Journal of Information Systems Engineering and Management
2025, 10(48s)
e-ISSN: 2468-4376
https://www.jisem-journal.com/

Research Article

Formic Acid Burns Induce Progressive Oxidative Stress and Inflammation: A Pilot Comparative Study with Thermal Burns

Kamal Anas,[1*] Yefta Moenadjat,[2] Insan Sosiawan Tunru,[3] Diniwati Mukhtar,[4] Himmi Marsiati,[4] Ekowati Handharyani,[5] Arni Diana Fitri,[5] Indra Kusuma,[6] Aan Royhan,[7] Marisa Riliani[7], Sri Rahayu Ningsih.[7]

[1] Department of Surgery Faculty of Medicine Universitas YARSI (kamal.anas@yarsi.ac.id)
[2] Department of Surgery, Faculty of Medicine Universitas Indonesia (yefta.moenadjat@ui.ac.id)
[3] Department of Pathological Anatomy Faculty of Medicine Universitas YARSI (insan.sosiawan@yarsi.ac.id)
[4] Doctoral Program in Biomedical Sciences of Universitas YARSI (diniwati.mukhtar@yarsi.ac.id) (himmi.marsiati@yarsi.ac.id)
[5] Teaching Animal Hospital of Institut Pertanian Bogor (ekowatieko@apps.ipb.ac.id), (arnifi@apps.ipb.ac.id)
[6] Department of Physiology Faculty of Medicine Universitas YARSI (indra.kusuma@yarsi.ac.id)
[7]Department of Anatomy Faculty of Medicine Universitas YARSI (aan.royhan@yarsi.ac.id) (s.ningsih@yarsi.ac.id)
*Corresponding author (aan.royhan@yarsi.ac.id)

ARTICLE INFO

Received: 29 Dec 2024

Revised: 12 Feb 2025

Accepted: 27 Feb 2025

ABSTRACT

Background: Formic acid is a corrosive chemical used in industry that may cause severe chemical burns. Contact with this corrosive agent shows a distinct characteristic of thermal burn. This pilot study aimed to investigate whether a short 20–second exposure to formic acid is sufficient to induce skin damage.

Methods: An experimental burn proceeded using porcine as a model. Skin damage following exposure to formic acid for 20 seconds was compared to thermal burn. Histo-morphological change, inflammatory pathway, and oxidative stress were the variables of interest.

Results: Heated metal exposure caused immediate coagulative necrosis with eschar formation, whereas formic acid exposure led to progressive tissue discoloration and delayed necrosis. Histopathological analysis revealed epidermal disorganization and inflammatory cell infiltration in the formic acid group. NF–κB and HMGB1 expression were significantly increased after formic acid exposure, indicating sustained inflammation. An immediate increase of 8–isoprostane levels peaked 6 hours postburn (heated metal), whereas formic acid led to a gradual but persistent increase of 8–isoprostane.

Conclusion: A short 20–second exposure to formic acid can induce oxidative stress and inflammation in porcine skin, with a delayed but progressive injury pattern distinct from thermal burns. These findings provide forensic and clinical insights into chemical burn pathophysiology, warranting further investigation into long–term effects and therapeutic interventions.

Keywords: Formic acid burns, Oxidative stress, 8–Isoprostane, HMGB1, NF–κB

INTRODUCTION

Formic acid, a corrosive liquid supposed to be used to solidify the latex – the raw material – of the para rubber tree, leads to chemical burn following contact, a progressive type to deeper skin layers and beneath, an also systemic effect.[1–3] Epidemiologically, the incident is highly infrequent, as all types of chemical burns ranged between 3–5%.[4] However, Indonesia is the world's largest producer of natural rubber, with vast plantations spread across the country. Formic acid is widely spread, easily found in the market, and has a great potential for misuse. The misuse referred to an emerging issue in forensic medicine regarding workplace accidents in the rubber industrial field and intentional assaults.[1,2,5–8]

Unlike thermal burns with immediate and predictable injury patterns, formic acid burns continue to worsen over time with a definite slow progression followed by chronic inflammatory response, delayed healing, and increased morbidity.[9,10] The emerging problem underscores the need for a deep understanding of the patho-mechanism of

Journal of Information Systems Engineering and Management
2025, 10(48s)
e-ISSN: 2468-4376
https://www.jisem-journal.com/

Research Article

formic acid–induced burns. However, despite the profound impact, studies and references remain limited. Most studies focused on acid and chemical burns from a general perspective, but not each–acid specific.[11] To the author's knowledge, formic acid has unique chemical properties that contribute to its aggressive tissue penetration and prolonged inflammatory response.[9] A better understanding of the biological mechanisms, particularly oxidative stress and inflammatory response in molecular pathways, is crucial for developing strategies and targeted treatment. Thus, a study on skin–tissue damage, inflammatory pathways, and oxidative stress in animal models is needed.

