DIAGNOSTIC CONCORDANCE IN FORENSIC PATHOLOGY BETWEEN DIGITAL PATHOLOGY SYSTEMS AND OPTICAL MICROSCOPY

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This study evaluated the diagnostic agreement between a Digital Pathology System (DPS) and conventional optical microscopy in forensic pathology. A total of 1,220 slides from 90 forensic cases were digitized and reviewed using both methods by a forensic pathologist, with a washout period between readings. Agreement was analyzed using Cohen's Kappa coefficient under two scenarios: complete agreement and complete agreement plus minor discrepancies. High concordance was observed, with agreement rates of 93.03% (κ = 0.909) and 96.80% (κ = 0.957). Major organs such as the brain, heart, liver, and kidney showed agreement rates above 95% (κ = 0.949 – 1.000), while slightly lower agreement was found in lung and spleen due to histological complexity and postmortem changes. These findings suggest that DPS is a reliable alternative to conventional microscopy in forensic histopathology, though further validation is recommended for specific tissues.

Keywords: Digital Pathology System (DPS), Forensic Pathology, Diagnostic Agreement, Whole Slide Imaging (WSI), DPS Validation.

I. INTRODUCTION

Forensic pathology plays a vital role in medico-legal investigations by providing histological evidence for determining cause and manner of death. In Vietnam, its development remains limited due to a lack of trained personnel and inadequate diagnostic infrastructure. A national survey reported that only 23 of 63 provincial forensic centers are capable of conducting histopathological examinations, primarily due to insufficient resources and facilities.[1]

Digital Pathology Systems (DPS), which allow digitization and remote interpretation of whole slide images (WSI), offer a potential solution to these challenges. DPS has been validated for clinical use by major organizations including the College of American Pathologists (CAP), the Japanese Society of Pathology (JSP), and the Korean Society of Pathologists (KSP).[2-5] Multiple international studies have demonstrated high diagnostic concordance between DPS and traditional optical microscopy, with reported kappa values often exceeding 0.90.[6-8]

Despite this, evidence remains scarce regarding DPS application in forensic pathology, where tissue degradation and postmortem changes complicate interpretation. This study aims to assess the diagnostic concordance between a manually operated DPS and conventional microscopy in forensic histopathology, using a pilot system deployed in Thái Nguyên Province that integrates the Olympus BX43 microscope, Microvisionner software, and VRPACS platform.

This study aimed to evaluate the diagnostic concordance between a Digital Pathology

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System and conventional optical microscopy in forensic histopathology, using Cohen's Kappa coefficient across organ systems under predefined evaluation scenarios.

II. MATERIALS AND METHODS

1. Subjects

Study population and sample size: The study included 90 forensic autopsy cases, from which 610 tissue blocks were obtained. These blocks were processed to yield a total of 1,220 hematoxylin and eosin–stained glass slides, either collected from archival material or newly prepared for analysis.

The inclusion criteria were: Availability of complete case documentation; Adequate formalin fixation and tissue processing; Suitability of histological sections for digital scanning; Institutional approval for research use of the material.

The exclusion criteria included: Cases with severely autolyzed tissue; Inadequate fixation or damaged glass slides;Unrecoverable missing blocks or missing metadata.

2. Methods

Study duration: The study was conducted from February 2024 to February 2025.

Study location: Forensic Center of Thai Nguyen Province, and A Hospital, Thai Nguyen.

Study type: Diagnostic validation study.

Study metrics: Agreement rate and Cohen's Kappa coefficient.

Study design

The diagnostic validation process was conducted based on international guidelines, including those from the College of American Pathologists (CAP), the Japanese Society Pathology (JSP), the Korean Society Pathology (KSP).[2-5]

All case files, tissue blocks, and glass slides used for forensic histopathology examinations were collected from the Forensic Center of Thai Nguyen Province between March 2023 and March 2025.

All case information, tissue blocks, and slides were coded according to a predefined protocol.

Glass slides with inadequate quality were recut and restained using the standard H&E histopathology method.

High-quality slides were digitized using whole slide imaging with an Olympus BX43 microscope integrated with Microvisionner software.

WSI images were transmitted, stored, and displayed on the VRPACS digital pathology management system, an adaptation of radiology PACS.

Diagnostic assessment

A forensic pathologist with over five years of experience in optical microscopy-based forensic histopathology diagnosis was selected.

The pathologist was trained to proficiently interpret digital slides using the VRPACS system.

Slides were randomly reviewed using optical microscopy, and a diagnosis was recorded.

