*Research Article*

# Evaluation of the Neuroprotective Effect of Erythropoietin in Patients with Moderate Head injury

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## Abstract
**Background:** EPO is a potentially attractive neuroprotective agent following TBI. In experimental models, EPO has improved outcome after TBI, subarachnoid hemorrhage, stroke, global ischemia, and other CNS disorders. **Methods:** 40 patients (according to sample size) of either sex, aged between 18-60 years old with acute moderate TBI (GCS 9-12) admitted to ICU within 8 hours, were enrolled in this prospective randomized double blind study. 20 patients received Epo as intravenous infusion of 40,000 IU/50ml over 30 min on 3 consecutive days after trauma, starting within 8 hr of trauma in addition to the standard therapy and the rest received standard therapy and Sodium Chloride (placebo) 50 ml over 30 min on 3 consecutive days after trauma, starting within 8 hours of trauma. **Results:** Regarding to GOS we found that 5 (25%) patients in EPO group and 16 (80%) patients in control group get score 4 (moderate disability) and 15 (75%) patients in EPO group and 4 (20%) patients in control group get score 5 (good recovery). After 6 months 2 (10%) patients in EPO group and 13 (65%) patients in control group get score 4. and 18 (90%) patients in EPO group and 7 (35%) patients in control group get score 5. **Conclusion:** Early administration of high dose of EPO (40.000 IU) within eight hours of trauma for three successive days improved patient outcome by prevention of secondary brain insult through its anti-apoptotic, anti-inflammatory and anti-oxidant effects.
**Keywords:** Neuroprotective effect - Erythropoietin - Moderate head injury.

## Introduction
Traumatic brain injury (TBI) is a growing epidemic throughout the world and may present as major global burden in 2020.This burden has even found to be very high in some countries. In addition, TBI not only increases the overall morbidity and mortality, but also, imposes substantial impact on quality-of-life. Moreover, patients who survived from the primary insult, some may still have a long-term disability. In addition, the most of the victims are of younger age group[1,2]. The clinical outcomes of TBI are directly related to the severity of the primary and secondary lesions sustained by the patient. Primary lesions are those related to the initial impact (lacerations, contusion, fractures, and diffuse axonal injury). Secondary lesions are those which developed after the initial trauma, including hematomas, edema, and pathological processes cascades that cause ischemia resulting in a worsening of the clinical condition[3]. Exogenous erythropoietin (EPO) provides neuroprotection. Studies have demonstrated that EPO treatment is capable of reducing blood–brain barrier (BBB) breakdown and edema after TBI. These results suggest that multiple pathways may be involved in the mechanism of EPO neuroprotection, including its effects on EPOR in both neural and non-neural cells in the brain[4,5]. Concentrations of S-100β protein, the β subunit of a calcium binding protein present mainly in glial and Schwann cells, increase in human blood and cerebrospinal fluid after a wide range of diseases or conditions leading to brain damage. Increased concentrations in blood and cerebrospinal fluid have been reported in patients with traumatic brain injury [6].

## Patients and methods

After obtaining approval of the local ethics committee in El-Minia university hospital and written informed consent from the first degree relatives of the patients prior to entry into the study, 40 patients (according to sample size) of either sex, aged between 18-60 years old with acute moderate TBI (GCS 9-12) admitted to ICU within 8 hours, were enrolled in this prospective randomized double blind study. This study carried out at the (ICU), of El-Minia University Hospital between February 2012 to February 2014.

***Inclusion criteria:***

- Patients are expected to stay ≥ 48 hours
- Patients have hemoglobin not exceeding the upper limit of the applicable normal reference range in clinical use which is 16 g/dl in male and 14 g/dl in female

***Exclusion criteria:***

We excluded in our study patients with the following criteria:

- GCS < 9 or >12.
- History of deep venous thrombosis, pulmonary embolism or other thromboembolic event.
- A chronic hypercoagulable disorder, including known malignancy.
- Treatment with EPO in the last 30 days.
- First dose of study drug unable to be given within 8 hours of primary injury.
- Pregnancy or lactation or 3 months post-partum.
- Uncontrolled hypertension.
- Acute myocardial infarction.
- Known sensitivity to mammalian cell derived products.
- Hypersensitivity to the active substance or to any of the additives.
- Pure red cell aplasia.
- End stage renal failure (receives chronic dialysis).
- Severe pre-existing physical or mental disability or severe co-morbidity that may interfere with the assessment of outcome.
- Spinal cord injury.

