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Possible immuno-modulatory effects of tocilizumab in patients with refractory status epilepticus

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Abstract. – OBJECTIVE: Refractory Status Epilepticus (RSE) is a neurologic emergency that carries a high risk of mortality and morbidity. Every year, there are about 200,000 cases in the United States, affecting people of all ages. This study aimed to investigate the possible immuno-modulatory effect of tocilizumab in RSE patients receiving conventional anti-epileptic drugs.

PATIENTS AND METHODS: 50 outpatients who fulfilled the inclusion requirements for RSE were recruited in this randomized, controlled, and prospective study. The patients were divided into two groups randomly (n=25); the control group received standard RSE treatment, consisting of propofol, pentobarbital, and midazolam, and the tocilizumab group received standard RSE treatment plus tocilizumab. A neurologist evaluated each patient at the beginning of the therapy and after 3 months. Before and after treatment, serum nuclear factor kappa B (NF-κB), interleukin-6 (IL-6), tumour necrosis factor-alpha (TNF-α), interleukin-1β (IL-1β) and serum electrolytes were assessed.

RESULTS: The tocilizumab group showed a statistically significant reduction in the level of assessed parameters in comparison with the control group.

CONCLUSIONS: Tocilizumab might be a novel adjuvant anti-inflammatory medication in managing RSE.

Key Words:
Epilepsy, IL-6, Inflammation, Tocilizumab.

Introduction

Status Epilepticus (SE) is defined as a neurologic emergency characterized by high rate of morbidity and mortality[1].

SE is characterized by ongoing or recurrent seizures brought on by the ineffectiveness of seizure termination mechanisms[2]. RSE is a condition in which seizures continue despite the use of first-line drug (IV benzodiazepine) and second-line drug (IV antiepileptic drugs)[3]. It is one of the more severe and protracted forms of SE. It can be found[4,5] in 29-43% of SE cases. The pathogenesis of epilepsy is known to be excitotoxicity, which is triggered by excessive glutamate receptor activation and results in Na^+ and Ca^{++} influx through the plasma, which causes dendrite degeneration and cell death[6]. Recent research[6] has revealed that immuno-modulatory and neuro-inflammatory processes, which are regularly triggered in human epileptogenic brain regions and are undoubtedly engaged in animal models of epilepsy, contribute to the pathogenesis of epilepsy. Blood brain barrier (BBB) leakage through inflammation of the CNS has been linked to epilepsy progression, and it has been demonstrated that BBB leakage plays a role in both the advancement of epileptogenesis and the production of seizures[7]. The etiology of epileptic seizures has also been linked to possible cytokine involvement. The activation of proinflammatory cytokine production and inflammatory interactions in the brain following the occurrence of seizures have been studied in experimental models and clinical research. Epilepsy patients' histopathological investigations grab attention because the insight they provide highlights the role of inflammation in epileptogenesis[8].

Additionally, it has been demonstrated that the long-term consequences of inflammatory mediators contribute to changes in the permeability characteristics of the BBB. It has been demonstrated[9] that ionic imbalances in the extracellular environment can lower seizure threshold. It is thought that inflammatory processes involving glial activation play a substantial part in epileptogenesis in considering the various ways that demonstrate glial cells' contribution to the process. Release of inflammato-

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ry proteins, most likely cytokines and chemokines, hallmarks this activation[10]. Proinflammatory cytokines including IL-1, IL-2, and IL-6 are found in healthy brain tissue at relatively low concentration levels and have been demonstrated[8] to rise following generalized seizures. *In vivo* research[8] showed that the hippocampus' synthesis and secretion of proinflammatory cytokines such IL-1, IL-6, and TNF- increased immediately after tonic-clonic seizures. The possibility for these exogenous inflammatory mediators to lower seizure thresholds exists, which may change neurotransmitter release or uptake, channel activity, and glia-associated regulation of extracellular environments, such as potassium level[11,12].

Tocilizumab is an IL-6 receptor antagonist and an immuno-modulatory drug. Tocilizumab has been studied[13] for its potential to treat acute seizures, as well as some refractory autoimmune neurologic diseases.

