ORIGINAL ARTICLE

Synergistic Effect of Noise and Toluene Upon Hematological Alterations, Pulmonary Oxidative Stress and Inflammation

Effet Synergique du Bruit et du Toluène sur les Altérations Hématologiques, le Stress Oxydatif Pulmonaire et l'Inflammation

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Abstract

Aim: Study the synergistic effects of noise-toluene co-exposure on hematological parameters, oxidative stress and pulmonary architecture in rats.
Methods: 24 male Wistar rats were divided into four groups: control, noise exposure, toluene exposure and co-exposure noise-toluene. Biological analyses were performed to determine hematological parameters and evaluate markers of oxidative stress. Histopathological sections were observed to assess pulmonary tissue damage and the degree of inflammatory response.
Results: Hematological analysis revealed a significant decrease in red blood cells (RBCs), accompanied by an increase in white blood cells (WBCs) and platelets. Histological examination of lung tissues revealed inflammation with alveolar wall damage and peribronchial immune cell infiltration in the co-exposed group, which correlated with increased vascular permeability and pulmonary edema. Exposure to toluene and noise resulted in significant disruption of the pulmonary tissue structure accompanied by oxidative stress.
Conclusion: The results suggest that combined exposure to toluene and noise causes structural and functional changes in lung tissues and alterations in hematological parameters.

Keywords: Noise - Toluene - Lung – Coexposition - Oxidative stress - Pulmonary inflammation

Résumé

Objectif : Etudier les effets synergiques de la co-exposition bruit-toluène sur, les paramètres hématologiques, le stress oxydatif et sur l'architecture pulmonaire chez le rat.
Méthode : 24 rats mâles Wistar ont été divisés en quatre groupes : témoin, exposition au bruit, exposition au toluène et co-exposition bruit-toluène. Des analyses biologiques ont été réalisées pour déterminer les paramètres hématologiques et évaluer les marqueurs du stress oxydatif. Des coupes histopathologiques ont été observées pour évaluer les lésions tissulaires pulmonaires et le degré de réponse inflammatoire.
Résultats : La coexposition bruit-toluène a entraîné une augmentation significative de la peroxydation lipidique et une diminution de l'activité de la catalase et de la SOD. L'analyse hématologique a révélé une diminution significative des globules rouges, accompagnée d'une augmentation dans le nombre de globules blancs et de plaquettes. L'examen histologique des tissus pulmonaires a révélé une inflammation avec des lésions des parois alvéolaires et une infiltration de cellules immunitaires péribronchiques dans le groupe co-exposé, ce qui était corrélé à une augmentation de la perméabilité vasculaire et à un œdème pulmonaire.
Conclusion : Les résultats suggèrent que l'exposition combinée au toluène et au bruit entraine un changement structural et fonctionnel au niveau des tissus pulmonaires et des modifications dans les paramètres hématologiques.

Mots clés: Bruit- Toluène- Poumons- Coexposition - Stress oxydatif- Inflammation pulmonaire.

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Introduction

Simultaneous exposure to noise and toluene is prevalent in industrial settings, representing a significant occupational hazard across sectors, such as manufacturing, construction, and industries involving solvent use, including painting (1,2). Workers are frequently subjected to this dual exposure, necessitating a comprehensive understanding of potential health implications.

Combined exposure presents a significant risk as it can elicit adverse effects on multiple physiological systems (3-5). The renal system is also at considerable risk, with evidence indicating that toluene exposure may induce nephrotoxicity, characterized by tubular damage, glomerular dysfunction, and alterations in renal oxidative stress markers (6,7). Moreover, the central nervous system is highly susceptible to the neurotoxic effects of both noise and toluene, potentially resulting in cognitive impairment, neurobehavioral deficits, and disruption of neurotransmitter homeostasis. These neurotoxic effects may involve oxidative stress, neuroinflammation, and apoptosis, which collectively impair neural function and contribute to the development of neurodegenerative disorders (8,9,5). The cardiovascular system is particularly susceptible to the combined effects of noise and toluene exposure, which can result in hypertension and alterations in heart rate variability. Noise exposure induces endothelial dysfunction, a factor that contributes to the development of atherosclerosis and other cardiovascular diseases. Oxidative stress is a critical mediator of the adverse effects associated with exposure to noise and toluene. (10,4,11).

