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Role of N-acetylcysteine in non-acetaminophen-related acute liver failure: an updated meta-analysis and systematic review

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Abstract

Background The American Association for the Study of Liver Diseases recommends that N-acetylcysteine (NAC) may be beneficial in non-acetaminophen-related drug-induced liver injury. A subsequent review and analysis reported the current evidence to be inconclusive. Herein, we present an updated review and meta-analysis.

Methods We evaluated prospective, retrospective and randomized controlled trials that compared outcomes in patients of all ages with acute liver failure (defined as abnormal liver enzymes along with elevated international normalized ratio >1.5, with or without hepatic encephalopathy) receiving NAC with the outcomes in a control group. The primary outcome was to compare the overall survival in the 2 groups. Secondary outcomes included difference in length of hospital stay, transplant-free survival, and post-transplant survival.

Results Seven studies (N=883) that met the inclusion criteria were included in this analysis. The mean age of patients in the NAC group was 21.22 years compared with 23.62 years in the control group. The odds of overall survival were significantly higher in the NAC group than in controls (odds ratio [OR] 1.77, 95% confidence interval [CI] 1.3-2.41). Post-transplant survival (OR 2.44, 95%CI 1.11-5.37) and transplant-free survival were also better in the NAC group than in the control group (OR 2.85, 95%CI 2.11-3.85). Patients in the control group had statistically significant odds of a longer inpatient stay (mean difference 7.79, 95%CI 6.93-8.66).

Conclusion In patients with non-acetaminophen-related acute liver failure, NAC significantly improves overall survival, post-transplant survival and transplant-free survival while decreasing the overall length of hospital stay.

Keywords N-acetylcysteine, acute liver failure, drug-induced liver injury, acetaminophen

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Introduction

Acute liver failure is a rare, life-threatening disease characterized by acute liver dysfunction in patients who have no previous history of underlying liver disease. It is a rapidly progressive disorder with a reported incidence of around 2000-3000 cases per year in the US, with mortality as high as 30% [1]. The term was originally used by Trey and Davidson in the 1970s. The International Association for the Study of the Liver further classifies liver failure into hyperacute liver failure, occurring within 10 days of the inciting event, fulminant, occurring within 10-30 days, and subacute hepatic failure, occurring within 5-24 weeks [2].

Acetaminophen remains the most common etiology of acute liver failure in the US, followed by other drug-induced liver injury and hepatitis B virus [1]. Medical management usually involves supportive measures that depend on the underlying etiology. Liver transplant remains the only effective treatment, but given the limited number of organs available, other treatment modalities have been sought.

N-acetylcysteine (NAC) has been the drug of choice for the treatment of acetaminophen-related liver failure since the 1970s. It is a thiol-containing derivative of amino acid cysteine. NAC helps neutralize free oxygen radicals and replenishes cytoplasmic and mitochondrial glutathione stores by acting as a glutathione substitute and directly combining with reactive metabolites. It serves as a source of sulfate, thus enhancing

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Conflict of Interest: None

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non-toxic sulfate conjugation and preventing hepatic damage [3-5]. It has also been suggested that NAC may have a vasodilatory and inotropic role, thus improving perfusion and oxygenation to vital organs during shock-like states [4,6]. While the role of NAC in acetaminophen-induced liver failure is pivotal, in 2011 the American Association for the Study of Liver Diseases (AASLD) guidelines suggested NAC may also be beneficial in non-acetaminophen-related drug-induced liver injury [7]. The evidence for this recommendation came largely from a double blinded randomized trial by Lee *et al*, which showed that intravenous NAC improved transplant-free survival in non-acetaminophen-related acute liver failure. The majority of patients in the study had drug-induced liver injury (DILI) [8]. Hu *et al* published a meta-analysis in 2015, which included 4 prospective studies. Their results showed an insignificant difference in overall survival between the NAC and control groups. However, the NAC group was found to have better transplant-free survival and post-transplant survival [9]. Chughlay *et al*, in their meta-analysis in 2016, concluded that the current evidence is inconclusive to determine whether there is a role for NAC in non-acetaminophen-related DILI [3].

