ISSN: 0369-8963

Visualization of Micro crack pattern of dry blood stains on solid target surfaces.

Hemraj Verma[1], Dr. Surya Shekhar Daga[1], Dr. Umema Ahmed[1], Dr Girish Kumar Mathur[1], Ms. Pooja Rawat[1,] Gunjit Singhal[1]

Department of Forensic Science, Vivekananda Global University, Rajasthan.

Abstract: Pictorial or visual identification of deposited bloodstain pattern, matters specially where the blood found is in very less quantity where chemical test may destroy the only remanent of blood stain, i.e., skeletonised remain of blood stain on washed or wiped surface of vehicle or weapon. Authors, summarized the interpretation made by visual identification of micro-cracks inside remnant drops, smear or tiny blood streak on various solid target surfaces commonly encountered at crime spot i.e., ceramic tiles, metal surfaces, cloth articles, and wood. Instead of large cracks on dry pool of clotted blood, these micro-cracks are hard to visualise by naked eyes hence we used microscopic photography to document these observations at total magnification of 40x or 100x magnification. We observed relative variations in crack size on single target surface in addition to absence and smaller to large crack size variations on different target surfaces and associated them with possible reasons for such variations.

Key words: Bloodstain pattern analysis, Blood clotting, Drying, Absorption, Micro-cracks, Marangoni convections, Hydrophilic surface, Hydrophobic surface, Porous and non-porous Surface.

 https://doi.org/10.5281/zenodo.14054158

Introduction:

Bloodstains encountered at crime spot are often in the form of dry stain or remnant skeletonised stain during the documentation. Due to its sticky property outer boundary of impacted blood drop get pinned on most surfaces after deposition. Often very small quantity of bloodstains, creates a confusing scenario for the investigator to whether chemically test it or not. We tried to address this problem by highlighting the typical primary visual identification features of dried bloodstains encountered at crime spot by observation under microscope. The multi-disciplinary nature of blood stain pattern analysis makes it a very interesting scientific area where rheology, trigonometry, biology, biochemistry, physiology, all together play their role in crime scene investigation. Non-repeatability and non-replicability of any particular crime event, makes bloodstain pattern analysis a disputable subject to fit in the definition of science where replicability of experiment is a must. This area helps to ascertain the said statement by eye witness to be corelated with what physical evidence otherwise indicate. Blood has two components one is cellular and another is liquid(plasma). Red blood cells in blood were first observed by Antonie van Leeuwenhoek in 1699, and was termed as red globules, either individual or in clumping [1]. Blood is a non-Newtonian fluid due to red blood cells, and have shear thinning property. it's viscosity (shear rate resistance to flow in response to shear stress) i.e., viscosity decreases with increase in shear rate. Red blood cell has higher density than plasma (approximately 1100kg/cubic meter) [2-4]. On the other hand, blood plasma behaves like a Newtonian fluid hence at higher shear rates plasma property also have its effect on viscosity. Human RBC's cluster forming tendency is at low shear rates is known as "rouleaux". Du No"uy, observed surface tension reduction for blood serum as a function of time [5]. In

 https://doi.org/10.5281/zenodo.14054158

ISSN: 0369-8963

presence of plasma proteins (fibrinogen and globulin), the cells tend to aggregate despite having similar negative charges on themselves (Rampling,1988). Use of Adobe Photoshop® for measuring length and width ratio of deposited bloodstain on target surface briefed by Mathew Noedel [6]. Sergeant Mark reynolds briefed an about Microsoft Office Excel® 2003 Auto shape function-based measurement for blood satin measurement [7]. After deposition on solid target surface blood stain undergoes a series of changes. Color change occurs after drying and clotting due to further change of hemoglobin in methemoglobin and heamochrome. Deposited blood stain has thick central region and lighter periphery. Often this central thick clotted blood get flakes off and shaded after drying and gelation due to large crack formation in central thick region of dry blood stain. In addition to these large central cracks, Authors observed micro cracks inside both thin and thick clotted blood stains as well as in side remnant dry blood stain on various target surfaces. We found that micro crack pattern inside clotted blood stain was found to be a characteristic phenomenon in case of clotted dry blood stain specially on both porous and non-porous target surfaces. Drying and gelation of deposited bloodstain is followed by impact and spreading on various solid surfaces. In due course of drying by vaporization and gelation of deposited blood on various solid surfaces involve the movement of R.B.C and other cellular component's movement toward periphery by Marangoni convections. Further drying and gelatin sheet formation causes stretch and thus resulted in 'crack pattern inside thick region of dry blood stains on various solid surfaces caused by stress induced by water evaporation. These crack patterns were observed in 2013 by Bouzeid and Brutin [8]. Brutin, in 2011 observed that the Marangoni convection pulls R.B.C. towards the rim or outer periphery of the drop. They observed the Marangoni flow inside the motion inside a drop of human blood by a digital camera [9]. We observed micro-crack patterns on variety of blood streak, smear, and bloodstained non porous target surfaces in dried blood stains at 40X and 100X magnification. In addition to these micro-cracks, we observed prominent, and large

 https://doi.org/10.5281/zenodo.14054158

cracks emanating from center of the drop and further progression toward periphery as the time elapsed [10]. These large cracks are visible by naked eyes inside central dense region of the dry human blood drop deposited on ceramic tile, due to central collective cohesive force drawing red cells toward center of the drop **(fig.01).** Micro crack pattern can be used as an identification feature of blood stain on various solid target surfaces along with peculiar shine, and gelation. Authers identified these micro-cracks inside various solid target surfaces i.e., metal rod, stainless steel, cloth, ceramic tile, cemented floor surface, dry soil smeared metal surface and wood. Summarized data is presented in pictorial form by digital camera by microscopic photographs at total magnification up to 40x and 100x magnification.

Sampling Methodology:

We used air dried bloodstains deposited on various target surfaces, were observed followed by self-withdrawal of blood. Dry blood-stained area is selected for the observation and further microscopic observations were done on both porous as well as common non-porous surfaces. Results thus obtained were documented by digital camera and compound microscope. Findings summarized and presented in form of microscopic observation at 40x magnification and 100x magnification along with digital Photograph.

 https://doi.org/10.5281/zenodo.14054158

Observations:

