Benefits of N-acetylcysteine on liver functions in living donor hepatectomy

Mona A Ammar, Amr M Hilal Abdou
Department of Anesthesia and Intensive Care, Faculty of Medicine, Ain Shams University, Cairo, Egypt

ABSTRACT

Background and Aims: The proportion of patients undergoing living donor liver transplantation is high especially in countries without or with limited cadaver organ sharing programs. The aim of this study was to evaluate the post-hepatectomy effect of using N-Acetylcysteine (NAC) infusion in living donors undergoing donor hepatectomy. Methods: In a prospective randomised non-blinded study, 50 healthy donors were enrolled; following hepatectomy patients were randomised into 2 groups: Group NC receiving NAC 150 mg/kg diluted in 100 ml glucose 5% over 40 minutes, followed by NAC 12.5 mg/kg in 500 ml glucose 5% over 4 hours. This was followed by NAC 6.25 mg/kg for 2 post-operative days, Group C (Control group) received ringer acetate infusion at same rate for 2 days. The primary outcome was serum lactate levels. Secondary outcomes were liver function tests, serum creatinine and urine output on intensive care unit (ICU) admission (0 hr.), after 24 hours and 48 hours, length of ICU stay. Results: Our study revealed significant reduction in serum lactate in Group NC at 0, 24 and 48 hours compared to C group (P = 0.017, 0.002, 0.014). INR values showed significant reduction after 48 hours in Group NC compared to Group C (P = 0.049). Total Bilirubin, ALT, and Creatinine, urine output and ICU stay showed no statistical difference between the 2 groups. Conclusion: The NAC protocol is a safe, cost-effective tool for improvement of post hepatectomy liver function and early stabilisation of the metabolic profile.

Key words: Donor, liver, living, N-acetylcysteine, transplantation

INTRODUCTION

By the end of 2018, Egypt will have an average 3000 registered patients for living donor liver transplantation, which means that there will be same number of donors who will be voluntary donors especially when an established cadaveric programme for organ donation is not established. Thus there is an increased risk related to liver resection in these healthy donors which magnifies the importance of ensuring optimal graft size and function to maintain the donor safety. In our centre, Ain Shams Specialized Hospital we advocate a separate team for donors’ evaluation and assessment of fitness in all medical, surgical and psychological fields. One of the most dramatic complications following donors’ hepatectomy is postresection liver failure which is dependant on several factors, such as the extent of resection, vascular injury and ischaemic/reperfusion injury (IRI). In our practice, for the past 10 years, we have noticed a transitional impairment of liver function (elevated liver enzymes, total and direct bilirubin, and elevated serum lactate levels) following donor liver resection. Several drugs have been investigated for liver regeneration with proven benefit of N-Acetylcysteine (NAC) on rats with steatohepatitis. There have been several uses of NAC in treatment of acetaminophen toxicity and prevention of post radiocontrast renal injury, sepsis and cardiac surgeries. Being a rich source of sulfhydryl (SH) group, this increases the free radical scavenger glutathione (GSH) stores; NAC also plays an important role in decreasing the toxic damage...
induced by free radicals associated with impaired liver function following hepatic resection.\textsuperscript{12}

We conducted this study aiming to study the effects of using NAC post hepatic resection in donors and its possible benefit to the post-operative liver functions.

**METHODS**

The liver transplantation programme at Ain Shams University Hospitals relies on living related donors. Extra care during anaesthetic management of the donor is warranted because of clear ethical considerations.

This prospective randomised non-blinded study complies with declaration of Helsinki, performed after obtaining approval from the ethical committee of the Ain Shams University FWA 000017585, FMASU R10/2018. All participants provided written informed consent prior to entering the study. The trial was registered at Clinicaltrials.gov with reference number NCT03634566. This study was performed at Ain Shams University Specialized Hospital during the period of April 2018 and April 2019. The authors hypothesised that immediate post hepatic resection use of NAC will improve the liver function tests (ALT, Bilirubin, INR, Serum Lactate) reduce post-operative intensive care admission rates, in addition to lower rates of post-operative hepatic complications.

