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Role of amifostine in preventing liver injury caused by paracetamol intoxication

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Abstract

This investigation attempted to examine the possible protective impact of amifostine, alone or in combination with N-acetylcysteine (NAC), in paracetamol-induced liver injury. Fifty-six adult male Wistar rats (weighing 180–200 g) were randomly divided into seven groups (n = 8 per group): sham, paracetamol, amifostine, NAC, paracetamol + NAC, paracetamol + amifostine, and paracetamol + NAC + amifostine. Liver injury was assessed through serum AST and ALT activities, tissue glutathione and nitric oxide (NO) levels, and histopathological scoring. Severe hepatocellular injury was detected only in the paracetamol + NAC group. The paracetamol + NAC + amifostine group exhibited significantly lower hepatocyte damage compared with both paracetamol + NAC and paracetamol + amifostine groups, ranking second only to the control group. Mild to moderate regenerative changes were noted in the livers of the paracetamol + NAC and paracetamol + amifostine groups. No signs of fibrosis, iron storage, or steatosis were detected in any experimental group. Tissue NO levels were lowest in the paracetamol + NAC + amifostine group, reaching statistical significance compared to all other groups. The combined administration of amifostine and NAC provided superior protection against paracetamol-induced liver injury compared with standard NAC therapy, supporting the potential role of amifostine as an adjunct treatment.

Keywords: Amifostine, antidote, emergency service, N-acetylcysteine, paracetamol, toxicology

Introduction

Paracetamol is among the most frequently prescribed drugs in the class of analgesic and antipyretic agents, and it represents the leading cause of acute hepatic failure in the United States [1]. The hepatotoxic effects linked to high doses of acetaminophen (paracetamol, N-acetyl-p-aminophenol) were first reported by Davidson and Eastham [2]. Following this initial description, a series of systematic toxicological studies on paracetamol were conducted.

In certain instances, patients with paracetamol intoxication required liver transplantation, prolonged monitoring and treatment in intensive care units, or even hemodialysis due to acute kidney failure despite receiving standard therapy. Rarely, additional systemic complications involving other organs have also been reported [3].

It has been reported that paracetamol intoxication imposes an annual economic burden of nearly 87 million dollars in the United States [4]. N-acetylcysteine (NAC), the only clinically approved antidote for acetaminophen toxicity, exerts its hepatoprotective effect primarily through replenishment of intracellular glutathione (GSH) stores, detoxification of the reactive metabolite N-acetyl-p-benzoquinone imine (NAPQI), and direct scavenging of reactive oxygen species. In later stages, NAC also contributes to mitochondrial energy metabolism

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by providing substrates to the Krebs cycle, thereby limiting oxidative stress and cell death [5].

Amifostine (WR-2721) is a phosphorylated thiol prodrug that is dephosphorylated in tissues to its active metabolite WR-1065, which acts as a potent free radical scavenger, antioxidant, and cytoprotective agent. It has been widely studied as a radioprotector and chemoprotector due to its ability to neutralize reactive oxygen species, preserve DNA integrity, and maintain cellular redox balance. Recent experimental studies have demonstrated that amifostine exerts hepatoprotective effects against various toxic agents, including methotrexate, acrylamide, and acetaminophen [6-8].

Although Rasouli et al. recently demonstrated that amifostine and its active metabolite WR-1065 could mitigate acetaminophen-induced hepatotoxicity in Wistar rats [8], that study did not include a comparison with N-acetylcysteine.

To our knowledge, the present research is the first to systematically evaluate and directly compare the protective effects of amifostine alone, NAC alone, and their combination in a standardized model of paracetamol-induced liver injury.

In light of this, the aim of our study was to assess the therapeutic efficacy of NAC [9]—a well-recognized treatment option—against that of amifostine (WR-2721), a phosphorylated thiol compound known for its protective activity against hepatic and renal toxicity caused by anticancer therapies [10]. Furthermore, the study sought to determine whether their combined use might provide an additional benefit in a preclinical experimental setting.