After a washout period of at least two weeks, the slides were randomly reassessed using the VRPACS digital pathology system.

Agreement classification

The diagnostic results from both methods were analyzed and classified into three levels of agreement:

Complete agreement: Diagnoses from both methods are identical.

Partial agreement with minor discrepancy: Discrepancies considered minor if they do not affect the determination of the cause of death. A predefined list of minor discrepancies was established.

No agreement (major discrepancy): Major discrepancies occur when there is a fundamental difference in the nature of the lesion, leading to a complete change in the cause of death determination.

Two scenarios were considered:

Scenario 1: Agreement based solely on complete agreement.

Scenario 2: Agreement including both complete agreement and minor disagreement.

Cohen's Kappa coefficients and their standard errors (SE) were computed for each scenario, along with confidence intervals for agreement rates.

Statistical Analysis

Data were processed and analyzed using R statistical software (version 4.3.2) and Microsoft Excel. Diagnostic agreement between the Digital Pathology System and conventional optical microscopy was assessed using Cohen's Kappa coefficient, a statistical measure that quantifies the level of agreement beyond chance. Kappa values were calculated for two evaluation scenarios: (1) based on complete agreement only, and (2) including both complete agreement and minor discrepancies.

Diagnostic agreement rates were calculated as the percentage of slides with concordant diagnoses out of the total number of slides assessed. For each organ and evaluation scenario, the following statistical parameters were determined: 95% confidence intervals (CIs) for agreement rates, absolute CI width, standard error (SE), and SE as a percentage of the agreement rate (%SE).

Similarly, Cohen's Kappa coefficients were reported with their corresponding 95% CIs, standard errors, and %SE. Confidence intervals were estimated using the approximate normal distribution based on SE values. Organ-specific analyses were performed to evaluate the stability and variability of diagnostic concordance across different tissue types.

All analyses were conducted with a significance threshold set at $p < 0.05$.

3. Research ethics

The study is part of the research project titled *"Study on histopathological lesion characteristics and the role of digital pathology in forensic examination",* which has been approved by the Biomedical Research Ethics Committee of Hanoi Medical University under Certificate No. 1141/GCN-HMUIRB, issued on January 18, 2024.

The research solutions and tools used in this study have been evaluated, approved, and authorized for implementation by the Thai Nguyen Department of Health under Decision No. 2587/QĐ-SYT, dated November 24, 2023, and by the Thai Nguyen Provincial People's Committee under Decision No. 591/QĐ-UBND, dated March 27, 2024.

III. RESULTS

Table 1 presents the agreement rates between DPS and optical microscopy across different organs. The overall agreement rate in Scenario 1 was 93.03%, while in Scenario 2, it increased to 96.80%.

Table 2 presents the agreement rates along with their 95% confidence intervals (CIs), absolute CI widths, and standard errors (SE) expressed as a percentage of agreement rate for each organ under both evaluation scenarios. In Scenario 1, the overall agreement rate was 93.03% (CI: 91.50% – 94.56%), while in Scenario 2, it increased to 96.80% (CI: 95.62% – 97.98%). Stomach and skin exhibited the highest agreement (100%) with the narrowest CI width, whereas lung and spleen had the lowest agreement rates (86.41% and 86.00%, respectively) with wider CI ranges.

Table 1. The agreement rates between DPS and optical microscopy

Organ	Total Organs	Total Slides	Complete Agreement	Disagreement		Scenario 1		Scenario 2	
				Minor	Major	Agreement Rate (%)	Kappa	Agreement Rate (%)	Kappa
Lung	92	184	159	13	12	86.41	0.826	93.48	0.913
Heart	84	168	155	5	8	92.26	0.908	95.24	0.951
Liver	82	164	150	9	5	91.46	0.900	96.95	0.955
Brain	75	130	125	3	2	96.15	0.956	98.46	0.979
Stomach	63	126	124	2	0	98.88	0.979	100	1.00
Adrenal gland	63	126	120	3	3	95.24	0.932	97.62	0.968
Kidney	59	118	120	3	3	95.24	0.890	96.61	0.949
Pancreas	45	90	87	1	2	96.67	0.949	97.78	0.962
Spleen	25	50	43	4	3	86.00	0.821	94.00	0.920
Skin	32	64	62	2	0	96.88	0.970	100	1.00
Total	610	1220	1135	46	39	93.03	0.909	96.8	0.957

Table 2. Agreement Rates and Stability Metrics by Organ Across Different Evaluation Scenarios

