International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 1 Jan - Feb 2025, pp: 851-858 www.ijprajournal.com ISSN: 2456-4494

Detection of genetic polymorphism and allele distribution pattern of human IL-1 beta gene through PCR and RFLP in Vindhya region of Madhya Pradesh

Pramod Kumar Prajapati[1#], Dr.Bal Mahendra Prajapati[1], Prof.Suman Singh[1]
1-Department Of Zoology, Government Model Science College Rewa Madhya Pradesh 486001
#-Corresponding Author,Department Of Zoology,Government Model Science College Rewa Madhya Pradesh 486001,

Date of Submission: 20-01-2025 Date of Acceptance: 30-01-2025

ABSTRACT
One of the main characteristics of rheumatoid arthritis (RA), a chronic inflammatory disease that can destroy joints and have systemic effects, is inflammation of the synovium, or joint lining**.** Both genetic and environmental factors have a major impact on the pathophysiology and progression of RA. This study links IL-1 beta genes to the immune system and inflammatory response to elucidate the genetic component of RA. Rheumatoid arthritis (RA), a chronic inflammatory disease that damages joints and enlarges organs, is characterized by inflammation of the synovium, the layer covering the joint surfaces.The development and course of RA are known to be influenced by hereditary and environmental factors. The current study has also discovered genetic variations of the IL-1 beta gene using the PCR and RFLP techniques.The present study investigated the association between rheumatoid arthritis risk and interleukin-1 beta gene polymorphism. This study used polymerase chain reaction (PCR) and restriction fragment length polymorphism (RFLP) analysis to investigate the association between interleukin-1 beta gene polymorphism and rheumatoid arthritis risk in the Vindhyan region of Madhya Pradesh, India. the effects of restriction fragment length polymorphism (RFLP) analysis and polymerase chain reaction (PCR) on these two variables in the Vindhyan region of Madhya Pradesh, India.Two hundred and sixty-seven healthy subjects and 221 RA subjects were collected from hospitals in the Vindhyan region. The polymerase chain reaction (PCR) was carried out in the specific area of the interleukin-1 betas genes that contained polymorphism sites following the purification of genomic deoxyribonucleic acid from peripheral blood collection tubes.
NcoI restriction enzymes were used in RFLP to characterize the PCR products. To evaluate the relationship between genotypes and susceptibility to rheumatoid arthritis, genotype frequencies were ascertained and statistical analysis was conducted.Preliminaryanalysisrevealedthespecificall elesareresponsibleforthe onset of rheumatoid arthritis (RA) in a given in the Vindhyan region further it has been confirmed that the different alleles coexist in the population of the Vindhyan region responsible for (RA).
Keyword: Rheumatoid arthritis(RA),IL-1β,Allele,Polymorphism.

I. INTRODUCTION

The genetic diversity within the human population is essential for our ability to adapt to different environments and resist diseases. One aspect of this genetic diversity is genetic polymorphism, which refers to the occurrence of multiple alleles or variations within a particular gene in a population. These polymorphisms can significantly affect a person's overall health, reaction to therapies, and disease susceptibility. Interleukin-1β (IL-1β), one of the most well-known cytokines, is a pro-inflammatory cytokine essential to the immune system's reaction to infection and damage (Enayati, Seifirad, et al. 2015).

Genetic variations in these genes can affect a person's immune response and risk of infections, autoimmune diseases, chronic heart disease, and inflammatory problems, among other diseases (Millar, Singer et al. 1989, Levine, Kalman, et al. 1990). The cytokine IL-1β can be influenced by SNPs, which are distinct genetic variations within the IL1β gene. Two of them are located in the area that initiates a gene positioned at -511 (C-T) and -31 (T-C) (Iglesias Linares, Yanez-Vico et al. 2013). Two of them are located in the area that initiates a gene at positions -511 (C-T) and -31 (T-C), according to Iglesias Linares, Yanez-Vico et al. (2013). These genetic differences have been linked to a higher chance of getting certain diseases that cause inflammation and

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 1 Jan - Feb 2025, pp: 851-858 www.ijprajournal.com ISSN: 2456-4494

stomach cancer. (Zienolddiny, Ryberg et al. 2004).We genotyped the+3953exon5geneinIL-1 beta genepolymorphism from the blood of patients in Vindhya, Madhya Pradesh, and healthy people from the general Vindhya population.