Material and Methods

A total of 56 adult male Wistar rats (180–200 g) were included in the experiment. The study design was evaluated and approved by the Adnan Menderes University Local Ethics Committee for Animal Experiments (ADÜ-HADYEK) on August 1, 2012 (Approval No: B.30.2.ADÜ.0.00.00.00/050.04/2012/030). All animals were housed under standardized conditions and randomly assigned to seven groups of equal size two weeks prior to the start of the protocol. The rats were accommodated in Makrolon Type 4 cages within a controlled laboratory setting, maintained under a 12 h light/12 h dark cycle, at 24 ± 1 °C temperature, and 50%–70% relative humidity.

After a two-week acclimatization period, the experimental procedures described below were conducted on these groups. Before liver toxicity assessment, the rats were fasted overnight for approximately 16 hours and weighed before the procedure.

Experiment Groups and Their Design

Seven groups were formed by random assignment, with eight rats in each group:

1. Sham (n= 8): No paracetamol intoxication was induced; each rat received only 8 ml of physiological saline (SF).
2. Paracetamol (n= 8): Rats were administered 1 g/kg of paracetamol dissolved in 8 ml of SF via orogastric gavage.
3. Amifostine (n= 8): No paracetamol intoxication was induced; animals received intraperitoneal delivery of amifostine at a dose of 100 mg/kg.
4. NAC (n= 8): During paracetamol intoxication, rats were treated with an intraperitoneal bolus of N-acetylcysteine at 140 mg/kg, reflecting the clinically used dosage.
5. Paracetamol + NAC (n= 8): Rats received 1 g/kg of paracetamol (dissolved in 8 ml of SF) by orogastric gavage, followed 30 minutes later by intraperitoneal administration of NAC at 140 mg/kg.
6. Paracetamol + Amifostine (n= 8): Animals were given 1 g/kg of paracetamol (dissolved in 8 ml of SF) by orogastric gavage, followed after 30 minutes by intraperitoneal injection of amifostine at 100 mg/kg.
7. Paracetamol + Amifostine + NAC (n= 8): Rats received 1 g/kg of paracetamol (dissolved in 8 ml of SF) by orogastric gavage, followed 30 minutes later by intraperitoneal administration of NAC (140 mg/kg) combined with amifostine (100 mg/kg).

All rats received intramuscular ketamine hydrochloride (Ketalar, Eczacıbaşı; 50 mg/kg) to induce general anesthesia while maintaining spontaneous respiration. The abdominal area was prepared with 10% povidone-iodine, and a midline laparotomy of approximately 4 cm was performed under sterile conditions, leaving only the incision site exposed. Under aseptic circumstances, nearly 4 ml of blood was obtained from the vena cava for biochemical testing and placed into both plain and heparinized tubes. Following blood collection, the animals were euthanized.

Paracetamol (Parol 500 mg tablet; Atabay İlaç Fab. A.Ş., Turkey) was obtained in tablet form through commercial sources. N-acetylcysteine (Asist ampoule, 300 mg/3 mL [10%]; Hüsnü Arsan İlaç Sanayi, Turkey) was provided as an injectable preparation, and amifostine (Ethyl 500 mg vial, 10 mL; Er-Kim İlaç Sanayi ve Tic. A.Ş.) was purchased in ampoule form.

All animal groups were monitored for 24 hours. At the end of this period, about 4 ml of blood was drawn again from the vena cava for biochemical analyses and divided into heparinized and plain tubes. Plasma separation was carried out after centrifugation of the samples at 4000 g for 10 minutes. The erythrocyte pellets were rinsed three times with cold physiological saline, resuspended in an equal volume of water, and stored at −20 °C until further analysis. The same biochemical markers were measured in serum samples and compared with hemoglobin (Hb) levels. Nitric oxide (NO) concentrations were quantified after centrifugation. Liver damage assessment was performed by measuring serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities with a commercially available diagnostic kit.

