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Brain-derived neurotrophic factor (BDNF) Single nucleotide Gene Polymorphism And Nerve growth factor Are Risk Factors That Increase The Severity Of Allergic Rhinitis

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Abstract

Allergic rhinitis is a health issue that affects nasal membranes, causing congestion and sneezing. It is caused by type 2 helper cells that produce Immunoglobulin E (IgE) that reacts to allergens. Brain-derived neurotrophic factor (BDNF) Single nucleotide gene polymorphism and Nerve growth factor (NGF)are useful biomarkers for allergic phenotypes, and NGF plays a role in the pathophysiological mechanisms underlying airway inflammation and hyperresponsiveness. Our goal is to improve the diagnosis and treatment of this condition by studying if NGF levels differ between people with allergic rhinitis and healthy people, as well as the novel link between BDNF rs6265 polymorphisms and their severity. To achieve this, we conducted a case-control study among 120 subjects with rhinitis of both sexes. Three groups of individuals were created: those with mild allergic rhinitis, those with moderate-to-severe allergic rhinitis, and a control group diagnosed according to clinical examination and skin prick tests for allergen sensitivity. The present study conducted complete blood count (CBC), C-reactive protein (CRP), Interleukin-1β (IL-1β), Interleukin-6 (IL-6), Nerve growth factor (NGF), and genotyping for BDNF rs6265 polymorphism. This study explored the link between NGF, BDNF gene variations, and allergic rhinitis severity in adults from Upper Egypt. The results showed that higher NGF levels were associated with more severe allergic rhinitis, but skin prick tests were more closely associated with the condition's severity. Inflammatory markers IL-6 and IL-1β levels increased with severity, confirming their established roles in allergic rhinitis. Contrary to existing research, the most common genotype for BDNF rs6265 in this population was GG. The research found that BDNF rs6265 gene polymorphism is associated with an increased risk of moderate-to-severe AR. The most prevalent genotype in the current research was GG. NGF levels were higher in AR cases than in controls and can act as a potential biomarker in the diagnosis of AR. IL-1β and IL-6 may also be biomarkers in the diagnosis of AR.

Keywords: BDNF gene Polymorphism, NGF, Allergic Rhinitis, CRP, IL-1β and IL-6

1. Introduction

Most frequently affecting the nasal membranes, a condition known as allergic rhinitis (AR) is characterized by symptoms like congestion and sneezing. It is an international health concern; epidemiological surveys indicate that 10–40% of people worldwide, possibly up to 50% in certain nations [1], suffer from AR. Due to the non-life-threatening nature of the disease, the burden of AR is usually underestimated. On the other hand, it may have a substantial negative impact on quality of life and well-being in addition to causing physical symptoms [2]. subsequently suggested to cause exhaustion and fluctuations in mood [3], impair cognitive function [4], and cause anxiety and depressive disorders [5], it has a significant impact on numerous aspects of daily life, including sleep [6], work/school [7], and social life. It is commonly known that type 2 helper (Th2) cells cause allergic reactions (AR), which in turn cause B cells to secrete particular immunoglobulin E or IgE, that acts against Allergens in the air [8]. Due to the infiltration of inflammatory cells and the inflammatory cytokines that they secrete, such as interleukin-4 (IL-4)/interleukin-5 (IL-5)/interleukin-13 (IL-13), the microenvironment of the immune system has altered, which has an impact on the normal growth pattern of the epithelium [9]. This includes dysfunction of the epithelial barrier, nasal sensitization, elevation of nasal

