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Original Research Article

Histopathological Features of Abrasions- An Autopsy Study

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Key words

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Abstract

Background: The extent of skin injury can range from an abrasion to more serious situations like laceration. Because of the uncertain and variable results of the naked eye examination of the wound it is important to study these injuries microscopically. **Materials and Methods:** A prospective 2 years study was carried out consisting of 104 cases with abrasions which were examined from the deceased along with control skin samples at the mortuary of the Medical College and Hospital run by Municipal Corporation of Greater Mumbai, Maharashtra. **Results:** Out of 104 cases, railway accident cases (n=52, 50%) were the highest. Males (n=93) were affected more than females (n=11). Younger age group was affected more (21-30 years and 30-40 years age group). The average duration for the formation of reddish brown scab is within a range of 6-48 hours. Neutrophilic infiltration increases from 1 hour and reaches peak (n=30, 28.84 %) in 24-30 hours. Mononuclear cells infiltration reaches peak (n= 17, 16.34 %) at 36-72 hours. Twelve cases out of 104 showed presence of mast cells and majority of the cases (n=6) were in 5 days duration group. Fibrosis was seen in 21 cases and majority of the cases (n=15) were seen in 5 days duration group. **Conclusion:** Even though the gross appearance provides an approximate age of wound, histopathology remains the mainstay for the definitive diagnosis. Hence, the importance of microscopic examination of wounds cannot be over emphasized and is very helpful in the administration of medico legal cases.

1. Introduction

The history of trauma parallels the history of the evolution of man. With his aggressive instincts, creative ability and endless ambition to conquer the environment without any regards to the price he has to pay to achieve his goals.

The term trauma refers to an injury inflicted by force on a living tissue. It also applies to emotional and mental stress that results into a disorganized psychic or behavioural state.^[1] Literally, the word trauma means a deeply

distressing or disturbing experience, emotional shock following a stressful event or a physical injury, which may be associated with physical shock, and sometimes, leads to long-term neurosis. The origin of the word trauma is from the Greek word, which means 'wound'. The word 'injury' means an instance of being injured, or the fact of being injured, or harmed or damage.¹ Indian Penal Code (IPC) (Section 44) has defined injury as any harm, whatever illegally caused to any person

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in body, mind, reputation or property. Section no 319 of IPC states that a person is guilty of causing 'hurt' whoever causes bodily pain, disease or any infirmity to any person. Hence, hurt or injury can be caused without causing any wound but wound will invariably cause hurt and injury.

The forensic aspect of trauma deals with an accurate description of injury, assessment of its severity and its interpretation in relation to the law in force at the time.¹ It is a challenging task for investigating agency to reveal the mystery and for judiciary to award a deterrent sentence to the guilty. The circumstances become more complex when such exercise also involves socio-economic factors in addition to medico legal components of the case.² In a case of death by physical violence there is always one major point to be determined by the forensic pathologist: whether the injuries were inflicted before death (ante mortem) or after it (post mortem).³ For the reconstruction of events, it is important to time the wound which means to estimate the interval between trauma and death.³

Like any other body part, skin is also affected by injury. The extent of injury can range from an abrasion to more serious situations like laceration. Loss of integrity of large portion of the skin may lead to major disability or even death. Because of the uncertain and variable results of the naked eye examination of the wound it is important to study these injuries microscopically. In forensic practice, it is necessary to evaluate the causal relationship between death and any wound.⁴ In this study we have attempted to examine the histopathology of the abrasions at different time intervals with the help of special stains.

2. Materials and methods

This two years prospective study was carried out on the dead bodies with well demarcated injury and were received for medico legal autopsy at the mortuary of the Medical College and Hospital run by Municipal Corporation of Greater Mumbai, Maharashtra.

Inclusion Criteria: It includes the cases in which the time of infliction of the injury is known.

Exclusion Criteria:

1. Cases where the injuries are infected.
2. Cases suffering from other associated debilitating diseases and chronic illness like diabetes, peripheral vascular diseases, and septicemia.
3. Decomposed bodies.

