BIOLOGY, MEDICINE, & NATURAL PRODUCT CHEMISTRY ISSN 2089-6514 (paper)
Volume 13, Number 2, October 2024 | Pages: 601-608 | DOI: 10.14421/biomedich.2024.132.601-608 ISSN 2540-9328 (online)

Comparison of Interleukin-1β-511 C/T Polymorphism in Schizophrenia of Batak and Minangkabau Ethnicities in Pematang Siantar

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Manuscript received: 25 June, 2024. Revision accepted: 04 December, 2024. Published: 31 December, 2024.

Abstract

This research is an overview of a comparative study on the Interleukin-1β-511 C/T polymorphism in individuals with schizophrenia of Batak and Minangkabau ethnicities in Pematang Siantar, Indonesia. The study aims to investigate genetic differences among these ethnic groups to better understand susceptibility to schizophrenia, which is influenced by multiple factors including genetics. The research uses a comparative categorical analytic approach with a cross-sectional method and involves DNA isolation and polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) examinations. Based on the results, significant demographic differences between the Batak and Minangkabau ethnic groups, particularly in gender distribution, age, onset of illness, duration of illness, and PANSS scores. Notably, it finds a significant difference in the occurrence of the C allele and the T allele between the two ethnic groups, with the C allele being more prevalent in the Minangkabau group and the T allele more so in the Batak group. This suggests that the IL-1β -511 C/T polymorphism may play a differential role in the susceptibility to schizophrenia among these ethnicities, indicating a potential for ethnic-specific risk factors or protective elements.

Keywords: Polymorphism; Schizophrenia; Batak; Minangkabau; ethnicities.

Abbreviations: ODGJ: mental disorder; ODS: people with schizophrenia.

INTRODUCTION

Mental disorders remain a serious issue to date, with approximately 35 million people experiencing depression and 21 million suffering from schizophrenia in 2016. There has been an increase in the number of individuals affected by mental disorders, reaching 7 per thousand households, estimating around 450 thousand individuals with severe Mental Disorders (ODGJ). According to the Indonesian Law No. 18 of 2014, ODGJ are individuals experiencing disturbances in behavior, thoughts, and feelings manifested in symptoms and/or significant behavioral changes that can cause suffering and impair their ability to function as humans (NMA, 2021).

Schizophrenia is a severe mental disorder characterized by profound disturbances in thinking processes, affecting language, perception, and self-awareness. The disease is marked by the presence of two or more symptoms, including delusions, hallucinations, disorganized speech, catatonic behavior, and negative symptoms. Schizophrenia typically affects over 21 million people worldwide and it has been estimated that approximately 7 individuals per 1000 will develop Schizophrenia during their lifetime (Orrico-Sánchez et al., 2020). Schizophrenia has been identified as one of the major health disorders worldwide. This mental disorder has significant effects on the global population, impacting over 21 million individuals, with 50% of sufferers not receiving appropriate treatment, and 90% of those untreated occurring in developing countries.

The causes of this disease are multifactorial, including genetic, neurodevelopmental, social, immunological, and degenerative factors in the central nervous system (Kahn, 2020). Genes play a significant role in schizophrenia, as schizophrenia patients exhibit rare gene variations resulting from gene segment deletions or duplications. Many genes have been selected as indicators extensively associated with schizophrenia (Damanik et al., 2020). Ven and Susser (van der Ven & Susser, 2023) and Chung et.al (Chung et al., 2023) has examined recent trends in the incidence of psychotic disorders, demographic characteristics, and comorbid psychiatric and medical conditions among six racial or ethnic groups.