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mucous, and dysfunction of the cilia cells in the nasal mucosa. An important part of allergic diseases is played by interleukin-1β (IL-1β), one of the fundamental elements in the occurrence and development of inflammation [10]. Research has shown that allergen-induced inflammation and its clinical manifestations, including runny nose, itching, redness, and rashes, may be closely associated with IL-1β. Further research has revealed a significant increase in IL-1β levels in allergic diseases [11], and inhibiting the IL-1β signaling pathway may somewhat lessen the intensity of inflammation. Consequently, it has been suggested that IL-1β may serve as a biomarker for AR. Activated T cells and fibroblasts produce the lymphokine interleukin (IL)-6, which has grown to be a crucial biomarker for the identification of infectious diseases and inflammation. According to related research, IL-6 is implicated in allergies and has a significant impact on how they arise and develop [12]. The nasal mucosa secretes neurotrophins, such as brain-derived neurotrophic factor (BDNF) and nerve growth factor (NGF) [13]. The BDNF gene is located on chromosome 11p13, and because of alternative splicing, it generates several transcripts [14]. Genetic variations in BDNF have been connected to the pathophysiology of allergic rhinitis (AR), atopic asthma, and eczema in several populations [14]. It has been demonstrated that the functional polymorphism rs6265 (Val66Met) controls intracellular transport and influences BDNF secretion [15]. This polymorphism has been linked to asthma in German children and other genetic variations in BDNF [16]. However, other research indicates that although BDNF is a useful biomarker for allergic phenotypes, allergic diseases are not linked to genetic variations in BDNF [17], [18]. How genetic variations in the BDNF gene relate to allergic disorders is still unknown. According to reports, nerve growth factor (NGF) regulates nerve growth, neurotransmission, differentiation, and survival of neurons [19]. Additionally, NGF has multidirectional effects on the mediation of several pathophysiological processes, such as the production of catecholamines, inflammatory responses, and immune responses [21–26]. According to reports, compared to mice in the control group, the nasal lavage fluids of AR mice exhibit significantly higher expressions of NGF, IL-6, and vasoactive intestinal peptide (VIP) [27]. NGF is primarily found in inflammatory cells and is persistently and highly expressed in the nasal epithelium and submucosal tissues of AR patients, according to prior research [28–29]. It has also been demonstrated that after allergen stimulation, the level of serum NGF significantly increases in AR subjects [30]. Also, it has been suggested that NGF plays a role in the pathophysiological mechanisms underlying airway inflammation and hyperresponsiveness in AR patients [31] both the upper and lower airways are mediated by NGF [32]. Up till now, No study has been done on the upper Egypt population on BDNF rs6265 polymorphisms and NGF as a possible blood biomarker in allergic rhinitis. So, the purpose of the present study was to identify the NGF concentration in the blood of patients with allergic rhinitis and control subjects, as well as look into the possible association between BDNF rs 6265 polymorphisms and allergic rhinitis, its severity, and various phenotypes.

2. Materials and Methods

2.1 Study design.

In collaboration with the ENT and medical biochemistry departments at Sohag University, a prospective case-control study was carried out. Before starting the study, every patient received information about the possible advantages and disadvantages of skin testing, and they all signed written informed consent forms to take part. The Sohag University Hospitals Local Ethical Committee gave its approval and the study was carried out in compliance with the Declaration of Helsinki. The Sohag University Faculty of Medicine's ethical committee approved the study's protocol. This approval is registered with the IRB under the number Soh-Med-23-04-09PD. It is also accessible on ClinicalTrials.gov under the ID NCT05907733. Furthermore, all research participants provided written consent following receipt of all information.

2.2 Study population.

This study included a total of 120 subjects with rhinitis, of both sexes. Sample size calculation was done by G*Power 3 software [32]. A calculated sample of 60 respondents (case and control) was needed with an error probability of 0.05% and 80% on a one-tailed test (type 1 error).

- The exclusion criteria Patients taking non-steroidal anti-inflammatory drugs, smokers, those with immunological diseases, those who are pregnant or nursing, and those with vasomotor rhinitis.
- The clinical examination, with a focus on rhinitis symptoms, and a comprehensive allergic history were performed on the participants. Age at which rhinitis symptoms first appear, probable triggers, frequency, and intensity of symptoms.
- Three groups of patients were created: one for mild allergic rhinitis, one for moderate-to-severe allergic rhinitis, and one for control. Guidelines for allergic rhinitis and its impact on asthma were established in 2001 and based on these criteria; allergic rhinitis was diagnosed and categorized as mild, moderated, or severe.

• All patients undergo skin prick tests (SPTs) using commercial extracts for a standardized panel of food and inhalant allergens.

2.3 Laboratory Workup

Under strictly sterile conditions, peripheral vein blood samples were drawn into chemistry tubes and blood collection tubes containing EDTA. For ten minutes, samples were centrifuged at 5,000 g. The plasma was separated and then kept cold (-80 °C) until analysis.

2.4 Complete blood count (CBC).

We used the Cell Dyne 3700 Ruby TM, an automated cell counter from Abbott Diagnostics (USA), to perform the CBC. To do this, we took 2ml of blood in a tube with EDTA and put it into the Cell Dyne Ruby TM. The machine then gave us the CBC results.

2.5 C-type reactive protein.

The CRP Latex (lot CR286F, Spinreact, Spain) was used to estimate the amount of C-reactive protein (CRP). Samples and reagents were allowed to reach room temperature. 50 µL of a 1ml serum sample and one drop of each positive and negative control were added to separate circles on a slide test. CRP-latex reagent was added next to the samples. The slide was placed on a rotator for 2 minutes. After removing the slide, a macroscopic examination was carried out to check for the presence or absence of visible agglutination, which indicates the CRP concentration. The approximate CRP concentration in the patient sample is calculated as follows: 6 x CRP Titer = mg/L.