These stressors lead to excessive generation of reactive oxygen species (ROS), which overwhelm the body's antioxidant defenses and cause cellular damage. This oxidative imbalance is a fundamental driver of inflammatory processes that contribute to tissue injury and progression of chronic diseases. In particular, given their extensive exposure to both oxygen and environmental pollutants, the lungs are highly vulnerable to oxidative damage. This underscores the importance of understanding the interplay between oxidative stress and respiratory health in the context of industrial exposure. (12-14)

This study aims to evaluate pulmonary inflammation, oxidative stress, and histological alterations induced by concurrent exposure to 85 dB(A) noise and 300 ppm inhaled toluene in rats.

Methods

Animals and treatment

Twenty-four male Wistar rats obtained by the Pasteur Institute of Tunis were randomly divided into four groups (Control group, Noise group, Toluene group and Noise-Toluene group) of six animals each and maintained in animal housing a week before the start of the experiments to ensure an adaptation period. Animals were placed in number of two per cage to avoid stress factors and received a commercial pellet diet (Industrial Society of Food, Sfax, Tunisia) and water ad libitum. Rats were kept under closely controlled environmental conditions (12 h light/ dark cycle, room temperature 23 ± 1°C).

The control group was also transferred to a separate chamber located in a noise-free environment, with background noise levels maintained below 40 ± 5 dB (A) and free from toluene exposure. Noise group exposed to 85 dB (A), toluene group exposed to 300 ppm and combined group exposed to noise and toluene.

All experimental procedures followed the guidelines outlined in the Care and Use of Animals and were approved by the Ethics Committee of the National School of Veterinary Medicine of Sidi Thabet (Réf: 162020/FMT) according to the International Council for Laboratory Animal Science (ICLAS) recommendations.

Experimental Protocol

Following the established protocol described in our recent study by Ben Attia et al. (4), rats were subjected to daily exposure to noise and/or inhaled Toluene for six weeks (5 days/week) from 8:00 am to 2:00 pm. This exposure was conducted within specially designed Plexiglas chambers, incorporating a generation system, an exposure system, and a monitoring system, as detailed by Hinners et al. (15).

Noise Exposure

The acoustic exposure system was established using the Audacity 2.3.2 audio software, where the noise level was consistently set at 85dB (A) to deliver an octave-band noise (8-16 kHz) through a sound speaker. This speaker was uniformly positioned to ensure that all rats were subjected to the same intensity of noise. The noise level was meticulously monitored with an integration sound level meter with Class 1 accuracy, Type 2238 Bruel and Kjaer, ensuring precise exposure as per the established protocol (16).

Toluene Exposure

A concentration of 300 ± 10 ppm was maintained for six hours within the exposure chamber. This was achieved by injecting liquid toluene (SMSBio, Sud médical services, Tunisia) using an isocratic pump into a mixing vessel connected to the chamber. Air circulation was facilitated by a fan at the inlet, and an adjustable airflow was ensured by a centrifugal fan at the outlet. The toluene concentration was continuously monitored and analyzed using gas chromatography-mass spectrometry (Agilent Technologies 6890 N Network GC System). The chamber also featured several circular openings to allow researchers to monitor and sample the toluene levels, with the temperature kept stable at 23±1°C, monitored via a digital thermometer.

To explore the potential interactions between noise and toluene, a co-exposure experiment was conducted, following a protocol similar to the individual exposure experiments for both noise and toluene. In this setup, the simultaneous generation of noise and toluene allowed

for the examination of their combined effects on the subjects, providing valuable insights into the interaction between these two environmental stressors.

Sample collection

At the end of the experiment, rats were anesthetized using ketamine chlorhydrate (50 mg/kg) and sacrificed by decapitation.

Hematological Parameters
Hematological analyses, including measurements of red and white blood cells and platelets, were conducted using an I-sens automated system (i-Smart 30 Pro Electrolyte Analyzer, UK).