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Preparation of Tissue Samples and Pathological Samples

Liver tissues were homogenized at 4 °C in 50 mM phosphate buffer (pH 7.4) at a ratio of 1:10 (w/v). The buffer contained 0.2 M phenylmethylsulfonyl fluoride (PMSF) as a protease inhibitor, 1 M Ethylenediaminetetraacetic acid (EDTA), and 1 µM leupeptin. The homogenates were centrifuged at 10,000 rpm for 5 minutes, and the resulting supernatants were transferred into Eppendorf tubes and preserved at –80 °C until analysis.

Hepatic tissue fragments were immersed in 10% neutral buffered formalin for 24 hours, subsequently embedded in paraffin blocks using routine histological methods. Sections 4 µm in thickness were prepared with a microtome, stained with hematoxylin–eosin, and mounted with Entellan for microscopic evaluation.

Histopathological examination of liver samples from all experimental groups included assessment of hepatocyte injury, regenerative alterations, reticular framework status, focal necrosis, and fibrosis (Trichrome–Masson staining). Iron accumulation and hepatic steatosis were also evaluated. The severity of findings was graded semi-quantitatively as absent (–), mild (+), moderate (++), or severe (+++).

GSH Measurement Method

Specimen glutathione levels were determined in line with the procedure described by Beutler et al. The precipitating solution consisted of metaphosphoric acid, disodium EDTA, and sodium chloride (NaCl). Disodium hydrogen phosphate (Na_2HPO_4) was used to prepare the disodium phosphate solution, while the DTNB reagent was prepared with 5,5'-dithio-bis (2-nitrobenzoic acid) and sodium citrate.

Reduced glutathione standards, ranging from 1 to 60 mg/dl, were used to generate the calibration curve. Both standards and tissue samples were analyzed at 412 nm using a UV-160 Shimadzu spectrophotometer against a blank. Findings were reported in terms of mg of GSH per gram of wet tissue [11].

NO•(nitrous oxide) Measurement Method

Nitric oxide (NO) concentrations in tissue samples were quantified according to the method of Navarro-Gonzalves et al. Initially, proteins were removed from the supernatant. A glycine–sodium hydroxide (NaOH) buffer was prepared, and copper sulfate ($CuSO_4$) was added to activate cadmium granules. A sulfanilamide solution containing 37 % hydrochloric acid together with sulfanilamide was used, while N-(1-naphthyl) ethylenediamine dihydrochloride (NED) reagent was obtained by mixing N-(1-naphthyl) ethylenediamine dihydrochloride. Standard solutions ranging from 2 to 80 µg were prepared with sodium nitrite ($NaNO_2$).

For analysis, NO levels in tissues were evaluated using the Griess assay. Standards and samples were read in an enzyme-linked immunosorbent assay (ELISA) microplate reader set at 540 nm. Concentrations were automatically calculated by the instrument, and results were expressed as µM per gram of wet tissue [12].

Statistical Methods

All data were analyzed using the Statistical Package for the Social Sciences (SPSS, version 21). The distribution of univariate variables was checked with the Kolmogorov–Smirnov and Shapiro–Wilk tests, whereas multivariate normality was examined using the Mardia, Doornik, and Omnibus tests, along with coefficients of variation. Variables consistent with a normal distribution were assessed using parametric methods, while non-parametric techniques were applied when normality assumptions were not satisfied.

For comparisons among groups, one-way ANOVA (Brown–Forsythe) and one-way ANCOVA were employed, and post hoc analyses were carried out with LSD and Games–Howell tests. For non-parametric variables, the Kruskal–Wallis H test with Monte Carlo simulation was used, followed by pairwise comparisons according to the method of Miller (1966).

Categorical variables were compared using the Pearson Chi-square test or Fisher's exact test with Monte Carlo simulation. Continuous variables were summarized as mean ± standard deviation (SD) or as median with interquartile range (IQR), while categorical variables were described as numbers and percentages. All analyses were performed within a 95% confidence interval, and p-values < 0.05 were regarded as statistically significant.