Organ	Scenario 1					Scenario 2				
	Agreement Rate (%)	95% CI Lower (%)	95% CI Upper (%)	Absolute CI Width (%)	SE % of Agreement Rate	Agreement Rate (%)	95% CI Lower (%)	95% CI Upper (%)	Absolute CI Width (%)	SE % of Agreement Rate
Spleen	86.00	79.36	92.64	13.28	5.58	94.00	87.91	100	12.09	3.65
Lung	86.41	82.07	90.75	8.68	3.01	93.48	89.91	97.04	7.13	1.98
Liver	91.46	87.42	95.50	8.08	2.41	96.95	94.32	99.58	5.26	1.60
Heart	92.26	88.18	96.34	8.16	2.30	95.24	92.02	98.46	6.44	1.75
Kidney	95.24	89.18	97.26	8.08	2.52	96.61	93.34	99.88	6.54	1.89
Adrenal gland	95.24	92.01	98.47	6.46	2.05	97.62	95.28	99.97	4.96	1.44
Pancreas	96.67	93.78	99.56	5.78	1.99	97.78	95.16	100	4.84	1.49
Brain	96.15	93.14	99.16	6.02	1.82	98.46	96.64	100	3.36	1.33
Skin	96.88	94.13	99.63	5.50	2.14	100	100	100	0.00	0.00
Stomach	98.88	96.26	100	3.74	1.37	100	100	100	0.00	0.00
Total	93.03	91.50	94.56	3.06	0.98	96.8	95.62	97.98	2.36	0.69

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Table 3. Cohen's Kappa Values and Associated Stability Metrics by Organ Across Different Evaluation Scenarios

Organ	Scenario 1						Scenario 2					
	Kappa	95% CI Lower	95% CI Upper	CI Width	SE	SE % of Kappa (%)	Kappa	95% CI Lower	95% CI Upper	CI Width	SE	SE % of Kappa (%)
Spleen	0.821	0.698	0.944	0.246	0.0626	7.63	0.920	0.832	1.000	0.168	0.0448	4.87
Lung	0.826	0.763	0.889	0.126	0.0324	3.92	0.913	0.866	0.960	0.094	0.0243	2.66
Kidney	0.890	0.840	0.940	0.100	0.0257	2.89	0.949	0.913	0.985	0.072	0.0185	1.95
Liver	0.900	0.864	0.936	0.072	0.0185	2.06	0.955	0.929	0.981	0.052	0.0134	1.40
Heart	0.908	0.879	0.937	0.058	0.0147	1.62	0.951	0.931	0.971	0.040	0.0102	1.07
Adrenal gland	0.932	0.892	0.972	0.080	0.0201	2.16	0.968	0.941	0.995	0.054	0.0138	1.43
Pancreas	0.949	0.908	0.990	0.082	0.0208	2.19	0.962	0.933	0.991	0.058	0.0151	1.57
Brain	0.956	0.926	0.986	0.060	0.0154	1.61	0.979	0.958	1.000	0.042	0.0108	1.10
Skin	0.970	0.935	1.00	0.065	0.0181	1.87	1.00	1.000	1.000	0.000	0.000	0.00
Stomach	0.979	0.950	1.00	0.050	0.0147	1.50	1.00	1.000	1.000	0.000	0.000	0.00
Total	0.909	0.890	0.928	0.038	0.0095	1.04	0.957	0.944	0.970	0.026	0.0067	0.70

Table 3 presents the Cohen's Kappa values along with their standard errors (SE), SE as a percentage of Kappa, and 95% confidence intervals (CIs) for each organ under both evaluation scenarios. In Scenario 1, the overall Kappa value was 0.909 (CI: 0.890 – 0.928), increasing to 0.957 (CI: 0.944 – 0.970) in Scenario 2. Stomach and skin exhibited the highest Kappa values (1.000), indicating perfect agreement, while spleen and lung had the lowest Kappa values (0.821 and 0.826, respectively). The CI width was narrowest for high-agreement organs and wider for tissues with lower concordance, such as spleen (CI width: 0.168 in Scenario 2).