Biochemical Assays in Plasm
Biochemical assays for parameters such as cholesterol, triglycerides, lactate dehydrogenase (LDH), and creatine kinase (CK) were performed on plasma samples utilizing the COBAS INTEGRA 400 Plus automated analyzer (Switzerland), following established standard procedures.

Lung Wet/Dry Weight Ratio
To assess lung edema, the right lung of each rat was excised, washed with a 0.9% NaCl physiological solution, and weighed. Subsequently, the lung tissue samples were dried at 80 °C for 48 hours to determine the dry weight and calculate the wet/dry ratio. This parameter was calculated using the methodology outlined by Kouki et al. (17).

Histological study
The inferior lobe of the left lung was bisected for histopathological examination and assessment of oxidative stress markers. Rats lungs were perfused and immersed in the fixative solution (10 % neutral buffered formalin) for 48 h, dehydrated through graded series of ethanol, embedded in paraffin, sectioned with a microtome (Leica Biosystems RM2245, Great Britain) to obtain 4-5 µm-thick paraffin sections and then stained with hematoxylin and eosin (H&E).

Oxidative stress markers determination
A portion of the lung was weighed and washed with ice-cold saline immediately and kept at -80 °C until analysis. The lung tissue was homogenized (ULTRA-TURRAX® IKA-WERK, Germany) in ice-cold PBS buffer (pH =7.2) and centrifuged for 20 min (10,000 rpm at 4 °C). The supernatants were collected and maintained at -80 °C until subsequent biochemical analysis.

Lipid peroxidation determination
The determination of lipid peroxidation (LPO) was based on the detection of malondialdehyde (MDA), which was determined using the Buege and Aust (18) method. Samples were mixed with a BHT-TCA solution (1% -20 %, v/v) and centrifuged at 1,000 rpm for 10 min. The supernatant was blended with TBA-Tris solution (120 mM- 26 mM, v/v) in an acidic solution (0.6 M HCl). The mixture was heated at 100 °C for 15 minutes and then cooled at room temperature. Absorbance was measured at 532 nm using a UV-spectrophotometer (Labomed, UV-2650, Inc, USA).

Catalase activity
Catalase (CAT) activity was measured according to the method of Aebi et al. (19). This assay is based on the ability of this enzyme to degrade hydrogen peroxide (H2O2), which results in a decrease in the absorption of the reaction mixture at 240 nm.

Superoxide dismutase activity
Superoxide dismutase (SOD) activity was evaluated as described by Beyer and Fridovich (20) using epinephrine (50 mM carbonate buffer, pH 10.2) and bovine catalase (0.4 U/mL). This method is based on the inhibition of the auto-oxidation of epinephrine to adrenochrome in the presence of SOD. Absorbance was measured at 480 nm.

Total protein level determination
The protein concentration in lung samples was determined using the Bradford method (21) with bovine serum albumin (BSA) as the standard. Absorbance readings were measured at 595 nm.

Statistical analysis
The results are expressed as mean ± standard error of mean the data collected were statistically analyzed using Mann-Whitney U test. A value of $P < 0.05$ was considered to be significant.

Result

Effects of noise and toluene exposure on hematological parameters

Table1 summarizes the hematological parameters for the control and exposed rats.

Table 1. Complete blood counts

Groups	Red Blood Cells ($\times 10^6$ µL)	White Blood Cells ($\times 10^3$ µL)	Blood platelets ($\times 10^3$ µL)
Control	8.42 ± 0.36	8.20 ± 0.89	871.50 ± 53.37
Noise exposure	8.41 ± 0.70	11.86 ± 1.14 *	1217.33 ± 83.82 *
Toluene exposure	6.63 ± 0.62 *	12.16 ± 1.41 *	987 ± 33.66 *
Co-exposure Noise-Toluene	6.56 ± 0.80 *	13.65 ± 2.38 *	1407.17 ± 155.25 *

* p< 0.05 vs control group

The data reveal a significant reduction in red blood cell (RBC) counts in the toluene exposure and coexposure groups relative to the control group, with counts of 6.63 ± 0.62 × 106/µL and 6.56 ± 0.80 × 106/µL compared to 8.42 ± 0.36 × 10^6/µL ($p < 0.05$ for both comparisons). Furthermore, there was a significant increase in white blood cell (WBC) counts across all exposure groups compared to the control group. Platelet counts were also significantly elevated in the all exposure groups

compared to the control group. These findings indicate significant alterations in hematological parameters.