***Initial assessment:***

Patients were assessed immediately after admission by a primary survey according to ABCDE protocol to exclude any life threatening conditions. A blood sample was obtained for complete blood cell count and chemistry, urinalysis, urinary toxicology screen, and a beta-human chorionic gonadotropin value in all females of childbearing age. Cross-matching 2-4 units of RBCs were prepared, depending on the severity of the trauma and shock. Urinary and gastric catheters were inserted. Temperature, ECG, blood pressure and oxygen saturation monitors were applied.

***Secondary Survey:***

Detailed patient history was taken, for detection of medical/surgical history and events leading up to trauma. Detailed physical examination of the patient was done.

***Grouping criteria:***

The patients included in the study were allocated randomly to 2 groups 20 patients in each group.

***Group I:*** patients with GCS (9-12) received Epo as intravenous infusion of 40,000 IU/50ml over 30 min on 3 consecutive days after trauma, starting within 8 hr of trauma in addition to the standard therapy (head position, sedation, dehydrating measures, temperature control, euvolemia, seizer control, parenteral and enteral feeding and antibiotics).

***Group (II):*** received standard therapy and Sodium Chloride (placebo) 50 ml over 30 min on 3 consecutive days after trauma, starting within 8 hours of trauma.

**Parameters assessed:**

1- ***General assessment:***

Continuous ECG monitoring.

Heart rate (beat/min), MAP (mmHg) and Oxygen saturation on admission, 30 minutes, 1 hour, 2, 4, 12, 24, 36, 48, 60, 72, 84, 96 hours after admission.

Central venous pressure monitoring.

Urine output monitoring/hour.

Acute Physiology and Chronic Health Evaluation (APACHEII) score[7,8].

Sequential Organ Failure Assessment (SOFA).

2- **Neurological assessment:**

**(GCS)**[9]: GCS will be recorded on admission, 30 minutes, 1 hour, 2, 4, 12, 24,

36, 48, 60, 72, 84, 96 hours and 120 hours after admission.

**Glasgow Outcome Scale (GOS)** [10].

**3- Radiological assessment:**

CT scan is done for diagnosis and for follow up to exclude new lesion or expanding hematoma. Abdominal sonar to exclude associated trauma. Plain X ray on the body to exclude associated fractures.

**4- Laboratory investigations:**

**Routine ICU investigation:** Complete blood count, renal function tests, liver function tests, blood sugar, arterial blood gases and serum electrolytes.

**S100-β:** Venous blood samples were collected on admission, 24 hours and 48 hours of admission for detection of S100β by (ELIZA).

**Laboratory work:**

On admission 2ml of venous blood withdrawn and placed in plain tube which left to be clotted at room temperature for 30 minutes, and then centrifuged at 300 rpm for 10 minutes. The separated serum was kept frozen at -20˚C for assay of S100β protein by ELIZA. Then 2 ml of venous blood were collected after 24 hrs and after 48 hrs of admission and separated serum was used also for assay of S100β protein by ELIZA.

**Statistical analysis:**

Data were collected from patients on admission and during ICU stay. Data were collected, entered and processed on compatible computer using SPSS version 21 (Chicago, IL, USA).

## Results

Regarding the demographic and clinical characteristics of the studied patients, there was no significant statistical difference associated with age, sex, APACHE II score at admission (table 1).

