(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
9 September 2011 (09.09.2011)

PCT

(10) International Publication Number
WO 2011/107875 A2

(51) International Patent Classification:
A61H 1/02 (2006.01)

(21) International Application Number:
PCT/IB2011/000492

(22) International Filing Date:
7 March 2011 (07.03.2011)

(25) Filing Language: Italian

(26) Publication Language: English

(30) Priority Data:
PI2010A000023 5 March 2010 (05.03.2010) IT

(71) Applicant *(for all designated States except US)*: **TONLORENZI, Daniele** [IT/IT]; Via Monteverde 39, I-54033 (IT).

(72) Inventor; and

(75) Inventor/Applicant *(for US only)*: **BRUNELLI, Marcello** [IT/IT]; Via Rio Pozzale, 133, I-56021 Cascina (IT).

(74) Agent: **CELESTINO, Marco**; ABM AGENZIA BREVETTI & MARCHI, Viale Giovanni Pisano, 31, I-56123 Pisa (IT).

(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

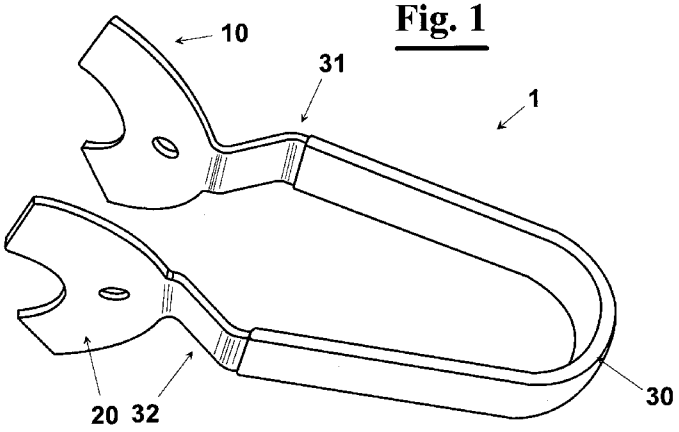
(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

— *without international search report and to be republished upon receipt of that report (Rule 48.2(g))*

(54) **Title**: DEVICE FOR TREATING OR PREVENTING CEREBRAL DISEASES, ARTERIAL HYPERTENSION, CEREBRAL STROKE, NEURODEGENERATIVE DISEASES.

Fig. 1



(57) **Abstract**: A device (1) for treating or preventing cerebral diseases, arterial hypertension, cerebral stroke, neurodegenerative diseases, provides a means for causing a mouth hyperextension and activating the proprioceptive sensations of the mandibular muscles. The means for causing a mouth hyperextension comprise a central portion (30) including a "U"- folded metal lamina of about 1-2 mm of thickness, for example 1.2 mm. At the end (31.32) of the central portion (30) are provided two curved laminar portions (10.20) that are arranged to be located, in use, at the upper wall (41) and at the lower wall (42) of the oral cavity of a patient (50). More in detail, the curved laminar portions (10.20) can be connected to the central portion (30) through a respective stair portion (11.21). This allows the central portion (30) to go beyond the dental arches (51, 52) of the patient (50), in order to arrange the laminar portions (10.20) adjacent to the palate and to the space between the tongue and the lower dental arch of the patient, or directly on the tongue itself. The metal of the device (1) has such a hardness that allows, in use, to stretch the mandibular elevator muscles.

WO 2011/107875 A2

TITLE

DEVICE FOR TREATING OR PREVENTING CEREBRAL DISEASES, ARTERIAL HYPERTENSION, CEREBRAL STROKE, NEURODEGENERATIVE DISEASES.

DESCRIPTION

Field of the invention

The present invention relates to a device for treating or preventing cardiovascular diseases, arterial hypertension, cerebral diseases, cerebral stroke, neurodegenerative diseases.

Description of the prior art

As well known, the trigeminal nerve is the fifth of the cranial nerves, and it conveys most of sensory information coming from the skull, from the face, from the oral cavity, from the conjunctiva and from the dura mater and provides motor innervation to the jaw muscles.

The trigeminal system comprises fibres that convey sensory information of different modes, that project to different brainstem nuclei.

Peripherally, the trigeminal system consists of three main branches: the ophthalmic branch, the mandibular branch and the maxillary branch. The ophthalmic and maxillary branches are purely sensory nerves, whereas the mandibular branch contains both sensory and motor fibres. The three branches exit from the skull through three foramina, known as the superior orbital fissure, the foramen rotundum and the oval foramen.

The trigeminal nerve is therefore a mixed nerve that is functionally similar to a spinal nerve. Like in the case of the spine, the central branches of the sensory fibres and the motor axons penetrate into and exit from the brainstem at distinct locations, i.e. at a sensorial root (afferent root) and at a motor root (efferent root). The cellular

bodies of most trigeminal sensory fibres are located at the Gasser's ganglion (or semilunar ganglion).

In the human being, the skin of the face contains three types of receptors that convey information through the trigeminal nerve: the mechanoreceptors, the thermoreceptors and the nociceptors. The trigeminal nerve also innervates most part of the mouth mucosa, two-thirds of the front tongue portion, and the dura mater of the anterior and media cranial fossae. The trigeminal nerve also innervates the dental pulp, the gums and the periodontal membrane.

By stimulating the various sensory branches of the trigeminal nerve, during facial surgery, considerable effects have been remarked on cardiocirculatory parameters. The bradicardia response, i.e. a pressure blood reduction and apnea, that occurs by stimulating the trigeminal terminations, is called trigemino-cardiac reflex.

During a maxillo-facial surgery, the trigemino-cardiac reflex can cause symptoms such as hypothension, bradicardia and apnea. It has been observed that the trigemino-cardiac reflex is caused by a stimulation at any position of the trigeminal nerve, and that the trigemino-cardiac reflex disappears substantially immediately once the surgery is completed.

It has also been observed that a direct stimulation of the trigeminal terminations may cause synaptic excitators in the neurons of the vagal centre, which means that the vagus nerve is involved as efferent branch of the reflex.

In patients who experienced cranial base surgery, the cardiac frequency and pressure blood follow-up has shown a cardiac frequency average decrease ranging from 78 pulsations/minute to 48 pulsations/minute, with a pressure average decrease of 33%.

In order to avoid the above described symptoms during

maxillo-facial surgery, the patients are administered atropine, a cholinergic receptor blocker. In some cases a lowering the cardiac frequency required administering large atropine doses, in order to avoid a cardiocirculatory arrest.

However, few data are available at present about the physiological mechanisms that control the trigemino-cardiac reflex as well as about the possible therapeutic applications.

In US2011022126 a device is described for stimulating the trigeminal nerve by application of an electric current that produces a predetermined intensity magnetic field.

Instead, in US5693077 a thermal stimulus is used in order to cause to cause a reflex of the trigeminal nerve.

Summary of the invention

It is therefore a feature of the present invention to provide a device for non-invasively causing a trigemino-vagal reflex, in order to stimulate the proprioceptive afferences on the mandibular muscles.

It is another feature of the present invention to provide a device for non-invasively causing a trigemino-vagal reflex which allows improving the performances of athletes while making sport.

It is another feature of the present invention to provide a device for non-invasively causing a trigemino-vagal reflex for treating and preventing psychophysical stress in a subject who performs a sport.

These and other objects are achieved by a device for treating or preventing cerebral diseases, arterial hypertension, cerebral stroke, neurodegenerative diseases, comprising a means for causing an alternating hyperextension of the jaw, in order to cause a cyclical extension of the mandibular elevator muscles with a

non-invasive proprioceptive stimulation of regions that are innervated by the trigeminal nerve and with a subsequent stimulation of the trigemino-cardiac reflex.

By "alternating hyperextension" it is meant a succession of alternating mandibular open-close movements, wherein the opening movement has an extent suitable for stretching the elevator muscles. This way, by cyclically repeating such hyperextension, for example for 10/15 minutes, an optimal stimulation is obtained which causes a considerable trigemino-cardiac reflex.

In particular, the means for causing a hyperextension of the jaw may comprise:

- a first portion that is arranged to be brought into contact with the upper wall of the oral cavity of the patient;
- a second portion that is arranged to be brought into contact with the lower wall of the oral cavity of the patient;
- a connection portion arranged between the first and the second portions, which is arranged to apply a contrast force upon approaching of said first and second portions.

Advantageously, the connection portion is arranged to maintain the first and the second portions at a rest distance and to allow the first and the second portions to approach each other upon application of a closing force from the rest distance to a maximum closing distance. More in detail, the rest distance corresponds to the distance between the lower wall of the oral cavity and the upper wall of the oral cavity at the height of the inner dental arch of a patient at a maximum mandibular extension.

In particular, a means can be provided for adjusting the rest distance.

Advantageously, the above described contrast force has

a maximum value set between 14N and 25N.

In particular, the above described adjustment means of the rest distance is housed in a handgrip.

In a preferred exemplary embodiment, the connection portion is made of a resilient material, in particular of steel, such as stainless 301.

In particular, the means for causing a hyperextension of the jaw comprises:

- at a first end, a first substantially "U"-shaped laminar portion, said first laminar portion arranged to be brought into contact with the upper wall of the oral cavity of the patient;
- at a second end, a second substantially "U"-shaped laminar portion, said second laminar portion arranged to be brought into contact with the lower wall of the oral cavity of the patient;
- a laminar connection portion connecting said first and said second shaped laminar portions, said laminar connection portion made of a resilient material.

In an advantageous exemplary embodiment, the above described lamina has a thickness set between 1 mm and 2 mm, advantageously between 1.1 mm and 1.5 mm, for example 1.2 mm.

In particular, the lamina may have a length set between 15 cm and 25 cm.

According to another aspect of the invention the device, as above described, may be used for treating or for preventing cerebral diseases.

In a further aspect of the invention the device, as above described, can be used for treating or preventing hypertension.

Even in another aspect of the invention the device, as above described, can be used for treating or preventing cerebral stroke.

Furthermore, the device as above described, can be used for treating or preventing neurodegenerative diseases.

Brief description of the drawings

The invention will be now shown with the following description of an exemplary embodiment thereof, exemplifying but not limitative, with reference to the attached drawings in which:

- Fig. 1 shows a perspective view of a possible exemplary embodiment of a device, according to the invention, for treating or preventing cerebral diseases, arterial hypertension, cerebral stroke, neurodegenerative diseases;
- Fig. 2 shows the device of Fig. 1 in a perspective elevational side view;
- Fig. 3 shows the device of Fig. 1 in a top plan view;
- Fig. 4 shows an elevational side view of the device of Fig. 1 in operation conditions;
- Fig. 5 shows an elevational front view of the device of Fig. 1 in operation conditions;
- Figs. 6 and 7 diagrammatically show perspective views of two possible exemplary embodiments of the device of Fig. 1;
- Figs. 8A to 8C show respective time plots of the artery systolic pressure (SP), artery diastolic pressure (DP) and cardiac frequency (CF) measured with a sphygmomanometer, in subjects treated with the device of Fig. 1, the asterisk "*" indicates $p<0.05$ between the data B, representative of the basal value, and the corresponding points;
- Figs. 9A to 9C show respective time plots of the data obtained in patients treated with the device of

Fig. 1 (■) and in control conditions (●) by sphygmomanometer measurements, the asterisk "*" indicates $p<0.0001$ between corresponding values of the two curves;

- Figs. 10A to 10C show respective time plots of the values of SP, of DP, and of CF obtained in the subject while masticating a chewing-gum (▒) and in control conditions (●) by manual sphygmomanometer measurements;

- Figs. 11A to 11C show respective time plots of the values of SP, DP and CF obtained in patients treated with device 1 (■,n=9) or when masticating a chewing-gum (▒,n=9) by sphygmomanometer measurements ("*" indicates $p<0.01$ between corresponding values of the two curves);

- Figs. 12A to 12C show respective time plots of values of SP, of DP and of CF obtained in patients treated with an ice cream stick (▒,n=9) and in control conditions (●,n=9) by a sphygmomanometer measurement step;

- Figs. 13A to 13C show respective time plots of values of SP, of DP and of CF obtained in patients treated with an ice cream stick (▒, n=9) and with the device of Fig. 1 (■, n=9) with sphygmomanometer measurements, the asterisk "*" indicates a pressure difference $p<0.001$ between corresponding data of the two curves;

- Figs. 14A to 14C show respective time plots of values of SP in control conditions (Fig. 14A) by applying the device of Fig. 1 (Fig. 14B) and while masticating a chewing-gum (Fig. 14C), respectively, the arrows indicates the period (10 minutes) during which the various treatments have been performed, n=9;

- Figs. 15A to 15C show the graphs of the data

obtained with the "Finapres" system and that describe the DP time plot in control conditions (Fig. 15A), by applying the device of Fig. 1 (Fig. 15B) and while masticating a chewing-gum (Fig. 15C). [The arrows indicate the period (10 minutes) during which the various treatments have been applied. n=9]

- Figs. 16A to 16C show time plots of data obtained by the Finapres system, which are representative of CF in control conditions (Fig. 16A), by applying the device of Fig. 1 (Fig. 16B) and while masticating a chewing-gum (Fig. 16C),respectively. [The arrows indicate the period (10 minutes) during which the various treatments have been applied, n=9];

- Fig. 17 shows a time plot of the average blood pressure of a subject treated with IM for 5 minutes. Note the oscillatory trend throughout all the observation period, where (*) indicates $p<0.05$;

- Figs. 18A to 18E show a time plot of the diameter (measured in μm) of arterioles of order 5, 4, 3, 2 and 1, respectively, in mice treated with mandibular hyperextension (IM) for 5 minutes, (*) indicates $p< 0.05$;

- Fig. 19 shows a time plot of the percentage deviation from a respective basal trend of the average blood pressure (PAm) and of the diameter of the arteries of order 2 in patients treated with IM for 5 minutes;

- Fig. 20 shows the deviation of PAm in patients treated with IM for 10 minutes (the asterisk (*) indicates $p<0.05$);

- Figs. 21A to 21E show respective time plots of diameter (measured in μm) of the arterioles, of order 5, 4, 3, 2 and 1, respectively, in mice treated with IM for 10 minutes;

- Fig. 22 shows the graph of the percentage deviation from the basal trend of the average blood pressure (PAm) and of the diameter of the arteries of order 2 in patients treated with IM for 10 minutes;
- Fig. 23 shows a time plot of the deviation of PAm in patients treated with IM for 15 minutes;
- Figs. 24A to 24E show respective time plots of diameter (measured in μm) of the arterioles, respectively of order 5, 4, 3, 2 and 1 in patients treated with IM for 15 minutes;
- Fig. 25 shows a time plot of the percentage deviation from the basal trend of the average blood pressure (PAm) and of the diameter of the arteries of order 2 taken as a representative example in patients treated with jaw hyperextension for 15 minutes;
- Figs. 26A to 26E show a time plot of the diameter (measured in μm) of arterioles of order 5, 4, 3, 2 and 1, respectively, in the control mice.

Detailed description of some exemplary embodiments

With reference to Figs. 1 to 4, a possible embodiment of a device 1 for causing a mouth hyperextension and activating the proprioceptive sensations of the mandibular muscles of a patient 50, comprises a central portion 30 including a "U"-folded metal lamina of about 1-2 mm thickness, for example 1.2 mm.

At ends 31 and 32 of central portion 30, device 1 may be also equipped with curved laminar portions 10 and 20 that are adapted to be arranged, in use, at an upper wall 41 and at a lower wall 42 of the oral cavity of a patient 50.

More in detail, curved laminar portions 10 and 20 may be connected to central portion 30 through respective step portions 11 and 21. This allows central portion 30 to go

beyond dental arches 51 and 52 of patient 50, in order to arrange laminar portions 10 and 20 respectively adjacent to the palate and to the area between the tongue and the lower dental arch of the patient, or directly on the tongue itself, or in the sublingual region. The hardness of the metal of which device 1 is made is such that it allows, in use, to stretch the mandibular elevator muscles.

For instance, device 1 may be made of steel, in particular of stainless steel 301, or in another spring steel in order to apply a resilient contrast force that may be set between about 14 N and about 25 N.

In figures 6 and 7 two possible exemplary alternative embodiments are diagrammatically shown. Fig. 6 relates to a unit 200 for adjusting the rest distance and the maximum force opposed by the jaws, comprising for example inside adjustment means of known type, such as screw, lever, or cam adjustment means. An electromechanical adjustment means may also be provided operated by a program means, which modifies the rest length and/or the maximum force according to a predetermined program.

In Fig. 7, device 1 has a handgrip 15 by which the user holds device 1. Furthermore, a means 13 may be provided for adjusting a rest distance that corresponds to the distance between the lower wall of oral cavity 42 and the upper wall of oral cavity 41 at the height of the inner dental arch of a patient at a maximum mandibular extension.

TEST DATA (TRIAL 1)

Twenty healthy patients have been considered, 6 males and 14 females, aged between 24 and 26, and have been treated with a mandibular stretching test by device 1, above described, in order to stimulate the proprioceptive afferences of the mandibular muscles.

The cardiac frequency (CF), the artery systolic pressure (SP) and the artery diastolic pressure (DP) of each of the 20 patients was measured.

In particular, the test data have been obtained by examining two different groups (group A and group B) of the patients, as described hereafter.