Effects of noise and toluene exposure on plasma Cholesterol, Triglycerides, LDH, and CK Levels

Table 2 presents the plasma concentrations of cholesterol, triglycerides (TG), lactate dehydrogenase (LDH), and creatine kinase (CK) in rats

Table 2. Plasma biochemical parameters of different group

Groups	Cholesterol (mmol/L)	Triglyceride(mmol/L)	Lactate dehydrogenase(U/L)	Creatine kinase (U/L)
Control	1.20± 0.08	0.9 ± 0.08	344.33 ± 50.53	368.5 ± 27.31
Noise exposure	1.31± 0.07 *	1.3 ± 0.07 *	365 ± 21.93	1489.5 ± 139.21*
Toluene exposure	1.15± 0.08 €	1.045 ± 0.08	583 ± 54.01 *	1430 ± 169.68 *
Co-exposure Noise- Toluene	1.45± 0.05 *	1.4 ± 0.05 *	604.5 ± 112.65 *	1857.83 ±142.39 *

* p< 0.05 vs control group, € p< 0.05 vs noise-toluene group

In the control group, the mean total cholesterol concentration was 1.20 ± 0.08 mol/L. This value was significantly elevated in the noise group and co-exposure group, with concentrations of 1.31 ± 0.07 mol/L and 1.45 ± 0.05 mol/L, respectively ($p < 0.05$ for both comparisons). Similarly, triglyceride levels were 0.9 ± 0.08 mmol/L in the control group and increased significantly in the noise group and coexposed group to 1.3 ± 0.07 mmol/L and 1.4 ± 0.05 mmol/L, respectively ($p < 0.05$ for both comparisons).

Additionally, plasma LDH levels were significantly higher in the toluene group and co-exposure group, with values of 583 ± 54.01 U/L and 604.5 ± 112.65 U/L, respectively, compared to the control group (344.33 ± 50.53 U/L; $p < 0.05$ for both comparisons). Noise exposure alone also significantly increased total CK concentration from 368.5 ± 27.31 U/L in the control group to 1489.5 ± 139.21 U/L ($p < 0.05$). The combination of noise and toluene further elevated CK levels to 1857.83 ± 142.39 U/L ($p \leq 0.05$).

Macroscopic evaluation of pulmonary tissue

Macroscopic analysis (Figure 2) of lung tissues from the control group revealed a uniform pink coloration and smooth surface texture, with no signs of edema, hemorrhage, or other visible abnormalities, indicative of normal pulmonary tissue. In contrast, the lungs of the noise-exposed rats displayed significant pathological changes, including pronounced edema and a noticeable darkening of the tissue, accompanied by an uneven surface and areas of discoloration, suggesting compromised pulmonary function. Additionally, the lungs of rats exposed to toluene and those co-exposed to both toluene and noise exhibited distinctive pathological features, such as localized hemorrhagic foci, and extensive lung edema. These findings were markedly different from the normal appearance observed in the control group.