**Table (1): Demographic data of the study groups**.

| Characters | | Group (I) N=20 | Group (II) N=20 | P-value |
|---|---|---|---|---|
| **Age (years)** | **Range**<br>**Mean±SD** | 18-38<br>25.6±6.32 | 18-40<br>26.6±7.05 | 0.6 |
| **Sex** | **Male**<br>**Female** | 19 (95%)<br>1 (5%) | 18 (90%)<br>2 (10%) | 0.5 |
| **APACHE Score** | **Range**<br>**Mean±SD** | 6-19<br>10.7±3.13 | 6-14<br>11.5±3.12 | 0.4 |

**Haemodynamics changes:** There was no significant difference in the mean HR (figure 1), mean arterial blood pressure (mmHg) (MAP) (figure 2) and arterial oxygen saturation ($Spo_2$) (figure 3) between the two groups all over the times of the study.

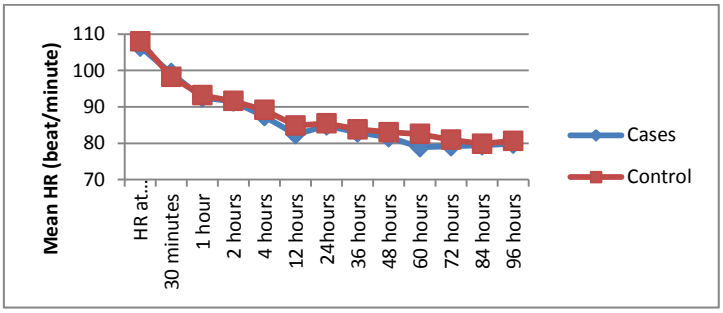


**Figure (1): Heart rate changes in the study groups.**

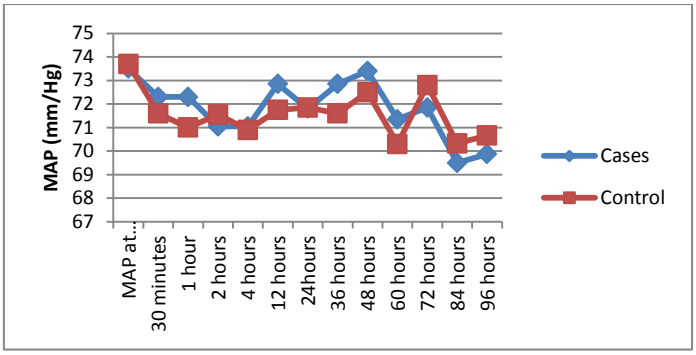


**Figure (2): Mean arterial blood pressure changes in the study groups.**

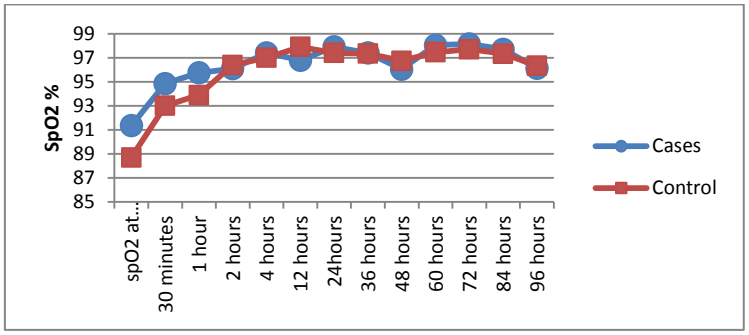


**Figure (3): Arterial oxygen saturation changes in the study groups.**

**Changes in sofa scale:** When comparing the two groups as regards to Sofa scale it was found that there was no significant difference between the two groups at admission data, but there was significant difference between the two groups on the first, second, third, fourth and fifth days of admission as shown in table (2).

**Table (2): Changes in sofa score in the study groups (Mean ±SD).**

| Time | Group (I) N=20 | Group (II) N=20 | P- value |
|---|---|---|---|
| **On admission** | 4.35±1.26 | 5.00±1.29 | 0.1 |
| **1st day** | 3.25±1.71 | 4.60±1.78 | 0.02* |
| **2nd day** | 3.00±2.22 | 4.60±2.43 | 0.03* |
| **3rd day** | 2.73±2.49 | 4.70±2.51 | 0.01* |
| **4th day** | 2.33±0.51 | 3.14±1.18 | 0.007* |
| **5th day** | 2.12±0.51 | 3.33±2.16 | 0.01* |

*= significant (P-value<0.05)

**Changes in Glasgow coma scale**: Regarding to changes in GCS there was no significant difference between the two groups at admission, 30 minutes, one hr. and 24 hrs. after admission, but there was significant difference between the two groups as regarding to the following data 36, 48, 60, 72, 84, 96 and 120 hrs. of admission.Within group, the GCS gradually increases and there was significant difference between the admission data and the following data starting at 36 hrs. in group (I) and there was no significant difference between the admission data and the following data in group (II) as shown in table (3).