The present study aimed to investigate the possible immuno-modulatory effects of tocilizumab in RSE patients receiving conventional anti-epileptic drugs.

Patients and Methods

Patients

Fifty patients who fulfilled the inclusion criteria were included in the study as shown in Figure 1. They were recruited from the outpatient clinic from Mansoura University Hospital.

Inclusion Criteria

- Age: 15-75 years old.
- Gender: Male and female.
- Newly diagnosed RSE patients who are scheduled to receive anti-epileptic drugs.
- Patients with normal renal and hematological functions.

Exclusion Criteria

- Old age >75 years old.
- Pregnant or lactating females.
- Severe kidney dysfunction patients when GFR level <30 ml/min.
- Hepatic patients (ALT or AST >3 times ULN).
- Neutropenia when neutrophil count < 500 cells/mm^3.
- Thrombocytopenia when platelet count < 50,000 cells/mm^3.
- Patients taking immunosuppressant drugs, and anti-inflammatory drugs affecting serum cytokines.
- Cancer patients taking chemotherapy.
- Patients with inflammatory bowel diseases.
- Patients with a known hypersensitivity to any of the used drugs.

Study Design

This was a prospective, randomized, and controlled clinical study to investigate the possible immuno-modulatory effect of tocilizumab in RSE patients receiving conventional anti-epileptic medications compared to patients receiving conventional anti-epileptic therapy for treatment of RSE plus tocilizumab.

This clinical study was granted approval by the National Research Ethics Committee at Tanta University with the approval number 34233/11/20. The current study was carried out in accordance with the 1964 amendments to the Helsinki Declaration's associated ethical principles. All patients provided their informed consent before the present study was registered in 2022 at www.ClinicalTrials.gov with the reference code NCT05346666. The randomization was made using a computer random number generator to select random permuted blocks.

Patients whose inclusion criteria were fulfilled and provided informed consent were randomly divided into 2 groups (n = 25), as shown in the CONSORT flow diagram (Figure 1). The control group received the standard course of treatment for SE, which included phenobarbital (loading dose was 5-15 mg/kg over one hour) and propofol (Pofol^R 10 mg/ml ampoule, Dong Kook, Korea; loading dose was 3-5 mg/kg with maintenance dose of 1-15 mg/kg/h). Midazolam (Dormicum^R 15 mg ampoule, Hoffman La Roche, France; loading was 0.2 mg/kg with maintenance doses between 0.05 and 2.0 mg/kg/h), and infusion rates could be continued at 0.5-15 mg/kg/h. Selected dose for tocilizumab was based on previous studies[9].

Tocilizumab group received the same treatment in control group in addition to tocilizumab (ACTEMRA^R 200 mg vial, Hoffman La Roche, France) that was started at a dose of 4 mg/kg administered twice monthly for 3 months.

Sample Size Calculations

Calculation of sample size was based on data from a previous study by Dastan et al[14], taking into consideration dropout rates.

Follow-Up

Weekly meetings and daily phone calls were used to check in on patients. To rule out any organic dysfunction, patients underwent liver function tests, renal function tests, com-

