A Study to Compare and Contrast Animal Blood to Human Blood Product

Daniel V. Christman, Medical Investigator
Snohomish County Medical Examiner's Office
Everett, Washington

At the time the term Bloodstain Pattern Analysis was coming into its own, another term was making its debut in the law enforcement arena; bloodborne pathogens. While bloodstain pattern analysis was being taught, primarily in North America with some regularity, the issue of biohazard safety was not a paramount concern. Is whole human blood product a safe medium to be utilized in teaching bloodstain pattern analysis? For that matter, is animal blood a safe medium to handle? If not, what is a suitable substitute? Some teachers of this scientific discipline have replaced whole human blood with animal blood for instructional purposes. Some students have reported using cow and horse blood during the experimentation and teaching process. Many crime scene reconstructive techniques have undergone critical review in the judicial process and in time the use of animal blood will come under some close scrutiny by counsel. Are we ready to respond to legal questioning like:

- "How do these blood products differ in composition?"
- "Do human and animal blood maintain different flight characteristics?"
- "Do the resulting individual bloodstains exhibit any differences?"
- "What experimentation has been done to document any differences or similarities in the two?"

Using human and veterinarian hematology data, this paper will document, compare and contrast the composition of animal and human blood. Using standardized testing procedures, the differences and similarities of actual bloodstains will be presented with regards to the effect of differing target surfaces and angles.

In the past, most instructors have had little or no problem obtaining whole human blood from blood banks to use in teaching blood pattern analysis. Many blood banks have now restricted the use of whole human blood product for private experimentation. For the past four years, the blood banks in the greater Seattle, Washington area have denied private researchers access to blood products. Most of the expired blood from local blood banks is sold to large medical research groups for use in testing medical equipment. Bloodborne pathogens and liability

issues were cited as the primary reasons for this change in policy. Even though these facilities screen their blood thoroughly, with the onset of the human immunodeficiency virus (HIV) and the threat of hepatitis B (HBV) blood banks no longer feel comfortable providing blood products even to those with whom they have an established working relationship. By contacting local blood banks and plasma donation centers, instructors can determine how accessible blood products are in their locale. Some crime scene reconstructionists and bloodstain pattern instructors have found whole human blood products to be scarce if not impossible to obtain. A search for a suitable substitute has resulted in the use of animal blood in place of whole human blood products. Animal blood is easy to obtain, relatively inexpensive and in some cases free. Many animal processing facilities will cooperate by providing fresh, whole animal blood product upon request. Currently, there are a number of bloodstain pattern analysts and crime scene reconstructionists using animal blood to teach new analysts and conduct controlled experimentation. Those queried in this research reported using bovine (cow), equine (horse), swine (pig) and sheep (lamb) blood in replacement of whole human blood.

Throughout judicial history, many successful crime scene reconstruction techniques and processes have undergone critical review. Courts world-wide are seeing the power of bloodstain pattern analysis manifest itself in many high profile cases. Hence, attorneys are surfacing in 40 hour basic bloodstain pattern analysis training to get a better understanding of this scientific discipline. As a result of the legal community's interest in bloodstain pattern analysis, one could reasonably predict future judicial query to include questioning about the following:

- What specific type of blood was used to recreate a crime scene or specific bloodstain pattern? And if animal blood was used:
- How does animal blood differ from human blood?
- How do resulting bloodstains differ when studying the various blood types?

The following research was conducted in an effort to obtain a better understanding of how whole animal blood differs from whole human blood where bloodstain pattern analysis is used for crime scene reconstruction. Several areas of study were reviewed:

- Chemical and physical composition of blood.
- Differing types of impact (low, medium and high) subjected to the various fluids.
- Review of resulting stain patterns.

Materials and Methods

This paper will examine and document various liquid models to replace whole human blood for experimentation and teaching. For the purpose of this project, animal blood consisted of a sample of blood from the following species: bovine (cow), equine (horse), swine (pig) and sheep or lamb blood. Human blood consisted of post mortem blood drawn from the peripheral vascular system at time of autopsy. Standard Calligraphy Ink was used as the final liquid model to compare to the other bloods. In this study, both human and animal blood was treated with an anticoagulant (heparin) or a preservative (Sodium Floored) immediately after being drawn from the body.

Animal blood is relatively easy to obtain in large quantities especially in large bodied animals like bovine and equine species. Animal processing facilities can provide whole blood as can butchers who arrange slaughter service for private farms. In this study, animal blood was obtained and treated at the time of slaughter, by the author or by veterinarians in a clinic setting.