**Table (3): Changes in Glasgow coma scale in the study groups (Mean ±SD).**

| Time | Group (I) N=20 | Group (II) N=20 | P- value |
|---|---|---|---|
| **On admission** | 9.30±0.65 | 9.15±0.36 | 0.3 |
| **30 min after admission** | 9.00±1.55 | 9.15±0.36 | 0.6 |
| **1 hour** | 9.05±1.79 | 8.70±0.92 | 0.4 |
| **24 hours** | 9.50±1.60 | 8.60±1.31 | 0.06 |
| **36 hours** | 9.95±1.53# | 8.50±1.67 | 0.007* |
| **48 hours** | 10.65±2.01# | 8.50±2.39 | 0.004* |
| **60 hours** | 10.80±2.50# | 8.20±2.66 | 0.003* |
| **72 hours** | 11.35±2.81# | 8.40±3.18 | 0.004* |
| **84 hours** | 10.71±3.63# | 10.00±3.09 | 0.007* |
| **96 hours** | 11.14±3.93# | 9.66±3.61 | 0.04* |
| **120 hours** | 11.60±3.78# | 8.50±4.04 | 0.02* |

*=P-value <0.05 as compared between the two groups

# =P-value < 0.05 as compared to the admission value in the same group

**Hemoglobin changes (Hb):** We observed that there was no significant difference between the studied groups as regard to (Hb) concentration on the first day of admission, but on the third day of admission (Hb) concentration was significantly increased in EPO group as it was (13.98±1.05) when it was compared with control group as it was (11.75±1.28) with P-value= 0.001. When we compared (Hb) concentration on the fifth day of admission between the two groups we found that (Hb) concentration was significantly increased in EPO group as it was (13.12±0.63) when it was compared with control group as it was (10.65±0.17) with P-value= 0.0001as shown in figure (4).

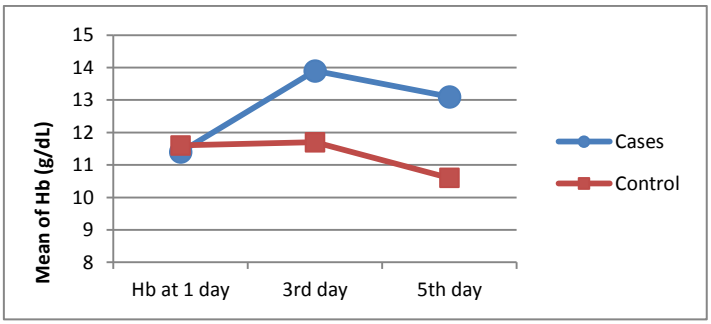


**Figure (4): Hemoglobin changes in the study groups.**

**Hematocrit changes (Hct):** When we compared (Hct) percentage between the two groups we found that there was no significant difference between the studied groups as regard to (Hct) percentage on the first day of admission, but on the third day of admission (Hct) percentage was significantly increased in EPO group as it was (38.86±3.22) when it was compared with control group as it was (35.40±1.14) with P-value= 0.0001. As regard to (Hct) percentage on the fifth day of admission we found that it was significantly increased in EPO group as it was (38.20±1.48) when it was compared with control group as it was (33.25±2.21) with P-value= 0.005 as shown in figure(5).

