Clinical Research

Tigecycline reduced tumor necrosis factor alpha level and inhospital mortality in spontaneous supratentorial intracerebral hemorrhage

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ABSTRAK

Latar belakang: *Luaran perdarahan intraserebral spontan supratentorial (PISS) masih buruk. Respons inflamasi sekunder akibat cedera otak dan prosedur bedah diyakini sebagai penyebabnya. Penelitian ini bertujuan untuk mengetahui aktivitas antiinflamasi tigesiklin dengan menghitung kadar TNF-α, dan efek neuroproteksi yang dicerminkan oleh angka kematian di rumah sakit.*

Metode: *Pasien dengan PISS yang akan dilakukan evakuasi hematoma, dirandomisasi untuk jenis antibiotik profilaksis tigesiklin (n=35) atau fosfomisin (n=37). Pada semua subjek diukur kadar TNF-α sebelum pembedahan serta hari ke-1 dan ke-7 pascabedah. Pada hari ke-7 dilakukan pemeriksaan* CT Scan *ulang.* Skor Glasgow outcome scale *(GOS) dan lama rawat dicatat pada saat keluar rumah sakit. Data dianalisis dengan uji Mann-Whitney atau uji kai kuadrat. Efektivitas klinis relatif dinilai dengan menghitung number needed to treat (NNT).*

Hasil: *Didapatkan perbedaan bermakna pada proporsi subjek yang mengalami penurunan kadar TNF-α pada kelompok tigesiklin dibanding fosfomisin pada hari ke-7 pascabedah (62% vs 29%, p=0,022). Pengurangan edema pacsa operasi berbeda tidak bermakna pada kedua kelompok (86% vs 80%, p=0,580). Tigesiklin menunjukkan efektivitas klinis mengurangi luaran buruk (GOS ≤ 2 (20% vs 38% ; p=0,096; OR=0,41; NNT=6) dan inhospital mortality (17% vs 35%; p=0,083; OR=0,49; NNT=5). LOS ≥ 15 hari (40% vs 27%; p=0,243; OR=1,81; NNT=8).*

Kesimpulan: *Tigesiklin memiliki kemampuan antiinflamasi dan neuroproteksi, serta memperbaiki luaran klinis pada PISS yang dilakukan evakuasi hematoma.*

ABSTRACT

Background: The outcome of patients with spontaneous supratentorial intracerebral hemorrhage (SSICH) is unsatisfactory. Inflammatory response secondary to brain injury as well as those resulted from surgical procedure were considered responsible of this outcome. This study was intended to elucidate the anti-inflammatory activity of tigecycline by measuring TNF-α level and its neuroprotective effect as represented by inhospital mortality rate.

Methods: Patients with SSICH who were prepared for hematoma evacuation were randomized to receive either tigecycline (n=35) or fosfomycine (n=37) as prophylactic antibiotic. TNF-α level was measured in all subjects before surgery and postoperatively on day-1 and day-7. A repeated brain CT Scan was performed on postoperative day-7. The Glasgow outcome scale (GOS) and length of stay (LOS) were recorded at the time of hospital discharge. Data were analyzed using Mann-Whitney and Chi square test. Relative clinical effectiveness was measured by calculating the number needed to treat (NNT).

Results: There was a significant difference regarding the proportion of subject who had reduced TNF-α level on postoperative day-7 between the groups receiving tigecycline and fosfomycine (62% vs 29%, p=0.022). Decrease brain edema on CT control (86% vs 80%, p=0.580). Tigecycline administration showed a tendency of better clinical effectiveness in lowering inhospital mortality (17% vs 35%; p=0.083; OR=0.49; NNT=5) and worse clinical outcome / GOS ≤ 2 (20% vs 38% ; p=0.096; OR=0.41; NNT=6). LOS ≥ 15 hari (40% vs 27%; p=0.243; OR=1.81; NNT=8).

Conclusion: Tigecycline showed anti-inflammatory and neuroprotective activities. These activities were associated with improved clinical outcome in patients with SSICH after hematoma evacuation.

Keywords: inhospital mortality, SSIH, tigecycline, TNF-α

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The mortality rate at day-7 post-spontaneous supratentorial intracerebral hemorrhage (SSICH) reaches 34.6%, and becomes 50.3% in the first month of treatment.[1] Unfortunately, patients who can survive will suffer neurological disabilities with poor quality of life.[2] Referring to a population-based study in the United States, the inhospital mortality for SSICH patients undergoing hematoma evacuation is 27.2%, and those without hematoma evacuation is 32%.[3] Among the causes of unsatisfactory clinical outcome, is secondary brain injury as the consequence of untreated inflammatory response,[4] as well as additional injury/inflammation due to surgical procedure[5] that can lower the benefit of hematoma evacuation.[6] Until now, SSICH management is still considered unsatisfactory.[4,7]

