



Original Article

## Estimation of Post mortem Interval from Renal Histopathological Changes: Effect of Mechanical Trauma in Medico-Legal Autopsies

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### ABSTRACT

**Background:** Estimation of postmortem interval (PMI) is a crucial component of forensic investigations. Conventional methods based on gross postmortem changes are often influenced by environmental and intrinsic factors, limiting their accuracy. Histopathological evaluation of internal organs, particularly the kidney, offers a more objective approach. However, the presence of mechanical trauma may alter tissue morphology and affect interpretation.

**Aim:** To estimate the postmortem interval using histopathological changes in the kidney and to evaluate the influence of mechanical trauma on these changes.

**Material and Methods:** This prospective study was conducted on 150 medico-legal autopsy cases with known time of death at UCMS and GTB Hospital, Delhi. Cases were categorized into six groups based on PMI (3–39 hours). Kidney tissue samples were collected, processed, and stained with hematoxylin and eosin. Histopathological changes in glomeruli, proximal convoluted tubules, and distal convoluted tubules were graded from G0 to G4. Statistical analysis was performed using ANOVA and Tukey's post hoc test.

**Results:** A progressive increase in degenerative histopathological changes was observed with increasing PMI. Grade 1 changes predominated in early intervals (3–9 hours), while higher grades (G3 and G4) were more frequent beyond 27 hours. No Grade 0 changes were observed. The mean PMI increased significantly across grades ( $p = 0.001$ ), demonstrating a strong correlation between histological changes and time since death. However, trauma-related factors such as hypovolemic shock and ischemia produced overlapping features, including tubular necrosis and nuclear loss.

**Conclusion:** Renal histopathology is a reliable adjunct for estimating postmortem interval, showing a significant correlation with time since death. However, mechanical trauma acts as an important confounding factor by inducing ante-mortem changes that may mimic or accelerate postmortem autolysis. Therefore, histopathological findings should be interpreted in conjunction with clinical history, type of injury, and environmental conditions for accurate PMI estimation.

**Keywords:** Postmortem interval; Kidney histopathology; Autolysis; Mechanical trauma; Forensic pathology; Tubular necrosis; Medico-legal autopsy.

### INTRODUCTION

One objective of postmortem examination is to estimate time since death. It plays crucial role in investigation by the investigating officer.

The estimating time since death gives a primary idea of time of assault to the police and to check whether it is reliable with the alibi of the suspect or not. [1]

The conventional methods of ascertaining the time since death are based on naked eye observations of the gross changes in a dead body occurring after death. Various gross changes in the body after death are loss of corneal reflex and changes in eye, cooling of the body, post mortem hypostasis, rigor mortis, decomposition and other putrefactive changes. Some clue regarding time of death is also gathered from the condition of food in stomach, intestine and urine in bladder. (2, 3, 4, 5)

Forensic pathologists are trying to establish time passed since death by studying degenerative changes in organs and tissues at different intervals such as bone marrow (6), spleen (7) brain tissue (8) and Gingival tissue (9).

Immediately or shortly after death, many degenerative changes commence in the body and progress in fairly orderly manner until the body disintegrates. Each change has its own time, factor or rate. Unfortunately, these rates of development of post mortem changes are strongly influenced by unpredictable endogenous and environmental factors. (10)

The renal cortex is composed of glomeruli, Proximal Convoluted Tubules (PCT), Distal Convoluted Tubules (DCT) in cortical labyrinth and Collecting Tubules (CT) within medullary rays. Glomeruli are composed of tufts of capillaries in contact with mesangial matrix and glomerular epithelium. (11)

Tufts are enclosed in Bowman's capsules, composed of visceral and parietal epithelia separated by a potential space Bowman's space. The urinary pole of glomeruli leads into PCT that are tortuous tubules lined by pyramidal cells with basally located, round, vesicular nuclei with prominent nucleoli. The luminal surface of these epithelial cells is lined by a mucopolysaccharide brush border of microvilli. The bulk of the renal cortex is composed of these tubules. Distal convoluted tubules are lined by low cuboidal epithelial cells, resulting in larger luminal diameters than in PCT. These cells have centrally located nuclei and no brush borders on luminal surface. Within the medullary rays the CT are lined by low cuboidal epithelium with scalloped-edged luminal borders, pale cytoplasm, and dark, spherical centrally located nuclei. (11)

Body tissues are affected by autolysis at variable rates depending on sensitivity of their cells to anoxia and cellular concentration of proteolytic enzymes. Renal proximal convoluted tubules and renal medulla are tissue reported to be rapidly altered. (12, 13)

Very few studies have been conducted by histopathological studies to estimate time since death in Delhi.