Newer studies have since been published that were not included in previous meta-analyses [10-12]. In our meta-analysis we sought to include all available studies, including randomized control trials and retrospective studies evaluating the efficacy of NAC in non-acetaminophen-related acute liver injury. The primary outcome was to compare the overall survival in patients presenting with acute liver failure who received NAC vs. those who did not. Secondary outcomes included differences in length of hospital stay, transplant-free survival, and post-transplant survival.

Materials and methods

Study selection criteria

We looked at studies assessing the efficacy of NAC in non-acetaminophen-related acute liver failure. Acute liver failure was defined as abnormal liver enzymes along with an elevated international normalized ratio >1.5, with or without the presence of encephalopathy, in a patient who previously had no evidence of liver disease.

Data collection and extraction

We searched the following databases: MEDLINE, PubMed, Ovid journals, Embase, Cumulative Index for Nursing and Allied Health Literature, ACP Journal Club, DARE, International Pharmaceutical Abstracts, old MEDLINE, MEDLINE Non-Indexed Citations, OVID Healthstar, and Cochrane Central Register of Controlled Trials (CENTRAL). The search was performed for the years 2000 to December 2019. Abstracts were manually searched in the major gastroenterology journals for the past 3 years. The search terms used were non-acetaminophen-related acute liver injury, acute liver failure, N-acetylcysteine, drug-induced liver injury, transplant-free survival, overall survival, mortality, morbidity, length of hospital stay, complications, meta-analysis, and systematic review. The reference lists of all eligible studies were reviewed to identify additional studies. The retrieved studies were carefully examined to exclude potential duplicates or overlapping data. Titles and abstracts selected from the initial search were first scanned, and the full papers of potential eligible studies were reviewed. Two authors (SW and HS) independently searched and extracted the data into an abstraction form. Any differences were resolved by mutual agreement. Preferred Reporting Items for Systematic reviews and Meta-Analysis guidelines (PRISMA) statement guidelines were followed for conducting and reporting meta-analyses. The PICOS scheme was followed for reporting inclusion criteria.

Study selection criteria

We included prospective and retrospective studies in our meta-analysis. The patient population could range anywhere from neonates to adults. NAC could be administered orally or intravenously. The following information was extracted from the study: authors, year of publication, place, study design, number of patients receiving NAC, dose and route of NAC administration, age, length of stay, overall survival, transplant-free survival, post-transplant survival in NAC and control group. Articles were excluded if: 1) they were not written in English; 2) no outcomes were reported; or 3) they represented review articles or studies published as abstracts only.

Statistical analysis

Microsoft Excel was used for data collection. Statistical analysis was performed using Rev-Man 5.3 (Cochrane Collaboration, Oxford, UK). We conducted a random-effect meta-analysis when there was significant heterogeneity; otherwise, we used the fixed-effect model. For effect sizes the odds ratio (OR) for dichotomous outcomes and standardized mean difference (SMD) for continuous variables were calculated using a random-effect model in cases of significant heterogeneity between estimates. The heterogeneity among studies was tested using the I^2 statistic and Cochrane's Q test [13]. An I^2 value of 0-39% was considered as non-significant heterogeneity; 40-75% as moderate heterogeneity; and 76-100% as considerable heterogeneity. A P-value >0.05 was considered to reject the null hypothesis that the studies were heterogeneous. The effect of publication and selection bias on the summary estimates was tested using the Begg-Mazumdar bias indicator [14]. In addition, funnel plots were constructed to evaluate potential publication bias [15,16].

Results

The initial search identified 28 reference articles, of which 7 were selected and reviewed. Data were extracted from 7 studies (N=883) that met the inclusion criteria. The study was conducted based on the PRISMA guidelines. The schematic diagram for study selection criteria is mentioned in Fig. 1. All studies were published as full articles. All the pooled estimates given were calculated using fixed- and random-effect models. The mean age of patients in the NAC group was 21.22 years, compared to 23.62 years in the control group. Table 1 shows the etiology of acute liver failure in our patient population. The P-value for chi-square heterogeneity for all pooled accuracy estimates was considered significant if <0.05. The agreement between reviewers for the collected data gave a Cohen κ value of 1.0.