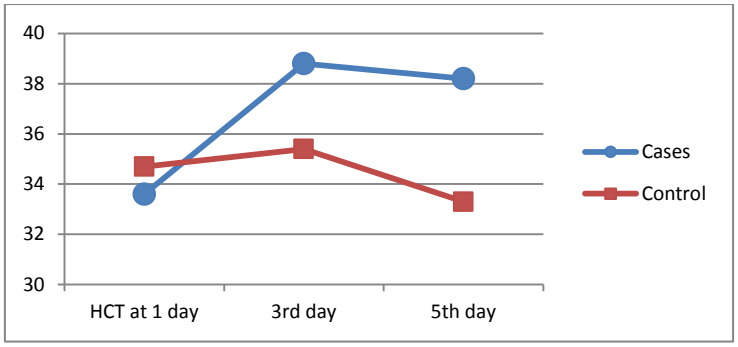


**Figure (5): Changes in Hct concentration in the study groups.**

**Changes in the level of serum S 100β (pg/ml) in the study groups:** Comparing the level of serum S100β between two groups there was no significant difference in admission value and the value measured 24hrs after admission between the two groups.There was significant difference in the values measured 48 hrs after admission between the two groups.The concentration of S100β was decreased from admission time to 24 hrs and 48hrs after admission in group (I) with significant statistical difference as shown in table (4). But we found no significant statistical difference when we compared concentration of S100β at admission with its concentration 24 hrs and 48 hrs after admission in group (II).

**Table (4): Changes in the level of serum S 100β (pg/ml) in the study groups (Mean ±SD).**

| Time | Group (I) N=20 | Group (II) N=20 | P- value |
|---|---|---|---|
| **On admission** | 380.60±284.73 | 389.40±330.26 | 0.9 |
| **After 24 hrs.** | 285.90±208.59# | 375.10±314.33 | 0.2 |
| **After 48 hrs.** | 168.15±98.41# | 379.60±318.53 | 0.007* |

*=P-value <0.05 as compared between the two groups

# =P-value < 0.05 as compared to the admission value in the same group

**Change of Glasgow outcome scale (GOS):**Regarding to GOS we found that 5 (25%) patients in group (I) and 16 (80%) patients in group (II) get score 4 (moderate disability) and 15 (75%) patients in group (I) and 4 (20%) patients in group (II) get score 5 (good recovery). After 3 months 4 (20%) patients in group (I) and 15 (75%) patients in group (II) get score 4, and 16 (80%) patients in group (I) and 5 (25%) patients in group (II) get score 5. After 6 months 2 (10%) patients in group (I) and 13 (65%) patients in group (II) get score 4. and 18 (90%) patients in group (I) and 7 (35%) patients in group (II) get score 5.As shown in table (5), (6).

**Table (5): Glasgow outcome scale in group (I)**

| GOS | Group (I) No of patients (%) | | |
|---|---|---|---|
| | At discharge | 3 month | 6 months |
| **Score (4)** | 5 (25%) | 4 (20%) | 2(10%) |
| **Score (5)** | 15 (75%) | 16 (80%) | 18 (90%) |

**Table (6): Glasgow outcome scale in group (II)**

| GOS | Group (II) No of patients (%) | | |
|---|---|---|---|
| | At discharge | 3 months | 6 months |
| **Score (4)** | 16 (80%) | 15 (75%) | 13(65%) |
| **Score (5)** | 4(20%) | 5(25%) | 7 (35%) |

Score (4) = moderate disability
Score (5) = good recovery

**Correlation between GOS and S100β on admission, 24 hrs. and 48 hrs.** Correlation between GOS and S100β level in EPO group was done by Spearman's rho correlation test and showed that there was weak negative correlation between S100β measured on admission,24 hours and 48 hours after admission and GOS after 6 months with r value= -.275, -.318, -.072 respectively as shown in figures (6), (7) and (8).