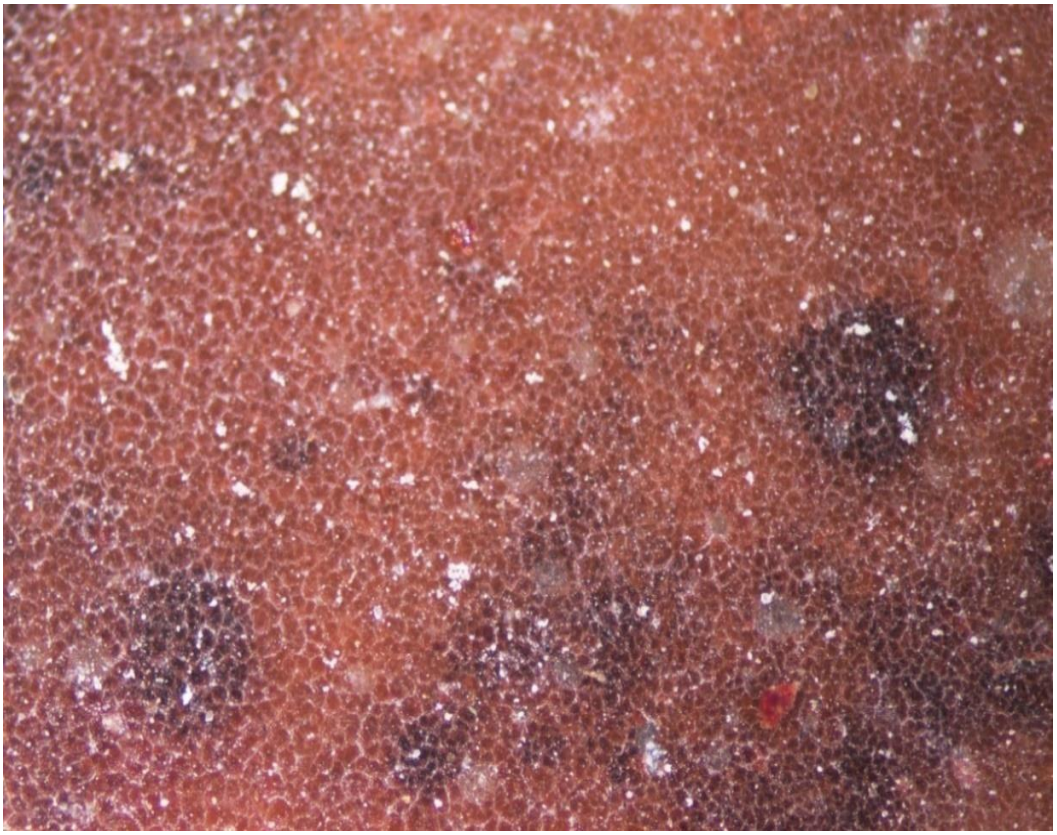


Fig.01: Crack pattern dried human blood at 40x magnification deposited blood drop on ceramic tile surface.

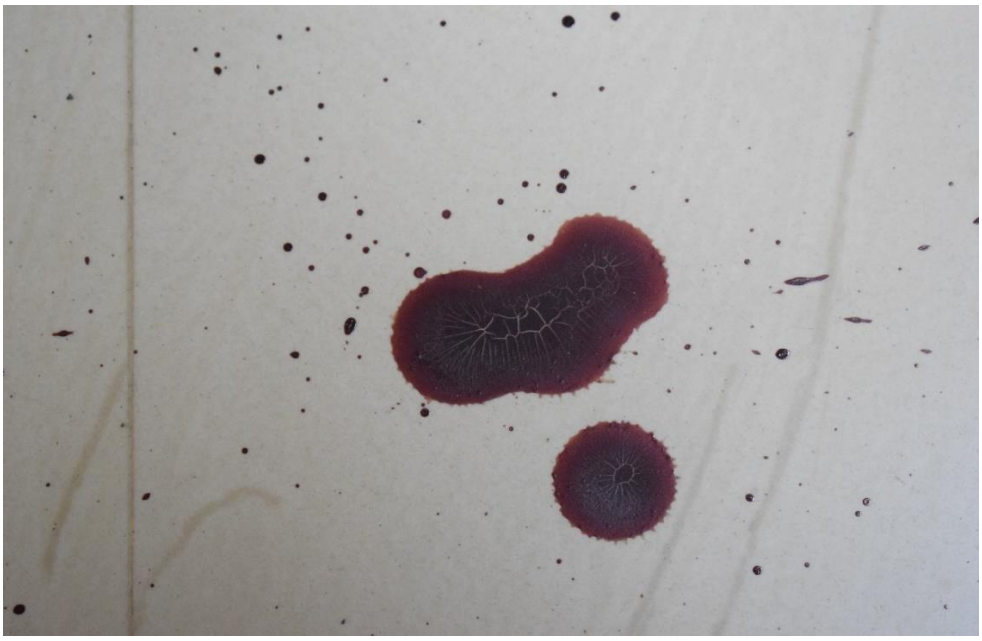


Fig.01: Large crack pattern inside center of dried human blood drops deposited on ceramic tile surface.

https://doi.org/10.5281/zenodo.14054158

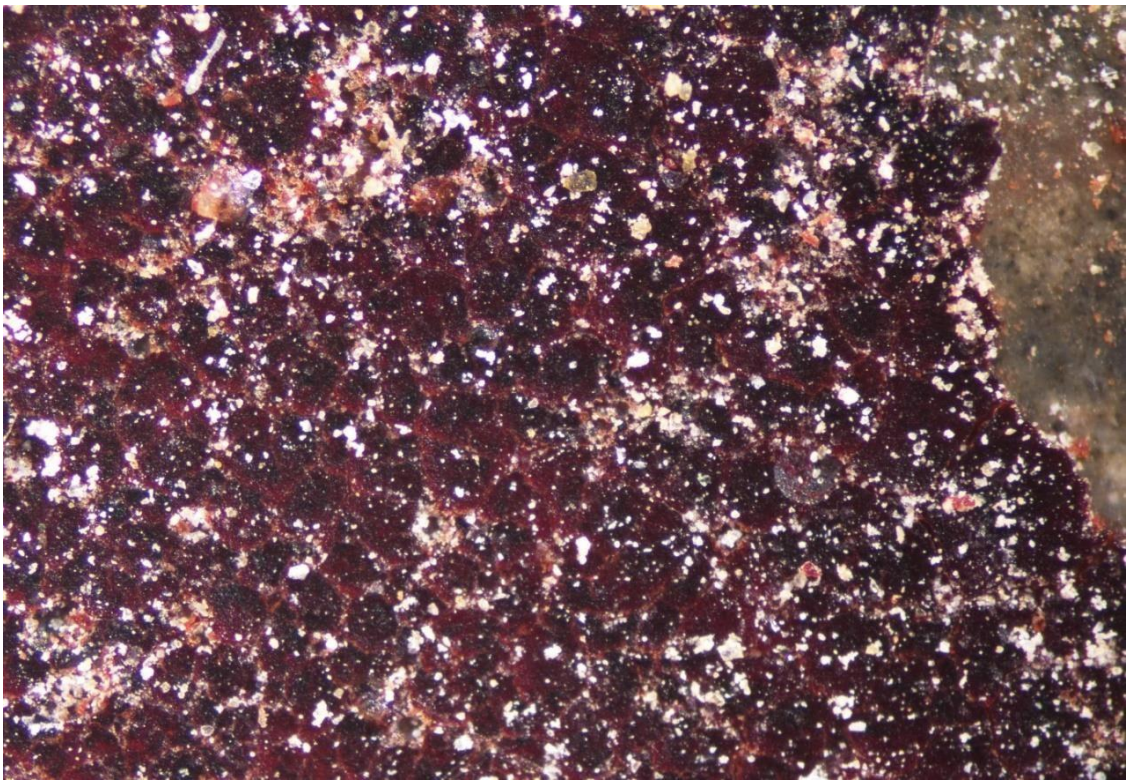


Fig.02: Crack pattern of dried human blood smear at 40x magnification on rough cemented floor surface.