2.6. *Assay of serum IL-1β:*

The IL-1 β ELISA kit was utilized (catalog no. E0143Hu Biotech Co., Ltd, Shanghai, China). Every step was completed according to the manufacturer's protocol. Within ten minutes of adding the stop solution, the absorbance of the color change at a wavelength of 450 nm must be detected using the Stat Fax 2600 Microplate Reader, USA, to measure the optical density (OD). Using the standard concentration and matching OD values, find the linear regression equation for the standard curve. The concentration of the corresponding sample can then be found by applying the sample OD values to the regression equation.

The detection range is 20-6000pg/ml [33].

2.7 An assay of serum IL-6:

(subreddit.com, Shanghai, China; catalog no. 201-12-0091) was utilized. Human interleukin 6 (IL-6) levels in samples are measured using the double-antibody sandwich enzyme-linked immunosorbent assay (ELISA) kit. Using a Stat Fax 2600 Microplate Reader, the OD was measured at 450 nm wavelengths. We Calculated the linear regression equation for the standard curve using the concentration and corresponding OD values. Next, use the sample OD values on the regression equation to determine the concentration of the corresponding sample.

The detection range is 1.5 pg/ml to 300 pg/ml [33]

2.8 Assay of Nerve growth factor in serum:

Using an Enzyme Immunosorbent Assay (ELISA) kit obtained from Sinogeneclon Biotech Co., Ltd. in China (catalog No. SG-11226), nerve growth factor (NGF) was found in serum samples. The kit uses an enzyme-linked immune sorbent assay (ELISA) with double-antibody sandwiching to determine the concentration of human nerve growth factor (NGF) in samples. The absorbance of the color change at a wavelength of 450 nm was measured using a Stat fax 2100 Microplate Reader, USA. This measurement must be completed within 15 minutes of adding the stop solution. Using the standard concentration and matching OD values, find a linear regression formula for the standard curve. Next, use the regression equation and the sample OD values to find the concentration.

Detection range: 16pg/ml – 1000 pg/ml [34].

2.9 Genotyping

2.9.1 DNA Extraction, a DNA quick blood genomic extraction kit (ABT blood DNA Mini Extraction kits cat No.ABT003) was used to extract DNA from whole blood while following the manufacturer's recommendations. Before analysis, the collected DNA was kept at -20 °C.

2.9.2 *Brain-derived neurotrophic factor (BDNF)*, an allelic discrimination assay was done at real-time PCR to genotype of BDNF gene. Sequences were provided by Thermo Fisher Scientific catalog number (4351379) (TaqMan Master Mix, Applied Biosystems), the Master Mix of qPCR (300). TCCTCATCCAACAGCTCTTCTATCA(G/A)GTG TTCGAAAGTGTC.AGCCAATGAT (F-primer) is the context sequence for the BDNF rs6265 gene. 12.5 ul of the master mix was mixed with 12.5 µl of the master mix was mixed with 1.25 µl of the SNP genotyping assay primer mix and 6.25 µl of DNAase-free water. Each sample received 5µl of genomic DNA extract, and the negative control reaction received 5µl of DNAase-free water PCR amplification was done by thermal cycling conditions were adjusted as follows: Hold 10 min at 95° C for AmpliTaq Gold Enzyme Activation, 40 Cycles of denaturation (at 92 °C for 15 sec), and annealing extension (at 60 °C for 1 min for each). Loaded the reaction plate into the thermal cycler, and then the run was started. After PCR amplification, we performed an endpoint plate read using (Applied Biosystems StepOneTM Instrument,

Singapore). The Sequence Detection System (SDS) Software uses the fluorescence measurements made during the plate read to plot fluorescence (Rn) values based on the signals from each well. The plotted fluorescence signals indicate which alleles are in each sample.

2.10. Statistical analysis

We use Statistical Program for Social Science (SPSS) version 26(Armonk, NY: IBM Corp). The gathering of data was recorded and evaluated on an IBM-compatible computer. Tests of significance such as the CHI square test, Mann-Whitney, and Kruskal-Wallis H test were utilized for comparing different parameters in our participants. The receiver operating characteristic (ROC) curve and area under the curve (AUC) analysis were used to assess the diagnostic value. The significance level was less than 0.05 in all statistical tests.