The theory suggests that individuals genetically predisposed to schizophrenia when exposed to infections

during pregnancy, disrupt normal brain development, mediated through the immune response to these infections (Llorca-Bofí et al., 2024). The main candidates implicated in these infections are cytokines associated with inflammation (called proinflammatory cytokines). Cytokines are soluble polypeptide signaling proteins involved in initiating and sustaining immune responses and serve as important mediators of crosstalk between the brain and the immune system. Infections will activate proinflammatory cytokines (along with other immune factors), such as Tumor Necrosis Factor (TNF)-α, interleukin (IL)-1β, and IL-6, which play crucial roles in the early defense against infection and the initiation and/or development of inflammation (Dinarello, 2018). Furthermore, fetal exposure to proinflammatory cytokines has been associated with lesions in fetal brain white matter and developmental abnormalities, such as periventricular leukomalacia, cerebral palsy, and mental retardation (Amin et al., 2020). White matter abnormalities and premorbid motor and cognitive disturbances are often found in the course of schizophrenia, indicating that fetal exposure to inflammation can cause residual neurodevelopmental symptoms associated with schizophrenia even in the absence of genetic vulnerability to schizophrenia.

Cytokines also play a complex role in schizophrenia, either caused by or resulting from other neuropathological processes. In the brain, cytokines may be involved in regulating several neurotransmitters, such as serotonin, noradrenaline, dopamine, and glutamate (Lesh et al., 2018). Cytokine interactions with dopamine and glutamate are known to be relevant to the pathophysiology of schizophrenia. Several studies measuring levels of proinflammatory cytokines, such as interleukin (IL)-1, -6, and TNF-α, in peripheral blood or cerebrospinal fluid of schizophrenia patients have shown dysregulation of these cytokines in schizophrenia. IL-1β is an agonistic proinflammatory cytokine on the interleukin-1 receptor. IL-1β is involved in the development of the nervous system, where it can have neurotoxic effects and act as a protective agent for the nervous system (Liu et al., 2021; Reale et al., 2021).

The gene encoding interleukin-1β (IL-1β) is located on chromosome 2q14. IL-1β has also been proposed to modulate synaptic plasticity underlying learning and memory in the adult nervous system, with inhibitory or enhancing effects depending on the concentration where it is produced (Damanik et al., 2020). Some association studies in Caucasian samples suggest that genetic variability in IL-1β may increase the risk of schizophrenia, particularly the -511 C/T variant. Such as the study conducted on 356 individuals from 89 families affected by schizophrenia, found an association between the -511 T allele in the IL-1β gene promoter region with psychosis, indicating a possible role of this gene in vulnerability to schizophrenia spectrum disorders (Harrison, 2015). According to Lesh (Lesh et al., 2018) on 533 schizophrenia patients in Japan showed the first evidence that IL-1β gene polymorphism is associated with vulnerability to schizophrenia in the Japanese population.

The population of North Sumatra Province, according to ethnic groups, consists of indigenous North Sumatran residents, indigenous immigrant residents, and foreigners. The Batak ethnic is one of the indigenous North Sumatran residents. Based on studies by Tampubolon in 2015 and Puspasari in 2019 on the characteristics of inpatients with schizophrenia at Prof. Dr. M. Ildrem Medan Psychiatric Hospital, it is known that the highest proportion of schizophrenia patients belongs to the Batak ethnic. Pro-inflammatory studies related to ethnicity have also been conducted in Indonesia, especially in North Sumatra. Some studies include the levels of Tumor Necrosis Factor- α and its relationship with cognitive function in schizophrenia in the Malayan-Mongoloid ethnic group by Amin et al (Amin et al., 2020). Another study on the relationship between INF-γ+874 A/T gene polymorphism and schizophrenia in Batak ODS (people with Schizophrenia) with control groups by Damanik et al (Damanik et al., 2020) and a study by Saragih et al (Saragih et al., 2021)on the difference in the frequency of appearance of G allele and C allele variant –G174C IL-6 in the Batak ODS group with the control group.

West Sumatra ranks fourth highest in the prevalence of Schizophrenia in Indonesia. In North Sumatra, the Minangkabau ethnic is one of the indigenous immigrant residents. The Minangkabau ethnic ranks second as the largest minority ethnic group in Medan city after the Chinese ethnic group. Minangkabau society values life highly. In line with the view of life being beneficial to their families and communities, work is highly regarded. Work is a necessity for Minang society. Wealth can shield a family from shame. There are many customary events like weddings that require significant costs. Therefore, hard work is highly prioritized. This may be related to triggering factors for schizophrenia in the Minangkabau ethnic.