Considering that inflammation has pivotal role in the development of brain injury, controlling inflammatory response would be one of the promising treatment in SSICH management.[8] However, administration of steroidal anti-inflammatory drugs is proven ineffective, as well as increased infection as its complication.[9] Animal studies have shown anti-inflammatory and neuroprotective activities of tetracycline derivatives.[10,11] Among several inflammatory markers, tumor necrosis factor–alpha (TNF-α) has been known as a good marker of brain injury in SSICH patients.[12,13] Low or reduced TNF-α level has been proven to be associated with better outcome of intracranial hemorrhage (ICH) in animal study[14] as well as in human subjects.[13,14] A new tetracycline derivative, tigecycline, reduces TNF-α level and shows neuroprotective activity in animal studies.[15] However, anti-inflammatory and neuroprotective activities of tigecycline in SSICH patients has not been well studied. This study was aimed to evaluate whether tigecycline could reduce TNF-α level as well as reduce brain injury in human subject with SSICH who underwent hematoma evacuation.

METHODS

This was a randomized controlled trial (RCT) to elucidate the anti-inflammatory and neuroprotective activities of tigecycline in SSICH patients who underwent hematoma evacuation at 11 hospitals in Jakarta. Randomization was carried out by block system, with the size of four. Patients who were prepared for hematoma evacuation over a period of August 2012 to November 2014, were randomized to receive either 100 mg tigecycline or 2 g fosfomycine as prophylactic antibiotic. Antibiotic was administered intravenously 30 minutes before the surgery. The protocol of this study has been approved by Medical Ethics Committee, Faculty of Medicine, Universitas Indonesia/Cipto Mangunkusumo Hospital (No. 493/PT02.FK/ETIK/2012).

SSICH diagnosis was made based on clinical condition and the results of CT scan imaging. Surgical intervention was indicated if the hematoma volume was ≥30 mL and the clinical finding showed a decreased level of consciousness. The volume was measured by following formula: ½ (A × B × C) with A as the greatest diameter of hematoma on CT scan, B as the vertical diameter (90°) to A, and C is the amount of slices on the CT scan by which the hematoma were seen (counted in centimeters).[16] The surgical procedures were hematoma evacuation with craniotomy or craniectomy. Sample size was calculated by following formula:[17]

$$n_1 = n_2 = \frac{(Z_\alpha\sqrt{2PQ} + Z_\beta\sqrt{P_1Q_1 + P_2Q_2})^2}{(P_1Q_1)^2}$$

By taking P_1=0.4 as the proportion of the effect of standard treatment, and P_2=0.7 as proportion of the effect of tigecyclin, α=0.05 and β=0.1, a minimum of 22 subjects were needed in each group.

Subject characteristics and outcomes were recorded as numeric and/or categorical data. The subject characteristics were age, sex, Glasgow coma scale (GCS) score, hematoma volume and its mass effect, onset and duration of surgery, size of corticotomy, systemic factors which affected prognosis, as well as the plasma level of TNF-α before surgery. The outcomes of our study were TNF-α plasma level on postoperative day-1 and day-7, changes in brain edema level on CT scan, glasgow outcome scale (GOS) and length of stay (LOS) at the time of hospital discharge.

Brain edema level was measured by perihematoma hypodensity thickness on the CT scan before surgery and perihematoma hypodensity thickness/former hematoma area

in milimetres. The changing level of edema was measured by comparing the degree of brain edema on CT scan pre-surgery with repeated CT scan at day seven post-surgery. The plasma level of TNF-α was measured using ELISA, the reagent for TNF-α is produced by DRG in the United States. Normal value of TNF-α according to the manual book is 4.5–12.5 pg/mL, by which the abnormal value of TNF-α was determined if it is more than 12.5 pg/mL. Data from all of the randomized subjects were included in the analysis. Numeric data were presented as mean value and data distribution, categorical data were presented as proportion (%). Comparison of numeric data between the two groups was done by using Mann–Whitney test, meanwhile categorical data were tested using Chi square test. The statistical level of significance was set at $p<0.05$. Relative clinical effectiveness was calculated using relative risk (RR), odds ratio (OR), and number needed to treat (NNT).

RESULTS

The sample size of our study was 72 subjects, including 35 subjects in the treatment group (tigecycline) and 37 subjects in the control group (fosfomycine).

Subject characteristics

Age mean, GCS, and hematoma volume of subjects in both groups were equal. Risk factors for poor outcome such as elderly age (over 60 years), female, history of hypertension, and abnormal mean arterial pressure (MAP) were found greater in the tigecycline group. On the contrary, mid line shifting (MLS) of ≥10 mm, which is a sign of brain herniation, was greater in the fosfomycin group (Table 1).