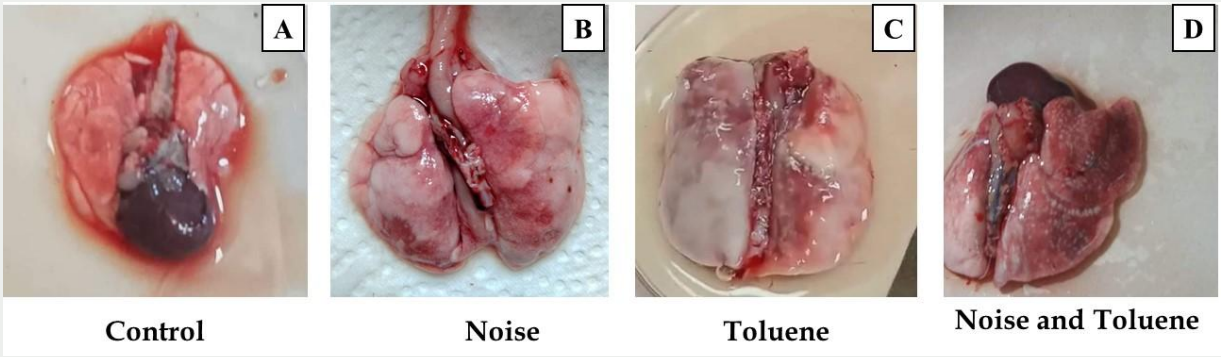

Figure 2. Macroscopic observation in rat lungs. (A) Control group, (B) Noise exposure group, (C) Toluene exposure group and (D) Co-exposed group of noise and toluene.

Pulmonary edema index assessment

The pulmonary edema index was assessed by measuring the wet/dry weight ratio of the lungs (Table 3).

Table 3. Pulmonary edema index measured by wet/dry weight ratio of lungs

Groups	Wet/Dry Weight Ratio (Mean ± SD)
Control	3.00 ± 0.04
Noise Exposure	4.62 ± 0.12*
Toluene Exposure	5.76 ± 2.01*
Co-Exposure Noise-Toluene	5.95 ± 1.36*

* p< 0.01 vs control group

This analysis revealed a significant increase in the ratio for the exposed groups compared to the control. Specifically, the control group exhibited a ratio of 3 ± 0.04. In comparison, the noise-exposed group had a ratio of 4.62 ± 0.12, the toluene-exposed group showed a ratio of 5.76 ± 2.01, and the group exposed to both noise and toluene demonstrated the highest ratio of 5.95 ± 1.36. These results indicate a marked elevation in pulmonary edema among the exposed groups, highlighting the impact of both individual and combined exposures on lung tissue.

Microscopic analysis of lung tissues

Histopathological examination of lung tissues (Figure 3) from the control group revealed a normal lung architecture, characterized by uniform alveoli and an intact alveolar epithelium. In the noise-exposed group, the lungs exhibited lymphoid nodules with surrounding inflammatory cells around the bronchioles, although the interalveolar septa remained intact, similar to the control group. Conversely, rats exposed to toluene and those subjected to both toluene and noise demonstrated pronounced pathological alterations. These included the accumulation of parenchymal debris, extensive mononuclear cell infiltration within the lung parenchyma, intra-bronchiolar cell debris, and inflammatory cell infiltration around the bronchioles. Additionally, significant fragmentation of the alveolar septa and obstruction of numerous alveoli were observed.

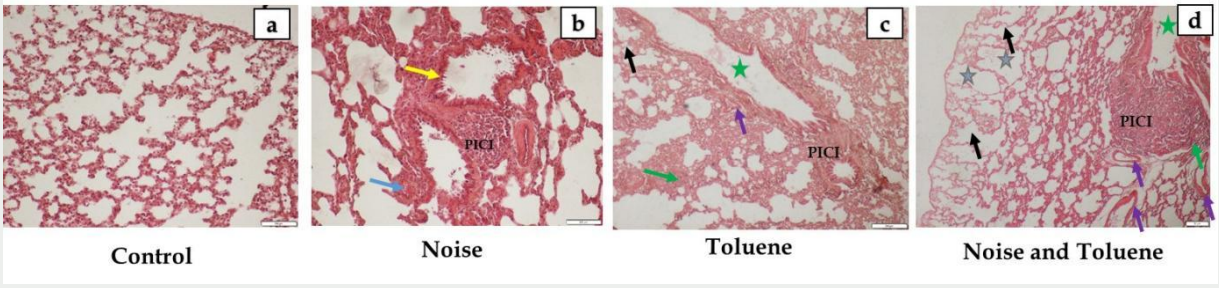

Figure 3. Microscopic observaion of lung tissue following exposure to noise (85 dB(A)) and/or inhalation of toluene (300 ppm). a: Control group; b, c, and d: Exposed group.
Yellow arrows denote inflammatory cells surrounding the bronchioles, purple arrows highlight dilated blood vessels. Black arrows indicate fragmentation of the alveolar septa, and blue arrows point to inflammatory nodules. Green stars indicate intra-bronchiolar cellular debris and gray stars denote interalveolar cellular debris. Magnification: x 10. Scale bar =50 µm.