- Bovine or cattle were routinely shot in the head with a small caliber rifle. The animal was then hoisted, by the hind legs, into the air. While the animal was hanging, a suprasternal incision was made at the base of the neck, the aorta was accessed through the incision and punctured with a knife. Fifty cups of blood were obtained in a clean five gallon bucket situated under the animal. Prior to the blood letting, 500 ml. of anticoagulant citrate dextrose solution was placed in the bucket. As the blood entered the bucket, it immediately mixed with the anticoagulant creating an even distribution. The blood was then placed in one liter bottles for refrigerated storage and eventual use.

- Equine blood was obtained by veterinarians at a large animal clinic. An arterial blood sample was obtained by using a sterile syringe and transferring the blood into a purple top tube which contained heprin, an anticoagulant. The blood was then placed in one liter bottles for refrigerated storage and eventual use.

- Swine and sheep blood were obtained at the time of slaughter by the author. These animals were shot in the head with a small caliber rifle. An incision was made in the neck region making the arterial system readily accessible. A large bore needle and syringe was used to enter the arterial system and obtain the blood sample. The blood was then

transferred to a purple top tube which contained heprin, an anticoagulant. The blood was then placed in one liter bottles for refrigerated storage and eventual use.

- Standard Calligraphy Ink was purchased and used as the only non-blood liquid model. The exact chemical composition was not available at the time this paper was presented.

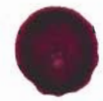
Additionally, human and animal hematology texts were used to compare and contrast the chemical and physical composition of each type of blood. Specific lab values were compared to determine if there was a real difference between the differing types of blood. A blood smear of each blood type was prepared using a standard *Wright Stain* and the slides were examined microscopically.

EXPERIMENTATION:

Bloodstain Shape v. Impact Angle

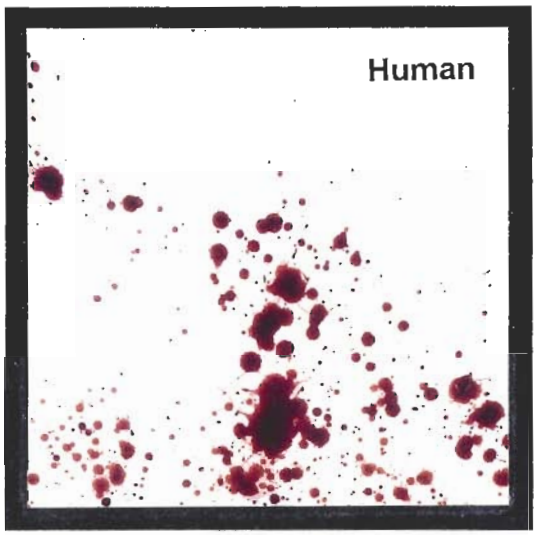
Single blood droplets of each type of blood were disengaged from a clean medicine dropper onto paper targets and the associated stains were studied. Human, bovine, equine, swine, sheep blood and ink drops were positioned 12" above a clean, smooth paper target. The targets were manipulated to different degrees to create varying angles of impact including 90°, 50°, 30° and 10°. The resulting stains were then studied for differences and similarities.

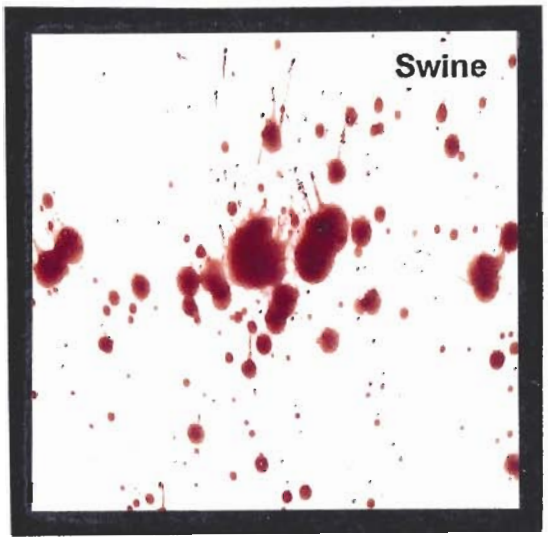
Comparison of Fluid Models Dropped at Increasing Angles

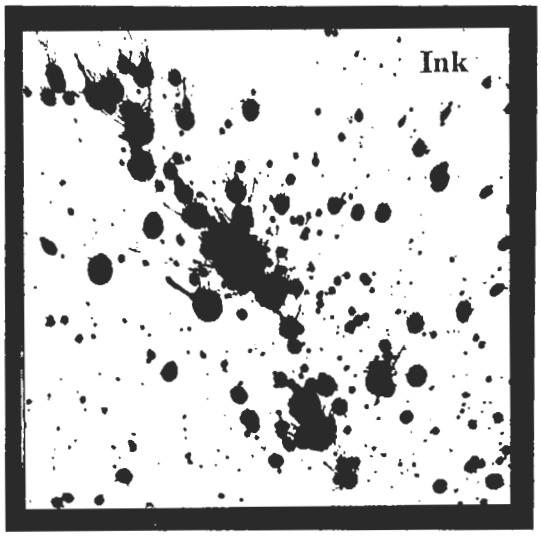
	Human	Bovine	Equine	Swine	Sheep	Ink
90°						
70°						
50°						
30°						
10°						