Eleven patients of a first test group, or unit A, have been treated with device 1 for 10 minutes, and the above parameters were measured: CF, SP and DP. Subsequently, the above parameters have been measured without using device 1. Nine patients of second test group, or group B, have been subjected to 4 different treatments. In a first case, the patients have been treated with device 1 for 10 minutes. In a second case, the patients masticated a chewing-gum. In a third condition, the patients biting a stiff rod between their teeth, and in a fourth operative condition, the patients were not treated at all. For each operation condition the parameters CF, SP and DP were measured.

Given the homogeneity of the results, the data related to the different test conditions were cumulatively used for statistical analysis.

A Finapres measure tool was used in order to follow the blood pressure by a non-invasive measurement.

The blood pressure time plot was obtained by means of a finger cuff designed for detecting blood pulsations. The pressure time plot has been used for deducing, by means of a suitable filter, the SP, AP and average pressure measured at the brachial artery. Furthermore, three electrodes were applied to the patients, one to the left shoulder, another to the right shoulder and a control electrode at the left hip in order to record an electrocardiograph (ECG).

Hemodynamic data and the ECG were taken by a specific

computer system and stored for successive analysis.

An automatic digital sphygmomanometer was also used. The measuring cuff was applied at the left arm of the patients and maintained there during the whole recording session. After making sure that the bracelet was at the same height as the heart, the hemodynamic parameters of the patients were automatically determined by an automatic inflating/deflating system and by an automatic Fuzzy Logic detection system. The range of the pressure detector was 20-280 mmHg and the range of the cardiac frequency was 40-180 pulsations/minute. The measure precision for pressure was 3 mmHg and for pulsations was 5%.

For each among 11 patients of the first group, CF, SP and DP values were determined 5 times, 3-4 minutes from one another, in order to obtain the basal values of the parameters that were taken into account, indicated in the time plots with B.

Device 1 was then tested for 10 minutes and the hemodynamic parameters were determined by a digital sphygmomanometer with the following timing: upon completion of 10 minutes (T), and after that at 5 (5'), 15 (15'), 30 (30'), 50 (50') and 80 (80') minutes.

The patients of group B followed the same procedure as the patients of group A; moreover, they received the cuff of the Finapres system at the middle finger of their right hand, as well as electrodes for recording the ECG graph. Subsequently, a pulsatility function of the finger cuff was activated and the recording started and continued for 30 minutes, whereas the manual recording was continued up to 80 minutes after completion of the treatment. The first 5 minutes of the Finapres recording matched the last 2 manual measurements that were made of the basal values before each treatment.

The patients of group B performed 4 recording sessions, on distinct and not necessarily consecutive days, during which, randomly, they were subjected to 4 different treatments: no treatment, application of device 1 for 10 minutes, mastication of a chewing-gum for 10 minutes and positioning and biting an ice cream stick between the teeth for 10 minutes.

The choice of a chewing-gum was made for creating a mouth movement as similar as possible to the movement made in presence of device 1 but without inducing a muscle hyperextension. The use of an ice cream stick matched with the positioning of device 1 in the mouth, but produced only a stimulation of the exteroceptive sensations.

The data given below are shown as a ±E.S. average. By using of the Anova statistical test for repeated measurements, the differences were evaluated among the different test conditions with time. By the Tukey post hoc test an evaluation was made of the time at which significant differences were observed.

In order to establish whether the proprioceptive stimulation can activate a vagal response, a mandibular hyperextension was imposed with device 1 for 10 minutes to the 20 patients belonging both to group A and To the 20 patients, the artery systolic pressure (SP), the artery diastolic pressure (DP) and the first (basal, B) cardiac frequency (CF) were measured, just after applying device 1 (T) and then after 5, 15, 30, 50 and 80 minutes. The basal values were obtained as above described.

In Fig. 8A a time plot is shown of the artery systolic pressure (SP). A progressive reduction is noted of the artery systolic pressure (SP) that is statistically significant (ANOVA for repeated measures

F6.19= 7.509, p<0.0001; Tukey post hoc test) starting from 5 minutes (5') after the treatment, with respect to basal values (B).

Fig. 8B shows a time plot of the artery diastolic pressure (DP). A reduction of DP is noted that is statistically significant (ANOVA for repeated measures: F6.19=2.590, p=0.0197; Tukey post hoc test) starting from 15 minutes (15'), after the treatment, with respect to basal values (B).

In Fig. 8C a time plot is shown of the cardiac frequency (CF). A decrease is observed of CF which is statistically significant, (ANOVA for repeated measures F6.19=8.809 p<0.0001, Tukey post hoc test) starting from 15 minutes after the treatment with respect to basal values (B). The symbol "*" indicates p<0.05 between B, the basal value, and the corresponding points.

When patients are not treated at all (control conditions) the values of SP, DP and CF do not show, instead, any significant change, as shown in Figs. 9A - 9C with the symbol "●". More in detail, the graphs shown in Figs. 10A to 10C show a time plot of the data obtained in patients treated with device 1 (■, n=20) and in control conditions (●, n=20) by a sphygmomanometer. More in detail in Fig. 10A shows SP values, Fig. 10B shows DP values and Fig. 10C shows CF values. The symbol "*" indicates a difference p<0.0001 between the respective values of the two curves. By the ANOVA test for repeated two-way measures, a significant difference was remarked of the SP trend (F6.38=9.803, p<0.0001; Tukey post hoc test: p<0.05) (Fig. 9A), of the DP trend (F6.32=7.628, p<0.0001; Tukey post hoc test: p<0.05) (Fig. 9B) and of the CF trend (F6.32= 8.629, p<0.0001; Tukey post hoc test: p<0.05) (Fig. 9C) after the application of device 1 (■) with respect to the control

conditions (●).

In order to evaluate if a mastication movement can cause a change of the cardiac parameters, 9 of the 20 test patients were asked to masticate a chewing-gum for 10 minutes. Summary data of SP, DP and CF manual recordings are shown in Figs. 11A to 11C. More in detail, the graphs show a time plot of the data obtained in patients treated with chewing-gum (▒, n=9) and in control conditions (●, n=9) by manual sphygmomanometer measurements. Fig. 11A shows SP values, Fig. 11B shows DP values and Fig. 11C shows CF values.

After 10 minutes masticating chewing-gum, no statistically significant differences of the three parameters were observed with respect to the basal values, and with respect to the SP plot (ANOVA for repeated measures: $F_{1.16}=0.336$, $p=0.571$; Tukey post hoc test: $p<0.05$), to the DP plot (ANOVA for repeated measures: $F_{1.16}=0.465$, $p=0.507$; Tukey post hoc test: $p<0.05$) and to the CF plot, all obtained in control conditions (ANOVA for repeated measures: $F_{1.16}=0.465$, $p=0.507$; Tukey post hoc test: $p<0.05$) (Figs. 11A-11C).

On the contrary, the ANOVA test for repeated two-way measures has revealed statistically significant differences between the plots obtained treating the patients with device 1 (■, Figs. 12A-12C) and the plots obtained with chewing-gum mastication (▒,Figs. 12A-12C) both for the SP ($F_{6.16}=3.745$, $p=0.0022$; Tukey post hoc test: $p<0.05$), and the DP ($F_{6.16}=2.974$, $p=0.01$; Tukey post hoc test: $p<0.05$) and CF plots ($F_{6.16}=6.294$, $p<0.0001$; Tukey post hoc test: $p<0.05$).

The use of device 1 causes a mouth hyperextension, in addition to a mastication movement and an actuation of the exteroceptive sensations of the palate

periodontal front portion. The tests carried out by chewing-gum mastication excluded that the trigemino-cardiac reflex is caused by mastication.

In order to exclude that this reflex is induced by exteroceptive stimulation, the SP, DP and CF of 9 of the 20 test patients were measured before and after biting an ice cream stick, i.e. a stiff element, between the upper and lower dental arches of the front portion of the mouth. This allowed to stimulate only the exteroceptive sensations of the patients. Even in this case, DP, SP and CF were determined manually.

As shown in the diagrams of Figs. 12A to 12C, after 10 minutes of treatment with the ice cream stick (▒) no statistically significant differences of the three parameters were observed with respect to the basal values and with respect to the plots obtained in the control conditions (●) for the SP (ANOVA for repeated measures: $F_{6.16}=0.342$, $p=0.573$; Tukey post hoc test: $p<0.05$), the DP (ANOVA for repeated measures: $F_{6.16}=3.180$, $p=0.108$; Tukey post hoc test: $p<0.05$) and the CF (ANOVA for repeated measures: $F_{6.16}=0.197$, $p=0.667$; Tukey post hoc test: $p<0.05$).

On the contrary, as shown in Figs. 13A to 13C, the ANOVA test for repeated two-way measures detected statistically significant differences between the time plots obtained with device 1 (■) and the time plots obtained by using the ice cream stick (▒), both for the SP ($F_{6.22}=5.170$, $p<0.0001$; Tukey post hoc test: $p<0.05$) the DP ($F_{6.22}=4.614$, $p=0.0003$; Tukey post hoc test: $p<0.05$) and the CF ($F_{6.22}= 6.284$, $p<0.0001$; Tukey post hoc test: $p<0.05$).

Nine of the 20 test patients were treated with device 1, masticated chewing-gum and were placed in control conditions while measuring the cardiac

parameters, also with the Finapres system. The use of this system allowed continuously measuring SP, DP and CF parameters for a time not longer than 30 minutes, since the finger cuff applied to the patients may cause pain after a longer time period.

As shown by the plots of figs. 14A-14C, 15A-15C and 16A-16C, SP, DP, as well as CF data, there is a matching with the corresponding values measured by a manual sphygmomanometer.

In fact, all the plots obtained by the Finapres measurements show that there is no decrease of any of the 3 above parameters from minute 1 to minute 5 and from minute 5 to minute 15, and up to 5 minutes after end of the treatment, which corresponds to the period between point B and point 5' of the plots that summarize the sphygmomanometer measurements, as previously shown. From minute 20 of Finapres data corresponding to point 5' of the previous plots, a decreasing tendency of SP is observed, whereas in the case of DP and CF, such decrease is observed from minute 30 of the Finapres recording, which corresponds to point 15' of the previous plots.

The above described data show a statistically significant SP reduction with respect to basal values starting from 5 minutes after ending the mandibular stretching and up to 80 minutes, and show also statistically significant reduced DP and CF values with respect to the basal values starting from 15 minutes after the treatment, up to 80 minutes.

The SP, DP and CF decreases are not due to relaxed and calm conditions in which the patients were placed during the trial, since the measurements that were taken in the same conditions, but without applying device 1, did not point out any change of the parameters measured

during the whole recording. Furthermore, statistically different values are obtained if the data obtained with device 1 are compared with the data related to control conditions.

The SP decrease is observed occurring before the DP and CF decreases. This allows deducing that the mechanisms that cause such effects are different. Actually, it is possible that a more important neuronal recruiting is needed to stimulate the vasomotor centre, which can cause a CF and DP reduction, that would cause the significant reduction observed after 15 minutes after the end of the *stretching*.

The use of device 1 thus caused a new and surprising effect, i.e. a considerable reduction of the values of SP, DP and CF.

On the contrary, the data obtained in case of chewing-gum mastication, or in case of an exteroceptive stimulation of the periodontal region obtained by making the patients bite an ice cream stick between the front dental arches, did not show any significant variations of the three observed parameters, which means that the proprioceptive stimulation obtained by device 1 is the cause of a response referable to the trigemino-cardiac reflex.

The collected data globally show that a proprioceptive stimulation of the mandibular region, as obtained by device 1, causes a prolonged trigemino-cardiac reflex. Therefore, device 1 can be applied for treating, or preventing cardiac, cardiovascular, pathologies, arterial hypertension, etc.

TEST DATA (TRIAL 2)

The experiments were carried out on Wistar male mice, having a body weight of 250-300 g, and randomly

divided into the following test groups:

1. group I5min (n=9): mice subject to basal observation for 15 minutes, then to mandibular hyperextension (IM) for 5 minutes and to 80 minutes of recovery (post-IM).

2. group I10min (n=9): mice subject to basal observation for 15 minutes, then to IM for 10 minutes and to post-IM for 80 minutes.

3. group I15min (n=9): mice subject to basal observation for 15 minutes, then to IM for 15 minutes and to post-IM for 80 minutes.

4. control group (n=9): mice subject only to surgical procedure, and to microcirculation observations for 110 minutes.

A catheter was inserted into the left femoral artery for recording the systemic blood pressure and for arterial blood gas analysis, whereas a catheter was inserted into the left femoral vein for injecting a fluorescent tracer.

During the whole duration of the trials, the body temperature of the mouse was monitored and maintained at 37.0±0.5°C through a special heated stereotaxic holder.

The observation of the pial microcirculation was carried out in a cranial window opened proximate to the fronto-parietal cortex according to a method previously described by *Lapi et al.* (2007).

Briefly, a cut of 1 cm was made in the skin, in order to expose the skull. The skin edges were retracted by suture, thus forming a "well" for a perfusion liquid with two ducts, i.e. an introduction duct a discharge duct, in order to allow a flow and an inlet and outlet liquid flux, and to keep the cerebral surface continuously supplied with liquid. Finally, the craniotomy was performed by removing the bone and

cutting the dura mater away, in order to expose the pial vessels.

In the perfusion liquid [artificial cerebrospinal liquid (LCS): 119 mM NaCl, 2.5 mM KCl, 1.3 mM $MgSO_4 \cdot 7 H_2O$, 1.0 mM NaH_2PO4, 26.2 mM $NaHCO_3$, 2.5 mM $CaCl_2$ and 11.0 mM glucose], a gaseous mixture was bubbled, which contained 10% O_2, 6% CO_2 and 84% N_2 at pH 7.38±0.02, and the temperature was maintained at 37.0±0.5°C.

The stimulation of the mandibular branch of the trigeminal nerve was induced by positioning device 1, according to the invention. The opening of the mouth of the animal was fixed up to reaching a maximum allowable mandibular extension, beyond which the mandibular muscles would have been fatigued. Muscle fatigue was evaluated in preliminary trials where the mice were subjected to different opening ranges of the mouth, and the tension of the mandibular muscles was measured electromiographically.

The pial vessels were observed by a fluorescence microscopy technique for in vivo and real-time studying the microvascular changes in a specific tissue, in this case the pia mater. Fluorescence microscopy made it possible to obtain very detailed data, be measuring the diameter and the length of each pial arteriol in order to geometrically characterize the micro vascular pial network.

An optical microscope was used (Leitz Orthoplan) which was equipped with long distance lens [2.5x numerical aperture (NA) 0.08; 10x, NA 0.20; 20x,NA 0.25; 32x, NA 0.40], of a couple of eyepieces (10x) and of a system of filters. The epi-illumination was supplied by a 100 W mercury lamp comprising special fluorescein isothiocyanate filter bound with destane of molecular weight 70'000 Da (FD70, tracer used at the concentration

of 50 mg/100 g body weight) and of a thermal filter (Leitz KG1). The images of the microvascular field were taken by a high light gain digital video camera (DAGE MTI 300 RC) and recorded with a computer system (Pinnacle DC 10 plus, Avid Technology, MA, USA) for the following off-line analysis.

The microvascular measurements were made off-line by a computer system equipped with a specific software. The arteriolar pial network was geometrically characterised in basal conditions. The diameters and the length of the pial vessels were measured, by the MIP-CNR computerised method (*Lapi et al.*, 2007). Each observed microcirculation was mapped, by joining together the pictures of the vessels directly taken from the computer in stop-frame conditions.

Firstly the terminal arterioles from which the capillary vessels depart were identified, then highest order arterioles were identified and recorded. The vessels were classified following by the *Strahler* method according to the diameter.

At the end of the recovery period, the red blood cells speed was measured at a capillary level by a MIP computer system in an observed area of the cranial window of 150x150 μm, and expressed in mm/s.

Finally, the average systemic blood pressure (PAm) was recorded as the arithmetic mean between diastolic and systolic pressure, by an electronic system (Pressure Monitor BP-1, WPI) connected to the catheter which is located in femoral artery; the values of the ematic gas were determined every 30 minutes on blood arterial samples (ABL5; Radiometer, Copenhagen, Denmark).

The data are expressed as ±E.S average. For the Gaussian distribution analysis of the data the Kolmogorov-Smirnov method was used. Since the data had a

normal distribution, the groups were compared by a parametric test (ANOVA and Bonferroni post-hoc test). The statistical significance was fixed at $p<0.05$.

In all the 36 mice (n=36) which were used for the investigation, the arterioles of the pial microcirculation were classified according to the diameter and to the length using the images recorded in the first 15 minutes of (basal) observation. The capillary vessels were classified as 0-order vessels, whereas terminal arterioles, from which the capillary vessels depart, were classified as order 1 vessels. Then, backwards along the arteriolar tree, larger vessels were present, which were classified in increasing order, up to arterioles of order 5, which are the largest vessels that were observed in the tests.

As shown in table 1 below, vessels of all 5 orders are present in the sample, in different amounts: since order 5 vessels are located peripherally in the cranial window, it was difficult to observe them and therefore they are not present in all the tests, while the order 2 vessels were present in a greater amount.

Table 1 hereafter shows the total number of the arterioles belonging to the five orders, the diameter and the length of the vessels. The diameter ranges do not overlap ($p<0.05$ [*], ANOVA and Bonferroni post-hoc).

