

Original Research Paper

Determination of Time Elapsed since Death from Changes in Morphology of Red blood cells in Ranchi, Jharkhand

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Abstract

Determination of 'time elapsed since death' (TSD) is one of the important content of the post-mortem report. Although the changes in morphology of red blood cells are 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. In present series of studies in majority of cases the shape of RBCs became dimorphic, and peripheral area became pale in 12 to 24 hrs after death. In 18 to 36 Hrs normal central pallor lost and 24 to 48 Hrs majority of RBC lysed.

Key Words: RBC, Dysmorphic, Central pallor, Periphery pale, Time Elapsed 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, determining 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 RBCs 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 red blood cells (RBC) 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.

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.

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Only those cases whose TSD were known by relatives, police or doctors and verified by other post-mortem changes, were 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:

1. **Red Blood Cells:** Morphology of red blood cells (RBC) was noted in following manner:
 - a. **Integrity:** Intact, Mixture of intact & lysed, and Lysed & not recognizable
 - b. **Shape:** Intact, Slightly dysmorphic and grossly Dysmorphic
 - c. **Central Pallor:** Intact, Reduced and Lost
 - d. **Periphery:** Red and Pale

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:

Group	Time elapsed since death
I	0—03Hrs
II	06—12 Hrs
III	12—18 Hrs
IV	18—24 Hrs
V	24—36 Hrs
VI	36—48 Hrs
VII	>48 Hrs

Inclusion Criteria:

- When TSD was known by the relatives, police or doctors
- When it is verified by other post-mortem changes

Exclusion Criteria:

- a. Body was grossly affected with septicemia.
- b. Anaemia

- c. Nutritional deficiency
- d. Malignancy of blood
- e. Blood disorder.
- f. Charred body

Observations:

a. Integrity

In all cases examined up to 12 hours after death the RBCs were found to be intact, where as in between 12 hours to 18 hours after death in 94.7% cases cells were intact and in 5.3 % cases the mixture of intact and lysed RBCs was found in the slides.

In the cases examined in between 18 hours to 24 hours after death in 58.8% cases cells were intact whereas in 41.2% cases the mixture of intact and lysed RBCs was found.

Among the cases examined between 24 hours to 36 hours after death in 55.6% slides mixture of intact and lysed RBCs and in 33.3 % slides all cells were found lysed & unrecognizable.

In between 36 hours to 48 hours after death in 16.7% cases mixture of intact and lysed RBCs was present where as in 83.3% RBCs were completely lysed & unrecognizable. No RBCs was found intact after 48 hours of death.

b. Shape

Out of three cases examined in first 3 hours of death the shape of the RBCs was found to be normal in all cases.

Among the cases examined during 3 hours to 6 hours after death RBCs were normal in 38.9% cases slightly dysmorphic in 61.1% cases where as in the cases examined in between 6 hours to 12 hours after death RBCs were found normal in 7.1% cases, slightly dysmorphic in 60.7% cases and grossly dysmorphic in 32.2% cases.

In between 12 hours to 18 hours after death in 12.5% cases RBCs were slightly dysmorphic and in 87.5 % cases it was found grossly dysmorphic where as it was found to be grossly dysmorphic in all cases examine during 18 to 24 hours after death.

In the cases examined during 24 hours to 36 hours after death RBCs were found to be grossly dysmorphic in 55.6% cases, mixture of dysmorphic and lysed in 11.1% cases and they were completely lysed & unrecognizable in 33.3% cases.

Among the cases examined between 36 hours to 48 hours after death mixture of dysmorphic and lysed RBCs were found in 16.7 % slides and it was completely lysed in 83.3% slides. Beyond this period cells were lysed in all the slides.

c. Central pallor

Within 6 hours after death central pallor of RBCs were normal in 81.8% cases, reduced in 18.2% cases.

In between 6 hours to 12 hours after death they were normal in 14.3% cases, reduced in 82.1% cases and lost in 3.6% cases where as in between 12 hours to 18 hours after death central pallor was reduced in 62.5% cases and lost in 37.5% cases.

The central pallor was reduced in 20.6% cases and lost in 79.4% cases examine during the 18 hours to 24 hours after death where as in between 24 to 36 hours after death it was reduced in 11.1% cases, lost in 55.6% cases and cells were found lysed in 33.3% cases.

Among the cases examined in between 36 to 48 hours central pallor was lost in 16.7% and cell were found lysed in 83.3%.

In all the cases examined beyond this period cells were found lysed.

d. Periphery of the cell

In the cases examined during first 6 hours after death periphery of the RBCs were found normally red in 90.9% case.

Among the cases examined during 6 hours to 12 hours after death the periphery of RBCs were found to be red in 64.3% and pale in 35.7% cases where as it was found red in 7.5% cases and pale in 92.5% cases during 12 to 18 hours after death.