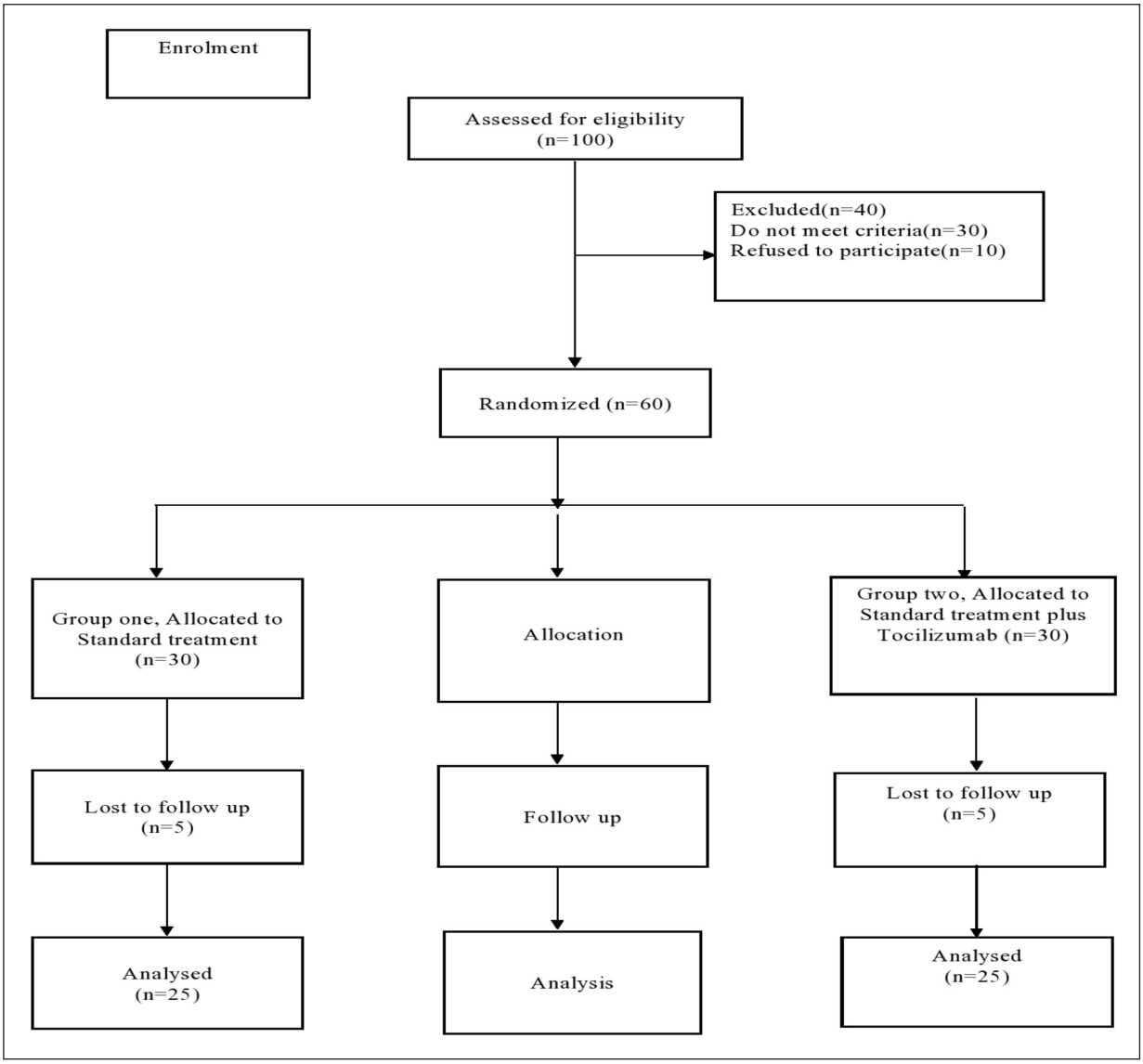

Figure 1. CONSORT diagram presenting the participated patients in this study.

plete blood counts and a full medical history at their initial visit. Additionally, IL-6, IL-1β, NF-κb, and TNF-**α** were measured as serum inflammatory biomarkers. The included cases were scheduled for routine follow-up visits. Determination of RSE prognosis was made by the Modified Status Epilepticus Severity Score (mSTESS: 0-2 = low risk; 3-4 = intermediate risk; >4 = high risk)[15].

Therapeutic Assessments

The primary endpoint was to study the effects of tocilizumab on the prognosis of RSE using mSTESS.

The secondary endpoint was determination of changes in inflammatory biomarkers and serum electrolytes.

Sample Collection

Between 8 and 10 a.m. before the study started and three months after the intervention, 10 mL of blood were withdrawn from every patient. 2 mL of blood were gradually transferred into EDTA tubes for complete blood count analysis. Into plain and clean test tubes, the remaining 8 mL were gradually transferred, where they were given time to clot before being centrifuged for 10 minutes at 4,500 g. (Hettich Zentrifugen EBA 20, Westfalia, Germany).