Medium Velocity Impact Spatter Comparisons

One cc of each type of blood was subjected to a medium velocity impact force. The static blood was positioned on a horizontal plane and a small steel hammer was introduced to the blood pool. Paper targets situated 10" around the perimeter of the blood pool collected the associated stain pattern to allow a permanent record of the event. A three and one-quarter inch square of the paper target was collected to represent the spatter pattern.

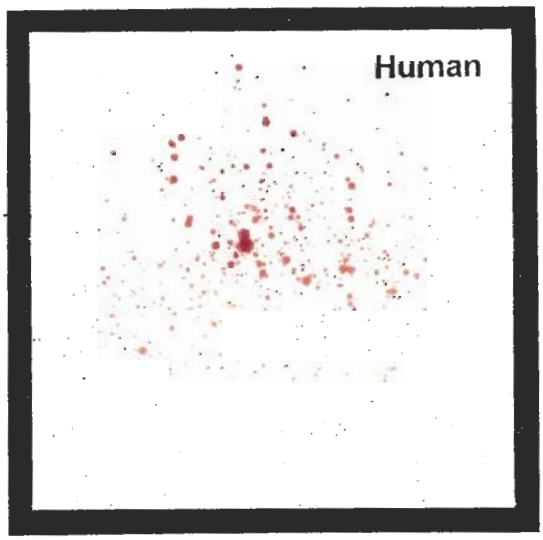


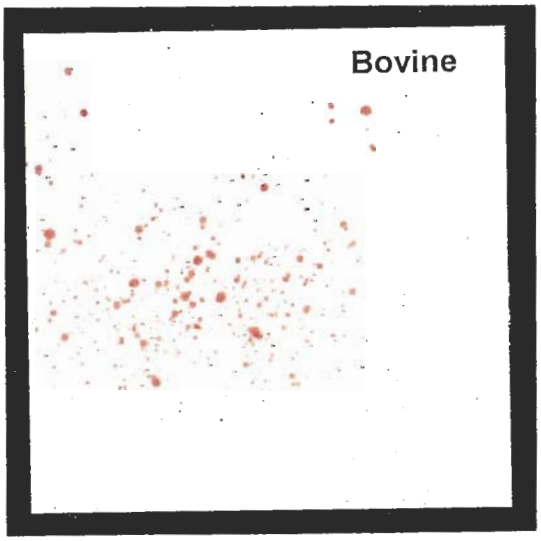


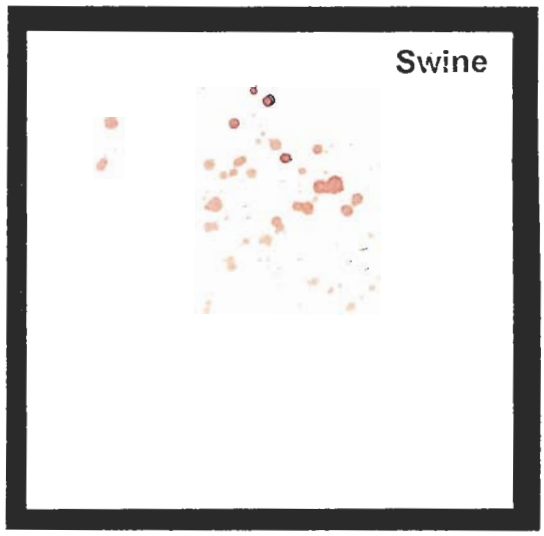


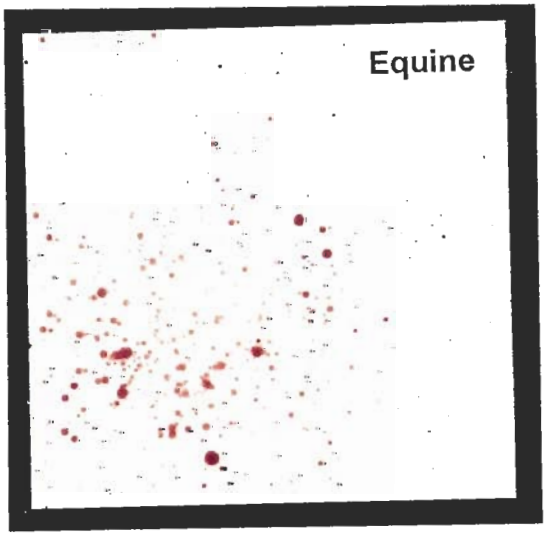
High Velocity Impact Spatter Comparisons