Primary outcome

The primary outcome was overall survival in our meta-analysis. The odds of survival were 1.77 times higher (95% confidence interval [CI] 1.30-2.41; P<0.001) in the NAC group compared to the control group (Fig. 2) shows the forest plot of odds in individual studies. Publication bias calculated using the Begg-Mazumdar indicator gave a Kendall's tau b value of 0.524 with a P-value of 0.108, indicating no publication bias.

Secondary outcomes

Transplant-free survival

Transplant-free survival was defined as the percentage of patients who did not receive a transplant and survived. The odds of transplant-free survival favored the NAC group compared to the control group (OR 2.85, 95%CI 2.11-3.85; P<0.001). Fig. 3 shows the forest plot of odds in individual studies. The Begg-Mazumdar indicator gave a Kendall's tau b value of 0.238 with a P-value of 0.359, indicating no significant bias. Fig. 4 shows the funnel plot for bias.

Post-transplant survival

The odds of post-transplant survival favored the NAC group compared to the control group (OR 2.44, 95%CI 1.11-5.37; P=0.03). Heterogeneity calculated using chi^2 was 1.23 and I^2 was 19%, indicating no significant heterogeneity.

Table 1 Etiologies of acute liver failure

Etiology	N-acetylcysteine	Control	Total
Drug-induced liver failure	48	45	102
Viral hepatitis	115	98	213
Autoimmune	15	17	32
Metabolic	29	16	45
Other (infection, undetermined, pregnancy-related etc.)	139	145	284

Length of stay

Length of stay details were provided in 5 studies. The mean difference in length of stay favored the control group as compared to the NAC group (mean difference: 7.79, 95%CI 6.93-8.66; P<0.001).

Discussion

The management of patients with acute liver failure remains a challenge. In view of the limited number of organs available, aggressive and early medical optimization remains key. Medical options mostly include supportive care.

Previous studies and meta-analyses suggested that NAC could play a supportive role in patients with non-acetaminophen-related acute liver failure [3,9]. This article presents an updated meta-analysis, with a total of 7 studies being included. The last meta-analysis was conducted in 2015 by Hu *et al* and included 4 studies [9]. Our results were different from those of Hu *et al* and showed significant improvement in overall survival in patients with non-acetaminophen-related acute liver failure treated with NAC.

The route of delivery of NAC was intravenous in all the studies included in our analysis, except for that of Mumtaz *et al*, who administered NAC orally. NAC was administered as a continuous infusion in 3 studies, while in the other 3 studies a loading dose was given prior to the continuous infusion [4]. A systematic review comparing oral with intravenous NAC administration showed similar levels of hepatotoxicity in rats with acetaminophen overdose, and the route of dosing did not make much difference [17]. NAC is a well-tolerated drug and has a low adverse effect profile. The adverse effects that have been reported include allergic reactions (bronchospasm, rash), cardiac pathologies (arrhythmias) and other generalized symptoms (dizziness, peripheral edema) [18].

Our results support the findings reported by Hu *et al* showing that transplant-free survival and post-transplant survival were better in the NAC group as compared to the control group [9]. On the other hand, another variable that had not been included in prior analyses, length of hospital stay, was found in our study to be shorter for patients in the NAC group compared to the control group.

Two of the 3 most recent prospective trials showed a significantly better overall survival in the group of patients in whom NAC was prescribed [10,12]. There was only one study conducted in children (Parkas *et al*) that showed no significant difference in overall survival. However, that study reported significantly better survival when NAC was initiated in patients with Grade I and II hepatic encephalopathy [11].