**Aim:** - To estimate the post-mortem interval by histopathology changes in the Kidney.

## **MATERIAL AND METHOD**

The present study was conducted in the Department of Forensic Medicine and Department of Pathology, UCMS and GTB Hospital, Delhi during November 2014 to April 2016.

### **Sample size**

Considering a range of duration of post mortem interval from 3-39 hours, to estimate a difference of 10 % of median time of 21 hours ( $\alpha= 5\%$ ), a sample size of 135 cases were required, but we took 150 cases so that every group had at least 25 cases. These cases were divided into six groups according to time since death with the interval of 6 hours -

- Group 1 – From 3 to <9 hours
- Group 2 - From 9 to <15 hours
- Group 3 – From 15 to <21 hours
- Group 4 – From 21 to <27 hours
- Group 5 – From 27 to <33 hours
- Group 6 – From 33 to 39 hours.

Criteria for selection of subjects

### **Inclusion criteria**

All cases with the alleged history of mechanical trauma, where time of death was known and verified by hospital records were taken.

### **Exclusion criteria**

- Those Cases having known history of any pathology of Kidney were excluded.
- Any injury or pathology grossly observed during post mortem examination was excluded.
- Cases in which any sign of natural disease was found during histopathological examination were excluded.

## METHOD

Total of 150 cases, irrespective of age and sex brought to the mortuary of UCMS & GTB Hospital for medico legal autopsies were taken. The autopsy was done by standard procedure/method. Both kidneys were separated and examined for any gross abnormality and sign of disease. Tissue measuring 2.5 cm x 2.5 cm from right kidney having capsule, cortex and medulla was taken. The sample was kept in 10% formalin for 24 hrs for fixation. Small pieces or blocks of tissues each measuring approximately 1cmx1cmx2mm was taken for histopathological examination and was processed by the standard methods of processing for histopathological study. (14)

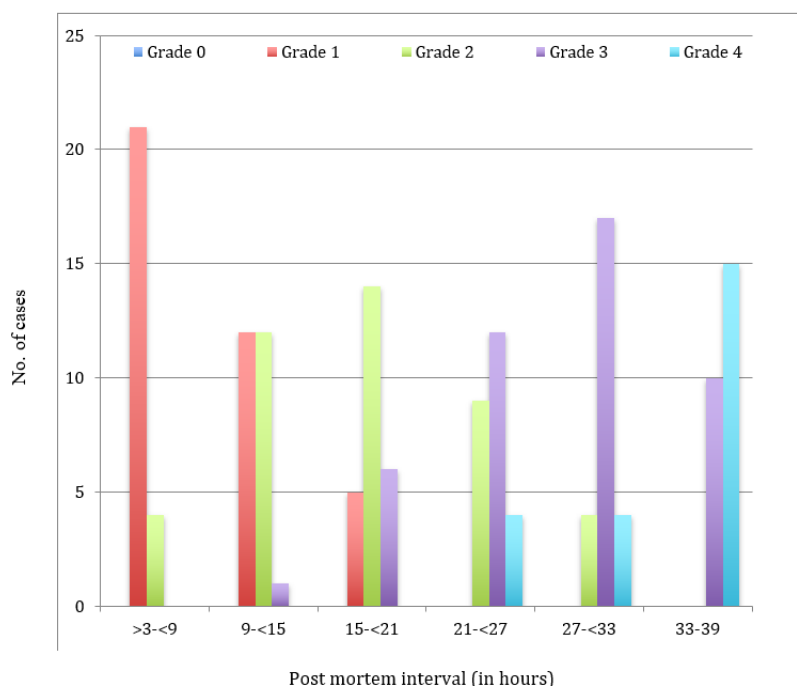
The paraffin sections of tissues were labeled during the process of block making. From the block of tissues, sections were cut at 4-5 micron thickness with a rotating microtome. The sections were placed in warm water at 50°C in a tissue floatation bath for spreading out and will be mounted on glass slide smeared with albumin glycerin solutions. The slides were stained by Haematoxylin and Eosin stain. The stained slides were examined under microscope for studying the various histopathological changes that take place at different postmortem interval under 10x magnification and then 40 x magnifications. The stained slides were observed according to the grading. (15)