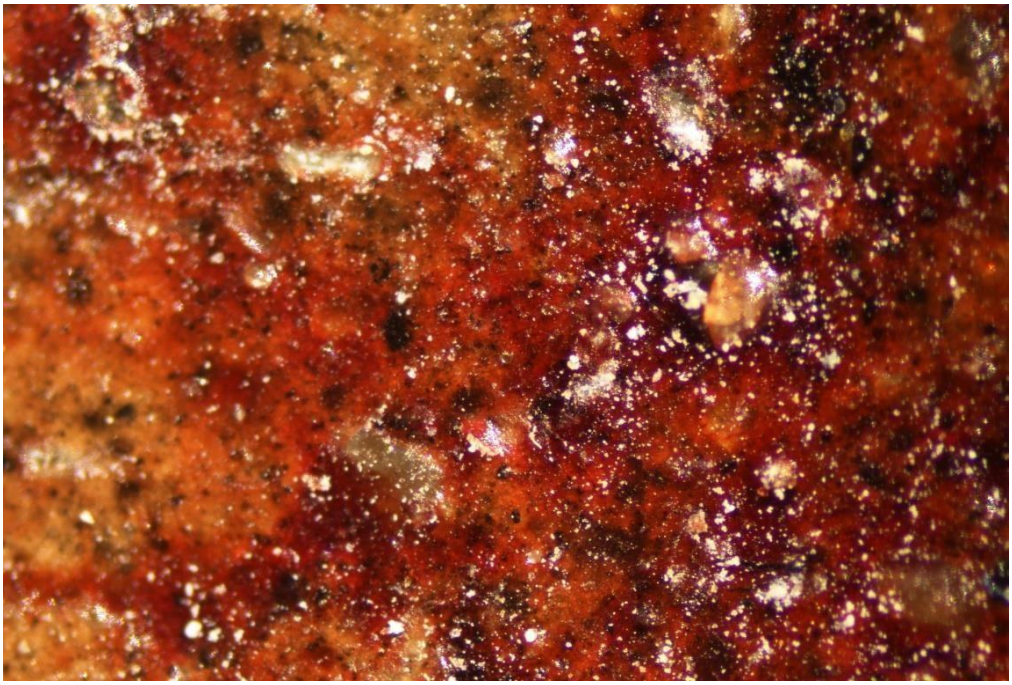


Fig.03: Absence of crack pattern on relatively porous cemented floor at 40x magnification.

https://doi.org/10.5281/zenodo.14054158

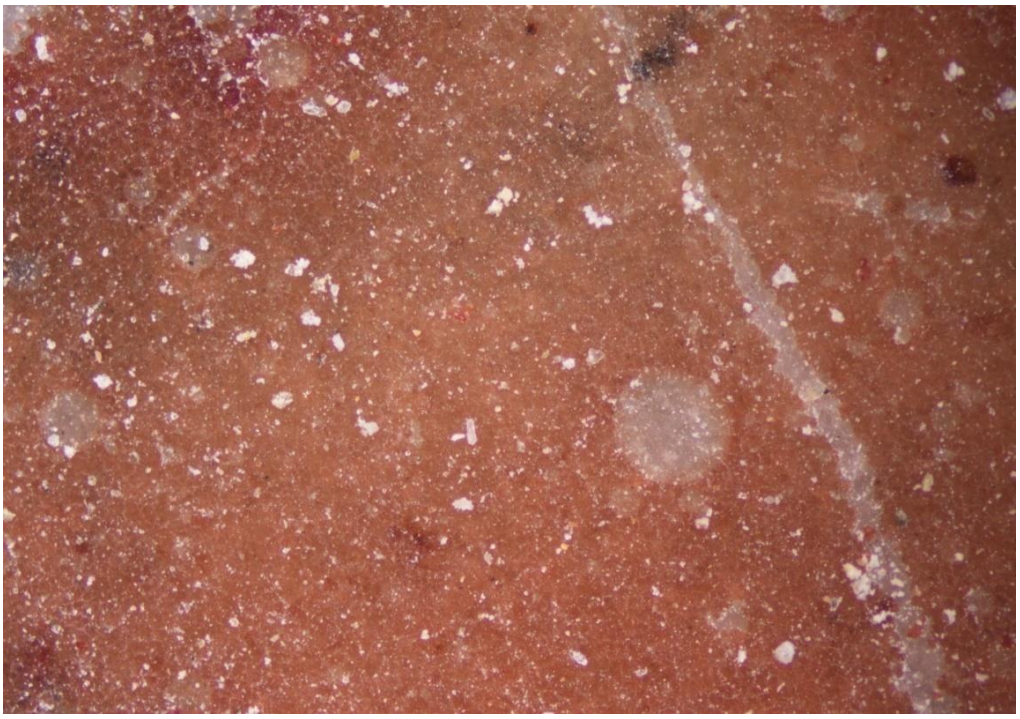


Fig.04: Micro-cracks around dry voids due to air bubble inside fresh and dried anti-mortem human blood-stained granite floor tile at 40x magnification.

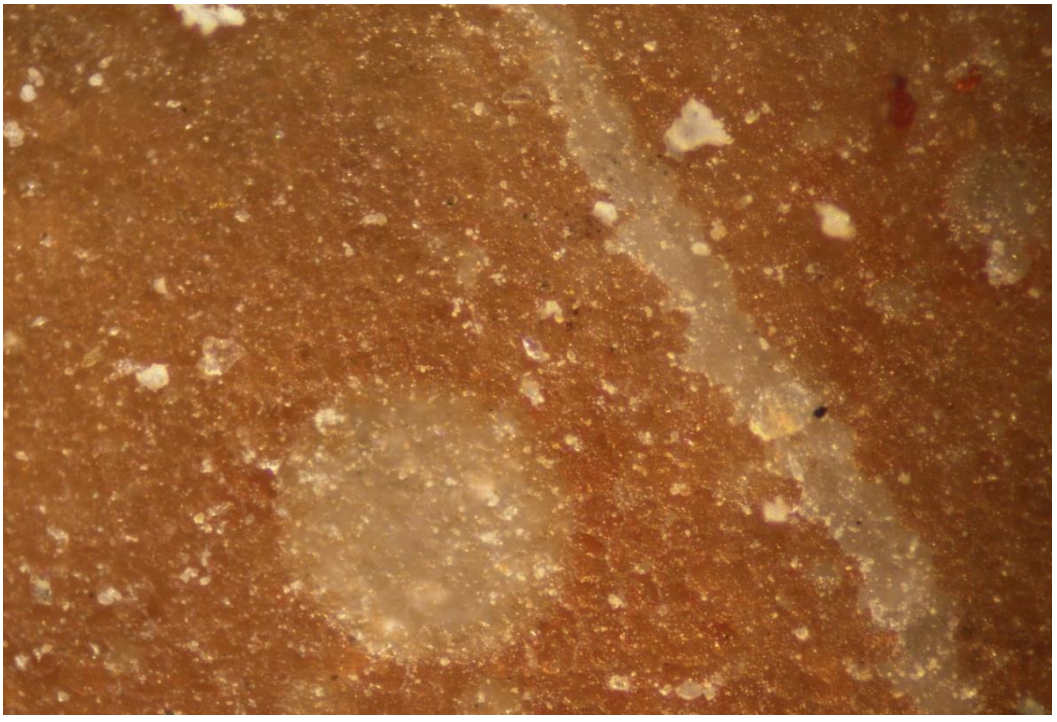


Fig.05: Note the tiny crack pattern around void caused by tiny air bubble inside dried smear of blood stained granite tile at 100x magnification.