3. Results

Our sample consists of (120) participants in a 1:1 design (30) Mild-moderate cases, (30) Sever cases of allergic rhinitis, and (60) healthy controls. Among them, 30 Mild-moderate cases (mean age 38.83±11.44years, range 20-60years; 16 males (53.3%)), and 30 severe cases of allergic rhinitis (mean age 41.31±10.4years, range 21-59 years; 13 (43.3%)). Sixty there were also groups of normal controls that were both gender and age correspond to the patient groups.

The study SNP followed the Hardy-Weinberg equilibrium Equation (HW). Hardy-Weinberg equilibrium was performed by chi-square (test of goodness of fit) with one-degree freedom using Gene pop software version 4.7 in Mild –Moderate cases are (p-vale is 0.925 >0.05, $\chi 2 = 0.157$), in severe cases are (p-vale is 0.689 >0.05, $\chi 2 = 0.745$) and in healthy control are (p-vale is 0.759 >0.05, $\chi 2 = 0.552$). This means our study groups follow the Hardy-Weinberg equilibrium.

3.1 In our result there were no significant differences between cases and the controls regarding age, BMI, and SBP (P= 0.628, 0.625, 0.047) respectively. There was a significant difference between cases and control regarding DBP (p=0.04). (Table 1).

*P < 0.05 (significant) **P < 0.01 (highly significant) ***P < 0.001 (very highly significant) NS: Nonsignificant p >0.05. P between three groups, P1 between Mild -moderate vs severe cases.

3.2 There was a non-significant difference between cases and the controls regarding the distribution of Gender and Smoking (P= 0.719, 0.488) respectively. There was a significant difference between Mild -moderate and severe cases of AR regarding skin prick test (p=0.0001). (Table 2).

3.3 There were increases in the level of CRP, IL-1β, IL-6, and Nerve Growth factor(NGF) in serum of Mild -moderate and severe cases than their level in controls as p=0.0001. Hg, WBCs, and platelets levels in the blood of Cases and controls show non-significant differences as (P= 0.04,0.818, 0.408) respectively. (Table 3)(figure A).

3.4 We found that were are excellent correlations between serum NGF and the following: Disease duration, IL-1β, IL-6, and CRP (Table 4)(figure B).

3.5 The most common gene variant in study groups was GG followed by GA and AA but there was a non-significant difference between cases and the controls regarding the genotyping distribution of the BDNF gene (P= 0.891)& G allele was the most common in study groups (Tables 5).

3.6 Independent predictors for the development of Severe allergic rhinitis. Univariate binary logistic regression analysis revealed that NGF, IL-1 serum level, and IL-6 were substantially linked to the occurrence of severe allergic rhinitis. NGF (odds ratio: 0.973, confidence interval 95%: 0.963-0.984, P =0.0001), IL-1 serum level (odds ratio: 0.995, confidence interval 95%: 0.992-0.997, P =0.0001) and IL-6 serum level (odds ratio: 0.875, confidence interval 95%: 0.785-0.977, P =0.017).

3.7 Multivariate binary logistic regression analyses do not confirm that (Table 6, 7).

3.8 Serum NGF was asignificant predictor test for allergic rhinitis with P= 0.000, sensitivity 85.2%, specificity 98.3%, and accuracy rate 91.2 %. (Table 8) (figure C).

3.9 There were non-significant differences between the homozygous G/G, A/A, and heterozygous G/A and BDNF gene polymorphism regarding the serum Nerve growth Factor concentration (P = 0.439). (Table 9).

Table (1) Clinical and Demographic variables of the study groups

Groups		Age	Duration of symptoms	BMI (kg/m^2)	DBP	SBP
Mild –Moderate cases (n=30)	Mean± SD	38.83±11.44	6.35± 2.84	25.2±2.5	78.33±8.4	121.17±12.1
	Median (Range)	39(20-60)	10(2-12)	25(19-31)	80(60-90)	120(100-150)
Severe cases (n=30)	Mean± SD	41.31±10.4	6.9± 2.01	24.6 ± 3.24	75.7±8.7	115.3±13.4
	Median (Range)	42.5(21-59)	7.5(3.5 -11)	24.5(19-31)	72.5(60-90)	115(90-150)
Control (n=30)	Mean±SD	39.9±10.6	0	25.3± 3.1	80.6±8.8	121.7±12.6
	Median (Range)	41.5(21-60)	0	25.3(19-32)	80(60-95)	120(90-150)
	p-value p1	0.628 0.403	0.0001 0.262	0.625 0.444	0.04 0.218	0.047 0.055

*P < 0.05 (significant) **P < 0.01 (highly significant) ***P < 0.001 (very highly significant) NS: Non-significant p >0.05. P between three groups, P1 between Mild -moderate vs severe cases.BMI (Body Mass Index), DBP(diastolic blood pressure) and, SBP (Systolic Blood pressure).

