To study the serum level of ST-2 in COPD patient in a tertiary care center of India

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KEYWORDS

COPD, ST-2 , Smokers, Non-smokers.

ABSTRACT

Background: In COPD patients, the abnormal inflammatory immune responses are accelerated by the IL-33/ST2 pathway activation after chronic insult from smoking, air pollution and biomass fuel exposure. Thus IL-33/ST2 act as a central driver in the pathogenesis of COPD. ST-2 is a heterodimeric receptor which in necessary for IL-33 induced downstream signal inflammatory pathway. So ST-2 receptor is a key regulator of inflammation in COPD. **Aim & Objective**: we aim to find out responder phenotype for anti-ST-2 therapy by the estimation of serum ST-2 levels in COPD patients in a tertiary care center of India. The objective of the present study is to assess the serum level of ST-2 in COPD patients and healthy control subjects with cigarette smoking and without cigarette smoking. **Material and method:** This Prospective study was done at Shri Guru Ram Rai Institute of Medical and Health Sciences, Dehradun. In order to estimate serum ST2 level by ELISA method 150 COPD patients and 150 healthy control were enrolled from Shri Mahant Indresh hospital, Dehradun. **Result:** The concentration of ST-2 is significantly higher in COPD patients as compared to healthy control. **Conclusion:** It may be concluded that ST-2 may be a potential biomarkers for anti ST-2 therapies to reduce the rate of exacerbations in COPD and improve their quality of life.

Introduction:

COPD is characterized by airflow limitation in lungs due to chronic inflammation caused by significant exposure to irritants such as cigarette smoke, air pollution, biomass fuel and old infection with tuberculosis [1]. Pathological changes in lung parenchyma and airway are associated with inflammation along with parenchymal destruction, loss of alveolar tissue (emphysema) and chronic bronchitis[2,3] . Inflammatory cytokines are released from inflammatory cells which leads to a series of abnormal innate inflammatory immune responses by activation of IL-33/ST-2

pathway, i.e. a key regulator of COPD pathogenesis. After cell necrosis, the IL-33 is released from nucleus into circulation and binds with the ST-2 receptor which induces the dimerization of TIR domain (Toll Interleukin 1) of ST-2 and downstream signal via MyD88 adaptor [3]. MyD88 interact with IRAK 1/4 and recruits TRAF6 which in turn activates NF-κB and MAPK (mitogen activated protein kinase). Apart from this binding of IL-33 to ST-2 receptor also induces the INF γ expression by regulating TNFα and IL-1β and mediate type 2 inflammatory immune responses[4].
ST-2 is a heterodimer receptor which is also known as IL1RL1 (interleukin 1 receptor like 1 family) and IL-1RAcP (IL-1R-combined protein)[4,5]. ST-2 also leads to activation of natural killer T-cells (NK), type 2 innate lymphocytes cells (ILC 2), basophils and dendritic cells[5].All these cells are involved in type 2 innate immune responses[5]. Furthermore, inflammation is aggravated by ST-2 expression in goblet cells and epithelial cells[3]. So ST-2 plays an important role in pathogenesis of COPD and ST-2 concentration is increased in COPD patients. Thus ST-2 can be gold standard biomarker of COPD and may be targeted as therapeutic agent to improve strategies of treatment by interrupting binding of IL-33 to ST-2 receptor via blocking the MyD88 dependent inflammatory pathway[4]. Special attention has been paid to develop antibodies against ST-2 receptor (anti-ST-2) therapy as a treatment regimen. Moreover, currently some ongoing clinical trial are already on phase 1, phase 2 and phase 3 based on anti-ST-2 therapy for COPD treatment[3]. Astegolimab an anti-ST-2 monoclonal antibody (already used in asthmatic patient) is under clinical trial (NCT03615040, NCT050505037929 and NCT05595642) for COPD patients[3]. In India, apart from smoking , other factors are also involved in progression of COPD such as biomass fuel, occupational exposure, premature birth , old infection such as tuberculosis[6,7]. So we can find the responder phenotype for anti-ST-2 therapy by estimation of serum level of ST-2 in smokers and non-smoker COPD patients. Thus, we aim to find out responder phenotype for anti-ST-2 therapy by the estimation of ST-2 in COPD patient in a tertiary care center of India.