https://doi.org/10.5281/zenodo.14054158

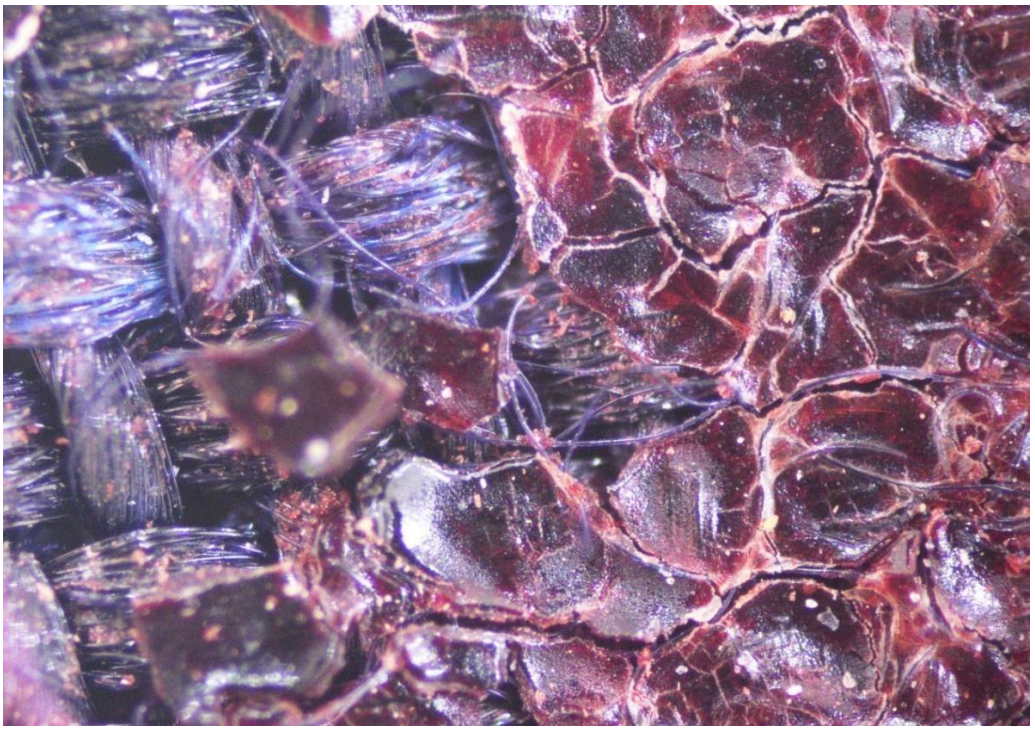


Fig.06: Crack pattern on deposited thick dry blood stained, synthetic thick fabric at 40x magnification.

Fig.07: Large cracks on dried, thick clotted human blood at 40x magnification on polyester cloth.

https://doi.org/10.5281/zenodo.14054158

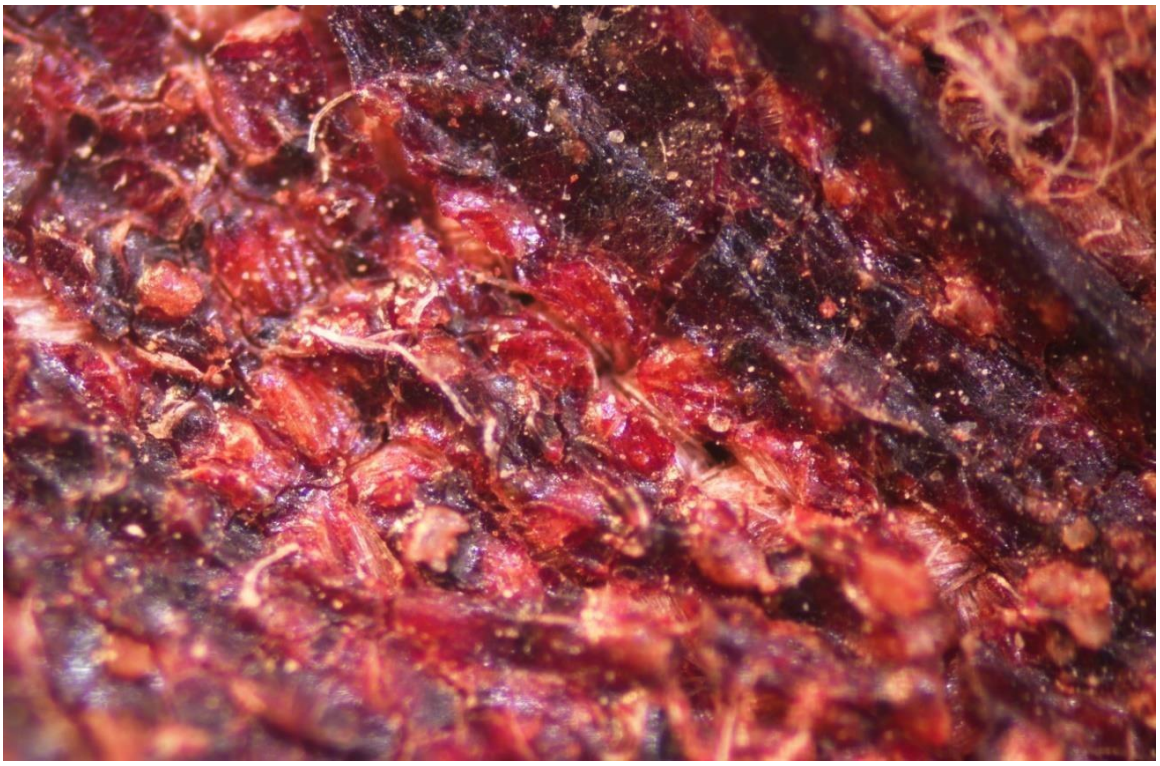


Fig.08: Relatively thin layer of dried human blood showing absence of significant crack pattern on polyester cloth at 40x magnification.

Fig.09: Blood drop dried on dicot wooden stick without bark, showing skeletonized blood drop.

https://doi.org/10.5281/zenodo.14054158

ISSN: 0369-8963

Fig.10: Microcracks inside dry blood stain deposited on dicot wood surface at 40x magnification.

Fig.11: Micro-cracks also observed in remnant skeletonized bloodstain of human blood on wood surface at 100x magnification.

 https://doi.org/10.5281/zenodo.14054158

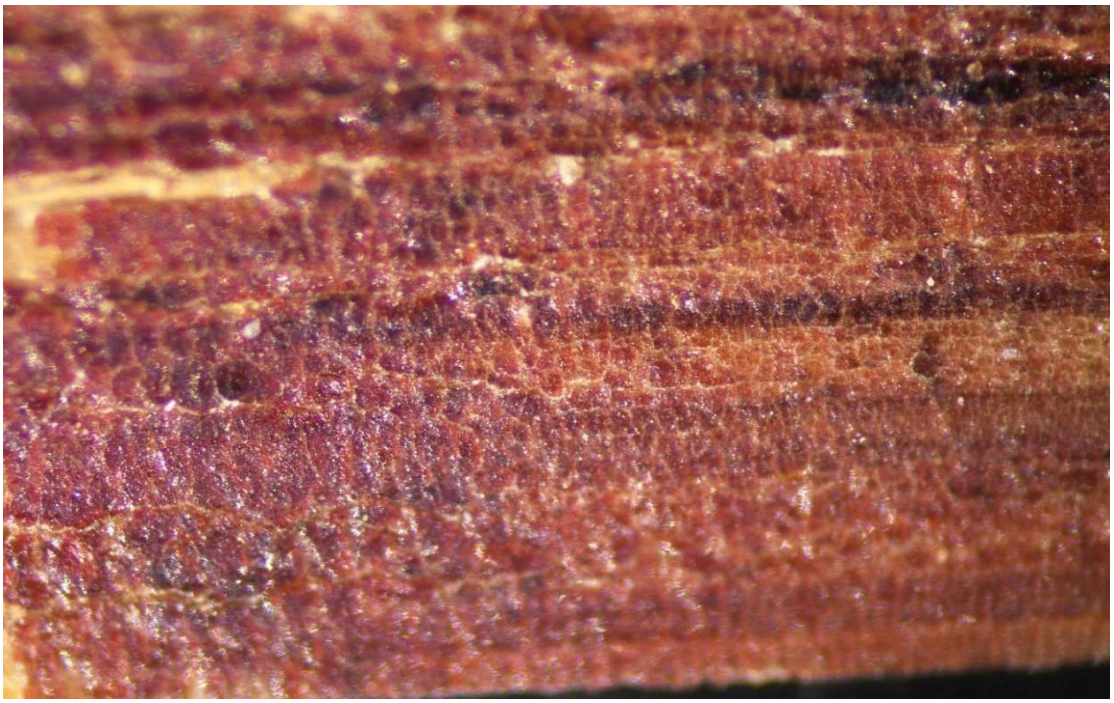


Fig.12: Micro-cracks inside thin dry blood smear on wooden surface at 100x magnification.

