplex virus and two neurodegenerative disorders. With regards to the IgM concentrations the present results coincided with other studies like Jasminka and Azra who conducted their work on a total of 118 patients with MS in which they found that a wide majority of the samples tested positive for the virus in its IgG form but when they tested the same samples for IgM only one sample 0.84% out of the 118 tested positive.[17]

The majority of other studies that looked at IgM levels for HSV found much lower levels of positive samples than IgG for instance the researchers Tada and Khandelwal found that out of a total of 150 patients only 15.66% tested positive for the IgM antibody[18]. While out of the 118 patients that Jasminka and Azra tested only 0.84% turned out positive[17]. This trend can probably be traced back to the fact that the majority of humans are infected by the herpes virus at a young age[19]. A large number of these cases are transmitted maternally from mother to child[1]. The IgM antibody appears at the beginning of the initial infection, disappearing after a few days to give way to the appearance of other types of antibodies[20]. For this reason the likelihood of collecting a sample from a potentially infected patient during the small time frame in which the IgM antibody is readily available in their serum is unlikely. The sample must be collected during the acute phase of the initial infection in order to obtain adequate amounts of IgM to clinically test positive. For this reason the majority of the studies including the present study found the majority of the samples testing negative for HSV IgM antibody.

### Conclusion

The results of this study indicate that IgM levels are not sufficient enough to establish a clear association between the neurodegenerative diseases studied and the herpes simplex virus since the majority of the samples resulted in negative results due in part to the short time frame that the IgM antibody may readily be available for detection during an immunological reaction.

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