Individual genetic differences among different ethnic groups can determine who is susceptible to schizophrenia or not, and identifying these variations can help provide personalized care to patients. Personalized care also involves prevention and treatment for patients, based on their genetic makeup and environment. Genome-wide association studies (GWAS) have proven to be a useful technique in associating variations in the genome with specific diseases using statistical procedures (Falola et al., 2017)

The presence of schizophrenia patients is considered disruptive and even hazardous. Research on interleukin-1β –511 C/T polymorphism in individuals with schizophrenia (ODS) is still very limited, and the research has only been conducted in a few countries with varying results. Meanwhile, in Indonesia, particularly in

North Sumatra, there has been no previous research comparing interleukin-1β –511 C/T polymorphism in ODS of Batak and Minangkabau ethnicities. Based on the above background, the researcher is interested in investigating whether there are differences in interleukin-1β –511 C/T polymorphism in individuals with schizophrenia of Batak and Minangkabau ethnicities in Pematang Siantar.

MATERIALS AND METHODS

This study is comparative categorical analytic research with non-paired one-time measurement comparing the groups of individuals with schizophrenia of Batak and Minangkabau ethnicities using a cross-sectional method. The research was conducted at the Mental Health Polyclinic of dr. Djasamen Saragih Regional General Hospital in Pematang Siantar. The target population in this study is individuals with schizophrenia. The accessible population consists of patients who seek treatment at the Mental Health Polyclinic of dr. Djasamen Saragih Regional General Hospital in Pematang Siantar from December 2023 to January 2024. The research sample comprises Schizophrenia patients who seek treatment at the Mental Health Polyclinic of dr. Djasamen Saragih Regional General Hospital in Pematang Siantar from December 2023 to January 2024. The sampling method used is non-probability sampling, specifically Consecutive sampling, where all subjects who come and meet the selection criteria are included in this study until the required number of subjects is met.

Demographic Characterization

The demographic characteristics analyzed in the groups of Individuals with Schizophrenia of Batak and Minangkabau ethnicities are gender, age, age of onset, duration of illness, and total PANSS score following method by Opler et.al (Opler et al., 2024). In the healthy control group, the demographic characteristics analyzed are gender and age.

Inclusion and exclusion Criteria

The inclusion Criteria include:

- Individuals with schizophrenia of Batak ethnicity diagnosed based on PPDGJ-III criteria.
- Individuals with schizophrenia of Minangkabau ethnicity are diagnosed based on PPDGJ-III criteria.
- Having PANSS scores ranging from 80 to 120 and age between 18 to 45 years old.
- Two generations of first-degree family members are of Batak ethnicity.
- Two generations of first-degree family members are of Minangkabau ethnicity.
- Cooperative and willing to be interviewed

The exclusion criteria include:

- Having a history of other psychiatric disorders.
- Having a history of neurological diseases, endocrine disorders, and autoimmune diseases and having a history of alcohol and other addictive substance use (except nicotine and caffeine).

Sample

The sampling technique used in this study is consecutive sampling, where all subjects who come and meet the selection criteria are included in the study until the required number of subjects is met. Sample size calculation for comparing the interleukin-1β – 511C/T polymorphism in Batak and Minangkabau ethnicities with a non-paired categorical-comparative 2x2 table. Sample size calculation is determined by establishing the effect size (P1-P2). This study requires 97 individuals with Schizophrenia Batak and Minangkabau ethnicity respectively. data collection was carried out by the researcher and assisted by the team, preceded by screening using inclusion and exclusion criteria. Individuals who met the inclusion criteria and did not have exclusion criteria were explained about the aims and objectives of the research. Then the selected subjects were asked to sign an agreement/informed consent to take part in the research. Subjects diagnosed with schizophrenia using the PPDGJ III diagnostic criteria were assessed using the PANSS interview where a PANSS score between 80-120 indicated that people with schizophrenia were cooperative enough to take part in the research. After that, blood is drawn from the subject and then laboratory examinations, statistical analysis, and preparation of results reports are carried out.