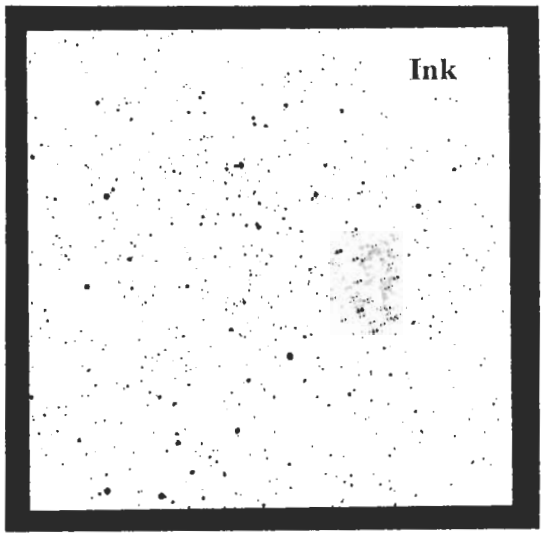
Each type of blood was subjected to a high velocity impact force. A quantity of each type of blood was placed in a plain, 12 cc syringe. The excess blood was discharged, which left a small residual amount in the cylinder. The plunger was drawn open and a rapid discharge of the remaining blood occurred. The resulting patterns represent a bloodstain pattern consistent with classic high velocity impact spatter patterns. The patterns were captured on adjacent 90° paper targets placed 12" away from the terminal end of the syringe. A three and one-quarter inch square of the paper target was collected to represent the spatter pattern.





