Original Article

Study of serum level of kisspeptin and interferon-beta in genital wart patients

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Abstract

Background: Researchers are interested in genital wart (GW) studies due to their increased incidence. In a single experimental research, virally infected mouse models showed elevated kisspeptin levels and low interferon levels. **Objective:** The objective of the study was to evaluate the serum levels of kisspeptin and interferon (INF)-beta in GW patients. **Patients and Methods:** Forty patients with GWs and forty healthy participants of comparable age and sex as a control group were included in this case–control study. Serum levels of kisspeptin and IFN-beta were measured using ELISA during the period from December 2021 to April 2022. **Results:** Kisspeptin was significantly higher among cases than controls, whereas IFN-beta level was lower among cases than controls ($P < 0.001$). There were no significant relations between kisspeptin and IFN-beta levels and the clinical data for the studied participants, and there was no significant correlation between both ($P > 0.05$). **Conclusion:** The reported increased kisspeptin level which was associated with decreased interferon-beta level in patients with GWs might indicate a new insight into viral infection pathogenesis. Further research including all steps in kisspeptin/G protein-coupled receptor 54 pathway is required. Targeted therapy for this pathway may be of value for those patients.

Key words: Genital warts, interferon-beta, kisspeptin

Introduction

Researchers worldwide are interested in genital wart (GW) pathogenesis which might allow better understanding of more protective and treatment measures. The rapidly increased incidence of GWs, being the most common sexually transmitted disease worldwide, and the carcinogenic potential of human papillomaviruses (HPVs) are the reason for this interest.[1] There are around 15 high-risk mucosal HPV strains that are firmly linked to cervical cancer. The most carcinogenic viruses are HPV-16 and HPV-18. These viruses are now known to be related to a specific group of genital malignancies.[2] There are more than 100 different varieties of HPV, of which between 30 and 40 are related to the skin and mucosa in the anogenital region. Approximately 90% of GW cases are caused by HPV types 6 and 11.[3] GWs are generally asymptomatic except for some dyspareunia and discomfort in case of vulvar warts and penile warts. Perianal and intra-anal warts might cause bleeding. Vaginal warts might be associated with vaginal discharge, bleeding, obstruction of the birth canal, and neonatal infection which may result in juvenile-onset recurrent papillomatosis.[4]

In response to the attack by various viruses, host cells create and release signaling proteins known as interferons, which alter the immune system's response to viral infections.[5] Regarding the ability of interferon therapy to eradicate the virus from the compromised cells, it was demonstrated that patients with condylomas and T-helper lymphocyte insufficiency showed a good improvement after receiving interferon therapy.[6] The human brain and other genetically related species manufacture the protein known as kisspeptin, which is encoded by the KISS-1 gene. It plays a crucial role in reproduction.[7] The primary receptor of neuropeptide hormone kisspeptin, G protein-coupled receptor 54 (GPR54), is vital for immune response monitoring and regulation.[8] Several

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studies have suggested that kisspeptin/GPR54 has a part in immunological modulation in response to immune stress caused by lipopolysaccharide (LPS).[9] However, the exact role of kisspeptin/GPR54 pathway in innate immunity and host defense against viral pathogen infection is still unknown. Virally infected mice had elevated levels of kisspeptin, which were linked to lower levels of interferon-beta.[10] We conducted this study to assess the serum levels of kisspeptin and interferon-beta in patients with GWs.

Patients and Methods

A case–control study was carried out on eighty participants, during the period study from December 2021 to April 2022. All participants included in this study were divided into Group I, including forty patients with GWs, and Group II, including forty healthy participants of comparable age and sex as a control group.

Ethical consideration

After explaining the purpose of the present study to the participants, their written informed consent was obtained. The Committee for Human Rights in University Research approved the study.

Inclusion criteria

Inclusion criteria were immunocompetent patients with newly diagnosed GWs of both sexes.

