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Comparison of traditional and modern postmortem interval estimation techniques in forensic investigations

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Abstract

In the field of death investigation, practical determination of the time since death, or postmortem interval (PMI), is the most important. Several approaches have been devised to allow PMI estimation including the classical methods of the developmental stages and temperature of the body, and molecular biological methods. This paper is intended to present sufficiently, perform and analyze the state of the art of such methods, discuss their strengths and weaknesses, and outline the developments' progress in the context of forensic science, especially the prospective areas of estimating more correctly and reliably the duration of postmortem interval.

Keywords: forensic science, postmortem interval (PMI), Time of death.

Introduction

Postmortem interval estimation (PMI) is an important part of forensic investigations because it provides an estimated time frame until death and helps reconstruct the events surrounding a person's death. Precise PMI estimations may assist detectives in determining the timeline of a crime and minimizing a list of prospective suspects. Furthermore, PMI data helps detect if the dead body was relocated or altered post-mortem, which is crucial in murder examinations. In the absence of dependable PMI information, critical aspects of a case may remain ambiguous, affecting the integrity of the process of justice. A classic example of the application of traditional methods is in cases of stab wounds or gunshot injuries. A forensic pathologist would meticulously document the external wounds, their dimensions, and trajectory. Internally, the dissection would involve tracing the wound pathways through tissues and organs, identifying damaged structures, and estimating blood loss. For instance, in a suspected homicide by stabbing, the pathologist would identify the number and depth of stab wounds, determine the type of weapon used (if possible, based on wound characteristics), and identify critical organ damage leading to death. This process relies heavily on the pathologist's anatomical knowledge, visual inspection, and manual dissection techniques[1,2]. A compelling example of modern methods in action is in cases of sudden cardiac death with no obvious macroscopic findings. Traditionally, such cases might have been attributed to "natural causes" without specific identification of the underlying pathology. However, with modern techniques, a more precise diagnosis can be achieved. For instance, in a young athlete who collapses and dies suddenly, a traditional autopsy might reveal an enlarged heart, but the exact cause of arrhythmia might remain elusive[3]. However, Estimating PMI is difficult because to several variables that impact the breakdown process, such as temperature, humidity, body size, and the surrounding environment. Consequently, forensic scientists use several tools, from conventional physical measurements to sophisticated molecular and biochemical technologies, to enhance the precision of PMI predictions. Conventional techniques have always relied on visible changes in the cadaver, including blind spots and phases of decomposition. But these methodologies are influenced by environmental and personal variability, often resulting in erroneous estimations. Recent science has increasingly prioritized biological, molecular, and computational methodologies[4].

1. Traditional Methods of PMI Estimation: Conventional PMI evaluation techniques rely on discernible physiological alterations that transpire in the body post-mortem. These approaches are simple to use and can be straightforward to use in the field, and therefore have been widely used in forensic pathology for many years. While being influenced by several variables, including environmental circumstances, these approaches remain valuable, particularly during the first postmortem phase.

I. Algor Mortis (Body Cooling)

Postmortem althomy concerns the cooling of the after death. The process of the body temperature changing over time to be more similar to the environmental temperature can be used to determine hypothermia and therefore is a good method of estimating post-mortem interval, but only during the early stages after death.

- Mechanism: When an organism's metabolic activities cease, it cools at a reliably calculable rate depending on ambient temperature, body mass and clothing..
- Timeline: The average cooling process is around 1.5-2°C per hour under typical climatic circumstances, however it may vary according to factors such as humidity, exposure, and overall temperature. Consequently, mathematical models such as Henssge's Nomogram are often used to provide a more precise estimation of PMI, taking into account body weight and outside variables.
- Obstacles: Temperature-dependent PMI estimations are constrained by environmental factors, giving them less trustworthy in certain contexts. For instance, graves discovered in aquatic environments or next to thermal sources have irregular cooling rates[5].