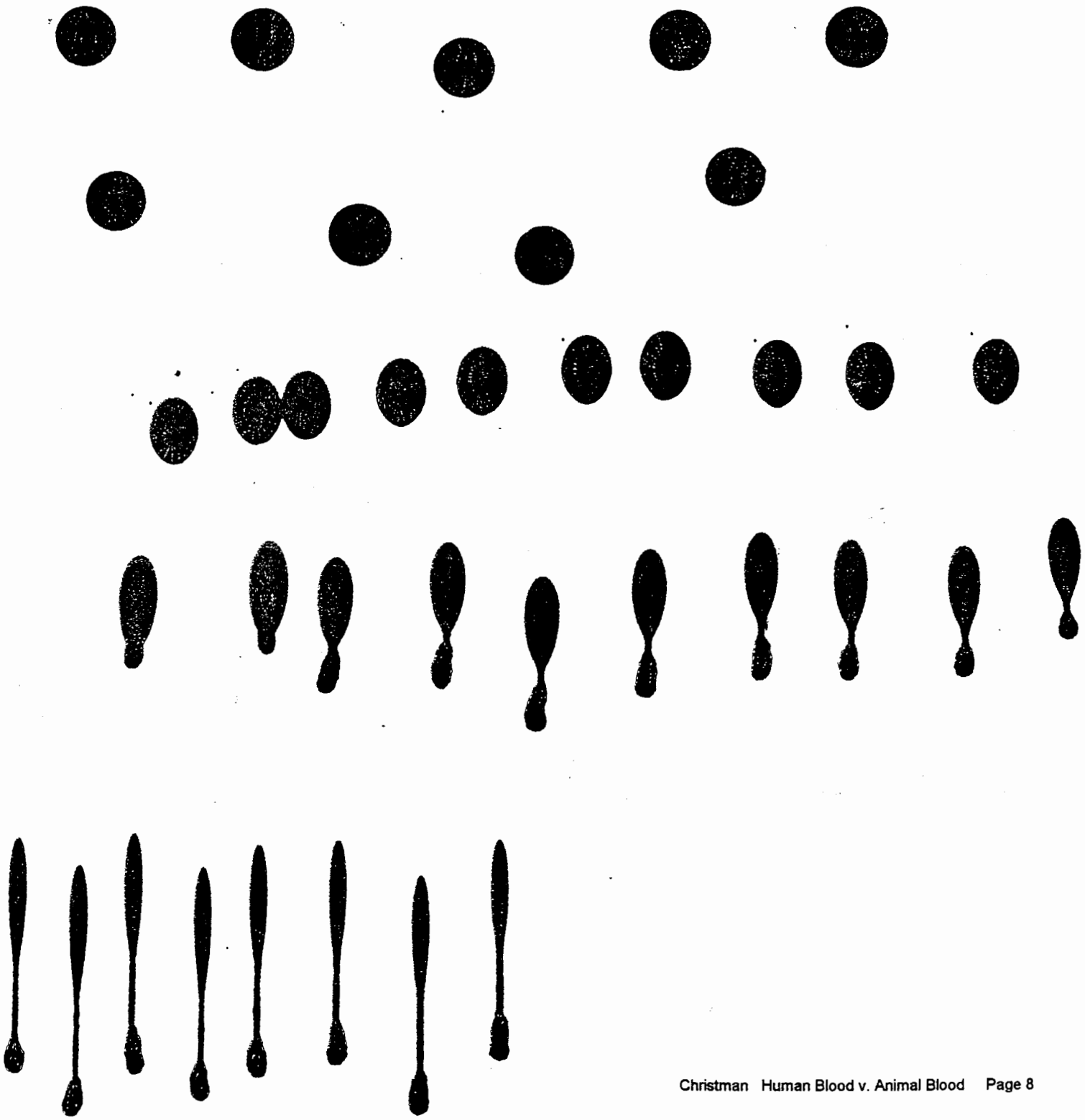






Reproducibility

Single blood drops of similar volume were dropped from similar distances onto paper targets. The plane of the targets were manipulated to different degrees so to create varying angles of impact including 90°, 80° 60°, 40°, and 10°. Fifteen drops of each type of blood were dripped onto the different targets and allowed to dry at room temperature. A random sampling of the drops were measured and calculated to determine the angle of impact. Reproducibility becomes an important point in analyzing the various drops. Studying only one or even a few bloodstains of each blood type would not take into account the possibility for variances, so multiple random bloodstains were sampled.



HUMAN - 10°			
	Width	Length	Angle
1	4	23	10.0°
2	4	24	9.5°
3	4	23	10.0°
4	4	21	10.9°
5	4	22	10.4°
Mean 10.16°			

BOVINE - 10°			
	Width	Length	Angle
1	5	27.0	10.6°
2	5	26.5	10.8°
3	5	26.0	11.0°
4	5	25.0	11.5°
5	5	24.0	12.0°
Mean 11.18°			

HUMAN - 30°			
	Width	Length	Angle
1	7	16	25.9°
2	7	18	22.8°
3	7	15	27.8°
4	7	15	27.8°
5	7	16	25.9°
Mean 26.04°			

BOVINE - 30°			
	Width	Length	Angle
1	7	16	25.9°
2	8	18	26.3°
3	8	18	26.3°
4	7	16	25.9°
5	7	15	27.8°
Mean 26.4°			

HUMAN - 50°			
	Width	Length	Angle
1	10	13	50.2°
2	11	15	47.1°
3	11	15	47.1°
4	10	14	45.5°
5	10	14	45.5°
Mean 47°			

BOVINE - 50°			
	Width	Length	Angle
1	8.5	11.5	47.6°
2	10.0	13.0	50.2°
3	10.0	13.0	50.2°
4	10.0	13.0	50.2°
5	9.5	12.00	52.3°
Mean 50.1°			

HUMAN - 90°			
	Width	Length	Angle
1	12	12	90°
2	12	12	90°
3	12	12	90°
4	12	12	90°
5	11	11	90°
Mean 90°			

BOVINE - 90°			
	Width	Length	Angle
1	11.5	11.5	90°
2	11.5	11.5	90°
3	11.0	11.0	90°
4	11.0	11.0	90°
5	11.5	11.5	90°
Mean 90°			

EQUINE - 10°			
	Width	Length	Angle
1	4	21	10.9°
2	4	22	9.5°
3	4	20	11.5°
4	4	21	10.9°
5	4	22	10.4°
Mean 10.82°			

SHEEP - 10°			
	Width	Length	Angle
1	4.5	22	11.8°
2	5.0	25	11.5°
3	4.5	23	11.2°
4	5.0	26	11.0°
5	4.5	25	10.3°
Mean 11.16°			

EQUINE - 30°			
	Width	Length	Angle
1	8.0	16.0	30.0°
2	7.0	16.0	25.9°
3	7.0	16.5	25.1°
4	7.0	16.0	25.9°
5	6.5	15.0	25.6°
Mean 26.5°			

SHEEP - 30°			
	Width	Length	Angle
1	8	19	24.9°
2	9	21	25.3°
3	8	20	23.5°
4	9	20	26.7°
5	8	20	23.5°
Mean 24.78°			

EQUINE - 50°			
	Width	Length	Angle
1	9.0	13	43.8°
2	9.0	13	43.8°
3	9.5	13	46.9°
4	9.5	13	46.9°
5	9.0	13	43.8°
Mean 44.42°			

SHEEP - 50°			
	Width	Length	Angle
1	11	15	47.1°
2	11	16	43.4°
3	10	14	45.5°
4	10	14	45.5°
5	10	12	45.5°
Mean 45.4°			

EQUINE - 90°			
	Width	Length	Angle
1	11.5	11	90°
2	11.0	11	90°
3	11.0	11	90°
4	11.0	11	90°
5	11.0	11	90°
Mean 90°			

SHEEP - 90°			
	Width	Length	Angle
1	12	12	90°
2	13	13	90°
3	13	13	90°
4	12	12	90°
5	13	13	90°
Mean 90°			

a cubic millimeter of human peripheral venous blood usually contains about 5.1 to 5.8 million erythrocytes in males and 4.3 to 5.2 million erythrocytes in females. Cattle maintain 6.0 - 10.5 million erythrocytes per cubic millimeter.