METHOD

This experimental study proceeded using a healthy 20 week aged 50 kg weighted porcine (Sus scrofa domesticus) model as their anatomical and physiological similarities to human skin, including comparable epidermal thickness, dermal composition, and wound healing processes, making them widely accepted model for cutaneous burn studies.[12,13] The study conducted in Teaching Animal Hospital, Institut Pertanian Bogor.

Sample preparation

Prior to the intervention, a two–week acclimatization was applied.[14–16] The treatment proceeded in the porcine 's back; the right side was designated for tissue sampling 30–, and 60–minutes following exposure, while the left was designated for sampling 3– and 6–hours following exposure. Three conditions were investigated: control, heated metal plate–induced burn, and formic acid burn (figure 1).

The treatments

The thermal burn was induced using a heated metal plate of a 2 cm diameter (100°C) and applied to the skin for 5 minutes. Formic acid burns were induced using a cotton pad soaked in 90% formic acid and were applied to the skin for 20 seconds. After a designated period (30 minutes, 1 hour, 3 hours, and 6 hours following exposure), the skin in the treatment area of a 5 cm diameter was excised as the specimen for further investigations. The treatment was performed under general anesthesia.

The specimens

The specimen was divided into three parts: 1) for histopathology using hematoxylin and eosin (HE) staining; 2) for investigating stress oxidative using 8–isoprostane as a marker; 3) for investigating inflammatory pathway using nuclear factor kappa beta (NFKβ) and high mobility group box1 protein (HMGB1) as a marker.

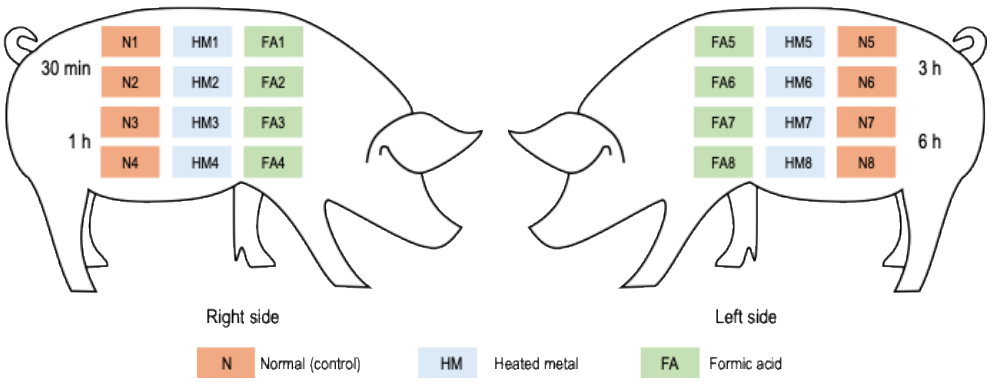

Figure 1 The diagram illustrates the distribution of treatment areas on both sides of the pig's body

Right side: specimens were collected at 30 minutes and 1 hour after exposure. Left side: specimens were collected at 3 hours and 6 hours after exposure. The treatments included normal skin (N, orange), heated metal (HI, light blue), and formic acid (FA, light green). Each treatment was applied to specific regions, and samples were excised at designated time points for further analysis.

Specimen preparation

1. HE stains for histomorphology study

Journal of Information Systems Engineering and Management
2025, 10(48s)
e-ISSN: 2468-4376
https://www.jisem-journal.com/

Research Article

The specimen was fixed in formalin and embedded in paraffin for histological analysis. Sections of 4 µm were cut using a dermatome and mounted on a glass object. The slides were deparaffinized, rehydrated, and stained with hematoxylin for nuclear visualization, followed by eosin for cytoplasmic contrast. The stained sections were dehydrated, cleared with xylene, and mounted with a coverslip. Histomorphology changes were observed under a light microscope.

2. 8–isoprostane for the study of oxidative stress

The specimens were homogenized and processed to extract proteins for ELISA analysis. The concentration of 8–isoprostane, a biomarker of oxidative stress, was measured using the General 8–isoprostane, 8–iso–PGF2A ELISA KIT (BZ–08240300–EB, Bioenzy) following the manufacturer's protocol. Absorbance readings were taken at 450 nm using a microplate reader. The results were expressed in pg./100 mg of tissue and statistically analyzed to compare differences between groups and time points.

3. Immunohistochemistry (IHC) staining method

Paraffin–embedded tissue sections of 4 µm were deparaffinized, rehydrated, and subjected to antigen retrieval. Endogenous peroxidase activity was blocked using 3% hydrogen peroxide. The sections were then incubated with HMGB1 antibody (GTX101277, Genetex) and anti–RELA (NF–kappa B p65) antibody (MBS821459, MyBioSource) at 1:100 dilutions. A secondary antibody conjugated with HRP (horseradish peroxidase) was applied, followed by detection using DAB (3,3′–diaminobenzidine) chromogen. The sections were counterstained with hematoxylin, dehydrated, and mounted for microscopic examination. Immunoreactivity was assessed semi–quantitatively.