Sample size calculation was done using PASS programme (PASS version 2011, NCSS, LLC. Kaysville, Utah, USA) setting -1 error (alpha) at 0.05 and the power (1-β) at 0.8. Based on result from a previous study\textsuperscript{13} showed that the serum lactate level on day 1 among NAC group was 26.2 ± 7.7 mg/dL, while for the control group, it was 36.3 ± 15 mg/dL. Calculation according to these values produced a minimal sample size of 22 cases per group, with considering a 10% drop out rate. The sample was decided as 25 cases per group. Sample of 50 consecutive healthy adult more than 21 years ASA I or II donors’ candidate for right lobe hepatectomy for living donor liver transplantation (LDLT) were included and written informed consent was taken.

All donors have the right to abort and refrain from donation any time during their preparation till the morning of operation. Detailed explanation of the surgical hazards, hospitalisation, medication needs, and mortality rates (which is less than 0.5%) was done by a senior surgical staff. Exclusion criteria included reported allergy to NAC, All patients received conventional pre-operative assessment according to unit protocol to exclude any comorbidities or contraindication to donation. Potential donors were assessed in three phases as per Ain Shams Center of Organ Transplantation (ASCOT) protocol.\textsuperscript{14}

Standard anaesthetic technique started with routine monitoring tools, including invasive arterial blood pressure through left radial artery catheter, urine output hourly.

Donor operation consisted of right lobe hepatectomy with or without middle hepatic vein; intraoperative abdominal duplex ultrasound was done to delineate the middle hepatic vein and its tributaries and line of dissection.

Following hepatectomy patients were randomised using a computer generated list into two groups: Group NC received NAC 150 mg/kg diluted in 100 ml glucose 5% over 40 minutes followed by 12.5 mg/kg in 500 ml glucose 5% over 4 hours, and later 6.25 mg/kg for first 2 post-operative days. Group C (Control group) received ringer acetate continuous infusion at same rate for 2 days. At the end of operation all patients were fully recovered and transferred to surgical ICU for post-operative care.

Perioperative standard care including the anaesthetic technique, surgical team, dissection and intensive care management were set to avoid any heterogeneity in the study and to assure adequate standardisation of care.

All donors received standard post-operative care in the ICU, including full monitoring of haemodynamics, urine output, and routine laboratory profile done daily. Initiation of therapy included antibiotics, antiemetic, and analgesia in addition to continuation of NAC infusion. Fluid management consisted ringer acetate 40-60 ml/kg/day to maintain urine output of 100-200 ml/hr.

Generally, patients were started on liquid diet on day one post-operatively, care taken to achieve normovolemic balance with minimal urine output of 1 ml/kg/hr during the ICU stay.

The primary outcome was serum lactate levels. Secondary outcomes were liver function tests (Total Bilirubin, ALT levels, INR), serum creatinine and urine output in ml/hr recorded on
ICU admission (0 hr.), and after 24 hours and 48 hours, length of ICU stay, post-operative morbidity or mortality.

50 patients’ data were collected, revised, coded and entered to the Statistical Package for Social Science (IBM SPSS) version 23 (Chicago, USA). The quantitative data were presented as mean, standard deviations and ranges when their distribution found parametric. Also qualitative variables were presented as number and percentages. The comparison between groups regarding qualitative data was done by using Chi-square test. The comparison between two independent groups with qualitative data and parametric distribution were done by using Independent t-test. The confidence interval was set to 95% and the margin of error accepted was set to 5%. So, the $P$ value was considered significant at the level of $<0.05$.

RESULTS

The enrollment period started in April 2018 till April 2019. Herein, we included 50 patients’ scheduled for right lobe hepatectomy for living donor liver transplantation (22 males and 28 females) of mean age 26 years and weight 78 kilograms, Table 1 describes the donor characteristics in both NC and C groups with no significant statistical differences. The intraoperative period was uneventful in all patients, with no recorded haemodynamic instability, no blood products given, no need for inotropic support and no mortality recorded, all patients were successfully extubated and transferred to SICU.

Upon admission to ICU serum lactate, ALT, total bilirubin, Creatinine and international normalisation ratio (INR) were recorded. Our results revealed significant reduction in serum lactate in Group NC at 0, 24, 48 hours compared to group C ($P = 0.017, 0.002, 0.014$). Similarly INR values also showed significant reduction after 48 hours in Group NC as compared to Group C ($P = 0.049$) [Table 2 and Figure 1].

Regarding other parameters such as total bilirubin, ALT, and Creatinine there we no statistical difference between the two groups ($P > 0.05$) similarly urine output also did not show any statistical difference between two groups [Table 3]. With respect to the average ICU stay; there was no difference between the two groups ($P = 0.286$) [Figure 2] with average 2.4 days.

