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Cold atmospheric pressure plasma for treatment of chronic wounds: drug or medical device?

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### Journal of Wound Care

**Cold atmospheric pressure plasma for treatment of chronic wounds: drug or medical device?**

---Manuscript Draft---

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<td>Axel Kramer, MD, PhD Barbara R Conway, PhD Konrad Meissner, MD, PhD Fritz Scholz, PhD B H Rauch, PhD Anke Moroder Alexander Ehlers Alfred J. Meixner, PhD Claus-Dieter Heidecke, MD, PhD Lars Ivo Partecke, MD Manfred Kietzmann, PhD Ojan Assadian, MD, DTMH</td>
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I would be grateful if you could highlight the changes you have made to the text in yellow (not using tracked changes) and include a summary of the revisions you have made when you resubmit your article.

Reviewer #1

C1. In Germany, the society for plasma-medicine has recently undertaken activities to implement a standard for regulation of plasma devices, the DIN SPEC 91315. It is recommended to include this in the discussion.
A1: We sincerely thank the respected reviewer for pointing out to this important standard. We have included now a brief statement highlighting the DIN SPEC 91315, and we have added the standard’s reference to the reference list. The added paragraph summarises the intention of the standard as follow: “In this respect and as a first step, the technical standard DIN SPEC 91315:2014-06 was developed, which characterizes the basic physical and technical performance parameters of CAP sources to be used for bio-medical or biological experiments and for further development to become medically applicable plasma sources.”

C2. Moreover, wound therapies are in general hard to place, as for instance they affect biochemical compositions of the wounds. Hence, it might be time to review our approach to medical devices in the wound care sector in general.

A2: We completely agree with this important remark. The authors discussed about adding further thoughts about this aspect, however, we had to acknowledge that a brief highlight of this matter would be insufficient, as it may leave too many legal, medical, and regulatory aspects unexplained, and a full discussion on the difficulties with current registration procedures, particularly for wound management devices, would inflate the manuscript inappropriately. Therefore, we kindly ask the respected reviewer not to elaborate on this aspect and keep the topic for a separate manuscript in future.

Reviewer #2

C1. This manuscript has a large number of references, often with 4 or 6 supporting each statement. In reviewing titles of the references and searching for abstracts and papers, it appears that a number of these references may not be primary sources, but are perhaps reviews that have referenced other primary sources supporting the statements. Where definitive statements are made, this reviewer recommends that primary sources should be cited. An example, among others, is the section in Background covering mutagenicity, supported by references 53-56. 53 and 54 have, respectively, the following titles: "Hydroxyl radicals attack metallic gold" and "A solid state redox buffer as interface of solid-contact ISE - a strategy to improve the reproducibility and stability of potentials". These do not appear to be primary sources for the scientific data on mutagenicity. Reference 57 also does not appear, based on the abstract, to be a primary source of data on "the latter was as result of directly exposing DNA". Reference 58 does not appear, based on the abstract, to refer to coagulation but rather it refers to skin disinfection and the thermal aspects of this. Reference 59 is a report of an in vitro study on plasma, ozone and hydrogen peroxide using a device that is identified by reference to previous papers (refs 22 and 34) which does not appear to support the assertion that this is a plasma skin regeneration device. The device is not identifiable; it should be identified and supported by clinical references if they are available. Reference 60 is not the MEDDEV Borderline Guideline. Reference 61 is not a description of the decision by the European Court. The references cited stop at 61 in the references list but the numbering in the manuscript continues up to 90.

A1: We thank the respected reviewer for pointing out the many mistakes with the reference list. Indeed, we are inconsolable about this inexcusable error! After re-checking the references, we have noticed that the complete reference list was muddled up with previous versions. Regretfully, it seems that multiple versions of the manuscript and inadequate use of the automated reference manager software have resulted in chaotic references. We have completely re-arranged the references, which should be all corrected now. Again, we whole heartedly apologize for the inconvenience caused.

Additional Information:

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Cold atmospheric pressure plasma for treatment of chronic wounds: drug or medical device?

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Declaration of interest

None of the authors have any competing interests.

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Abstract

Background. The use of cold atmospheric pressure plasmas (CAPs) as a new therapeutic option for chronic wounds appears to be promising. Currently, uncertainty exists regarding their classification as medical device or medical drug.

