



## DIFFERENCES IN INTERLEUKIN-23 SERUM LEVELS TOWARDS THE SEVERITY OF ACNE VULGARIS IN WOMEN

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### ABSTRACT

**Background:** Acne vulgaris is a chronic inflammatory disease in the pilosebaceous gland of the skin which can cause depression and anxiety. IL-23 is a cytokine that involved in autoimmune diseases, including acne vulgaris. IL-23 affects the differentiation process of Th17 which further produces IL-17, an important component in the pathogenesis of acne vulgaris. IL-17 plays a significant role in the grade of skin damage caused by the inflammation process. **Objective:** To find out differences in IL-23 serum levels and the grade of AV severity in women. **Methods:** An observational study with a cross-sectional design approach conducted from June until October 2020 in 15 women with AV aged 15-25 years old, not consumed anti-inflammatory drugs, antibiotics, retinoid group of drugs and free from other inflammatory diseases. Subjects were divided into three groups based on the AV severity grade: specifically mild (n=5), moderate (n=5) and severe (n=5). Subjects had venous blood taken for IL-23 serum content by the ELISA method. Data on IL-23 serum content in the three groups were tested using the One-Way ANOVA method to determine the difference. **Results:** Average age from the 3 groups was 21 (17-23) years old. The results showed that there is no significant difference in IL-23 rate from the 3 groups of AV severities (p=0,490). **Conclusion:** : There is no significant difference of IP-23 rate between the 3 groups of AV severities in women.

**Keywords:** IL-23, Acne vulgaris, Grade of acne vulgaris severity.

### INTRODUCTION

Acne vulgaris (AV) is a high prevalence disease especially in South East Asia. According to the Global Burden of Disease Study, the prevalence of AV cases is higher in women at 9.81% as opposed to men at 8.96%.<sup>1</sup> Acne vulgaris is a chronic inflammation of the pilosebaceous unit due to its long course, there is a recurrence pattern and it has manifestations such as acute or late onset. Otherwise, acne vulgaris has psychological and social effects towards the quality of life of the affected.<sup>2</sup>

The cause of AV are multifactorial, influenced by hormonal, genetic, psychology, age, gender, cosmetics, personal hygiene and climate/environment. AV had different manifestations in every individual according to the severity. Indonesian Cosmetic Dermatology Study Group (KSDKI) used the Lehmann's classification to assess the degree of severity of AV. Lehmann's Classification was divided into mild, moderate and severe by calculating the inflammatory and non-inflammatory lesions found on the face.<sup>3</sup>

The pathogenesis of AV involved the hyperproliferation of epidermal follicles, increased

the production of sebum, bacterial colonisation *Propionibacterium acnes* (*P. acnes*), *Staphylococcus epidermidis* (*S. epidermidis*), *Pityrosporum ovale* (*P. ovale*) and inflammation. Inflammation in AV induced by an immunologic response to *P. acnes* that caused stimulation of keratinocyte via *Toll-Like Receptors* (TLRs).<sup>4</sup> Keratinosit Assisted stimulated keratinocyte by innate immune cells that produces TNF. TNF worked together with the *Neural Wiskott-Aldrich Syndrome Protein* (N-WASP) and *Nuclear Factor* (NF) to influenced the expression of IL-23.<sup>5</sup> Active IL-23 played an important role in the differentiation process of Th17 where Th17 in turn produces IL-17 that played a role in AV chronic inflammation.<sup>6</sup>

IL-23 is an important cytokine for the autoimmune and chronic inflammatory process. There had been evidence of elevated IL-23 levels in the two conditions. Studies shown the role of IL-23 towards the different autoimmune diseases and chronic inflammation, however there was no study identified the difference between IL-23 and AV. Therefore, the researcher was interested in the study of IL-23 levels to the severity of AV in women.



## METHODS

This research is an Observational Research using the *cross-sectional design* approach. The subject in this research are women affected by AV chosen using *Consecutive sampling* at the ages of 15-25 years old, have not taken antibiotics, anti-inflammatory and retinoid group medications, and have no inflammatory disease history nor autoimmune disease based on patient history.