Table 1

Order	Arterioles (N)	Diameter (μm)	Length (μm)	Subject (N)
5	16	62.0±5.0	1.169±420	36
4	35	46.0±3.5	987±250	36
3	87	62.0±3.6	481±120	36
2	148	62.0±2.4	360±100	36
1	120	62.0±2.5	140±83	36

Group I5min, consisting of mice treated with 5 minutes of mandibular hyperextension (IM) (n=9) has shown an average systemic blood pressure (PAm) of 110±2 mmHg in basal conditions. During the application of the divaricator, PAm is reduced and reaches after 5 minutes of the IM a value of 99±2.1 mmHg, which is significantly lower than the basal value(ANOVA and Bonferroni post-hoc test, $p < 0.05$; variation ratio:10±2%) (Fig. 17).

Upon removal of device 1, the trend of the pressure became complex: in the first 5 minutes post-IM, PAm value was significantly increased with respect to the basal values (122±2.3 mmHg, which is about 11±3% of the basal value, (ANOVA and Bonferroni post-hoc test, $p<0.05$); at 15 minutes post-IM it was decreased down to basal values of 110.2±2 mmHg, and then significantly increased again up to 137.5±2.4 mmHg (25±4% with respect to the basal value, ANOVA and Bonferroni post-hoc test, $p<0.05$), at 45 minutes post-IM. After this period, PAm progressively decreased down again to the basal values of 110.1±1.8 mmHg, at 80 minutes (Fig. 17).

At the cerebral level, the arterioles of all 5 orders had shown a not significant diameter decrease (ANOVA and Bonferroni post-hoc test, p=ns) during the 5 minutes of the IM: arterioles of order 5 changed from an average diameter in basal conditions of 57±1.2 μm to a diameter of 55.2±0.9 μm after 5 minutes of the IM. Order 4 arterioles changed from 46.6±1.1 μm in basal conditions to 45.2±0.8 μm, which corresponds 3±1% of the basal value; order 3 arterioles changed from 34.2±1.1 μm in basal conditions to 34.2±1.1 μm, order 2 arterioles changed from 24±0.9 μm in basal conditions to 22.5±0.6 μm and order 1 arterioles changed from 11±0.2 μm in basal conditions to 10.3±0.1 μm, thus showing a reduction of about 6±2% of the basal values for the

three last orders (Figs. 18A-18E).

In the first 5 minutes post-IM, the pial arterioles with diameter of order 5 and 4 significantly increased up to 65.5±1.1 μm and to 53.9±0.9 μm, respectively (ANOVA and Bonferroni post-hoc test, $p<0.05$), which corresponds to a variation ratio, with respect to the basal value, of 15±3% and of 16±3%, respectively. The diameters of order 3, 2 and 1 arterioles increased significantly to 42±0.9 μm, to 29.4±0.8 μm and to 13.4±0.1 μm, respectively (ANOVA and Bonferroni post-hoc test, $p< 0.05$), which corresponded to a variation ratio of 23±4%, 22±4% and 22±4%, respectively. Such increased value was maintained up to 45 minutes post-IM, then the arterioles progressively recovered their basal diameter at 80 minutes (Figs. 18A-18E).

Furthermore, the speed of the red blood cells in the capillary vessels as calculated at the end of the observation (0.22±0.02 mm) was not significantly different from the value that was observed in the controls (ANOVA and Bonferroni post-hoc test, p=ns).

To sum up, during the 5 minutes of the IM treatment, a considerable PAm decrease was observed associated with a vasoconstriction of the pial arterioles, which turned out to be significant in the lowest order vessels. During post-IM step, the PAm fluctuated and then returned to the basal value, after 80 minutes. The pial arterioles dilated during the first 45 minutes post-IM and at 80 minutes returned to the diameter basal value. The arteriolar diameter change was significant in the lowest order arterioles.

In Fig. 19 the PAm time plot is compared with the diameter change of the order 2 arterioles. The two parameters appears not to have similar time plots.

Group I10min consisted of animals which were treated

with IM for 10 minutes (n=9) after having been observed for 15 minutes in basal conditions (106±2.5 mmHg PAm value). In these mice, PAm significantly decreased down to 93.3±2.5 mmHg in the first 5 minutes of the IM, (ANOVA and Bonferroni post-hoc test, $p<0.05$), which corresponds to a reduction ratio of 12±3% with respect to the basal values observed in I5min, and in the following 5 minutes, PAm continued decreasing down to the value of 83.7±2.0 mmHg (ANOVA and Bonferroni post-hoc test, $p<0.05$ vs basal), which is 21±2% lower than the basal conditions: such decrease went on for all the post-IM period and at 80 minutes it was still 83.8±2.3 mmHg, which is significantly less than the basal values (ANOVA and Bonferroni post-hoc test, $p<0.05$) (Fig. 20).

During the first 5 minutes of the IM, the pial arterioles with diameter of order 5 and 4 (average diameter: 64±1.1 μm and 45.2±1.1 μm, respectively, in basal conditions) were significantly decreased down to 58.8±1.1 μm and to 41.4±1.1 μm, respectively (ANOVA and Bonferroni post-hoc test, $p<0.05$), which corresponds in both cases to a variation ratio of 8±3% with respect to the basal values. The arterioles with diameters of order 3, 2 and 1 (average diameter in basal conditions: 31±0.8 μm, 24.8±0.7 μm and 16.5±0.3 μm, respectively) decreased significantly down to 25.4±0.7 μm, 20±0.7 μm and 13.2±0.3 μm, respectively (ANOVA and Bonferroni post-hoc test, $p<0.05$), which corresponds to a variation ratio of 18±3%, 19±3% and 20±3% of the basal values, respectively. For all 5 orders of the arterioles, the reduction of the diameter recorded in the first 5 minutes of the IM remained substantially unchanged in the subsequent 5 minutes (Fig. 20).

In the post-IM phase, a progressive increase of the diameter of the pial arterioles was observed during the

first 45 minutes. The diameter order 5 and 4 arterioles significantly increased up to the values of 70.4±1.1 μm and 49.6±1.2 μm, respectively (ANOVA and Bonferroni post-hoc test, $p<0.05$), already at 5 minutes of recovery, which is equal to an increase ratio of about the 21±2% of the basal values ($p<0.05$), then values of 78±0.9 μm and 54.6±1 μm, respectively, were achieved at 45 minutes, and such dilation level was maintained during the remaining 35 minutes of observation.

The diameter of order 3, 2 and 1 arterioles progressively increased up to the values of 40.6±0.7 μm, 32.4±0.5 μm and 21.6±0.5 μm, respectively (ANOVA and Bonferroni post-hoc test, $p<0.05$), which corresponds to a variation ratio of 30±2%, 30±2% and 31±2% of the basal values, respectively, at 45 minutes post-IM. Such a dilation was kept until the end of the observation period, when the diameters were 40.3±0.9 μm (order 3), 32.6±0.6 μm (order 2) and 21.6±0.5 μm (order 1).

The vasodilatation determined a significant increase of the speed of the red blood cells in the capillary vessels, with respect to the controls. Such speed, calculated at the end of the observation, was 0.35±0.03 mm/s (ANOVA and Bonferroni post-hoc test, $p<0.05$).

Therefore, for 10 minutes of the IM a significant PAm decrease was observed associated with a significant reduction of the diameter of the pial arterioles. In the post-IM step, while the pressure values remained significantly low, the arteriolar diameters underwent a remarkable vasodilatation, which particularly concerned order 2 arterioles, as shown in Fig. 22.

The mice treated with IM for 15 minutes after an observation period of 15 minutes in basal conditions, have shown in the first 10 minutes of the IM a progressive PAm decrease down to a value of 90±2.1 mmHg,

which is significantly less than the basal values (PAm basal: 114±2.5 mmHg; ANOVA and Bonferroni post-hoc test, $p<0.05$), and which was substantially maintained during the following 5 minutes of treatment and all the post-IM period (Fig. 23).

The diameters of order 5 arterioles (basal diameter: 60±1 μm) and order 4 arterioles (basal diameter: 47±0.9 μm) had a progressive decrease with respect to the basal values, which was significant even after only 5 minutes of IM (57±1.1 μm and 43.3±0.9 μm, respectively, ANOVA and Bonferroni post-hoc test, $p<0.05$), and which was maintained in the following 10 minutes of IM; then, a significant increase followed (ANOVA and Bonferroni post-hoc test, $p<0.05$) that after 5 minutes brought the diameter of the vessels up to 65.7±0.8 μm and to 50.8±0.8 μm, respectively, and after 45 minutes up to 69.4±0.7 μm and 54.5±0.9 μm, respectively. Such increase went on during the following 35 minutes post-IM. Even order 3 arterioles (basal diameter: 33.3±0.7 μm), order 2 arterioles (basal diameter: 22.5±0.6 μm) and order 1 arterioles (basal diameter: 14±0.5 μm) shown a progressive decrease with respect to the basal values, which was significant already ever since after 5 minutes (27.6±0.7 μm, 18.2±0.6 μm and 11.2±0.4 μm, respectively; ANOVA and Bonferroni post-hoc test, $p<0.05$), the decreased value remained the same during the following 10 minutes of the IM, then a significant increase followed (40.1±0.7 μm, 27.2±0.7 μm and 16.9±0.5 μm, respectively; ANOVA and Bonferroni post-hoc test, $p<0.05$) ever since at 5 minutes post-IM. At 45 minutes, the arteriolar diameters had increased to the values of 43±0.9 μm, 29.3±0.7 μm and 18.3±0.6 μm, respectively (ANOVA and Bonferroni post-hoc test, $p<0.05$); these values were maintained until the end of the observation

period (Fig. 19).

Furthermore, at 80 minutes post-IM, the speed of the red blood cells in the capillary vessels had significantly increased with respect to the controls (0.33±0.04 mm/s; ANOVA and Bonferroni post-hoc test, $p<0.05$).

In conclusion, for 15 minutes IM PAm significantly decreased, as well as the diameter of the pial arterioles. After removing the divaricator, PAm maintained values significantly lower than basal values, and the arterioles shown a remarkable vasodilatation which concerned more significantly order 2 arterioles, as shown in Figs. 24A to 24E.

Finally, in 9 mice that were observed for about 110 minutes without undergoing any IM action, it was observed that PAm maintained substantially the same value during the whole duration of the observation, and at the cerebral level each order of arterioles did not show any significant diameter change (Figs. 26A to 26E).

On the contrary, The pial arterioles showed the normal rhythmic diameter fluctuations (vasomotion) (table 2), which disappeared, instead, in case of vasodilation or in case of vasoconstriction. Furthermore, all the capillary vessels were perfused and the speed of the red blood cells remained at 0.23±0.04 mm/s.

Table 2 hereafter shows the fluctuating values of the diameters of the observed vessels of each order.

Table 2

Order	Frequency (Hz)	Amplitude (% average diameter)
5	0.055±0.002	10-30
4	0.020±0.050	30-50
3	0.030±0.020	60-80
2	0.120±0.050	60-80
1	0.150±0.050	60-80

The physiological parameters recorded at the beginning and at the end of the observation, such as hematocrit level, pH, pCO_2 and pO_2, did not immediately show significant change in the different test groups (data not shown).

The above described test data showed a new and surprising effect of mandibular hyperextension (IM) on the systemic blood pressure and on pial microcirculation and, therefore, they showed what modifications occur in the cerebral microcirculation as a consequence of a proprioceptive stimulation of the trigeminal nerve in terms of variations of the diameter of the pial arterioles.

The test data were obtained using the mouse pial microcirculation, which represents a valid experimental model for observing in vivo the cerebral blood flow, since it is characterised by surface arterioles, anastomotic arterioles and penetrating arterioles which attain the underlying cortical layer; therefore, the study of these vessels provides an indirect evaluation of the blood flow even at the cortical level. Furthermore, since the cerebral vessels of the mouse have similitude with human, it is possible to obtain test data that may be extended to humans (*Lee, 1995*).

Normally, the non-invasive imaging methods used for studying in vivo the cerebral flow, such as Laser Doppler and functional magnetic resonance, do not provide the flow value in a predetermined region of the cerebral tissue and do not discriminate between a type of vessel and another.

The results showed that the proprioceptive stimulation of the trigeminal nerve causes a decrease of the systemic blood pressure and a tone alteration of the pial arterioles during the application of the divaricator, which causes IM. In the post-IM phase a complex response resulted according to the duration of the treatment. The real duration of the effects may be even longer. The observations continued up to 80 minutes post-IM, and since the results could have been observed even for a longer time, further tests can be carried out in order to determine how long after IM treatment a reversion of the effects may be observed.

In particular, it was observed that the proprioceptive stimulation of the trigeminal nerve, independently from the duration of the IM, causes a significant decrease of the systemic blood pressure and a decrease of the diameter of the pial arterioles during the whole IM treatment, which is more important for lowest order vessels. In the post-IM period, instead, the trend of the pressure diverged from the trend of the variation of the arteriolar diameters and turned out to be strictly related to the duration of the IM. In fact, after 5 minutes of IM, the systemic blood pressure showed a three-step trend, characterised by an increase, a decrease, a new increase which lasted until 45 minutes post-IM, and a return to basal values at 80 minutes. The diameter changes of the pial arterioles have shown a more regular trend, since the diameters increased until

45 minutes post-IM, and then decreased and attained the basal values again, at 80 minutes.

With longer IM (10 and 15 minutes) the systemic blood pressure remained unchanged at significantly lower values than the those observed in basal conditions, both during IM and during the 80 minutes of post-IM observation.

The investigation of the arteriolar network made it possible to observe a significant increase of the arteriolar diameter, during the whole post-IM period, which was more important in the lowest order arterioles. The dilation, in turn, was associated with an increased flow in the capillary vessels.

Therefore, the response of the cerebral vessels to a reduction of the systemic blood pressure induced by a proprioceptive stimulation of the trigeminal nerve is much more complex than what the regulation mechanisms of the cerebral circle could lead to suppose. Actually, the initial pressure fall observed during IM causes at first a vasoconstriction and only afterwards a vasodilation.

The vasoconstriction observed during the IM period is probably due to the activation of the trigeminal proprioceptive afferences, with the activation or liberation of vasoconstrictors mediators. It is important to note that, during this period, the systemic blood pressure decreases. It is therefore possible to suppose also that an activation of the baroceptive reflex may take place. Afterwards, during the post-IM period, the decrease of the systemic blood pressure is associated with a vasodilatation of the cerebral arterioles. Such regulation response of the vascular tone could be caused by the liberation of acetylcholine which acts on the vascular endothelium and releases the relaxing factor (EDRF). The increased blood flow within

the vessels could induce endothelial NOS activation, as previously shown by Iadecola et al. (1994).

The nitric oxide, once liberated by the endothelial cells, could in turn induce the activation of a soluble guanylate cyclase which would activate the mechanism of cGMP formation in the vascular smooth muscle cells with a subsequent reduction of calcium fluxes and the relaxation of the smooth muscle.

As well known from the scientific literature, an increase of the neuronal activity caused by the stimulation of the trigeminal nerve is associated with an early release of neuronal nitric oxide that, as a consequence of its high reactivity, is more effective on the arteries immediately adjacent to the activated neuronal pool. Furthermore, most arterioles of the lowest orders (2 and 1) penetrate into the underlying cortical layer and are therefore at direct contact with the cortical neurons. This could explain why the smallest arterioles are more reactive than those having a larger diameter. The higher dilation of the lowest order arterioles could assist a more intense flow distribution at the most active cortex regions.

The data recorded in this study about the variation of the systemic blood pressure matches with what was previously observed in normal pressure subjects, where an IM of 10 minutes caused significant and long-lasting reduction of the systolic and diastolic pressure, and of the cardiac frequency. The data concerning the IM-induced arteriolar diameter changes indicated that the proprioceptive activation of the trigeminal nerve causes a typical tone modulation of the cerebral arterioles, which is probably enhanced by specific mechanisms that seem partially different from the mechanisms that are actuated at cerebral level when a

peripheral nerve is stimulated. As well known, an electric stimulation of the femoral nerve causes a dilatation of the pial arterioles, both during the treatment and in the post-stimulation period.

Therefore, it seems that specific regulation mechanisms of the cerebral circle are activated during the proprioceptive stimulation of the trigeminal nerve, which may be interesting to investigate more in detail.

The above commented data allow concluding that, device 1, according to the invention, makes it possible to non-invasively prevent or delay the onset of such diseases as hypertension, as well as the risk of cerebral stroke.

The foregoing description of exemplary embodiments will so fully reveal the invention according to the conceptual point of view, so that others, by applying current knowledge, will be able to modify and/or adapt such exemplary embodiments for various applications, without further research and without parting from the invention, and it is therefore to be understood that such adaptations and modifications will have to be considered as equivalent to the specific embodiment. The means and the materials to perform the different functions described herein could have a different nature without, for this reason, departing from the scope of the invention. It is to be understood that the phraseology or terminology that is employed herein is for the purpose of description and not of limitation.

CLAIMS

1. Device for treating or preventing cerebral diseases, arterial hypertension, cerebral stroke, neurodegenerative diseases, **characterised in that** it comprises a means for causing an alternating hyperextension of the jaw, in order to cause a cyclical extension of the mandibular elevator muscles with a non-invasive proprioceptive stimulation of regions that are innervated by the trigeminal nerve and with a subsequent stimulation of the trigemino-cardiac reflex.

2. Device, according to claim 1, wherein said means for causing a hyperextension of the jaw comprises:
- a first portion that is arranged to be brought into contact with the upper wall of the oral cavity of the patient;
- a second portion that is arranged to be brought into contact with the lower wall of the oral cavity of the patient;
- a connection portion arranged between the first and the second portions, said connection portion arranged to apply a contrast force upon approaching of said first and second portions.