These variables were subjected to statistical analysis. The Shapiro–Wilk test, ANOVA and Post–hoc Tukey HSD were used.

RESULTS

The macroscopic evaluation of porcine skin following exposure to heated metal and formic acid reveals distinct patterns of tissue alteration over time. In the normal control group, the skin maintains a consistent color and texture without visible changes across all time points. In contrast, exposure to heated metal results in immediate and pronounced tissue damage, characterized by forming a dark eschar with a surrounding erythematous border at 30 minutes. Over time, this necrotic area becomes more defined, with increased tissue hardening and darkening, indicating progressive coagulative necrosis. Conversely, the formic acid–exposed skin exhibits subtle but progressive changes. Initially, the affected area appears like normal skin, but by 3 to 6 hours, mild yellowish discoloration and textural alterations become evident, suggesting ongoing tissue reaction and damage (figure 2).

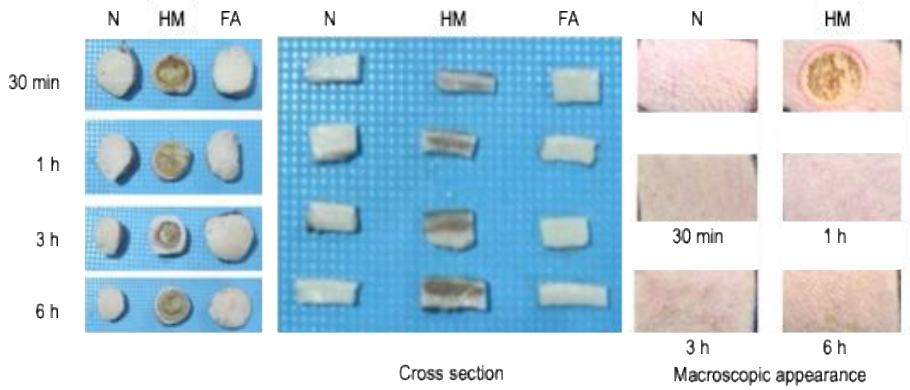

Figure 2. Macroscopic and cross–sectional appearance of porcine skin following exposure to heated metal and formic acid at different time points (30 min, 1 h, 3 h, and 6 h). The normal (control) group shows no visible change. Heated metal–induced burns showed immediate coagulative necrosis with eschar formation, which was darkened and hardened over time. Formic acid exposure initially appears like normal skin then gradually changes to yellowish discoloration and subtle textural changes.

Journal of Information Systems Engineering and Management
2025, 10(48s)
e-ISSN: 2468-4376
https://www.jisem-journal.com/

Research Article

Cross–sectional analysis showed deep, well–demarcated necrosis in the heated metal group, while the formic acid group showed diffuse, progressive tissue damage. Cross–sectional analysis further supports these findings, demonstrating distinct injury patterns between thermal and chemical insults. The heated metal group showed deep tissue penetration with a clear demarcation between necrotic and viable tissue, consistent with coagulative necrosis. Over time, the eschar thickens and becomes more rigid, indicating extensive protein denaturation and cell death. In contrast, formic acid exposure results in more diffuse and less sharply demarcated damage, with gradual softening and loss of structural integrity in affected areas. These findings suggest that while thermal injury induces immediate, localized, and well–defined necrosis, chemical injury progresses more gradually, affecting tissue viability over an extended period (figure 2).

After formic acid exposure, hematoxylin–eosin (HE) staining revealed distinct histopathological alterations in the epidermis and dermis. The stratum basale exhibited signs of cellular degeneration or structural disorganization, likely indicative of direct chemical injury. Depending on the severity of the exposure, this damage led to apoptosis or focal necrosis. Inflammatory responses were evident, as demonstrated by the increased infiltration of mononuclear inflammatory cells in the dermis, suggesting a native immune response to chemical–induced tissue injury (figure 3).

Moreover, the basal cells displayed increased mitotic activity, likely as a rapid adaptive response to cellular stress and damage. This phenomenon does not represent pathological hyperplasia, as seen in malignancies or chronic processes, but rather a transient reactive proliferation triggered by inflammatory and stress signals. The epidermal structural integrity appeared compromised, with potential disruptions in cell adhesion and organization, further supporting the notion of acute cellular response to chemical insult (figure 3).