Objective. Because the classification of CAP has medical, legal, and economic consequences as well as implications for the level of pre-clinical and clinical testing, the correct clarification is not an academic debate, but an ethical need.

Method. A multidisciplinary team of researchers and lawyers has analyzed the physical and technical characteristics as well as legal conditions of the biological action of CAP.

Results. It is concluded that the mode of action of the locally generated CAP with its main active components, being different radicals, is pharmacological and not physical in nature.

Conclusion. Depending on the intended use, CAP should be classified as a drug, which is generated by use of a medical device directly at the point of therapeutic application.
Background

Highly energetic physical plasmas comprise a mixture of reactive ionized particles, which can be physically adjusted to body temperature by mixing protons, resulting in so-called "cold plasma". Cold atmospheric pressure plasmas (CAPs) are generated under atmospheric pressure at ambient temperatures ranging from 20°C to 50°C.\textsuperscript{1,2}

Interest in medical applications of CAPs is rapidly increasing. CAPs ignited a technological spark in industry, biotechnology, and in the field of hygiene, for indications where the use of antimicrobial active agents or heat sterilization is limited. This includes decontamination of thermo-labile medical devices, food, packaging materials, waste-water, or indoor air.\textsuperscript{3} For such applications, the question does not arise whether CAPs are a drug or a medical device.

On the other hand, plasma has shown promising antiseptic results on skin and mucosal membranes in infection-related diseases in dermatology\textsuperscript{4-6} and dentistry\textsuperscript{7}, and has also been found to exert anti-carcinogenic effects similar to those of oxygen radicals, mainly by dose-dependent anti-proliferative\textsuperscript{8} and apoptotic activity.\textsuperscript{9-13} First reports on the use of argon plasma for tumor removal date back to 1989\textsuperscript{14}, followed by successful ablation of non-neoplastic Barrett's mucosa\textsuperscript{15} and other neoplastic diseases.\textsuperscript{16-21} A plasma-activated medium has even been claimed to exert anti-tumor effects.\textsuperscript{22,23}

Results of in-vitro,\textsuperscript{24} in-vivo\textsuperscript{24-28} and initial clinical studies on chronic wounds of domestic animals\textsuperscript{29,30} and humans\textsuperscript{31-35} indicate that CAP may be a promising alternative to conventional treatment options, based on the following biological premises:

- A wound cannot heal as long as it is infected\textsuperscript{36}. CAP may support wound healing through its antiseptic efficacy in vitro\textsuperscript{37-39}, ex vivo\textsuperscript{40}, in vivo\textsuperscript{41} and in humans.\textsuperscript{31,32,42-44}

- CAP may facilitate the transformation of the chronic wound from a stagnating wound to an acute healing wound, i.e. by inflammatory\textsuperscript{45,46} and proliferation-supporting stimuli \textsuperscript{47,48}, including stimulation of neovascularization.\textsuperscript{29,49,50}

However, while the medical applications of CAP are promising, it is vital to ensure the safety and reliability of applied CAPs in humans before it may be considered for routine clinical
practice. Therefore, aside from the clinical effectiveness, the safety for patients and users is an important consideration during the registration and approval processes with health authorities. Based on the intended use and the potential risks associated with a medical intervention, be it supportive, diagnostic, or therapeutic, products may be classified as medical devices, or medical drugs.

In this respect and as a first step, the technical standard DIN SPEC 91315:2014-06 was developed, which characterizes the basic physical and technical performance parameters of CAP sources to be used for bio-medical or biological experiments and for further development to become medically applicable plasma sources. However, findings on mutagenicity and transient expression of pre-mutagenic active compounds, even though the latter was as result of directly exposing DNA, suggest that the current risk assessment for plasma applications is still incomplete and there is a need to consider also energy, penetration depth, and the body’s detoxification capacity must also be considered. In addition to classification issues, both pre-clinical and clinical investigations need to be preceded by tests of clinical effectiveness and the exclusion of chronically toxic, mutagenic, and carcinogenic risks.