- G-0: No Change
- G-1: Mild (Architecture maintained, mild cloudy swelling and disruption of tubular epithelium)
- G-2: Moderate (Architecture maintained, more cloudy swelling and disruption of epithelium, glomeruli swollen)
- G-3: Severe (Architecture disturbed, cloudy swelling and disruption of epithelium is prominent. Collapse of glomeruli)
- G-4: Very Severe (Complete collapse of glomeruli, marked disruption of epithelium. Nuclei are fragmented)

## RESULT AND OBSERVATION

Total 150 cases were examined in which males were 76% (n=114) and females were 24% (n=36). Degenerative changes in kidney were studied in Glomerulus, Proximal convoluted tubule and distal convoluted tubule. In Glomeruli necrotic changes were studied. In Proximal Convoluted Tubule cell disruption with loss of nuclei was studied. In Distal convoluted Tubule shedding of cells with loss of nuclei was studied and histological changes were graded.

As shown in Fig.1, it was observed that Grade 0 was not found in any case in present study it is because in present study samples were taken from the cases where time since death was at least 3 hours. Grade 0 is classified as no change in histopathological examination. It suggests that some histopathological changes start immediately after death or within 3 hours of death.



**Fig. 1: Cluster bar diagram showing distribution of histopathological grades of Kidney in relation to post mortem interval.**

At post mortem interval >3 and <9 hours, 21 cases showed histopathological changes of Grade 1 which correspond to 84% of total cases, 4 cases showed histopathological changes of Grade 2 which correspond to 16% of total case.

At post mortem interval 9 to <15 hours, 12 cases showed histopathological changes of Grade 1 which correspond to 48% of total cases, 12 cases showed histopathological changes of Grade 2 which correspond to 48% of total cases and 1 case showed histopathological change of Grade 3 which correspond to 4% of total cases.

At post mortem interval 15 to < 21 hours, 5 cases showed histopathological changes of Grade 1 which correspond to 20% of total cases, 14 cases showed histopathological changes of Grade 2 which correspond to 56% of total cases and 6 cases showed histopathological changes of Grade 3 which correspond to 24% of total cases.

At post mortem interval 21 to < 27 hours, 9 cases showed histopathological change of Grade 2 which correspond to 36% of total cases, 12 cases showed histopathological change of Grade 3 which correspond to 48% of total cases, 4 cases show histopathological change of Grade 4 which correspond to 16% of total cases.

At post mortem interval 27 to < 33 hours, 4 cases showed histopathological change of Grade 2 which correspond to 16% of total cases, 17 cases showed histopathological change of Grade 3 which correspond to 68% of total cases, 4 cases showed histopathological change of Grade 4 which correspond to 16% of total cases.

At post mortem interval 33 to 39 hours, no case showed histopathological change of Grade 1, 5 cases showed histopathological change of Grade 2 which correspond to 20% of total cases, 6 case showed histopathological change of Grade 3 which correspond to 24% of total cases and 14 cases showed histopathological change of Grade 4 which correspond to 56% of total cases.

It was observed that as the post mortem interval is increasing the degenerative changes also increases. To find any statistical significance between the groups ANOVA test was applied. P value .001 was obtained which was statistically significant. Table 1 shows that, the histopathological changes of Grade 1 observed in minimum 3 hours and maximum 18 hours. Maximum numbers of cases (10 out of 17) were observed at 8 hours after death. Mean time since death of Grade 1 was 10 hours.

**Table 1: Mean, Mode, and Range of Postmortem interval in relation to histopathological grades of Kidney.**

Histopathological grades of Kidney	Minimum duration (Hrs)	Maximum duration (Hrs)	Mean time since death (Hrs)
0	-	-	-
1	3	18	10
2	6	31	17
3	4	39	26
4	21	39	33

Histopathological changes of Grade 2 observed in minimum 6 hour and maximum 31 hours after death. Maximum numbers of cases were observed at 20 (4 out of 44) hours. Mean time since death of Grade 2 was 26 hours.

Histopathological changes of Grade 3 observed in minimum 4 hours and maximum 39 hours after death. Maximum numbers of cases (4 out of 45) were observed at 32 hours. Mean time since death of Grade 3 was 26 hours.