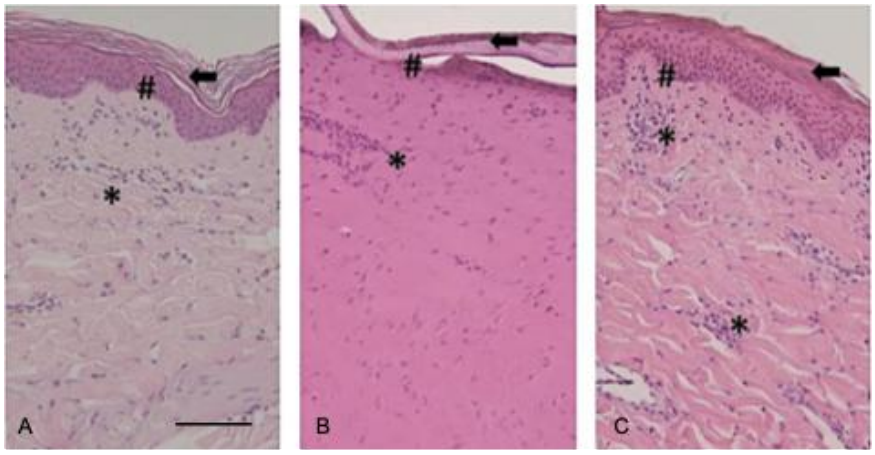

Figure 3. Histopathological features of the skin following normal (A), heated metal (B) and formic acid (C) exposure at 6 hours, stained with hematoxylin–eosin (HE). The stratum corneum (←) and entire epidermis of heated metal (B) exhibited necrosis and detachment, accompanied by diffuse and dense protein denaturation in the dermal collagen. Mononuclear inflammatory cell infiltration (*) was evident. Coagulative degeneration or necrosis is observed in the stratum corneum (←) of formic acid epidermis (C), along with structural disorganization of the stratum basale (#) in the epidermis. Increased infiltration of mononuclear inflammatory cells (*) is evident in the dermis, indicating an active inflammatory response. Scale bar = 100 µm.

The immunohistochemical staining of HMGB1 reveals distinct differences in expression between normal, heated metal exposure, and formic acid–treated skin samples. In the normal sample (A), the epidermis and dermis remain structurally intact and exhibit no detectable HMGB1 expression. Similarly, in the heated metal–treated sample (B), despite extensive necrosis in the epidermis and degeneration in the dermis, HMGB1 staining remains negative, indicating no significant translocation or activation of this damage–associated molecular pattern (DAMP) protein following thermal injury (figure 4).

Conversely, in the formic acid–treated samples (C and D), a notable increase in HMGB–1 expression is observed, particularly in the cytoplasm of epidermal epithelial cells. This suggests active cellular stress responses and potential HMGB1 translocation, possibly contributing to inflammatory signaling following chemical exposure. Cytoplasmic

Journal of Information Systems Engineering and Management
2025, 10(48s)
e-ISSN: 2468-4376
https://www.jisem-journal.com/

Research Article

HMGB1 in formic acid–treated samples but not in thermal injury samples implies differential tissue damage and immune activation mechanism, potentially highlighting the distinct inflammatory pathways triggered by chemical versus thermal insults (figure 4).

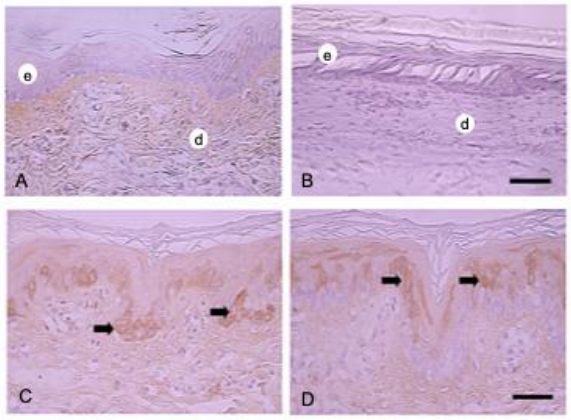

Figure 4**.** Immunohistochemical staining of HMGB1 in skin samples 6 hours after exposure. (A) Normal skin shows intact epidermis (e) and dermis (d) with negative HMGB1 staining. (B) Heated metal exposure results in epidermal necrosis and dermal degeneration, with no HMGB–1 expression. (C, D) Formic acid exposure shows positive HMGB1 expression (→) in the cytoplasm of epidermal epithelial cells, indicating cellular stress response. Staining method: indirect immunohistochemistry. Scale bar = 50 μm.

The immunohistochemical staining for NFKβ expression in skin samples reveals distinct patterns across treatment conditions. In the normal group (A), both the epidermis (e) and dermis (d) exhibit normal histological architecture with no detectable NFKβ expression. Similarly, in the heated metal exposure group (B), severe necrosis and degenerative changes in the epidermis are observed in the dermis. However, despite this extensive tissue damage, NFKβ remains negative, indicating that thermal injury alone may not trigger a significant NFKβ–mediated inflammatory response within the observed timeframe (figure 5).

In contrast, formic acid exposure (C, D) results in weak positive NFKβ expression, particularly in the cytoplasm of epithelial cells in the epidermis (e) (indicated by arrows). This suggests that formic acid exposure elicits a mild activation of NFKβ potentially as part of an inflammatory or stress–induced response. The weak positivity indicates a lower level of transcriptional activation compared to what might be expected in a more severe or prolonged inflammatory setting. This finding suggests that while formic acid exposure initiates an inflammatory cascade, it does so with limited NFKβ activation within the six–hour observation period (figure 5).

