

Air Plasma Spray for First Aid

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Abstract

Hemorrhage during trauma occurred in emergency situations is a significant challenge. It may be life threatening if it is not treated swiftly. A new device which can effectively stop bleeding to save life of injured person, especially in battlefield situations and accidents, is presented. A plasma generator is designed to generate a low temperature air plasma spray for treating wounds. The spectral spike at 777.4 nm in the emission spectrum of the plasma plume and the spatial distribution of this emission line's spectral intensity indicate that abundant atomic oxygen is generated and sprays out of the generator by about 25 mm. Atomic oxygen carried by the plasma spray can quickly activate the cascading of coagulation processes and works as dry disinfectant to advance healing. Tests on blood droplets reveal the strong dependence of blood clotting on the amount of atomic oxygen applied in the plasma treatment, which is maneuvered by increasing the plasma treatment time or decreasing the exposure distance; in both approaches, the degree of blood clotting increases. Treated smeared blood samples show that an increase of the erythrocyte concentration and a drastic decrease of the platelet count are also correlated to the increase of atomic oxygen dose applied in the plasma treatment. The results reveal the mechanisms of air plasma blood coagulation and wound healing. As animal models, pigs were used in the tests of stopping wound bleeding from a cross cut in the ham area, from a hole in an ear's saphenous vein, and from cuts to arteries in an ear and in a real leg, all stopped swiftly. Moreover, both artery cuts were secure to remove tourniquet; downgrade of tourniquet necessary wound in under 2 minutes was demonstrated. The healing progress of cross cut wounds was observed. The healing time was shortened to about half. This battery power plasma spray can be carried to or placed at anywhere available for first aid applications. It stops bleeding swiftly to save life, and also downgrades tourniquet necessary wound to extend the golden period of saving the remaining part below tourniquet.

Keywords

Plasma Spray, First Aid, Coagulation, Wound Healing, Atomic Oxygen

1. Introduction

Bleeding, even from an external hemorrhage, may be life threatening if it is not treated swiftly [1]. New methods and devices which can rapidly stop bleeding, and so help to save the life of an injured person under emergency situations, especially in battlefield situations [2], are of significance.

Blood coagulation involves platelet activation and coagulation cascade. After wound is bleeding, the vasoconstriction is taken place and oxidants are released in the vascular lumen to enhance platelet agglomeration at the wound location to act on blood clotting. Moreover, oxidants fragment platelets to form a fibrin clot which keeps blood coagulation in homeostasis. Thus the treatment to speedup coagulation has to reduce the blood pressure at wound site to slow down bleeding, to clot injured blood vessels to stop bleeding, to heal the cause of bleeding and prevent complications, and to relieve symptoms [3] [4].

When atomic oxygen interacted with H_2O , similar oxidants to those released in the vascular lumen are generated. It suggests that atomic oxygen treatment could speedup blood clotting and clot formation and an atomic oxygen generator could serve purpose.

Plasma can effectively convert electromagnetic energy into kinetic energy of electrons, which is needed for exciting and dissociating molecular oxygen to produce chemically reactive oxygen species (ROS), including molecular oxygen in metastable states and atomic oxygen. Microwave discharge [5], high frequency inductive discharge [6], RF plasma jet [7] [8], or dc/low frequency capacitive arc discharges [9] [10] have been applied for plasma generation at atmospheric pressure. A gas flow is introduced to stabilize the discharge and to push ionizations out of the discharge region to form a plasma torch. A microwave plasma torch [5] has a rather high temperature. The inductive plasma torch [6] and non-transferred dc plasma torch [8] employ high current power supply and require very high gas flow rate to achieve stable operation. Consequently, the structures of these torch devices are relatively large, and the temperatures of the plasma torches are high.