The proportion of erythrocytes in a sample of blood is called the *hematocrit* which is determined by first centrifuging a blood sample in a hematocrit tube until the erythrocytes are packed at the bottom of the tube and then measuring the ratio or percentage of the packed erythrocyte volume to the total sample volume. Although there is *considerable variability*, normal hematocrit samples of human peripheral venous blood average about 46.2% in males and 40.6% in females. The hematocrit of human arterial blood is generally slightly lower than venous blood, and the hematocrit of blood in the very small vessels of the body is considerably lower than blood in the large arteries and veins.

Normal Hematocrit Values

	Male	Female
Human	42% - 50%	40% - 48%
Equine	32% - 52%	N/A
Bovine	24% - 46%	N/A
Swine	24% - 50%	N/A
Sheep	35% - 45%	N/A

- **Leukocytes** (loo'-ko-sites) or white blood cells are also formed elements that defend the body against invasion by foreign organisms or chemicals and remove debris that result from dead or injured cells. Leukocytes are present within the blood in smaller numbers than erythrocytes; a cubic millimeter of human blood usually contains between 5000 and 10,000 leukocytes.

- **Platelets** (plate'-lets) are small cytoplasmic fragments that measure about 2.5 microns in diameter and number about 250,000 to 400,000 per cubic millimeter of human blood. Platelets play an important role in the process of blood clotting.

Some have debated the effects of things like the hematocrit on bloodstain patterns. Analysts have written papers and presented at conferences about how an individual's hematocrit effects the resulting bloodstain pattern. There is certainly no doubt that the more formed elements in a fluid, the thicker the fluid becomes. One type of ink used in this study contained glycerol and was so thick that it never dried. The resulting stains were still measured and studied. The length-width ratio was calculated and the angle of impact was verified even on this thick fluid.

SWINE - 10°			
	Width	Length	Angle
1	5	13	12.5°
2	5	25	9.5°
3	5	24	12.0°
4	5	24	12.0°
5	5	24	12.0°
Mean 12.0°			

INK - 10°			
	Width	Length	Angle
1	4.5	28.0	9.2°
2	4.5	28.0	9.2°
3	5.0	24.0	12.0°
4	4.0	25.5	9.0°
5	4.5	23.0	11.2°
Mean 10.12°			

Swine - 30°			
	Width	Length	Angle
1	8.0	17.0	28.0°
2	8.0	17.0	28.0°
3	8.0	17.5	27.2°
4	7.5	17.0	26.1°
5	8.0	17.0	28.0°
Mean 27.46°			

INK - 30°			
	Width	Length	Angle
1	10	21	28.4°
2	10	22	27.0°
3	10	20	30.0°
4	10	22	27.0°
5	10	21	28.4°
Mean 28.16°			

SWINE - 50°			
	Width	Length	Angle
1	11	14	51.7°
2	11	14	51.7°
3	10	13	50.2°
4	10	13	50.2°
5	10	14	45.5°
Mean 49.86°			

INK - 50°			
	Width	Length	Angle
1	14	19.0	47.4°
2	12	17.0	44.9°
3	11	16.5	41.8°
4	12	17.0	44.9°
5	12	12.0	44.9°
Mean 44.78°			

SWINE - 90°			
	Width	Length	Angle
1	12	12	90°
2	12	12	90°
3	13	13	90°
4	12	12	90°
5	12	12	90°
Mean 90°			

INK - 90°			
	Width	Length	Angle
1	13	13	90°
2	12	12	90°
3	13	13	90°
4	13	13	90°
5	13	13	90°
Mean 90°			

Discussion

Blood is defined as the *circulating tissue* of the body; the fluid and its suspended formed elements that are circulated through the heart, arteries, capillaries and veins; blood is the means by which 1) oxygen and nutritive materials are transported to the tissues, and 2) carbon dioxide and various metabolic products are removed for excretion. The blood consists of a pale yellow or gray fluid, plasma, in which are suspended red blood cells (erythrocytes), white blood cells (leukocytes) and platelets. *Stedman's Medical Dictionary*