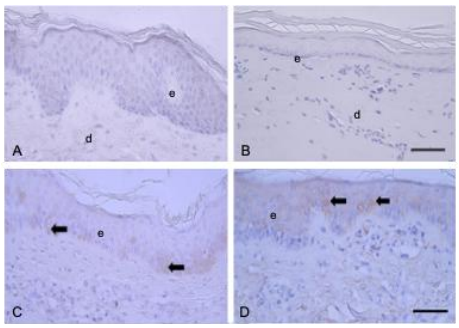

Figure 5. Immunohistochemical staining of NFKβ expression in skin samples after 6 hours exposure under different conditions. (A) Normal group: epidermis (e) and dermis (d) appear normal with no NFKβ expression. (B) Heated metal exposure: necrosis is observed in the epidermis (e) with degeneration in the dermis (d), but no NFKβ expression identified (C, D) Formic acid exposure: weak positive NFKβ expression (←) is observed in the cytoplasm of epithelial cells in the epidermis (e), indicating mild activation. Scale bar = 50 μm.

Journal of Information Systems Engineering and Management
2025, 10(48s)
e-ISSN: 2468-4376
https://www.jisem-journal.com/

Research Article

The ELISA study for 8–isoprostane levels indicate significant differences among treatment groups over time. The normal group exhibits a gradual increase in 8–isoprostane levels, reflecting baseline oxidative stress. In contrast, the formic acid group shows a more pronounced rise, particularly at 3 and 6 hours, suggesting that chemical exposure induces oxidative stress more rapidly than in the normal group. The heated metal group demonstrates the highest levels of 8–isoprostane at all time points, with a marked increase at 6 hours, indicating that thermal injury results in the most severe oxidative damage (figure 6).

Statistical analysis confirms these findings, with the Shapiro–Wilk test ($p = 0.313$) confirming normality and ANOVA yielding a highly significant result ($F = 42.02$, $p = 3.35 \times 10^{-17}$). Post–hoc Tukey HSD analysis identifies significant differences, particularly between the normal control and heated metal groups at later time points. The heated metal 6–hour group exhibits the highest oxidative stress, significantly differing from both the normal and formic acid groups. Additionally, the formic acid 30–minute group differs significantly from the normal 30–minute group, suggesting that formic acid induces oxidative damage early.

These findings highlight the varying degrees of oxidative stress induced by different injury mechanisms, with thermal exposure producing the most pronounced effects over time (figure 6).

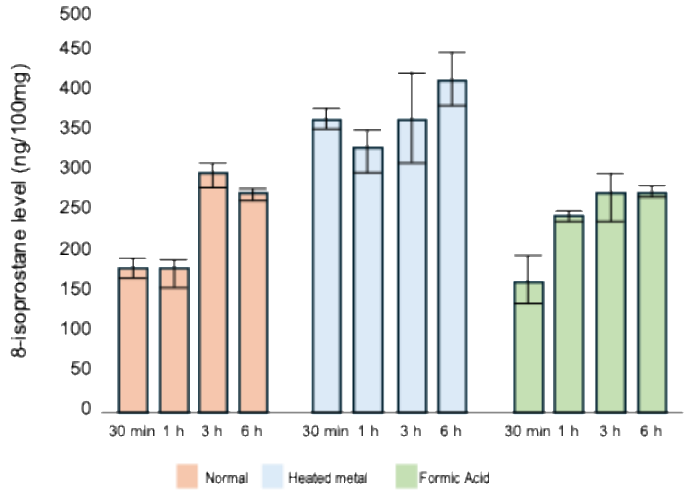

Figure 6. 8–isoprostane levels (ELISA) in different treatment groups (Normal, Heated Metal, and Formic Acid,) at 30 minutes, 1 hour, 3 hours, and 6 hours. The heated metal group shows the highest 8–isoprostane levels, particularly at 6 hours, indicating severe oxidative stress. The formic acid group also shows a significant increase in 8–isoprostane levels compared to the normal group, suggesting notable oxidative damage, although not as severe as the heated metal group. Statistical analysis confirms significant differences among groups ($p < 0.05$).

DISCUSSION

This study highlights the differential effects of thermal and chemical injury on porcine skin, particularly regarding macroscopic appearance, histopathological changes, inflammatory responses, and oxidative stress markers. The findings demonstrate that while both heated metal and formic acid induce tissue damage, the mechanisms and progression of injury differ significantly.

The macroscopic evaluation revealed that exposure to heated metal shows an immediate and severe tissue necrosis, characterized by the formation of eschar with a well–demarcated boundary. This finding aligned with previous study describing thermal burns as causing coagulative necrosis, where protein denaturation occurs rapidly due to direct heat application.[12,13,17] Over time, this necrotic tissue hardens, reinforcing the irreversible nature of thermal injury.