Histopathological changes of Grade 4 observed in minimum 21 hours and maximum 39 hours after death. Maximum numbers of cases (6 out of 23) were observed at 36 hours. Mean time since death of Grade 4 was 33 hours.

It was observed that if we compare each grade with other grade, the mean time since death with standard deviation in each grade were statistically significant with p value .001 by Turkey's post hoc test (Table 2).

**Table 2: Tukey's Post hoc test for histopathological grades of Kidney**

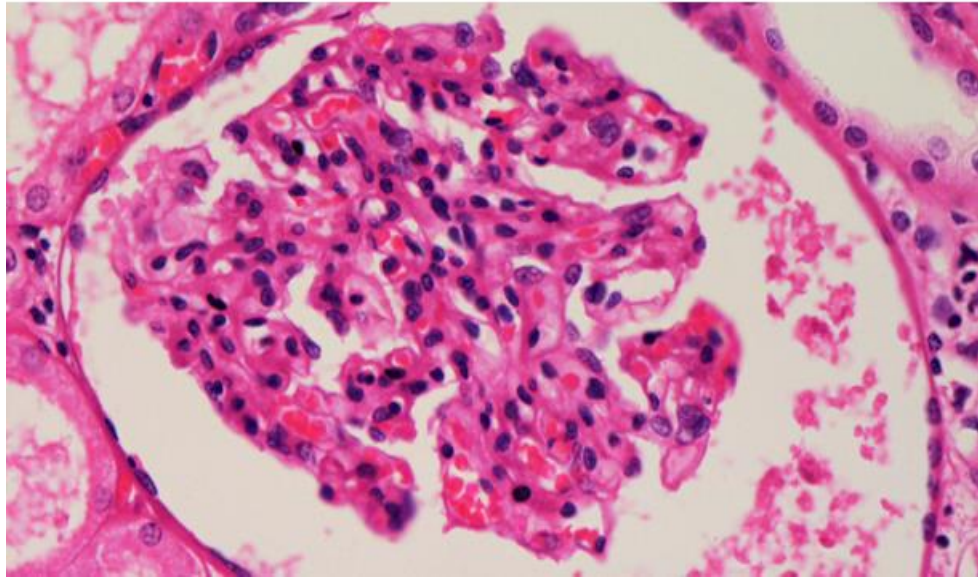
Histopathological grades of Kidney	Post Mortem interval Mean ± Standard Deviation	P value
Grade 1	10.2±4.0	.001
Grade 2	17.6±6.3	.001
Grade 3	26.8±8.5	.001
Grade 4	33.0±5.6	.001

**Table 3: Comparison of histopathological changes in Kidney with other study**

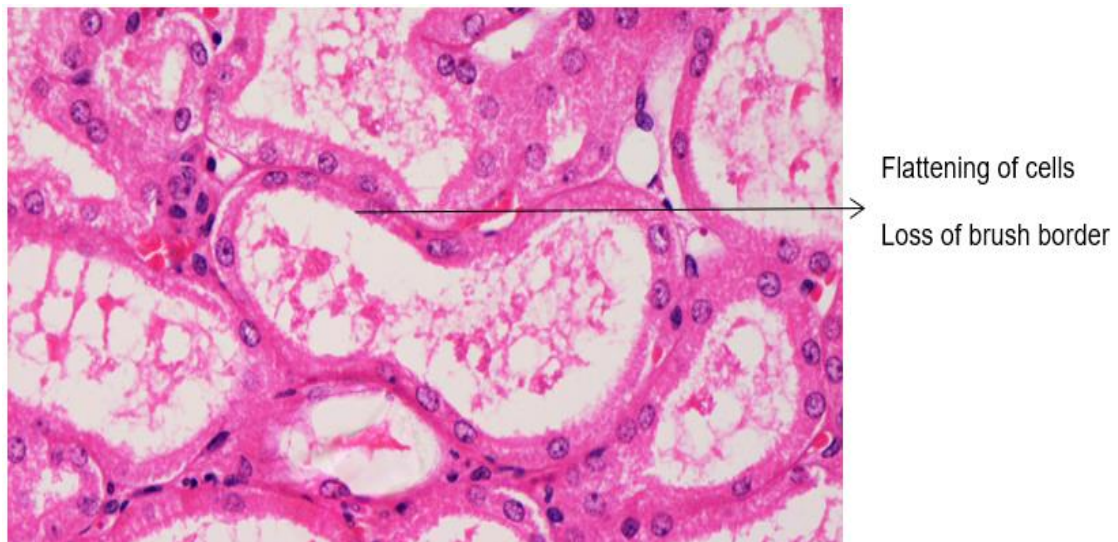
Present study	Vinita Kushwaha et al
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>3 - <9 hours	Loss of brush border, Flattening of cells	Up to 12 hours	Cloudy swelling, disruption of epithelium
9 - <15 hours	Partial loss of cells	13 - 18 hours	More swelling, glomeruli swollen
15 - <21 hours	PCT tubular cell disruption	19 - 24 hours	Collapse of glomeruli
21 - <27 hours	Loss of nuclei of PCT, DCT focal loss of nucleus	25 - 30 hours	Glomeruli swollen
27 - <31 hours	PCT necrotic, DCT shedding	31 - 39 hours	More swelling, glomeruli swollen
33 - 39 hours	Glomeruli necrotic		