DNA Isolation Technique

Blood samples were taken in the amount of 5 cc from the anterior cubital vein. The blood will be put in vacutaner which contains ethylenediamine tetraacetic acid (EDTA) and stored at 4-8^{o}C until DNA isolation is carried out. The method used for DNA isolation is salting out. A total of 2 ml of a peripheral blood sample containing EDTA was included falcon tube, then 6 ml red blood cells (RBC) lysis solution (blood ratio: RBC = 1:3), RBC lysis solution containing 199mM EDTA, 100 mM KHCO3, and 1.45 NH4Cl. The tube was homogenized by turning it upside down and incubated at room temperature 27^{o}C for 10 minutes. Next, the tube was inverted again before centrifugation was carried out at a speed of 1500 rotations per minute(rpm) for 10 minutes at room temperature to obtain a white supernatant and pellet (precipitate), then supernatant thrown away. The above procedure is repeated 3 times until you get a pellet that is free from red blood cells.

After that, the formed pellets are added to the cell lysis solution (CLS) containing 10 mM Tris-HCl, 0.25 mM EDTA, and 20% SDS. 1.334 ml of CLS was added and carried out up and down with a transfer pipette slowly until the mixture became homogeneous. The mixture was incubated deeply in water bath at a temperature of 37^{o}C for 30-60 minutes. Next, the mixture

is added protein precipitation (PP) (5 M ammonium acetate) as much as 867 µl and carried out vortex enough. The mixture was centrifuged again at 3000 rpm for 15 minutes at a temperature 4ºC is thus obtained supernatant and brown pellets on the tube walls. The supernatant was transferred into a new tube containing 767 µl of cold isopropanol solution, then the tube was turned back and forth 25-30 times until DNA strands were seen floating in the isopropanol solution. Next, the DNA in isopropanol was incubated overnight at 20ºC. After overnight incubation, centrifugation was carried out at 3000 rpm for 5 minutes. The supernatant is then thrown away. Next, 867 µl of cold 70% ethanol was added for washing and carried out inverted, then centrifuged again at 3000 rpm for 5 minutes at a temperature 4ºC until obtained supernatant and DNA pellets. DNA pellets were dried for 2 hours at room temperature or 1 hour at 37°C. Drying is carried out by placing the tube in an inclined position on the base tissue dry. The DNA pellet was dissolved by adding 300 µl of TE solution (10mM Tris-HCl, 0.25 EDTA) and then incubated for 2 hours at 37^0C inside the water bath. Next, the DNA was transferred to a 1.5 ml Eppendorf tube and stored at -20^0C, and the DNA was ready for analysis for the next stage.

Detection of Interleukin -1β -511 C/T Polymorphism

Genomic DNA is extracted from samples that have undergone freezing using standard methods. The single base polymorphism at position -511 in the interleukin-1β promoter region was read by PCR. For each allele, the PCR reaction was carried out on template DNA with a pair of oligonucleotide primer, namely 5' TGGCATTGATCTGGTTCATC 3' (forward) and 5' GTTTAGGAATCTTCCCACTT 3' (reverse). PCR cycling conditions were denaturation at 94°C for 5 minutes followed by 30 cycles at 94°C for 30 seconds, 55°C for 30 seconds, and 72°C for 7 minutes. The PCR product was stored at 37°C for 1 night with 10 units of Ava 1 restriction enzyme for IL-1β and will produce products at 190 bp + 114 bp (CC) and 304 bp (TT).

Data analysis

Data normality tests will be carried out using the Kolmogorov-Smirnov test for a sample size of more than 50. If both variables are normally distributed and the X2 condition is met, a Chi-Square test will be carried out. After that, the Odds Ratio (OR) value will be calculated. An OR value of more than one is a risk factor, an OR equal to one or above one indicates that the independent variable is not a risk factor, and an OR of less than one indicates that the independent variable is a protective factor. Data processing and analysis is carried out with the Statistical Software SPSS ver 21.