II. Rigor Mortis (Muscle Stiffness)

Rigor mortis is the stiffening of muscles due to biochemical changes that occur after death, particularly the lack of ATP, which is required for muscle relaxation[6].

- Mechanism: Muscle stiffness begins within 2-6 hours postmortem and becomes fully developed after 12-24 hours. The stiffness dissipates as decomposition sets in, typically around 36-48 hours. External factors, such as temperature, body mass, and the cause of death, can affect the onset and duration of rigor mortis.
- Use in PMI: Rigor mortis is helpful in the short-term estimation of PMI (within the first 48 hours), though its onset and duration can be influenced by conditions like cold environments (delayed onset) or high physical exertion prior to death (accelerated onset)[7].

III.Livor Mortis (Postmortem Lividity)

Livor mortis, or hypostasis, is the gravitational pooling of blood in the lower portions of the body, leading to a purplish-red discoloration of the skin [8].

- Mechanism: Livor mortis begins within 30 minutes to 2 hours after death and becomes fixed after 8-12 hours. The patterns of lividity can indicate whether the body has been moved after death and provide clues about the time of death.
- Timeline: Unfixed lividity means the body was moved before livor became fixed, typically within the first 12 hours. Once lividity becomes fixed, it suggests that the body has remained in the same position for at least 12 hours.
- Hurdles: The livor mortis gets influenced by external factors such as external temperature, position of the body, and leakage of blood. Cold environments may interrupt the process but high temperatures could speed it up[9].

IV.Decomposition

It is the breakdown of tissues as a result of autolysis and decay (decomposition). This presents a substantial advance in long-term PMI estimates through insights into the rate and level of decomposition[10].

- Stages:

Fresh Stage: Autolysis begins, and rigor mortis sets in.

Bloat Stage: After 2-3 days, bacterial activity produces gases, causing bloating.

Active Decay: Tissues liquefy, and skin begins to peel after 5-10 days.

Advanced Decay: The body dries out, and skeletonization begins.

- Use in PMI: Decomposition is significantly affected by temperature, humidity, insect activity, and the conditions of burial, submersion, or exposure to air. Advanced forensic methodologies may examine volatile molecules emitted during decomposition to more accurately determine the post-mortem interval in remains discovered in advanced stages of decay.
- Challenges: Decomposition rates fluctuate significantly based on external variables. Consequently, it is challenging to depend only on eye inspection for PMI assessment[11].

2. Modern Methods of PMI Estimation:

Contemporary techniques to estimating postmortem interval (PMI) have developed quickly over the years; enabling biochemical, molecular and technological advances in developing innovations aimed towards enhancing the accuracy of PMI estimation especially when morphological characteristics are less reliable[12].

I. Biochemical Methods

Biochemical approaches focus on the presence of specific indicators in body fluids, tissues or other biologic materials that either deteriorate at a known rate (e.g. enzymes) or change over time after death (e.g. decomposition markers). While these procedures are particularly useful during the early postmortem period, they also offer more precision than standard treatments[13].

A. Vitreous Humor Analysis:

Due to its relatively “hard” texture and being an elastic solid that has no liquid like properties, the vitreous humor is normally unaffected by environmental factors, allowing it to serve as a good medium in post mortem investigations. Concentrations of electrolytes, such as potassium, rise in the vitreous humor in a linear fashion post-cardiac arrest[14].

- Mechanism: After death, cell membranes break down, releasing intracellular potassium into the vitreous humor. This increase occurs at a predictable rate, allowing PMI to be estimated up to 72 hours after death[15].

B. Enzymatic and Protein Degradation

Enzymes and proteins are degraded at known rates after death - for example, enzymes such as creatine kinase and lactate dehydrogenase are degraded in muscle tissue, and certain proteins are degraded in organs such as the brain and liver - measuring these markers can help estimate PMI, especially during the early stages of decomposition[16].