**KIDNEY HISTOPATHOLOGIC CHANGES- GRADE 1**

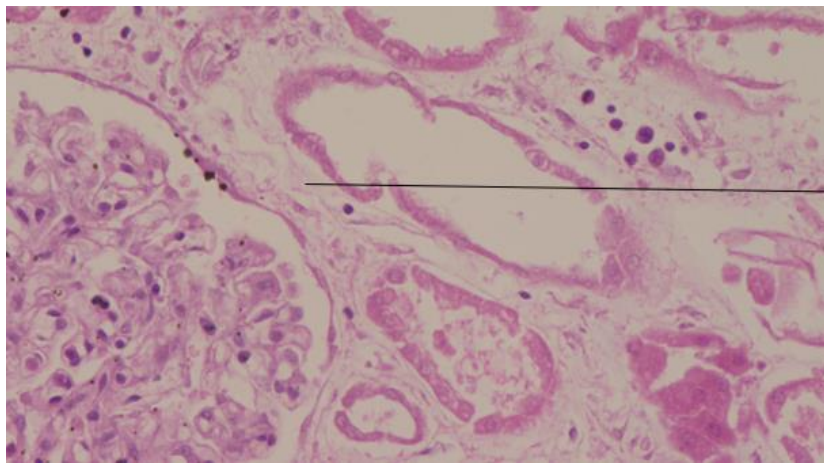


**Fig 2 : Showing unremarkable glomeruli (H & E Staining X 400)**



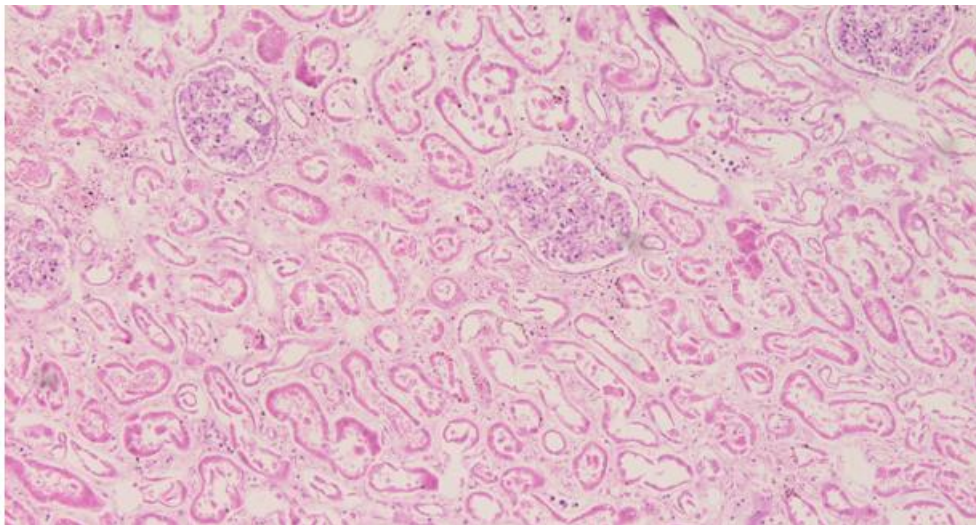
**Fig 3 : Showing flattening of cells and loss of brush border of proximal convoluted tubule .(H & E Staining X 400 )**