Two aliquots of serum were separated: one was frozen at -80°C for cytokine analysis and the other was used for standard kidney and liver function tests.

Biochemical Analysis

Using spectrophotometric kinetic method, the liver enzymes alanine aminotransferase (ALT) and aspartate transaminase (AST) were measured in serum samples[16]. Serum creatinine levels, a measure of kidney function, were calculated using the Jaffé reaction[17]. 2 mL of blood in EDTA tubes were examined using an automated hematology analyzer to determine the complete blood count.

Using enzyme-linked immunosorbent assay (ELISA) kits, serum levels (expressed as pg/ml) of NF-κb (catalogue no. 201-12-0691), TNF-α (catalogue no. 201-12-0083), IL-1 (catalogue no. 201-12-0144), and IL-6 (catalogue no. 201-12-0091) were measured according to manufacturer instructions (Sunredio, Shanghai, China).

Statistical Analysis

The study was conducted with an 80% power level and a 95% level of significance. Statistical Package for Social Sciences (SPSS) version 26 for Windows was used to code, process, and analyze the collected data (IBM Corp., Armonk, NY, USA). Prior to and after therapy, significant differences within the group were found using paired Student's *t*-tests and significant differences between the groups were determined by unpaired Student's *t*-tests. The Pearson's correlation coefficient was used to correlate the assessed parameters. Categorical data were subjected to the Chi-square test. All *p*-values were two-tailed, with $p < 0.05$ considered statistically significant.

Results

Clinical and Demographic Data

This study included 50 RSE patients who were classified into two groups. Group 1 (the control group) received conventional treatment (propofol, pentobarbital, and midazolam); group 2 (the tocilizumab group) received conventional treatment and tocilizumab. Their baseline data was presented in Table 1. There were no significant variations between the studied groups in demographic data such as age ($p = 0.80$), sex ($p = 0.87$), and body mass index ($p = 0.87$).

Effect of the Studied Medications on Modified Status Epilepticus Severity Score (mSTESS) in the two Study Groups

Table II shows that there was no statistical significance between all the patients at the start of

Table I. Comparison of the basic post-operative conditions of four groups (mean).

Parameter	Control group (N=25)	Tocilizumab group (N=25)	*p*-value
Age (years)	34.76 ± 9.10	35.44 ± 10.44	0.80
Sex (M/F)	12/13	13/12	0.87
BMI (Kg/m^2)	27.55 ± 3.78	26.94 ± 3.75	0.87

Data presentation as mean ± SD. Control group: RSE patients treated with conventional treatment (propofol, pentobarbital, and midazolam); Tocilizumab group: RSE patients treated with conventional treatment plus tocilizumab; F: female; M: male; BMI: body mass index.

Table II. Analysis of Modified Status Epilepticus Severity Score (mSTESS) in the two study groups.

	Control group (N=25)			Tocilizumab group (N=25)			
Parameter	Before therapy	After therapy	*p*-value[a]	Before therapy	After therapy	*p*-value[a]	*p*-value after therapy[b]
mSTESS	4.64 ± 1.22	3.52 ± 1.08	0.0001*	4.68 ± 1.34	1.64 ± 0.90	0.0001*	0.0001*

Data presentation as mean ± SD. Control group: RSE patients treated with conventional treatment (propofol, pentobarbital, and midazolam); Tocilizumab group: RSE patients treated with conventional treatment plus tocilizumab; mSTESS: Modified Status Epilepticus Severity Score. [a,b]: level of significance within and between groups respectively; *: statistically significant ($p < 0.005$).

Table III. Analysis of serum electrolytes in both groups before and after therapy.