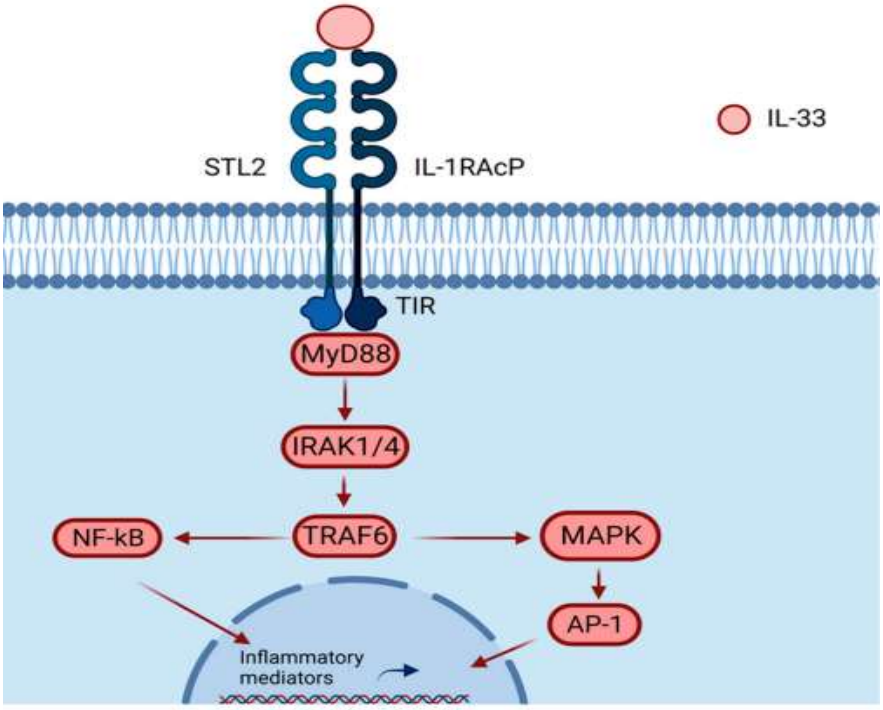

Figure 1: IL-33 binding to the ST2 activate MyD88 into its intracellular domain (TIF). MyD88 binding activates the IL-1R-associated kinase (IRAK) and TRAF6, NF-κB or AP-1 pathway

Activations of NF-κB and AP-1 initiate inflammatory cytokine expression and abnormal inflammatory responses[3].

Material and method: - The aim and objective of present study is to assess the serum level of ST-2 in COPD patients and healthy control subjects with cigarette smoking and without cigarette smoking. This Prospective study, done at Shri Guru Ram Rai Institute of Medical and Health Sciences, Dehradun. This study included 150 COPD patients and 150 healthy control subjects enrolled from Shri Mahant Indresh hospital, Dehradun. Participants were more than 40 years of age at the time of enrolment into the study. COPD diagnosis was made on spirometry with post bronchodilator FEV_1/FVC <0.70 as per GOLD guidelines in Respiratory department of Shri Mahant Indresh hospital[7]. COPD patients had a smoking history of more than 10 pack years. Each COPD patient was on regular treatment for COPD and had a documented history of ≥ 2 moderate or ≥ 1 severe COPD exacerbation within 12 month prior to enrolment. All patients enrolled were willing and able to give consent, and also willing and able to comply with study protocol. All questionnaires and Investigations which are mentioned were done at Shri Mahant Indresh hospital. After overnight fast blood samples was collected from each subject by venipuncture with standard blood collection technique in a plain vial for the analysis of ST-2. The ST-2 was analyzed by using ELISA method. The normal range for ST-2 was between 0.31- 20ng/ml [8]. Here sST-2 (soluble ST2) form of ST-2 was measured. The data obtained was analyzed by using SPSS statistical software for Windows. The mean and standard deviation of ST-2 in COPD (smokers and non-smokers) and healthy control (smoker and non-smokers) was calculated .Comparisons of ST-2 between healthy control and COPD patients and between healthy smokers versus non-smokers, COPD smokers versus non-smokers were analyzed using the student's t-test and Pearson's correlation coefficient to calculate the p-value or to find out significant difference.