RESULTS AND DISCUSSION

Gender is a categorical variable and will be presented in a frequency distribution. This variable with the phenotype of the ODS group and the Control group of the Batak ethnic and the Minangkabau ethnic was tested using an unpaired comparative hypothesis 2x2 table with conditions chi-square fulfilled so the test used chi-square.

Table 1 shows the demographic characteristics of the ODS (people with schizophrenia) group of the Batak and Minangkabau ethnicities. The Batak ODS group with the most subjects was 63 women (52.5%) and 57 men (47.5%). The median age was 31 years with a minimum value of 19 years and a maximum value of 44. The median of disease onset was 30 years with a minimum value of 17 years and a maximum value of 44 years. The median duration of illness was 10 years with a minimum value of 2 years and a maximum value of 20 years. The median PANNS score is 101 with a minimum score of 82 and a maximum score of 120.

Table 1. Demographic Characteristics of Batak ODS and Minangkabau ODS.

Gender	Ethnic Group		
	Batak (n=120)	**Minangkabau (n=120)**	**P value**
Man	57 (47.5)	63(52.5)	
Woman	63 (52.5)	57(47.5)	0.001*
Age (Years)	31 (19-41)	30(19-44)	
Onset	30 (17-44)	29.5(19-44)	
Long time of illness	10(2-20)	10(1-20)	<0.001**
Total Score Panss	101 (82-120)	99.5(86-110)	

*Chi-square with continuity correction
**Mann_Whitney U
ODS = people with Schizophrenia

In the ODS group of the Minangkabau ethnicities, the majority of subjects were 63 men (52.5%) and 57 women (47.5%). The median age is 30 years with a minimum value of 19 years and a maximum value of 44 years. The median disease onset was 29.5 years with a minimum value of 19 years and a maximum value of 44 years. The median duration of illness was 10 years with a minimum value of 2 years and a maximum value of 20 years. And

the median PANNS score is 99.5 with a minimum score of 86 and a maximum score of 110. The results of the analysis show that there are significant differences between the two ethnic groups in terms of gender and value =0,000. There were also significant differences in age, onset, duration of illness, and total PANSS scores in the two ethnic groups value =0,000.

In terms of demographic characteristics, the largest sample was women in the ODS group of the Batak ethnic and men in the ODS group of the Minangkabau ethnic, namely 63 people. There were significant differences in gender between the two groups, and significant differences were found in age, onset, duration of illness, and total PANSS scores in the two ethnic ODS groups. The median age in the ODS group of the Batak ethnic was 31 years, with a minimum age of 19 years, and a maximum value of 44 years, while the age in the ODS group of the Minangkabau ethnic was found to be a median of 30 years, with a minimum of 19 years and a maximum value of 44 years. Charlson et al. (Charlson et al., 2018) stated that no gender differences were observed in global prevalence and around 70.8% of schizophrenia disorders were found in those aged 25-54 years with the highest prevalence in their 40s and decreasing in older age groups.

The median onset in the ODS group of the Batak ethnic is 30 years, with a minimum value of 17 years and a maximum of 44 years. In the ODS group of the Minangkabau ethnic, the median onset was 29 years with a minimum of 19 years and a maximum of 44 years. The median duration of illness in the ODS group of the Batak ethnic is 10 years with a minimum of 2 years and a maximum of 20 years, and in the ODS group of the Minangkabau ethnic the median duration of illness is 10 years with a minimum of 1 year and a maximum of 20 years. Schizophrenia can occur in all age groups, where schizophrenia disorder is characterized by a pro-moral stage that lasts for several years and causes social consequences. The onset of schizophrenia shows a sharp increase reaching a peak at the age of 15 to 25 years in men, while in women the onset is at the age of 15 to 30 years at the first peak and the second peak at the age of 44 to 49 years (Charlson et.al., 2018). The most common initial onset of this disease is 15-30 years of age and is a chronic disease that causes disruption to the patient and family and has a major social and economic impact.