Parameter	Control group (N=25)			Tocilizumab group (N=25)			*p*-value after therapy[b]
	Before therapy	After therapy	*p*-value[a]	Before therapy	After therapy	*p*-value[a]	
Na+ mmol/L	138.4 ± 2.51	139.2 ± 2.54	0.24	138.6 ± 2.95	139.9 ± 3.96	0.10	0.44
K+ mEq/L	4.41 ± 0.74	4.34 ± 0.66	0.71	4.21 ± 0.69	4.25 ± 0.61	0.78	0.62
Ca mg/dL	9.14 ± 0.65	9 ± 0.92	0.47	9.14 ± 0.93	9.04 ± 0.80	0.48	0.33

Data presentation as mean ± SD. Control group: RSE patients treated with conventional treatment (propofol, pentobarbital, and midazolam); Tocilizumab group: RSE patients treated with conventional treatment plus tocilizumab; (Na^+): serum sodium; (K^+): serum potassium; (Ca^{++}): total serum calcium. [a,b]: level of significance within and between groups respectively; *: statistically significant ($p<0.005$).

therapy ($p > 0.05$). After 12 weeks, there was a significant decrease in mSTESS in the control group (4.64 ± 1.22 ± 0.74 *vs.* 3.52 ± 1.08, $p < 0.0001$). Moreover, tocilizumab group showed a statistically significant decrease in mSTESS after treatment (4.68 ± 1.34 *vs.* 1.64 ± 0.90, $p < 0.0001$), by paired Student's *t*-test.

Between group comparisons by unpaired Student's *t*-test, tocilizumab group showed a greater significant reduction in mSTESS in comparison with the control group ($p < 0.0001$).

Analysis of Serum Electrolytes in the two Study Groups

Table III shows that control group showed no significant differences in all measured electrolytes after treatment as follow: serum Na^+ (138.4 ± 2.51 *vs.* 139.2 ± 2.54, $p = 0.24$), serum K^+ (4.41 ± 0.74 *vs.* 4.34 ± 0.66, $p = 0.71$), and serum Ca^{++} (9.14 ± 0.65 *vs.* 9 ± 0.92, $p = 0.47$), using paired *t*-test.

Tocilizumab group revealed that there were no significant changes in all measured electrolytes after three months of therapy as follow: serum Na^+ (138.6 ± 2.95 *vs.* 139.9 ± 3.96, $p = 0.1$), serum K^+ (4.21 ± 0.69 *vs.* 4.25 ± 0.61, $p = 0.78$), and serum Ca^{++} (9.14 ± 0.93 *vs.* 9.04 ± 0.80, $p = 0.48$), using paired *t*-test.

Between group comparisons by unpaired *t*-test, Table III shows that there was no significant difference between the studied groups.

Effect of Studied Medications on Serum Inflammatory Biomarkers

Table IV showed that control group showed a significant decrease in all measured biomarkers after treatment as follow: serum IL-6 (89.47 ± 6.005 *vs.* 37.28 ± 1.02, $p = 0.0001$), serum IL-1β (110.6 ± 5.25 *vs.* 79.24 ± 5.42, $p = 0.0001$), serum TNF- alpha (366.6 ± 14.01 *vs.* 92.76 ± 5.50, $p = 0.0001$), and serum NF-κB (17.50 ± 1.002 *vs.* 6.46 ± 0.65, $p = 0.0001$), using paired *t*-test.

Table IV. Analysis of the inflammatory markers in the two study groups.

Parameter	Control group (N=25)			Tocilizumab group (N=25)			*p*-value after therapy[b]
	Before therapy	After therapy	*p*-value[a]	Before therapy	After therapy	*p*-value[a]	
IL-6 (pg/ml)	89.47 ± 6.005	35.14 ± 4.13	0.0001*	88.99 ± 4.86	23.90 ± 2.85	0.0001*	0.0001*
TNF-α (pg/ml)	366.6 ± 14.01	92.76 ± 5.50	0.0001*	367.8 ± 13.26	57.27 ± 5.84	0.0001*	0.0001*
IL-1β (pg/ml)	110.6 ± 5.25	79.24 ± 5.42	0.0001*	110.7 ± 5.41	82.87 ± 5.08	0.0001*	0.018*
NF-κB (ng/l)	17.50 ± 1.002	6.46 ± 0.65	0.0001*	16.76 ± 1.87	3.162 ± 1.35	0.0001*	0.0001*