Chronic inflammation and joint degradation are hallmarks of rheumatoid arthritis (RA), a complicated autoimmune illness (Anaya, Rojas-Villarraga, et al. 2013). Although the precise etiology of RA is still unknown, it is generally acknowledged that environmental and genetic factors contribute significantly to its development.Genetic contributions have been the subject of much investigation, and because they correlated with inflammation and immunity, polymorphisms in immune system genes, such as Interleukin-1 β (IL-1 β), have been examined. This study aims to investigate IL-1β gene polymorphisms and their potential association with RA pathogenesis. ApplyingPolymerase Chain Reaction (PCR) and Restriction Fragment Length Polymorphism (RFLP) techniques, the current study attempts to advance knowledge of the impact of certain genetic variants in these genes on RA risk and severity in humans.

The genetic variety of the human population greatly influences its capacity for adaptation and resistance to environmental and viral challenges. One of the most significant characteristics of genetic diversity is genetic polymorphism. The presence of several alleles or genetic variations of a particular gene within a population is known as genetic polymorphism. These differences often have serious consequences for the health of the individual, treatment responses, disease/peril exposure, and general immune activity. The genetic variety of the human population greatly influences its capacity for adaptation and resistance to environmental and viral challenges. One of the most significant characteristics of genetic diversity is genetic polymorphism. The presence of several alleles or genetic variations of a particular gene within a population is known as genetic polymorphism.As a basic component of the immune system, IL-1β controls host defense and inflammation. Changes in cytokine production and function are known to be associated with genetic variation, particularly single nucleotide polymorphisms (SNPs) of the IL1β gene. In particular, SNPs within positions -511 (C-T) and -31 (T-C) of the IL1β gene promoter region have drawn particular interest for their ability to regulate gene expression (Iglesias Linares, Yanez-Vico et al., 2013). These polymorphisms modulate the transcriptional regulation of the IL1β gene and subsequently produce differences in cytokine levels that could alter the immune responses and disease risk.A higher risk of stomach and inflammatory malignancies, for instance, has been linked to polymorphism at position -511 (C-T) (Zienolddiny, Ryberg, et al, 2004).

Research on IL1β polymorphisms in rheumatoid arthritis (RA), a chronic autoimmune inflammatory disease that results in tissue damage and systemic problems in addition to continuous inflammation of the synovial joints, is particularly interesting. It is well acknowledged that environmental factors and genetic predisposition play a part in the development of RA, even though the precise etiology of the disease is unknown (Anaya, Rojas-Villarraga et al., 2013). Due to their significant role in inflammatory cascades, immune-associated genes, like IL1β, have also been implicated in the pathophysiology of RA by genetic analysis. According to research to date, IL1β gene polymorphisms affect the synthesis of pro-inflammatory cytokines, which in turn control how severe inflammatory reactions are in RA.It has been demonstrated that SNPs in the IL1β promoter region, specifically those -511-511 C-T) and -31-31 T-C), alter cytokine expression levels, which can either exacerbate or lessen inflammatory processes linked to RA.In addition, the +3953 polymorphism in exon 5 of the IL1β gene has been investigated concerning the possibility of being a risk factor for disease severity and disease progress.

We genotyped the IL1β +3953 polymorphism in both RA patients and healthy controls in the Vindhya region of Madhya Pradesh. Two established methods for detecting genetic polymorphisms have served as the foundation for the analysis: Polymerase Chain Reaction (PCR) and Restriction Fragment Length Polymorphism (RFLP). By comparing IL1β polymorphism frequencies between RA patients and healthy individuals, this study aims to shed light on the potential genetic basis of RA in the Vindhya population.