Conversely, formic acid exposure for 20 seconds led to a more insidious pattern of damage. Initially, the affected skin appeared normal, but over time, a yellowish discoloration and progressive tissue breakdown became evident. This delayed response suggests that formic acid induces a sustained chemical reaction rather than immediate protein denaturation.[18] Histopathologically, formic acid exposure resulted in structural disorganization of the epidermis and increased mononuclear inflammatory cell infiltration in the dermis. The presence of inflammation at later time

Research Article

points suggests ongoing tissue injury, possibly due to prolonged penetration and interaction with cellular components. These findings confirm that even a brief 20–second exposure to formic acid is sufficient to cause significant skin damage, reinforcing its potential role as a hazardous chemical agent.[9,19]

Immunohistochemical analysis of HMGB1 expression provided further insight into the inflammatory response triggered by each type of injury. In thermal injury, HMGB1 expression was negligible despite extensive tissue necrosis, aligning with the concept that thermal burns cause rapid and irreversible cell death, minimizing active inflammatory signaling from damaged cells.[20–25] In contrast, formic acid exposure led to significant HMGB1 translocation to the cytoplasm of epidermal cells, indicating stress–induced cellular response. As HMGB1 functions as a damage–associated molecular pattern (DAMP) protein, its upregulation suggests active inflammation and immune cell recruitment.[22,25,26]

Similarly, NFKβ expression differed between treatment groups. While heated metal exposure did not activate NFKβ, formic acid exposure resulted in weak but detectable NFKβ expression in the epidermis. NFKβ is a key transcription factor involved in inflammation and cell survival,[27] suggesting that formic acid may activate a delayed inflammatory cascade, whereas thermal injury primarily causes direct coagulative necrosis without strong NFKβ–mediated responses.[27–30]

The ELISA analysis of 8–isoprostane, a biomarker of oxidative stress, further supported the distinct mechanisms of injury.[31–34] The highest levels of 8–isoprostane were observed in the heated metal group, particularly at 6 hours post–exposure, indicating severe oxidative stress and lipid peroxidation due to thermal damage. In the formic acid group, oxidative stress markers also increased, but to a lesser extent and with a more gradual rise. This suggests that while formic acid exposure induces oxidative damage, it does so progressively rather than instantaneously. Importantly, these findings indicate that a 20–second exposure to formic acid is sufficient to trigger oxidative stress responses, further supporting its potential for inducing significant chemical burns.[30]

Statistical analysis confirmed significant differences in 8–isoprostane levels between treatment groups, reinforcing the idea that thermal injury induces the most severe oxidative stress. In contrast, chemical injury follows a more prolonged course. These findings align with previous studies suggesting that oxidative stress is crucial in tissue damage progression following chemical burns.

From a clinical and forensic perspective of view, the differential effects of heated metal and formic acid exposure have important implications. The distinct injury patterns observed can aid in forensic investigations of burn–related crimes, particularly in distinguishing between thermal and chemical burns. The delayed tissue damage seen in formic acid exposure may explain why some victims initially underestimate the severity of the injury, leading to late medical intervention. Additionally, the intense inflammatory response associated with formic acid exposure suggests that therapeutic strategies targeting inflammation and oxidative stress may be beneficial in managing chemical burns.

This study's limitations include using a porcine model, which, although anatomically and physiologically like human skin, may not fully replicate the complex biological responses observed in human burn injuries. Additionally, the sample size was limited to two pigs, which may affect the generalizability of the findings. The study also focused on a relatively short observation period (up to 6 hours), which may not capture the long–term inflammatory and oxidative stress responses following exposure. Furthermore, while ELISA and immunohistochemistry provided valuable insights into protein expression, additional molecular analyses, such as gene expression profiling, could have further elucidated the underlying mechanisms of injury. Future studies with larger sample sizes, extended observation periods, and more comprehensive molecular assessments are needed to validate and expand upon these findings.

CONCLUSION

This study demonstrates that heated metal and formic acid exposure induce significant oxidative stress and inflammatory responses in porcine skin through distinct mechanisms. While heated metal caused immediate coagulative necrosis with severe oxidative damage, formic acid exposure for just 20 seconds triggered a delayed but progressive injury characterized by increased HMGB1 and NFKβ expression and oxidative stress. These findings highlight the potential of formic acid to cause chemical burns with ongoing tissue damage, emphasizing the need for early medical intervention. From a forensic perspective, the distinct injury patterns observed can aid in

Journal of Information Systems Engineering and Management
2025, 10(48s)
e-ISSN: 2468-4376
https://www.jisem-journal.com/

Research Article

differentiating between thermal and chemical burns. Further research with larger sample sizes and extended observation periods is needed to understand the long–term effects better and optimize treatment strategies for such injuries.

Ethical Approval Statement

This study was approved by the Animal Ethics Committee of the School of Veterinary Medicine and Biomedical Sciences, IPB University, under ethical approval number 257 KEH/SKE/IX/2024. All experimental procedures involving animals were conducted by institutional guidelines and adhered to ethical principles for animal research. The study utilized porcine models to investigate the effects of formic acid exposure on skin tissue, including histopathological analysis and biomarker evaluation. The research was carried out at the Teaching Animal Hospital, IPB Bogor, ensuring compliance with ethical standards for animal welfare.