Exclusion criteria

Patients before and around the age of puberty, patients with precocious and delayed puberty, patients with fertility disorders, patients with AIDS, hepatitis C virus, hepatitis B virus, or other sexually transmitted infections, and patients with malignant transformation were excluded from the study.[10]

All the chosen cases in the current study underwent the following: complete history taking with personal information: name, age, and sex; present history: course (progressive, regressive, or stationary), recurrences,[11] and duration of the clinically apparent lesion (according to the patient own wards); family history; and history of medical conditions or drug use. A general examination was conducted to find any excluding factors. Warts were diagnosed clinically and examined dermatologically for size, number, shape, and location.

Laboratory investigations

Under strict aseptic conditions, 2 ml of venous blood was drawn and slowly transferred into a plain tube. The samples were centrifuged at 1000 × g for 15 min after being allowed to clot for 30 min. Separated serum was stored in aliquots and maintained frozen at −80°C. Following that, the ELISA method (Sunred Biological Technology, Shanghai, China) was utilized for measuring the serum levels of kisspeptin and interferon-beta.

Statistical analysis

Using an IBM personal computer and the statistical software SPSS version 23, the results were gathered, tabulated, and statistically evaluated (IBM Corp., 2013: Armonk, NY, USA). Analytical statistics included the Chi-square test (χ^2), Mann–Whitney test (U), Kruskal–Wallis (K) test, and Spearman's correlation (r), with $P = 0.05$ being deemed statistically significant. Descriptive statistics included percentages (%), means ($\bar{x}$), and standard deviations.

Results

Demographic data of the studied groups

The mean age of the studied cases was 30.4 ± 9.57 years and among controls was 34.0 ± 11.4 years. In the cases group, the number of males is equal to the number of females, while in the control group, 55% were female and 45% were male with no significant differences between groups regarding their age and sex [Table 1].

Clinical data and clinical criteria of warts among the studied cases

Seventy percent of the patients had progressive course, 30% had stationary course, and 30% had recurrent warts [Table 2]. The mean duration of GW disease was

Table 1: Comparison between the studied groups regarding demographic data (*n*=80)

Variables	Studied groups (*n*=80)		*U*	*P*
	Cases (*n*=40)	Control (*n*=40)		
Age (years)				
Mean±SD	30.4±9.57	34.0±11.4	1.33	0.182
Median	29.5	32.5		
Range	18-54.0	18-58.0		
Sex				
Male	20 (50.00)	18 (45.00)	χ^2=0.201	0.654
Female	20 (50.00)	22 (55.00)		

Significance level at P<0.05. U=Mann-Whitney test; χ^2=Chi-square test; SD=Standard deviation

Table 2: Clinical data and clinical criteria of wart among the studied cases (*n*=40)

Studied variables	Studied patients (*n*=40), *n* (%)
Course	
Stationary	12 (30.0)
Progressive	28 (70.0)
Duration (months)	
Mean±SD	6.48±6.06
Median	5.00
Range	1-30
Family history	
Positive	12 (30.0)
Negative	28 (70.0)
Recurrence	
Yes	12 (30.0)
No	28 (70.0)
Clinical criteria of wart	
Site	
Labia	20 (50.0)
Penis	10 (25.0)
Penis and scrotum	7 (17.5)
Perianal	3 (7.50)
Shape	
Verrucous	13 (32.5)
Filiform	4 (10.0)
Cauliflower	6 (15.0)
Flat papules	17 (42.5)
N	
Mean±SD	4.55±2.54
Median	4.00
Range	1.00-12.0
Size/mm	
Mean±SD	5.05±1.86
Median	5.00
Range	2.00-9.00

SD=Standard deviation

6.48 months and ranged from 1 to 30 months. Positive family history presents in 30% of the patients. Regarding the site of warts, in females, all patients had labial warts and no other sites were found in females. Furthermore, in males, 25% of the studied patients had penile warts, followed by 17.5% having penis and scrotum warts, and only 7.5% of them had perianal warts. Regarding the shape of warts, 42.5% of the studied patients had flat papules, 32.5% had verrucous papules, 15% had cauliflower growths, and only 10% had filiform GWs. The mean number was 4.55, with a range of 1–12 warts, and the mean size was 5.05, with a range of 2–9 mm.