For medical applications, low temperature non-equilibrium air plasma would be better usage of the electron kinetic energy gained from the electromagnetic sources for producing ROS, rather than for heating the plasma effluent. One such plasma generator [11] implements airflow and magnetic field to rotate elongated arc discharges. It functions as a blood coagulator by using atomic oxygen, carried in the plasma effluent to activate erythrocyte-platelet interactions, to stop blood bleeding from external wounds [12]. Chen *et al.* [13] and Kuo *et al.* [14] showed that this air plasma spray, carrying significant amount of atomic oxygen, could clot anti-coagulated whole blood samples in less than 20 seconds, which is much less than 30 minutes for an untreated sample to reach complete coagulation. Using pigs as the animal model, air plasma bleeding control was demonstrated [15] [16].

In the present work, a portable device which generates air plasma spray for first aid is described in Section 2. The coagulation and healing mechanisms of the plasma treatment are discussed in Section 3. In Section 4, the efficacy of the air plasma spray to stop bleeding is demonstrated. Summary and conclusion are presented in Section 5.

2. Plasma Spray Generator

In plasma coagulation applications, it is desirable to have plasma 1) generated in open air with large spatial extent, 2) carrying a significant amount of free radicals, such as reactive oxygen species, and 3) non-equilibrium at low thermal temperature. In the following, the design of a plasma generator which produces an air plasma spray meeting these conditions is described.

2.1. Arc Discharge Module and Air Plasma Spray

Shown in **Figure 1** is a schematic of a discharge module, which consists of a pair of concentric electrodes, a cylindrical frame, and a position holder [11]. The central electrode, a cylindrical copper (or tungsten) rod of diameter "d", is inserted through the ring shape outer electrode of an inner diameter "a" and through a tight fit position holder keeping the central electrode along the central axis of the cylindrical frame. This holder has large openings to permit the airflow entering from the bottom of the cylindrical frame to pass through it. The size (diameter) of the module can vary with the specific application. The gap between the electrodes is in the range of 0.8 to 1 mm.

At atmospheric pressure, the air discharge is an arc mode, which generally evolves into a constricted arc and develops hot spots on the electrode surfaces. An airflow with flow rate of ~1.5 ℓ /s at gap and a nozzle are introduced to elongate and rotate the discharge path to prevent arc constriction and hot spot formation. The nozzle also works to spread out the plasma plume, as well as to cover the high voltage (HV) central electrode for safety; it increases the size of the plasma plume which expands from the exit of the nozzle by about 25 mm (*i.e.*, about 35 mm away from the gap). A significant increase of the discharge path length works to reduce the arc temperature and the sparkling and heating of the electrodes. It also increases the generation region of the reactive species, which is particularly important to those with short lifetimes, such as atomic oxygen.



Figure 1. A schematic of a plasma generator module.

2.2. Power Supply

The air breakdown voltage at atmospheric pressure is about 3 kV/mm. As the discharge is initiated, the current increases rapidly to cause a sudden drop of the voltage which is the signature of arc discharge having a negative V-I characteristic. It is found that the voltage maintaining the discharge in the 1mm gap between the electrodes can be much lower than 350 V. Thus, a high voltage (HV) induction (ignition) coil can be used to trigger [17] and to control the timing of the discharge. With the aid of a trigger, two types of portable and light weight power supplies, one with 120 VAC input and one with 12 VDC input, are made to run a handheld plasma spray as shown in **Figure 2**. The battery powered plasma spray can be used in open fields where an AC outlet is not accessible.

2.3. Emission Spectroscopy

The presence of atomic oxygen in the plasma spray was examined via its emission spectroscopy [18] [19]. At atmospheric pressure, 777.4 nm and 844.5 nm lines are commonly found in non-equilibrium air discharges, where 777.4 nm lines are usually more intense than 844.5 nm lines. O'Connor *et al.* [20] developed a method using the intensity ratio I_{844}/I_{777} to determine the discharge gas temperature. In a scan of the emission spectroscopy as shown in **Figure 3(a)**, the 777.4 nm lines have a dominant spectral peak and the 844.5 nm lines are missing; it indicates that the discharge gas has low



Figure 2. A handheld plasma spray.