Changes of TNF-α plasma level

Changes of the TNF-α level were analyzed based on three aspects, i.e. mean value, proportion of subject with normal level, and with decreased level. Changes of brain edema were measured by comparing the degree of brain edema using CT scan figures before and after surgery. Changes of TNF-α plasma level are shown in Table 2.

On post operative day one, TNF-α levels in the tigecycline group from all the three aspects were improved. Median was decreased from 14.5 (3.1–95.8) pg/mL to 12.4 (4.3–85.0) pg/mL, proportion of subjects with normal level

Table 1. Subject characteristic

	Group	
	Tigecycline	Control
Subject amount, n(%)	35 (49)	37 (51)
Gender		
Male, n(%)	19 (54)	26 (70)
Female, n(%)	16 (46)	11 (30)
Age		
Mean (SD)	52.8 (9.1)	51.8 (8.9)
>60 years, n(%)	9 (26)	5 (14)
Poor Risk Factors		
Female, n(%)	16 (46)	11 (30)
Elderly (age >60 years)	9 (26)	5 (14)
Abnormal MAP, n(%)	32 (91)	29 (78)
Hypertension, n(%)	32 (91)	32 (86)
GCS		
Median (min–max)	9 (6–13)	9 (5–14)
Hematome volume		
Median (min–max) mL	45 (30–90)	50 (30–90)
Volume ≥60 mL, n(%)	21 (60)	22 (59)
Midline shift		
Median (min–max) mm	5 (0–15)	5 (0–20)
≥10 mm, n(%)	7 (20)	12 (32)
Brain edema		
Moderate degree, n(%)	17 (48)	15 (40)
TNF-α level before intervention		
Median (min–max) pg/mL	14.5 (3.1–95.8)	15.9 (3.9–64.6)
Normal Level, n(%)	12 (34)	14 (41)
Leukocyte level		
Mean (SD) /µL	14.17 (3.48)	15.48 (5.54)
Surgical onset		
Less than 24 hours, n(%)	22 (63)	27 (73)
More than 48 hours, n(%)	6 (18)	6 (16)
Surgical duration		
Median (min–max) minutes	160 (60–360)	160 (60–420)
Corticotomy diameter		
Median (min–max) mm	10 (5–30)	15 (3–30)

MAP= mean arterial pressure; GCS= Glasgow coma scale; TNF-α= tumor necrosis factor–alpha; SD= standar deviation

Table 2. Changes of the TNF-α and brain edema

	Tigecycline group	Control group	p	RR	OR
TNF-α –Day 1					
Median (min-max) pg/mL	12.4 (4.3-85)	16.9 (4.0-84.4)	0.247		
Normal level, n (%)	19 (56)	11 (37)	0.124	0.70	0.46
Decreased level, n (%)	22 (65)	12 (43)	0.085	0.61	0.41
TNF-α – Day 7					
Median (min-max) pg/mL	15.1 (2.1–60.5)	15.9 (2.6–97.0)	0.346		
Normal level, n (%)	13 (48)	11 (44)	0.764	0.85	0.85
Decreased level, n (%)	16 (62)	7 (29)	0.022	0.54	0.26
Brain edema degree					
Decreased, n (%)	24 (86)	20 (80)	0.580	0.7	0.68

TNF-α= tumor necrosis factor–alpha

Table 3. Glasgow outcome score and length of stay

	Group		RR	OR	NNT	p
	Tigecycline n(%)	Control n(%)				
GOS						
GOS 1	6 (17)	13 (35)	0.49	0.38	5	0.083
GOS ≤2	7 (20)	14 (38)	0.53	0.41	6	0.096
LOS ≥15 days	14 (40)	10 (27)	1.48	1.81	8	0.243

GOS= Glasgow outcome score; LOS= length of stay; RR= relative risk; OR = odds ratio; NNT= number needed to treat

of TNF-α was increased from 34% to 44%, and the proportion of subjects with decreased level was greater compared to the proportion of subjects having increased level (65% vs 35%). On the contrary, in the control group, median was increased from 15.93 (3.9–64.6) pg/mL, the proportion of subjects with normal level of TNF-α was decreased from 41% to 37%, and the proportion of subjects with decreased level was lower compared to the proportion of subjects having increased level (43% vs 57%). The changes of TNF-α level between the two group were statistically not significant. However, the data showed that the tigecycline reduced the TNF-α level, as well as the surgical procedure increased the TNF-α level.

On post operative day seven, the proportion of subjects with normal TNF-α level in the tigecycline group was increased from 34% (before tigecycline administration) to 48%. However, it was decreased compared to post operative day one from 56% to 48%. The proportion of subjects with normal TNF-α level in the control group was also increased from 41% (before fosfomycin administration) to 44% The differences of the proportion of normal level of TNF-α between the two group were statistically not significant (chi square resulted p=0.764 (95% CI=0.560-1.531). However, the differences of the proportion of subjects with reduced TNF-α level between the two group were statistically significant (62% vs 29%; p=0.022; 95% CI=0.313–0.941).