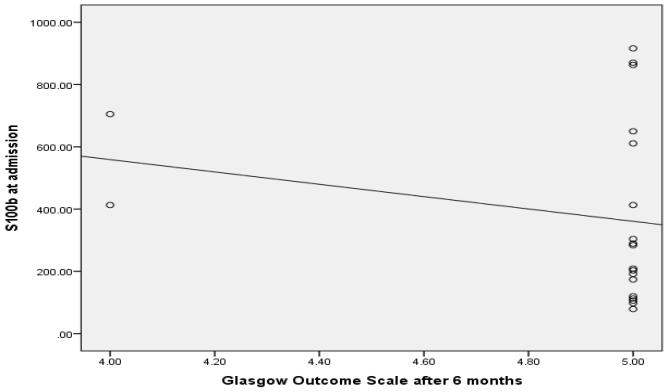


**Figure (6): Correlation between GOS and S100β on admission in group (I).**

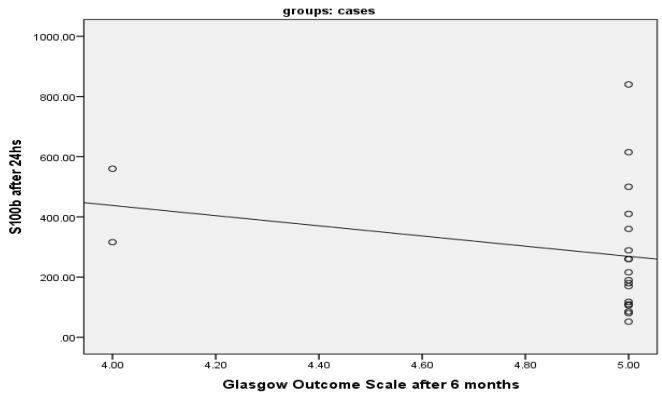


**Figure (7): Correlation between GOS and S100β after 24 hrs in group (I).**

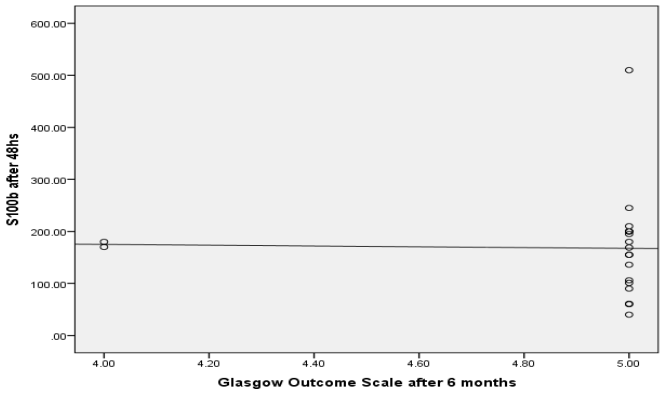


**Figure (8): Correlation between GOS and S100β after 48 hrs in group (I).**

## Discussion

EPO is a potentially attractive neuro-protective agent following TBI. In experimental models, EPO has improved outcome after TBI, subarachnoid hemorrhage, stroke, global ischemia, and other CNS disorders. EPO has been found to improve outcome from spinal cord injury and ischemia in some experimental studies. In experimental TBI studies, EPO has been shown to have neuroprotective effects when given early post-injury, and to have effects that enhance neurological recovery even when given at later times after injury. The early neuroprotective mechanisms are probably complex, involving anti-inflammatory, anti-apoptotic, and vascular actions. The time window for EPO induced neuroprotection after experimental TBI is at least 6 h post-injury. The late mechanisms of EPO that may enhance neurological recovery may include both neurogenesis and angiogenesis[11].

Low level expression of EPO and its receptors has long been reported in the brain especially after brain injury. Cerebral EPO is up regulated by harmful condition such as hypoxia. It was also shown that in vitro rhEPO could protect neuronal cells against hypoxic injury. Consequently, the possibility arise that brain's EPO may provide protective role and therefore administration of EPO could be useful to reduce consequences of brain injury.[12]

While EPO and EPOR are only weakly expressed in normal adult brain, expression of EPO and the EPOR is greatly increased in response to different types of brain injury. EPOR expression is significantly upregulated as early as 1 day and up to 7 days after TBI, whereas EPO is transiently elevated at days 1 and 2. The upregulated EPO and EPOR may reflect the brain's survival response to injury. Prolonged increased EPOR expression suggests that the endogenous EPO concentration may not be sufficient for cell survival after injury, and increased EPOR provides a platform for treatment with exogenous EPO. This is in agreement with results of many studies indicating that exogenous EPO provides neuroprotection and neurorestoration in several animal models, including TBI[13].