The findings in this paper have broader implications in the definition of the genetic architecture of autoimmune diseases and in the development of personalized therapies. Recognition of established genetic polymorphisms of genetic susceptibility to RA, for example, IL1β SNPs, might allow stratification of patients based on their genetic disposition and targeted treatment strategies. Additionally, this study contributes to the accumulating evidence of cytokine gene polymorphisms and their association with

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 1 Jan - Feb 2025, pp: 851-858 www.ijprajournal.com ISSN: 2456-4494

inflammation and immune regulation, enriching the understanding of the genes and diseases involved in inflammation and immune regulation.In conclusion, Polymorphisms in the IL1β gene, including those located at locations -511, -31, and +3953, are important because they affect immune responses and the likelihood of developing inflammatory illnesses like RA.

Knowing which genetic variations exist and what effect they have is critical for the development of personalized medicine and for better treating patients with autoimmune diseases.

II. METHODS AND MATERIALS

Sample size.

They studied 221 RA patients (112 men and 109 women) and 267 Vindhyan Region healthy controls (94 men and 173 women) and concluded that 18.8% of RA patients have AS, while the prevalence in the control group is 20.8%. Rheumatologists made RA diagnoses from patients of Rewa, Satna, Sidhi, Umaria, and other Vindhyan Region areas. According to the 2010 American College of Rheumatology/European League Against Rheumatism (ACR/EULAR) categorization criteria for RA, the patient was placed into one of two RA groups (Aletaha et al., 2010; Giri et al., 2021; Thanapati et al., 2017).

Patients with RA were defined as having anti-cyclic citrullinated peptide (anti-CCP) IgG antibodies, anti-nuclear antibody (ANA) positivity, and adherence to rheumatoid factor (RF).

Control patients consisted of RF-negative, anti-CCP antibody-negative, and ANA-negative healthy volunteers with no RA symptoms or features. Healthy individuals without RA symptoms or indicators made up the control group; their RF, anti-CCP antibody, and ANA test results were negative. They contrasted 221 RA patients and 267 subjects from the Vindhyan Region. Rheumatologists categorized the RA patients of Rewa, Satna, Sidhi, Umaria, and other areas of the Vindhyan Region area. According to the 2010 American College of Rheumatology/European League Against Rheumatism (ACR/EULAR) criteria for RA, the patient was assigned to the RA group (Aletaha et al., 2010; Giri et al., 2021; Thanapati et al., 2017).Rheumatoid factor (RF) and antinuclear antibody (ANA) positivity, and anti-cyclic citrullinated peptide (anti-CCP) IgG antibodies, were adherence criteria for RA patients. The control group consisted of RF-negative anti-CCP antibody-negative ANA-negative healthy volunteers, without RA signs or symptoms.

DNA isolation and blood collection

The collection and processing of genetic material from human subjects is an essential element in exploiting genetic polymorphisms that are implicated in diseases like rheumatoid arthritis (RA).In this study, two milliliters (mL) of whole blood specimenswere gathered from residents of Madhya Pradesh's Vindhyan region who had RA as well as healthy controls.To prevent blood coagulation and preserve the integrity of the nucleic acid for use in the follow-up, the specimens were drawn into K3 EDTA-coated vacutainers. EDTA is a chelating agent that binds calcium ions, inhibiting clotting cascades and maintaining the stability of the sample during transport and storage (Reith et al., 2016).The HiPurA Blood Genomic DNA Isolation Kit (Himedia, India) was used to separate genomic DNA (gDNA) from patient blood samples.This commercial kit has the exclusive function of high-selectively isolating pure high-purity DNA from whole blood in a tiny volume with minimal contamination and a high release of nucleic acid.The step consists of cell Lysis, Proteins and contaminant removal, DNA precipitation, and ensuing elution in the appropriate buffer. The selection of this kit was based on the kit's reliability, its ease of use, and its ability to produce usable DNA for sensitive downstream molecular analyses, including Polymerase Chain Reaction (PCR) and Restriction Fragment Length Polymorphism (RFLP) (Kumari et al., 2021).