Result and Discussion:

Table-1: Comparison of serum sST-2 levels (ng/ml) of COPD with healthy control and COPD (smoker versus non-smokers) , healthy control (smoker versus no-smokers)

Group		**Mean±SD**	**p-value**
COPD		10.72±7.44	<0.0001***
Control		3.57±2.28	
COPD	**Smokers**	11.55±7.884	0.021
	Non-smokers	8.365±5.46	
Control	**Smokers**	4.36±2.09	<0.0001***
	Non-smokers	1.31±0.816	

P<0.01=significant*, P<0.001=very significant, P<0.0001=extremely significant*****

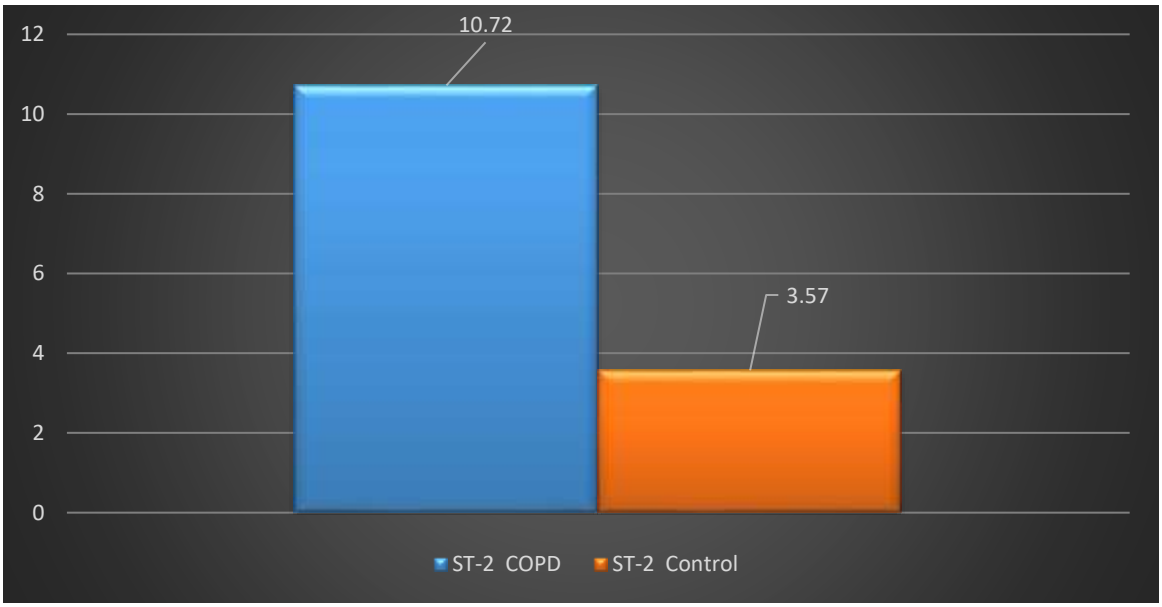

Figure 2: Comparison of ST-2 (ng/ml) in COPD and control group

Figure 3: Comparison of ST-2(ng/ml) in Smoker and non-smoker COPD and control groups