The median total PANSS score in the ODS group of the Batak ethnic is 101 with a minimum score of 82 and a maximum score of 120, while in the ODS group of the Minangkabau ethnic the median PANSS score is 99.5 with a minimum score of 86 and a maximum of 110. The total PANSS score was divided into 3 parts, namely low (<75), medium (75-94), and high (>95), in patients with a medium total PANSS score, compared to a low PANSS total score (Purnama et al., 2023). Schizophrenia is not a common mental disorder, affecting 20 million people worldwide. Schizophrenia is a complex mental disorder that generally occurs earlier in men and is influenced by genes with mild to moderate effects and non-genetic risks such as environmental and psychological factors that change the chemical structure of the brain (Nasution et al., 2023).

Differences between the IL-1β -511 C/T Variant Allele in ODS of the Batak and the Minangkabau ethnicities

IL-1β -511 C/T variant, the allele consists of two, namely the C allele and the T allele. The allele variable is a categorical variable, where the data is presented in a frequency distribution. The relationship between the IL-1β -511 C/T gene allele and schizophrenia is expressed odds ratio/OR obtained from the risk estimate chi-square test (Table 2). Batak ODS group, the frequency of occurrence of the C allele is 98 times (40.8%) and the T allele is 142 times (59.2%). For the ODS group of the Minangkabau ethnic, the frequency of occurrence of the C allele was 127 times (52.9%) and the T allele 113 times (47.1%). The p-value was found to be 0.008 with the OR being 0.614 with a 95% confidence interval between 0.428 to 0.881 (Table 2).

Table 2. Differences between the IL-1β -511 C/T Variant Allele in ODS of the Batak and the Minangkabau Ethnicities.

Allele	Ethnic Group		P value	OR (95% CI)
	Batak (n=120)	Minangkabau (n=120)		
C	98(40.8)	127(52.9)	0.008	0.614(0.428-0.881)
T	142(59.2)	113(47.1)		

*ODS= people with Schizophrenia

Based on the research results, it was found that the frequency of occurrence of the C allele in the ODS group of the Batak ethnic was 40.8% and the T allele was 59.2%. Meanwhile, in the ODS group of the Minagkabau ethnic, the frequency of occurrence of the C allele was 52.9% and the T allele was 47.1%. The occurrence of the C allele in the ODS group of the Minangkabau ethnic is 29 times more than in the ODS group of the Batak ethnic. Furthermore, the occurrence of the T allele in the ODS group of the Batak ethnic was 29 times more than in the ODS group of the Minangkabau ethnic. Furthermore, based on chi-square analysis, the p-value = 0.008 was obtained, indicating that there was an insignificant relationship between the allele and

schizophrenia. The OR value obtained was 0.614 with a 95% CI between 0.428-0.881. This shows that in the ODS of the Batak and Minangkabau ethnic, the T allele was found to have a 0.008 times probability of experiencing schizophrenia compared to individuals who had the C allele. The OR value was below 1, so the T allele was a protective factor that caused schizophrenia. Zanardini et.al's research stated that the proportion of the C allele appeared significantly more frequently in the ODS group than in healthy controls with a P value of 0.047. Furthermore, Sasayama et.al stated that male ODS was at an average age of 43.3 years and female ODS was at an average age of 44.8 years, finding that the proportion of the C allele was more frequent in the healthy control ODS group. The significant difference in the IL-1β -511 C/T polymorphism can be a risk factor for schizophrenia in the Minangkabau community compared to the Batak community.