Data presentation as mean ± SD. Control group: RSE patients treated with conventional treatment (propofol, pentobarbital, and midazolam); Tocilizumab group: RSE patients treated with conventional treatment plus tocilizumab. IL-6: interleukin 6; TNF-α: tumor necrosis factor alpha; IL-1β: interleukin 1 beta; NF-κB: nuclear factor kappa b. [a,b]: level of significance within and between groups respectively; *: statistically significant ($p<0.005$).

Tocilizumab group revealed that there was a statistically significant decrease in all measured biomarkers after three months of therapy as follow: serum IL-6 (88.99 ± 4.86 *vs.* 23.90 ± 2.85, $p = 0.0001$), serum IL-1β (110.7 ± 5.41 *vs.* 82.87 ± 5.08, $p = 0.0001$), serum TNF- alpha (367.8 ± 13.26 *vs.* 57.27 ± 5.84, $p = 0.0001$), and serum NF-κB (16.76 ± 1.87 *vs.* 3.162 ± 1.35, $p = 0.0001$), using paired *t*-test.

Between group comparisons by unpaired *t*-test, no significant difference between the studied groups was found in their baseline values (Table IV). After three months of therapy, tocilizumab group showed a statistically significant decrease in all measured biomarkers in comparison with the control group ($p < 0.0001$) except for IL-1β ($p < 0.018$).

Discussion

RSE is typically defined as the presence of acute convulsive seizure which is resistant to at least two anti-epileptic drugs, including one non-benzodiazepine medication[18]. The term new onset refractory status epilepticus (NORSE) refers to the clinical manifestation rather than the precise diagnosis[19].

To our knowledge, this study is considered to be the first controlled and randomized trial to evaluate the effectiveness of tocilizumab in the management of RSE. Case series and case reports[13,20] are the only documentation available for tocilizumab therapy in status epilepticus patients[13,20].

Our results were in line with those published by Jun et al[20] who looked at the effects of using tocilizumab in seven adult patients who had NORSE. The usage of tocilizumab was introduced if status epilepticus persisted despite using these immune therapies, along with anesthetics and anti-seizure drugs. With the exception of one instance, administration of tocilizumab was followed by the cessation of SE following a 2-10-day latency period[20].

Cantarín-Extremera et al[13] examined the effect of tocilizumab in 2 children with RSE. They showed that both cases had elevated IL-6 levels in both serum and cerebral spinal fluid (CSF) and a positive response to treatment with tocilizum[13].

According to the results of the same clinical study, Son et al[21] reported that tocilizumab treatment caused SE to stop in 7 out of 9 patients. Early response to treatment was indicated by the fact that patients who responded to tocilizumab typically displayed clinical or electrophysiological improvement by the end of the second cycle of treatment[21].

Tocilizumab's mode of action in neurological disorders is not entirely understood. Under normal circumstances, the drug cannot penetrate the BBB, but it is speculated that it might do so if the BBB is dysfunctional[22-24]. In the setting of protracted seizures, elevated cytokine levels may facilitate BBB disruption and consequently raise tocilizumab permeability[20]. According to previous reports[25], blocking IL-6 receptors can attenuate some of the disruptive effects of IL-6 on the BBB.

Blood brain barrier (BBB) leakage has been found[23,26] to have a role in both the development of epileptogenesis and the triggering of seizures, and it has been suggested[26] that CNS inflammation caused by BBB leakage contributes to the progression of epilepsy. Recent evidence[26] that limbic seizures increase messenger RNA (mRNA) of inflammatory cytokines in mouse forebrain has suggested that cytokines are involved in the etiology of epilepsy. Additionally, seizures stimulate the release of interleukin-6 (IL-6) and tumor necrosis factor (TNF) from rat hippocampus segments, and an increase in IL-1 immunoreactivity has been discovered[26] in human epileptic tissue.