**KIDNEY HISTOPATHOLOGIC CHANGES- GRADE 2**



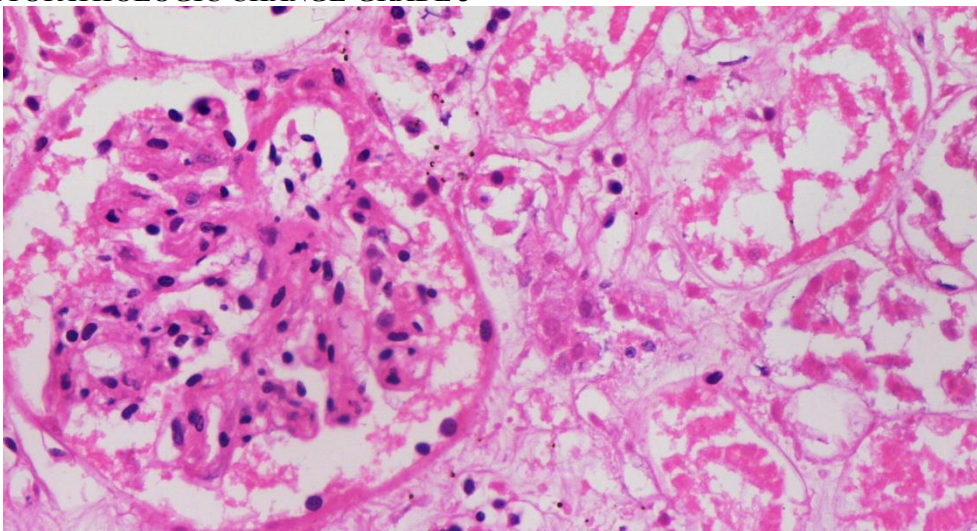
→PCT tubular cell  
disruption,  
Focal loss of nuclei

