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(71) Applicants *(for all designated States except US)*: **UNIVERSITA' DEGLI STUDI DI PADOVA** [IT/IT]; Via VIII Febbraio 1848, 2, I-35122 Padova (IT). **CONSORZIO RFX** [IT/IT]; Corso Stati Uniti, 4, I-35127 Padova (IT).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **LEONARDI, Andrea** [IT/IT]; Via Foscari, 8, I-35127 Padova (IT). **DELIGIANNI, Velika** [GR/IT]; Via Marin, 16, I-35122 Padova (IT). **MARTINES, Emilio** [IT/IT]; Via Martiri della Libertà, 21, I-35137 Padova (IT). **ZUIN, Matteo** [IT/IT]; Via Marin, 16, I-35122 Padova (IT). **CAVAZZANA, Roberto** [IT/IT]; Via Pascoli, 17, I-35125 Padova (IT). **SERIANNI, Gianluigi** [IT/IT]; Via Ferrari, 18, I-35020 Masera' Di Padova (IT). **SPOLAORE, Monica** [IT/IT]; Via Torino, 19, I-30035 Mirano (IT).

(74) Agent: **CINQUANTINI, Bruno**; NOTARBARTOLO & GERVASI S.p.A., Corso di Porta Vittoria 9, I-20122 Milan, (IT).

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[Continued on next page]

(54) Title: PLASMA DEVICE FOR TREATING LIVING TISSUES

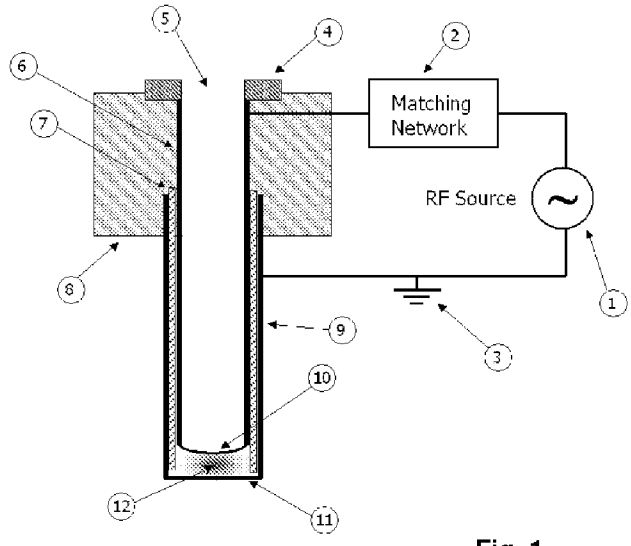

Fig. 1

(57) Abstract: Plasma device for treating living tissues consisting of a duct through which a noble gas or the like flows, in which a pair of electrodes is positioned close to the outflow opening, the pair of electrodes being at a potential difference varying in the course of time at a high frequency, and one electrode being connected to a reference potential, so that a low temperature plasma is generated thus avoiding the occurrence of electric discharges between the device and the tissue subjected to treatment.

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Declarations under Rule 4.17:

— *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*

— *of inventorship (Rule 4.17(iv))*

Published:

— *with international search report*

PLASMA DEVICE FOR TREATING LIVING TISSUES

DESCRIPTION

Field of the invention

The present invention relates to a device for treating living tissues based on plasma technology.

State of the art

The use of plasma is already known for treating living tissues. Specifically, one use is to achieve a loss in mutual adhesion between cells and in the attachment thereof to their substrate by effect of the interaction between plasma and cells.

Another use consists in inducing programmed cell death (apoptosis).

Finally, a further use is to sterilise the treated area by killing bacteria therein without significantly damaging the cells forming the area.

Either one or the other effect is obtained by varying the current, the voltage and the frequency in the plasma devices as well as the duration of the treatment.

Specifically, the plasma devices provided by the known art are based on the outflow of an inert gas that flows through a nozzle or hollow needle, so that an electric discharge of the “glow discharge” type or of the “electric arc” type is generated between said needle and the surface to be treated, as these are set at different potentials. The build-up of the electric discharge is allowed by the ionization of a fraction of the gas, that is by the generation of plasma. In turn, it is the current flow that maintains the process of plasma generation over time through the ionization of the neutral atoms by the electrons carrying such a current.