Risk factors for schizophrenia start from developmental processes, neurodegenerative and neurotransmitter disorders to autoimmune processes or infections (Dinarello, 2018). Cytokines and other growth factors are associated with the genesis of schizophrenia or pathology and it is found that the central nervous system (CNS) contains and utilizes the receptors. Cytokines are polypeptides and protein monomers or glycolized polymers that are the result of many genes. Genetic variations in IL-1, especially the -511 C/T mutation (rs 16944) can increase the risk of schizophrenia and the IL-1β -511 C/T polymorphism is a factor in schizophrenia (Damanik et al., 2020). Immune dysfunction in schizophrenia is related to the pathomechanism of the disorder so increased levels of IL-1β and IL-6 are associated with schizophrenic behavior (Jordan et al., 2021). Increased levels of IL-1β and IL-6 are associated with chronic schizophrenia in old age. However, confounding factors are age, gender, smoking, and antipsychotic drugs have not been shown to influence serum cytokines.

Differences between the IL-1β -511 C/T Variant Genotype in ODS of the Batak Ethnic and the Minangkabau Ethnic

The IL-1β -511 C/T variant is a combination of C and T alleles, consisting of CC, CT and TT genotypes. The genotype variable is a categorical variable, the data is presented in the form of a frequency distribution which will be tested using logistic regression, choosing the TT genotype as a comparison (Table 3).

Table 3. Differences Between IL-1β -511 C/T Variant Genotypes in ODS of the Batak and the Minangkabau Ethnicities.

Genotype	Ethnic Group		P value	OR (95% CI)
	Batak (n=120)	Minangkabau (n=120)		
CT	60(50)	56(46.7)	0.063	0.865 (0.479-1.562)
CC	21(17.5)	37(30.8)	0.080	0.534 (0.264-1.079)
TT	39(32.5)	27(22.5)	comparison	

*ODS= people with Schizophrenia

In the Batak ODS group, the CT genotype frequency was 60 (50%), CC was 21 (17.5%) and TT was 39 (32.5%). For the ODS group of the Minangkabau ethnic, the frequency of the CT genotype was 56 (46.7%), CC was 37 (30.8%), and TT was 27 (22.5%). From the results of analysis using logistic regression, the p-value for genotype (CT vs TT) was 0.063. OR = 0.865 with 95% CI between 0.479 - 1.562. The p-value for genotype (CCvs TT) was 0.080. OR = 0.534 with a 95% CI between 0.264 - 1,079. This means that there is no significant difference between the genotypes (CT vs TT) and (CT vs TT) and the incidence of Schizophrenia. In this study, the frequency of appearance of the CT genotype in the ODS group of the Batak ethnic was 50%, the CC genotype was 17.5% and the TT genotype was 32.5%. Meanwhile, in the ODS group of the Minangkabau ethnic, the occurrence of the CT genotype was 46.7%, the CC genotype was 30.8% and the TT genotype was 22.5%. The frequency of appearance of the CT genotype in the ODS group of the Minangkabau ethnic is lower than that of the ODS group of the Batak ethnic, however, the frequency of occurrence of the CC genotype in the ODS group of the Minangkabau ethnic is higher than that of the ODS group of the Batak ethnic. Based on the results of logistic regression analysis, the chi-square value for genotype (CC vs TT) was 0.063 with OR = 0.865 with a 95% CI between 0.479 - 1,562. The p-value for genotype (CC vs TT) was 0.080. OR = 0.534 with a 95% CI between 0.264 - 1.079. This means that there is no significant difference between genotype (CT vs TT) and (CT vs TT) and the incidence of schizophrenia.

Based on Hazumi et.al (Hazumi et al., 2022) stated that all subjects were Japanese individuals who had no biological relationship submitted in their self-reports, and were recruited from the outpatient clinic of the National Center of Neurology and Psychiatry Hospital Tokyo, Japan. As a result, the proportion of the CC genotype in the ODS group was 21%, CT 48% and TT 31%. The non-ODS group with the proportion of the CC genotype was 17%, CT 47%, and TT 36%. The study stated that the p-value = 0.35 for genotype distribution, meaning

that the IL-1β-511 C/T genotype was not related to the incidence of schizophrenia, but the incidence of schizophrenia was associated with the C allele only. Furthermore, the groups of patients were found bifrontal temporal gray matter volume deficit, and white matter tissue deficits are common in carriers of the 2 genotype alleles, namely TT or CT. In contrast, interleukin-1β polymorphisms had no effect on brain morphology in healthy subjects (Isaacs et al., 2022; Kraus et al., 2024)(Chrusciel et al., 2022).

