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Microwave Tissue Processing: A Rapid, Reliable, and Cost-Effective Alternative to Conventional Methods

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Abstract:

Background: Timely and accurate histopathological diagnosis depends on effective tissue processing. While conventional tissue processing (CTP) is widely used, its time-intensive nature can delay diagnosis. Microwave tissue processing (MTP) has emerged as a faster alternative with potential advantages in efficiency and chemical usage.

Objective: To compare MTP and CTP in terms of processing time, histological quality, reagent consumption, and sectioning performance, and to evaluate MTP's suitability for cancer-related diagnostics.

Methods: Sixty tissue samples (kidney, liver, and breast) were processed by MTP or CTP. Four blinded pathologists assessed histological quality using a 5-point scale. Processing time, reagent

use, and sectioning characteristics were recorded. Data were analyzed using independent t-tests or Mann–Whitney U tests ($p < 0.05$).

Results: MTP significantly reduced processing time (1–2 hours vs. 12–16 hours; $p < 0.001$) and reagent consumption by ~45–50%. Histological quality was comparable or superior with MTP, particularly in breast tissue ($p < 0.03$). However, CTP provided better sectioning integrity and ribbon formation ($p < 0.02$).

Conclusion: MTP offers a rapid, cost-effective, and diagnostically reliable alternative to conventional processing, though improvements in sectioning quality are needed for routine adoption.

Keywords: Histology, Microwaves, Tissue Processing, Pathology, Surgical, Comparative Study, Biomedical Technology, Diagnosis

Introduction

Histopathology is the cornerstone of disease diagnosis, providing detailed microscopic insights into cellular and tissue architecture. The accuracy of a histopathological diagnosis depends heavily on the quality of tissue processing, which prepares specimens for microscopic examination. The standard, conventional tissue processing (CTP) method involves a lengthy sequence of steps: fixation, dehydration, clearing, and paraffin infiltration [1, 2]. While highly effective, this multi-step process is time-consuming, often requiring 12–16 hours, which can delay patient diagnosis and treatment [3].

To overcome these limitations, microwave tissue processing (MTP) has been developed as an accelerated method. MTP utilizes microwave energy to generate heat through the rapid rotation of polar molecules, thereby expediting the diffusion of reagents into the tissue [4, 5]. This technique promises to reduce processing time, improve laboratory efficiency, and lower the consumption of hazardous chemicals, making it particularly appealing for urgent diagnostic needs, such as intraoperative consultations, and in settings with limited resources [6, 7].

Previous studies have shown that MTP can significantly shorten turnaround times while maintaining histological quality comparable to CTP [8, 9]. However, the widespread adoption of

MTP has been hindered by concerns regarding its consistency, reliability across different tissue types and thicknesses, and the potential impact on downstream diagnostic applications like immunohistochemistry [10, 11].

To address the limitations of conventional methods and evaluate microwave processing as a viable alternative, this study provides a comprehensive comparative analysis of both techniques. This study aims to systematically compare the efficacy of microwave tissue processing (MTP) with conventional processing techniques (CTP), with a specific focus on processing time, tissue morphology, staining quality, and overall laboratory workflow.

Although microwave-assisted tissue processing has been evaluated in various international settings, there is a lack of studies in low-resource environments, such as Sudan and East Africa, where rapid and cost-effective methods could significantly impact diagnostic turnaround times. Additionally, previous studies have not extensively assessed sectioning quality and practical feasibility across multiple tissue types.

To the best of our knowledge, this is the first comparative study of microwave and conventional tissue processing conducted in Sudan. This research aims to determine the diagnostic and practical feasibility of microwave tissue processing, thereby informing its potential role in modern histopathology laboratories.

Materials and Methods

Study Design and Setting

This was a comparative, experimental study conducted over a 10-week period in a histopathology laboratory equipped with both a microwave tissue processor and a conventional automated processor. The study aimed to systematically compare the efficacy and practicality of microwave-assisted tissue processing (MTP) against conventional tissue processing (CTP).