The final decision on the correct classification of CAP has not only medical, legal, and economic consequences, but also fundamental implications for the required level of pre-clinical testing, and future development of this new technology. To date, the final classification of CAPs as a medical drug or medical device has been the subject of academic debate, resulting in uncertainty on the required test data for registration with the respective health authorities in Europe and the US, product liability, manufacturers’ responsibility, and future economic strategies. Therefore, a multidisciplinary team of physicians, surgeons, pharmacists, physicists, and lawyers familiar with the medical application of CAPs has critically evaluated the requirements for the classification of cold atmospheric pressure plasmas as a medical drugs or medical devices from different perspectives.
Differentiation between drug and medical device

In Europe and the US, medical drugs and medical devices are regulated by separate laws, which cannot be applied simultaneously. Dual labeling is generally not accepted by drug administration agencies and thus entities are regulated by either the medical devices or medicines legislation, but not both. Historically, medical drugs were distinguished from medical devices by their pharmacological, metabolic, and/or immunologic effects, while the mode of action of medical devices was predominately based on physical aspects.

Furthermore, based on the potential risk associated with the use of medical devices, they can be distinguished into 3 classes. Class I medical devices are defined as devices without risks to humans due to low invasiveness, temporary use (≤ 60 min), and application with non-critical skin contact. Class IIa medical devices bear potential risks as they are associated with moderate invasiveness, short-term uninterrupted or repeated use (≤ 30 d) within the human body (ophthalmological, intestinal, or surgically created body cavities). Class IIb medical devices have potential elevated risks, elicit systemic effects, or are intended for use beyond 30 days (i.e. non-invasive birth control). Finally, class III medical devices have the highest potential risks, e.g. technical solutions for long-term drug application, pacemakers, implants, or invasive birth control devices.

However, the intention of preventing or treating illnesses may apply to both medical drugs and medical devices, and therefore does not qualify as a distinguishing characteristic between the two. Thus, the only reason to objectively differentiate between a drug and a medical device is its pharmacologic, immunologic or metabolic effect, compared to the physical effect of the medical device, which may merely support pharmacologic, immunologic or metabolic effects. Pertaining to CAP devices, their categorization as drugs or medical devices therefore depends solely on their mode of action. For example, surgical CAP devices used for cutting or coagulating tissue in humans (so-called “beamers”) or plasma skin regeneration devices, were previously approved as medical devices class IIb, based on their physical (thermal) mode of action.
In contrast, the MEDDEV-Borderline guideline\textsuperscript{58} defines a pharmacologic effect as an interaction between molecules of the substance in question and a cellular component, such as a receptor, which either elicits a direct response or blocks another one in response to a third agent. The view therefore does not specifically demand an interaction of substance molecules with “cellular components of the host”, but merely requires “cellular components” only. A recent decision of the European Court \textsuperscript{59} supports the definition of a pharmacologic effect of a substance as an interaction with any cellular components within the host’s body, including foreign targets cells like bacteria, viruses, or parasites. This opinion is also supported by the European directive 2004/27/EG, a revised version of the directive 2001/83/EG section 2, subsection 2, which elevates the MEDDEV-Borderline guideline\textsuperscript{58} to the instrument of choice in cases of uncertainty for defining a new technology.

However, where pharmacological aspects outweigh the physical therapeutic use of CAP, its classification requires a more complex consideration. In the case of CAP, the transferability of obvious physical and biological effects between different CAP devices is complex due to various potential mixtures of reactive species and the application of energy during plasma application.

**Physical and chemical characteristics of CAP**

The different available technologies relevant for plasma medicine can be categorized either as direct or indirect physical plasmas. In the case of direct plasmas, the patient’s body serves as the second electrode through an electric voltage field between the head of the device and the skin surface to be treated, i.e. dielectrically impaired discharges (DBE).\textsuperscript{60,61} Indirect plasmas (plasma jets) arise between two electrodes, e.g., within a hand-piece, and are conveyed outward by an intense gas stream.\textsuperscript{1,62} Depending on the type of carrier gas (i.e., argon or helium containing atmospheric pressured ambient air plasma), the energy supply for plasma generation and the type of plasma generation, the plasma may appear as a visible flame-like beam from a nozzle.\textsuperscript{1} In both directly and indirectly generated plasma, the plasma contains charged particles (electrons, ions), excited atoms and molecules (i.e.,
singulett-oxygen), free radicals (atoms or molecules containing an unpaired electron), photons and electromagnetic fields, leading to the emission of visible UV or VUV radiation. The chemical composition and the physical characteristics of the generated plasma significantly depend on a number of variables such as pressure, gas mixture, design of the device, physical stimuli, and others. The reactive compounds, which become biochemically active, emerge either during the generation of the plasma in due course of interaction with molecules of the surrounding air, and/or with the medium, the bodily fluid, or the tissue to be treated.  