Kisspeptin and IFN-beta levels among the studied groups

Kisspeptin was higher in cases (204.2 ± 156.2) than in controls (54.7 ± 18.5), whereas lower IFN-beta level was detected in cases (66.8 ± 18.2) than in controls (180.2 ± 131.0) ($P < 0.001$) [Table 3].

Relation between kisspeptin and IFN-beta levels with clinical data of the studied patients

There were no significant relations between kisspeptin and IFN-beta levels with sex, onset, course, family history, recurrence, site, and shape of warts ($P > 0.05$) [Table 4].

Table 3: Comparison between the studied groups regarding kisspeptin and interferon-beta levels (*n*=80)

Studied variable	Cases (*n*=40)	Controls (*n*=40)	*U*	*P*
Kisspeptin				
Mean±SD	204.2±156.2	54.7±18.5	7.32	<0.001**
Median	129.4	49.5		
Range	57.1-597.7	10.1-100.7		
IFN-β				
Mean±SD	66.8±18.2	180.2±131.0	7.47	<0.001**
Median	67.8	131.1		
Range	15.7-99.0	96.2-776.3		

**Highly significant. *U*=Mann-Whitney test; SD=Standard deviation; IFN= Interferon

Correlation between kisspeptin and IFN-beta levels with age, disease duration, number of warts, size of warts, and IFN-beta among studied patients

There were no significant correlations between kisspeptin and IFN-beta levels with each other or with age, disease duration, number of warts, and size of warts among studied patients ($P > 0.05$) [Table 5].

Discussion

HPV infection with specific strains is what causes GWs.[3] After thorough searching, there are no published studies on kisspeptin hormone in GW sufferers. However, it was found that the primary receptor for the neuropeptide hormone kisspeptin, GPR54, is crucial for controlling immunological response through the regulation of interferon secretion.[12]

Therefore, the purpose of this study was to assess the blood levels of interferon-beta and kisspeptin in GW patients.

Our findings revealed that the levels of kisspeptin were significantly greater in the GW patients than in the controls. Up to our knowledge, this is the first study to demonstrate the relation between kisspeptin and GWs caused by HPV infection. In line with our results, Huang *et al*.[12] revealed that, during viral infections, kisspeptin levels in mouse serum significantly rose. They discovered kisspeptin to be a new virus-induced neuropeptide hormone engaging GPR54, which in turn activates calcineurin, which dephosphorylates TBK1 and prevents the synthesis of IFN-I.

In addition, Luedde *et al*.[13] studied 133 critically sick patients (94 of them having sepsis and 39 being admitted to the intensive care unit for other reasons) and revealed a significant rise in kisspeptin levels. Kisspeptin levels were not related to the cause of critical illness, but they correlate with the immune system's level of activation.

The relation between kisspeptin and immune system could be explained by Cerdeira *et al*.,[14] who

Table 4: Relation between kisspeptin and interferon-beta levels with clinical data of the studied patients (*n*=40)

Studied variables	Kisspeptin level, mean±SD	Test *P*	IFN-β level, mean±SD	Test *P*
Sex				
Male	206.9±171.2	*U*=1.25, *P*=0.211	62.7±16.9	*U*=0.602, *P*=0.547
Female	217.1±154.6		70.8±18.9	
Course				
Stationary	187.8±148.6	*U*=0.742, *P*=0.458	69.5±16.3	*U*=0.384, *P*=0.701
Progressive	221.3±167.2		65.6±19.2	
Family history				
Positive	233.3±188.1	*U*=0.050, *P*=0.960	56.6±22.1	*U*=0.481, *P*=0.631
Negative	201.4±148.6		71.2±14.6	
Recurrence				
Yes	195.0±148.5	*U*=0.681, *P*=0.496	69.1±18.9	*U*=0.897, *P*=0.370
No	230.8±176.3		64.8±17.8	
Site				
Labia	217.2±154.6	*K*=2.18, *P*=0.535	70.8±18.9	*K*=2.80, *P*=0.423
Penis	240.9±197.6		64.4±11.8	
Penis and scrotum	193.9±156.7		62.1±25.4	
Perianal	99.5±59.9		58.9±11.6	
Shape				
Verrucous	276.8±196.9	*K*=2.38, *P*=0.496	65.1±21.4	*K*=1.24, *P*=0.743
Filiform	139.2±28.1		60.7±25.5	
Cauliflower	218.4±159.2		64.1±12.1	
Flat papules	192.6±158.2		70.5±16.2	