Figure 3. (a) Photon counts of the emission lines from 700 to 900 nm, the arrow is located at 777.4 nm; and (b) spatial distribution of 777.4 nm radiation intensity of the plasma spray.



temperature. The 777.4 nm line is attributed to the 5P state of the atomic oxygen (OI). Presented in **Figure 3(b)** is the spatial distribution of the 777.4 nm radiation intensity ranging from $<10^4$ to $>10^9$ (in Rayleighs). The distribution extends out axially to about 25 mm from the nozzle of the module, where the intensity of the 777.4 nm line is about 10^5 R. Thus the apparent photon emission from a slice of the plasma plume at 25 mm away from the cap is about 10^{15} m⁻²·sec⁻¹, where the calculation assuming low optical thickness represents a minimum bound on the average flux.

The thermal temperature (<320 K) of the plasma effluent is much lower than the excitation temperature of electrons channeled in the arc loop, which is estimated via emission spectroscopy to be higher than 7700 K. This plasma spray is non-equilibrium due to the following factors associated with the design and the operation, 1) the discharge is run in a periodic mode with a duty cycle less than 10%, 2) the airflow pushes the discharge along an elongated path, 3) the elongated arc loop is vibrating by the air pressure in the nozzle, 4) the triggered discharge is maintained at low voltage. Because the discharge extends to 30 to 35 mm away from the discharge gap, some of the energetic electrons and OI are generated locally in the plasma plume, rather than being conveyed from the gap region of the electrodes.

3. Air Plasma Blood Coagulation and Wound Healing Mechanisms

Blood is a fluid tissue that includes 60% of a liquid portion known as blood plasma, and 40% of formed elements or blood cells [21]. Blood plasma, a protein-salt solution up to 95% of water by volume, suspends red blood cells (RBC), white blood cells (WBC), and platelets alike. It contains albumin (the chief protein constituent), fibrinogen (responsible in part for blood clotting), globulins (including antibodies) and other clotting proteins [21]. Formed elements consist 86.6% of RBC, 10.4% of platelets, and 3% of WBC. RBC contains hemoglobin, a complex iron-containing protein that carries oxygen and participates in carbon dioxide exchange. Platelets plays a vital role in the early response to vascular injury; at wound site, platelets become activated, agglomerate, and forming blood clots adhering to injured blood vessel wall components; it also secretes mediators that attract WBC [22]-[24]. WBC consists of neutrophils, eosinophils, basophils, monocytes, and lymphocytes [21]. The average lifetime for WBC is hours to days, for RBC 120 days, and for platelet 9 days. Platelets are more fragile cells and 3 - 4 times smaller than RBC [21] [25] [26].

Both formed elements and blood plasma contribute to blood coagulation during hemorrhage. Blood coagulation involves platelet activation and coagulation cascade. When the platelets encounter the break situation of the blood vessel, external molecules touching the platelets trigger platelet activation; it is followed by the coagulation cascade, which is a complicated step-by-step blood clotting process. Several proteins (fibrinogen, tissue factor, calcium, etc.) and molecules, called coagulation factors, play important roles in the coagulation cascade. In the following, the role of atomic oxygen in plasma treatment to speed up blood coagulation and wound healing is explored.

3.1. In-Vitro Tests of Air Plasma Blood Coagulation

Blood samples used in tests were mixed with 3.2% sodium citrate solution at 9:1 ratio (in volume). Each sample was set on a glass slide. The sodium citrate solution is a commonly used reagent to prevent premature blood coagulation, it chelates calcium ions to prolong the natural clotting time [27] [28] to more than 25 minutes.

The effects of heat and atomic oxygen radical on blood clotting were studied and compared by tests on blood droplet samples [13]. Test of heating effect was performed by treating a blood droplet set in a well with a hot airflow of a hair dryer for 16 sec; it raised the sample temperature from 20°C to about 61°C. A photo of the sample taken after a hot air treatment is presented in Figure 4(a). No noticeable blood clot can be identified.

The atomic oxygen flux carried by the plasma spray decreases exponentially in distance, the total amount of atomic oxygen applied to the blood droplet sample in treatment will be proportional to the total exposure time and inversely proportional to the exposure distance. Hence, the plasma treatments on blood droplet samples at different exposure distances and durations can reveal the dependency of coagulation cascade on the applied atomic oxygen flux.