A total of 104 cases with abrasions were examined from the deceased along with control skin samples. The history regarding causation of wounds and survival period was taken from the police, the relatives and the hospital records. For the study, after recording all general particulars of the deceased, gross examination of the abrasion was done and photograph of that abrasion was taken for record. Skin sample of size 2cm X 1cm X 0.5 cm from the site of abrasion and a sample of normal skin from the site of central incision made during the medico legal autopsy was taken and preserved in 10% formalin for histopathological examination. The time limit for medico legal autopsies varies with circumstances related with the case and it has been performed at the earliest after obtaining required documents (i.e. inquest report, statement of relatives etc.) from the investigating officer of the case. The abrasions were grouped into three different time intervals:

1. Injuries up to 2 days of survival: they are further divided into interval of 6 hours, from 0 to 48 hours.
2. Injuries of duration 2-5 days of survival: they are further divided into time interval of 24 hour.
3. Injuries of duration more than 5 days of survival.

The abrasions were categorized into the above mentioned groups with the help of a colorimetric scale NNDV (Nuzzolese, Neri, Di Vella).

Fixation and staining: The tissues were kept for 24 hrs in the 10 % neutral buffered formalin for fixation. The paraffin blocks were made and 5 micron thick sections were cut by microtome as per the standard technique. They were then stained with Haematoxylin and Eosin, Van Gieson's connective tissue stain, Toluidine blue stain techniques. The control sections were also subjected to the same procedure and techniques.

3. Results

In the present study the maximum number of cases were observed in the survival period of 0-6 hrs (n=60, 57.69%) followed by the survival period of > 5 days (n=17, 16.34 %). The minimum number of cases were observed in the survival period of two to three days (n=3, 2.88 %) and two cases (1.92 %) in 24-30 hrs. Out of 104 cases, railway accident cases (n=52, 50%) were highest followed by road traffic accident cases (RTA, n=34, 32.69 %). Fall from the height cases were 13 (12.5%). Hanging and building collapse cases were 2 (1.92%) while one case was of physical assault (0.96%). **Table no 1 and 2** give a detailed distribution of cases in relation with different survival time and

cause wise distribution of abrasions respectively. It was observed that the number of cases involving the males (n=93, 89.42%) were more than females (n=11, 10.57%) and the maximum number of cases were observed in the age group of 21-30 years (n=37, 35.57%) followed by the age group of 31-40 years (n=24, 23.07%). The minimum number of cases were observed in the age group of less than ten years (n=2, 1.92%). **Table no 3** describe age group wise distribution of cases.

Table no 1: Distribution of total cases according to the survival time.

Sr.no	Survival Time	Total	Percentage (%)
1	0-6 hrs	60	57.69
2	6-12 hrs	7	6.73
3	12-18 hrs	1	0.96
4	24-30 hrs	2	1.92
5	30-36 hrs	1	0.96
6	36-42 hrs	1	0.96
7	42-48 hrs	1	0.96
8	2-3 days	3	2.88
9	3-4 days	5	4.8
10	4-5 days	6	5.76
11	>5 days	17	16.34
Total		104	100

Table no 2: Cause wise distribution of cases.

Sr. No	Cause of Abrasion	No of Cases	Percentage (%)
1	Railway accident	52	50
2	Road traffic accident	34	32.69
3	Fall from height	13	12.5
4	Physical assault	1	0.96
5	Hanging	2	1.92
6	Building collapse	2	1.92
Total		104	100

Table no 3: Showing age wise distribution of cases.

Age group (years)	Number of cases	Percentage (%)
<10	2	1.92
11-20	7	6.73
21-30	37	35.57
31-40	24	23.07
41-50	14	13.46
51-60	12	11.53
61-70	4	3.84
>71	4	3.84
Total	104	100

Out of 104 cases, 60 cases which were in 0-6 hrs duration, of which abrasions in 53 cases (88.34%) were reddish in colour with bleeding and oozing and in seven cases (11.66 %) they showed reddish brown scab with dried blood. Seven out of 104 cases (6.73%) which were observed in 6-12 hrs duration group, abrasions in four cases (57.14%) were reddish in colour with oozing and slight bleeding, while in three cases (42.86%) they showed reddish brown scab with

dried blood. One case (0.96%) was observed in 12-18 hrs duration group and abrasion in that case (100%) showed reddish brown scab with dried blood.