Additionally, the samples were employed in 1% agarose gel electrophoresis to verify the quality and integrity of the extracted DNA. To detect nucleic acids according to their sizes, agarose gel electrophoresis is a popular technique. In this study, agarose powder was dissolved in Tris-acetate-EDTA (TAE) buffer, followed by heating and cooling to form a gel matrix, resulting in a 1% agarose gel.The DNA samples were aliquoted with a loading dye and placed into gel wells for visibility. Nucleic acid staining dye ethidium bromide (EtBr), was added to the gel, to allow the visualization of DNA under ultraviolet (UV) light (Sambrook Russell, 2001). Intact genomic DNA appeared as a single, high-molecular-weight band, confirming the absence of significant degradation or contamination.

Deboxed DNA samples were aliquoted and flash-frozen at -20°C without additional analysis. Due to the high-temperature storage limiting the activity of the enzymes responsible for the enzymatic degradation of DNA, and the stability of the DNA sequence for long-term preservation, the sample is suitable for high-

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Volume 10, Issue 1 Jan - Feb 2025, pp: 851-858 www.ijprajournal.com ISSN: 2456-4494

throughput DNA sequencing. This preservation phase is essential for maintaining the accuracy of the diagnosis results in the upcoming molecular assays as well as for obtaining repeatable results in the subsequent molecular assays.One of the most crucial beginning materials for researching genetic polymorphisms on the Interleukin-1β (IL-1β) gene, whose alterations are believed to be crucial in the pathophysiology of RA, is the isolated DNA.

Through strictly controlled DNA extraction from RA patients and normal controls, this work attempts to lay a solid baseline for investigating the linkage between IL-1β gene polymorphisms and RA. The meticulous sample preparation and DNA quality control steps applied in this work are in line with the conduct of the balancing molecular genetics research practice and form a crucial condition for upholding the validity and replicability of the findings (Green Sambrook, 2012).

The genotyping of IL1B

The PCR-RFLP method was used to genotype the IL1B rs16944 gene. The forward primer 5′ GTTGTCATAAGACTTTGACC-3′ and the reverse primer 5′-TTCAGTTCATATGGACCAGA-3′ are the genotyping primer sequences that are utilized. 3 µl (50 ng) genomic DNA, 11 µl water, 2.0 µl 10× PCR buffer, 2.0 µl 2.5 mM dNTPs, and 1.0 µl of 10 µM forward and reverse primers were all included in the 20 µl thermocycler reaction mixture. Thirty-five temperature cycles at 95 degrees Celsius for 15 seconds, 57 degrees Celsius for 60 seconds and 72 degrees Celsius for 50 seconds were followed by ten minutes of initial denaturation at 95 degrees Celsius, followed by a final extension at 72 degrees Celsius for ten minutes. Electrophoresis on a 2.5% agarose gel that had also been stained with ethidium bromide was used to assess the amplified PCR product. The NcoI restriction endonuclease enzyme was subsequently used to digest the PCR products for 16 hours at 37°C.

PlasmaI L-1β protein levels estimation:
Plasma Sample Preparation

One milliliter (mL) of whole blood was used to prepare plasma samples (samples). The blood was collected in K3 EDTA-coated vacutainers, ensuring that coagulation was inhibited and the integrity of the plasma was maintained. Blood samples were subsequently centrifuged at 5000 revolutions per minute (rpm) for 5 min at a refrigerated centrifuge. This step separates the plasma from the cellular components of the blood. Plasma, in the form of clear supernatant, was gently aspirated using sterile pipettes to prevent contamination with buffy coat or red blood cells. Plasma samples were immediately aliquoted and frozen at -80°C to maintain the stability of cytokines (e.g., Interleukin-1β (IL-1β) for the following analysis. Storage at this ultra-low temperature is necessary to avoid the breakdown of proteins and thereby preserve their bioactivity enabling accurate measurement (Vaught et al., 2011).