All blood, whether animal or human is well described by the above definition. The species studied in this project have similar chemical and physical composition. Some animals, such as llamas, some birds and reptiles, have nucleated red blood cells and were not represented in this study. Because of their small body size, animal with nucleated red blood cells do not usually provide enough blood volume for experimentation. The blood types studied here contain:

Plasma

The liquid portion of animal and human blood, plasma, is comprised of about 91% water. The portion of the plasma that is not water, consists of various dissolved or colloidal materials, hormones, cellular products, and metabolic end products such as urea. The plasma also contains plasma proteins like albumins, globulins (some of which act as antibodies in immune responses and others that serve as transport molecules) and fibrinogen (important to blood clotting.) The plasma proteins act as buffers that help stabilize the pH and contribute to osmotic pressure and viscosity of the plasma. Various ions, including sodium (Na^+), chloride (Cl^-) and bicarbonate (HCO_3) are present in the plasma. The plasma also contains food materials such as carbohydrates (e.g. glucose), amino acids and lipids. Gases like oxygen, nitrogen and carbon dioxide can also be found in the plasma.

Formed Elements

The portion of blood that is not plasma consists of formed elements which include erythrocytes or red blood cells (RBC), various types of leukocytes, or white blood cells (WBC) and platelets.

- **Erythrocytes** (e-rith'-ro-sites) are small, circular, biconcave discs approximately 7-8 microns in diameter. Erythrocytes are the most numerous of the formed elements found in the blood and contribute to the total viscosity of blood. Although normal values are varied,

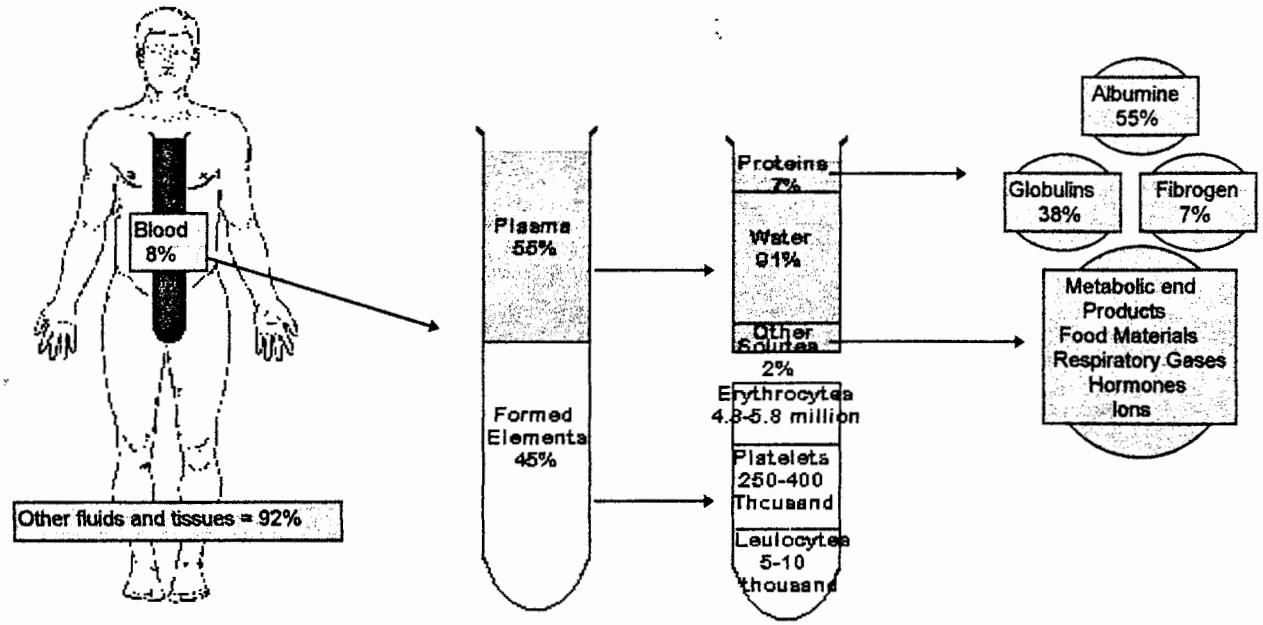


Figure 1 *The values indicated are approximate, and in many cases there are individual variations and or variations by sex.*

Other Variables

There are many forces that have an effect on blood. Some of these have a considerable effect while other are inconsequential. The crime scene reconstructionist will be concerned with those major variables that illicit a significant effect. Minor variables consist of states that effect negligible change and are essentially unnoticed by the bloodstain pattern analyst. The margin of error is insignificant when incorporating the crude method of measurement employed by some analysts. The major variables that are paramount to fluid dynamics and identified in this study are as follows:

- Gravity

Blood, like any fluid, is acted upon by gravity, that is blood respects the physical laws of gravity and reacts accordingly. Lividity, an internal bloodstain, is acted upon by the effects of gravity. Lividity patterns settle in the small vessels of the dependent areas of the body, secondary to gravity's draw. External bloodstains as flow patterns on a wall, are also created secondary to gravity and flow to dependent regions.