Three samples treated at the same exposure distance of 25 mm for 8, 12, and 16 sec, respectively, are presented in Figures 4(b)-(d) for comparison. The sample temperatures were less than 55°C. A shell, formed on each blood sample surface, can be clearly seen. The photos indicate that the degree of blood clotting increases with the increase of the exposure time from 8 to 16 sec. Three samples treated at three different exposure distances of 25, 30, and 40 mm for the same exposure time of 16 sec are presented in **Figures 4(d)-(f).** Again, the sample temperatures were less than 55° C. As shown, the degree of blood clotting decreases as the exposure distance increases from 25 to 40 mm. These results deduce that the degree of blood clotting increases with the increase of the atomic oxygen flux delivered by the plasma spray.



Figure 4. Blood samples treated (a) by a heated airflow; by a plasma spray at a fixed exposure distance of 25 mm with three exposure times of (b) 8 sec; (c) 12 sec; and (d) 16 sec; and by a plasma spray with a fixed exposure time of 16 sec at two increased exposure distances of (e) 30 mm and (f) 40 mm.



3.2. Tests on Smeared Blood Samples-Cell Count Dependency

Untreated (control) and plasma spray-treated smeared blood samples were prepared for cell staining and microscopy analysis, which identified cell types and performed cell counts [14]. The results of cell counts from samples treated with four different exposure times of 2, 3, 4, and 10 s and two different exposure distances at 25 mm and 40 mm are presented in **Figure 5**. As shown, at a fixed exposure distance of 25 mm, the concentration of RBC/platelets increases/decreases monotonically as the exposure time increases. Cell counts of RBC and platelets with 10 s exposure time at 25 mm and 40 mm exposure distances were compared. As shown, the treatment at longer exposure distance causes less changes on the concentrations of RBC and platelets.

The correlation between the rapid reductions of the platelet counts and clot formation time (presented in Section 3.1) of a treated sample suggests that atomic oxygen, delivered by the plasma spray, rapidly induce oxidants in blood plasma to fragment platelets for forming blood clot.

3.3. Mechanism of Air Plasma Blood Coagulation

The results of the tests on blood droplet samples presented in **Figure 4** and the microscopy analysis of smeared blood samples presented in **Figure 5** correlate the speedy coagulation, the increasing of RBC, and decreasing of platelet counts, to the increase of atomic oxygen flux in the treatment. It suggests that atomic oxygen carried by the plasma effluent rapidly trigger coagulation cascade.

When interacted with H_2O , atomic oxygen can generate large amount of reactive oxygen species (free radicals and hydrogen peroxides), which are expected to function similarly to those oxidants, produced or released in the vascular lumen. Several key steps in coagulation cascade are then triggered by the oxidants. These oxidants stimulate RBC-platelets and WBC interactions. The interactions influence the concentration



Figure 5. Changes of RBC and platelet concentrations of plasma-treated sample with the increase of the exposure time from 3 to 10 sec in the cases of two exposure distances at 25 and 40 mm.

of cells suspended in blood; the observations of the microscope presented in Figure 5 show the increase of RBC concentration in line with the increase of the exposure time. Enhanced adeninenucleotides released by aggregated RBC trigger platelet adherence/ agglomeration [29]-[36], where fibrinogen acts as a bridge to link activated platelets together. Subsequently, globular complexes are formed to trap RBC and platelets; the viscosity of blood samples will also be affected by oxidants that presumably contribute to albumin denaturation [37] as well as other proteins found in the blood. Consequently, the blood flowability is decreased and coagulation is rising. Additionally, platelets are fragmented by oxidants to induce thrombin, which converts the soluble fibrinogen, large and complex glycoprotein, into insoluble fibrin strands to form fibrin gel holding activated platelets to form homeostatic plug.