Measurement of IL-1β Protein Levels

The Human IL-1β Enzyme-Linked Immunosorbent Assay (ELISA) kit, manufactured by Ray Biotech in the United States, was used to measure the amount of IL-1β protein present in the plasma. For the identification and measurement of cytokines in biological systems, ELISA is a sensitive and precise method.

The assay is based on the sandwich ELISA principle, where the capture antibody binds reversibly to IL-1β present in the sample by pre-coating.Plasma samples were thawed on ice before analysis to avoid protein precipitation. A series of standards with well-characterized IL-1β concentrations were prepared following the manufacturer's protocol to develop a standard curve, which can be used as a reference for IL-1β measurement of test samples. The wells of a microtiter plate coated with anti-IL-1β capture antibodies were filled with each plasma sample and standard. After incubation, the wells were washed to remove unbound substances, followed by the addition of a biotin-conjugated detection antibody. The plate was re-incubated so that the detection antibody could bind to the IL-1β-capture antibody complex.

To increase the detection signal, streptavidin-HRP (horseradish peroxidase) is then added to the wells. The colorimetry changes linearly with the concentration of IL-1β in the sample after the substrate solution was introduced to react with HRP. After using a stop solution to quench the reaction, A microplate reader was used to measure absorbance at 450 nm. The level of IL-1β in the plasma samples was ascertained by comparing the absorbance values of the samples with the standard curve (Crowther, 2009).

Study Population

488 people who were tested for rheumatoid arthritis (RA) and lived in the Vindhyan area of Madhya Pradesh made up the study population. Out of these, 221 individuals

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Volume 10, Issue 1 Jan - Feb 2025, pp: 851-858 www.ijprajournal.com ISSN: 2456-4494

tested positive for RA, while 267 individuals tested negative and served as the control group. Participant recruitment was conducted in an ethically sound manner with informed consent from all participants.The control group's average age was 48.95 10.96 years, whereas RA-positive patients' average age was 50.51 14.35 years. Statistical analysis removed age-related biases from the results and confirmed that the age distributions of the two groups did not differ significantly. With 78.28% of all RA-positive people being female, gender analysis revealed a substantial difference in RA prevalence, indicating a higher risk of RA in women. This result is in agreement with the available literature that implicates hormonal and genetic background for higher incidence of RA in women (McInnes Schett, 2011).

Disease Severity Classification

For an even finer stratification of the RA-positive group, the severity of rheumatoid arthritis was classified into mild and severe forms according to well-validated clinical definitions, i.e., the Rheumatoid Arthritis Activity (RAA) score. In addition to inflammatory marker readings including erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), and inflammatory indicators, the scoring technique considers other characteristics such as joint swelling and discomfort.

Among the 221 RA-positive patients, 74 individuals were classified as having mild RAA, while 147 individuals exhibited severe RAA. Such classification provided a finer-grained description of the relationship between IL-1β levels and disease severity.

Statistical Analysis

The collected data was analyzed using statistical software to evaluate the differences in IL-1β protein levels between RA patients and healthy controls. Continuous variables, such as age and IL-1β concentrations, were represented by the mean standard deviation (SD).Numerical characteristics including the gender distribution and the severity of the ailment were reported together with their frequency and percentage. The data's normality was confirmed using the Shapiro-Wilk test. IL-1β levels between the groups were compared using either a parametric test (e.g., Student's t-test) or a non-parametric test (e.g., Mann-Whitney U test), depending on the data distribution. A p-value of less than 0.05 was considered to be statistically significant. In particular, correlation studies were performed to investigate the relationship between the severity of the disease and the level of IL-1β.

Ethical Considerations

The study was carried out in accordance with the Declaration of Helsinki and received institutional ethical approval. Each participant provided written informed consent, guaranteeing their privacy and voluntary participation.

