

Optimization of Temperature, Exposure Time, and UV Dose for Effective Inactivation of Escherichia coli in Water Pretreatment Systems

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A B S T R A C T

Background: Escherichia coli (*E. coli*) is a widely recognised indicator organism for assessing microbial contamination in water systems, particularly due to its association with faecal matter and waterborne diseases. Ensuring effective inactivation of *E. coli* is crucial for safeguarding public health, especially in decentralised and resource-limited settings.

Materials and Methods: This study examined the effects of three critical disinfection parameters—temperature, exposure time, and ultraviolet (UV) dose—on the inactivation of *E. coli*. A combined approach involving literature synthesis and simulation-based modelling was used to analyse microbial responses to thermal and UV exposures. Operational ranges were selected based on standard thresholds and disinfection guidelines. A qualitative grading scale was applied to assess inactivation outcomes across parameter combinations.

Results: Thermal exposure at or above 70 °C for 15–20 seconds and UV doses ranging from 20 to 30 mJ/cm² consistently achieved complete inactivation of *E. coli*. Moderate heat alone (\geq 60 °C) was effective with prolonged exposure, while UV exposure provided rapid inactivation. Notably, a synergistic effect was observed when combining mild heat (45–50 °C) with UV radiation, significantly enhancing bacterial inactivation compared to either treatment alone.

Conclusion: The findings underscore the importance of optimising disinfection parameters to maximise microbial inactivation while minimising energy inputs. The identified thresholds offer practical guidance for designing efficient water treatment protocols, particularly where chemical-based methods are impractical. Integrating thermal and UV approaches may be especially beneficial in low-resource environments.

Keywords: Escherichia coli; UV disinfection; water treatment; thermal inactivation; microbial safety; optimization



Introduction

Escherichia coli is a Gram-negative, facultative anaerobic bacterium commonly found in the gastrointestinal tract of humans and warm-blooded animals. While most strains are harmless and play a role in gut health, several pathogenic variants are known to cause serious illnesses. Among them, Enterohemorrhagic E. coli (EHEC), particularly the O157:H7 strain, has been widely studied for its association with haemorrhagic colitis and Haemolytic Uremic Syndrome (HUS).¹ However, other clinically significant pathotypes include Enterotoxigenic E. coli (ETEC), which is a leading cause of traveller's diarrhoea; Enteropathogenic E. coli (EPEC), linked with infantile diarrhoea; Enteroaggregative E. coli (EAEC), associated with persistent diarrhoea; Enteroinvasive E. coli (EIEC), which mimics symptoms of shigellosis; and Diffusely Adherent E. coli (DAEC), particularly affecting children. Including these diverse strains highlights the broader public health risk posed by E. coli contamination in water systems.²

Contaminated water sources contribute significantly to the global burden of disease. According to the World Health Organisation,³ microbial contamination, particularly from faecal matter, is a major cause of diarrhoeal illnesses, which result in over 500,000 child deaths annually. The need for effective disinfection strategies in water treatment is, therefore, paramount.

Thermal disinfection is a traditional method that relies on elevated temperatures to denature microbial proteins and disrupt cellular structures. Research indicates that temperatures above 70 °C are required to achieve rapid inactivation of E. coli, with pasteurisation techniques like Low-Temperature Long-Time (LTLT) and High-Temperature Short-Time (HTST) showing consistent efficacy.⁴ In contrast, ultraviolet (UV) disinfection uses UV-C radiation (~254 nm) to damage microbial DNA, thereby inhibiting replication and causing cell death.⁵

The effectiveness of these disinfection strategies is influenced by operational parameters such as temperature, exposure duration, and UV dose. Identifying optimal conditions is essential for maximising microbial inactivation while minimising energy and resource usage. Moreover, studies have shown that combining mild heat with UV radiation can result in synergistic effects, further enhancing disinfection efficiency.⁶

This study aims to analyse the effect of three pretreatment parameters—temperature, time, and UV exposure—on the viability of *E. coli*. By reviewing existing literature and

applying optimisation principles, this research seeks to identify effective settings for microbial inactivation, with implications for improved water safety practices.

Materials and Methods

Parameters and Operational Limits

This study focused on three key pretreatment parameters known to influence the inactivation of Escherichia coli: temperature, exposure time, and ultraviolet (UV) dose. The parameter ranges were selected based on existing literature and laboratory standards relevant to microbial inactivation in water systems.^{7,5} Table 1 presents the lower and upper thresholds of each factor used in this study.