Sample Collection and Grouping

A total of 60 tissue samples were collected from the surgical department, comprising three distinct tissue types: kidney, liver, and breast. Following collection, all specimens were fixed in 10% neutral buffered formalin for 24 hours at room temperature to ensure proper tissue preservation. The samples were then randomly allocated into two equal groups to minimize selection bias:

- **Microwave Processing Group (n=30):** 10 kidney, 10 liver, and 10 breast samples.
- **Conventional Processing Group (n=30):** 10 kidney, 10 liver, and 10 breast samples.

Tissue Processing Protocols

Microwave Tissue Processing (MTP) Protocol: This was performed using a microwave tissue processor (e.g., Leica ASP300S). The protocol was designed for rapid processing and involved the following steps:

1. Dehydration: Samples were sequentially immersed in a graded series of ethanol solutions (70%, 80%, 90%, and 100%). Each step lasted for 2–5 minutes, with a total dehydration time of 10–20 minutes.
2. Clearing: A clearing agent (xylene) was used for 5–10 minutes to remove residual ethanol.
3. Infiltration: Tissues were infiltrated with molten paraffin wax at approximately 60°C for 10–15 minutes. The total processing time for this group ranged from 1 to 2 hours.

Conventional Tissue Processing (CTP) Protocol:

This was performed using a standard automated processor (Leica TP1020), adhering to the following established timeline:

1. Dehydration: Tissues underwent sequential immersion in graded ethanol solutions: 70% ethanol for 1 hour, 80% for 1 hour, 90% for 1 hour, and two baths of 100% ethanol for 1 hour each. Total dehydration time was approximately 5–7 hours.
2. Clearing: Samples were immersed in two changes of xylene for approximately 1 hour each.
3. Paraffin Infiltration: Tissues were infiltrated with molten paraffin wax in two baths, with each bath lasting 1–2 hours. The total processing time for this group ranged from 12 to 16 hours.

Evaluation of Outcomes

After processing, all samples were embedded in paraffin molds and sectioned at 4–5 µm using a rotary microtome. The sections were then stained with hematoxylin and eosin (H&E). Four experienced pathologists, who were blinded to the processing method, independently assessed the quality of each stained section. The following parameters were evaluated using a standardized 5-point scoring scale (1 = poor, 5 = excellent):

- **Histological Quality:**

- Clarity of cytoplasmic and nuclear staining
- Uniformity of staining
- Clarity of cellular detail

- **Morphological Preservation:**
  - Preservation of overall tissue architecture
  - Presence or absence of artifacts (e.g., tissue shrinkage, nuclear bubbling)
- **Sectioning Quality:**
  - Ease of cutting and handling
  - Tissue section integrity (e.g., absence of microtome chatter, tears)
  - Quality of ribbon formation

Additionally, quantitative data on processing time and reagent consumption (ethanol, xylene, and paraffin wax volumes) were recorded and compared between the two groups.

Statistical Analysis

Statistical analysis was conducted using SPSS software. The mean histological scores and quantitative data between the MTP and CTP groups were compared using independent samples t-tests or Mann–Whitney U tests, depending on data distribution. A p-value of < 0.05 was considered statistically significant.

Results

Processing Time

Microwave Tissue Processing (MTP) showed a remarkable and statistically significant reduction in processing time across all examined tissue types when compared to Conventional Tissue Processing (CTP). On average, MTP completed the processing in approximately 1.5 hours (ranging from 1 to 2 hours), while CTP required around 14 hours (spanning 12 to 16 hours) ($p < 0.001$ for liver, kidney, and breast tissues; independent t-test) (Table 1). This dramatic 85–90% decrease highlights MTP's potential to greatly accelerate diagnostic workflows, making it especially valuable in clinical situations where rapid turnaround is critical.