Effects of noise and toluene exposure on oxidative stress parameters in lung tissue

Table 4 illustrates the results of the MDA assay alongside the activities of superoxide dismutase (SOD) and catalase (CAT) in lung tissues. The MDA assay indicated a significant elevation in lipid peroxidation levels in rats exposed to toluene and those co-exposed to both noise and toluene compared to the control group. Specifically, MDA levels were 1.64 ± 0.258 nmol/mg protein for the toluene group and 1.75 ± 0.138 nmol/mg protein for the co-exposed group, relative to 0.513 ± 0.055 nmol/mg protein in the control group ($p \leq 0.01$). Noise exposure alone resulted in a non-significant increase in MDA levels (0.72 ± 0.089 nmol/mg protein).

Table 4. Oxidative stress parameters in lung tissue

Groups	MDA (nmol/mg prot)	Catalase (U/mg prot)	SOD (U SOD/ min/mg prot)
Control	0.513 ± 0.055	0.135 ± 0.040	6.77 ± 0.627
Noise exposure	0.72 ± 0.089	0.149 ± 0.015	7.24 ± 3.82
Toluene exposure	1.64 ± 0.258 *	0.039 ± 0.003 *	3.012 ± 0.663 *
Co-exposure Noise-Toluene	1.75 ± 0.138*	0.034 ± 0.009 *	2.677 ± 0.489 *

* p< 0.01vs control group

The data of catalase activity revealed a significant decrease in the activity of this enzyme in both the toluene-exposed (0.039 ± 0.003 U/mg protein) and co-exposed groups (0.034 ± 0.009 U/mg protein) compared to controls (0.135 ± 0.040 U/mg protein), with p-values < 0.01. In contrast, noise exposure alone led to a non-significant increase in catalase activity (0.149 ± 0.015 U/mg protein).

Results showed a significant reduction in SOD activity in rats exposed to toluene alone and in combination with noise compared to the control group ($p \leq 0.01$). Specifically, the co-exposed group exhibited a notable decrease in SOD activity (2.677 ± 0.489 U SOD/min/mg protein) relative to the toluene-only group (3.012 ± 0.663 U SOD/min/mg protein) and the control group (6.77 ± 0.627 U SOD/min/mg protein). These results highlight the differential effects of various exposure conditions on oxidative stress markers and antioxidant enzyme activities in lung tissues.

Discussion

Occupational exposure to both toluene and noise poses a significant challenge to respiratory health, highlighting the need for thorough investigation of their combined effects. Toluene, a well-known respiratory toxin, interacts with noise exposure to amplify structural and functional damage to the pulmonary tissues.

This study aimed to unravel the impact of these environmental stressors through a comprehensive approach integrating biochemical assays with detailed histopathological assessments to provide a clearer understanding of their joint influence on respiratory physiology. The observed alterations in hematological parameters following exposure to toluene, noise, and their combination elucidate the systemic stress imposed on the body, particularly in blood physiology. A significant reduction in red blood cell (RBC) count was observed in both toluene and combined toluene and noise groups, suggesting that toluene, whether acting independently or in conjunction with noise, may impair erythropoiesis or enhance hemolysis. This decrease in RBCs could be attributed to oxidative damage affecting erythrocytes or the suppression of bone marrow function, which may contribute to the impaired production of new red blood cells.

Conversely, elevation in white blood cell (WBC) counts across all exposure groups indicated a heightened immune response, likely triggered by the toxic and inflammatory effects of both toluene and noise. The increase in WBCs aligns with previous studies, where rabbits exposed to noise at 100 dB and toluene at 1000 ppm for 14 consecutive days exhibited similar trends: an increase in WBC counts following simultaneous exposure to both agents, and a decrease in RBC counts when exposed to either stressor alone (22).