Fig.13: Absence of any such cracks inside thin dry stain of plant resin on dicot wood at 40x magnification.

https://doi.org/10.5281/zenodo.14054158

Fig.14: Another wooden stick with blood like resin stain without any crack inside dry resin stain at 40x magnification.

Fig.15. Blood smeared stainless steel knife, allowed to dry.

https://doi.org/10.5281/zenodo.14054158

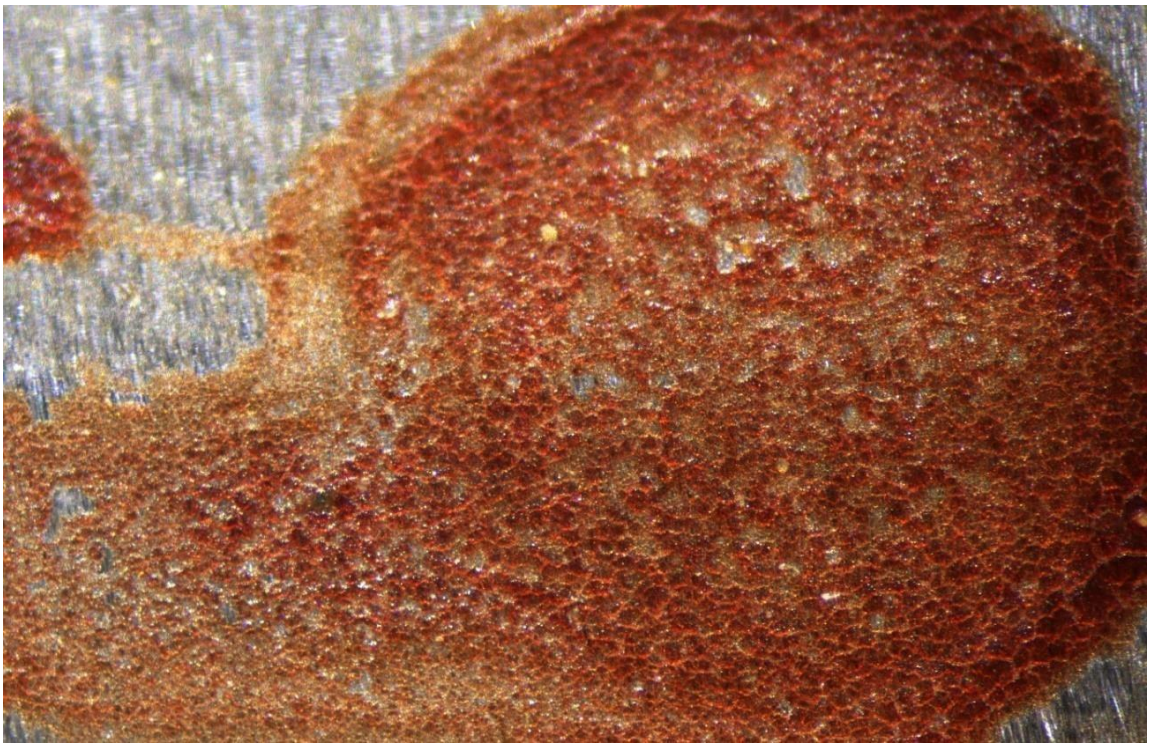


Fig.16: Note the large cracks on the thick rim of dried blood drop deposited on stainless steel blade of knife at 40x magnification.

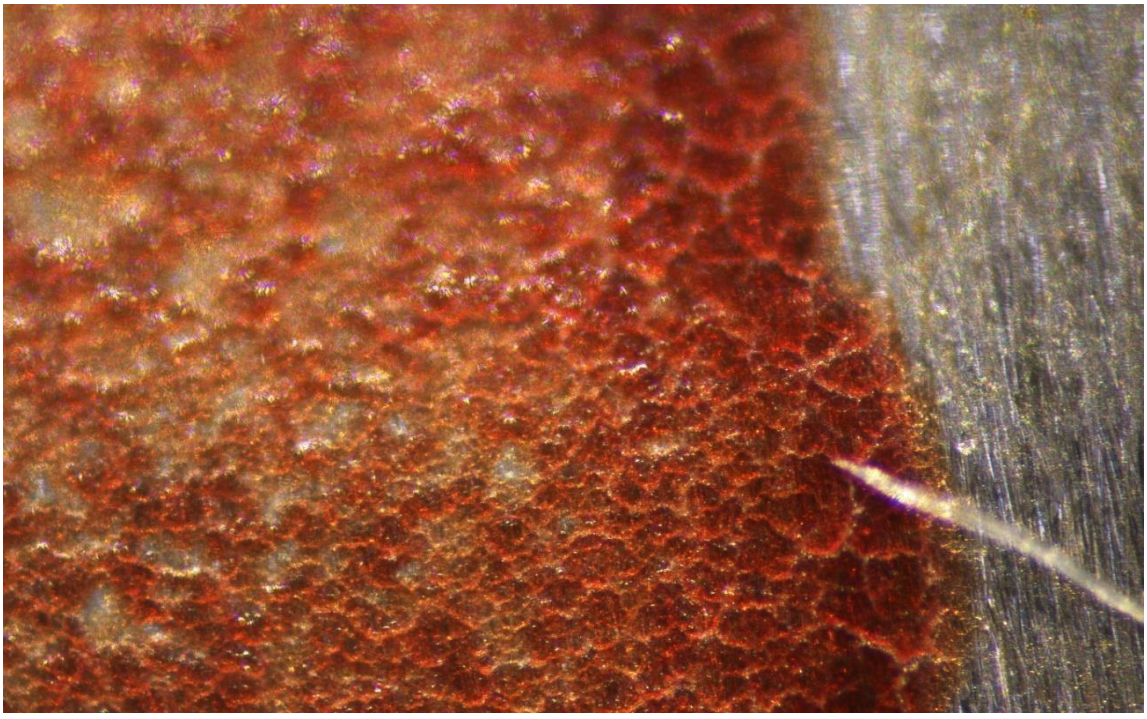


Fig.17. Blood stain at 100x magnification, note the tiny crack pattern inside lamella and thick cracks inside rim part of the blood drop deposited on stainless steel knife blade surface.

