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## Original Research Paper

# Determination of Time since Death from Changes in Morphology of White Blood Cells in Ranchi, Jharkhand

[1]Binay Kumar, [2]Tulsi Mahto, [3]Vinita Kumari, [4]Aman Kumar

## Abstract

Determination of 'time elapsed since death' (TSD) is one of the important content of the post-mortem report. Irreversible changes occur in the WBCs in the internal environment due to non-availability of oxygen, accumulation of carbon dioxide, pH change and accumulation of toxic products. Although the changes in morphology of white blood cells are also variable, depending on different factors like other parameters used for the purpose of determination of time since death, but it is less variable as compared to others. The study sample comprised of 150 medico-legal autopsies conducted in the department of Forensic Medicine & Toxicology, Rajendra Institute of Medical Sciences, Ranchi, Jharkhand, during June 2006 to September 2007. Blood samples were collected from heart chambers and slides were prepared on spot at the time of autopsy. Slides were stained by Leishman's stain and examined under light microscope. In present study neutrophils were recognized up to 30 Hrs Lymphocytes up to 36 hrs, Eosinophils up to 20.30 hrs and monocytes up to 19.20 Hrs. In no case basophil was observed.

**Key Words:** WBC, Neutrophils, Lymphocyte, Eosinophils, Monocytes, Lysed, Time Since Death (TSD)

## Introduction:

Determination of 'time elapsed since death' (TSD) helps in the investigation of complex and mysterious cases to unearth the truth for the administration of justice in many ways. In general, determination of the time of death is extremely difficult, and accuracy is almost impossible.

Although by careful study of different macroscopic, microscopic, chemical and biological parameters, the TSD can be determined in considerably narrow range.

Irreversible changes occur in the WBCs in the internal environment due to non-availability of oxygen, accumulation of carbon dioxide, pH change and accumulation of toxic products. [1]

The changes in morphology of White blood cells (WBC) are also variable depending on different factors, like other parameters used for the purpose of determination of TSD but it is less variable as compared to others.

## Corresponding Author:

[1]Assistant Professor,
Department of Forensic Medicine & Toxicology,
All India Institute of Medical Sciences, Patna
E-mail: dr.binay_rimsranchi@yahoo.co.in
[2]Professor,
[3]Ex Junior Resident (Non-Academic)
[4]Assoc. Prof,
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## Materials and Methods:

The study sample comprised of 150 medico-legal cases for autopsies conducted in the Department of Forensic Medicine & Toxicology, Rajendra Institute of Medical Sciences, Ranchi, Jharkhand, during June 2006 to September 2007.

Only those cases in which TSD were known by relatives, police or doctors and verified by other post-mortem changes were included in this study. Cases in which bodies grossly affected with septicemia, anemia, and nutritional deficiency, Malignancy of blood, blood disorders, and charred bodies were not included in this study.

Blood samples were collected from heart chambers and slides were prepared on spot at the time of autopsy. Slides were stained by Leishman's stain and examined under light microscope. The study was based upon variation in White Blood Cells.

Morphology of all types of white blood cells (WBC) i.e. neutrophils; lymphocytes, Eosinophils and monocytes were noted in following manner:

**Normal, slightly dysmorphic, grossly dysmorphic, mixture of dysmorphic & Lysed and Lysed.**

For the purpose of classifying the observation systematically, the dead bodies were grouped in the following manner based on the known time elapsed since death:

**Table A: Groups of Dead Bodies**

| Group | Time elapsed since death |
|---|---|
| I | 0—06Hrs |
| II | 06—12 Hrs |
| III | 12—18 Hrs |
| IV | 18—24 Hrs |
| V | 24—36 Hrs |
| VI | 36—48 Hrs |
| VII | >48 Hrs |

## Observations:

**1. Neutrophils:**

In our study among the cases examined during the first 6 hours after death in 91% cases morphology of neutrophils were found to be normal and in 9% cases they were slightly dysmorphic. In 6 to 12 hours after death they were normal in 60.7% and slightly dysmorphic in 39.2% cases.

Where as in 12 to 18 hours after death they were normal in 12.55% cases, slightly dysmorphic in 25% and were grossly dysmorphic in 62.5% cases. (Table 1)

In between 18 to 24 hours neutrophils were slightly dysmorphic in 2.9% cases, grossly dysmorphic in 79.5% cases, mixture of dysmorphic and lysed in 14.7% and were completely lysed in 2.9% cases.

They were found to be grossly dysmorphic in 22.2% cases, mixture of dysmorphic and lysed in 22.2% and in 55.6% cases found to be completely lysed in 24 to 36 Hrs. In all cases examined after 36 hours of death neutrophils were found to be lysed.

Neutrophils were found to be recognizable latest by 30 hours in present study.

**2. Lymphocytes:**

Out of all the cases examined during the first 6 hours after death lymphocytes were found to be normal in 95.5% of cases and cells were slightly dysmorphic in 4.5% where as they were normal in 82.1% cases and slightly dysmorphic in 17.9% cases during 6 hours to 12 hours after death. In 12 to 18 hours after death cells were normal in 35% cases, slightly dysmorphic in 20% cases and grossly dysmorphic in 45%.