Results

Liver Hepatocyte Injury

No hepatic cell injury was detected in the control animals. In the paracetamol group, two rats exhibited mild hepatocellular alterations, while no moderate or severe injury was observed. In the amifostine-treated group, hepatocyte integrity remained preserved without any detectable injury. In the NAC group, mild hepatocyte damage was noted in a single rat, whereas moderate injury appeared in two animals from the paracetamol + amifostine group.

In the paracetamol + NAC group, three animals exhibited varying degrees of hepatocellular damage, including one case each of mild, moderate, and severe injury. In contrast, in the paracetamol + amifostine + NAC group, only one rat showed minimal hepatocyte involvement. Severe hepatocellular injury was observed exclusively in the paracetamol + NAC group. Overall, following the control group, the lowest frequency of hepatocyte injury was recorded in the paracetamol + amifostine + NAC group.

NO•(nitrous oxide) Measurement Method

In the paracetamol + amifostine group, two rats exhibited moderate liver regeneration. In the paracetamol + NAC group, mild regeneration was detected in one animal and moderate regeneration in three animals. No evidence of liver regeneration was identified in the remaining groups.

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Spotty Necrosis in Liver

In the paracetamol group, two rats exhibited mild spotty necrosis. In the NAC group, one animal showed mild necrosis, while in the paracetamol + amifostine group, two rats demonstrated mild to moderate changes. Within the paracetamol + NAC group, mild necrosis was identified in two rats and moderate necrosis in one rat. In the paracetamol + amifostine + NAC group, minimal spotty necrosis was observed in two animals (Table 1).

According to the results, the decline in tissue NO concentrations detected in the paracetamol + NAC + amifostine group suggests a potential protective effect of amifostine (Table 2).

Table 1. It is possible to compare hepatocyte injury, regeneration, and localized necrosis among groups

Histopathological parameter		Sham n(%)	Paracetamol n(%)	Amifostine n(%)	NAC n(%)	Paracetamol+ amifostine n(%)	Paracetamol+NAC n(%)	Paracetamol+ NAC+amifostine n(%)
Hepatocyte Injury	Mild (+)	-	2(25%)	-	1(12.5%)	-	1(12.5%)	1(12.5%)
	Moderate (++)	-	-	-	-	2(25%)	1(12.5%)	-
	Severe (+++)	-	-	-	-	-	1(12.5%)	-
Regeneration	Mild(+)	-	-	-	-	2(25%)	3(37.5%)	-
	Moderate(++)	-	-	-	-	-	-	-
	Severe (+++)	-	-	-	-	-	-	-
Spotty Necrosis	Mild(+)	-	2(25%)	-	1(12.5%)	-	2(25%)	2(25%)
	Moderate (++)	-	-	-	-	2(25%)	1(12.5%)	-
	Severe (+++)	-	-	-	-	-	-	-

Table 2. Statistical analysis of indicators of cell injury in blood and tissues

Biochemical parameter	Control	Paracetamol	Amifostine	NAC	Paracetamol+ amifostine	Paracetamol+ NAC	Paracetamol+NAC+ amifostine	Pairwise comparisons
Liver NO(µM/g)*	0.5±0.1a	0.4±0.1	0.4±0.1	0.7±0.2	0.4±0.1	0.5±0.1b	0.3±0.1c	a vs b > 0.005, a vs c < 0.001
Liver GSH(mg/g)*	7.1±0.7a	8.5±2.8	2.4±0.4	0.7±0.0	5.6±2.8	4.8±1.0b	2.4±0.5c	a vs b = 0.002, a vs c p < 0.001

Discussion

This research underscores the importance of exploring alternative therapeutic options for paracetamol-induced liver injury, with particular emphasis on the potential contribution of amifostine. Previous experimental studies have induced paracetamol intoxication and compared outcomes with NAC, while others have examined the protective effects of NAC and various alternative agents against kidney and liver injury through different mechanisms [13-15]. More recently, amifostine has been evaluated in a preclinical setting of acetaminophen-related hepatotoxicity, where it demonstrated hepatoprotective properties [8].