Acknowledgement

The authors would like to express their gratitude to Universitas YARSI for funding this research. We also extend our appreciation to Teaching Animal Hospital, IPB Bogor and Integrated Laboratory of Universitas YARSI for providing the facilities and technical support necessary for conducting this study. Special thanks to the research staff and laboratory personnel for their valuable assistance throughout the experimental process.

Conflict of interest

The authors declare no conflict of interest related to this research. This study was conducted independently, without any influence from funding sources, and there were no financial, personal, or professional relationships that could have biased the results or interpretations of the study.

The role of author

No	Contributor role	Definition	Author 1 2 3,...*
1	Conceptualization	Ideas, formulation or evolution of overarching research goals and aims.	**Author 1,2, 3, 4**
2	Data curation	Management activities to annotate (produce metadata), scrub data, and maintain research data (including software code, where it is necessary for interpreting the data) for initial use and later reuse.	**Author 1, 7**
3	Formal analysis	Application of statistical, mathematical, computational, or other formal techniques to analyze or synthesize study data.	**Author 1, 7**
4	Funding acquisition	Acquisition of financial support for the project leading to this publication	**Author 1**
5	Investigation	Conducting a research and investigation process, specifically performing the experiments, or data/evidence collection.	**Author 1, 5, 6**
6	Methodology	Development or design methodology; creation of models.	**Author 1, 2, 3, 4, 5, 6**

Journal of Information Systems Engineering and Management
2025, 10(48s)
e-ISSN: 2468-4376
https://www.jisem-journal.com/

Research Article

7	Project administration	Management and coordination responsibility for the research activity planning, and execution.	**Author 1, 5**
8	Resources	Provision of study materials, reagents, materials, patients, laboratory samples, animals, instrumentation, computing resources, or other analysis tools.	**Author 1, 5, 6**
9	Software	Programming, software development, designing computer programs, implementation of the computer code and supporting algorithms; testing of existing code components	**Author 7**
10	Supervision	Oversight and leadership responsibility for the research activity planning and execution, including mentorship external to the core team	**Author 2, 3, 4, 5**
11	Validation	Verification, whether as a part of activity or separate, of the overall replication/reproducibility of results/experiments and other research outputs	**Author 1, 2, 6, 7**
12	Visualization	Preparation, creation and/or presentation of the published work, specifically visualization/data presentation	**Author 1, 7**
13	Writing – original draft preparation	Creation and/or presentation of the published work, specifically writing the initial draft (including substantive translation).	**Author 1, 7**
14	Writing – review and editing	Preparation, creation and/or presentation of the published work by those from the original research group, specifically critical review, commentary, or revision – including pre– or post-publication stages.	**Author 1, 2, 3, 7**

REFERENCE

1. Sigurdsson J, Bjijrnsson A, Th Gudmundsson S. Formic acid burn-local and systemic effects report of a case. Burns 1983;9:358–61.
2. Chan TC, Williams SR, Clark RF. Formic Acid Skin Burns Resulting in Systemic Toxicity. Ann Emerg Med 1995;3:383–6.
3. Naik SM, Goutham M, Bhat RS, Bhat C, Appaji M, Chidananda K, et al. Acute Formic Acid Poisoning: A Case Series Analysis with Current Management Protocols and Review of Literature. Int J Head Neck Surg 2014;5(3):104–7.