3.4. Air Plasma Wound Healing Mechanism

The role of atomic oxygen in wound healing process is explained as follows. Hypoxia [38] acts a key factor to stimulus tissue repair by creating an oxygen gradient from the hypoxic tissue of wound to the nearby unbroken tissue [39]. The central area of the wound is most hypoxic, and the oxygen gradient increase toward the uninjured tissue progressively. With the supply of atomic oxygen from the plasma spray, more oxygen provided in respiratory burst can be shared in other action such as producing superoxide (SOD), cell metabolism and raising tissue oxygen tension in the wound healing. Atomic oxygen also provides the oxygen in the blood by reaction of catalase which plays a protection role avoiding cells damaged by H_2O_2 . More significantly, atomic oxygen together with generated oxidants work for disinfection concurrently with blood coagulation in the wound treatment. Thus the period of the inflammatory phase in wound healing is shortened. Harman [40] points out that cells are aging via accumulating free radical and oxidative damage over time. In order to maintain the normal function, skin tissue is metabolized when it is aging. Because the treated area accumulates sufficient atomic oxygen, this area is aging faster than surrounding tissue. The skin tissue increases the metabolism to speed up the generation of new tissue. The new skin tissue grows under the ageing tissue, and replaces the position after aging tissue is peeled.

3.5. Discussion

The experimental results presented in this work are consistent with the published data on the effect of oxidants on blood coagulation. Therefore, the clotting process is explained to be attributed to the stimulation of RBC-platelets and/or WBC interactions by the oxidants created during the interaction of blood and atomic oxygen (as well as other likely ROS). It may be further confirmed by future studies, focusing on how the atomic oxygen (and ROS) carried by the plasma spray influences each individual cell type; the samples will be single cell type, either RBC, WBC, or platelets, as well as blood plasma, in order to verify their actual involvement in blood clotting, blood coagulation, and fibrinolysis.



4. Blood Coagulation and Wound Healing with Animal Models

Two 3-month-old male pigs weighing around 25 kg [15] and two 6-month-old male pigs weighing around 40 kg [16] were used to explore the coagulation efficacy of the air plasma spray; the two smaller pigs were used in the experiments of cross cut wound and hole in a saphenous vein; and two larger ones used in artery cut and in the observation of the healing progress of a cross cut. The wound introduced on one pig was treated by the plasma spray; the exposure distance was fixed at 25 mm except in the case of cross cut for healing observation, the exposure distance was fixed at 30 mm.

4.1. Demos of Air Plasma Blood Coagulation

Row (a) of **Figure 6** demonstrates a cross cut case; a scalpel was used to make a cross cut, which consisted of two straight cuts cross to each other, in the ham area. The size of each straight cut was about 10 mm in length and 5 mm in depth. The cut was then treated by the plasma spray continuously; a ruler is attached to the plasma generator to fix the exposure distance at 25 mm. After 13 s of the plasma treatment, bleeding was stopped as shown. The other pig was an untreated control whose wound bleeding was stopped naturally. The bleeding lasted for more than 4 minutes. As the exposure distance increased to 30 and 40 mm, the needed treatment times also increased to 17 and 22 s, respectively.

A saphenous vein from a pig ear was first identified. As shown in Row (b) of **Figure 6**, a needle was used to punch a hole in this vein; when blood flow started, it was treated immediately by the plasma spray which stopped bleeding in 15 s; the hole in the vein was sealed completely. The plasma treatment may irritated the exposure area around the wound, causing skin reddish as shown; but this irritation disappeared in two days and no apparent side effects were observed. In the photos, the circular orange plate was an ID tag. The bleeding time of the untreated control in the other pig was measured to be about 88 s.



Figure 6. Photos showing (row (a)) cutting by a scalpel, treating this cross cut for 13 s, and bleeding-stopped cross cut wound; (row (b)) punching a hole in an ear's saphenous vein, plasma treatment for 15 s, and hole is sealed; (row (c)) cutting an ear artery, plasma treatment for 12 s, and cut is sealed.

Before cutting an artery in an ear with a scalpel, the ear was tied with a tourniquet, shown in Row (c) of Figure 6, to reduce the blood pressure. The plasma spray was run with 2-s on/4-s off alternately. Bleeding was stopped after 6 runs of plasma on-off treatment. The total plasma treatment time was 12 s, though the total time spent in this treatment was 32 s. The cut to the artery was secure even after tourniquet was removed. In the untreated case, it took 1 minute to stop the bleeding naturally and the cut was not secure without tourniquet.