Whereas lymphocytes were normal in 2.9% cases, slightly dysmorphic in 5.9% cases and grossly dysmorphic in 91.2% cases during 18 to 24 hours of death.

Out of cases examined during 24 to 36 hrs after death neutrophils were found normal in 11.1% cases, dysmorphic in 22.2% cases, mixture of dysmorphic and lysed in 22.2% and were lysed in 44.5% cases.

They were found lysed in all the cases examined beyond 36 hours after death. (Table 2) Lymphocytes were found recognizable latest by 36 hours after death in present study.

**3. Eosinophils:**

During first 6 hours after death the eosinophils were found to be normal in 50% cases, slightly dysmorphic in 45.45% cases, grossly dysmorphic in 4.5% cases where as the cells were found normal in 3.6% cases, slightly dysmorphic in 28.6 % cases grossly dysmorphic in 60.7% cases and were lysed in 7.1% cases in 06-12 Hrs. (Table 3)

Present study showed that in between 12-18 hrs after death the eosinophils were found dysmorphic in 45%, mixture of dysmorphic and lysed in 5.1% cases and were completely lysed in 50% of the cases where as they were found to be dysmorphic in 5.9% of the cases, mixture of dysmorphic in 2.9% and were found lysed in 91.2% of case examined during the period of 18 to 24 hours.

In all cases eosinophils examined beyond 24 hours after death were found to be lysed. Eosinophils were found to be recognizable in latest by 20.30 hours after death in present series of cases.

**4. Monocytes:**

In this study out of the total cases examined, in first 6 hours after death in 45.5% cases morphology of the monocytes were found to be normal, in 50.1% cases they were recognizable but slightly dysmorphic, and in 4.5% cases lysed. Where as they were normal in 7.1% case, slightly dysmorphic in 28.6%, and grossly dysmorphic in 64.3% of cases examined during 6-12 hours after death. (Table 4)

Among the cases examined during the 12-18 hrs after death the monocytes were found to be slightly dysmorphic in 5%, grossly dysmorphic in 37.5%, mixture of dysmorphic and lysed in 12 .5% and lysed in 45% cases.

They were found dysmorphic in 2.9% cases and lysed in 97.1% of cases examined during 18 to 24 hours after death. In all cases examined beyond 24 hours after death the monocytes were found to be absent. In the present study monocytes were found to be recognizable latest up to 19.20 hours.

## Discussion:

Rajesh Bardale in his study observed that neutrophils up to 20-24 hrs, lymphocytes up to 30 hrs eosinophils up to 21 hrs and monocytes are identifiable up to 18 Hrs after death. [2] Penttila A, Lahio K. stated that when corpses were kept at +4°C the lymphocytes seemed to be most resistant and basophils the least resistant to the effects of autolysis. [3]

H Dokgoz et al found that eosinophils and monocytes were identifiable up to 72 hrs, neutrophils up to 96 hrs and lymphocytes up to

120 Hrs after death in non-refrigerated cadavers. [4] Rajesh Bardale had not found morphology of any cell identifiable beyond 30 hrs contrary to other studies. [1, 2, 4]

The reason might be that degenerative cellular changes occur earlier and more rapidly in cadaveric blood than in vitro blood of controls [1, 2- 5] or might be attributable to environmental and temperature difference. [2, 3]

Platt et al in their study of cerebrospinal fluid cells found that if the post-mortem duration is greater than 12 hour, the cells become vacuolated and cannot be identified. [6]

Wyler D et al found that the post-mortem cell count in cerebrospinal fluid correlates to the time after death and can be described mathematically (Polynomial curve of third order). [7]

## Conclusion:

The present study proves that changes in the morphology of white blood cells can be helpful as supplementary procedure for estimating time since death. It is also a very simple procedure and interpretation of above mentioned findings is easy.

## References:


1. **Babapulle CJ, Jayasundera NPK .**Cellular changes and time since death. Med Sci Law 1993; 33:213-22.
2. **Rajesh Bardale, Dixit P.G.** Evaluation of Morphological Changes in Blood Cells of Human cadaver for the estimation of Postmortem interval. Medico-Legal Update Vol. 7 No. 2 (2007-04-2007-06).
3. **Penttila A, Laiho K.** Autolytic changes in blood cells of human cadavers. II. Morphological studies. Forensic Sci Int. 1981 Mar-Apr; 17(2):121-32.
4. **H Dokgoz et al.** Comparison of morphological changes in white blood cells after death and in vitro storage of blood for the estimation of postmortem interval. For Sci Int.124 (2001) 25-31.
5. **Lahio K, Penttila A.**Autolytic changes in blood cells and other tissue cells of human cadavers I. Viability and ion studies. Forensic Sci Int 1981; 17:109-20.
6. **Platt M S et al.** Postmortem cerebrospinal fluid pleocytosis. Am J Forensic Med Pathol 1989; 10: 209-12?
7. **Wyler D, Marty W, Bar W.** Correlation between the post-mortem cell content of cerebrospinal fluid and time of death. Int J Legal Med. 1994; 106(4):194-9.