Said devices are defined having a unipolar configuration, as said needle represents an electrode, whereas the surface to be treated forms the other electrode of the circuit that is therefore established, whereby the plasma is generated outside the device and the treatment takes place by direct contact, or nearly, of the plasma with the tissue to be treated.

It is therefore apparent that the potential of the second electrode is not irrelevant. Specifically, the circuit consisting of the needle/tissue does not have a reference potential, with the subsequent risk of a possible generation of electric discharges concentrated between the needle and the tissue with the subsequent damaging of the tissue itself.

Another problem depends on the fact that, as the production of plasma is associated to a gas heating, when it is produced externally, the gas flow results potentially detrimental for the substrate to be treated.

Summary of the invention

It is the object of the present invention to overcome all of the aforesaid drawbacks and to provide a device for treating living tissues for the purpose of obtaining effects such as sterilization, cell detachment, cell apoptosis or tissue necrosis, as well as other possible non-destructive interactions.

The object of the present invention is a plasma device for treating living tissues, which, according to claim 1, includes:

- a pair of electrodes one of which is connected to a reference potential;
- an electric generator connected to said electrodes, adapted to generate a potential difference varying over time therebetween;
- a duct, one end of which is connected to a gas source and the other end of which contains said electrodes, so that the gas is converted to plasma just before exiting the duct.

The gas employed to generate plasma may be helium, argon or another noble gas, or a mixture thereof, and it may contain higher or lower fractions of nitrogen, oxygen or other reactive gases.

Said device may be applied in the sterilization of tissues, avoiding the damaging of the possible substrate, as the aforesaid device forms a bipolar system, including at least one pair of electrodes required for the generation of plasma.

Specifically, the two electrodes are connected to an electric radiofrequency generator, and one of the two electrodes is connected to a reference potential that may be equivalent to ground.

Accordingly, the plasma is produced within the device itself, from which a flow exits substantially consisting of neutral atoms or molecules and active chemical species, thereby avoiding that the electrodes come into contact with the substrate to be treated.

Advantageously, said device results safe due to the absence of electric discharges between the device and the tissue, which therefore does not represent the second electrode and, furthermore, a low temperature of the gas is ensured, it being ideal for the substrate not to be damaged during treatment.

Therefore, a specific object of the present invention is a plasma device for treating living tissues, as described in greater detail in the claims.

The dependent claims disclose preferred embodiments of the invention.

Brief description of the Drawings

Further features and advantages of the invention will become more apparent in light of the detailed description of a preferred though not exclusive embodiment of a plasma device for treating living tissues, which is shown by way of non-limitative example with the aid of the accompanying drawings in which:

Figure 1 shows the schematic diagram of a plasma device;

Figure 2 shows the decreasing trend of the number of bacterial colonies as the treatment time increases with the present device, with a 90% reduction in a treatment of about 1 minute.

Detailed description of a preferred embodiment of the invention

A preferred embodiment of the device, which is shown in Figure 1, provides for a pair of electrodes 6 and 9 consisting of two concentric nozzles, one inserted into the other and reciprocally insulated by means of an tube 7 made of insulating material, which is placed between the two and is as long as the electrode 9.

The inner nozzle 6 forms the duct through which the gas flows and ends internally to the outer nozzle 9, so that a special area 12 is formed for the generation of plasma. Therefore, the latter results between said tube 7 made of insulating material and the inlets of both electrodes.

The inlet of the nozzle forming the inner electrode 6 is overlapped by a grid 10 made of conductive material and the inlet of the nozzle forming the outer electrode 9 is overlapped by a grid 11 made of conductive material. Both grids are galvanically connected with respective electrodes. Said grids are adapted to improve the generation of plasma and with reference to the specific application, they are from a few tenths of millimetre to some tens of millimetre away from one another.

Said electrodes and therefore said grids are subjected to a potential difference, as they are electrically connected to said electric generator 1, which is specifically an electric radiofrequency generator, by means of a coupling circuit 2, designated as Matching Network, which serves to minimise the power reflected towards the electric generator and to increase the voltage to the required values to ionize the

gas.