https://doi.org/10.5281/zenodo.14054158

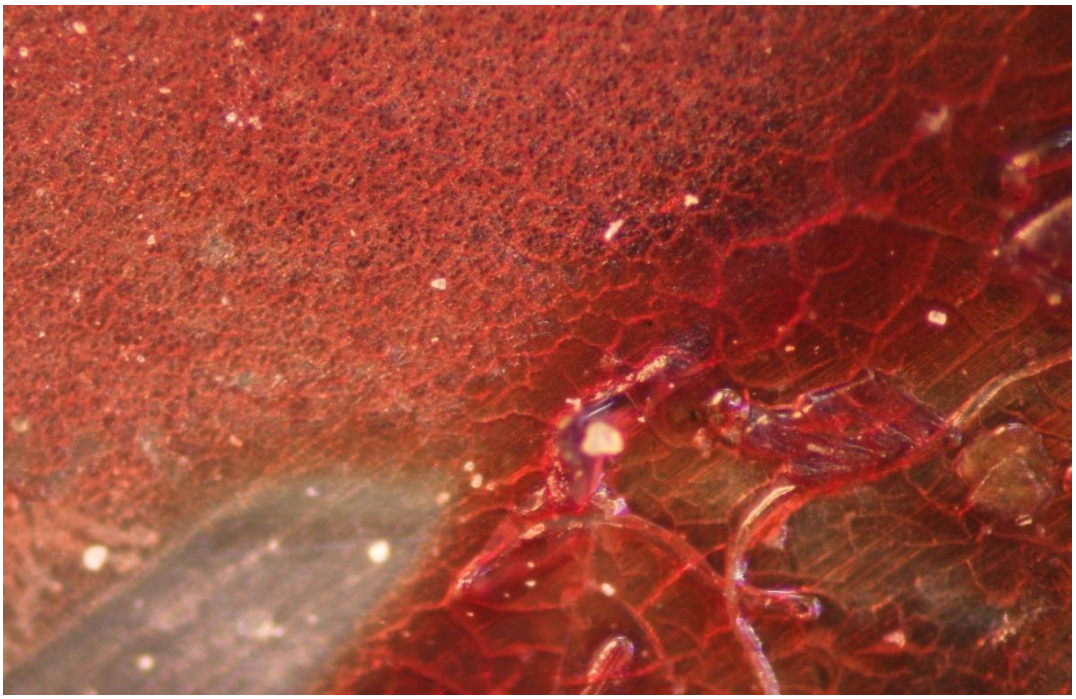
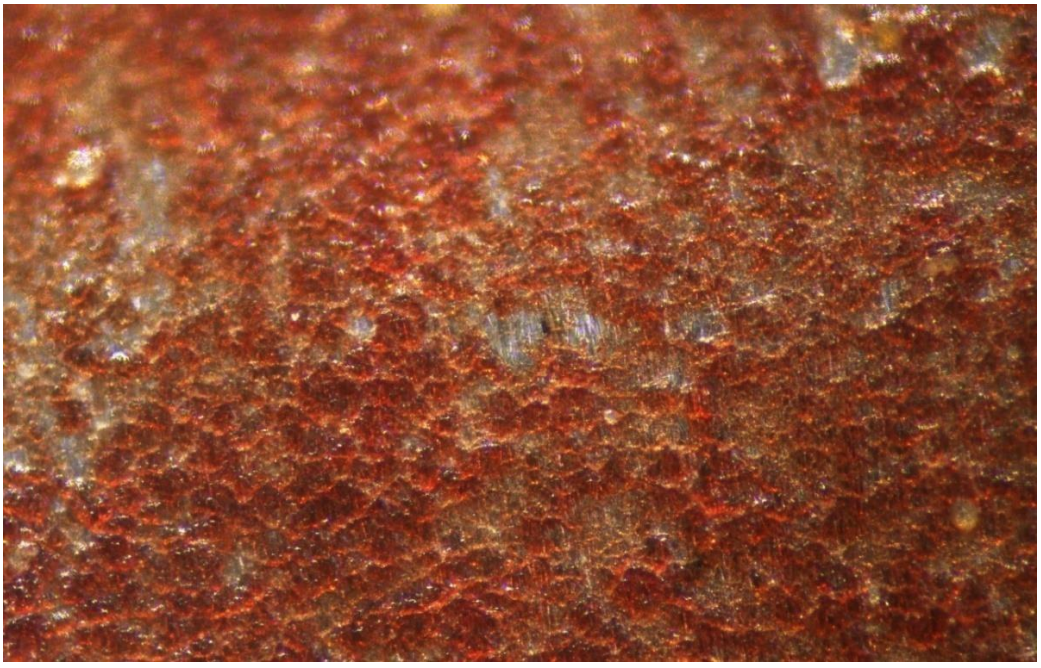


Fig.18: Large size of cracks on thick border of clotted bloodstain due on gelation and clotting before complete drying on stainless steel knife blade surface at 40x magnification.



https://doi.org/10.5281/zenodo.14054158

Fig.19: Relatively small cracks inside thin dry lamella of the blood streak on hydrophilic stainless blade surface of the knife at 100x magnification.

Fig.20: Smaller cracks on relatively thin layer of plasma rich human dry blood smear at left side and large cracks on thick red blood cell rich layer at 40x magnification.

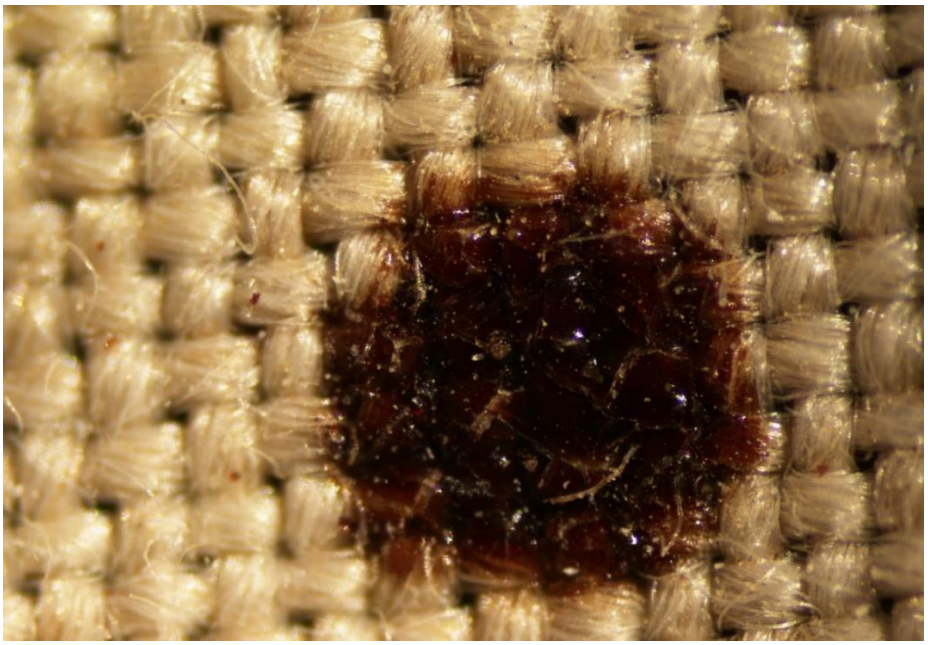


Fig.21: No cracks inside tiny thick human blood droplet in the microliter range on polyester cloth surface at 40x magnification possibly due to gelation and clotting.

https://doi.org/10.5281/zenodo.14054158

Fig 22: Note the absence of crick inside thin tiny dry clotted blood droplet on polyester cloth at 40X magnification possibly due to fast absorption of liquid component before drying.