In addition to the direct plasma effects due to the composition of the CAP at the effect site, there are also subsequent secondary actions within the tissue, based on radical formation generated by CAP, similar to physiologically generated radicals. Hydroxyl radicals (•OH) may act as second messengers in T-cell activation, and induce apoptosis. Nitrogen monoxide (NO) acts as a vasodilator, influences tissue oxygen metabolism, and can prevent excessive amplification of Th1 cells. NO and peroxynitrite (ONOO⁻) promote angiogenesis, influence cell proliferation and differentiation. These examples of reactive oxygen and nitrogen species are also expected in CAP or in interaction of plasma with fluids, and could substantially contribute to generating the biological effects of CAP. The major difference between plasma species and endogenous radical production is the increased amounts of oxygen and nitrogen reactive species during CAP exposure. Although in endogenous metabolism, radicals are produced, increased generation of oxygen species can lead to direct cell damage. The effect of electrons and ions in generating radicals in CAPs are probably not necessarily of radical nature, but rather consequential reactions of the body following oxidative stress. Radicals, in particular, can exert ionizing effects on tissue components due to their tendency towards electron donation or uptake. Radical-driven reactions attack a variety of chemical bonds during this process. Hydrogen peroxide can be considered a pharmacologically relevant oxidation product. The reactions following plasma use described above compare to the phenomena seen after the exposition of mononuclear blood cells to culture media, in that similar anti-proliferative and cytotoxic effects could be elicited by long-term CAP treatment of cell suspensions. This is in
line with demonstrable anti-microbial effects of saline solution\textsuperscript{73} and water\textsuperscript{79} following prior treatment with CAP. So far, it remains questionable whether the “induction of the phosphate starvation response regulon PhoP by Argon plasma”\textsuperscript{80} is a primary or secondary pharmacological effect.

The enhanced oxygen radical reactivity has two causes:

(a) In a thermodynamic capacity, oxygen radicals are relatively strong oxidants.

\[
E^\circ_{\text{OH/H}_2\text{O}} = 2.813 \text{ V}
\]

is significantly larger than the standard potentials of hydrogen peroxide and oxygen:

\[
E^\circ_{\text{H}_2\text{O}_2/\text{H}_2\text{O}} = 1.763 \text{ V},
E^\circ_{\text{O}_2/\text{H}_2\text{O}} = 1.229 \text{ V}. 
\]

(b) In a kinetic manner, oxygen radicals are, just like many other simply charged gas radicals, quite reactive, because they contribute to one-electron reactions, which in general can occur faster than complex redox reactions with oxidation number changes of more than \pm 1. Due to the known relationship between linear-free enthalpies, high constants for the speed of oxidation correlate with high standard potentials. Superoxide radicals bear a rather low oxidation potential (around \text{\textasciitilde}0.3 \text{ V}), and therefore are only weak oxidants and rather strong reductive agents. Nevertheless, the reactions of the superoxide ion in water are quite fast, because protonation leads to generation of the hydrogen peroxide radical \text{HO}_2, and the standard potentials for the formation of \text{H}_2\text{O}_2 or \text{H}_2\text{O} are rather high:

\[
E^\circ_{\text{\text{HO}_2/\text{H}_2\text{O}_2}} = 1.44 \text{ V},
E^\circ_{\text{\text{HO}_2/\text{H}_2\text{O}}} = 1.65 \text{ V}. 
\]

The risk potential of \text{OH} radicals is furthermore emphasized by the fact that they even attack seemingly inert substances, such as gold, liberating gold ions, rather than forming a stable surface of gold oxide, which could be expected following normal thermodynamic rules.\textsuperscript{81, 82}

### Classification of CAPs as medical drug or medical device

The classification of CAPs depends on its intended use. It is crucial to determine whether or not solely physically (thermal, such as in coagulation), or pharmacologically active substrates are being generated as part of the main effect. Even in the latter case, however, CAPs can
still be classified as medical device when used for cleaning of artificial surfaces, e.g. biofilm removal on dental implants.