Figure 1: A) Skin with mild inflammatory infiltrate with few mast cells (H&E 40x). B) and C) Toluidine blue highlighting the mast cells with magenta granules within cytoplasm (0-6 hours). D) Skin with dense fibrosis (H&E, 40x) E) EVG stain highlighting the fibrosis (beyond 5 to 7 days), inset showing fibrous band, brown in colour.

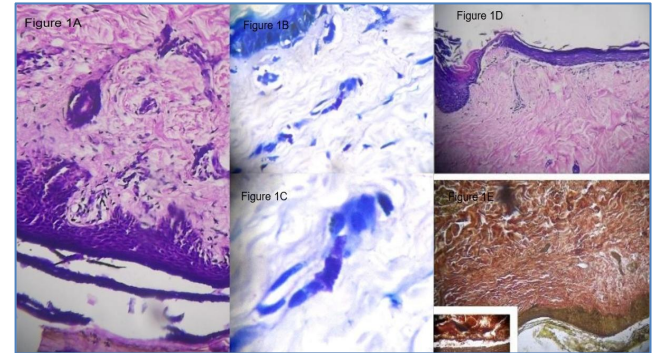
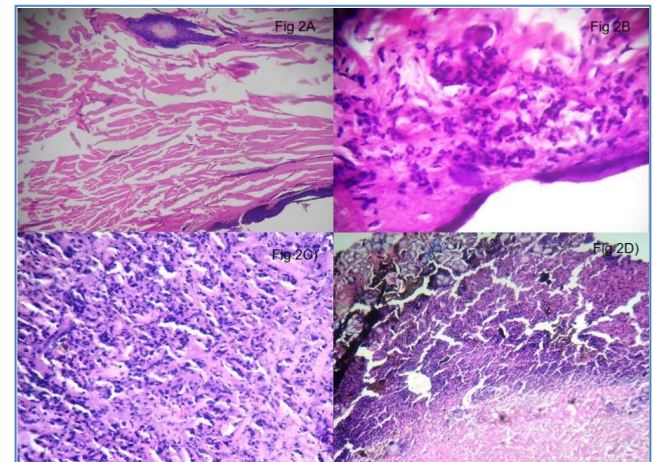


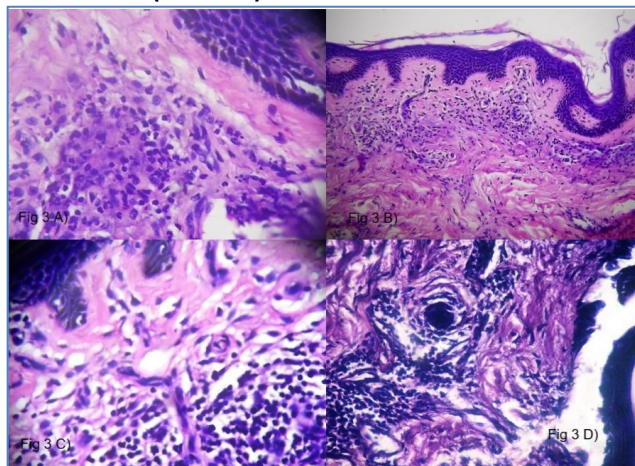
Figure 2: A) Skin with denuded epithelium with no inflammatory infiltrate (0-1 hour) H&E 40x. B) Skin with focal ulceration and underlying neutrophils (6-12 hours) H&E 40x. C) Moderate infiltration of neutrophils (12-24 hours) H&E 40x. D) Ulcerated skin with severe neutrophils admixed with extraneous material (24-30 hours) H&E 40x.



In 30-36 hrs, 36-42 hrs and 42-48 hrs duration group only one case (0.96%) was observed and abrasions in all these three groups (100%) showed reddish brown scab with dried blood. Out of three (2.88%) cases observed in 2-3 days duration group, abrasions in all the cases (100%) showed hard, brown scab. Out of five cases (4.8%) observed in 3-4 days duration group, abrasion in one case (20%) showed hard, brown scab, in two cases (40%) they showed hard brownish black scab and in two (40%) cases they showed partial shedding of the scab. Out of six cases (5.76%) observed in four to five days duration group, abrasion in one case (16.7%) showed hard, brown scab, in two cases (33.3%) they showed hard,

brownish black scab and in three cases (50%) they showed partial shedding of the scab. Out of 17 cases (16.34%) observed in more than five days duration group, abrasion in three cases (17.64%) showed hard black scab, in three cases (17.64%) they showed partial shedding of the scab, in four cases (23.52%) they showed complete shedding of the scab and in seven cases (41.17%) they showed depigmented skin.