Table (2) Sex, Smoking, and skin prick test distribution among the study group

Groups		Frequency	*p*-value *p*	*p1*
Mild -Moderate Cases (N=30)	Smoking • Non-smoking • Smoking	 19 (63.3%) 11 (36.7%)	0.719	0.566
	Gender • Male • Female	 16 (53.3%) 14 (36.7%)	0.488	0.621
	Skin Prick test • Positive • Negative	 19 (63.3%) 11 (46.7%)	0.0001***	0.0001***
Sever Cases (N=30)	Smoking • Non-smoking • Smoking	 18 (60%) 12 (40%)		
	Gender • Male • Female	 13 (43.3%) 17 (56.7%)		
	Skin Prick test • Positive 26 (86.7%) • Negative 4 (13.3%) (46.7%) Negative			
Control (N=60)	Smoking • Non-smoking • smoking	 41 (68.3%) 19 (31.7%)		
	Gender • Male • Female	 34 (56.7%) 26 (43.3%)		

*P < 0.05 (significant) **P < 0.01 (highly significant) ***P < 0.001 (very highly significant) NS: Non-significant p >0.05. P between three groups, P1 between Mild -moderate vs severe cases.

Table (3) Biochemical tests among the study group

Groups	Parameters	Mean± Std. Deviation	Median(Range)	*p*-value *p*	*p1*
Mild -moderate cases (N=30)	Hg (g/dl)	11.703±1.4806	5.8(9.2-15)	0.04	0.269
	WBCS (10^3/µl)	8.146±5.59293	31.7(3.9-35.6)	0.818	0.739
	Platelets (10^3/µl)	281.9±47.345	173(213-386)	0.408	0.333
	CRP (mg/L)	27.23±17.252	59(6-65)	0.0001***	0.270
	IL-1β (pg/ml)	2554.75±684.8073	2752.2(1456- 208.2)	0.0001***	0.615
	IL-6 (pg/ml)	346.62±105.2570	356.8(210.6-567.4)	0.0001 ***	0.579
	Nerve Growth factor (NGF) (pg/ml)	383.05±169.7176	689(100-789)	0.0001 ***	0.081
Severe cases (N=30)	Hg (g/dl)	11.19±1.17	4.1(8.9-13)		
	WBCs (10^3/ul)	8.43±5.48	31.18(4.42-35.6)		
	Platelets (10^3/µl)	275.27±53.08	259(205-367)		
	CRP (mg/L)	32.7±18.631	63(6-69)		
	IL-1β (pg/ml)	2543.15±761.33	3606.3(204-3810.3)		
	IL-6 (pg/ml)	324.91±63.34	266(199.3-465.3)		
	Nerve Growth factor (NGF) (pg/ml)	461.97± 180.4	660(100- 760)		
Control (N=60)	Hg (g/dl)	12.007± 1.54	7(8.3-15.3)		
	WBCs (10^3/ul)	7.243± 1.7	6.6(4.6-11.2)		
	Platelets (10^3/µl)	285.17± 53.37	193(195-388)		
	CRP (mg/L)	6± 0.0	0(6)		
	IL-1β (pg/ml)	349.39± 152.32	790.7(100-890.7)		
	IL-6 (pg/ml)	113.78± 41.33	155.3(56.7-212)		
	Nerve Growth factor (NGF) (pg/ml)	113.54± 36.89	147.7(70.5-218.2)		

*P < 0.05 (significant) **P < 0.01 (highly significant) ***P < 0.001 (very highly significant) NS: Nonsignificant p >0.05.P between three groups, P1 between Mild -moderate vs severe cases.