The oldest study in our analysis was a retrospective study conducted over 2 different periods: 1989-1994, when standard care was delivered, and 1995-2004, when patients received

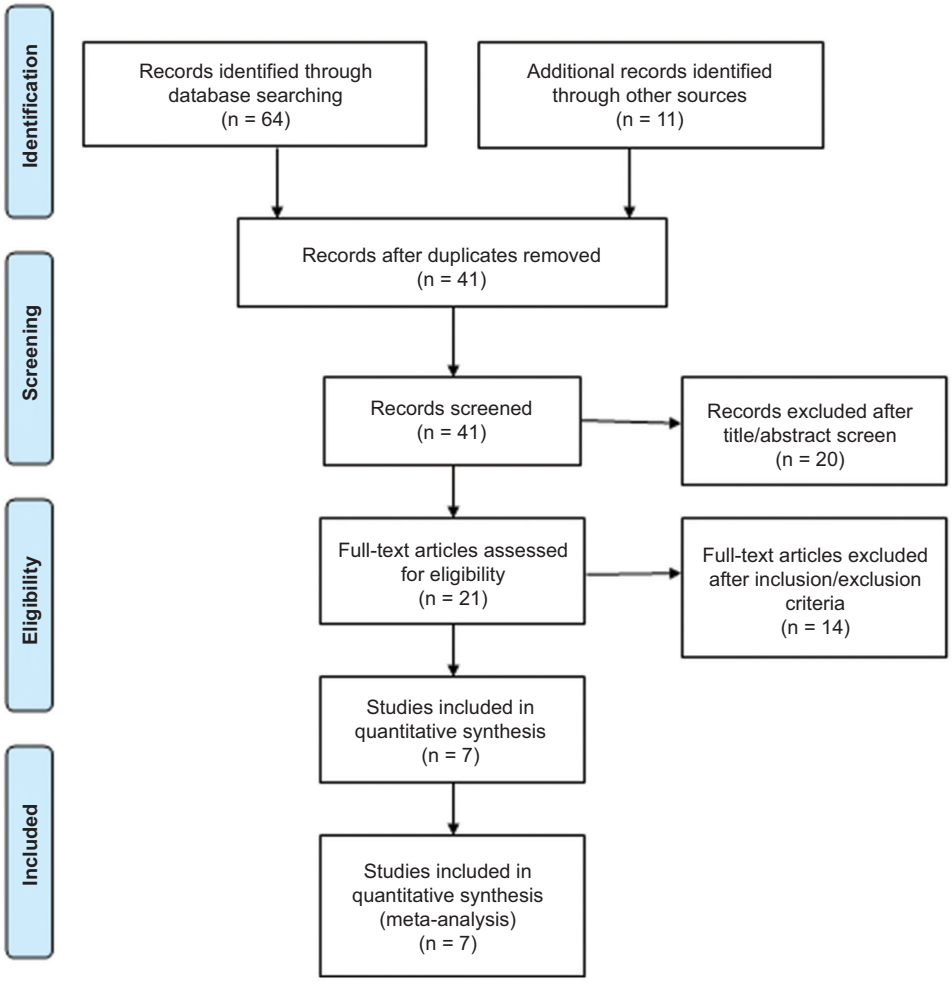

Figure 1 Study selection process in accordance with preferred reporting Items for systematic reviews and meta-analyses (PRISMA) guidelines

NAC. This study, conducted on children, reported a significant improvement in overall survival (10-year actuarial survival), transplant-free survival and post-transplant survival in the group that received NAC. However, there was some potential for bias, including a retrospective design, and a significantly higher percentage of jaundice, splenomegaly and ascites in the earlier group, raising the possibility that patients in this group may have had more advanced liver disease. The other potential for bias was the advances in healthcare between the 2 periods. This could be inferred from the data in the article regarding the higher rate of survival in the more recent period (2000-2004) [19].

Lee *et al*, in a randomized controlled trial, concluded that, in patients with non-acetaminophen acute liver failure and an early stage of hepatic encephalopathy, those in whom NAC was administered showed significantly better transplant-free survival. The results of this study suggested that earlier initiation of NAC is the key to improving outcomes, as patients with advanced coma did not benefit much from NAC and a greater proportion of those patients underwent transplantation [7]. Darwesh *et al* showed also showed significantly better transplant-free survival of adults in the NAC group, especially if treatment was started early, as 71% of the patients in the NAC group had grade 0 encephalopathy [12].