**Table 5: Change in Morphology of WBC V/S TSD**

| Time since death | Changes in Morphology of White Blood Cells |
|---|---|
| 0—06 hrs | Neutrophils - normal |
| | Lymphocyte - normal |
| | Eosinophils - normal |
| | Monocytes - normal |
| 06—12 Hrs | Neutrophils - Slightly dysmorphic |
| | Lymphocyte - Slightly dysmorphic |
| | Eosinophils - grossly dysmorphic |
| | Monocytes - grossly dysmorphic |
| 12—18 Hrs | Neutrophils - Grossly dysmorphic |
| | Lymphocyte- Slightly and Grossly dysmorphic mixed. |
| | Eosinophils - Lysed |
| | Monocytes - Lysed |
| 18—24 Hrs | Neutrophils - Grossly dysmorphic |
| | Lymphocyte - Grossly dysmorphic |
| | Eosinophils - Lysed |
| | Monocytes - Lysed |
| 24—36 Hrs | Neutrophils - mixture of lysed and dysmorphic cells |
| | Lymphocyte - Lysed |
| | Eosinophils - All Lysed |
| | Monocytes - All Lysed |
| 36-48 Hrs | Neutrophils - All Lysed |
| | Lymphocyte - Lysed |
| | Eosinophils - All Lysed |
| | Monocytes - All Lysed |
| > 48 Hrs | In case of lymphocyte also all Lysed |

**Table1**

**Morphological Changes in Neutrophils in Different Time Intervals**

| TSD (Hrs.) | Normal | Recognizable but slightly dysmorphic | Recognizable but grossly dysmorphic | Mixture of dysmorphic and Lysed | Lysed | Total |
|---|---|---|---|---|---|---|
| 0—6 | 20 (91%) | 2 (9%) | 0 | 0 | 0 | 22 |
| 6—12 | 17 (60.7%) | 11 (39.2%) | 0 | 0 | 0 | 28 |
| 12—18 | 5 (12.5%) | 10 (25%) | 25 (62.5%) | 0 | 0 | 40 |
| 18—24 | 0 | 1 (2.9%) | 27 (79.5%) | 5 (14.7%) | 1 (2.9%) | 34 |
| 24—36 | 0 | 0 | 2 (22.2%) | 2 (22.2%) | 5 (55.6%) | 9 |
| >36 | 0 | 0 | 0 | 0 | 17 (100%) | 17 |

**Table 2**

**Morphological Changes in Lymphocytes in Different Post- mortem Interval**

| TSD (Hrs) | Normal | Recognizable but slightly dysmorphic | Grossly dysmorphic | Mixture of dysmorphic and Lysed | Lysed | Total |
|---|---|---|---|---|---|---|
| 0—6 | 21 (95.5%) | 1 (4.5%) | 0 | 0 | 0 | 22 |
| 6—12 | 23 (82.1%) | 5 (17.9%) | 0 | 0 | 0 | 28 |
| 12—18 | 14 (35%) | 8 (20%) | 18 (45%) | 0 | 0 | 40 |
| 18—24 | 1 (2.9%) | 2 (5.9%) | 31 (91.2%) | 0 | 0 | 34 |
| 24—36 | 1 (11.1%) | 0 | 2 (22.2%) | 2 (22.2%) | 4 (44.5%) | 9 |
| >36 | 0 | 0 | 0 | 0 | 17 | 17 |

**Table 3**
**Morphological Changes in Eosinophils in Different Post-mortem Interval**

| TSD (Hrs) | Normal | Recognizable but slightly dysmorphic | Grossly dysmorphic | Mixture of dysmorphic and Lysed | Lysed | Total |
|---|---|---|---|---|---|---|
| 0—6 | 11 (50%) | 10(45.45%) | 1(4.5%) | 0 | | 22 |
| 6—12 | 1 (3.6%) | 8 (28.6%) | 17 (60.7%) | 0 | 2 (7%) | 28 |
| 12—18 | 0 | 0 | 18 (45%) | 2(5%) | 20 (50%) | 40 |
| 18—24 | 0 | 0 | 2 (5.9%) | 1 (2.9%) | 31(91.2%) | 34 |
| >24 | 0 | 0 | 0 | 0 | 26 (100%) | 26 |

**Table 4**
**Morphological Changes in Monocytes in Different Post-mortem Interval**

| TSD (Hrs) | Normal | Recognizable but slightly Dysmorphic | Grossly dysmorphic | Mixture of dysmorphic and Lysed | Lysed | Total |
|---|---|---|---|---|---|---|
| 0—6 | 11 (50%) | 11 (50%) | 0 | 0 | 0 | 22 |
| 6—12 | 2 (7.1%) | 8 (28.6%) | 18 (64.3%) | 0 | 0 | 28 |
| 12—18 | 0 | 2 (5%) | 15 (37.5%) | 5 (12.5%) | 18 (45%) | 40 |
| 18—24 | 0 | 0 | 1 (2.9%) | 0 | 33 (97.1%) | 34 |
| >24 | 0 | 0 | 0 | 0 | 26 (100%) | 26 |