In all slides examined in between 18 hours to 24 hours after death periphery of the cell was found to be pale.

Periphery of the RBCs were found to be pale in 66.7% and cells were lysed in 33.3% cases among the cases examined during 24 hours to 36 hours after death where as during 36 hours to 48 hours after death it was pale in 16.7% and cells were found to be lysed in 83.3% cases.

Discussion:

Rajesh Bardale observed RBC identifiable up to 18 Hrs postmortem. [2]

Penttila A, Lahio K states that when corpses were kept at +4°C the red cells were quite rapidly transformed from discoid configuration to crumbled discs, echinocytes and spherocytes, but no debris or burst cell configurations were seen. [3]

Besides the red blood cells many workers has worked on other different type of cells. H Dokgoz et all found that eosinophils and monocyte 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 states that he 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, 4, 5] or might be attributable to environmental and temperature difference. [2, 3]

Plat et al studied cerebrospinal fluid cells where they found that if the postmortem duration is greater than 12 hour, the cells become vacuolated and cannot be identified. [6]

Wylar D et al states 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:

From present study the changes in the morphology of red blood cells can be concluded. (Table A) It may prove 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:

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Table A: Change in Morphology of RBC

Time since death	Changes in morphology of red blood cells
0—03 hrs	Integrity - intact
	Shape - normal
	Central pallor - intact
	Periphery - red
03—06 Hrs	Integrity - intact
	Shape - slightly dysmorphic
	Central pallor - intact
	Periphery - red
06—12 Hrs	Integrity - intact
	Shape - slightly dysmorphic
	Central pallor - reduced
	Periphery - red
12—18 Hrs	Integrity - intact
	Shape - grossly dysmorphic
	Central pallor - reduced
	Periphery - red
18—24 Hrs	Integrity - intact
	Shape - grossly dysmorphic
	Central pallor - lost
	Periphery - pale
24—36 Hrs	Integrity - mixture of lysed and intact RBCs
	Shape - grossly dysmorphic
	Central pallor - lost
	Periphery - pale
36—48 Hrs	Completely lysed and not recognizable in majority of cases.
> 48 Hrs	Completely lysed and not recognizable in all cases.

**Table 1
Integrity of RBCs in Different Time Intervals**

TSD (Hrs.)	RBC intact	RBC intact and lysed mixed	RBC lysed & Not recognizable	Total
0—6	22 (100%)	0	0	22
6—12	28 (100%)	0	0	28
12—18	38 (94.7%)	2 (5.3%)	0	40
18—24	20 (58.8%)	14 (41.2%)	0	34
24—36	1 (11.1%)	5 (55.6%)	3 (33.3%)	9
36—48	0	1 (16.7%)	5 (83.3%)	6
>48	0	0	11 (100%)	11
Total				150

**Table 2
Shape of RBCs in Different Post-mortem Interval**

TSD (Hrs.)	RBCs normal	RBCs slightly dysmorphic	RBCs grossly dysmorphic	RBCs mixture of dysmorphic and lysed	RBCs lysed & unrecognizable	Total
0—3	3 (100%)	0	0	0	0	3
3—6	7 (38.9%)	11 (61.1%)	0	0	0	19
6—12	2 (7.1)	17 (60.7%)	9 (32.2%)	0	0	28
12—18	0	5 (12.5%)	35 (87.5%)	0	0	40
18—24	0	0	34 (100%)	0	0	34
24—36	0	0	5 (55.6%)	1 (11.1%)	3 (33.3%)	9
36—48	0	0	0	1 (16.7%)	5 (83.3%)	6
>48	0	0	0	0	11 (100%)	11
Total						150

Table 3
Central pallor of RBCs in Different Post-mortem Interval

TSD (Hrs.)	Central pallor normal	Central pallor reduced	Central pallor lost	Cells lysed	Total
0—6	18 (81.8%)	4 (18.2%)	0		22
6—12	4 (14.3%)	23 (82.1%)	1 (3.6%)	0	28
12—18	0	25 (62.5%)	15 (37.5%)	0	40
18—24	0	7(20.6%)	27 (79.4%)	0	34
24—36	0	1 (11.1%)	5 (55.6%)	3 (33.3%)	9
36—48	0	0	1 (16.7%)	5 (83.3%)	6
>48	0	0	0	11 (100%)	11
Total					150

Table 4
Periphery RBCs in Different Post-mortem Interval

TSD (Hrs.)	Periphery red	Periphery pale	Cells lysed	Total
0—6	20 (90.9%)	2 (9.1%)	0	22
6—12	18 (64.3%)	10 (35.7%)	0	28
12—18	3 (7.5%)	37 (92.5%)	0	40
18—24	0	34 (100%)	0	34
24—36	0	6 (66.7%)	3 (33.3%)	9
36—48	0	1 (16.7%)	5 (83.3%)	6
>48	0	0	11 (100%)	11
Total				150