Another prospective study (Mumtaz *et al*), conducted on an adult population, showed no significant benefit for the NAC group in terms of overall survival or length of hospital stay. This center lacked the capability for liver transplantation, and patients in the treatment group had worse baseline presentations. This was evident from the significantly higher bilirubin levels on presentation in the treatment group, with an increased percentage of patients requiring admission to the intensive care unit during their hospital stay. However, this was the only study in our analysis that administered NAC orally [4]. Oral NAC may have impaired absorption in the setting of liver failure, due to difficulty with tolerance (NAC associated nausea and vomiting), delayed gastric emptying (especially in cases of DILI), and concerns about intestinal failure [18].

It is notable that only one randomized controlled trial from a pediatric population was included in our analysis. This trial showed no difference in survival in the NAC group. However, it did show better transplant-free survival in the NAC group,

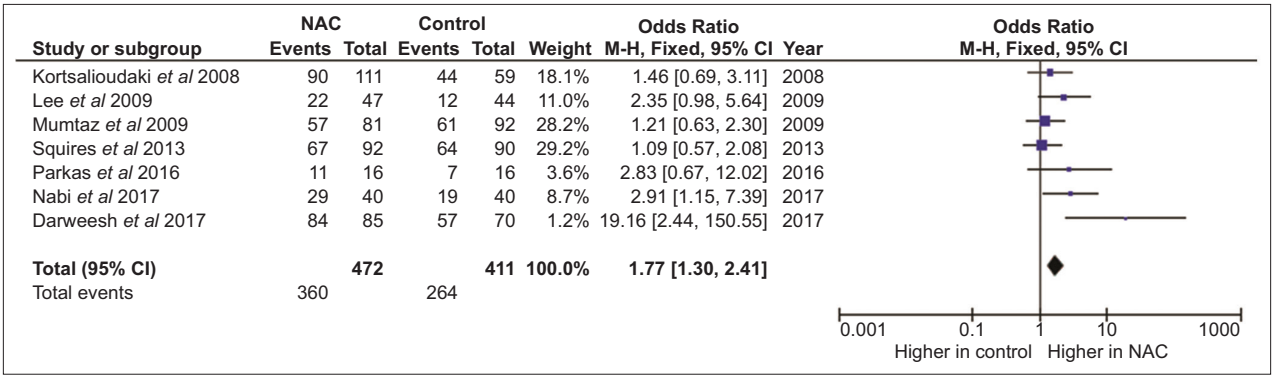

Figure 2 Forrest plot representing individual study proportions and the pool estimates of overall survival

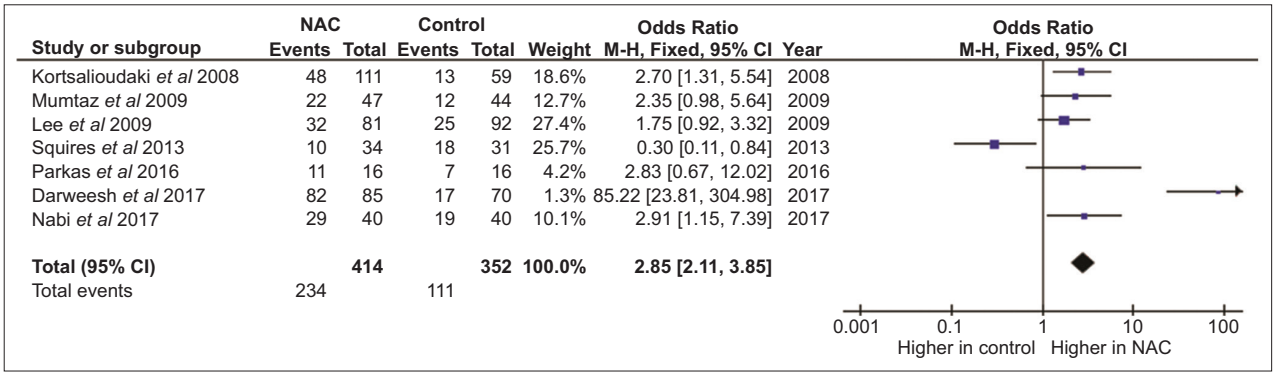

Figure 3 Forrest plot representing individual study proportions and the pool estimates of transplant-free survival