Histological Staining Quality

Using a standardized 5-point scoring system, histological evaluation revealed that staining quality in tissues processed with MTP was comparable—and in some cases superior—to those processed

by CTP. For liver and kidney tissues, there were no statistically significant differences in cytoplasmic or nuclear staining quality or in the clarity of cellular details ($p > 0.05$; Mann-Whitney U test). However, breast tissue samples processed via MTP received significantly higher scores for both cytoplasmic staining (4.9 ± 0.2 vs. 4.7 ± 0.3, $p = 0.03$) and nuclear staining quality (4.9 ± 0.2 vs. 4.6 ± 0.4, $p = 0.02$), suggesting enhanced tissue preservation and staining consistency in fatty tissues (Table 2).

Morphological Preservation and Cellular Detail

When assessing tissue morphology and cellular detail, MTP consistently outperformed CTP across all tissue types. Liver samples exhibited mean morphology scores of 4.8 ± 0.3 compared to 4.3 ± 0.4 with CTP ($p = 0.01$), and cellular detail scores of 4.7 ± 0.4 versus 4.2 ± 0.4 ($p = 0.02$). Similar improvements were observed in kidney tissue morphology (4.9 ± 0.2 vs. 4.4 ± 0.4, $p = 0.01$) and detail (4.8 ± 0.3 vs. 4.3 ± 0.3, $p = 0.01$), as well as in breast tissues (morphology: 5.0 ± 0.1 vs. 4.5 ± 0.4, $p = 0.005$; cellular detail: 4.9 ± 0.2 vs. 4.4 ± 0.3, $p = 0.005$) (Table 3). These findings indicate that MTP better preserves tissue architecture and cellular features, likely due to reduced tissue shrinkage and more uniform fixation.

Reagent Consumption

MTP demonstrated a clear advantage in reagent efficiency, substantially reducing the volume of chemicals needed for processing. Ethanol usage per sample was nearly halved in the MTP group (100 ± 10 mL) compared to CTP (175 ± 15 mL) ($p < 0.001$). Similarly, the clearing agent (xylene) consumption dropped from 40 ± 5 mL in CTP to just 12 ± 2 mL with MTP ($p < 0.001$). Paraffin wax usage was also lower with MTP (18 ± 3 mL) relative to CTP (25 ± 4 mL, $p = 0.01$). Overall, total reagent consumption decreased by approximately 45–50% when using microwave processing (Table 4), which not only cuts costs but also reduces chemical exposure and environmental impact.

Sectioning Quality

Despite the clear benefits of MTP in processing time, reagent use, and histological quality, Conventional Tissue Processing maintained superiority in sectioning quality parameters. Tissue section integrity, ease of sectioning, and ribbon formation consistently scored higher in the CTP group, with statistically significant differences (p-values between 0.001 and 0.02; Mann-Whitney

U test). For instance, liver tissue section integrity received a perfect score of 5.0 ± 0.0 following CTP, compared to 4.0 ± 0.5 with MTP ($p = 0.005$). Ribbon formation—a critical factor for producing continuous, high-quality sections—was notably poorer in MTP-processed blocks across all tissue types (Table 5). These findings suggest that while MTP enhances efficiency and most qualitative outcomes, further optimization is needed to improve block consistency and sectioning quality for advanced diagnostic applications.

Table 1: Processing Time Comparison (Hours)

Tissue Type	MTP Mean (Range)	CTP Mean (Range)	p-value
Liver	1.5 (1–2)	14 (12–16)	< 0.001
Kidney	1.5 (1–2)	14 (12–16)	< 0.001
Breast	1.5 (1–2)	14 (12–16)	< 0.001

Table 2: Histological Staining Quality Scores (Mean ± SD)

Tissue Type	Parameter	MTP Mean ± SD	CTP Mean ± SD	p-value
Liver	Cytoplasm Staining	4.7 ± 0.3	4.6 ± 0.4	0.45
Liver	Nucleus Staining	4.6 ± 0.4	4.6 ± 0.3	0.90
Liver	Cellular Detail Clarity	4.6 ± 0.3	4.5 ± 0.4	0.50
Kidney	Cytoplasm Staining	4.7 ± 0.3	4.7 ± 0.3	1.00
Kidney	Nucleus Staining	4.7 ± 0.3	4.7 ± 0.3	1.00
Kidney	Cellular Detail Clarity	4.8 ± 0.2	4.8 ± 0.2	1.00
Breast	Cytoplasm Staining	4.9 ± 0.2	4.7 ± 0.3	0.03*
Breast	Nucleus Staining	4.9 ± 0.2	4.6 ± 0.4	0.02*
Breast	Cellular Detail Clarity	4.8 ± 0.3	4.7 ± 0.3	0.15