3. Device, according to claim 2, wherein said connection portion is arranged to maintain said first and second portions at a rest distance and to allow said first and second portions to approach each other upon application of a closing force from said rest distance to a maximum closing distance, wherein said rest distance corresponds to the distance between the lower wall of the oral cavity and the upper wall of the oral cavity at the height of the inner dental arch of a patient at a maximum mandibular extension.

4. Device, according to claim 3, comprising an adjustment

means of said rest distance.

5. Device according to claim 2, wherein said contrast force has a maximum value set between 14N and 25N.

6. Device according to claim 3 wherein said adjustment means of said rest distance is housed in a handgrip.

7. Device, according to claim 1, wherein said connection portion is made of a resilient material.

8. Device, according to claim 1, wherein said means for causing a hyperextension of the jaw comprises:

- at a first end, a first substantially "U"-shaped laminar portion, said first laminar portion arranged to be brought into contact with the upper wall of the oral cavity of the patient;
- at a second end, a second substantially "U"-shaped laminar portion, said second laminar portion arranged to be brought into contact with the lower wall of the oral cavity of the patient;
- a laminar connection portion connecting said first and said second shaped laminar portions, said laminar connection portion made of a resilient material.

9. Device, according to claim 3, wherein said lamina has a thickness set between 1 mm and 2 mm.

10. Device, according to claim 3, wherein said lamina has a length set between 15 cm and 25 cm.

11. Use of the device according to any of claims 1 to 5 for treating or preventing cerebral diseases.

12. Use of the device according to any of claims 1 to 5 for treating or preventing hypertension.

13. Use of the device according to any of claims 1 to 5, for treating or preventing cerebral stroke.

14. Use of the device according to any of claims 1 to 5, for treating or preventing neurodegenerative diseases.

Fig. 1

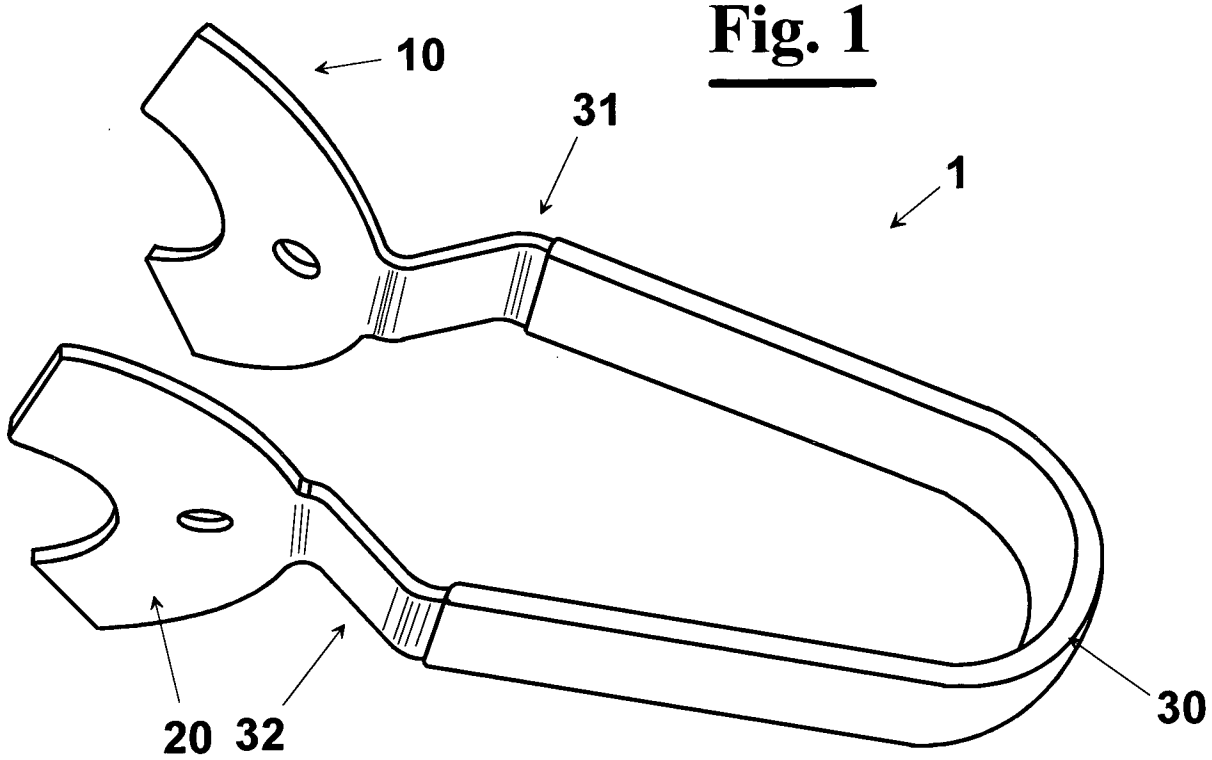


Fig. 2

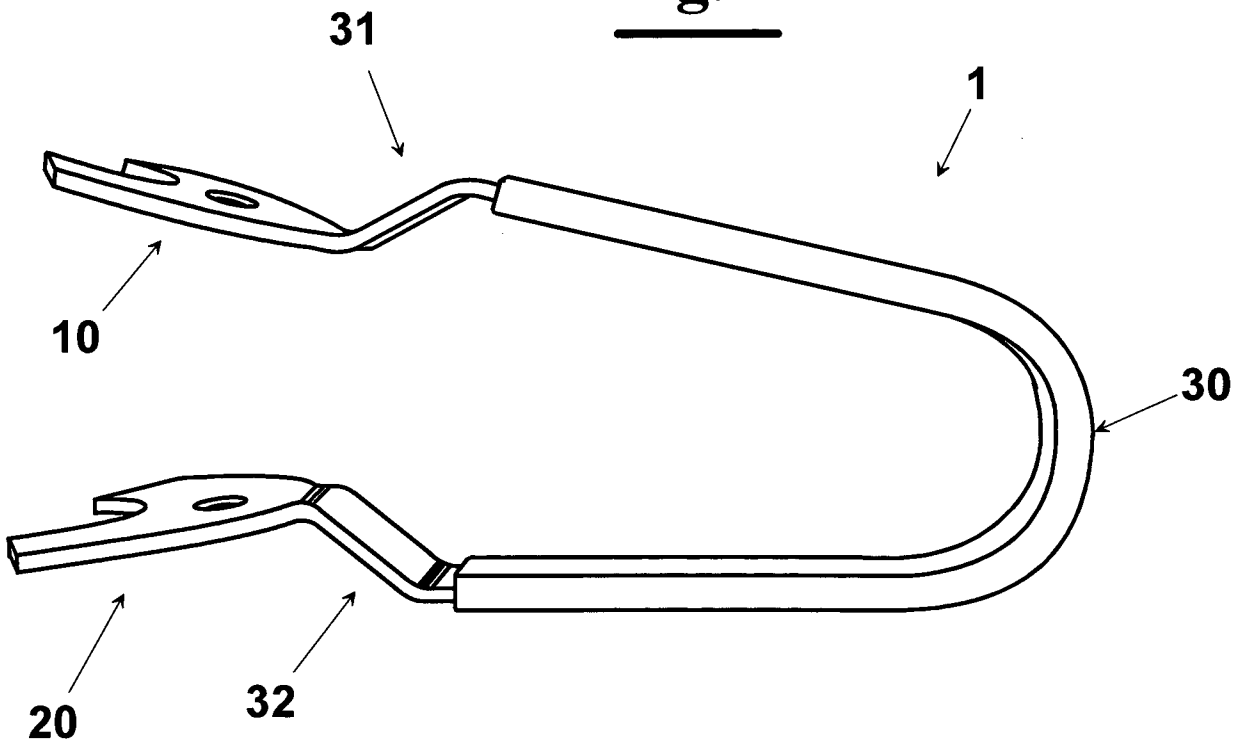


Fig. 3

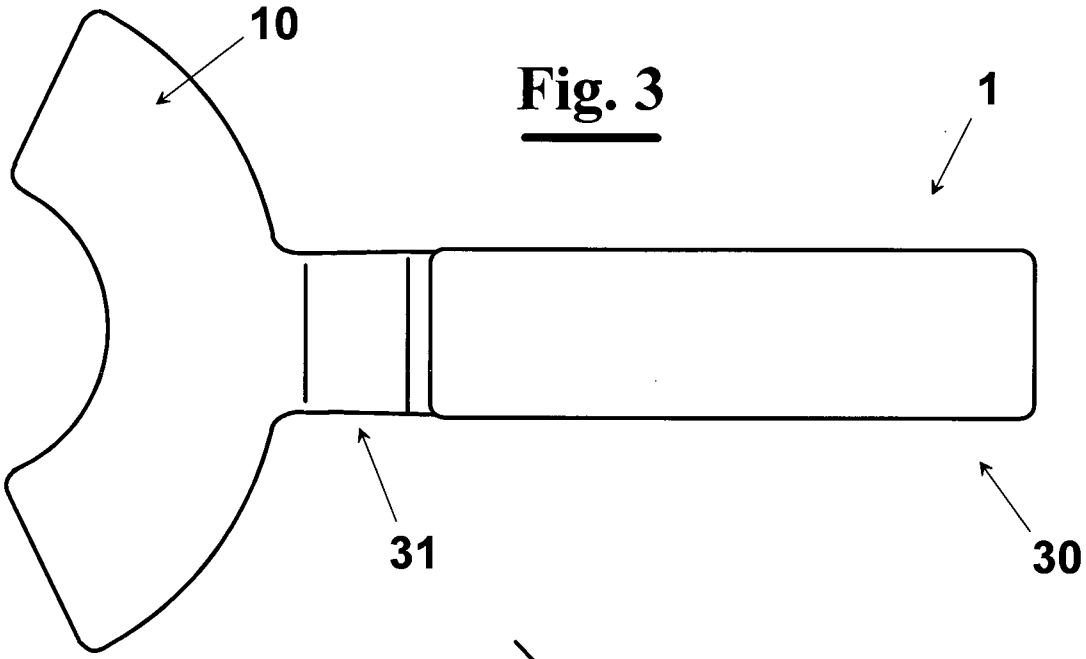


Fig. 4

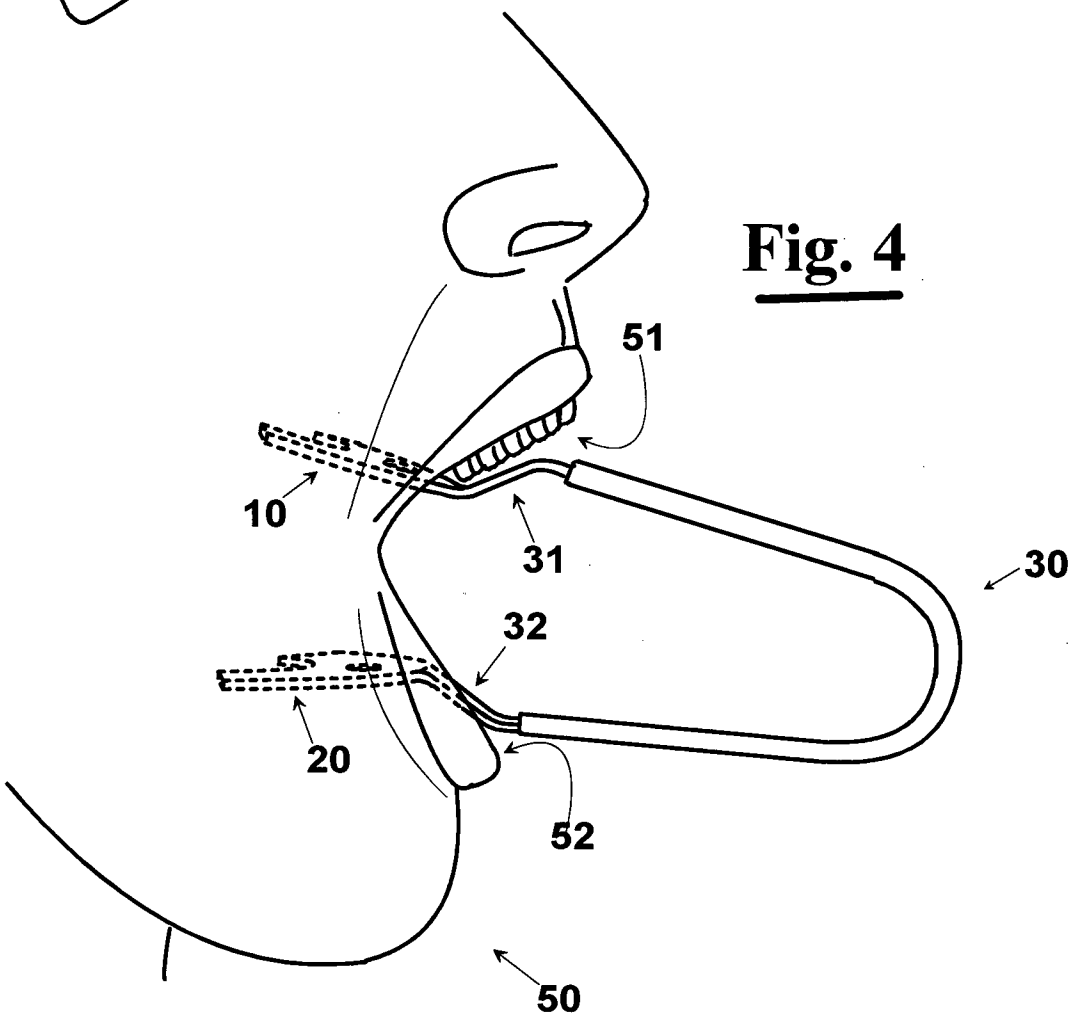


Fig. 5

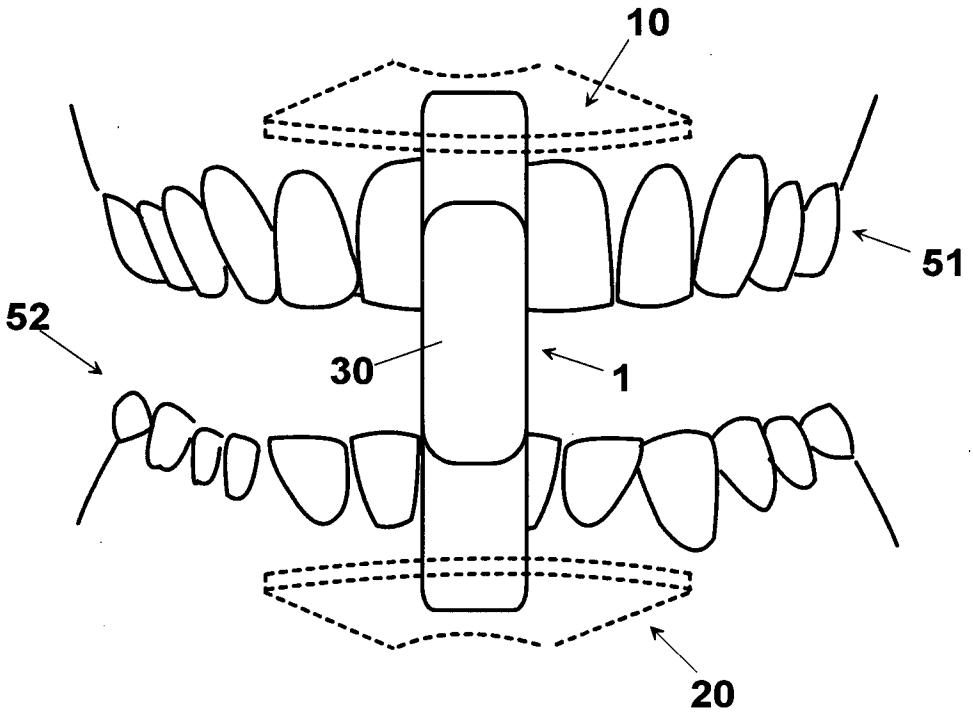
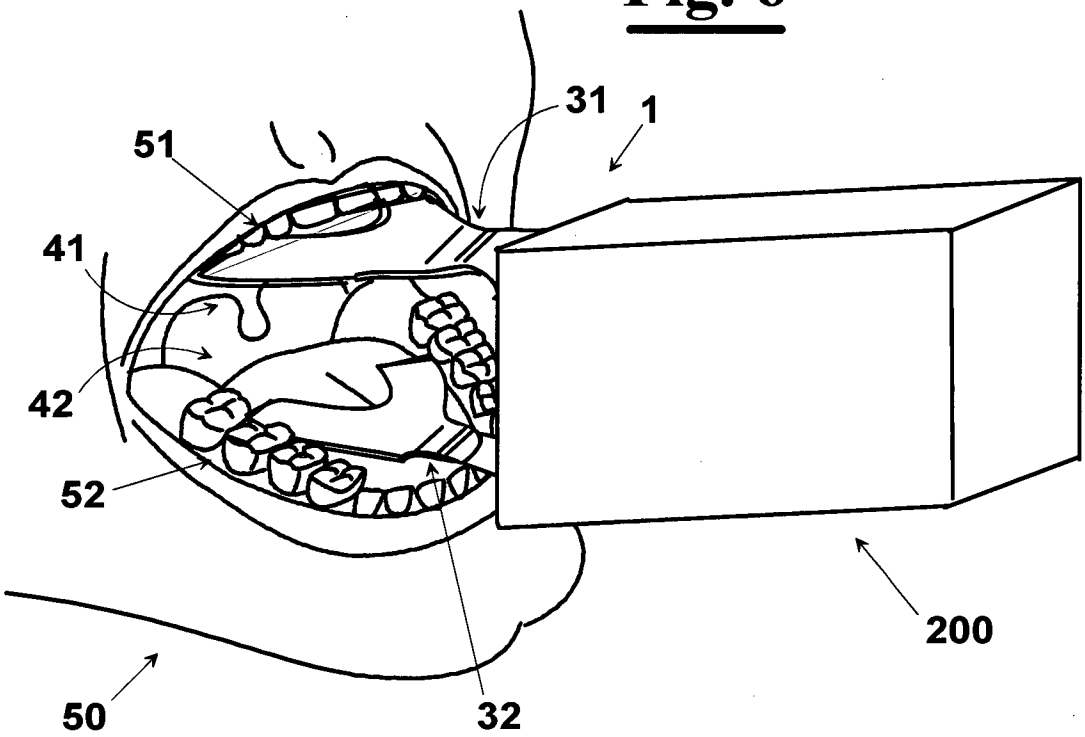