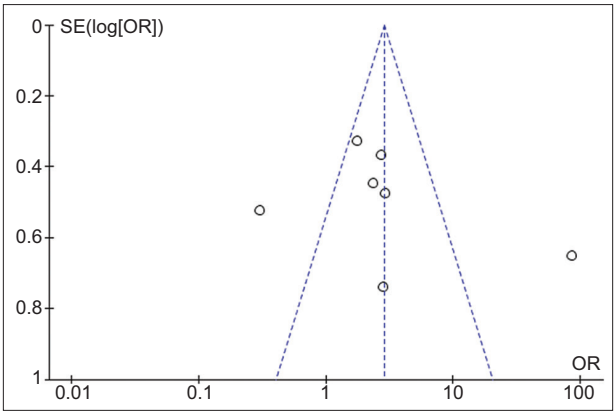

Figure 4 Funnel plot assessing publication bias for transplant-free survival

whereas the results for overall survival at the end of 1 year showed no significant difference [20]. Another prospective study of children also showed no significant difference in overall survival; however, survival was significantly better if NAC was given early, i.e., to patients with Grade I and II hepatic encephalopathy [11]. More randomized control trials need to be carried out before further concrete conclusions can be drawn regarding NAC administration in children; in the meantime, it may have some benefit early on and its use should be continued.

Our results support the use of NAC as an initial drug of choice in patients presenting with non-acetaminophen-related acute liver failure. In addition, our findings also suggest that NAC improves post-transplant and transplant-free survival. The reason for this trend in our study could be the larger number of patients and additional data being included from the studies by Parkas, Darwesh and Nabi *et al*, which added extra power to our meta-analysis.

The strengths of our meta-analysis include the fact that the literature review and data extraction were performed independently by 2 authors. Comparison of their analyses indicates excellent agreement. Publication bias was calculated using Kendall's tau equation. In addition, we used funnel plots to assess and report publication bias. The limitations of our meta-analysis are that only 2 studies were randomized controlled trials and only 1 was a multicenter trial; the others were mostly limited to a single institute. Studies with positive results tend to be published and cited. Moreover, smaller studies may show larger treatment effects compared to larger studies. While interpreting our results it should also be kept in mind that we included both children and elderly populations in our meta-analysis and, while there could be some overlap, the etiology of acute liver failure in children could differ from that in adults, leading to differences in outcome.

Currently, the AASLD recommends that NAC may play a role in acute drug-related failure to improve survival, but only 5% of the patients in our meta-analysis had clearly demarcated drug-induced liver failure. Our data support the use of NAC, not only in drug-induced liver failure, but also in other etiologies such as viral hepatitis, or any liver failure of

unknown origin. Further research may help clearly delineate the role of NAC in non-acetaminophen-related drug-induced acute liver failure, and to determine the ideal dosing regimens.

In conclusion, NAC improves survival in patients with non-acetaminophen-related acute liver failure. It also improves transplant-free survival, post-transplant survival and length of stay. Our data support the use of NAC in non-acetaminophen-induced acute liver failure and we propose it should be thought of as a first-line drug in acute liver failure of unknown origin, while patients are awaiting transplantation, especially in centers which cannot offer this. We also propose that it should be started earlier in the course of illness, as that has been shown to lead to better outcomes. Further studies are needed before more concrete conclusions may be drawn regarding its use in children, the ideal dosing regimen, and outcomes in specific etiologies of acute liver failure.

Summary Box

What is already known:

- American Association for the Study of Liver Diseases guidelines state that N-acetylcysteine (NAC) may be beneficial in non-acetaminophen-related drug-induced liver injury
- A previous meta-analysis in 2015 showed that NAC was associated with better transplant-free survival and post-transplant survival, but the difference in overall survival between NAC and control patients was found to be insignificant

What the new findings are:

- Based on this meta-analysis, in addition to transplant-free and post-transplant survival, the odds of overall survival were also significantly higher in the NAC group compared to the control group
- Only 5% of patients included in our meta-analysis had drug-induced liver failure; thus, our findings suggest that NAC could also be beneficial for other causes of acute liver failure, especially if administered early on
- Patients with non-acetaminophen-related acute liver failure who did not receive NAC had a longer hospital stay

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