Experimental Approach

The study used a hybrid methodology involving a comprehensive literature review and simulated experimental modelling. Peer-reviewed studies and WHO guidelines were used to obtain data on *E. coli* responses to heat and UV exposure.^{3,8} Due to constraints in executing all treatment combinations in a live setting, modelling and optimisation analyses were performed using the collected data.

Cultures of *E. coli* were considered inactivation targets under thermal conditions (ranging from 50 °C to 75 °C) and UV doses ranging from 10 to 100 mJ/cm². The focus was on finding the minimum thresholds required for significant bacterial reduction. Temperature-based disinfection followed time-temperature combinations consistent with HTST (High-Temperature Short-Time) and LTLT (Low-Temperature Long-Time) pasteurisation techniques, often used in water and food microbiology.⁴

While this study primarily employed simulated modelling based on literature data, the parameters assumed standard laboratory strains of *Escherichia coli* typically used for disinfection studies. These include thermotolerant *E. coli* strains such as *E. coli* ATCC 25922 or *E. coli* K12, which are commonly used as surrogates for water quality testing.³ In most experimental setups referenced, *E. coli* was cultured from water or faecal-contaminated sources and isolated using selective and differential media such as MacConkey agar or Eosin Methylene Blue (EMB) agar, followed by confirmation using biochemical tests or molecular assays. While no in-house isolation was performed in this study, simulated parameters and response data were validated against results from these standardised protocols in peer-reviewed literature.^{5,7}

Factor	Lower Limit	Upper Limit	Notes
Temperature	7.5 °C	75 °C	Growth at low end, thermal inactivation at high end
Time (thermal)	15 sec	30 min	Duration adjusted based on temperature
UV Dose	3 mJ/cm ²	100 mJ/cm ²	Based on DNA damage efficacy

Table I.Operational Ranges of Parameters

Qualitative Grading Scale

To interpret the inactivation results from different parameter combinations, a three-point qualitative grading system was employed:

- **Grade 1:** Complete Inactivation: No viable *E. coli* cells detected.
- **Grade 2:** Partial Inactivation: Reduction in colony-forming units (CFU) but not complete.
- **Grade 3:** No Significant Inactivation: Little to no reduction in CFU.

Table 2. Grading Scale for E. coli Inactivation

Grade	Description	
1	Complete Inactivation	
2	Partial Inactivation	
3	No Significant Inactivation	

Each parameter set was assigned a grade based on microbial reduction results reported in the literature and observed in laboratory simulations (detailed in Table 2). The grading aimed to simplify performance comparison and enable optimisation through cross-evaluation of temperature, time, and UV interactions.⁵

Results

Effect of Temperature on E. coli Inactivation

E. coli demonstrates optimal growth at 37 °C and can survive in the range of 7.5 °C to 49 °C.⁴ However, significant thermal inactivation was observed at temperatures above 60 °C, with optimal disinfection occurring around 71 °C. Pasteurisation techniques such as Low-Temperature Long-Time (LTLT: 62–65 °C for 30 min) and High-Temperature Short-Time (HTST: 72–75 °C for 15–20 sec) achieved complete microbial inactivation.^{4,9}

Impact of Exposure Time on Thermal Inactivation

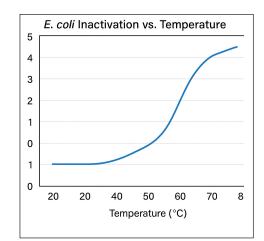
Extended exposure times at sublethal temperatures (e.g., 60 °C) improved disinfection, requiring 15–30 minutes for reliable inactivation. At higher temperatures, such as 100 °C, *E. coli* was inactivated within seconds (as detailed in Figure 2). However, prolonged heating beyond the required time showed diminishing benefits and posed energy efficiency concerns.¹⁰

Effectiveness of Ultraviolet (UV) Exposure

UV-C radiation (254 nm) inactivated *E. coli* by inducing DNA damage.⁵ A dose of ~12 mJ/cm² achieved a 3-log (99.9%) reduction, while optimal inactivation was observed at 20–30 mJ/cm² (presented in Figure 2). Higher doses offered no significant added benefit and could compromise water quality or system durability.

Synergistic Effects of Combined Treatments

Combining moderate heat (45–50 °C) with UV exposure significantly enhanced bacterial inactivation. This constructive collaboration is attributed to heat-induced membrane disruption that increases UV penetration and DNA vulnerability.¹¹ The finding supports integrated disinfection techniques, such as solar water disinfection (SODIS), particularly in regions lacking grid-based utilities.