- Viscosity

All fluids have viscosity which cause friction. Roughly speaking viscosity is a measure of the fluids resistance to shear when the fluid is in motion. In simple terms a fluid is a substance which cannot resist a shear force or stress without moving, as can a solid. As expected, viscosity and fluid flow vary with temperature, which is evident from the old saying "slow as

molasses in January." A familiar application is the viscosity grade given to motor oil (SAE 30W, 40W, 50W).

The viscosity of blood is dependent upon hydration. Known viscosity values of animal bloods are not readily available because veterinarian studies have proven that hydration directly affects how viscous the blood will be. The values for human blood and some other fluids are listed below.

Viscosity of Some Fluids

Liquid	Pl	cP
Water	1.00×10^{-3}	1.00
Blood, Whole Human	1.2×10^{-3}	1.7
Blood, Plasma Human	1.7×10^{-3}	2.5
Alcohol, Ethyl	1.2×10^{-3}	1.2

- Surface Tension

Surface tension may be the most familiar characteristics to the blood pattern analyst because this phenomena defines the sphere shape of a blood droplet in flight. In the course of basic bloodstain pattern education, instructors explain that the bloodstain is spheroid shaped because of the effect of surface tension. The term surface tension is used to identify the apparent stress in the surface layer of liquid. This layer behaves *like* a stretched membrane and can give rise to a pressure difference across a curved liquid surface (that is an air-liquid interface.) A fluid will not support a shear stress, yet we have witnessed water bugs skate along the surface of a pond. A razor blade or a needle, carefully placed on the surface of a glass of water, will float even though steel is some eight times denser than water. Since the liquid surface behaves *like* a membrane, we see why a liquid may form a meniscus in a capillary, and why blood droplets, like raindrops, are more or less spherical. This membrane or surface tension of a blood drop, must be compromised by some force, energy or object in order to create blood spatter. Because the physical and chemical composition of human and animal blood is so similar, surface tension values in the liquids are equally similar.

Surface Tension Values

Liquid	Temp	Surface Tension
Water	20°C	0.073 N/m
Water	100°C	0.059 N/m
Whole Blood	37°C	0.022 N/m
Blood Plasma	37°C	0.072 N/m

**Blood values for animal and human.*

Conclusions:

Bloodstain pattern analysis is an empirical science, one based greatly upon the observation skills of the analyst. If a sloppy measuring technique is employed or if the person taking the measurements has little experience in measuring stains, the results can give false testimony in reconstruction of crime scenes. One thing that is sure, blood is a fluid and whether it's human blood, animal blood, or ink for that matter, there is a negligible difference in the results. In conducting controlled experimentation and studying the resulting stains it became apparent the fluids used were reacting in a similar fashion. In comparing the mean scores for those stains tested, all of the fluid stains compared favorably to the "standard," human blood.

Bloodborne Pathogens:

One of the issues brought up early in this project, initially by the blood bank personnel, was that of bloodborne pathogens. With the advent of the human immunodeficiency virus (HIV) and other bloodborne threats like hepatitis B, the bloodstain pattern analyst runs a real risk of contracting one or more of these diseases. Proper handling of blood is crucial when handling blood or bloody items. Animals and their blood are not excluded from bloodborne pathogen issues. Zoonotic diseases are those diseases transferred from animal to man. The following is a partial list of those diseases and their respective hosts.

Bovine	Equine	Swine	Sheep
Brucelosis	Brucelosis	Brucelosis	Brucelosis
Leptospirosis	Encephalitis	Leptospirosis	
Rabies	*Eastern	Aracipilis	
Bovine Leukosis	*Western		
	*Venezuelan		

Most of these diseases are hardy and will survive for long periods of time outside of their host environment. Slaughter house workers are exposed to more blood than the most active blood pattern analyst. Many of these workers choose not to wear protective equipment and according to the United States Food and Drug Administration, Investigative Bureau, no known incident exists where these workers have contracted a zoonotic disease. Interestingly enough one zoonotic scientist noted, the greatest chance of risk in the slaughter house is when workers are exposed to "misted blood" where high pressure water hoses are used to wash out an animal (similar to misted blood from gunshot wounds.) It is good to be concerned about bloodborne diseases and take appropriate measures to protect yourself. However, in the world of domesticated food animals, those diseases are few and many have been irradicated in the United States.