4. Palao R, Monge I, Ruiz M, Barret JP. Chemical burns: Pathophysiology and treatment. Burns2010;36(3):295–304.
5. Bruze M, Gruvberger B, Fregert S. Chemical skin burns. In: Chew A, Maibach H, editors. Irritant Dermatitis. Berlin: Springer; page 53–61.
6. Bakovic M, Nestic M, Mayer D. Suicidal chemistry: combined intoxication with carbon monoxide and formic acid. Int J Legal Med 2016;130(3):723–9.
7. Perera V, Karunadasa K, Perera C. Self-inflicted formic acid burn: Rare form of acid burn (two case reports). Eur J Plast Surg 2012;35(2):199–200.
8. UK Health Security Agency. Formic acid Incident management [Internet]. 2024 [cited 2025 Jan 28]. Available from: chemcompendium@ukhsa.gov.uk
9. Burke R. Formic acid [Internet]. In: Burke R, editor. Applied Chemistry and Physics. CRC Press; 2020. Available from: https://taylorandfrancis.com/knowledge/Engineering_and_technology/Chemical_engineering/Formic_acid/
10. Wako FM, Pio G, Salzano E. Modeling Formic Acid Combustion. Energy Fuel 2022;36:14382–92.
11. Editors. Chemical Burns [Internet]. Burn and Reconstructive Centers of America Available from: https://burncenters.com/burns/burn-services/chemical-burns/
12. Seaton M, Hocking A, Gibran NS. Porcine models of cutaneous wound healing. ILAR J 2015;56(1):127–38.
13. Deng X, Chen Q, Qiang L, Chi M, Xie N, Wu Y, et al. Development of a porcine full-thickness burn hypertrophic scar model and investigation of the effects of Shikonin on hypertrophic scar remediation. Front Pharmacol 2018;9(JUN):1–12.
14. Obernier JA, Baldwin RL. Establishing an Appropriate Period of Acclimatization Following Transportation of Laboratory Animals. ILAR J [Internet] 2006;47(4):364–9. Available from: https://academic.oup.com/ilarjournal/article/47/4/364/810898
15. Bharati J, Kumar S, Devi SJ, Mohan NH, Gupta VK. Transcriptional dynamics of porcine granulosa cells during cellular acclimation to thermal challenge. J Thermal Biol [Internet] 2025;127:104064. Available from: https://www.sciencedirect.com/science/article/pii/S030645652500021X
16. Zhang Y, Sun L, Zhu R, Zhang S, Liu S, Wang Y, et al. Porcine gut microbiota in mediating host metabolic adaptation to cold stress. NPJ Biofilm Microbiom 2022;8(18):1–14.
17. Allgöwer M, Schoenenberger GA, Sparkes BG. Pernicious effectors in burns. Burns 2008;34(1 SUPPL.).
18. Kaushik S, Bird S. Topical chemical burns: Initial evaluation and management [Internet]. UpToDate2025;Available from: www.uptodate.com
19. ACDC - NIOSH. CDC - NIOSH Pocket Guide to Chemical Hazards: Formic acid [Internet]. 2019. Available from: https://beta.cdc.gov/niosh/npg/npgd0296.html#collapse_819535bae1ca30b564
20. Coleman LG, Maile R, Jones SW, Cairns BA, Crews FT. HMGB1/IL-1β complexes in plasma microvesicles modulate immune responses to burn injury. PLoS One 2018;13(3):1–23.
21. Huebener P, Pradere JP, Hernandez C, Gwak GY, Caviglia JM, Mu X, et al. The HMGB1/RAGE axis triggers neutrophil-mediated injury amplification following necrosis. J Clin Invest 2015;125(2):539–50.
22. Yang H, Antoine DJ, Andersson U, Tracey KJ. The many faces of HMGB1: molecular structure-functional activity in inflammation, apoptosis, and chemotaxis. J Leukoc Biol 2013;93(6):865–73.
23. Lantos J, Földi V, Röth E, Wéber G, Bogár L, Csontos C. Burn trauma induces early HMGB1 release in patients: Its correlation with cytokines. Shock 2010;33(6):562–7.
24. Wang H, Yang H, Czura C, Sama A, Tracey K. HMGB1 as a Late Mediator of Lethal Systemic Inflammation. Am J Respir Crit Care Med [Internet] 2001;164:1768–73. Available from: www.atsjournals.org
25. Chen R, Kang R, Tang D. The mechanism of HMGB1 secretion and release. Exp Mol Med2022;54(2):91–102.
26. Magna M, Pisetsky DS. The role of HMGB1 in the pathogenesis of inflammatory and autoimmune diseases. Molecular Medicine 2014;20(1):138–46.
27. Liu T, Zhang L, Joo D, Sun SC. NF-κB signaling in inflammation. Sign Transduct TargetTher 2017;2(e17023):1–9.
28. Mahung C, Wallet SM, Jacobs JE, Zhou LY, Zhou H, Cairns BA, et al. Multiplexed Human Gene Expression Analysis Reveals a Central Role of the TLR/mTOR/PPARγ and NFkB Axes in Burn and Inhalation Injury-

Research Article

Induced Changes in Systemic Immunometabolism and Long-Term Patient Outcomes. Int J Mol Sci 2022;23(9418):1–16.
29. George B, Suchithra T V., Bhatia N. Burn injury induces elevated inflammatory traffic: the role of NF-κB. Inflam Res 2020;70(1):51–65.
30. Jeschke MG, van Baar ME, Choudhry MA, Chung KK, Gibran NS, Logsetty S. Burn injury. Nature Rev Dis Prime 2020;6(11):1–25.
31. Bono R, Romanazzi V. Isoprostanes as Biomarkers of Disease and Early Biological Effect [Internet]. In: Preedy VR, Patel VB, editors. General Methods in Biomarker Research and their Applications. Springer Reference; 2015. page 383–404.Available from: http://www.springer.com/series/13842
32. Czerska M, Zieliński M, Gromadzińska J. Isoprostanes – A novel major group of oxidative stress markers. Int J Occup Med Environ Health2016;29(2):179–90.
33. Miller E, Morel A, Saso L, Saluk J. Isoprostanes and neuroprostanes as biomarkers of oxidative stress in neurodegenerative diseases. Oxid Med Cell Longev2014;2014.
34. van 't Erve TJ, Kadiiska MB, London SJ, Mason RP. Classifying oxidative stress by F2-isoprostane levels across human diseases: A meta-analysis. Redox Biol 2017;12:582–99.