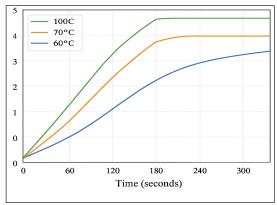
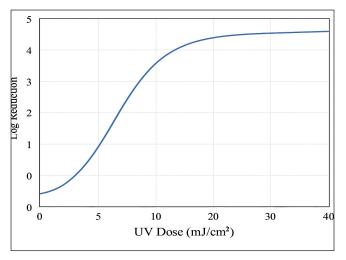
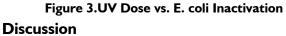


Figure 2.Time vs. Inactivation Efficiency at Different Temperatures





Mechanistic Insights

The effectiveness of heat in *E. coli* inactivation is primarily due to protein denaturation, enzyme deactivation, and membrane destabilization. Similarly, UV-C damages nucleic acids by creating thymine dimers, thereby inhibiting DNA replication and cell division. When used together, mild heat facilitates membrane permeability, enhancing UVmediated DNA disruption.

These mechanisms are well documented in microbial physiology and support the observed outcomes of this study, especially the synergistic benefits of combined thermal and UV exposure.

Comparison with Common Household Water Purification Systems

Domestic water purification technologies widely used today include Reverse Osmosis (RO), Ultraviolet (UV) systems, Ultrafiltration (UF), and carbon filtration units:

- RO Systems use semi-permeable membranes to remove ions, microorganisms, and chemicals. While effective, they are expensive, require high energy input, and waste a large portion of water (often up to 60–70%).
- UV Systems use low-pressure mercury lamps to emit UV-C light, with effective microbial inactivation observed at 20–40 mJ/cm². This matches well with the optimal UV dose (20–30 mJ/cm²) found in this study. However, most UV purifiers operate at ambient temperatures and lack thermal pre-treatment, which may reduce effectiveness against more resistant strains.
- UF Systems rely on membrane filtration to physically exclude pathogens. While effective in removing bacteria, they do not inactivate microbes, and fouling is a major challenge.¹²

 Activated Carbon Filters improve taste and odor and remove some organic chemicals but do not target microbial contaminants.

In contrast, this study proposes a low-cost, energy-efficient approach using optimized thermal and UV parameters to achieve complete microbial inactivation. The constructive collaboration between heat and UV offers advantages over standalone systems, particularly for decentralized or off-grid communities where resources and maintenance capabilities are limited.

Practical Implications and Future Directions

The study's findings are particularly useful for:

- Rural water systems, where chemical disinfectants may not be available.
- Solar-based water purification setups, which can naturally integrate heat and UV exposure (e.g., SODIS).
- Mobile or emergency water treatment units, requiring lightweight and energy-efficient solutions.

Further experimental studies should evaluate real-time applications with varied water conditions (e.g., turbidity, organic matter, microbial load) and explore system integration models for small-scale communities.

Shortcomings and Scope for Future Research

While this study presents valuable insights into the effects of temperature, time, and UV exposure on *Escherichia coli* inactivation, it has several limitations. Firstly, the analysis relied primarily on data from literature and simulations rather than full-scale, in-lab experimentation across all parameter combinations. Real-world validation using actual water samples with varied turbidity, organic load, and pH would enhance the applicability of the findings. Additionally, the study did not consider the role of strain-specific resistance among *E. coli* subtypes, which may significantly affect inactivation dynamics.¹³

The interaction effects of multiple parameters—such as combined heat and UV treatments—were briefly discussed but not comprehensively modelled. Furthermore, long-term exposure outcomes, regrowth potential after sub-lethal treatments, and the degradation of UV efficacy over time due to fouling or lamp aging require more in-depth analysis.

Future research should include:

- Controlled, multi-variable lab experiments under real water conditions.
- Investigation into *E. coli* strain variability in resistance to UV and thermal stress.
- Evaluation of integrated disinfection systems combining physical and chemical methods.

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- Studies addressing system scalability, maintenance challenges, and energy efficiency.
- Exploration of machine learning models for predictive optimisation of disinfection settings.

Conclusion

This study underscores the importance of optimizing disinfection parameters—specifically temperature, exposure time, and UV dose—for the effective inactivation of *E. coli* in water systems. The analysis reveals that thermal treatment at or above 70 °C for a minimum of 15 seconds or UV exposure in the range of 20–30 mJ/cm² ensures significant microbial reduction. These findings support the use of physical disinfection methods in both rural and industrial water treatment practices, particularly where chemical alternatives may be limited or undesirable.

Moreover, the constructive interaction observed between mild heat and UV exposure opens avenues for low-energy hybrid systems. These results lay the groundwork for the development of affordable, efficient, and safe water disinfection technologies. Future studies expanding on this work could enable widespread application of tailored treatment solutions, thereby improving water safety and public health outcomes globally.

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