Among the two electrodes, it is preferred that the electrode 9 and the grid 11 are galvanically connected to ground potential.

The free end of the inner electrode 6 is provided with a bayonet joint 5 of the known type, for an easy connection of the device to said gas source.

Both grids may be flat, concave or convex, although it is preferred for the grid 11 to be flat and for the grid 10 to be convex, the convexity being towards the grid 11.

The electrodes 6 and 9, which are insulated by the insulating tube 7, are held in position by a support structure 8.

Therefore, the gas flow 5 is introduced through said joint, which flows within the inner electrode 6 up to the end fitted with the grid 10. By flowing through the area 12, the gas is transformed into plasma by the effect of the electric field induced by the generator on the grids, which varies over time at a frequency between 1 and 30 MHz and displays a potential difference of the order of one kV.

The generator provides a power of the order of ten W, from 1 to 30 W, with an optimal value of 10 W, a considerable fraction of which is dissipated within the coupling circuit. The plasma flowing out from the device hits the cell substrate to be treated.

Some preferred gases are helium, argon mixed with air, oxygen, nitrogen and mixtures thereof and/or other gases, to form a flow in the range between 0.1 and 5 litres/minute. Specifically, the use of noble gases may be avoided, for example, by increasing the potential difference between the electrodes.

Therefore, said device may be applied, among other uses, to treatments aimed at the sterilization of the substrate, for example, for the therapy or the prevention of infections, to treatments aimed at detaching cells from the substrate, and to treatments to induce cell apoptosis.

A preferred application method provides that the optimum distance between the outer grid 11 and the substrate to be treated varies between 0 and 2 cm, whereas the optimum application time is in the range between 10 seconds and 5 minutes.

Advantageously, the low value of the power coupled with the plasma ensures that the latter is formed without the gas flow significantly heating. Thereby, the plasma results poorly ionized, i.e. most of the gas remains in the form of neutral atoms or molecules and in a non-equilibrium state, characterised by an electronic

population at high temperature, that is of the order of some electronvolts, thus of some tens of thousands of Kelvins, and a ion population at room temperature. A plasma displaying these features is usually defined as a “low temperature plasma”. Such a condition allows to prevent the occurrence of any thermal damage on the substrate to be treated.

Furthermore, including at least one pair of electrodes subjected to a known potential difference - one electrode (preferably the outer one) being set at a ground potential - ensures the total absence of electric discharges between the device and the substrate, thus avoiding the subsequent damaging thereof.

Experimental results with an application in the field of ophthalmology.

The device has been applied for times varying between 30 seconds and 5 minutes to bacterial suspensions in Luria placed in wells. Some untreated control samples have been kept in the same temperature conditions as those exposed to plasma. After the treatment, each sample has been plated on an appropriate agar substrate and incubated at 37°C for sixteen hours. The number of colonies grown on each culture plate has been considered representative of the number of bacteria that survived the treatment. The tests have been carried out on Escherichia Coli. The graph represents the number of residual bacterial colonies as the exposure time varies (the value for t = 0 is that relative to the control samples). In Figure 2, a clear decrease may be noted in the number of colonies as the treatment time increases with a reduction of 90% in about 1 minute.

A treatment has also been carried out on a pig’s eye on which a bacterial corneal ulcer had previously been induced. In this test, a 70% reduction in the bacterial charge has been noted as compared to control samples, after a treatment of 2 minutes, whereas the histological test under an optical microscope has not highlighted alterations of the cell structure of the corneal surface.

The specific modes to embody the invention described herein do not limit the content of this application, which includes all of the variants of the invention according to the claims.

CLAIMS

1. A plasma device for treating living tissues including:

- a pair of electrodes (6 and 9), one of which is connected to a reference potential;
- an electric generator (1) connected to said electrodes, adapted to generate a potential difference varying over time therebetween;
- a duct, one end of which is connected to a gas source and the other end of which contains said electrodes, so that the plasma is generated just before exiting the duct.

2. A device according to claim 1, wherein said gas source provides a gas mixture.

3. A device according to claim 2, wherein said gas mixture includes noble gases, such as argon and/or helium and/or neon.

4. A device according to claim 2, wherein said gas mixture includes reactive gases such as oxygen and/or nitrogen and/or steam and/or other gases according to the specific applications.