This immune response and the associated increase in WBCs could be attributed to oxidative stress induced by the combined exposure to toluene and noise. Such stress activates pro-inflammatory pathways and recruits immune cells (23), contributing to the elevated WBC count observed in the noise-toluene group. The findings of this study indicate sequential progression of events wherein chronic exposure to environmental stressors, specifically noise and toluene, elicits an inflammatory response, resulting in elevated white blood cell (WBC) counts. This phenomenon has been documented in previous studies that examined noise exposure (24). The combined inflammatory impact of toluene exposure likely exacerbated this response, as evidenced by the higher WBC counts in the noise-toluene group. Consequently, the interaction of these environmental stressors demonstrates a synergistic effect on the immune and hematological systems of the body, leading to significant alterations in hematological parameters.

The elevated platelet counts observed in the noise, toluene and combined toluene and noise groups further substantiated the concept of systemic stress and inflammation. Platelets, which are crucial for hemostasis, respond actively to vascular injury potentially triggered by endothelial damage resulting from oxidative stress associated with both noise and toluene exposure. Numerous studies have demonstrated that increased platelet counts are prevalent among individuals exposed to occupational noise and industrial solvents, underscoring their contribution to vascular and thrombotic complications under environmental stress conditions (25).

Specifically, the increase in platelet count due to noise exposure can lead to the formation of platelet aggregates, potentially resulting in vascular occlusion, as evidenced by the histological examination of cardiac tissues. Toukh et al. (26) identified a correlation between hypercoagulability and elevated levels of cortisol and corticosterone in plasma, linked to exposure to construction noise. This noise-induced stress is known to elevate blood catecholamines, which in turn stimulates thrombus formation and increases the risk of myocardial infarction (27). Collectively, these findings elucidate the complex interplay between environmental stressors and their impact on hemostatic processes, emphasizing the importance of monitoring platelet dynamics in the context of exposure to noise and toluene. Our study revealed that exposure to noise, toluene, or their combination induced significant alterations in plasma biochemical parameters, underscoring the deleterious effects of these environmental stressors on metabolic health. The significant increases in total cholesterol and triglyceride levels indicated that both noise and toluene may contribute to dyslipidemia, which is closely associated with an elevated risk of cardiovascular diseases.

Furthermore, elevated levels of lactate dehydrogenase (LDH) suggest tissue damage or cellular injury, indicating that both toluene exposure and the combined effects of noise and toluene can lead to necrosis in various tissues (28, 29).

Chronic noise exposure can elevate stress hormone levels and promote dyslipidemia by enhancing lipolysis and altering liver metabolism. Concurrently, toluene exposure generates reactive oxygen species (ROS) that exacerbate tissue injury and drive inflammatory processes (30).

Given the significant role of oxidative stress in mediating these effects, it is imperative to further investigate oxidative stress parameters in pulmonary tissue. This will allow us to elucidate how these environmental stressors contribute to oxidative damage in the lungs, and their potential implications for respiratory health.

Our findings revealed that exposure to toluene, either alone or in combination with noise, significantly increased the levels of malondialdehyde (MDA) in pulmonary tissue, coupled with a decrease in the activities of catalase and superoxide dismutase (SOD) enzymes. These observations underscore the oxidative damage induced by toluene exposure, especially when combined with an additional environmental stressor such as noise. Elevated MDA levels in the pulmonary tissue indicate heightened lipid peroxidation, a key marker of oxidative stress.

The significant reduction in catalase and SOD activities further illustrates the compromised antioxidant defense in the lung tissue of the exposed rats. Catalase and SOD are critical enzymes involved in ROS detoxification. SOD catalyzes the dismutation of superoxide radicals into hydrogen peroxide, which is subsequently decomposed by catalase (31). A reduction in these enzymatic activities suggests that the antioxidant system is overwhelmed by excessive ROS generated in response to toluene and noise exposure. This depletion of antioxidant defenses could enhance cellular susceptibility to oxidative damage, leading to potential long-term consequences, such as fibrosis, inflammation, and even carcinogenesis

in pulmonary tissue.