There was no recorded post-operative morbidity or mortality during the hospital stay, neither patients required post-operative surgical neither intervention nor ICU re-admission.

DISCUSSION

Donors’ safety is one of the main priorities as an anaesthetic team in management of living donor liver transplantation especially on a programme relying only on living donation. Post hepatectomy liver impairment is one of the serious complications following donation; which depends on several factors such as extent of resection, hepatic parenchymal status and prolonged ischaemia to the hepatic tissues. NAC being an antioxidant agent may reduce this insult, but no randomised controlled trial (RCT) is available to support of use of NAC in donor hepatectomy. Studies performed on the recipients showed that NAC is of benefit regarding the hepatic protection in Orthotopic LT.$^{[13,16]}$ The Liver is the primary organ responsible for clearance of serum lactate and impairment of lactate clearance is strongly associated with hepatic impairment and dysfunction.$^{[17,18]}$

In the process of liver resection in donors’ hepatectomy we observed elevated levels of lactate which reflects mitochondrial dysfunction and causes further acid base disturbances and acidosis. The consequences of lactic acidosis may affect the hepatic function in the post-operative period as described in several studies.$^{[19,20]}$ Effective lactate clearance post hepatectomy is associated with lower morbidity and mortality rates.

In our study starting the intraoperative infusion of NAC after liver resection significantly improved the lactate clearance values [Table 2 and Figure 1] compared to control group. Although lactate measurement as a single marker is not the ideal method for assessment of liver dysfunction,$^{[21]}$ early rise in serum lactate indicates accelerated hepatic dysfunction and gross pathological changes.

## Table 1: Demographic date age, sex and weight

| Parameter | NC | Group C | Test | $P$ |
|-----------|----|---------|------|-----|
| Sex (%)   | Female | 10 (40.0%) | 12 (48.0%) | 0.325* 0.569 |
|           | Male   | 15 (60.0%) | 13 (52.0%) |      |
| Age (years) | Mean±SD | 26.84±3.94 | 26.80±3.46 | 0.038• 0.970 |
|           | Range  | 22-35     | 21-33   |      |
| Weight (Kg) | Mean±SD | 78.00±4.35 | 75.88±5.13 | 1.575• 0.122 |
|           | Range  | 70-85     | 70-84   |      |

$P>0.05$: Non significant; $P<0.05$: Significant; *Chi-square test; •Independent t-test
With respect to our donors’ characteristics [Table 1] our results are similar to another study performed in Italy.\[22\] We did not find post-operative hepatic dysfunction in any of our patient. One of the reason may be that our center is not accepting donors with extended donor criteria. Other European centers\[23\] assessed the use of NAC protocol in recipients of LT which differ from our study in allocation of MELD scores and graft survival as we concentrated on the post-operative donors’ hepatic function. However, if we added together these demographic considerations, it would provide good results for the NC group which favors the effect of NAC protocol.

All earlier studies focused on the impact of NAC on recipients of LT and they have thus failed to express clinical benefit.\[24,25\] The reason that we used NAC protocol was because of a previous trial performed at King’s college\[15\] on Orthotopic LT focusing on the haemodynamic parameters and oxygen delivery. In this study, Bromely et al. proved that the significance of improvement of oxygen transport following the loading dose with no effect observed on the graft function, sepsis, or any other complication.
Table 3: Serum Creatinine and urine output (UOP) at 0, 24, and 48 h

| Parameter         | Group C NO=25 | Group NC NO=25 | Test* | P     |
|-------------------|---------------|----------------|-------|-------|
| Creatinine 0 h (mg/dl) |               |                |       |       |
| Mean±SD           | 0.96±0.15     | 0.93±0.19      | 0.760 | 0.451 |
| Range             | 0.8-1.2       | 0.8-1.3        |       |       |
| Creatinine 24 h (mg/dl) |               |                |       |       |
| Mean±SD           | 0.90±0.10     | 0.92±0.09      | -0.584| 0.562 |
| Range             | 0.8-1.2       | 0.8-1          |       |       |
| Creatinine 48 h (mg/dl) |               |                |       |       |
| Mean±SD           | 0.83±0.09     | 0.85±0.07      | -0.931| 0.356 |
| Range             | 0.7-1         | 0.7-1          |       |       |
| UOP 0 h (ml/h)    |               |                |       |       |
| Mean±SD           | 147.60±47.90  | 148.80±45.31   | -0.091| 0.928 |
| Range             | 80-250        | 80-250         |       |       |
| UOP 24 h (ml/h)   |               |                |       |       |
| Mean±SD           | 119.20±32.65  | 132.80±49.37   | -1.149| 0.256 |
| Range             | 80-200        | 80-250         |       |       |
| UOP 48 h (ml/h)   |               |                |       |       |
| Mean±SD           | 126.40±38.18  | 127.60±37.45   | -0.112| 0.911 |
| Range             | 90-200        | 90-200         |       |       |