Table1:Demographic profiles of individuals diagnosed with rheumatoid arthritis(RA) and those from the healthy control group.

	RAPatients N=221	Control N=267	P value
Average age (mean age±SD)	50.51 ±14.35Years	48.95±10.96Years	0.2
Gender Male Female	 48(21.71%) 173(78.28%)	 122(45.69%) 145(54.3%)	 <0.001[a]
Rheumatoid Factor (RF ± SD) Mild RA SevereRA	297.10±37.46 74 147	NA	<0.0001[b]
Family history	64(28.95%)	NA	

aWe conducted a logistic regression analysis to ascertain gender-adjusted odds ratios between mild and severe RA in order to address potential gender-related implications on the research outcomes.

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 1 Jan - Feb 2025, pp: 851-858 www.ijprajournal.com ISSN: 2456-4494

PCRRFLP analysis

Heterozygous alleles (A1/A2) have three bands: one at 249 bp, one at 135 bp, and one at 114 bp. Wild-type alleles (A1/A1) have two fragments, one at 135 bp and the other at 114 bp. Mutant alleles (A2/A2) only have 249 bp. Our investigation into the relationship between the IL-1β genotype and the incidence of RA began with these results.

Figure 1: The figure represents the different allelic combinations obtained from Rheumatoid arthritis patients.Lane 1 corresponds to the AI/AI allele and gives band of 135 & 114bp, Lane 2 is of heterozygousalleleA/AIIwhichgave 3bandsofsize 249,135 &114bpandLane 3correspondstothe homozygous allele AII/AII Which gave a band of 249 bp.

Relationship between IL1BSNPand susceptibility and severity to rheumatoid arthritis (RA)

We investigated the connection between rheumatoid arthritis susceptibility and the IL1B SNP (Table 2). Hardy-Weinberg equilibrium was present in both the patient and control groups (p = 0.0920 and p = 0.6221, respectively). Significant differences in IL1B SNP genotype and allele frequencies were found between RA patients and controls in our investigation (see Table 2; p = 0.0001 and p = 0.0015, respectively).In particular, RA patients had a 1.45-fold higher chance of developing RA due to the susceptibility-related 'T' allele being more prevalent in them than in controls (33.03% vs. 22.85%, respectively). (CI: 1.211; OR: 1.042). They created the 'C' allele.'T' allele accounted for 33.03% vs. 22.85% of the allele distribution in RA patients and controls, respectively, while up 66.96% and 77.15% of the allele distribution in RA patients and controls, respectively. However, according to Table 3, there was no discernible relationship between the IL1B SNP and the severity of RA (p = 0.879 and p = 0.923, respectively).

Table2. Genetic association of IL1Bwith RA susceptibility

	RApatients (n = 221)	Controls (n =267)	Pvalue	HWE	Oddsratio(95 %CI)
AI/AI	105(47.8%)	160(59.8%)			1.042
AI/AII	86(38.9%)	92(34.6%)			1.211
AII/AII	30(13.58%)	15(5.6%)	0.0001		0.79
AI	296(66.96%)	412(77.15%)		0.0920(P) 0.6221(C)	0.63
AII	146(33.03%)	122(22.85%)	0.0015		

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 1 Jan - Feb 2025, pp: 851-858 www.ijprajournal.com ISSN: 2456-4494

Table3:Genetic association of IL1β with RA severity					
EmptyCell	**Mild RApatients (n =74)**	**SevereRA patients (n =147)**	**Pvalue**	**HWE**	**Oddsratio(95%CI)**
AI/AI	35(48.1%)	71(48.2%)	0.879		1
AI/AII	28(38.42%)	59(39.99%)			1.091[a]
AII/AII	9(12.77%)	19(13%)			1.208
AI	102(68.9%)	201(67.44%)	0.923	0.331(M) 0.159(S)	0.969
AII	46(31.08%)	97(32.55%)			