5. A device according to claim 1, wherein said pair of electrodes includes two concentric nozzles (6) and (9) defining an inner and an outer electrodes, which are electrically insulated one from the other by means of an insulating tube (7) and are held in position by a support structure (8).

6. A device according to claim 1, wherein said electric generator (1) is a high frequency generator.

7. A device according to claim 1, further provided with a coupling circuit (2) that serves to minimize the power reflected towards the electric generator (1) and to increase the voltage up to values adapted to ionize the gas.

8. A device according to claim 1, wherein said reference potential is ground.

9. A device according to claim 1, wherein the inlet of said electrode (6) results internal to the outer electrode (9).

10. A device according to claim 1, wherein the inlet of the inner electrode (6) is overlapped by a grid made of conductive material (10) so as to be galvanically connected one to the other.

11. A device according to claim 1, wherein the inlet of the inner electrode (9) is overlapped by a grid made of conductive material (11) so as to be galvanically connected one to the other.

12. A device according to claim 10, wherein said grid (10) is flat or concave or convex.

13. A device according to claims 11 and 12, wherein the distance between said grids is in the range between a few tenths of millimetre and some tens of millimetre.

14. A device according to claim 5, wherein said insulating tube (7) is as long as the nozzle forming the outer electrode (9).

15. A device according to claim 1, wherein said electric generator is a radiofrequency electric generator having a power in the range between 1 and 30 W.

16. A device according to claim 1 or 15, wherein said electric generator has an optimum power of 10 W.

17. A device according to claim 7, wherein the set comprised of the electric generator and said coupling circuit is adapted to generate a potential difference between said electrode of the order of kV and a frequency between 1 and 30 MHz.

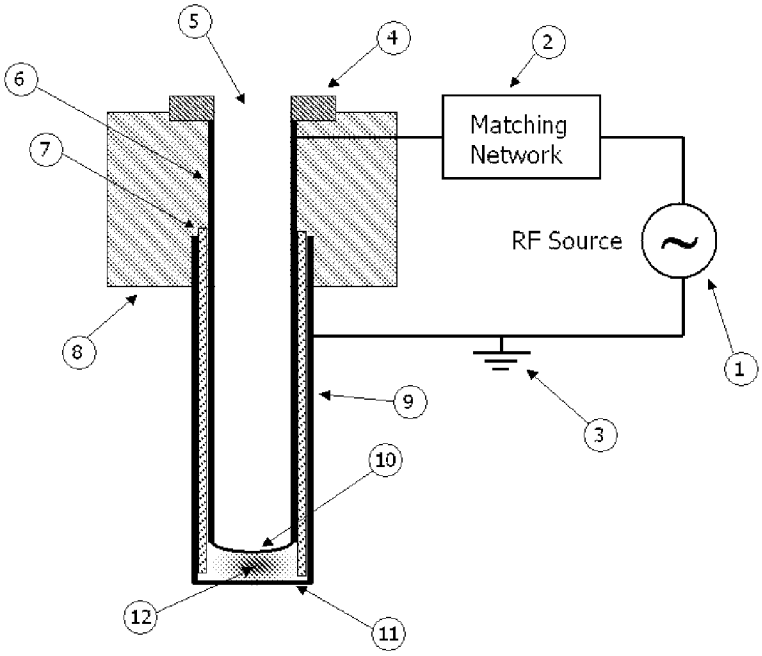
18. A device according to claim 1, wherein the gas flow flowing through said duct is in the range between 0,1 and 5 litres/minute.

19. A method of using the device according to the preceding claims, wherein a tissue is treated in order to sterilize the substrate and/or to detach the cells from the related substrate and/or induce apoptosis.

20. A method of using the device according to claim 19, wherein the inlet of the duct is held at a distance from the tissue to be treated in the range between 0 and 2 centimetres, in relation to the purpose of use.

21. A method according to claim 20, wherein the treatment time of the tissue to be treated varies between 10 seconds and 5 minutes in relation to the purpose of use.