Significance level at *P*<0.05. *U*=Mann-Whitney test; *K*=Kruskal-Wallis test; SD=Standard deviation; IFN= Interferon

Table 5: Correlation between kisspeptin and interferon-beta levels with age, disease duration, number of warts, size of warts, and interferon-beta among studied patients (*n*=40)

Studied variables	Kisspeptin level		IFN-β level	
	R	*P*	*r*	*P*
Age (years)	0.090	0.602	−0.125	0.443
Disease duration (months)	0.062	0.720	−0.073	0.652
Number of warts	−0.104	0.546	−0.123	0.450
Size of warts	0.062	0.717	0.158	0.329
IFN-β	0.329	0.144	-	-
Kisspeptin level	-	-	0.329	0.144

Significance level at $P<0.05$. *r*=Pearson correlation coefficient; IFN=interferon

demonstrated that natural killer (NK) 1 cells are induced to differentiate into NK3 cells by kisspeptin. In addition, NK cells are stimulated to produce transforming growth factor which facilitates the differentiation of peripheral NK cells into decidual NK cells. Furthermore, Shirshev *et al*.[15] revealed a considerable rise in the percentage of CD56-bright cells with increased percentage of L-selectin expression in kisspeptin-treated cultured cells when compared to controls. Kisspeptin was suggested to have an inhibitory effect on some LPS-induced pro-inflammatory cytokines (interleukin [IL]-1, tumor necrosis factor [TNF]-α, and IL-6) while generally having no effect on some anti-inflammatory cytokines.[16]

In the study by Ford and Thomas,[17] there were decreased levels of interferon-beta, indicating that HPV is able to affect the antiviral immune response by reducing the presence of interferons, causing a less effective antiviral state. In addition, the research by Syed and Ahmadpour[18] and Yang *et al*.[19] demonstrated a highly significant difference between GW patients and controls in terms of IFN-beta level, which was considerably lower in instances of GWs than controls ($P < 0.001$). The demonstrated lower IFN-beta levels in the cases group in comparison with the control group in the current study are in line with their findings.

On the other hand, the study by Lace *et al*.,[20] Lynde *et al*.(2020)[4] and Radi *et al*. (2018)[21] revealed that IFN-beta levels were noticeably higher in patients with GWs in comparison with controls. Trinchieri[22] indicated that IFN-I might have either protective or detrimental effect in infections and autoimmune disorders, as the host, pathogen, and environmental variables are responses for immunological homeostasis through the tight control of IFN-I response. This might provide an explanation for these contradictory data.

The levels of kisspeptin and IFN-beta did not significantly correlate with patient age, disease duration, number of warts, or wart size in this investigation. In a similar vein, the study by Huang *et al*.[12] reported no significant relationship between the animals' kisspeptin levels and their ages or the length of their illnesses ($P > 0.05$). Moreover, IFN-beta was strongly connected with disease duration, the number of warts, and the size of warts ($P = 0.05$) according to the study by Yang *et al*.[19] Erel *et al*.[23] indicated that age and serum kisspeptin levels had a substantial and negative correlation ($r = -0.458$). Compared to the other age groups over 25 years, the group between 20 and 24 years had the greatest amounts of kisspeptin ($P < 0.001$).

Limitations of the study

There are some restrictions on this study, including the small sample size, being a one-center investigation, and studying just two inflammatory indicators rather than evaluating many markers at once.

Conclusion

The observed elevated kisspeptin level in patients with GWs, which was correlated with a reduction in interferon-beta level, may indicate a novel understanding of the pathogenesis of viral infection. Further studies encompassing every stage of the kisspeptin/GPR54 pathway are necessary. For such patients, tailored medication for this pathway may be beneficial.

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Conflicts of interest

There are no conflicts of interest.

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