Total subject involved are 15 persons divided into 3 groups based on their severity of AV using Lehmann's criteria; mild, moderate and severe. Each group has 5 subjects. Data collection done by taken blood samples during the day. The obtained blood sample then given to the GAKI Laboratory to undergo centrifugation. Results of the centrifuge in the form of serum will be stored at  $-60^{\circ}$  Celsius to avoid sample damage. After collected, serum measured for IL-23 level using *high sensitivity* (HS) test kit using ELISA Method.

Data processed using a data processing program. The processed data distribution are observed and the normality test using *Shapiro-Wilk* due to the sample size of this research  $<50$  and continued with a comparative test using oneway ANOVA Difference Test. This research has obtained *Ethical Clearance* from the Health Research Ethics Commission (KEPK) Faculty of Medicine Diponegoro University, the serial number stated as. 190/EC/KEPK/FK UNDIP/VIII/2020.

## RESULTS

Data collection of the research done between July - October 2020. The subjects were 15 women affected by AV in Semarang City, divided into 5 women with mild AV severity, 5 women with moderate AV severity and 5 women with severe AV severity.

**Table 1.** Characteristics of Research Subjects

Variable	n	%	Mean ± SD	Median (min – max)	Degree of AV Severity			p	
					Mild	Moderate	Severe		
Age			20.40 ± 1.55	± 21(17-23)				0.001	
					20.60 ± 0.55	20.60 ± 2.19	20.00 ± 1.73		0.412
Occupation								0.192	
High school	2	13,3			0 (0%)	1 (20%)	1 (20%)		
Student									
College	13	86,7			5 (100%)	4 (80%)	2 (80%)		
student									
IL-23 (pg/ml)			57,56 ± 37,44	55,95 (5,31 – 125,8)				0.379	

The Median age of the research subjects were 21 (17 - 23) years old with the youngest was 17 years old and the eldest was 23 years old. From Table 1, indicated that the distribution of age data in the subjects of the research were not normal. The research subjects had not consumed any antibiotics, anti-inflammatory drugs and retinoid group medications. The research subjects also not diagnosed from autoimmune diseases or other chronic inflammation. From Table 1 also stated that

the occupation of the subjects were dominated by college students.

The analysis results shown that the subjects with mild AV had an average age of  $20.60 \pm 0.55$  years, subjects with moderate AV averages at  $20.60 \pm 2.19$  years and subjects with severe AV averages at  $20.00 \pm 1.73$  years, and from the results of the analysis of difference test the results were not significant in the three groups.

Up to 100% of research subjects diagnosed with mild AV were comprised of college students. 80%



of the subjects diagnosed with moderate AV were college students and the remaining 20% were high school students and from the analysis difference test shown no significant differences between the three groups.

**Table 2.** Data Distribution of IL-23 Levels Based on the severity of acne vulgaris. Significant if  $p < 0.05$

Degree of Severity	AV	Average $\pm$ SD (pg/mL)	p
Mild		40.38 $\pm$ 26.52	0.913
Moderate		64.58 $\pm$ 45.48	0.707
Severe		67.73 $\pm$ 39.69	0.930

Results of the analysis shown the average measurement of IL-23 levels on the mild AV group was at 40.38  $\pm$  26.52, moderate at 64.58  $\pm$  45.48, and severe at 67.73  $\pm$  39.69. The normality test shown that the distribution of data for IL-23 serum was normal.

The analysis was continued to find the difference of IL-23 serum levels between the 3 groups of degree of AV severity. Results of the analysis shown that there was no difference of the IL-23 serum levels between the 3 degree of AV severity ( $p=0,490$ ).