Table 4. Summary of Key Histopathological Findings in 10 Organs

Organ (number of samples)	Characteristic Histopathological Lesion	Number of samples	Proportion (%)
Lungs (n = 184)	Acute pulmonary edema	155	84.2
	Suppurative pneumonia/bronchopneumonia	95	51.6
	Alveolar hemorrhage	40	21.7
	Carcinoma	2	1.1
	Severe autolysis	46	25.0
Liver (n = 164)	Congestion – central lobular necrosis	105	64.0
	Fatty liver degeneration	55	33.5
	Focal necrosis	25	15.2
	Carcinoma	1	0.6
	Severe autolysis	41	25.0
Heart (n = 168)	Congestion, myocardial degeneration	110	65.5
	Acute myocardial infarction	22	13.1
	Infective endocarditis	1	0.6
	Severe autolysis	34	20.2
Brain (n = 130)	Global cerebral edema	100	76.9
	Large intracerebral hemorrhage	10	7.7
	Microhemorrhages	20	15.3
	Severe autolysis	13	10.0
Kidneys (n = 118)	Acute tubular necrosis	60	50.8
	Congestion – interstitial edema	45	38.1
	Renal infarction	1	0.8
	Glomerulonephritis/tubulointerstitial nephritis	12	10.2
	Severe autolysis	24	20.3

Organ (number of samples)	Characteristic Histopathological Lesion	Number of samples	Proportion (%)
Stomach (n = 126)	Mucosal hemorrhage	55	43.7
	Mucosal necrosis	10	7.9
	Acute gastritis/ulcer	15	11.9
	No significant lesion	46	36.5
	Severe autolysis	31	24.6
Adrenal glands (n = 126)	Acute adrenal hemorrhage	8	6.3
	Cortical necrosis	10	7.9
	No significant lesion	108	85.8
	Severe autolysis	30	23.8
Pancreas (n = 90)	Acute pancreatitis/hemorrhagic necrosis	5	5.6
	No significant lesion	85	94.4
	Severe autolysis	22	24.4
Skin (n = 64)	Subcutaneous hemorrhage/hematoma (hanging)	50	78.1
	Edema, perivascular inflammation	11	17.2
	Electrical injury	3	4.7
	Severe autolysis	8	12.5
Spleen (n = 50)	Congestion	15	30.0
	Lymphoid tissue necrosis	8	16.0
	Acute hemorrhage	7	14.0
	No significant lesion	20	40.0
	Severe autolysis	10	20.0

Table 4 shows that acute pulmonary edema, pneumonia, hepatic congestion, acute tubular necrosis, and cerebral edema were the most common findings, reflecting respiratory failure, infections, and multi-organ dysfunction as major causes of death. The proportion of severely autolyzed samples ranged from 10% to 25% depending on the organ, indicating the impact of decomposition on histopathological assessment.

IV. DISCUSSION

The results of this study demonstrate that the Digital Pathology System can achieve a very high level of diagnostic concordance with conventional optical microscopy in forensic histopathology. This high agreement, reflected by Cohen's Kappa values ranging from 0.909 to 0.957, underscores the reliability and practical feasibility of DPS, especially in provincial forensic centers. Tissues with homogeneous

structures and limited postmortem changes, such as the stomach, skin, and brain, showed near-perfect concordance, suggesting that digital slides can faithfully represent histological details. Conversely, lower concordance observed in lung and spleen tissues is likely attributable to complex microarchitecture, extensive vascular components, or rapid autolysis after death-factors that complicate image interpretation.

When compared to international studies, the level of agreement observed in this research appears higher. For instance, Goacher et al. (2017), in a systematic review of 38 studies, reported a mean concordance rate of 92.4% and a weighted mean Kappa of 0.76, lower than the 96.8% and 0.957 obtained in the present study.[6] Bauer et al. (2013) found a Kappa of 0.90 when validating whole slide imaging (WSI) for primary diagnosis in surgical pathology, which aligns with the more stringent scenario 1 in our study.[7] At a large academic center, Hanna et al. (2019) reported 99.3% diagnostic concordance using WSI for routine diagnoses but also noted a 19% decrease in efficiency compared to conventional microscopy-a factor not assessed in the current research.[8] These differences may stem from the nature of forensic pathology, where lesions are often macroscopically evident and carry strong diagnostic significance, resulting in less diagnostic ambiguity than in clinical pathology.

Among the examined organs, the lung and spleen demonstrated the lowest diagnostic concordance between digital pathology and conventional microscopy, with agreement rates of 86.41% and 86.00%, and corresponding Cohen's Kappa values of 0.826 and 0.821, respectively. These relatively lower values are attributable to the histological complexity and postmortem changes that affect these tissues in forensic settings. Lung tissue is particularly prone to features such as congestion, edema, hemorrhage, inflammation, and autolysis, which often overlap and reduce diagnostic clarity. Similarly, the spleen is highly vascular and undergoes rapid postmortem degradation, including hemolysis and architectural disruption, complicating the recognition of meaningful pathological changes.