Fig. 6



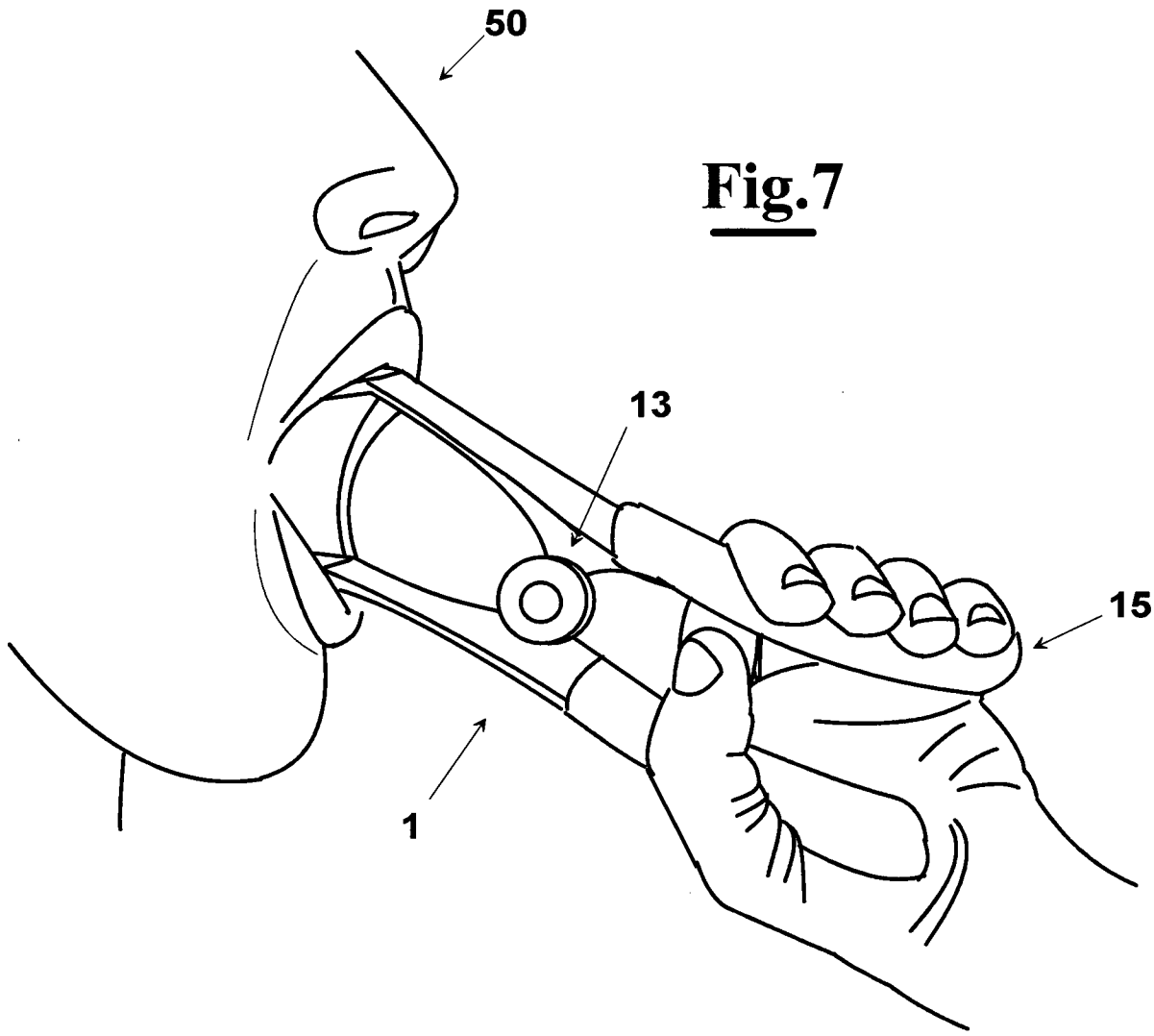


Fig.7

Fig. 8A

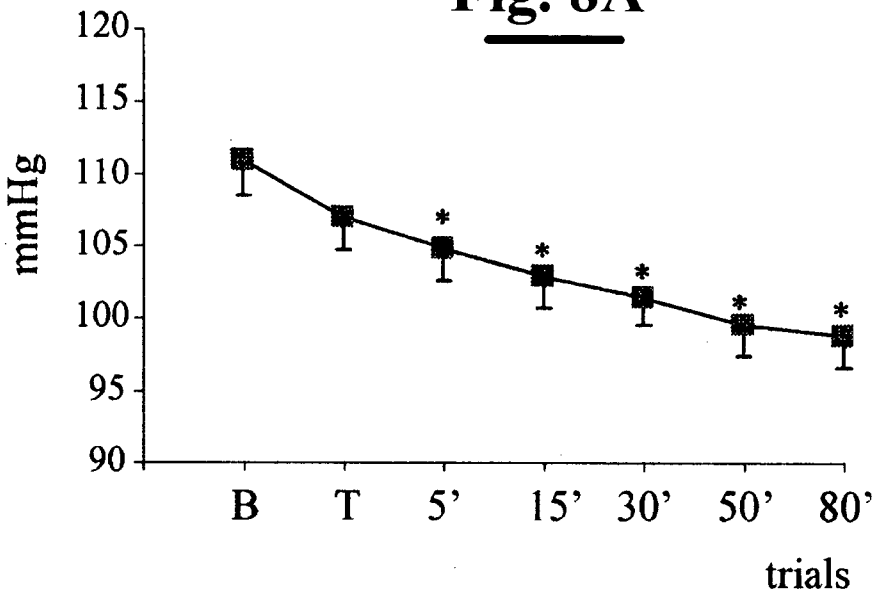


Fig. 8B

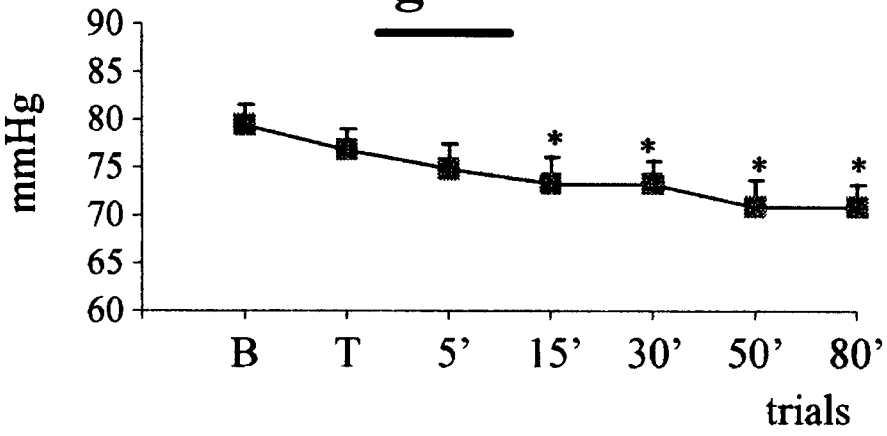


Fig. 8C

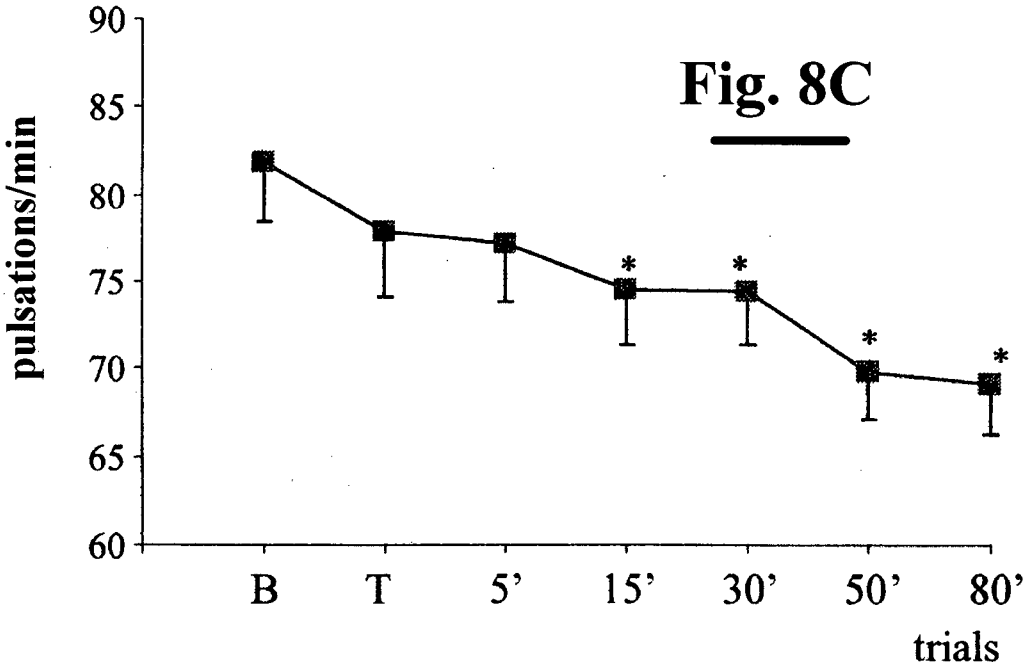


Fig. 9A

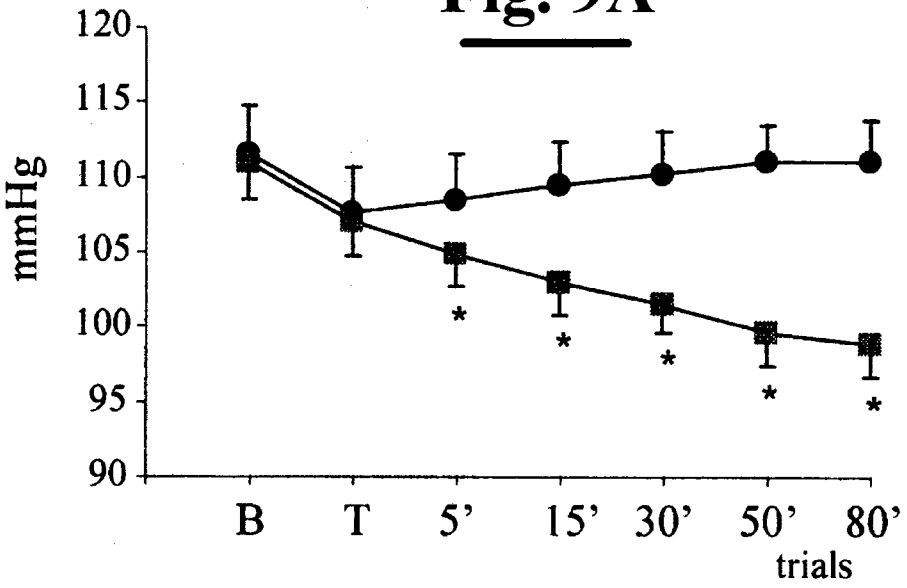


Fig. 9B

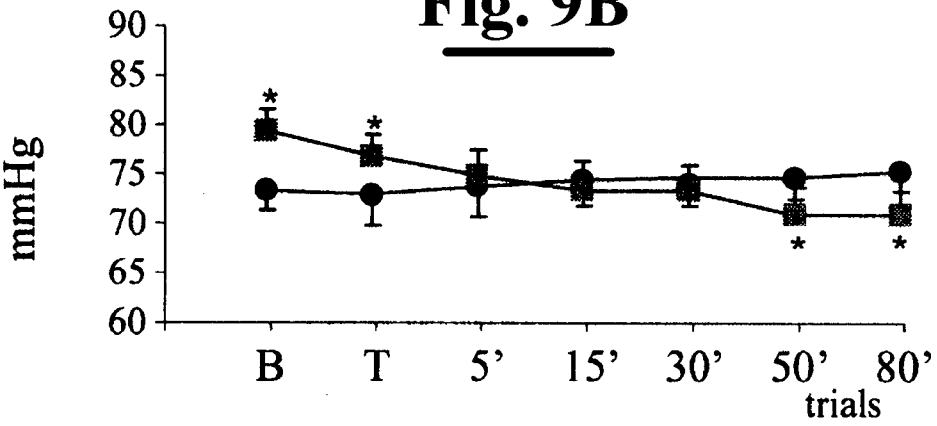


Fig. 9C

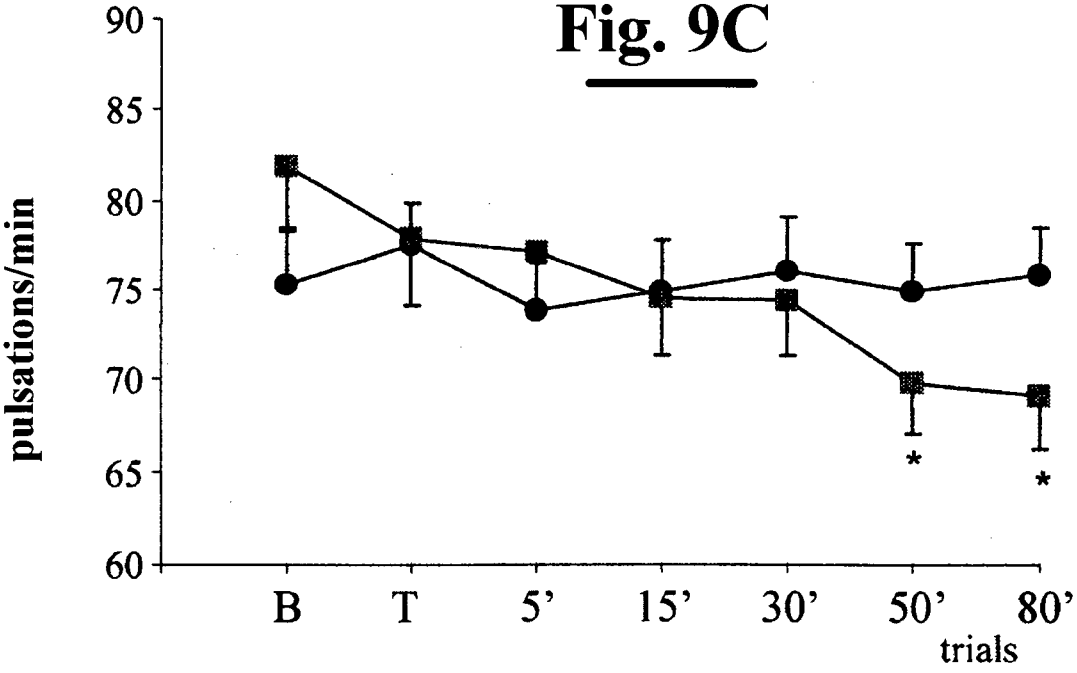


Fig. 10A

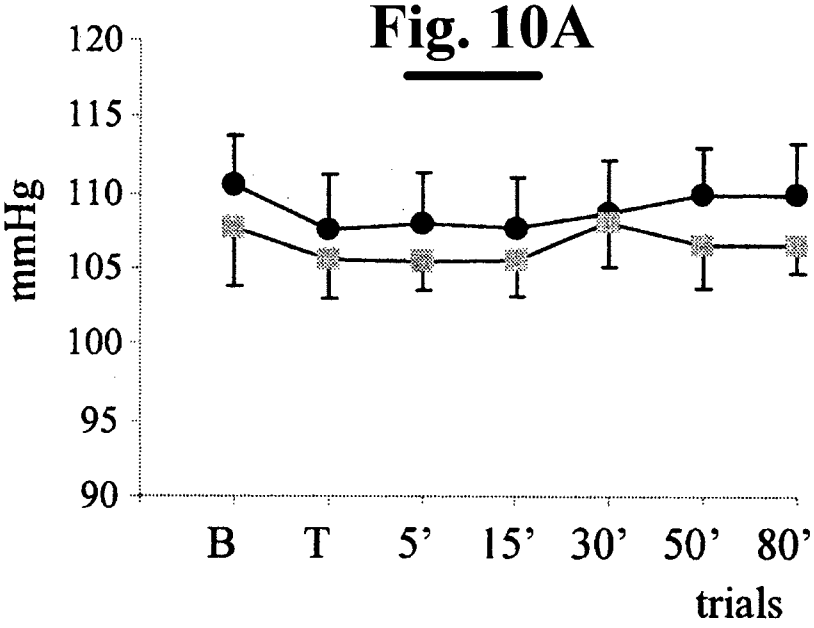


Fig. 10B