Table 3: Morphological Preservation & Cellular Detail Scores (Mean ± SD)

Tissue Type	Parameter	MTP Mean ± SD	CTP Mean ± SD	p-value
Liver	Tissue Morphology	4.8 ± 0.3	4.3 ± 0.4	0.01*
Liver	Cellular Detail	4.7 ± 0.4	4.2 ± 0.4	0.02*
Kidney	Tissue Morphology	4.9 ± 0.2	4.4 ± 0.4	0.01*
Kidney	Cellular Detail	4.8 ± 0.3	4.3 ± 0.3	0.01*
Breast	Tissue Morphology	5.0 ± 0.1	4.5 ± 0.4	0.005*
Breast	Cellular Detail	4.9 ± 0.2	4.4 ± 0.3	0.005*

Table 4: Reagent Consumption per Sample (Mean ± SD, mL)

Reagent	MTP Mean ± SD	CTP Mean ± SD	p-value
Ethanol (total)	100 ± 10	175 ± 15	<0.001
Clearing Agent (Xylene)	12 ± 2	40 ± 5	<0.001
Paraffin Wax	18 ± 3	25 ± 4	0.01*
Total Reagent Usage	130 ± 12	240 ± 22	<0.001

Table 5: Sectioning Quality Scores (Mean ± SD)

Tissue Type	Parameter	MTP Mean ± SD	CTP Mean ± SD	p-value
Liver	Tissue Section Integrity	4.0 ± 0.5	5.0 ± 0.0	0.005*
Liver	Ease of Sectioning	4.0 ± 0.6	5.0 ± 0.0	0.005*
Liver	Ribbon Formation	3.0 ± 0.7	5.0 ± 0.0	0.001*
Kidney	Tissue Section Integrity	4.0 ± 0.5	5.0 ± 0.0	0.005*
Kidney	Ease of Sectioning	3.0 ± 0.6	5.0 ± 0.0	0.001*
Kidney	Ribbon Formation	3.0 ± 0.7	5.0 ± 0.0	0.001*
Breast	Tissue Section Integrity	3.0 ± 0.6	4.0 ± 0.5	0.020*
Breast	Ease of Sectioning	3.0 ± 0.7	4.0 ± 0.5	0.020*