II. Molecular Methods

Molecular techniques analyze the degradation of DNA, RNA, and other molecular structures after death. These techniques are becoming increasingly popular as they provide more detailed insight into the biochemical changes that occur after death[17].

A. RNA Degradation:

RNA is more susceptible to degradation than DNA, making it a useful marker for estimating PMI, especially during the first hours and days after death. Different tissues have different rates of RNA degradation, and some mRNA molecules, such as those in brain, liver, and muscle, serve as reliable PMI indicators[18].

- Mechanism: Postmortem RNA degradation occurs in a time-dependent manner. Researchers have identified specific mRNA transcripts that degrade consistently, allowing for more precise PMI estimates[19].

B. DNA Degradation:

Although DNA is more stable than RNA, it also degrades after death. By analyzing fragmentation patterns of nuclear or mitochondrial DNA, forensic scientists can estimate PMI, especially if decomposition has progressed[20].

C. Forensic Entomology:

Forensic Entomoloy is based on the life cycles of insects and more specifically, the colonizing blowflies that infest the body after death. The larval stages of development, particularly the long-term PMI stages which last for more days to weeks, could be used to estimate time of PMI accurately[21].

- Mechanism: Blowflies are often the first insects to infest a corpse, depositing eggs within hours post-mortem. By identifying the bug species and examining their developmental stages (e.g., eggs, larvae, pupae), entomologists may estimate the duration of the body's exposure[22].

D. Microbial Succession (Thanatomicrobiome):

The study of microbial populations inhabiting cadaveric sites, referred to as thanatomicrobiome, is an emerging area of interest in forensic science. Just as with any other community, certain types of microorganisms appear in a sequential order and change with the environment (such as soil, water, or exposure to air), which enables this method to be used in estimating the postmortem interval, especially in the advanced stages of body decomposition[23,24].

E. Postmortem Imaging (MRI and CT Scans):

Non-invasive imaging modalities, including magnetic resonance imaging (MRI) and computed tomography (CT), have shown efficacy in forensic investigations. These methods enable forensic pathologists to see alterations inside the body, including tissue degradation, fluid buildup, and gas generation, without necessitating an intrusive dissection[25].

F. Artificial Intelligence (AI) and Machine Learning (ML):

AI and ML improve the accuracy of PMI predictions. AI systems could then analyze massive PMA datasets by synthesizing information from various approaches (i.e., molecular markers, microbial communities and environmental factors) to identify patterns of post-mortem change to give more accurate PMI estimates[26].

Table (1): Comparison of Traditional and Modern PMI Estimation Methods:

Category	Traditional Methods	Modern Methods
Effectiveness	- Useful for short-term PMI estimation (up to 48 hours for some techniques). - Accessible and low-tech, widely applied in field work	- Effective for both short-term (hours to days) and long-term PMI estimation. - Incorporates precise molecular, biochemical, and technological data
Examples	- Algor Mortis (Body Cooling) - Rigor Mortis (Muscle Stiffness) - Livor Mortis (Postmortem Lividity) - Visual decomposition	- Biochemical markers (potassium in vitreous humor, protein degradation) - RNA/DNA degradation analysis - Forensic entomology - Microbial succession
Accuracy	- Lower accuracy in unpredictable environmental conditions (e.g., extreme temperatures, water submersion). - Influenced by external factors (clothing, body mass, ambient temperature). - Effective mainly in early PMI estimation.	- High accuracy in both early and late PMI estimation, thanks to the precision of biochemical, molecular, and microbial markers.