P>0.05: Non significant; P<0.05: Significant; *Independent t-test

In our study, the use of NAC improved post-operative Lactate clearance levels by the first day in the ICU in addition to significant reduction of INR by the second day post operatively (P = 0.049) compared to control group not receiving NAC, which reflects the improvement of synthetic function of the liver, but no effects were observed on the other liver function tests such as total bilirubin, ALT. We feel that these values were due to the effect of intraoperative infusion of NAC; as it gives a potential time for the donors’ liver to replenish its own metabolic stores of GSH minimising the toxic effects of the free radicals.[10]

The benefits of NAC on the renal function following contrast agents is well known.[10] Our study did not show any effect on post-operative serum Creatinine levels and urine output during the ICU stay [Table 3], although nephrotoxic agents were not used. The adequate safety of the NAC, in addition to its low costs, positive effect as an antioxidant drug following hepatectomy in living donors’ LT makes the establishment of NAC protocol cost-effective especially in developing countries with average total cost of living donor LT of 15,000 $.

The limitations of the current study are the single center experience. Therefore, our results should be cautiously considered until further studies are conducted on living donor hepatectomy.

So what is the impact of this study on LDLT? Our study suggests that NAC could be beneficial in improving post-operative hepatic function by reducing serum lactate levels and stabilising the acid/base status of the living related donors, thereby optimising metabolic profile.

CONCLUSION

In a center that relies only on living organ living transplantation and where rendering donors’ safety a top priority, the NAC protocol is a safe anda cost-effective tool for improvement of post-hepatectomy liver function and stabilisation of the metabolic profile.

Financial support and sponsorship
Nil.

Conflicts of interest
There are no conflicts of interest.