Figure 3: A) Skin with underlying mixed inflammatory infiltrate (H&E 40x) B) Skin with mild mononuclear cell infiltrate (H&E 40x) C) Skin with moderate mononuclear cell infiltrate (H&E 40x) D) Skin with severe mononuclear cell infiltrate (H&E 40x)



Microscopically findings are shown in **Fig. 1-3**. Microscopically, hemorrhage was observed in 16 (15.38%) out of 104 cases. Out of 16 cases, 11 cases (10.57%) were observed in 0-6 hrs duration group, three cases (2.88%) were observed in 6-12 hrs duration group, one case each (0.96%) was observed in 42-48 hrs duration group and 4-5 days duration group. The presence of hemorrhage was probably due to bleeding and oozing in the abrasions within a range of 0-12 hrs of survival time period. Two cases (1.92%) which were observed beyond 42 hrs of survival time period also showed hemorrhage. Thus, the removal of scab could explain the presence of hemorrhage on microscopy.

In the present study, neutrophils were observed in 30 out of 60 cases (50%) in 0-6 hrs duration group, and thereafter in all the cases up to 5 days duration. The remaining 30 cases showed denudation of epithelium with no inflammation. The abrasions in these cases were in less than 60 minutes. The neutrophils were observed in all 17 cases (100%) of more than five days duration group. In the present study, mononuclear cells (MNC) were not observed in all 60 cases in 0-6 hrs duration group. They were

observed in all 7 cases (100%) in 6-12 hrs duration group, one case (100%) in 12-18 hrs group duration group and thereafter in all cases up to 5 days. They were observed in all 17 cases (100%) in more than 5 days duration group. Table no 7 gives the distribution of MNCs (mild, moderate and severe) according to the time interval.

Out of 104 cases, fibrosis was seen in 21 cases (20.19%). Out of these 21 cases, 15 cases (14.42%) were observed in more than five days duration group and three cases (2.88%) in 4-5 days duration groups. Two cases (1.92%) were observed in 3-4 days duration group and one case (0.96%) was observed in 2-3 days duration group. Mast cells were seen in 12 cases (11.53%). Six out of these 12 cases (5.76%), were observed in more than five days duration group. Two cases (1.92%) were observed in the survival period of 3-4 days while one case each (0.96%) was observed in survival period of 0-6 hrs, 6-12 hrs, 36-42 hrs and in 4-5 days.

In the present study, time range for the appearance of physical and microscopic inflammatory changes in abrasions were as follows. Bleeding was observed only during 0-6 hrs duration. Oozing was observed during 0-12 hrs and maximum abrasions showed oozing during 0-6 hrs. Dried blood was observed during 6-48 hrs, maximum abrasions showed dried blood during 12-18 hrs. Reddish colored abrasions with bleeding and oozing were observed from 0-12 hrs. Reddish brown scab with dried blood was observed from 6-48 hrs. Hard brown scab was observed from 2-5 days and hard brownish black scab was observed from 3-5 days. Hard black scab was observed from fifth day onwards. Partial shedding was observed from 3-4 days. Complete shedding was observed from 6-7 days onwards. Depigmented skin was observed from seventh day onwards.

Mild polymorphic infiltration was observed from 1-2 hours, which became severe in 24-30 hrs and then started decreasing from 30-36 hrs up to 48 hrs. Mononuclear cells infiltration appeared at 6-12 hrs. Moderate increase in mononuclear cells count was observed at 12-18 hrs. Significant increase in mononuclear cells count was observed at 36-48 hrs and remains as plateau in 2-3 days. They started decreasing from third day till fifth to seventh day. Collagen started appearing after 48 hrs onwards of infliction of injury and becomes dense beyond 5-7 days. Mast cell degranulation was observed at 0-6 hrs.