These findings are consistent with prior literature. Thrall et al reported limitations in interpreting pulmonary pathology using digital platforms, noting that variations in preservation and tissue architecture significantly affect interpretive consistency.[11] Goacher et al, in a systematic review of 38 validation studies, observed that tissues with microarchitectural complexity-including the lungs and lymphoid organs-tend to have lower diagnostic concordance between whole slide imaging (WSI) and conventional microscopy.[6] Wilbur et al further noted that hemorrhagic or necrotic tissues may present interpretive challenges on digital platforms, especially in low-contrast or degraded areas.[10]

The histopathological findings summarized in Table 4 of this study provide additional insights into the concordance results. The high frequency of lesions such as acute pulmonary edema, pneumonia, hepatic congestion, acute tubular necrosis, and cerebral edema reflects respiratory failure, severe infections, and multi-organ dysfunction as the predominant mechanisms of death in the studied cases. These common and easily recognizable lesions likely contributed to the high diagnostic agreement observed in most organs, as they offer clear histological patterns that are well captured by digital imaging.

However, the presence of a substantial proportion of samples with severe autolysis (ranging from 10% to 25% depending on the organ) highlights the challenges posed by

postmortem decomposition, which can obscure histological details and lead to diagnostic discrepancies, particularly in tissues like the lung and spleen.

It should also be noted that this study was conducted using samples collected from a single forensic center. As a result, the spectrum of pathological lesions may not fully represent the diversity of cases encountered in broader forensic practice, and the number of samples with complex or rare lesions (e.g., severe head trauma, congenital heart disease, metastatic cancer) was limited. This could limit the generalizability of the findings and may partly explain the overall high concordance observed, as the dataset predominantly included common, straightforward pathological patterns. Future multicenter studies with larger, more varied case collections, including challenging and uncommon lesions, are necessary to comprehensively validate the diagnostic performance of digital pathology systems in forensic settings.

The implications of these findings are particularly significant in the context of Vietnam. As of 2021, only 23 out of 63 provincial forensic centers had the capacity to conduct histopathological examinations, primarily due to a severe shortage of forensic pathologists (Hung, 2021).[1] Additionally, inadequate laboratory infrastructure, high initial investment costs, and a limited pace of digital transformation in the forensic sector remain key barriers. Against this backdrop, the successful application of DPS-particularly when integrated with VRPACS for image storage and transmission-offers a viable solution to bridge the gap in diagnostic capacity, enabling remote consultation and expanding access to forensic pathology services.

Nevertheless, several limitations must be acknowledged. First, all slide assessments were performed by a single pathologist, which may introduce observer bias and limits the evaluation of inter-observer variability. This limitation is due to the study's primary focus on intra-observer repeatability. Future studies should involve multiple pathologists to better assess reproducibility across observers. Second, the study was conducted at a single center with well-controlled slide quality, which may not reflect the variability found in other institutions with different resources and technical workflows. Multicenter studies across diverse settings are necessary to generalize the findings. Moreover, the digitization process relied on a manually operated scanner, which is susceptible to technical inconsistencies that may affect image quality and diagnostic accuracy.

To overcome these limitations, future research should explore the use of automated high-throughput slide scanners, standardized workflows, and integration of artificial intelligence (AI) algorithms to assist in lesion detection and classification. AI-supported DPS models, trained on forensic-specific digital datasets, could enhance diagnostic accuracy, reduce workload, and compensate for the shortage of trained personnel in remote or underserved areas.

V. CONCLUSION

This study confirms the high reliability of Digital Pathology Systems (DPS) in forensic histopathological diagnosis, demonstrating diagnostic agreement comparable to conventional optical microscopy, with only minor discrepancies in certain organ systems. The successful integration of DPS with the VRPACS image management platform highlights its potential for broader implementation in both forensic and routine histopathology practice.

While this study represents an important first step, we fully acknowledge its limitations-

particularly the single-center setting, the use of a single pathologist, and the imbalance in sample sizes across organs. To address these issues, we are firmly committed to conducting follow-up research with a multicenter design, involving multiple independent pathologists and a more balanced distribution of tissue samples. These steps will ensure more robust validation and enhance the generalizability of our findings.

Through these efforts, we aim to support the reliable and scalable adoption of digital pathology in forensic medicine, especially in resource-limited settings.

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