Here within present study we sought to understand effect of inflammation in serum level of ST-2 of COPD patients. Chronic irritation to airways triggers the abnormal innate immune responses by increased expression of ST-2 receptor after IL-33 binding to ST-2. Thus here in our study COPD patient had significantly higher serum level of ST-2 in COPD patient as compared to healthy control. In present study the mean value of ST-2 level in COPD patients was 10.72±7.44 ng/ml and in healthy control subjects was 3.57±2.28 ng/ml and the difference was statistically significant ($P < 0.001$**).** Present study findings are similar with the Xia J, (2014) et al. They found significant ($P< 0.001$) difference between COPD patient and control subjects ST-2 level in plasma[9]. Our study is also closely related to Yuan Z (2016) et al, Gabryelska A (2019) et al and Jiang M (2019) et al[4,5,10]. In a study performed by Tworek D (2018) et al, the sST2 was increased in serum ($p < 0.05$) of COPD patients compared with the healthy controls and ST2 was increased in the sputum($p = 0.059$) and in the serum of COPD patients with airway eosinophilia ($p < 0.001$)[11]. The hypothesis to use ST-2 as inflammatory disorder biomarker in COPD was given by Dattagupta A. (2018) et

al. after findings a higher ST2 in cardiovascular patients[12]. In the study conducted by Kraydashenko OV (2021) et al, the ST2 were highly upregulated in in patients with comorbid pathology group with COPD on the background of arterial hypertension[13]. Asthma and COPD share the same pathway of IL/ST2 involved inflammation. Yagami A. (2022) et al also state that all isoforms of ST2 was highly upregulated in asthma and Jayalatha AS(2021) et al quoted ST2 as a receptor for IL-33[14,15]. A study in a mice model performed by Allinne J (2019) et al quoted that the IL-33 and ST2 expression were up regulated when mice were exposed to HDM(house dust mite) and normalized with anti-IL-33 treatment[16] . Similarly, a study was also performed in a mice model by Zeng S. (2015) et al and they also found increased IL-33 and ST-2 mRNA expression exposed to intranasal infection with Respiratory Syncytial Virus (RSV) and anti-ST2 treatment inhibits RSV-induced lung histopathology[17]. The chronic exposure to cigarette smoke and exposure to any other form of toxic material are major factors involved in COPD pathogenesis and inflammatory responses[7]. However, to the best of our knowledge there is no Indian study on the estimation of serum ST-2 levels in COPD in Indian population. Thus here in this study we sought to estimate serum ST-2 level between COPD smokers versus non-smokers as well as in healthy control smokers versus non-smokers. In order to investigate effect of smoking in COPD and healthy controls airways, we compared ST-2 level between smokers and non-smokers healthy control subjects, the mean value of ST-2 was 4.365±2.09 ng/ml in smokers control and 1.310±0.818 ng/ml in non-smokers control and the difference was significant ($p<0.0001$). The mean ST-2 value in COPD smoker was11.55±7.88 ng/ml and COPD non-smokers was 8.365±5.46 ng/ml and the difference was not statistically significant (P=0.021). Tworek D (2018) et al quoted increased sST-2 in COPD ($p< 0.05$) patients as compared to healthy smokers[11]. Similarly in a study conducted by Yuan Z (2016) et al , the serum ST2 in slight and moderate COPD patients is twice higher than that of healthy smokers[4]. So we can consider anti- IL-33/anti-ST-2 monoclonal antibodies as inflammatory circuit breakers for IL-33/ST-2 accelerated inflammatory downstream signal in COPD patients. Similar statements were also given by Zhao J(2015) et al and Chen WY (2018) et al and Riera-Martínez L et al[3,18,19].

Conclusion:

The results of present **s**tudy highlights that the ST-2 can be used as key biomarker in order to investigate inflammation in COPD and anti-ST2 therapies may be a potential line of treatment for COPD patients as we found increased ST2 serum level. We can conclude that by blocking of IL-33/ST-2 pathway, maybe we will be able to restore irreversible inflammatory changes in COPD patients and remodeled tissue primed for exacerbations and thereby may be quality of COPD patients will be improved. To the best of our knowledge, this is first research conducted in India in COPD patients for the estimation of serum ST-2 levels and these findings need randomized control trials.

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