Short-term clinical outcome

Clinical outcomes were measured with GOS and LOS at the time of hospital discharge. GOS was quite applicable for measuring clinical outcome post spontaneous supratentorial intracerebral hemorrhage (SSIH), in spite of its rough characteristic. GOS consists of cognitive evaluation, disability, and social function.[18] Therefore, GOS measurement at the time of hospital discharge is a prognostic marker for clinical outcome at six months.[19] To present short-term clinical outcome, the study also included LOS (Table 3).

DISCUSSION

Intracerebral hematoma causes mass effects in the form of increased intracranial pressure and brain herniation, as well as a source of neurotoxic substances that damage the brain cells. All causes may affect each other and lead to unsatisfactory clinical outcomes. A previous study showed that hematoma volume is a powerful predictor of the 30-day mortality rate.[20] However, for most patients the usefulness of surgery is not well established. Hematoma evacuation can indeed reduce intracranial pressure and prevent brain herniation, as well as producing a significant amount of neurotoxic substances; however, the evacuation surgical procedure has been believed to cause additional brain injury.[5,6]

Surgical evacuation of hematoma for clinical deteriorating patients can be considered as a life-saving procedure.[21] It seems that there is no particular type of treatment, either surgical or medical, that can provide satisfactory clinical outcomes. Recent advances from basic science and pre-clinical studies regarding treatment strategies for SSICH show the role of cell replacement therapy, endogenous neurogenesis, and neuroprotection.[22] Neuroprotective agents were believed to provide brain cells protection from secondary brain injury that causes sustainable brain damage, as well as to improve neurological outcomes.[23] On animal models, anti-inflammatory drugs, as well as minocycline (the prototype of tigecycline), have been demonstrated to have neuroprotective activity and therapeutic effects.[24,25] This study was intended to elucidate the anti-inflammatory activity of tigecycline by measuring TNF-α level, and its effects on clinical outcomes by measuring the inhospital mortality rate and/or GOS score.

The findings in the present study confirm that tigecycline reduced TNF-α plasma level. This study noted a significant difference on the proportion of subjects with reduced TNF-α plasma level on the seventh day after administration/surgery between those in the tigecycline and fosfomycine group. About 62% of subjects in the tigecycline group showed reduced TNF-α plasma level, while only 29% of subjects in the control (fosfomycine) group showed reduced TNF-α plasma level.

These results are similar with the results of previous studies conducted in animal,[15] which indicate that tigecycline can reduce TNF-α plasma level. Since TNF-α is a proinflammatory cytokine,[26] the reduced plasma level could be assumed as a reflection of less inflammation process. Inflammatory response plays an important role as a cause of brain injury, either due to SICH or surgical procedures.[5,8] On the other hand, the non-steroidal anti-inflammatory drugs (rosiglitazone) and tigecycline have been shown to reduce the inflammatory response.[5,15]

Reduced TNF-α level or activity,[14,27] or moderate level of TNF-α[13] is associated with better clinical outcomes in intracerebral hemorrhage. The results of present study are consistent with the statements above. Inhospital mortality rate was reduced by 18%, i.e. the tigecycline group has recorded inhospital mortality rate by 17%, compared with 35% in the control (fosfomycine) group. A population-based study in US showed that the inhospital mortality rate was 27.2%.[28] Although statistically it was not significant, but it was clinically important. Subjects who received a single dose of 100 mg tigecycline would experience reduced risk of inhospital mortality by 2.6 times. In addition to the inhospital mortality rate, subjects in the tigecycline group also experienced better GOS score by 2.4 times.

This study protocol established a re-examination of CT scan on the seventh day after surgery. Considering that hematoma volume of more than 30 mL is associated with early death[29] and most of the patients (89%) die before the seventh day, re-examination of the CT scan on the seventh day become one of the limitations in this study. Assesment on adequacy of hematotoma evacuation and/or the presence of rebleeding can not be done. Other limitations in this study include the small sample size (72 subjects), a slight difference in the facilities, especially in intersive care unit and wards that exist among the hospitals that had become the study site and a slight difference in surgical procedure performed by different neurosurgeons. Although there were limitations, several factors which were believed to affect the outcomes, i.e. age,[30] gender,[31] GCS on admission and volume of the hematomawere comparable in both groups.[32,33]

In conclusion, a single dose of 100 mg tigecycline treatment before surgical procedure for hematoma evacuation of SSICH can significantly reduce TNF-α plasma level. Tigecycline treatment also shows lower inhospital mortality rate, which is clinically important. Further studies are necessary to asses the effectiveness and efficiency of tigecycline treatment for ICH by considering various limitations found in this study.

Conflict of interest

The author affirms no conflict of interest in this study.

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