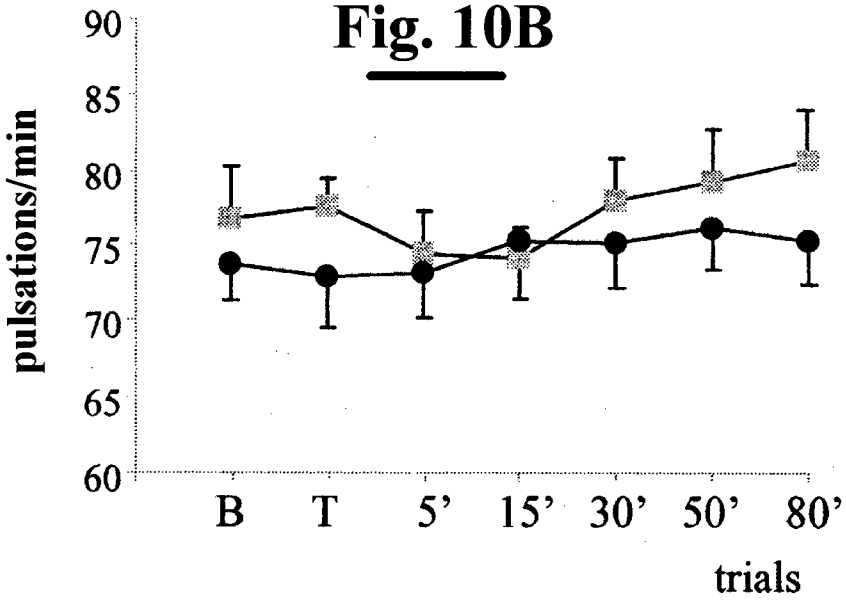


Fig. 10C

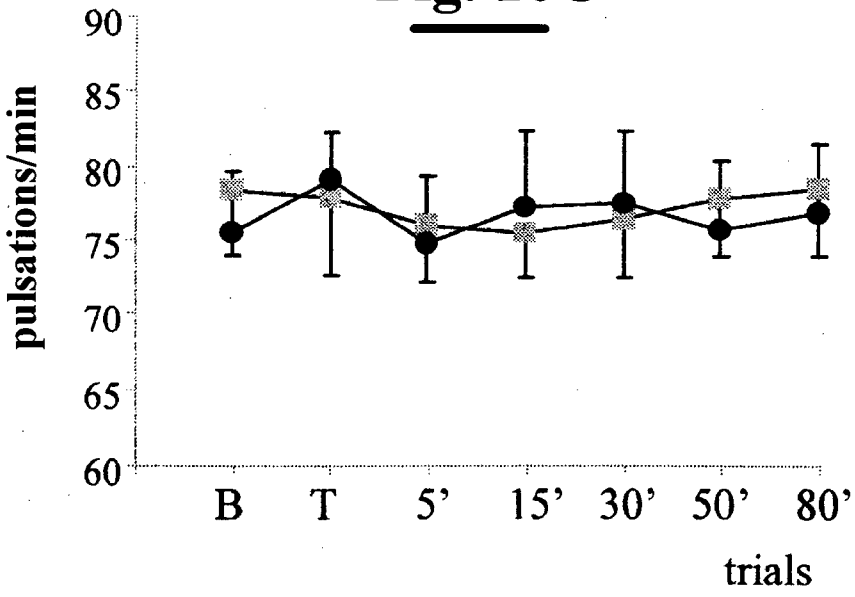


Fig. 11A

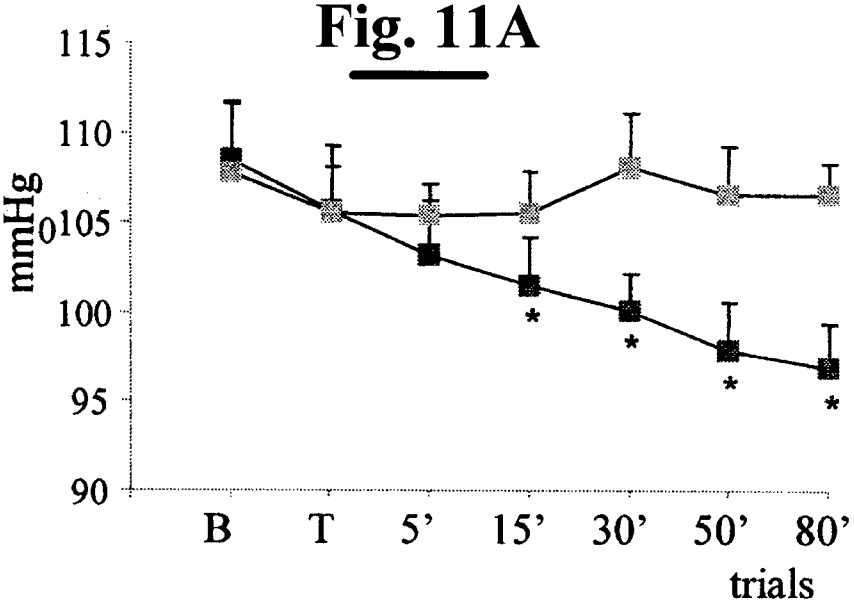


Fig. 11B

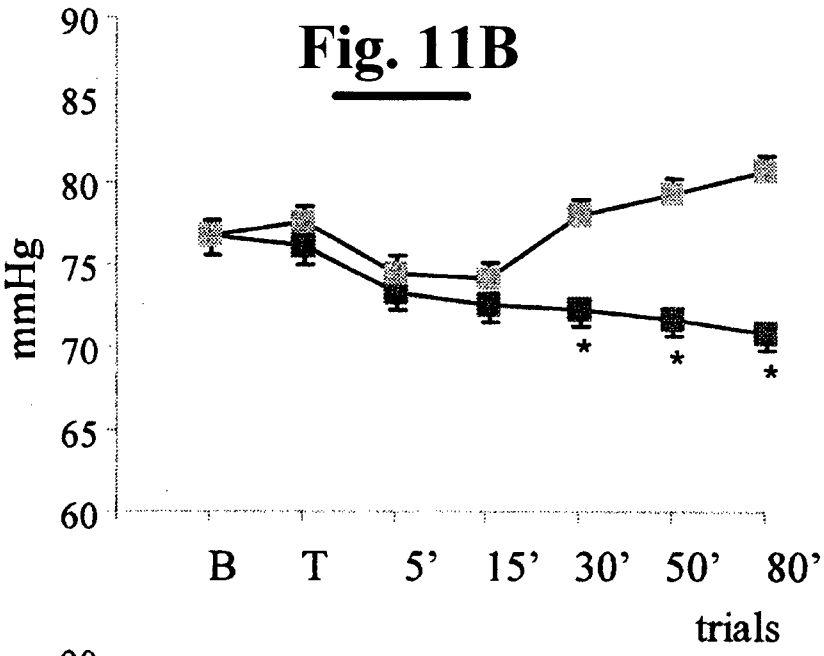


Fig. 11C

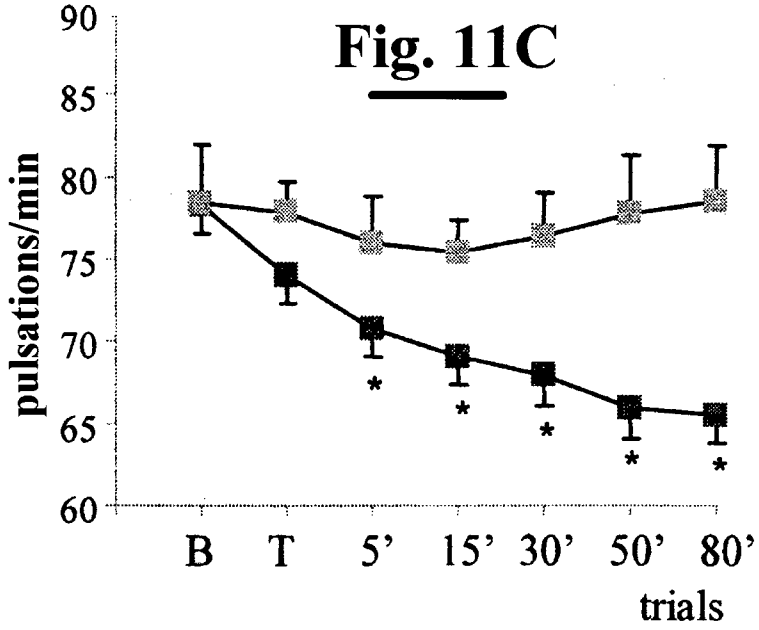


Fig. 12A

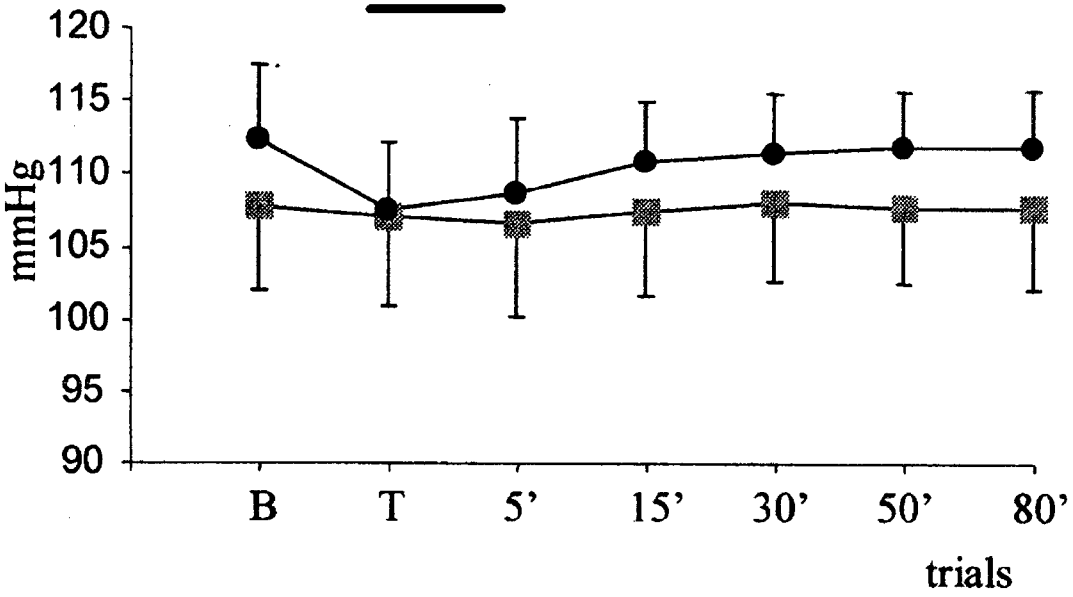


Fig. 12B

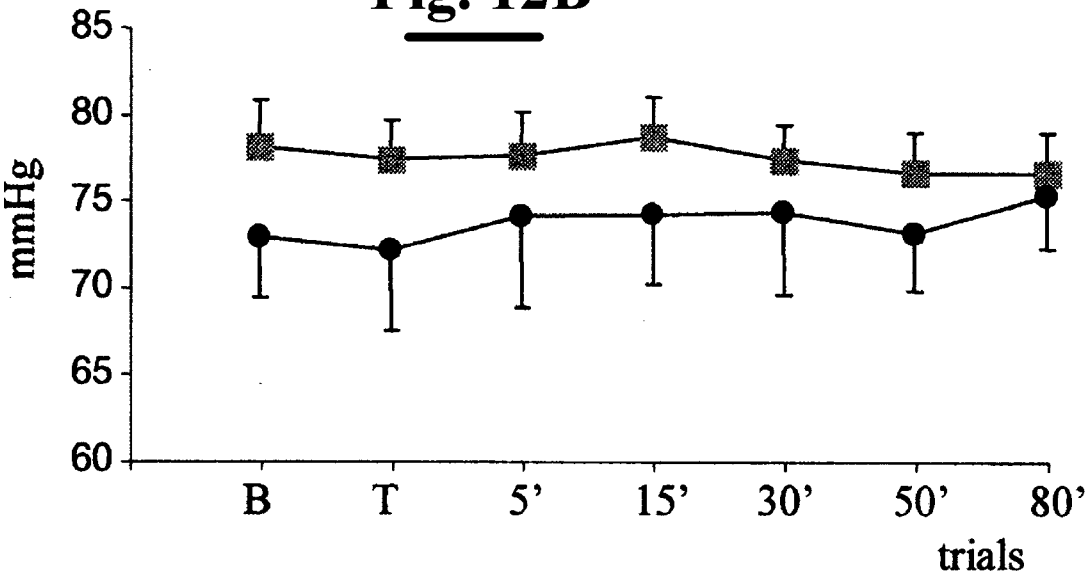


Fig. 12C

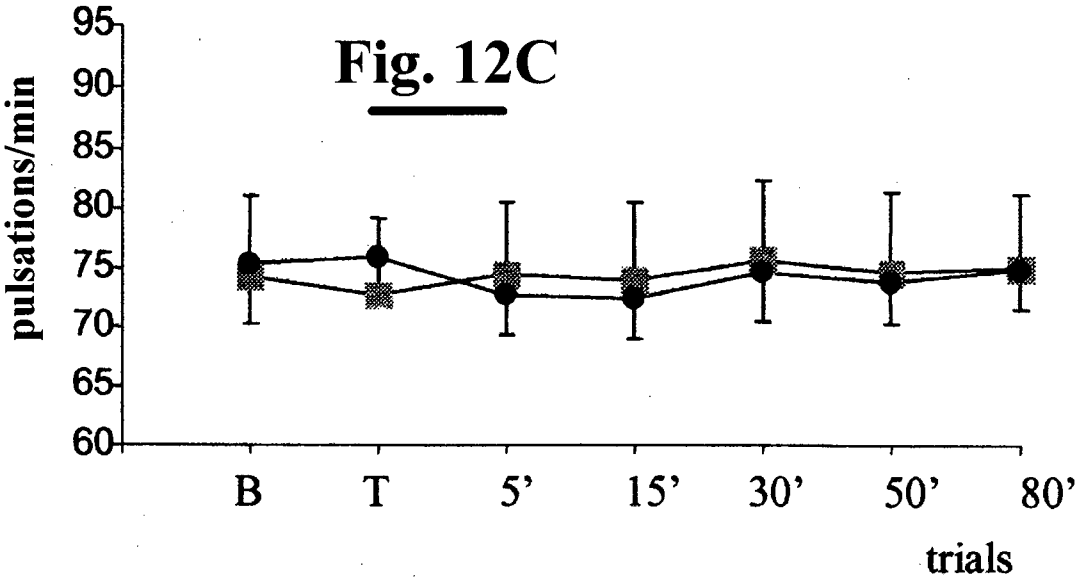


Fig. 13A

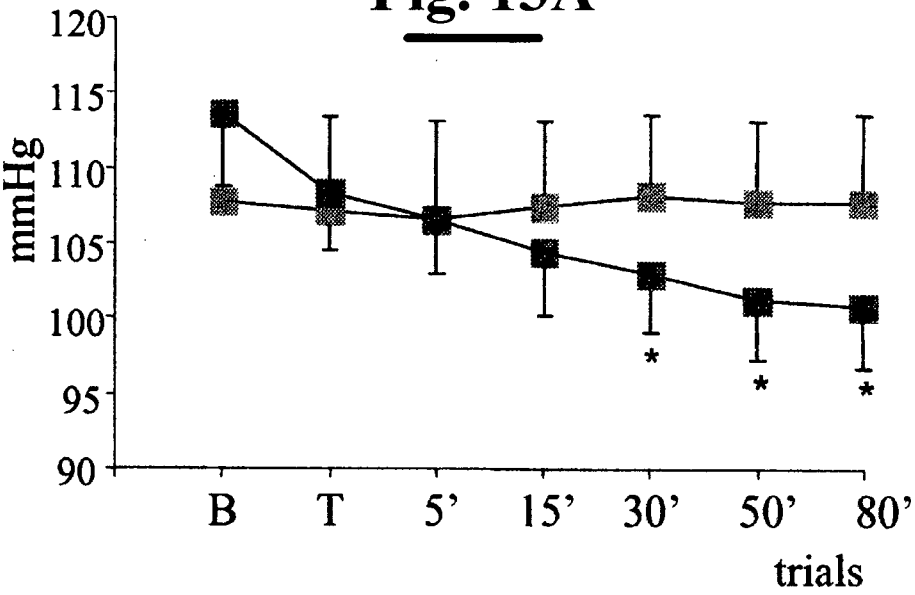


Fig. 13B