S100β is a 21-kD, calcium-binding protein that is mainly expressed in astroglial cells in the CNS. The S100β level in peripheral blood is known to be elevated in patients with various disorders of the CNS and also serves as a well-known biomarker for the severity of brain damage to predict the prognosis Since the peripheral S100β level has been used to suggest the extent of brain damage, the mechanisms underlying the increased peripheral S100β level have been investigated. Previous studies have suggested that S100β secrets increasingly from disrupted and /or activated glial cells into CSF and that S100β subsequently leaks into the blood stream through the damaged BBB[14].

As regard to identification of APACHE II score at admission we found that there was no significant deference between both groups. By using Sofa score we found that

there was no significant deference between both groups at admission data, but there was significant deference between the two groups on the subsequent days.

Our results were in agreement with Shiehmorteza et al., 2011[15], the purpose of their study was to evaluate antioxidant and anti-inflammatory effects of erythropoietin and its impact on organ function in traumatized patients. Twenty-six ICU-admitted traumatized patients within 24 hrs after trauma were randomly assigned to the EPO group which included 12 patients received EPO (300 units/Kg/day) subcutaneously and continued every other day for a total of 3 doses (days of 1, 3 and 5) and followed up for 10 days and control group not received EPO. Patients also underwent (APACHE II) scores, injury severity score based on data obtained within the first 24 hrs after ICU admission. (SOFA) as for organ dysfunction was assessed in the first and third and seventh days of admission and all participants were continuously monitored for their hemodynamic parameters. They found that ICU scores including APACHE II, and injury severity score scores were not significantly different between two groups, while a comparison of SOFA scores on the first, third and seventh day showed that EPO group had lower SOFA score which at least indicate that EPO has anti-inflammatory activity.

When our patients were monitored by frequent assessment of heart rate, mean arterial blood pressure and oxygen saturation we found that there was also no significant deference between both groups.

We found that intravenous administration of high dose of erythropoietin (40.000 IU) within 8 hours from trauma for 3 successive days to patients with moderate (TBI) results in significant clinical improvement which was assessed by GCS and improvement in the outcome which was assessed by GOS.

By comparing the changes in GCS between the two groups we found that there was no significant deference as regard to admission data and data measured 30 minutes, one hour and 24 hours after admission, but EPO group showed significant improvement in GCS at 36, 48, 60, 72, 84, 96, and 120 hours after admission. While within the group we found that GCS gradually increased and there was significant deference between the admission data and the following data starting at 36 hours in EPO group and there was no significant deference when we compared the admission data with the data measured after admission in control group.

Our results were in agreement with Saeid et al., 2012[12] in which a total of 54 patients (All of them men between 20-47 years old) were recruited to evaluate the use of rhEPO in outcome of DAI. They divided the patients into two groups 27 patients comprised intervention group that received 2000U rhEPO subcutaneously for six doses in two weeks (on days 0,2,4,6,8 and 10) and placebo group that received saline (0.9% NaCl). Follow-up neurologic outcome scoring was obtained on days 2,4,6,8 and 10, using the GCS showed differences of GCS from admission till the final step which proved that, GCS after administration of EPO was better than baseline.

Regarding to GOS which measured at discharge, 3months and 6months after discharge we found that: in EPO group five (25%) patients discharged with score 4 (moderate disability) and fifteen (75%) patients discharged with score 5 (good recovery) while in control group sixteen patients (80%) discharged with score 4 and four (20%) patients discharged with score 5. After three months of discharge; in EPO group four (20%) patients got score 4 and sixteen patients (80%) got score 5 while in control group fifteen (75%) patients got score 4 and five (25%) patients got score 5. Two patients in EPO group had moderate disability in comparison with thirteen (65%) patients in control group had moderate disability while eighteen (90%) patients in EPO group had good recovery in comparison with seven (35%) patients in control group had good recovery at the end of six months after discharge.There were no patients in both groups get score 3 (sever disability), score 2 (vegetative state) or score 1 (death).