4. Discussion

In the present study, maximum number of cases were observed in 0-6 hours duration group (n=60, 57.69%). This is similar to a study conducted by Vinay et al in which 40 cases were in 0-12 hours duration.⁵ Railway accident cases were highest (n=52, 50%) followed by RTA, (n=34, 32.69%). This is because local trains are most commonly used by the commuter for transport in metropolitan cities. Also, increasing population, overcrowding, poorly maintained roads, breaking traffic rules and lack of awareness among the people add to this. Study conducted by Vinay et al, found that RTA constituted majority of cases (89 cases out of 101, 75.4%), 10 cases due to fall from height and one case each due to railway injury and assault.⁵

Number of cases involving the males were more than females. This observation is consistent with Sandhu SS et al and Vinay et al.^{5,6,7} Sandhu et al found that out of 150 cases, 129 were males (86%) and 21 were females (14%).⁶ Vinay et al found that 89 cases (88.1%) were males and 12 cases (11.9 %) were females.⁵ This is mainly due to Male: Female ratio in our country. In most of the families, males only are the source of income. Lack of education among females and more aggressive behaviour among males also contribute to this. The maximum number of cases were observed in the age group of 21-30 years (n=37, 35.57%) followed by the age group of 31-40 years (n=24, 23.07%). This observation is consistent with the study of Sandhu SS et al and Vinay et al.^{5,6} Sandhu SS et al found that maximum cases (n=62%, 41.34%) were in 21-30 years followed by 31-40 years (n=36, 24%) age group.⁶ Vinay et al found 51 cases in the age group of 21-40 years.⁵ The reason behind this is that the younger age group is more energetic, earning hands in most families and thus prone to injuries. Other factors include changing lifestyle, ambitious behaviour and extreme level of competition.

a. Gross:

In the present study, bleeding was observed in 0-6 hours and oozing in 0-12 hours. These observations are consistent with the findings in the textbook of Nandy A, Mukherjee JB, Rao N, Vij K, Reddy KSN, Sandhu SS et al, Sharma et al and not consistent with the study of Kumar L et al.^{6, 8-14} In the present study, dried blood was observed during 6-48 hours but maximum abrasions showed dried blood during 12-18 hours. This is consistent with the finding in the textbook of Mukherjee JB, Rao N, Vij K, Reddy

KSN and study of Sandhu SS et al.^{6, 9-12} In the present study, reddish coloured abrasions were observed from 0-12 hours, maximum abrasions showed reddish coloured abrasion during 0-6 hours. These observations are consistent with findings in the textbook of Nandy A, Modi JP, Mukherjee JB, Dikshit PC, Vij K and study of Sandhu SS et al, Sharma et al and not consistent with the findings in the textbook of Pillay VV, Rao N, Reddy KSN and study of Kumar L et al.^{6,8-18}

Reddish brown scab with dried blood was observed from 6-48 hours, maximum abrasions showed reddish brown scab with dried blood during 18-48 hours. These observations are consistent with the findings in the textbook of Rao N, Bardale R and study of Sandhu SS et al.^{6, 10, 18} Hard, brown scab was observed from 2-5 days, maximum abrasions showed hard brow scab during 2-3 days. These observations are consistent with the findings in the textbook of Nandy A, Modi JP, Dikshit PC, Rao N, Sharma et al, Vij K, Bardale R, Reddy KSN and not consistent with the findings in the textbook of Mukherjee JB and Kumar L et al.^{8-16, 18} Hard, black scab was observed from the fifth day onwards, maximum abrasions showed hard black scab during 5-6 days. These observations are consistent with findings in the textbook of Nandy A, Dikshit PC, Vij K, Bardale R, Reddy KSN and not consistent with the findings in the textbook of Pillay VV and Kumar L et al.^{8, 11, 12, 14, 16-18}