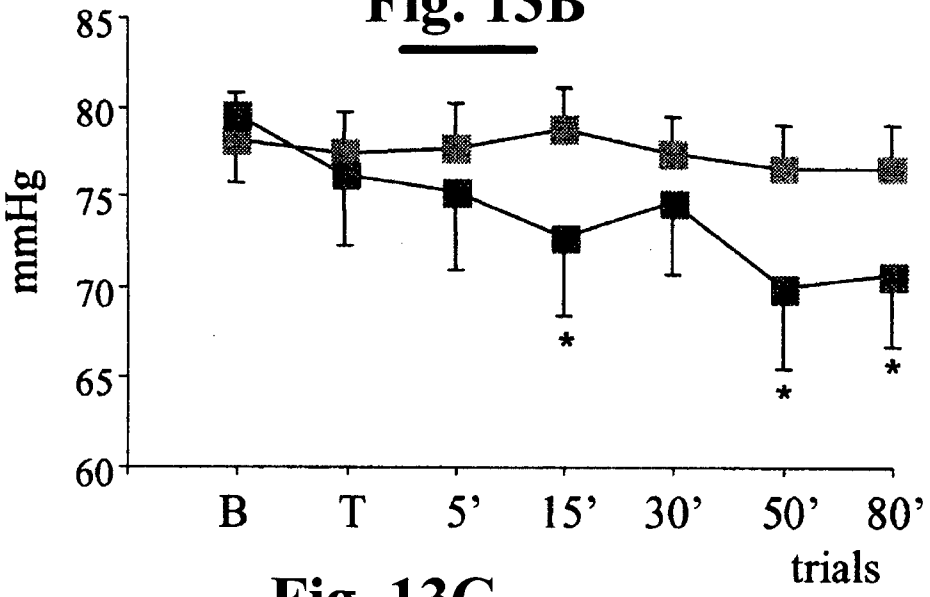


Fig. 13C

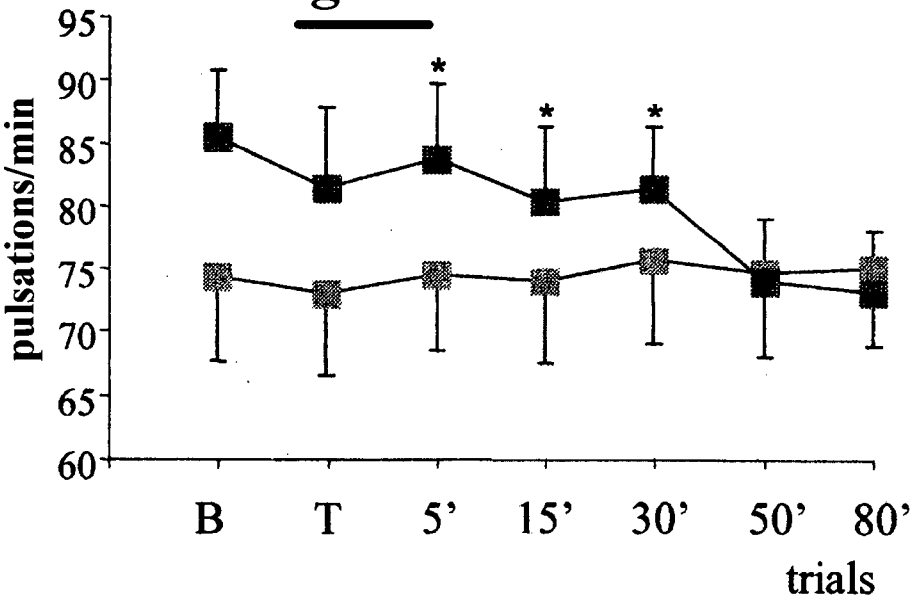


Fig. 14A

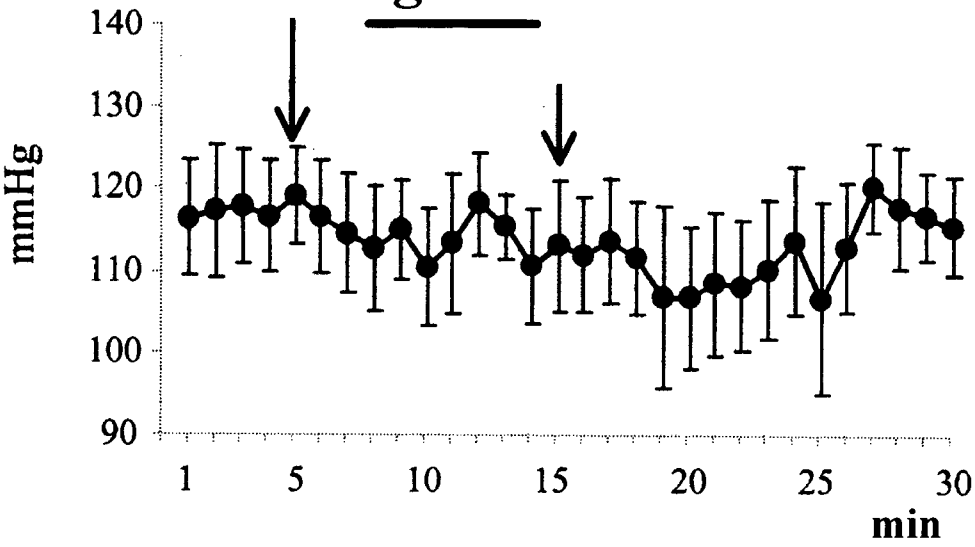


Fig. 14B

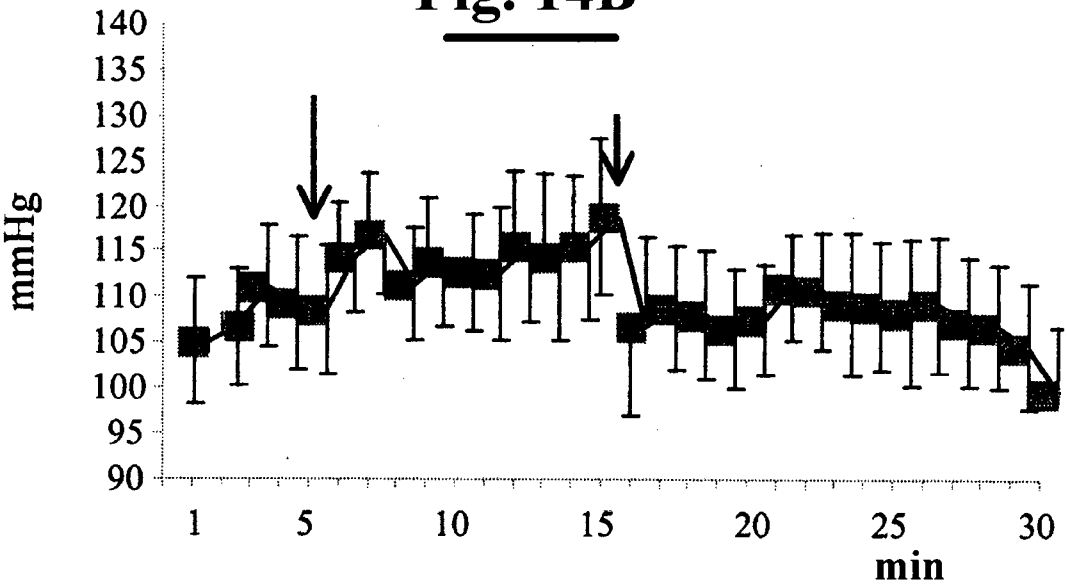


Fig. 14C

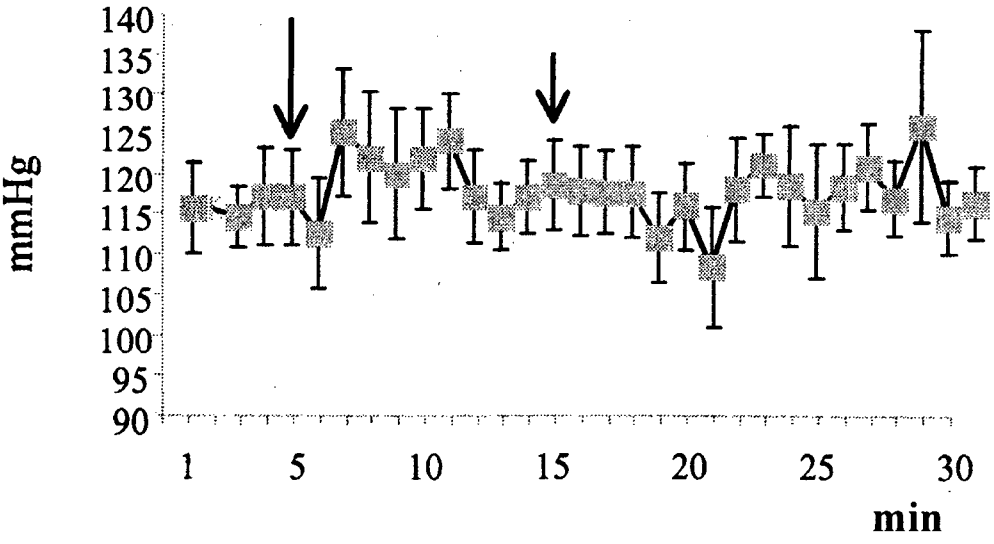


Fig. 15A

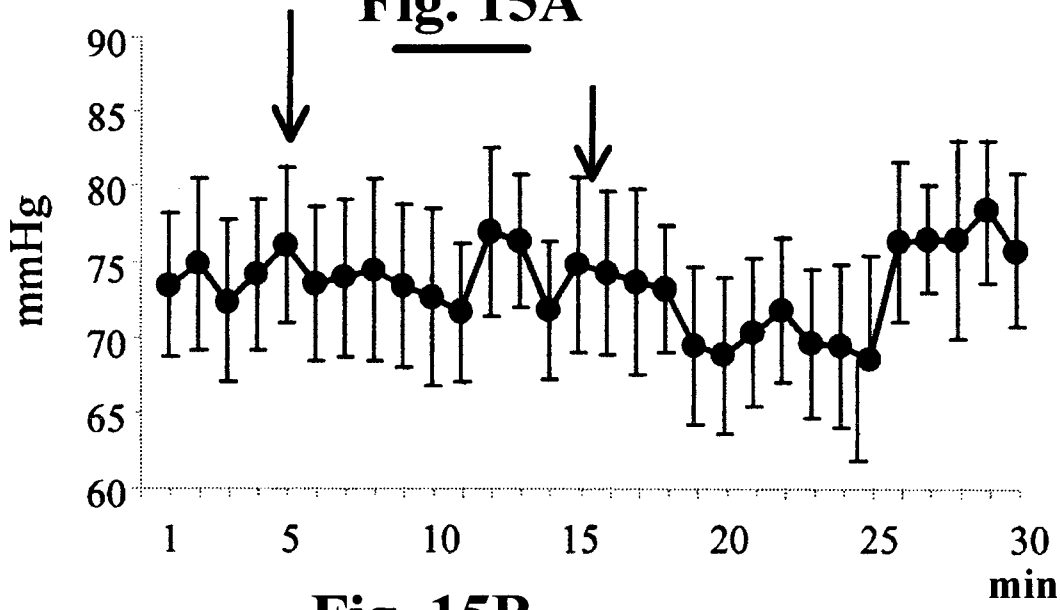


Fig. 15B

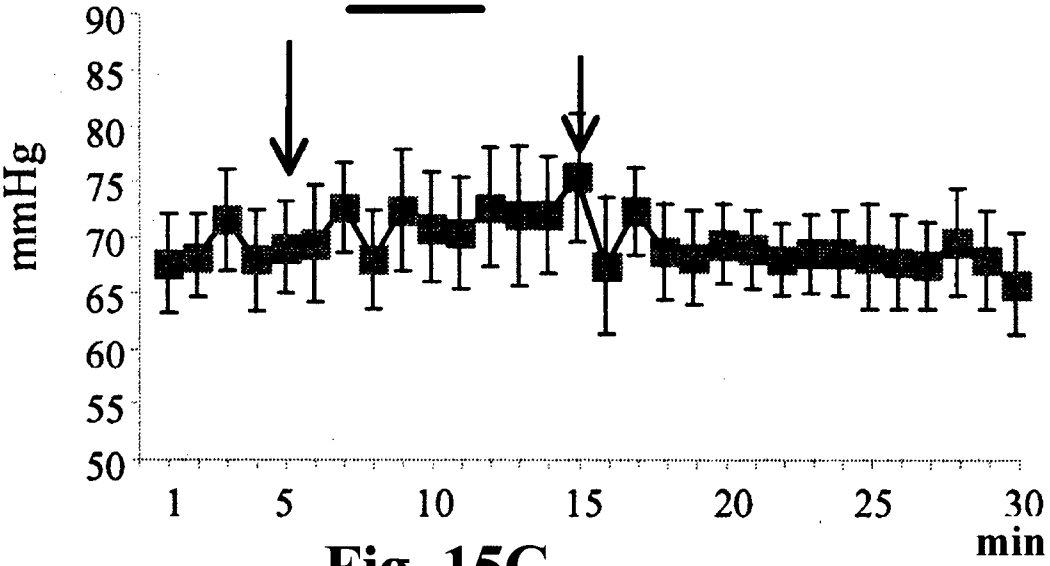


Fig. 15C

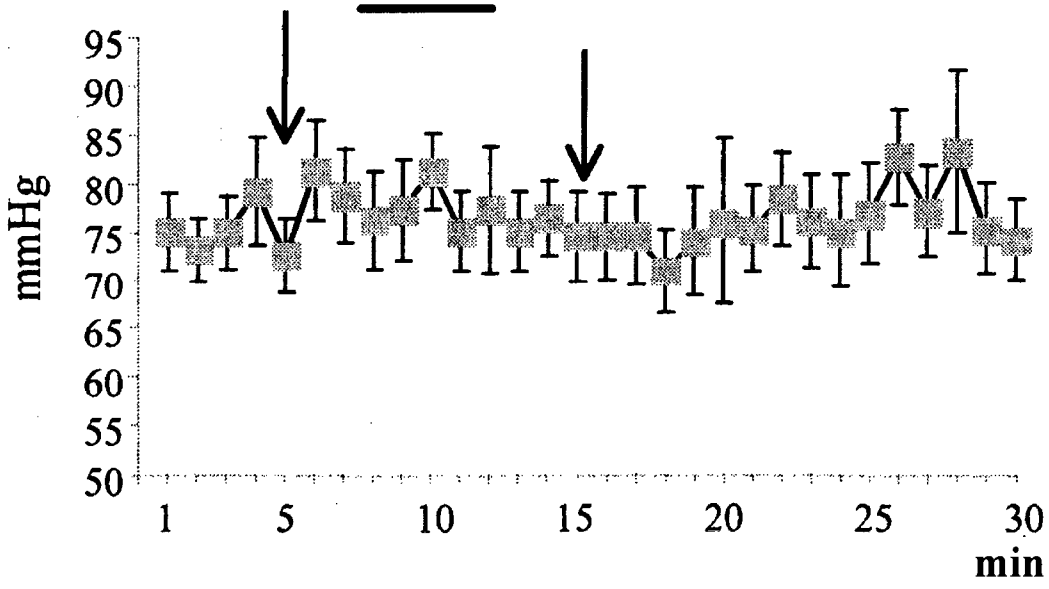
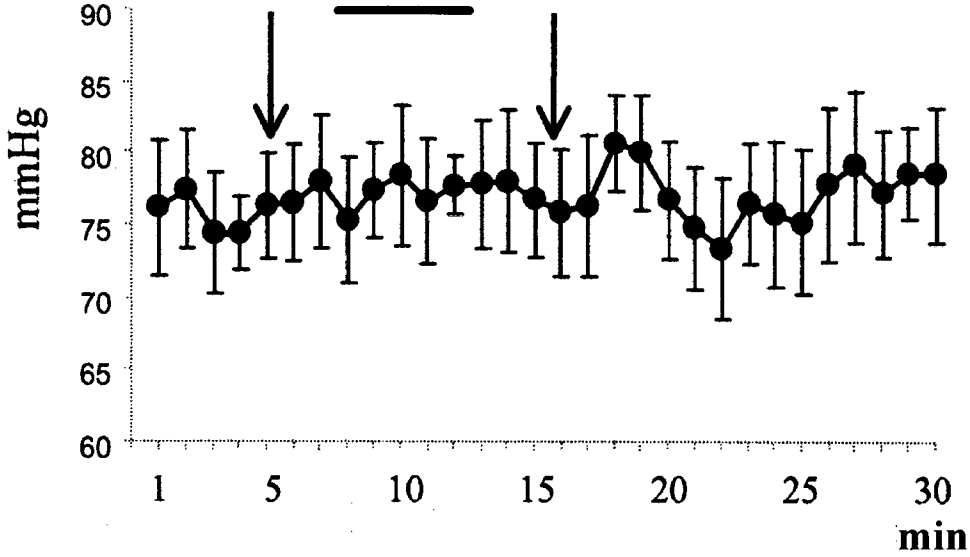