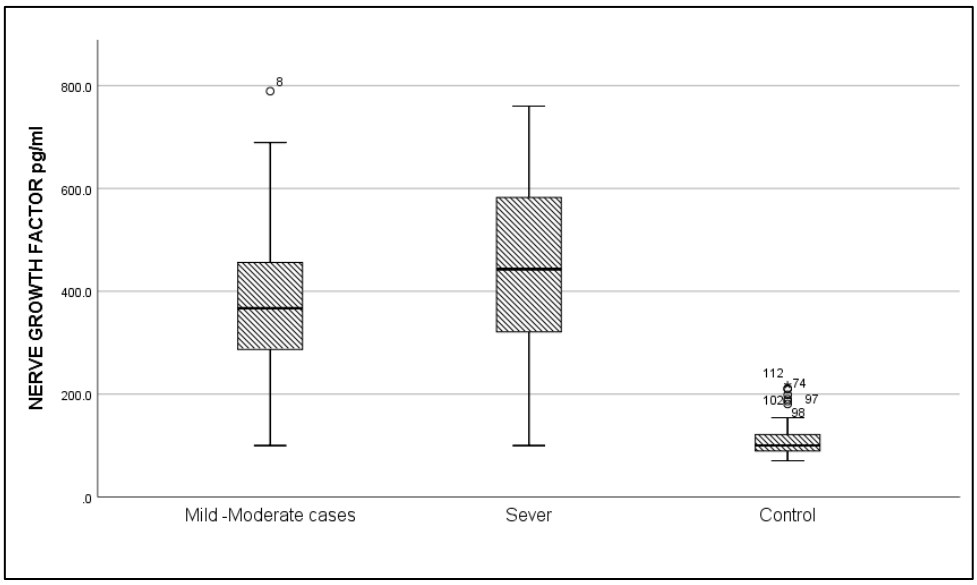

Figure (A): Box plot showing the difference of serum NGF in study groups.

Table (4): Spearman's rho correlation of age, duration of disease, BMI, IL-1β, IL-6, and CRP with serum Nerve Growth Factors

Correlation	Serum NGF
Age (n=120)	r= - 0.001 P=0.990 NS
BMI (n=120)	r=-0.041 P=0.695 NS
Duration of disease (n=120)	r= 0.740 P=0.0001***
IL-1β (n=120)	r=-0.719 P=0.0001***
IL-6 (n=120)	r=-0.769 P=0.0001***
CRP (n=120)	r=-0.735 P=0.0001***

*P < 0.05 (significant) **P < 0.01 (highly significant) ***P < 0.001 (very highly significant) NS: Nonsignificant p>0.05.0.2-0.4 positive mild correlation, 0.4-0.6 positive moderate correlation, 0.6-1positive excellent correlation.

Table (4) showed that there were positive excellent correlations between serum NGF and the following: Disease duration, IL-1β, IL-6, and CRP.

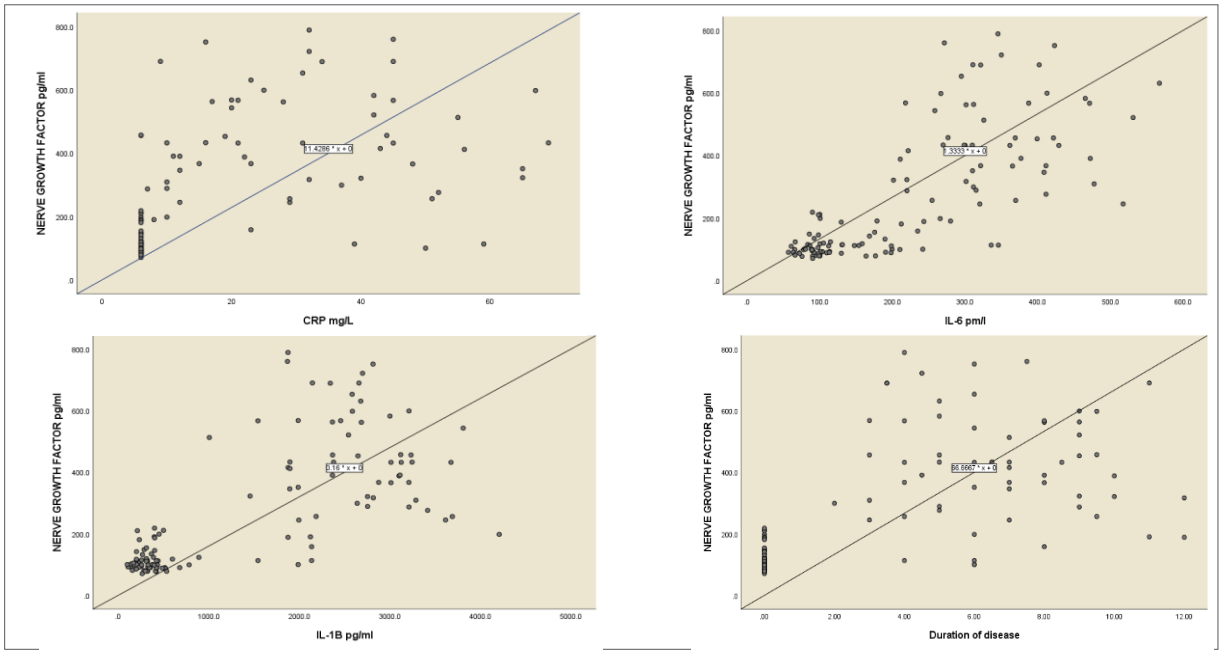

Fig (B) Correlation graph between serum NGF and Disease duration, IL-1β, IL-6, and CRP in all study group

Table (5) Distribution of genotypes, alleles of *BDNF* gene polymorphism in Cases and healthy control.