Our results were in agreement with Saeid et al., 2012[12] study. Their results showed a difference of GOS at the end of study when the patients were discharge from hospital and found that the rhEPO-treated patients improved earlier in which the difference between two groups occurred on day 10 and the better course of the rhEPO-treated patients continued throughout the rest of study period.

In contrast to our study Nirula et al., 2010[16] done a randomized, double-blind, placebo-controlled single-center trial in which patients either received EPO or saline placebo of equal volume. All blunt trauma patients ≥18 years of age with an admission GCS <13 and evidence of (TBI) on CT were eligible for this study. After obtaining informed consent from the family, patients were randomized to receive EPO (40,000 Units IV) or placebo administered once within 6 hours of the time of injury. Demographic and clinical data were obtained including age, gender; admission and ICU GCS. Secondary outcome measures included ICU length of stay, GCS at ICU discharge, and in-hospital mortality. They found that ICU length of stay was shorter for the EPO group by a mean of 2 days, but this did not reach statistical significance. They concluded that secondary outcomes of death, length of stay and GCS did not differ with the treatment and they recommended a larger trial with a dose-finding strategy to determine if EPO is neuroprotective in TBI patients. And said that their study demonstrated the safety of its use and a higher dosing is needed in future studies.

To evaluate the effect of EPO treatment on (Hb) concentration we measured (Hb) concentration on the first, third and fifth days after admission, we found that there was no significant difference between the studied groups as regard to (Hb) concentration at admission, but there was significant difference on the third and fifth days of admission being higher in EPO group. When we evaluated the effect of EPO treatment on (Hct) percentage by measuring (Hct) percentage on the first, third and fifth days after admission, we found that there was no significant difference between the studied groups as regard to (Hct) percentage at admission, but (Hct) percentage increased in EPO group on the third and fifth days of admission with a statistical significant difference.

Our results were in agreement with Xiong et al., 2010[5] on their study. They found that the baseline of hematocrit (HCT) was similar for all animals before injury. As compared to saline treatment, EPO treatment significantly increased HCT up to 1 week in the group of animal which received EPO at 1 day and HCT level increased up to 2 weeks in the group of animal which received EPO at days 1,2 and 3 post injury which returned to normal thereafter.

In our study the clinical improvement is confirmed by significant reduction in the serum level of S100β when comparing the admission values with the values measured after 24 and 48 hours of admission within EPO group. There was also reduction in the serum level of S100β when comparing the admission values with the values measured after 24 and 48 hours of admission within control group but it is of no statistically significance. When they examined the relationship between serum S100β levels and GOS using correlation analysis, we found that there was weak negative correlation between serum levels of S100β which were measured at admission, 24hrs. and 48hrs. After admission and GOS six months after discharge.

A study of a large cohort of patients was performed by Kleindienst et al., 2010[17] was in agreement with our results in which patients were enrolled into two groups, one presenting with isolated TBI requiring ventriculostomy and catheter placement (n=71) and the other presenting with subarachnoid hemorrhage (n=185). The normal S100β values had been established in control patients undergoing pituitary surgery treated with a lumbar drainage. Extracranial and brain injury were documented by CT, neurological function by the GCS and (GOS), as well as intensive care scores (APACHE). In TBI and subarachnoid hemorrhage subjects, they collected blood and CSF samples daily at 8

AM for up to 4 weeks post injury. They found that S100β in CSF was significantly increased up to day 7 following TBI and subarachnoid hemorrhage and in serum up to day 8 following TBI and up to day 14 following subarachnoid hemorrhage. To evaluate the value of S100β in the prediction of the outcome they found that within the groups, there was no consistent correlation between S100β concentrations in either serum or CSF and neurological function as assessed by GCS.

In conclusion as regard to EPO we noticed that it has a neuroprotective effect which was manifested clinically by improvement of GCS and GOS after six months of discharge.As regard to the serum level of S100β we noticed that there was significant reduction in the serum level of S100β in EPO group than control group with a weak negative correlation between it and GOS

The limitation of our study that we did not measure the CSF level of S100β and compare it with the serum level which might give an idea about the integrity of BBB and might have better correlation with the outcome and we also did not measure the serum level of S100β for more prolonged period.

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