Subsequently, a large scale test was performed by cutting an artery above the middle joint of a real leg. Tourniquet was applied to stop high-pressure arterial bleeds. The cut was treated by the plasma spray continuously for 60 s at an exposure distance of about 15 mm. Hemorrhage was controlled and tourniquet was removed. Manipulation of limbtotry to break clot was played, but wound did not re-bleed. This is the first demonstration of successful down grade of tourniquet necessary wound in under 2 minutes.

The plasma treatment time and the natural time (control) used to stop bleeding from each type of the wounds cut in the tests are recorded in **Table 1**.

4.2. Demo of Air Plasma Wound Healing

In wound healing observation, cross cut wounds were introduced in the ham area of two larger pigs; again, each cut was 10 mm in length and 5 mm in depth; one was untreated as a control and the other one was treated by the plasma spray with an intermittent exposure approach, by running the spray with 2-s on/2-s off alternately for 5 times.

The progress of the control (untreated cross cut) shown in the first row of Figure 7 indicates that the scab started peeling in the 8th day. The scab was diminishing in time; however, a small piece of the crust still remained in the wound area in the 14th observation day. The healing time of the cut treated with 10 s plasma exposure was found to be shortened. The scab started peeling in the 4th day, and the crust disappeared completely in the 8th day. The observation reveals that this air plasma spray has a positive impact on wound healing; it shortens cross cut wound healing time to about half.

Table 1. Bleeding control time of air plasma treatment.			
	Treatment		
		Plasma	Plasma
Wound	Plasma	(intermittent)	(intermittent)
	(continuous)	2-s on/4-s off	2-s on/2-s off
		On time	On time

18 s

13 s

15 s

60 s

8 s

8 s

12 s

Straight cut $(1 \text{ cm} \times 0.5 \text{ cm})$

Cross cut $(1 \text{ cm} \times 1 \text{ cm} \times 0.5 \text{ cm})$

A hole in an ear's saphenous vein

Artery cut in ear (tourniquet aided)

Artery cut in rear leg (tourniquet aided)



10 s

10 s

Natural (control)

190 s

240 s

88 s

60 s



Figure 7. Healing progress of untreated and plasma treated cross cut wounds in 14 days period.

4.3. Discussion

Further tests on wounds of artery cuts at different locations will be useful to establish the treatment procedure in the first aid. In future studies, it is also of interest to test the feasibility of air plasma spray to stop bleeding from artery wounds which cannot apply tourniquet. A surgical clamp may be used.

5. Summary & Conclusion

Blood coagulation and wound healing mechanisms by an air plasma spray are presented. Tests on blood droplets established a strong dependence of the blood clot on the atomic oxygen density delivered by this plasma spray. The microscope study of treated smeared blood samples correlates the decrease of the platelet count and the increase of the RBC count to the increase of the atomic oxygen flux in treatment. These correlations evidence the role of atomic oxygen flux in speeding up blood coagulation. When atomic oxygen interacts with H_2O , reactive oxygen species such as OH and H_2O_2 are produced. These oxidants target platelets to affect several key steps of platelet function, such as enhancing platelet aggregation and fragmenting platelets to form clotting surface for the subsequent steps of the coagulation. In addition, atomic oxygen enhances superoxide (SOD) and cell metabolism and raises tissue oxygen tension in the wound healing. It also provides disinfection to shorten the period of the inflammatory phase in wound healing.

Using pigs as animal models, rapid stop of wound bleeding by this air plasma spray was demonstrated. The tests also successfully demonstrate to downgrade tourniquet necessary wound in under two minutes. The consequence of plasma treatment on wound healing was also demonstrated. A single plasma treatment to stop bleeding of a cross cut wound also reduces the wound healing time to about half.

In conclusion, this air plasma spray has demonstrated its potential as an advanced first aid device. This handheld spray is designed to be run with two types of power supplies, which are portable and light weight. One power supply employs 120 VAC input and the other one with 12 VDC input. Both can be implemented in emergency rooms, ambulances, vehicles, and buildings. Moreover, the battery power plasma spray contains its own power source. It can be carried to or placed at anywhere available for first aid applications.

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