BDNF genotyping		Mild- Moderate cases N=30	Severe cases N=30	Controls N=60	Statistical Analysis
Genotypes	GG	15 (50%)	16 (53.33%)	35 (58.33%)	$p = 0.891$ Fisher's exact test
	GA	13 (43.3%)	13 (43.33%)	23(38.33%)	
	AA	2 (6.7%)	1 (3.33%)	2 (3.33%)	
Alleles	A	17 (14.16%)	15 (12.5%)	27(22.50%)	$p = 0.712$; $\chi^2 = 0.742$
	G	43 (35.83%)	45 (37.5%)	93(77.5%)	

*P < 0.05 (significant) **P < 0.01 (highly significant) ***P < 0.001 (very highly significant), NS: Nonsignificant p >0.05.

Table (6) Univariate analysis of factors predicting risk factors for allergic rhinitis

Parameters	OR (95% Confidence interval)	sig
Age	1.002 (0.969-1.036)	0.904
BMI	1.028 (0.911-1.160)	0.654 NS
Duration of the disease	0.0001 (0.0001-0.0001)	0.991 NS
Skin prick test Positive Negative	 1 5957063609.628 (0.0001-0.0001)	0.997 NS
NGF	0.973 (0.963-0.984)	0.0001***
Serum IL-1	0.995(0.992-0.997)	0.0001***
Serum IL-6	0.875(0.785-0.977)	0.017*
Serum CRP	0.0001 (0.0001-0.0001)	0.970NS
Genotypes of BDNF GG GA AA	 1 0.722 (0.344-1.516) 0.59 (0.093-3.77)	0.633 NS

Table (7) Multivariate analysis of factors predicting risk factors for allergic rhinitis

Parameters	OR (95% Confidence interval)	sig
Serum NGF	1.055(0.902-1.233)	0.503
Serum IL-1	0.983(0.944-1.023)	0.398
Serum IL-6	0.746(0.327-1.706)	0.488 NS

Table (8): Sensitivity, specificity, and accuracy rate of serum Nerve Growth Factor as predictors of allergic rhinitis:

Variable	Cut off point	AUC (Area under the curve)	P-value	Sensitivity (%)	Specificity (%)	PPV (%) (Positive predictive value)	NPV (%) (Negative predictive value)	Accuracy rate
Serum BDNF (pg/ml)	< 214.6	0.959	0.000***	85.2%	98.3%	98.1%	86.6%	91.2%

* P < 0.05 (significant), **P < 0.01 (highly significant), ***P < 0.001 (very highly significant), NS:Nonsignificant P > 0.05.

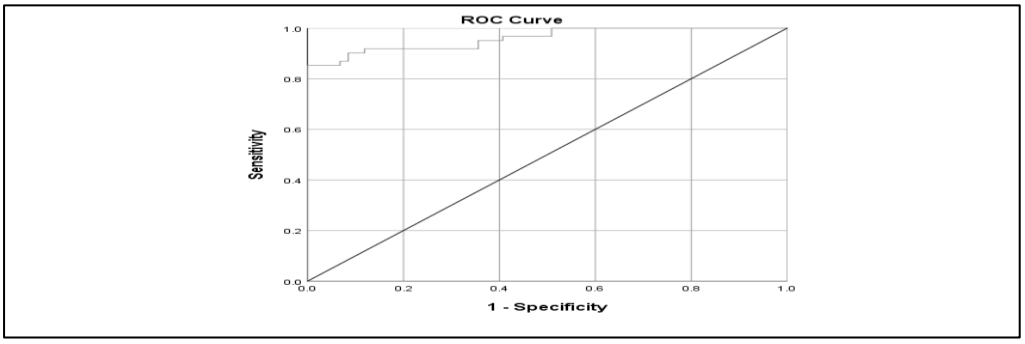

Fig (C) Receiver Operator Characteristic (ROC) Curve ofserum NGF.