Previous studies have reported similar patterns of oxidative stress in response to volatile organic compound (VOC) exposure, indicating that these environmental pollutants disrupt the pulmonary redox balance. However, this study provides unique insights into the combined effects of VOCs and noise, demonstrating a synergistic effect on oxidative stress markers, which could have significant implications for individuals subjected to multiple environmental stressors. These findings suggest that public health interventions should consider cumulative exposure risks, particularly in occupational settings, where simultaneous exposure to chemical and noise stressors is prevalent.

The results, which highlight oxidative stress as evidenced by elevated MDA levels and decreased catalase and SOD activities, suggest a primary pathway of lung tissue damage following toluene exposure, with this effect being intensified by concurrent noise exposure. These findings were corroborated by histological analysis, macroscopic observations, and permeability index measurements, which revealed significant inflammatory and structural alterations in pulmonary tissue. Histological analysis revealed notable peribronchial inflammatory cell infiltration, lymphoid nodule formation, and alveolar wall damage. Additionally, macroscopic examination revealed clear signs of inflammation, such as edema, hemorrhagic areas, and pus, while an elevated permeability index confirmed endothelial disruption and edema. Collectively, these observations corroborate the oxidative stress results, illustrating how oxidative damage translates into structural and inflammatory injury within lung tissue.

The observed edema, validated by permeability index measurements, is a critical marker of inflammation-driven damage; compromised endothelial barriers permit fluid leakage into interstitial and alveolar spaces (32). This fluid accumulation exacerbates tissue hypoxia and impairs gas exchange, further disrupting lung function (33). Macroscopic signs of inflammation, hemorrhage, pus formation, and tissue discoloration along with histological evidence of immune cell infiltration and lymphoid nodule formation indicate an aggressive immune response.

The combined oxidative and inflammatory processes compromise lung structural integrity, as evidenced by alveolar wall damage and intrabronchial debris, indicating a significant breakdown in the tissue architecture.

Together, these findings confirm and strengthen the hypothesis that combined exposure to toluene and noise potentiates oxidative and inflammatory responses, resulting in pulmonary dysfunction. This synergistic damage highlights the necessity for comprehensive exposure guidelines, especially in occupational settings where combined exposure is common. Future studies should explore additional oxidative and immune markers to further elucidate these mechanisms and establish a more robust foundation for interventions to protect against environmental stress.

This study posits that exposure to toluene, particularly in combination with noise, leads to a multifaceted pathological response characterized by oxidative stress and inflammation. The resultant dysregulation of hematological parameters, lipid profiles, and enzymatic activities suggests that these exposures may disrupt normal physiological processes, leading to significant metabolic dysfunction. The interplay between oxidative stress and inflammatory pathways may be a key driver of the observed changes, potentially contributing to the development of various health complications associated with environmental stress.

Conclusion

Combined exposure to noise and toluene induced several effects and a range of pathological changes by inducing inflammatory pathways than its components alone.

There are a number of industries in which combined exposure to various physical and chemical factors is quite common. However, a multidimensional approach should be necessary to mitigate health problems related to combined exposures (physical and chemical factors), this approach should emphasize technological advancements, public awareness, and changes in behavior and stringent regulatory measures. By adopting these strategies, society can effectively reduce the impact of combined exposure on health and improve the overall quality of life.

Perspective

To effectively mitigate these health risks, it is crucial to integrate environmental and occupational health strategies. Future research should focus on elucidating the underlying mechanisms of these interactions and developing targeted interventions to safeguard workers health.

- Based on these findings, the Occupational Safety and Health Administration may be directed to update workplace guidelines and protect workers by implementing strategies such as mandatory use of personal protective equipment, improved ventilation and training workers to detect the effects of noise and/or chemicals early.
- The most relevant question for the authorities is whether current workplace exposure standards for noise and/or toluene give adequate protection against damage caused by these scourges.

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