## DISCUSSION

Acne Vulgaris (AV) is a chronic inflammation of the pilosebaceous unit accompanied by blockage of keratin material.<sup>7</sup> Pathogenesis of AV includes numerous factors and one of the factors is the inflammatory process of AV triggered by *P. Acnes*.<sup>8</sup> This Microorganism trigger the production of cytokines, one of it is IL-17. IL-17 is a strong pro inflammation cytokine that influences the degree of severity of tissue damage caused by inflammation.<sup>6</sup> IL-17 is produced by differentiation of Th17.<sup>9</sup>

IL-17 produced by the differentiation of Th17 induced by IL-23. IL-23 is a cytokine produced by mononuclear cells. The level of IL-23 is systematically influenced by several factors, and research by L Li H dkk shown that the levels of IL-23 increased in cases of chronic inflammation and autoimmune diseases.<sup>5</sup>

The level of IL-23 at a chronic inflammation condition has been studied plentiful, such as the

research by Ahmed Aladl dkk whose study was about the relationship between IL-23 and psoriasis, atopic dermatitis, dan lichen planus. The research used a *case control* approach and *Random sampling* technique.<sup>10</sup> Whereas, this research used *cross sectional* approach and *Consecutive sampling* technique. However, until now there had been no research studying the relationship between IL-23 and the degree of AV severity. The purpose of this research was to find out the relationship between the level of IL-23 serum to the degree of AV severity in women by comparing the level of IL-23 of the grouped women affected by AV based on the degree of severity.

As a whole, the average level of IL-23 serum in this research was 57.56  $\pm$  37.44 pg/mL, compared to the research by Gerenova dkk, which stated that the average levels of IL-23 of the research subjects were normal (subjects that have no history of AV or chronic inflammation nor autoimmune disease) which was at 14.76  $\pm$  2.2 pg/mL. AV Cases can be concluded that there was an increase of IL-23 serum level according to the severity of AV. Any differences found in this research and the research done by Gerenova dkk is, Gernova dkk used 85 women and 8 men as subjects, other than that the research done by Gerenova dkk used subjects at an average age of 47.6  $\pm$  13.6 years old, whereas in this research the average age was at 20,40  $\pm$  1,55 tahun.<sup>11</sup>

This research produced a result of level of IL-23 to mild AV at 40.38  $\pm$  26.52 pg/ml, level IL-23 at moderate severity was at 64.58  $\pm$  45.48 pg/ml, and IL-23 level at severe degree of severity was 67.73  $\pm$  39.69 pg/ml. Based on the results of this data analyzed found  $p = 0,490$ . Thus, the bivariat analysis of the 3 groups in this research shown that there was no difference between the IL-23 levels between the groups of women based on the degree of AV severity. This is due to the fact there were several factors affecting the results which are genetics and the condition of the subjects.

The influence of genetics through hormones have been studied by Ekasari et al that proven CYP17 influences the degree of AV severity. Gene CYP17 was found in chromosome 10q24.3 that codes P450c17 $\alpha$  which is an enzyme involved in biosynthesis of androgen hormones and to mediate the activity of enzym 17 $\alpha$ -hidroksilase and 17,20-liase. Polymorphism is a type replacement of base T



to C in 5'-*untranslated region* (UTR) especially in the 34<sup>th</sup> base pair near the initiation of translation of gene that influences the severity of AV.<sup>12</sup> Research by He et al, proven that Chinese people have gene allele CYP17-34C/C homozygote which poses risks of acquiring severe AV. In this research, there was no identification of gen allele CYP17-34C/C to the point where it was not possible to explain the genetic trigger of IL-23.<sup>13</sup>

Other factors that influences the subject's condition are stress, menstruation cycle. Ekasari et al found that stress triggers the production of inflammatory mediator due to the increase of glucocorticoid and androgen adrenal which causes the increase of inflammatory cytokines.<sup>12</sup> According to Khosravisamani et al, menstruation also influences the severity of AV same as stress which shows the elevation of IL-23 levels in menstruating women.<sup>14</sup> In this research, there was no data related with the condition of menstruation cycles nor the level of stress on the research subjects where the trigger in the form of menstruation cycles and the level of stress cannot be ascertained.