In the present study, partial shedding was observed from 3-4 days onwards, maximum abrasions showed partial shedding on 5-7 days. These observations are consistent with the findings in the textbook of Nandy A, Modi JP, Mukherjee JB, Parikh CK, Dikshit PC, Vij K, Bardale R, Reddy KSN and study of Sandhu SS et al, Sharma et al and not consistent with the study of Kumar L et al.^{6,8,9, 11-16,18} Complete shedding was observed from 6-7 days onwards, maximum abrasions showed complete shedding on 7-8 days. These observations are consistent with the findings in the textbook of Mukherjee JB, Pillay VV, Vij K, Bardale R, Reddy KSN and study of Sandhu SS et al and not consistent with the study of Kumar L et al.^{6, 9, 11, 12, 14, 17, 18} In the present study, depigmented skin was observed from seventh day onwards, maximum abrasions showed depigmented skin during 7-10 days. These observations are consistent with the findings in the textbook of Nandy A, Mukerjee JB, Dikshit PC, Pillay VV, Vij K, Bardale R, Reddy KSN and study of Sandhu SS et al and not consistent with the study of Kumar L et al.^{6, 8, 9, 11, 12, 14, 16-18}

b. Microscopy:

In the present study, neutrophils started appearing from first hour but appreciable number of neutrophils appeared from 6-12 hours which is consistent with findings of Robertson I and Hodge PR, Mukherjee JB, Pillay VV, Vij K, Bardale R, Reddy KSN and study of Sharma et al.^{9,11-13,17-19} Significant increase in the number of neutrophils was seen in 24-30 hours which is consistent with the findings of Dann L, Glucksmann A and Tansley K, Vizian CB and Matoltsy AG, Vij K and not consistent with findings of Robertson I and Hodge PR, Bardale R and Reddy KSN as they observed this at 4-12 hours.^{11, 12, 18-21} In the present study, mononuclear cells were observed in 6-12 hours which is consistent with the findings of Mukherjee JB, Dikshit PC, Sharma et al, Bardale R, Reddy KSN and not consistent with the findings Vij K as they observed this at 24-48 hours.^{9,11-13,16,18}

In the present study, it was observed that the number of mononuclear cells increased significantly at 48 hours. This is consistent with the findings of Vij K, Sharma et al and not consistent with the findings of Bardale R and Reddy KSN as they observed this at 12-24 hours.^{11-13,18} Special stain Toluidine blue is a basic thiazine metachromatic stain with a high affinity for acidic tissue components. It is used to enhance the visualization of subtle injuries, particularly abrasions, that may not be easily detectable to the naked eye. When applied to tissue samples, toluidine blue selectively binds to nucleic acids in damaged cells. This binding highlights areas of cell injury, making abrasions more visible, under microscopic examination. Toluidine blue was used to stain mast cells (the mast cells were observed as metachromatic pink coloured cells with blue background). The mast cell degranulation was observed from 35 minutes i.e 0-6 hours of infliction of abrasion which was consistent with the findings of Reddy KSN and Sharma et al.^{12, 13}

Elastic van Gieson (EVG) stain combines two dyes: Elastic fibers are stained black or dark brown. Collagen fibers are stained red or pink. This staining pattern allows for the detailed examination of tissue architecture and the identification of structural disruptions caused by trauma such as abrasions. EVG stain when applied to abrasion samples highlights disruption of elastic and collagen fibers in the dermis, indicating extent of tissue damage. In our study, tissue sections stained with EVG stain highlighted collagen fibres as red too brown fibres. In the present study, collagen formation was observed by 2-3 days

(48 hours onwards) and it became dense beyond 5-7 days. This is consistent with the findings of Vij K, Bardale R, Reddy KSN, Sharma et al and not consistent with Dann L, Glucksmann A and Tansey K, Robertson I and Hodge PR as they observed collagen formation by the fifth day.^{11-13, 18, 20} The histopathological changes will be helpful to measure the age of abrasion more accurately, but further research is needed. Also, using the artificial intelligence there are future scope for research & innovation.²²⁻²⁵

5. Conclusion:

There are various factors influencing the healing of wounds like nutrition status and immunity of individual, presence of comorbidities, infection and age. Routinely, the age of wound is calculated using the available information from the textbooks and existing literature. Also, there is always some sort of observer variation. Even though the gross appearance provides an approximate age of wound, histopathology remains the mainstay for the definitive diagnosis. Hence, the importance of microscopic examination of wounds cannot be over emphasized and is very helpful in the administration of medico legal cases. Also, further studies with larger sample sizes along with Immunohistochemistry will surely help in improving wound dating accuracy.

Ethical Clearance: IEC approval is taken from the Institutional Ethical committee.

Contributor ship of Author: All authors equally contributed.

Conflict of interest: None to declare.

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