According to the Hardy-Weinberg formula, if the allele frequencies A and a are p and q, then ;

$(p+q) = 1$, it's mean $(p+q)^2 = 1$, In this formula, p^2, corresponds to the frequency of the homozygous AA genotype, q^2 to aa, and 2pq to Aa. This study has been tested according to the steps above using the help of online software. Table 4 shows the result of HWE values in ODS of Batak and Minangkabau with the IL-1β -511 C/T.

Table 4. HWE values in People with Schizophrenia Batak and Minangkabau with the IL-1β -511 C/T.

	P value	
	Batak	**Minangkabau**
Chi square If p<0.05-not consistent with HWE	0.054	0.080

From the results of the analysis using the HWE formula assisted by online software, the p-value obtained for the ODS of the Batak ethnic was 0.054 and for the ODS of the Minangkabau ethnicity it was 0.080, so it can be stated that the ODS of the Batak ethnic and the ODS of the Minangkabau ethnic are in accordance with the HWE rules (Hardy-Wienberg Equilibrium). In most population genetic estimates, HWE is assumed so that alleles determine the probability of the genotype and are not disturbed by anything. If the observed frequencies do not show a significant difference from the expected frequencies, the population is said to be in Hardy-Weinberg equilibrium. If not, there is a violation of the following formula assumptions and the population is not in HWE. If the assumptions are not met, then the estimate is inaccurate. When HWE is assumed to mean that the genotype probability is determined by the allele frequency, that is, there is no distortion of the transmission ratio, or selection of the genotype and if HWE is violated then statistical methods using allele frequencies may not be valid. The tests carried out observed that the chi square value using the HWE formula was found for the IL-1β -511 C/T variant polymorphism for the ODS of the Batak ethnic of 0.054 and the ODS of the Minangkabau ethnic of 0.080 and there were no individuals from each subject less than 5 so that it could be declared tribal ODS. Batak and ODS Minangkabau ethnicities are following the rules of HWE (Hardy-Wienberg Equilibrium). This shows that the allele frequency remains constant from generation to generation, that hereditary mechanisms do not change the allele frequency (Nasution et al., 2023; Purnama et al., 2023).

CONCLUSIONS

Based on the results of the analysis, it show that there are significant differences between the two ethnic groups in terms of gender and value = 0,000. There were also significant differences in age, onset, duration of illness, and total PANSS scores in the two ethnic groups p-value =0,000. According to chi-square analysis, the value of p = 0.008 is obtained, indicating that there is an insignificant relationship between the allele and schizophrenia. This shows that in the ODS of the Batak and Minangkabau ethnicities, the T allele was found to have a 0.008 times probability of experiencing schizophrenia compared to individuals who had the C allele. The OR value was below 1, so the T allele was a protective factor that caused schizophrenia. Logistic regression analysis, the chi-square value for genotype (CT vs TT) was 0.063 with OR = 0.865 with a 95% CI between 0.479 - 1.562. The p-value for genotype (CC vs TT) was 0.080. OR = 0.534 with a 95% CI between 0.264 - 1.079. This means that there is no significant difference between genotype (CT vs TT) and (CC vs TT) and the incidence of schizophrenia.

Acknowledgements: We are very grateful to the Mental Health Polyclinic of dr. Djasamen Saragih Regional General Hospital in Pematang Siantar for their assistance in this research.

Authors' Contributions: Raysha, Edy Fahcrial & I Nyoman Ehrich Lister designed the study. Raysha & Bayu Ariatama carried out the laboratory work. Raysha analyzed the data. Raysha & Edy Fachrial wrote the manuscript. All authors read and approved the final version of the manuscript.

Competing Interests: There are no competing interests.

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