		- Machine learning and AI integration improves predictive accuracy based on large datasets
Time Sensitivity	- Effective within the first 48 hours postmortem (e.g., rigor, livor mortis). - Beyond 48 hours, decomposition stages are used, but these are highly variable	- Effective across a wider PMI range (hours to weeks). - Microbial succession and RNA degradation allow for long-term PMI estimation
Reliability	- Less reliable for long-term PMI estimation. - External conditions (temperature, humidity) have significant influence.	- More reliable under varying environmental conditions. - Molecular markers (e.g., RNA, proteins) degrade at predictable rates regardless of external factors.
Speed of Analysis	- Rapid and straightforward; requires minimal equipment. - Suitable for field applications with basic tools.	- Requires laboratory equipment and time for analysis (e.g., RNA extraction, DNA sequencing). - Longer processing time compared to traditional methods
Technological Requirements	- Minimal technology needed (body thermometer, physical examination)	- Requires advanced lab technology (e.g., spectrometers, genetic sequencing, mass spectrometry, AI/ML algorithms).
Limitations	- Subject to environmental variations (temperature, humidity, clothing). - Less effective for PMIs beyond 48-72 hours. - Not applicable in advanced decomposition stages or long-term PMIs	- Requires specialized equipment and expertise (e.g., molecular biology techniques). - May not be accessible in all forensic settings (rural, low-resource environments). - Higher cost and time for analysis.
Applications	- Short-term PMI estimation (< 48 hours). - Most effective in early postmortem period (body cooling, stiffness, lividity).	- Both short-term and long-term PMI estimation. - Suitable for advanced decomposition stages where traditional methods fail

Future Technique

- RNA and DNA Degradation: Progress in molecular biology is enhancing the comprehension of RNA and DNA degradation patterns as indications of post-mortem interval (PMI). Research is concentrating on certain indicators that may provide more precise post-mortem interval estimations, even in degraded remains[28].
- Metabolomics: The examination of metabolic alterations in decaying cadavers is developing as a method for post-mortem interval assessment. Metabolomic profiles may indicate metabolic changes over time, providing a novel approach for ascertaining post-mortem interval (PMI) [29,32].
- Isotope Ratios: Stable isotope analysis, especially of carbon and nitrogen, is used to comprehend breakdown processes and environmental impacts. This technique may provide insights regarding PMI by examining variations in isotope ratios within bodily tissues[30,31].

- Predictive ModelsMachine learning methods are progressively used to forecast PMI using a mix of environmental data, breakdown phases, and other variables. Artificial intelligence models may include several elements to enhance precision and dependability[27].

Areas for Future Research

- Collaborative Research: Future investigations may improve via enhanced cooperation among forensic scientists, biologists, chemists, and data scientists to provide more precise techniques for estimating post-mortem interval (PMI). Cross-disciplinary research may provide creative answers and cohesive techniques[35].
- Protocol Development: Formulating standardized protocols for novel procedures and incorporating them into forensic practice will be essential. Standardization guarantees that novel methodologies are dependable and can be uniformly implemented across several instances[36].
- Privacy and Consent: With the advancement of forensic methods, it is imperative to address the ethical and legal concerns associated with the use of emerging technologies, especially those involving genetic and biometric information. Investigating the ethical ramifications and legal structures will be crucial[37].

Conclusions

PMI estimation has advanced significantly in recent years, transitioning from traditional, observable methods to more precise biochemical, molecular, and technological approaches. While challenges remain, particularly regarding environmental variability and resource accessibility, the integration of multiple methods and the use of AI offer a promising future for forensic science. Continued research into postmortem biological changes and the development of more sophisticated computational models will likely lead to more accurate and reliable PMI estimations, aiding forensic investigations worldwide.

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Author's Declaration

- We hereby confirm that all the Figures and Tables in the manuscript are original and have been created by us.
- We have obtained ethical clearance for our study from the local ethical committee at [Al-Nahrain University/College of Biotechnology]. This approval underscores our commitment to ethical research practices and the well-being of our participants.
- Ethical Clearance: The project was approved by the local ethical committee at [Al-Nahrain University/College of Biotechnology], ensuring adherence to ethical standards and the protection of participants' rights and welfare.

Author's Contribution Statement

All Author: Contributed to the conception and design of the study, data rearrangement and drafted the initial manuscript, collection all part of literature review and conducted some characteristics of the products.

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