Many studies[26,27] have demonstrated that drugs with anti-inflammatory effects could have a potential role in managing epilepsy. Kilinc et al[26] demonstrated that probiotic mixture supplementation over a long period of time protects against pentylenetetrazole (PTZ)-induced seizures, inflammation, and oxidative stress in rats and causes a significant decrease in several inflammatory cytokines involved in epilepsy, including IL-1, IL-6, and IL-17A[26]. Furthermore, fluoxetine has a stronger anti-seizure impact than serotonin. Additionally, serotonin and fluoxetine, but not sumatriptan, inhibit the PTZ-induced rises in IL-1 and IL-6 levels in serum and brain tissue[27]. Kilinc et al[28] revealed that Mast cell activation ameliorates pentylenetetrazole-induced seizures in rats.

After three months of treatment with tocilizumab, there was a statistically significant decline in serum IL-6 levels in comparison with the baseline and control group in the current study. These IL-6-mediated actions were inhibited by tocilizumab, which could account for our findings. The response of the acute phase and the shift from innate to adaptive immunity were both in part mediated by IL-6[29]. Seizures could also cause an increase in the levels of IL-6 in the CSF. Therefore, it was not

clear whether IL-6 itself was correlated with the appearance of SE in NORSE. However, due to its crucial inflammatory role, which may be acting before other inflammatory cytokines, like IL-1β, that have obvious direct pro-convulsant activity, a positive feedback link between seizures and inflammation may exist[30]. Our findings support Cantarn-Extremera et al[13] findings that the striking increase in IL-6 level during the acute clinical phase and its subsequent normalization following tocilizumab administration along with clinical improvement in their patients support the involvement of neuro-inflammation in patients with refractory repeated acute seizures[13]. In the WAG/Rij (Wistar Albino Glaxo/Rijswijk) rat absence epilepsy model, Leo et al[31] showed that tocilizumab had anti-absence and anti-epileptogenic properties. In WAG/Rij rats, tocilizumab co-administration with lipopolysaccharides or IL-6 demonstrated that tocilizumab prevented the severity of absence seizures brought on by both proinflammatory agents, demonstrating a protective central anti-inflammatory-like effect[31].

In comparison to the control group, the serum IL-1β level in the tocilizumab group was significantly statistically lower. After three months of treatment, there was a statistical and significant drop in IL-1β serum level in the two study groups in comparison with the baseline value. IL-1β expression in microglia and astrocytes after seizures has been seen found in studies[32] using experimental animal models, and IL-1β itself has been shown to increase neuronal excitability. It has been hypothesized that IL-1β causes seizures by stimulating the GluN2B subunit of the NMDA (N-methyl-D-aspartate) receptors, which are located on postsynaptic cells[33]. Using epilepsy models, Postnikova et al[34] discovered that GluN2B mRNA production rose 24 hours after seizures and that alterations in NMDA receptor might impair synaptic plasticity[34]. These results implied that uncontrolled IL-1β levels reduced physiological synaptic plasticity and resulted in neuronal impairment[35]. Another investigation conducted by Roseti et al[36] revealed that the pathophysiological levels of IL-1β in temporal lobe epilepsy reduced neurotransmission of GABA (Gamma-aminobutyric acid) by up to 30% and induced generation of seizure because of neuronal hyperexcitability[36]. Additionally, cytokine IL-1β was discovered to be significantly more abundant in CSF of the pediatric epileptic patients in comparison to the control group, indicating the crucial role of cytokines in the onset and exaggeration of epilepsy[37]. Because circulating IL-1β is so unstable, levels frequently appear to be around the normal range even in the presence of highly active systemic autoinflammatory diseases in most conventional reports[38-40], that may help to explain the low levels in the two groups at the beginning of treatment. This might be because of its presence in macrovesicle or because its half-life is so short[38-40].