Until now, there was no scientific evidence regarding the difference between IL-23 and the degree of AV severity. This research was limited because this was the first research that studied the differences between IL-23 serum to the degree of AV severity, and the size of the sample of this research was determined by the *rule of thumb*, by used 5 subjects for each group and based on data from this research, the researcher counted that the size of the sample is 109 persons per group. Further studies must be done with larger samples.

## CONCLUSION

There is no difference between the level of IL-23 serum in women to the degree of AV severity. Further study must be done to analyse other trigger factors such as genetic condition, menstruation cycle, eating patterns, and onset of AV.

## REFERENCES

1. Heng AHS, Chew FT. Systematic review of the epidemiology of acne vulgaris. *Scientific Reports*. 2020;10(1):1–29.
2. Moradi TS, Makrantonaki E, Ganceviciene R, Dessinioti C, Feldman SR, Zouboulis CC. Acne vulgaris. *Disease primers*. 2015; 1(1). 1-7.
3. Bernadette I, Kapantow GM, Yenny SW, Hidritiani R. Akne. 1 Ed. Jakarta. Badan FK UI. 2018. 1–59.
4. Maulinda S, Hindritiani R, Ruchiatan K, Suwarsa O. Perbandingan Kadar Interleukin-17 Serum Pasien Akne Vulgaris Tipe Papulopustular dengan Komedonal. *Journal Kedokteran Bandung*. 2016;48(3):160–3.
5. L LH, Yao Q, Mariscal AG, Wu X, Hülse J, Pedersen E, et al. Epigenetic control of IL-23 expression in keratinocytes is important for chronic skin inflammation. *Nature Communications*. 2018 ; 12(1).6-15.
6. Ebrahim AA, Mustafa AI, El-Abd AM. Serum interleukine-17 as a novel biomarker in patients with acne vulgaris. *Journal of Cosmetics Dermatology*. 2019; 18(6). 1-5.
7. Rimadhani M, Rahmadewi. Pengaruh Hormon terhadap Akne Vulgaris (Hormone Influence in Acne Vulgaris). *Jurnal Kedokteran Airlangga*. 2015;27(3):218–24.
8. Mochtar M, Murasmita A, Irawanto ME, Julianto I, Kariosentono H, Waskito F. The difference in interleukin-19 serum on degrees of acne vulgaris severity (tesis). *International Journal Of Inflammation. Dermatology Departement, Sebelas Maret University, Dr. Moewardi General Hospital, Surakarta*. 2018; 1-4.
9. McKenzie BS, Kastelein RA, Cua DJ. Understanding the IL-23-IL-17 immune pathway. *Trends in Immunology*. 2006 ;27(1):17–23.
10. Aladl A, Mosbeh A, Salama S. Assessment of Serum Interleukin 23 , 17 Level and its Relation to Disease Control in Psoriasis , Atopic Dermatitis and Lichen Planus : A Serological Study . *Scientific Journal of Clinical Research in Dermatology*. 2020;5(1):3–11.
11. Gerenova J, Manolova I, Stanilova S. Serum Levels Of Interleukin - 23 And Interleukin - 17 In Hashimoto's Thyroiditis. *Acta Endocrinol (Buchar)*. 2019;5(1):74-79.
12. Dhany P K, Tantari S, Arif W. Kadar Tumor Necrosis Factor- $\alpha$  Plasma Pada Berbagai Derajat Keparahan Akne Vulgaris di RSUD Dr. Saiful Anwar Malang. *Jurnal Kedokteran Universitas Brawijaya*. 2018; 5(2): 84–93.
13. He L, Yang Z, Yu H, Cheng B, Tang W,



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- Dong Y, et al. The relationship between CYP17 -34T/C polymorphism and acne in Chinese subjects revealed by sequencing. *Journal Dermatology*. 2006;212(4):338-42.
14. Khosravisamani M, Maliji G, Seyfi S, Azadmehr A, Abd Nikfarjam B, Madadi S, et al. Effect of the menstrual cycle on inflammatory cytokines in the periodontium. *Journal Periodontal Res*. 2014; 49(6): 1-6.