**Fig 4 : Showing cell disruption and loss of nuclei of proximal convoluted tubule (H & E Staining x 400)**

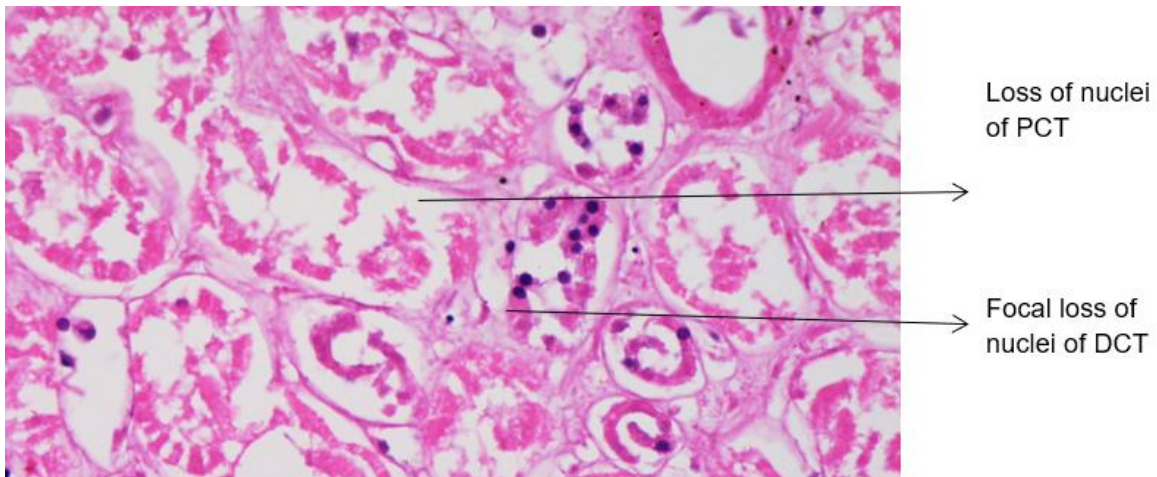


**Fig 5 : Showing degenerative changes (H & E Staining X 100)**

### **KIDNEY HISTOPATHOLOGIC CHANGE-GRADE 3**

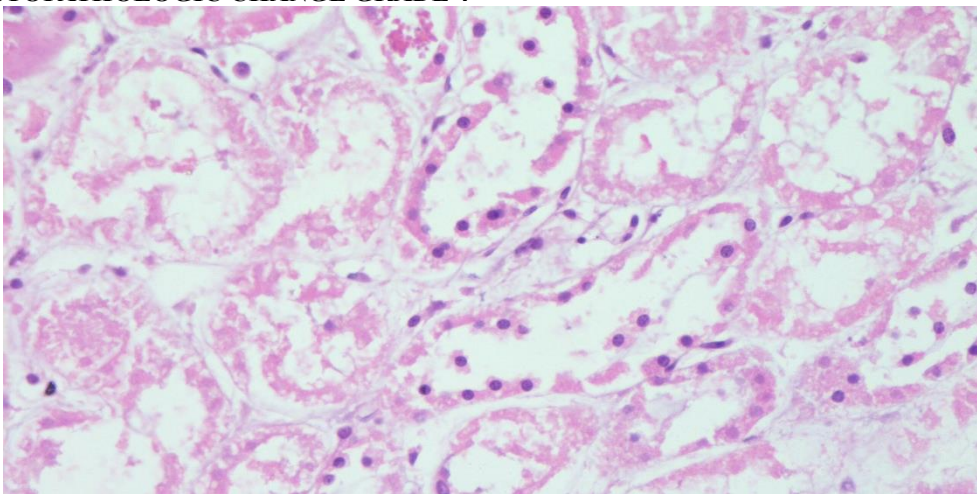


**Fig 6 : Showing necrotic PCT. (H & E Staining X 400)**

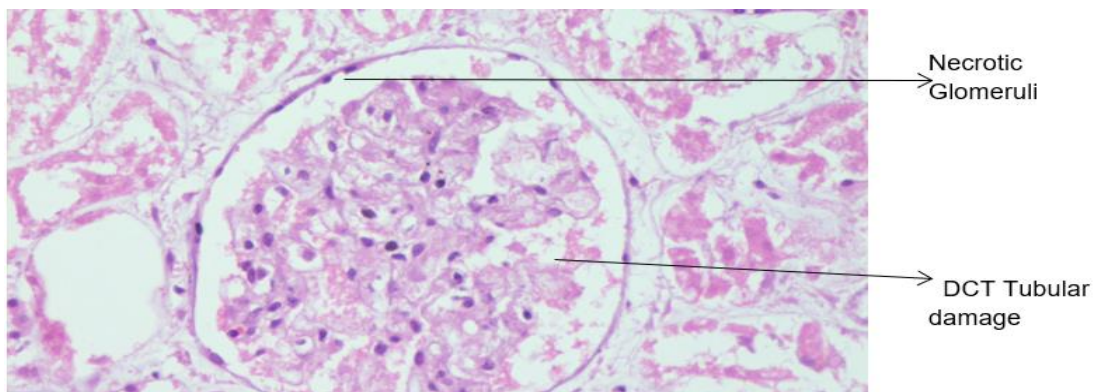


**Fig 7 : Show Disruption of tubular cells with loss of nuclei of PCT, focal loss of nuclei of DCT(H & E Staining X 400)**

**KIDNEY HISTOPATHOLOGIC CHANGE GRADE 4**



**Fig 8 : Show necrotic PCT, DCT tubular shedding .(H & E Staining X 400)\**



**Fig 9 : Show Necrotic glomerulus, DCT tubular shedding (H & E Staining X 400)**

**DISCUSSION**

One of the most important objectives of post mortem examination is to estimate time since death. The time of death is documented in death certificate in hospital deaths, but in non-hospital deaths, it is the responsibility of forensic pathologist to establish time of death correctly, because sometime relatives and others may mislead the investigating officials due to their malicious intention.

The present study was conducted department of Forensic medicine and department of Pathology of University College of Medical Sciences and Guru Teg Bahadur Hospital, Delhi. A total of 150 cases of different age and sex were taken. Out of 150 cases 114 were males and 36 females.

Only those cases where time of death was known and verified by hospital records were taken for study. All dead bodies were kept in morgue at 20-40°C temperature before postmortem examination.

The autopsy was done by standard procedure kidney was removed by Rokitsansky method. All the histopathological changes that take place in kidney tissues at different postmortem interval were noted and classified according to the grades (15).

At post mortem interval 9 to <15 hours in present study there was partial loss of cells of proximal convoluted tubule while Vinita Kushwaha et al found disruption of epithelium at 12 hours.