Table (9) Comparison between BDNF genotype and nerve growth factor in study groups

Parameter	GG N=66	GA N=49	AA N=5	p-value
Nerve growth Factor Mean ± SD Median (range)	 260.6± 202.16 712 (77-789)	 269.24±196.10 619.5(70.5-690)	 354.2± 261.36 633.8(117.6-751.4)	0.439NS

4. Discussion

As per the current research, this is the first study to examine the nerve growth factor blood level and BDNF rs6265 polymorphisms in adult patients with allergic rhinitis and healthy control subjects from the same ethnic group in Upper Egypt. About 10–40% of people have rhinitis, one of the most common chronic inflammatory illnesses [1]. It involves multiple etiological agents and can be induced by a variety of methods. Non-infectious rhinitis has historically been categorized as allergic or non-allergic based on its etiology [35]. The frequency of AR is 3.6% in Egypt [36]. The missense mutation known as "Val66Met," or polymorphism rs6265, changes the amino acid at position 66 in the protein from valine to methionine.

It has been discovered that BDNF genetic variations raise the likelihood of moderate-to-severe AR [37].

According to our research, there was a noteworthy distinction between mild-to-moderate and severe cases of AR in terms of the skin prick test results. The size of the wheal in the skin prick test is correlated with the intensity of allergic rhinitis symptoms, hence the results were contradictory [38]. Still, numerous research findings indicated no correlation between the degree of allergic rhinitis and the skin prick test [39, 40].

According to Reshaib et al. [41], elevated levels of IL-6 are indicative of more severe allergic rhinitis, which is consistent with our findings. The present investigation and several other studies have reported higher levels of C-reactive protein (CRP) in patients diagnosed with allergic rhinitis [42, 43]. Comparing patients with allergic rhinitis to controls and different subgroups of those with the condition, another study found no discernible variation in CRP levels [44].

How severe allergic rhinitis is largely dependent on IL-1β. According to research, patients with allergic rhinitis have higher levels of IL-1β than controls do, and these levels rise as the illness gets worse [45,46, 47] Significant inflammatory biomarkers for the pathophysiology and severity.

There are several ways in which nerve growth factor (NGF) increases the intensity of allergic rhinitis. The change of allergic rhinitis to bronchial asthma can be linked to NGF, which mediates Naso-bronchial interactions. It inhibits the release of epinephrine (EPI) through the JAK1/STAT1, p38, and ERK signaling pathways and increases the expression of synaptophysin (SYP) in adrenal medullary chromaffin cells (AMCCs) [48].

Moreover, NGF can stimulate Th2-type cytokines, like IL-5 and IL-13, from group 2 innate lymphoid cells (ILC2s). These cytokines are known to worsen allergic rhinitis by inducing inflammation [49] and are a significant inflammatory biomarker for the etiology and severity of allergic rhinitis this result agrees with what we found.

Also, individuals with allergic rhinitis have nasal mucosa expression of NGF, which is increased in response to nasal allergen stimulation. It works with neurotrophin receptors expressed in nasal tissue and peripheral blood eosinophils, including p75NTR, trkA, and trkB. It also has immunomodulatory effects on eosinophils [50].

Our results are consistent with these findings, which indicate that NGF influences immunological responses and neuroendocrine pathways to play a critical role in the pathophysiology and severity of allergic rhinitis [51]. An increased risk of moderate-to-severe allergic rhinitis (AR) is linked to the BDNF rs6265 gene polymorphism [37].

Specifically, the AA genotype of rs6265 is linked to an elevated risk of moderate-to-severe AR [52], which contrasts with our observation that the most prevalent genotype in our study groups is GG. Additionally, this mutation increases the amount of BDNF protein secreted in vitro [53].

Nevertheless, given the inconsistent findings of the research that are now accessible, the relationship between the BDNF rs6265 gene polymorphism and the severity of AR is still unknown [54].

The precise contribution of the BDNF rs6265 gene polymorphism to the severity of AR requires further investigation. There was no specific relationship between BDNF genotype and NGF in allergic rhinitis in current study. Further research would be needed to explore this relationship.

5. Conclusion

The present study, we observed a connection between the BDNF rs6265 gene polymorphism and a higher occurrence of moderate-to-severe allergic rhinitis. The results revealed that the GG genotype was the most common. Additionally, the results indicated that NGF could serve as a biomarker for diagnosing allergic rhinitis, as it was found in higher concentrations in allergic rhinitis cases than in controls. Therefore, allergic rhinitis can be diagnosed using this biomarker.

6. Conflicts of interest

There are no conflicts to declare".

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8. References

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