Impaired innate immunity, together with the development of sterile inflammation in the hepatic parenchyma, are major contributors to the progression of liver insufficiency following toxic paracetamol exposure. Early mitochondrial dysfunction results in hepatocyte death and the release of damage-associated molecules, including DNA fragments, heat shock proteins, and high-mobility group box 1 protein, which subsequently activate receptors on Kupffer cells. Upon activation, these cells release various cytokines and chemokines, promoting the recruitment of neutrophils and monocytes and thereby exacerbating tissue damage.

It is important to note that hepatocellular injury is mediated not only by infiltrating immune cells but also by cytokine- and chemokine-driven intracellular processes within hepatocytes. Such pathways involve upregulation of nitric oxide synthase and induction of acute-phase proteins, such as heat shock proteins and heme oxygenase-1. Recent findings also suggest that sterile inflammation may exert beneficial roles by facilitating the removal of cellular debris and promoting hepatocyte proliferation and tissue regeneration [16]. In our study, increased serum NO levels accompanied by decreased hepatic NO concentrations in the paracetamol + NAC + amifostine group compared with other groups may indicate a hepatoprotective effect of amifostine.

N-acetylcysteine (NAC) remains the sole clinically approved antidote for acetaminophen-induced hepatotoxicity, showing maximal benefit when given within the first eight hours of overdose [17]. Although NAC provides therapeutic effects even after 24 hours, its efficacy markedly diminishes with time [18]. The principal protective mechanism of NAC involves acting as a precursor for GSH biosynthesis and aiding in the detoxification of the reactive metabolite N-acetyl-p-benzoquinone imine (NAPQI) during metabolism [19]. In later

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stages, GSH contributes to limiting mitochondrial reactive oxygen species, while elevated doses of NAC further sustain mitochondrial function by supplying intermediates to the Krebs cycle [20]. In our research, the observation of increased serum GSH levels together with decreased tissue GSH concentrations in the paracetamol, NAC, and amifostine group—relative to the paracetamol-only and paracetamol + NAC groups—suggests that amifostine may either enhance GSH bioavailability or provide stronger support for its induction compared with paracetamol alone.

Overall, the findings demonstrated that the paracetamol + amifostine + NAC group showed statistically superior outcomes compared with both the paracetamol + NAC and paracetamol + amifostine groups. Accordingly, the combination of amifostine with NAC, administered as a single bolus dose in our experimental model, appeared to be more effective than the standard single-dose NAC therapy for paracetamol intoxication.

The enhanced protective effect observed with the combined administration of amifostine and NAC in preventing hepatic injury—compared with conventional therapy for paracetamol intoxication—underscores the need for further research into the potential efficacy of amifostine in other conditions involving hepatic damage, such as contrast-induced hepatotoxicity or toxin-related liver injury.

Conclusion

In conclusion, the combined use of amifostine and NAC appears to provide a superior therapeutic approach for paracetamol intoxication. Nevertheless, further studies with a broader scope, including the evaluation of maintenance dosing regimens for both agents, are warranted to validate and expand upon these findings.

Acknowledgements

This study was presented as an oral presentation at the Emergency Medicine Congress held in the Turkish Republic of Northern Cyprus on November 17–20, 2022.

Conflict of Interests

The authors declare that there is no conflict of interest in the study.

Financial Disclosure

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Ethical Approval

The Adnan Menderes University Local Ethics Committee for Animal Experiments (ADÜ-HADYEK) on August 1, 2012 (Approval No: B.30.2.A DÜ.0.00.00.00/050.04/2012/030).