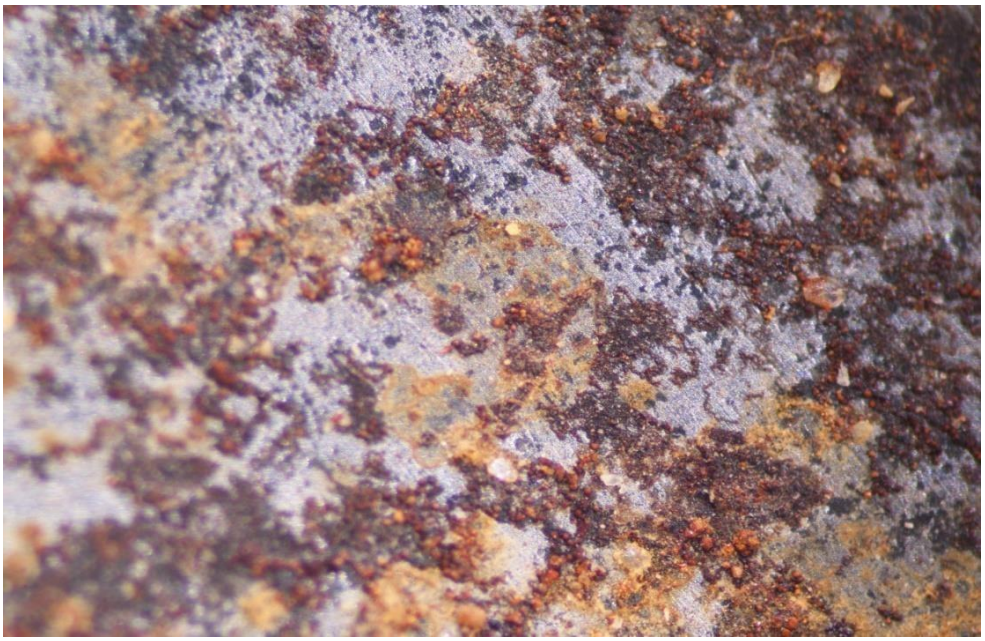


Fig.23: Cracks not visible inside blood smeared soil on rusted metal surface, light brown stains are of rust and dark red brown stains are of blood smeared soil on metallic surface at 40x magnification.

https://doi.org/10.5281/zenodo.14054158

ISSN: 0369-8963

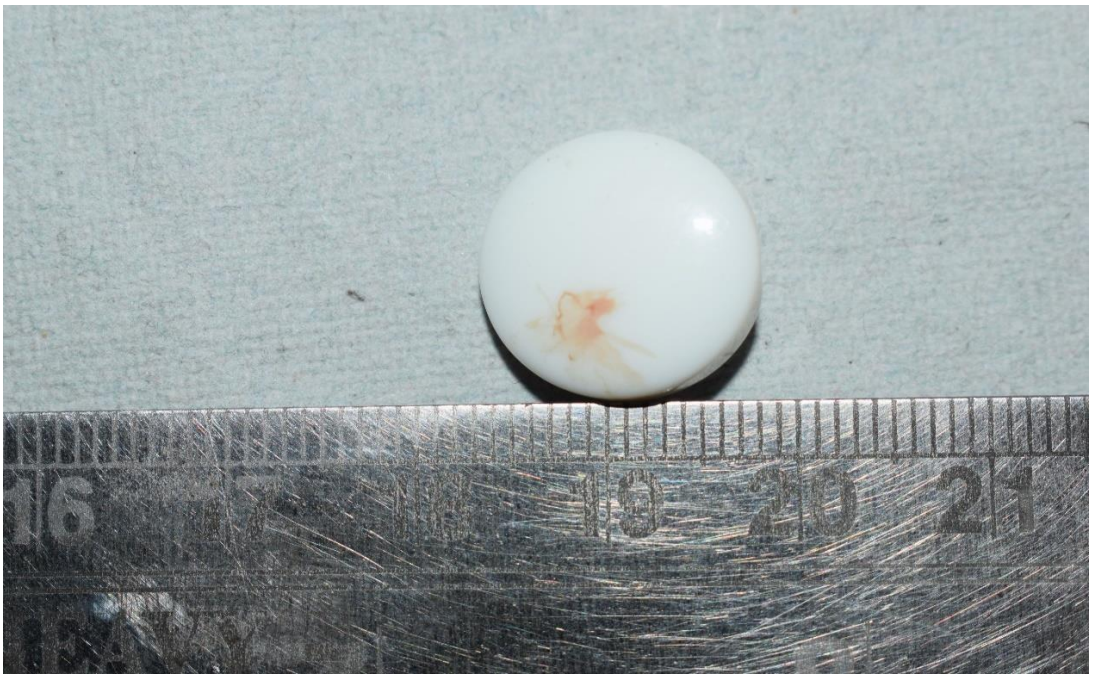


Fig.24: Skeletonized thin diluted blood smear on smooth surface of plastic button.

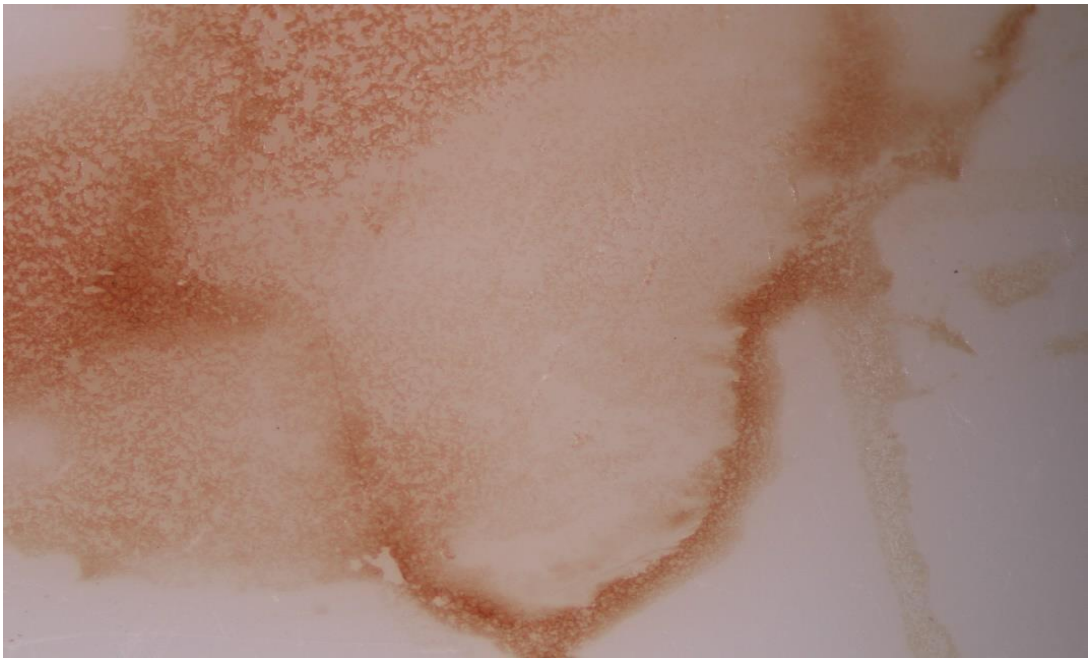


Fig.25: Thin diluted blood smear on plastic polymer smooth surface of button shows absence of cracks due to dispersal and drying of diluted blood prior to clotting at 40x magnification.

 https://doi.org/10.5281/zenodo.14054158

Discussion:

Laan et al. in 2016, identified different phases of drying of blood pool i.e., coagulation stage, gelation stage, rim desiccation stage, center desiccation stage, and final desiccation stage [11]. Identification and documentation of blood traces deposition on target surface, without chemical test is considered as preliminary. Authers observed similarity in micro-crack patterns on dry blood smear and drops on solid non-porous surfaces. Large pool of blood has large cracks visible with naked eyes whereas small blood drops, clots, various blood stained non porous objects including weapon and floor and relatively thin clotted blood smear have comparatively smaller crack pattern visible by microscopic observation at total magnification of 40x and 100x magnification [12-13]. Buildup of red cells at periphery were observed in small stains and smear on non-porous surfaces, **(fig.;15, 16, 17, 19, 21, 22 and 23)**, whereas porous hydrophobic surfaces show relatively large micro cracks when the deposited blood form thick clot before absorption of liquid component of blood (**fig.;02, 05, 06, 07, 08, 09, 10, 11, and 12).** On microscopic observation, cracks were not found on cloth and soil smeared rusted metal surface due to quick absorption by capillary effect of sand particles on drying before being clotted **(Fig.22, 23).** Although further analysis and interpretation is required to confirm the presence of blood stain yet the micro cracks are identified as visual characteristic for dry clotted blood stain. Since every bit of trace blood stain evidence i.e., skeletonized remains of blood stain on wiped or washed article needs careful processing due to integrity of the evidence [14-17]. Hence visual identification of micro crack pattern on remnant bloodstain serves as crucial step before applying any harmful or destructing chemical to identify or confirm dry blood stain. Relation between crack size and hematocrit value, effect of blood dilution on resultant cracks as well as effect of target surface on crack pattern need further evaluation (**Fig.24, 25**). It is observed that garments and soiled and rusted surfaces, post impact spreading of impacted blood drop occurs due to capillary action of soil particles and absorption on porous absorbent surfaces. In case of

 https://doi.org/10.5281/zenodo.14054158

non-porous surface impacted wet drop diameter remained same after drying due to pinning of outer periphery with the target surface [18].

Conclusion: Clotted blood show micro-crack patterns, on various solid target surfaces including cloth, wood, metallic surface, tiles and other nonporous surfaces after drying which may be used as visual identification feature of bloodstains. Microscopic examination of dry blood stain deposited on various solid surfaces revealed that relatively fast drying and clotting on hydrophilic and smooth stainless-steel surface have both small size cracks on the periphery and large cracks at the center or thick gelatinous region of the blood droplet and smear. Possible reason for smaller cracks on thin blood smear is fast drying time toward plasma rich thin smear at periphery even before gelation and clotting. Authors found that only thick clotted blood on the articles of clothing show crack pattern due to slow absorption by the garment in contrast to thin smear where blood is readily imbibed and dried before being clotted. **(fig.;06, 07, 21, and 22)**. Effect of hydrophilic and hydrophobic porous surface on deposited dry blood stain needs further study **(fig. 21 and 22)**. Rough and rusted metal surface have decreased spreading ratio due to long grooves results in less stretching upon drying and absence of cracks in thin smear in contrast to large cracks on thick blood smear. Skeletonization of dry blood stain is due to Marangoni flow on various non-porous surfaces. **(03, 04).** These crack patterns were not observed in lookalike plant resin stains **(fig.;13, 14).** A smooth surface bright coloured blood dry stain without smaller cracks were observed on absorbent surfaces, i.e., thick cloth, wood and sandy surfaces, possibly due to absorption of liquid component of blood before clotting and drying, and need further study. We observed that on non-absorbent metallic smooth surfaces crack size depends on thickness of stain and quantity of blood deposited on target surface (**Fig.18, 21, 22**). On smooth stainless steel surface red blood cell rich region of the deposited blood stain and smear has relatively large cracks in contrast to plasma rich thin blood smear **(fig.;15, 16, 17, 18, 19, 20).** Thick border of dry blood stain had large gelatinous cracks

https://doi.org/10.5281/zenodo.14054158

whereas relatively thin film of dry blood stain had smaller cracks gradually terminating in individual tiny flakes possibly due to aggregation of cellular components and further drying by evaporation. We were unable to find any crack inside the droplets in microliter range.

References:

1. A. van Leeuwenhoek. Concerning the circulation and stagnation of the blood in tadpoles. Philos. Trans. R. Soc. London, 22:447-455, 1702.
2. E. Ponder. The relation between red blood cell density and corpuscular hemoglobin concentration. Journal of Biological Chemistry, 144(2):333-338, 1942.
3. J. Lee and L. lee. A density method for determining plasma and red cell volume. Annals of biomedical engineering, 20(2):195-204, 1992.
4. O. Linderkamp, E. Friederichs, T. Boehler, and A. Ludwig. Age dependency of red blood cell deformability and density: Studies in transient erythroblastopenia of childhood. British journal of haematology, 83(1):125-129,1993.
5. A. Bejan and D. Gobin. Constructal theory of droplet impact geometry. International journal of heat and mass transfer, 49(15):2412-2419, 2006.
6. Matthew Noedel. Using Adobe Photoshop® Tools for Bloodstain Documentation: Abstracts of Papers Presented at the IABPA Annual 24.
7. Sgt. Mark Reynolds. Measurement of Bloodstains using Microsoft ® Excel Auto shapes: An Accurate and Precise Bloodstain Measurement System: Abstracts of the Scientific presentation given at the 2008 IABPA Annual Training Conference, Page11-12.
8. Bouzeid, W. and Brutin, D. (2013). Influence of relative humidity on spreading, pattern formation and adhesion of a drying drop of whole blood. Coll. And Surf. A,430:1-7
9. Brutin, D., Sobac, B., Loquet, B., and Sampol, J. (2011) Pattern formation in drying drops of blood. J. Fluid Mech.,667:85-95.
10. Laan, N., Smith, F., Nicloux, C., and Brutin, D. (2016) Morphology of drying blood pools. For. Sc. Int., 267:104-109.
11. Brutin D, Sobac B, Loquet B, Sampol J. Pattern formation in drying drops of blood. J Fluid Mech.2011; 667:85-95
12. Samir Kumar Bandyopadhyay*, Nabanita Basu. Review of Common Bloodstain Pattern Documented at a Crime Scene in the Event of Blunt Force Hit, AJCSIT (3)(1) (2015)045-063.

 https://doi.org/10.5281/zenodo.14054158

13. Karger B, Rand SP, Brinkmann B. experimental bloodstains on fabric from contact and from droplets. Int. J. Legal Med.2010;111(1):17-21.7.

14. R. Kannan and D. Sivakumar. Impact of liquid drops on a rough surface comprising microgrooves. Experiments in Fluids, 44(6):927-938,2008.

15. R. Kannan and D. Sivakumar. Drop impact process on a hydrophobic grooved surface, Colloids and Surfaces A: Physicochemical and Engineering Aspects, 317(1):694-704,2008.

16. M. Dube, B. Chabot, C. Daneault, and M. Alava. Fundamentals of fluid front roughening in imbibition. Pulp and Paper Canada-Ontario, 106(9):24,2005.

17. S. Michielsen, M. Taylor, N. Parekh, and F. Ji. Bloodstain pattern on textile surfaces: A fundamental analyses, 2015.

18. M. Marengo, C. Antonini, I. V. Roisman, and C. Tropea. Drop collisions with simple and complex surfaces, Current Opinion in Colloid & Interface Science, 16(4):292-302, 2011.

 https://doi.org/10.5281/zenodo.14054158