REFERENCES

1. Trotter JF, Campsen J, Bak T, Wachs M, Forman L, Everson G, et al. Outcomes of donor evaluations for adult-to-adult right hepatic lobe living donor liver transplantation. Am J Transplant 2006;6:1882-9.
2. Verbesey JE, Simpson MA, Pomposelli JJ, Richman E, Bracken AM, Garrigan K, et al. Living donor adult liver transplantation: A longitudinal study of the donor’s quality of life. Am J Transplant 2005;5:2770-7.
3. Hammond JS, Guha IN, Beckham J, Lobo DN. Prediction, prevention and management of postresection liver failure. Br J Surg 2011:98:1188-200.
4. Gomez D, Malik HZ, Bonney GK, Vong V, Toogood GJ, Lodge JP, et al. Steatosis predicts post-operative morbidity following hepatic resection for colorectal metastasis. Br J Surg 2007;94:1395-402.
5. Sigh SA, Vivekanathan P, Sharma A, Sharma S, Bharathy KG. Retrospective analysis of post-operative coagulopathy after major hepatic resection at a tertiary care centre in Northern India. Indian J Anaesth 2017;61:575-60.
6. Barth SA, Insellman G, Engemann R, Heidemann HT. Influences of ginkgo biloba on cyclosporin A induced liver peroxidae in human liver microsomes in comparison to vitamin E, glutathione and N-acetylcysteine. Biochem Pharmacol 1991:41:1521-6.
7. Yormaz S, Bulunoglu E, Kurutas EB, Cirakli H. The comparison of the effects of hepatic regeneration after partial hepatectomy, silybum marinaum, propofol, N-acetylcysteine and vitamin E on liver. Bratisl Lek Listy 2012:113:145-51.
8. Shimamoto K, Hayashi H, Tanai E, Morita R, Imaoka M, Ishii Y, et al. Antioxidant N-acetyl-L-cysteine (NAC) supplementation reduces reactive oxygen species (ROS)-mediated hepatocellular tumor promotion of indole-3-carbonil (I3C) in rats. J Toxicol Sci 2011;36:775-86.
9. Uzun MA, Koksal N, Kadioglu H, Gunerhan Y, Aktas S, Dursun N, et al. Effects of N-acetylcysteine on regeneration following partial hepatectomy in rats with nonalcoholic fatty liver disease. Surg Today 2009;39:592-7.
10. Hoffmann U, Fischereder M, Krüger B, Drobnik W, Krämer BK. The value of N-acetylcysteine in the prevention of radiocontrast agent-induced nephropathy seems questionable. J Am Soc Nephrol 2004:15:407-10.
11. Hein OV, Ohring R, Schilling A, Oellerich M, Armstrong VW, Kox WJ, et al. N-acetylcysteine decreases lactate signal intensities in liver tissue and improves liver function in septic shock patients, as shown by magnetic resonance spectroscopy: Extended case report. Crit Care 2004;8:66-71.
12. Vivot C, Van Ness K, Schwartz ME, Theise ND, Miller CM. N-acetylcysteine attenuates cold ischemia/reperfusion injury in the isolated perfused rat liver. Transplant Proc 1993;25:1983-4.
13. Sayed E, Gaballah K, Younis E, Yassen K, Abo El-Einen. The effect of intravenous infusion of N-Acetylcysteine in cirrhotic patients undergoing liver resection: A randomized controlled trial. J Anaesthesiol Clin Pharmacol 2017;33:450-6.
14. El-Meteini M, Dabbous H, Sakr M, Ibrahim A. Donor rejection before living donor liver transplantation: Causes and cost effective analysis in an Egyptian transplant center. Hepat Mon 2014;14:e13703.
15. Bromley PN, Cottam SJ, Hilmi I, Tan KC, Heaton N, Ginsburg R, et al. Effects of intraoperative N-acetylcysteine in orthotopic liver transplantation. Br J Anaesth 1995;75:352-4.
16. Thies JC, Koeppel TA, Lehmann T, Schemmer P, Otto G, Post S. Efficacy of N-acetylcysteine as hepatoprotective agent in liver transplantation; an experimental study. Transplant Proc 1997;29:1326-7.
17. Record CO, Chase RA, Williams R, Appleton D. Disturbances of lactate metabolism in patients with liver damage due to paracetamol overdose. Metabolism 1981;30:638-43.
18. Almenoff PL, Leavy J, Weil MH, Goldberg NB, Vega D, Rackow EC. Prolongation of the half-life of lactate after maximal exercise in patients with hepatic dysfunction. Crit Care Med 1989;17:870-3.
19. Del Portal DA, Shaffer F, Mikkelsen ME, Dorsey PJ, Gaienski DE, Goyal M, et al. Emergency department lactate is associated with mortality in older adults admitted with and without infections. Acad Emerg Med 2010;17:260-8.
20. Soliman HM, Vincent JL. Prognostic value of admission serum lactate concentrations in intensive care unit patients. Acta Clin Belg 2010;65:176-81.
21. MacQuillan G. Predicting outcome in acute liver failure: Are we there yet? [comment]. Liver Transplant 2007;13:1209-11.
22. Angelico M, Cillo U, Fagiuoli S, Gasbarrini A, Gavila C, Marianelli T, et al. for Liver Match Investigators. Liver Match, a prospective observational cohort study on liver transplantation in Italy: Study design and current practice of donor-recipient matching. Dig Liver Dis 2011;43:155-64.
23. Weismüller TJ, Negm A, Becker T, Barg-Hock H, Klempnauer J, Manns MP, et al. The introduction of MELD-based organ allocation impacts 3-month survival after liver transplantation by influencing pretransplant patient characteristics. Transpl Int 2009;22:970-8.
24. Hoffmann K, Bächler MW, Schemmer P. Supplementation of amino acids to prevent reperfusion injury after liver surgery and transplantation—where do we stand today? Clin Nutr 2011;30:143-7.
25. Jegatheeswaran S, Siriwardena AK. Experimental and clinical evidence for modification of hepatic ischaemia reperfusion injury by N-acetylcysteine during major liver surgery. HPB (Oxford) 2011;13:71-8.
26. Alito ML. N-Acetylcysteine passe-partout or much ado about nothing? Br J Clin Pharmacol 2006;61:5-15.