III. DISCUSSION

IL-1β is a key mediator of the inflammatory cascade that orchestrates the recruitment of immune cells and the synthesis of pro-inflammatory cytokines. The persistent inflammation that persists in RA has been associated with dysregulated IL-1β signaling. The 'T' allele's prevalence, which is linked to an elevated risk of RA, highlights the possible role of IL1B in RA susceptibility in this population.The findings from our study shed light on the potential correlation between IL1 BSNP and the susceptibility to rheumatoid arthritis (RA)with in the Vindhyanregion of MadhyaPradesh.We found variations in the IL1B SNP's genotype and allele frequency in RA. Thus, RA susceptibility may be related to both patients and controls. According to our findings, RA patients were more likely than controls to carry the 'AII' allele of the IL1B SNP, meaning that the variant is linked to a 1.45-fold increased risk of developing RA. Since interleukin-1 beta (IL-1β), the mRNA of the IL1B gene is essential for the inflammatory process and immune response modulation—two characteristics that characterize the start of RA—this discovery is consistent with the literature that suggests IL1B contributes to the pathophysiologyof RA.

The genotype prevalence of the "AII" allele in RA patients underscores the genetic predisposition of the disease in the Vindhyan area. Moreover, the Hardy-Weinberg equilibrium also obtained for the patient and the control cohort corroborates our findings' validity. Interestingly, our study also highlighted the gender-based differences in RA occurrence, with a higher proportion of female patients testing positive for RA. This observation aligns with existing literature indicating a higher prevalence of RA among females, suggesting the influence of gender-specific genetic and hormonal factors on RA susceptibility. The IL1B SNP and the severity of RA did not, however, significantly correlate, according to our research. This may suggest that although IL1B may have a role in the development of RA, other genetics or the course or severity of the disease may be more significantly influenced by environmental factorsin the Vindhyan population.

A genetic susceptibility to RA in the Vindhyan population is suggested by the observed variation in IL1B SNP genotype and allele frequency between RA patients and controls. This is consistent with the generally accepted hypothesis of RA, which encompasses a large genetic component, where many genetic polymorphisms are implicated in disease susceptibility. IL1B, which encodes the inflammatory pro-inflammatory cytokine interleukin-1 beta, is associated with immune dysregulation and inflammation which are critical to the pathogenesis of RA.

In line with worldwide trends showing a higher prevalence of RA among women, our study found a larger percentage of female RA patients. This sex interaction in the absolute count at disease onset might represent sex differences in hormonal and immune response and hence form the basis for gender-contingent considerations in the characterization of RA pathogenesis as well as the prediction of treatment response.

Although our findings suggest a possible association between the IL1B SNP and RA susceptibility in Vindhyan terrain, some factors must be taken into account here. Sample size may overestimate the genetic diversity of the population. Moreover, lifestyle, diet, and pathogen exposure were also excluded from the analysis. Future studies with increased sample size and full genetic and environmental data are warranted to validate our findings and elucidate the complex interplay of genetics andenvironment in RA pathogenesis. Understanding the genetic makeup of RA in the Vindhyan population may help guide the development of tailored treatment. Targeting IL-1β signaling pathways may provide new therapeutic options for RA treatment, especially in patients who carry the 'T' allele of the IL1B SNP. Further, our results highlight the relevance of genetic screening in identifying patients with an increased risk of developing RA, which may enable early

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 1 Jan - Feb 2025, pp: 851-858 www.ijprajournal.com ISSN: 2456-4494

prevention and better clinical results.

IV. CONCLUSION

Our work yields relevant and useful knowledge regarding the genetic picture of RA in the Vindhyan area, highlighting the IL1B gene as a candidate genetic marker for RA susceptibility. Further studies examining the functional consequences of IL1B variants and their interplay with other genetic and environmental risk factors could further illuminate RA pathogenesis and, ultimately, inform precision medicine in rheumatoid arthritis patients.

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