After three months of treatment, the serum TNF-α in the tocilizumab group was found to be statistically significantly decreased in comparison with the baseline and control group, according to the current study. These findings can be explained by the fact that there was a highly significant increase in TNF- α serum levels of epilepsy patients compared to controls. Activated astrocytes and microglia release the pro-inflammatory TNF-α cytokine. Extracellular glutamate levels are sensed by glial cells, which increase the release of TNF-α to upregulate synapses and maintain a specific neuronal excitatory level input when glutamate levels are low[41]. N-cadherin is an adhesion molecule involved in the organization and formation of inhibitory and excitatory synapses and has been previously reported[42] to be regulated by TNF-α. Additionally, it has been discovered[43] that TNF-α increases microglial glutamate release by enhancing glutaminase activity and gap junctions in microglia. Moreover, TNF-α increases the expression of AMPA (α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid) receptor, enhancing glutamatergic transmission. Increased AMPA receptors enable excessive calcium uptake, which results in neurotoxicity[43].

Our study revealed that there was a statistically significant decline in the serum NF-κB levels in the two study groups after three months of treatment in comparison with the baseline value, and the tocilizumab group serum NF-κB level was statistically lower than the control group. NF-κB is induced in glia and controls inflammatory processes which are responsible for exacerbation of inflammation-induced neurodegeneration[44]. Additionally, NF-κB plays a fundamental role as a key signal transducer at the blood-brain barrier level, influencing cellular permeability, intracellular trafficking, and endocytosis[45]. Stimulation of NF-κB signaling has been involved to activate inflammatory target proteins, such as PGE2 and COX-2 release that leads to cerebral vascular inflammation[46,47].

According to the findings of the present study, mSTESS was found to be significantly lower in the tocilizumab group in comparison with the control group. After three months of treatment compared to the baseline value, the mSTESS in

the two study groups decreased statistically significantly. Tocilizumab and antiepileptic medications may have reduced serum inflammatory cytokines, which may be the cause of this effect.

Regarding serum electrolytes, our results match with other studies[48,49] which reported there were no significant changes in serum potassium and sodium levels in epileptic patients. The study by Reynolds[48] revealed that epileptic patients with depression have higher serum sodium levels compared with healthy controls. Further studies are required to validate these results concerning serum electrolytes.

There were two clinical studies[13,20] that investigated the effect of tocilizumab in epilepsy, but they had small sample size, and they did not measure other inflammatory biomarkers. Our study had larger sample size with 25 patients in each group and measured inflammatory cytokines such as IL-6, IL-1β, TNF-α, and NF-κβ. Also, serum electrolytes and mSTESS scale were measured.

Strenghts and Limitations

To the best of our knowledge, this is the first clinical study to assess the role of the inflammatory cytokines in RSE and their change with treatment with tocilizumab. The current study has many strengths, including: the prospective nature of the study, being the first randomized controlled study with the two limbs are cases with RSE to illustrate the value of tocilizumab, and the prospective nature of the study itself.

The study, however, has some drawbacks. It is a single center study with a relatively small sample size, to start with. The short-term and long-term follow-up are also lacking. The upcoming studies should provide a thorough discussion of these drawbacks.

Conclusions

Depending on the findings of our study, it can be deduced that tocilizumab significantly reduces the frequency and severity of seizures in people with RSE. Additionally, the same medication significantly decreased serum levels of inflammatory and proinflammatory cytokines, which may have a fundamental role in the pathophysiology of epilepsy.

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Availability of Data and Materials

Data and materials in this study are available and transparent from the authors themselves upon reasonable request, as long as the claims are matched with field standards transparency.

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Conflict of interest

There is no conflict of interest declared by any of the authors.

Informed Consent

Written informed consent was obtained from every study subject.

Ethical Approval

The National Research Ethics Committee (Tanta University Ethical Committee, approval code: 34233/11/20) gave its approval for the study. The study complied with the 1964 amendments to the Helsinki Declaration's Ethical standards.

Authors' Contribution

All authors contributed significantly and equally to the work, whether it be in the conceptualization, study design, implementation, data collection, analysis, and interpretation, or in all of this area.

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