All therapeutic effects of CAP reported to date are mainly based on effects of radical-derived reactive products, which are associated with a pharmacological/immunological effect. Physical effects such as increased body temperature, electromagnetic fields, and (CAP source-dependent) UV or VUV radiation can potentially enhance these effects. However, therapeutic effects in healing of chronic wounds cannot be explained by physical effects alone. Moreover, the clinical course of chronic wound healing after the application of plasma in domestic animals and humans suggests that the plasma effect may trigger an intermediate phase of acute inflammation. The underlying immunological and biological mechanisms are not yet fully understood, but induction of integrins, NO, and increased phosphorylation of β-Catenin may indicate pharmacological and immunological effects of CAPs.

However, the complex composition of CAP and its generation within the plasma source at the time of application further corroborates its systematic classification within the classic drug category. In comparison with “classic” medical drugs, this may have the following consequences:

- The CAP compounds cannot be prepared and stored in a carrier substrate in defined doses with detailed knowledge about the exact composition before application on a patient.
- For CAP, storage times cannot be defined, as required for drugs or certain medical devices such as surgical or medical examination gloves. Instead, routine technical service and maintenance of the plasma-generating source will be required.
- Newly generated plasma intended for medical use can be uniquely different from another plasma in terms of its composition, depending on the type of plasma generation, the carrier gas used and environmental conditions. Therefore, the actual plasma for each current use needs to be thoroughly described in terms of its active components. During each use, narrow margins of tolerance need to be defined and
followed. Quantifiable parameters of action are: energy application (J/m²), temperature, field strength, UV spectrum, gas uses, and, associated with these, radicals in plasma. However, due to technical limitations, accurate quantification of radicals is not currently feasible. Moreover, it is technically challenging to maintain the energy application constant for a particular plasma source.

- Similar to a drug, CAPs intended for therapeutic application need to be composed of a defined content, which is unaltered by changes in environmental conditions, such as humidity of the ambient air. Therefore, plasma application needs to be independent of the surrounding atmosphere.

**Conclusion**

CAP as a new therapeutic option for chronic wounds or for neoplastic formations appears to be promising. Nevertheless, potentially deleterious long-term effects need to be ruled out. This applies particularly to mutagenic and carcinogenic risks of long-term plasma application. Therefore, before CAPs can safely be used in clinical applications for wound and skin treatment or treatment of tumors, it is imperative to further evaluate the interaction of living pro- and eukaryotic cells with CAP.

Analysis of physical and pharmacological properties of CAP supports its classification as a drug, which is generated by use of a medical device directly at the point of therapeutic application. In contrast to conventional drugs, CAP and its biologically active compounds are generated at the time of application by use of a medical device. Therefore, the effective plasma itself, which is generated at the treatment site could be regarded as a local redox modulator, which stimulates the cells and thus represents an applied pharmacological principle. The mode of action of the locally generated CAP with its main active components – the different radicals – is pharmacological and not physical in nature.
References

1. Weltmann, K.D., Kindel, E., Woedtke, T. von, Hähnel, M., Stieber, M., Brandenburg, R.  
   Atmospheric-pressure plasma sources: Prospective tools for plasma medicine.  

2. Isbary, G., Shimizu, T., Li, Y.F., Stolz, W., Thomas, H.M., Morfill, G.E., Zimmermann, J.L.  

   physical plasmas in the field of hygiene—relevance, significance, and future  

   Atmospheric pressure plasma in dermatology: Ulcus treatment and much more.  

   tissue-tolerable plasma on psoriasis vulgaris treatment compared to conventional  

   Randomized placebo-controlled clinical trial showed cold atmospheric argon  
   plasma relieved acute pain and accelerated healing in herpes zoster. Clin Plasma  


8. Lupu, A., Georgescu, N. Cell death in V79-4 and He-La cell lines, induced by  

   death in melanoma cells as compared with normal keratinocytes using a non-  


37. Daeschlein, G., von Woedtke, T., Kindel, E., Brandenburg, R., Weltmann, K.D., Jünger, M. *Antibacterial activity of an atmospheric pressure plasma jet against relevant


51. German Standardisation Committee. DIN SPEC 91315, **General Requirements For Plasma Sources In Medicine.** 2014; 1–16.