References

1. Lee WM. Acetaminophen and the U.S. Acute Liver Failure Study Group: lowering the risks of hepatic failure. Hepatology. 2004;40:6-9.
2. Davidson DG, Eastham WN. Acute liver necrosis following overdose of paracetamol. Br Med J. 1966;2:497-9.
3. Oliver LH, Lewis SN. Acetaminophen. In: Tintinalli JE, ed. Emergency Medicine. 7th ed. New York, NY: McGraw-Hill; 2010:1246-52.
4. Bond GR, Novak JE. The human and economic cost of paracetamol (acetaminophen) overdose. Pharmacoeconomics. 1995;8:177-81.
5. Liao J, Lu Q, Li Z, et al. Acetaminophen-induced liver injury: molecular mechanism and treatments from natural products. Front Pharmacol. 2023;14:1122632.
6. Akbulut S, Elbe H, Eris C, et al. Cytoprotective effects of amifostine, ascorbic acid and N-acetylcysteine against methotrexate-induced hepatotoxicity in rats. World J Gastroenterol. 2014;20:10158-65.
7. Karimi M, Ghasemzadeh Rahbardar M, Razavi BM, Hosseinzadeh H. Amifostine inhibits acrylamide-induced hepatotoxicity by inhibiting oxidative stress and apoptosis. Iran J Basic Med Sci. 2023;26:662-8.
8. Rasouli H, Razavi BM, Ghasemzadeh Rahbardar M, et al. Hepatoprotective effect of amifostine and WR-1065 on acetaminophen-induced liver toxicity in Wistar rats. Naunyn Schmiedebergs Arch Pharmacol. 2024;397:6001-15.
9. Hendrickson RG, Bizovi KE. Acetaminophen. In: Flomenbaum NE, Goldfrank LR, Hoffman RS, Howland MA, Lewin NA, Nelson NA, eds. Goldfrank's Toxicologic Emergencies. 8th ed. New York, NY: McGraw-Hill; 2006:333-43.
10. Feng M, Smith DE, Normolle DP, et al. A phase I clinical and pharmacology study using amifostine as a radioprotector in dose-escalated whole liver radiation therapy. Int J Radiat Oncol Biol Phys. 2012;83:1441-7.
11. Beutler E, Duron O, Kelly BM. Improved method for the determination of blood glutathione. J Lab Clin Med. 1963;61:882-8.
12. Navarro-Gonzálvez JA, García-Benayas C, Arenas J. Semiautomated measurement of nitrate in biological fluids. Clin Chem. 1998;44:679-81.
13. Kamanaka Y, Kawabata A, Matsuya H, et al. Effect of a potent iNOS inhibitor (ONO-1714) on acetaminophen-induced hepatotoxicity in the rat. Life Sci. 2003;74:793-802.
14. Fakurazi S, Nanthini U, Hairuszah I. Hepatoprotective and antioxidant action of Moringa oleifera Lam. against acetaminophen-induced hepatotoxicity in rats. Int J Pharmacol. 2008;4:270-5.
15. Terneus MV, Brown JM, Carpenter AB, Valentovic MA. Comparison of S-adenosyl-L-methionine (SAMe) and N-acetylcysteine (NAC) protective effects on hepatic damage when administered after acetaminophen overdose. Toxicology. 2008;244:25-34.
16. Jaeschke H, Williams CD, Ramachandran A, Bajt ML. Acetaminophen hepatotoxicity and repair: the role of sterile inflammation and innate immunity. Liver Int. 2012;32:8-20.
17. Larson AM. Acetaminophen hepatotoxicity. Clin Liver Dis. 2007;11:525-48.
18. Smilkstein MJ, Knapp GL, Kulig KW, Rumack BH. Efficacy of oral N-acetylcysteine in the treatment of acetaminophen overdose: analysis of the national multicenter study (1976 to 1985). N Engl J Med. 1988;319:1557-62.
19. Corcoran GB, Racz WJ, Smith CV, Mitchell JR. Effects of N-acetylcysteine on acetaminophen covalent binding and hepatic necrosis in mice. J Pharmacol Exp Ther. 1985;232:864-72.
20. Saito C, Zwingmann C, Jaeschke H. Novel mechanisms of protection against acetaminophen hepatotoxicity in mice by glutathione and N-acetylcysteine. Hepatology. 2010;51:246-54.