22. The use of the device according to one of claims 1 to 17 for a corneal ulcer sterilization.



1) Electric radiofrequency generator

2) Matching Network
3) Ground reference
4) Gas joint
5) Gas inlet
6) Inner electrode
7) Insulating tube
8) Support structure
9) Outer electrode
10) Inner grid
11) Outer grid
12) Plasma generation area

Fig. 1

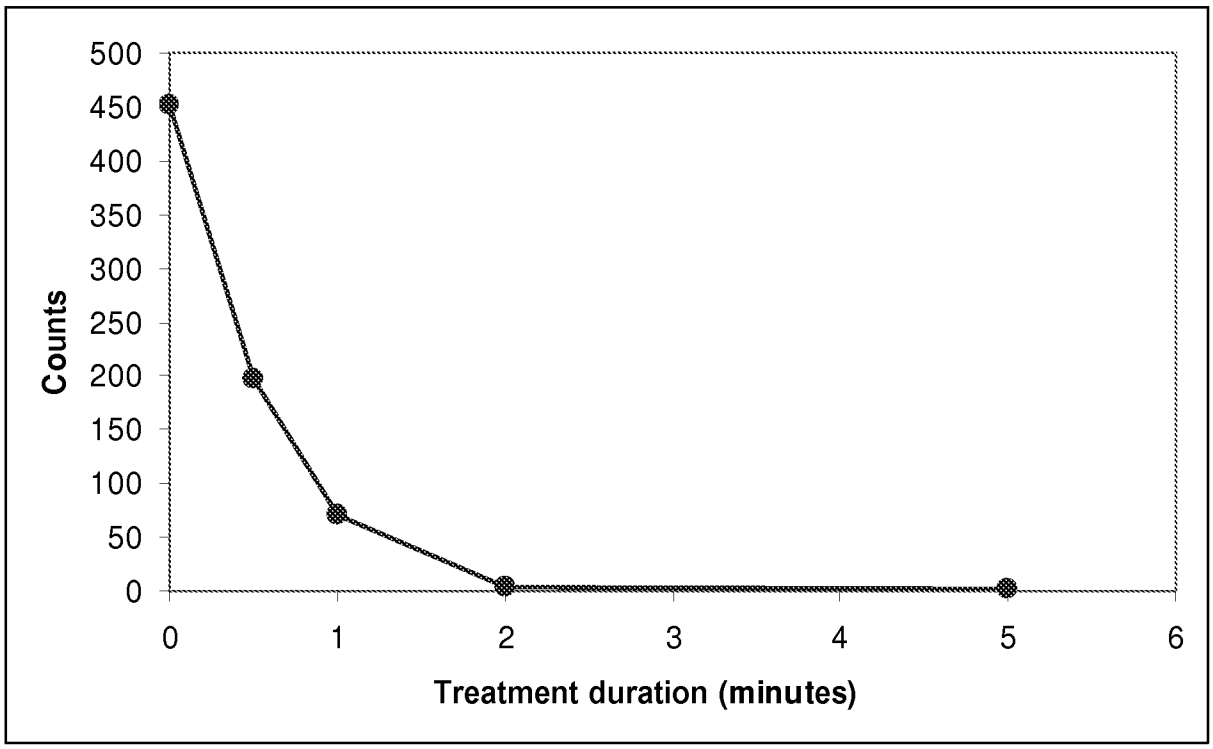


Fig. 2

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2008/063275

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61B18/00

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61B

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 01/62169 A (GYRUS MEDICAL LTD [GB]; GOBLE COLIN CHARLES OWEN [GB]; PENNY KEITH [GB]) 30 August 2001 (2001-08-30) pages 4,13,17,, column 33; figures 6,9,19	1-18

☐ Further documents are listed in the continuation of Box C.

☒ See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
30 December 2008	14/01/2009

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Edward, Vinod

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP2008/063275

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.: 19-22
because they relate to subject matter not required to be searched by this Authority, namely:

Due to the step of detaching cells, independent claim 19 relates to a method for treatment of the human or animal body by surgery (Rule 39.1(iv) PCT).

2. [ ] Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers allsearchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fees, this Authority did not invite payment of additional fees.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search reportcovers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.

[ ] The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.

[ ] No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/EP2008/063275

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
WO 0162169 A	30-08-2001	AT	293927 T	15-05-2005
		AU	3394401 A	03-09-2001
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