At post mortem interval 15 to <24 hours in present study there was disruption of proximal convoluted tubule cell disruption with focal loss of nuclei.

At post mortem interval 21 to <27 hours in present study there was loss of nuclei of proximal convoluted tubule was seen while there was focal loss of nuclei in distal convoluted tubule.

At post mortem interval 27 to <33 hours in present study glomeruli were unremarkable while distal convoluted tubule show shedding of cells.

At post mortem interval 33 to 39 hours in present study necrotic glomeruli, necrotic proximal convoluted tubule and shedding of distal convoluted tubule was seen. Vinita Kushwaha et al found collapse of glomeruli from 19 to 24 hours.

In present study the histopathological change of Grade 1 was found at post mortem interval 15 to <21 hours while Vinita Kushwaha et al found Grade 1 up to 31-34 hours. Histopathological change of Grade 2 was observed in present study at post mortem interval 27 to <33 hours while Vinita Kushwaha et al found it from 13 to 18 hours to 31 to 34 hours. Histopathological change of Grade 3 was found in present study in post mortem interval 9 to 33 hours while it was found from post mortem interval 13 to 24 hours by Vinita Kushwaha et al. Histopathological change of Grade 4 was found in present study from 21 to 39 hours while Grade 4 was not found in any post mortem interval by Vinita Kushwaha et al.

The difference of observations in present study from other studies may be because of inter observer variations. Some of the observations like sinus dilation, ballooning changes does not have any measuring criteria hence prone for subjective variation. Differences in atmospheric conditions like temperature and humidity also accounted for variations. Ante mortem factors like duration of stay in hospital also affect the observations because similar changes occur in diseased condition like hypovolemic and acute tubular necrosis.

## CONCLUSION

Histopathology of the Kidney provides a reliable estimation of Post-mortem interval. The present study does not show Grade 0 in any dead body because, in the present study, all the dead bodies sampled were those whose time since death was at least 3 hours. Grade 0 signifies no change in histopathology examination. It does not exclude that some histopathology changes started immediately after death or within 3 hours of death. Degenerative changes showed a positive correlation with time since death. The present study shows no gender-related changes.

The present study demonstrates that histopathological changes in the kidney show a progressive and statistically significant correlation with postmortem interval (PMI). Degenerative alterations such as tubular epithelial disruption, loss of nuclei, glomerular collapse, and eventual necrosis increased consistently with time since death, with higher grades (G3 and G4) predominating at longer PMIs ( $\geq 27$  hours). The absence of Grade 0 changes even at the earliest interval ( $>3$  hours) suggests that microscopic autolytic processes in renal tissue begin very early after death, supporting the sensitivity of kidney histology as an indicator of PMI.

Quantitative analysis further reinforces this relationship, with mean PMI increasing significantly across histological grades ( $p = 0.001$ ), indicating that renal histopathology can serve as a reliable adjunct tool in estimating time since death, particularly when used alongside conventional methods.

However, the study also highlights that mechanical trauma introduces important confounding effects. Ante-mortem factors such as hypovolemic shock, ischemia, and systemic hypoxia—commonly seen in blunt, sharp, and crush injuries—can produce renal changes resembling postmortem autolysis, including tubular necrosis and nuclear loss. This overlap may lead to overestimation of PMI if trauma-related alterations are not carefully distinguished from true postmortem changes.

## Limitations

### The findings must be interpreted in light of certain limitations:

- The study did not stratify cases based on type or severity of mechanical trauma, limiting precise evaluation of trauma-specific effects
- Environmental variables such as temperature and humidity, known to influence autolysis, were not standardized

- Histological grading was semi-quantitative and observer-dependent, introducing potential subjectivity
- Lack of biochemical or molecular correlation (e.g., enzyme degradation, proteomics)
- Possible influence of ante-mortem clinical conditions such as shock or hospital stay on renal morphology

### Recommendations

While kidney histopathology is a valuable and scientifically valid tool for PMI estimation, it should not be used in isolation. A multidisciplinary approach integrating histology, clinical history, environmental factors, and injury profile is essential for accurate interpretation, especially in cases involving mechanical trauma. For a more precise post-mortem interval estimation, further studies should use many dead bodies and parametric values for cellular changes.

### Conflict of interest: None

The ethical committee of UCMS gave clearance for the study.

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