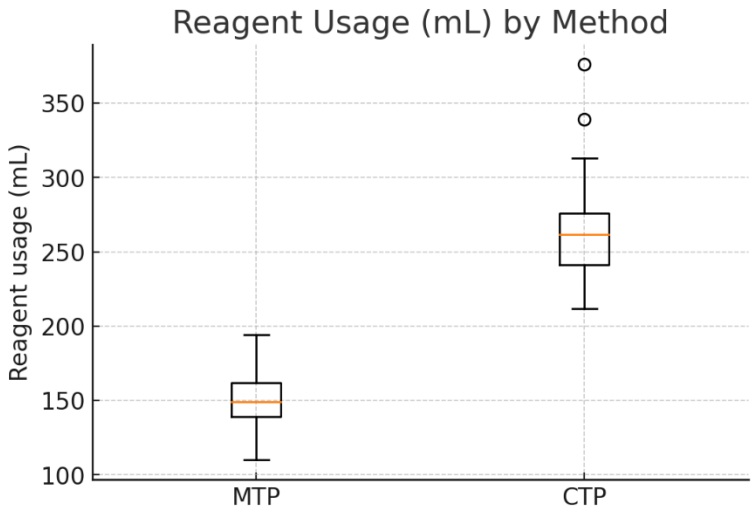

Figure 1. Reager

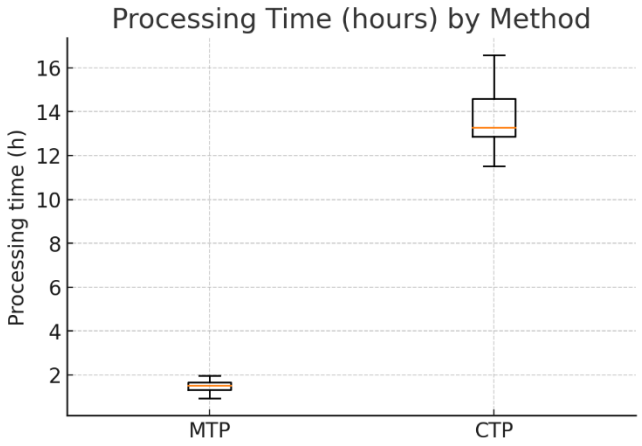

Figure 1. Processing Time Comparison).

Discussion

The findings of this study demonstrate that microwave tissue processing (MTP) is a viable, efficient alternative to conventional methods, offering significant reductions in processing time while maintaining high histological quality. Our results align with prior research [12,13], confirming that MTP can process tissues in 1-2 hours compared to the 12-16 hours required for conventional processing (CTP). This acceleration is particularly advantageous in time-sensitive

diagnostic scenarios, such as intraoperative frozen sections or urgent cancer diagnoses, where rapid turnaround directly impacts patient management.

Beyond speed, MTP preserved or even enhanced histological quality, especially in adipose-rich breast tissues, which exhibited superior nuclear and cytoplasmic staining ($p < 0.03$) compared to CTP. This improvement likely stems from the uniform heat distribution of microwave energy, which minimizes tissue shrinkage and fixation artifacts - common pitfalls of prolonged conventional processing [14]. Notably, liver and kidney tissues processed via MTP also showed comparable cellular detail and architectural preservation, reinforcing its diagnostic reliability across diverse tissue types.

An additional benefit of MTP is its reduced reagent consumption, cutting ethanol and xylene use by 45-50%. This reduction not only lowers costs but also decreases laboratory exposure to toxic chemicals, supporting safer and more sustainable histopathology practices [15]. Such efficiency is particularly valuable in resource-constrained settings, where reagent costs and disposal pose significant challenges.

However, MTP's sectioning quality lagged behind CTP, with poorer ribbon formation and tissue integrity ($p < 0.02$). This limitation, consistent with earlier reports [16,17], may arise from uneven paraffin infiltration or rapid cooling during microwave processing. To address this, future studies should investigate optimized embedding protocols, alternative paraffin formulations, or controlled cooling rates to improve block consistency. Until then, MTP may be best suited for diagnostic workflows where sectioning demands are less stringent, or as a complementary method alongside CTP.

In Summary, MTP represents a major advancement in histopathology, offering unparalleled speed, cost efficiency, and diagnostic-quality staining. While its current limitations in sectioning require further refinement, the technique holds immense promise for modernizing laboratory workflows, particularly in high-volume or urgent diagnostic settings. With targeted improvements, MTP could eventually supplant conventional methods as the gold standard for routine tissue processing.

This study was limited by its sample size (60 tissues) and the use of a single microwave processing protocol, which may not represent all equipment variations. Additionally, advanced diagnostic techniques such as immunohistochemistry and molecular testing were

not evaluated. Future research should involve larger, multi-center trials and explore protocol optimization to enhance sectioning quality.

Conclusion

Microwave tissue processing (MTP) is a highly effective and efficient alternative to the conventional method (CTP). It dramatically reduces processing time from hours to just a couple of hours, which can significantly accelerate diagnostic turnaround. Furthermore, MTP maintains excellent histological quality, and in some cases, provides superior results, particularly in fatty tissues like breast samples. A key benefit is the substantial reduction in reagent consumption, making it more cost-effective and environmentally friendly.

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