Fig. 16A



min

Fig. 16B

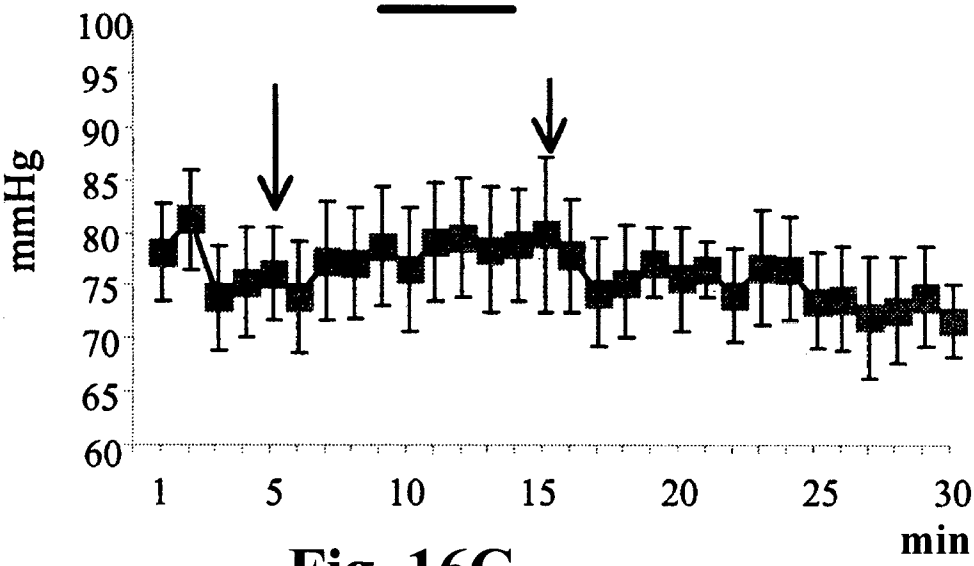


Fig. 16C

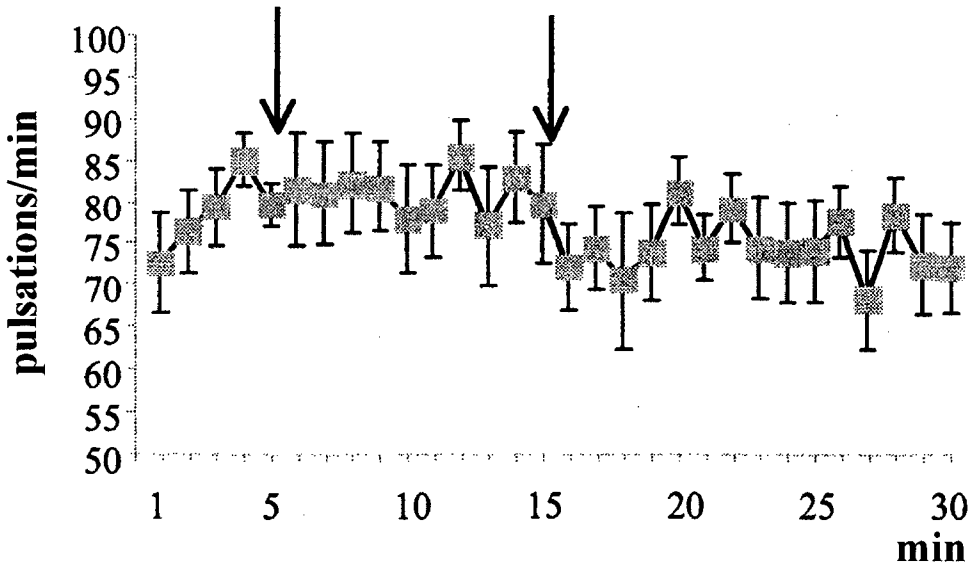


Fig. 17

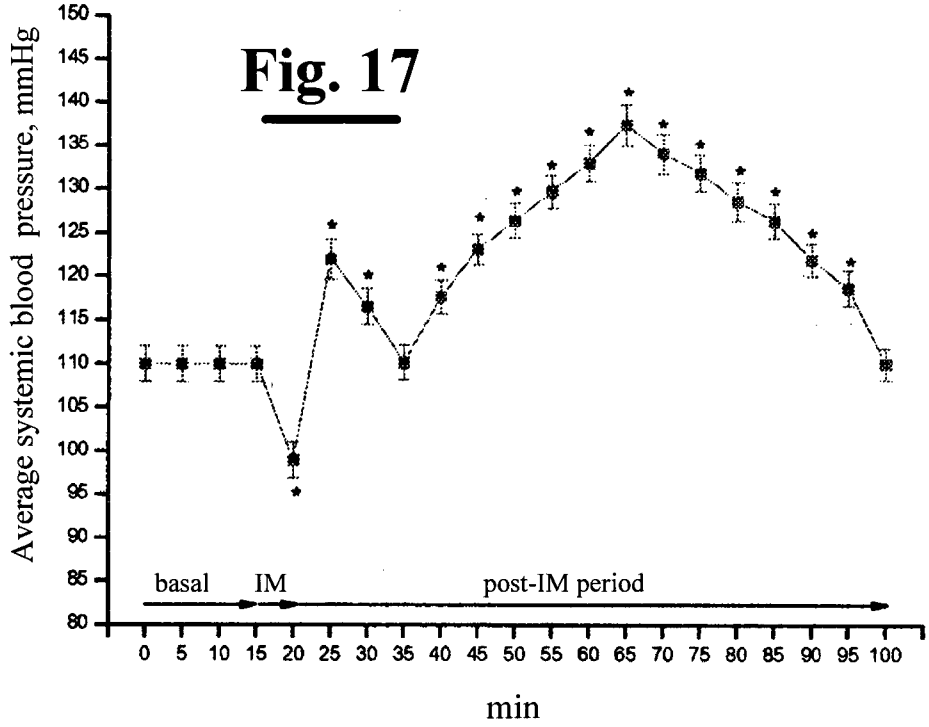


Fig. 18A

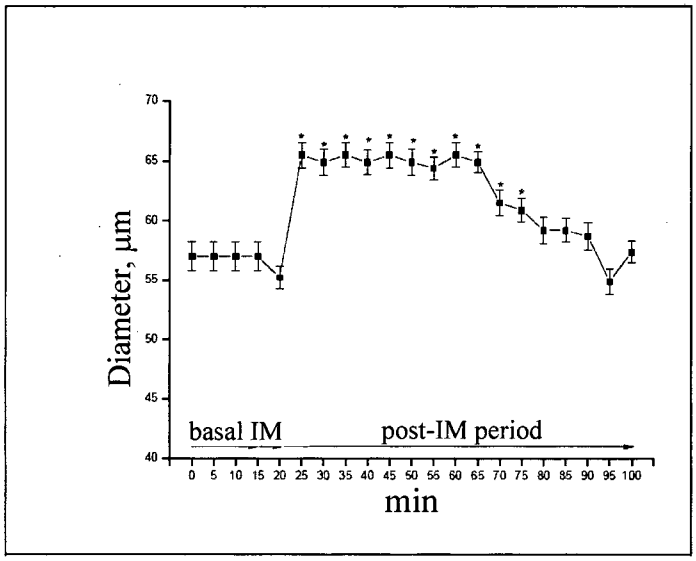


Fig. 18B

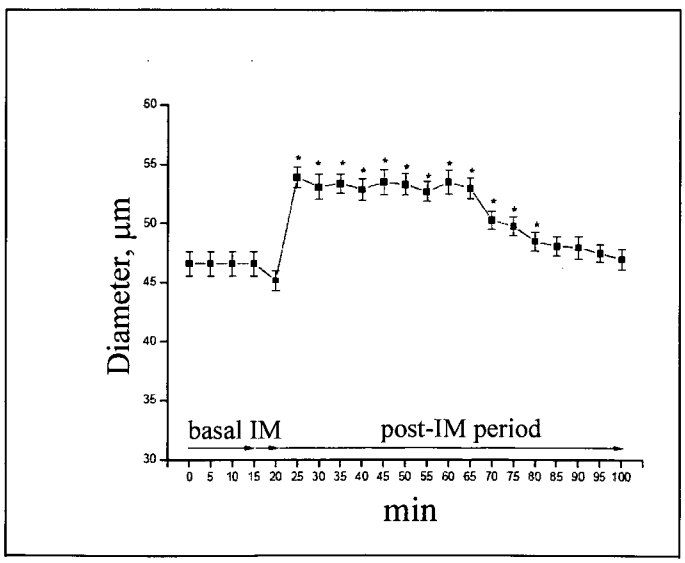


Fig. 18C

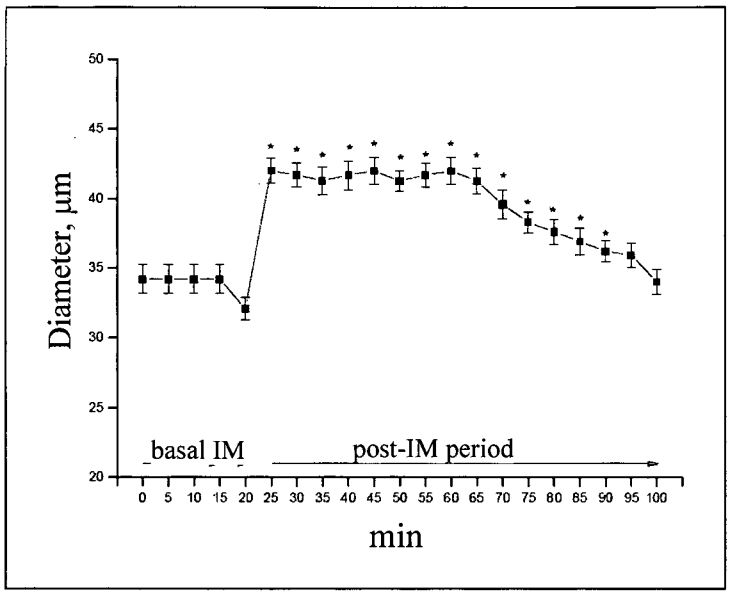


Fig. 18D

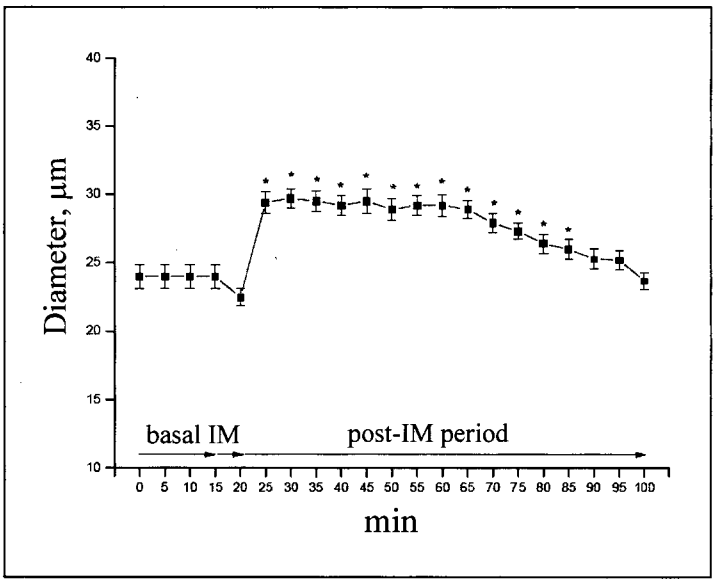


Fig. 18E

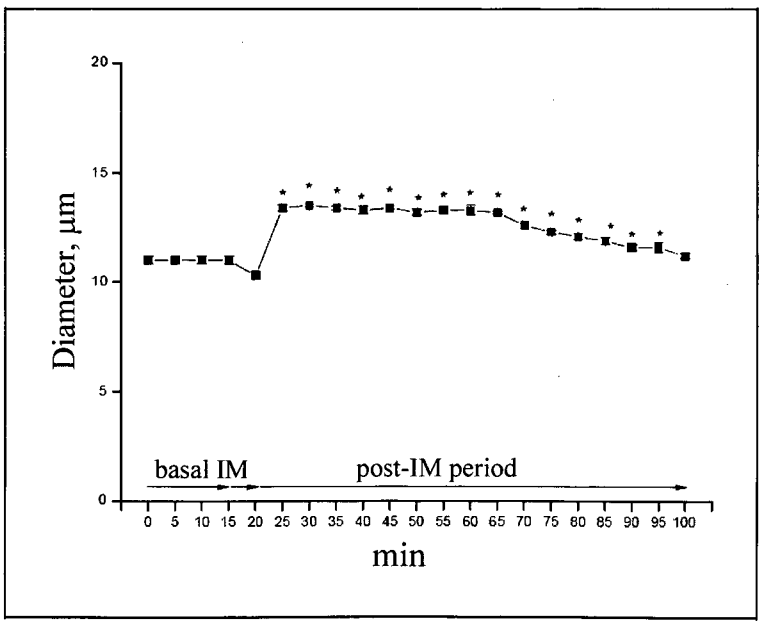


Fig. 19

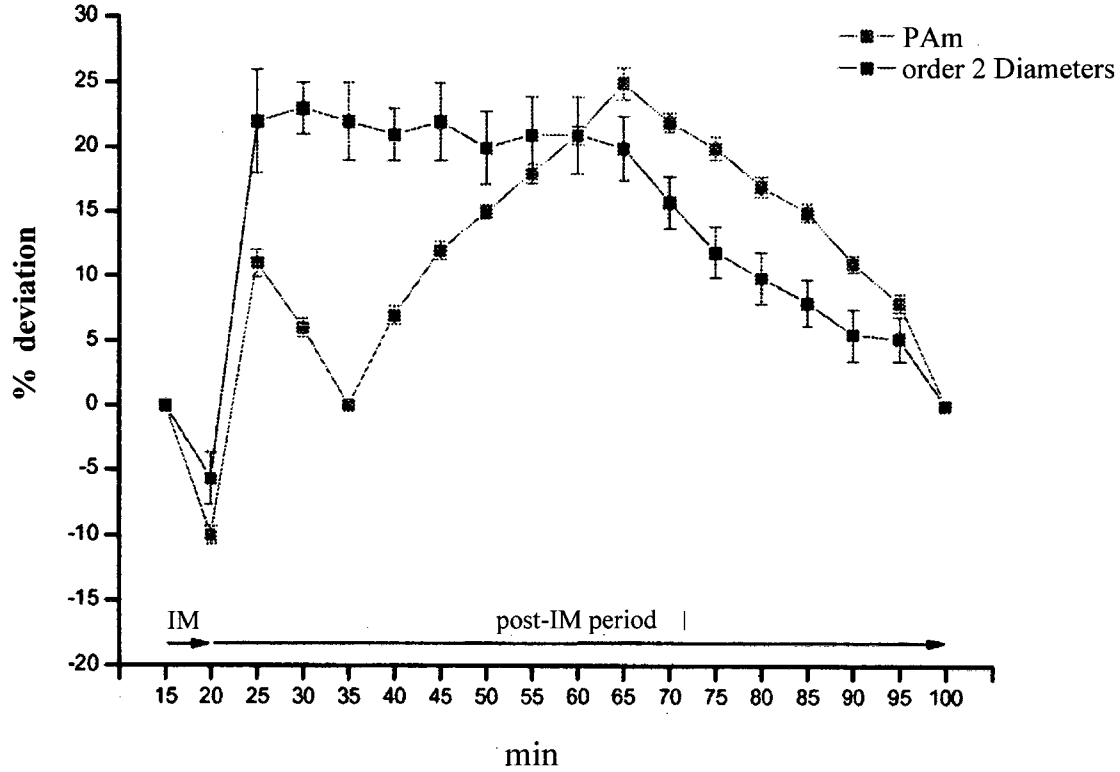

PAm
order 2 Diameters
% deviation
30
25
20
15
10
5
0
-5
-10
-15
-20
IM
post-IM period
15 20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100
min

Fig. 20

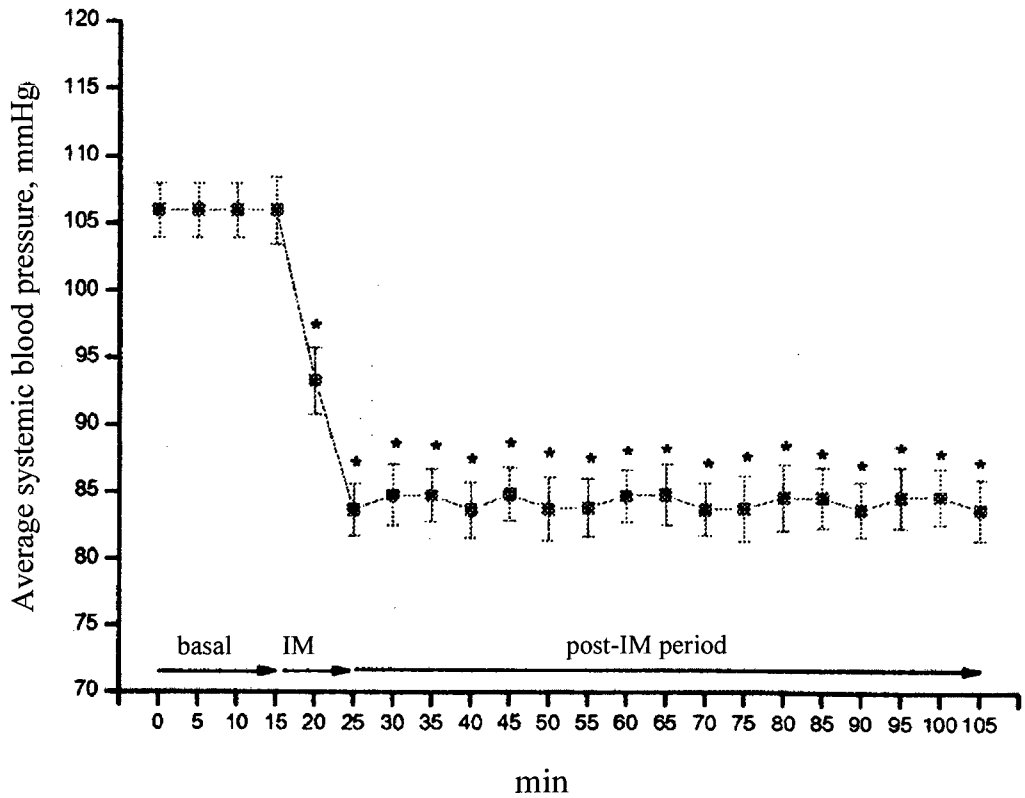

Average systemic blood pressure, mmHg
120
115
110
105
100
95
90
85
80
75
70
basal
IM
post-IM period
0 5 10 15 20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105
min

Fig. 21A

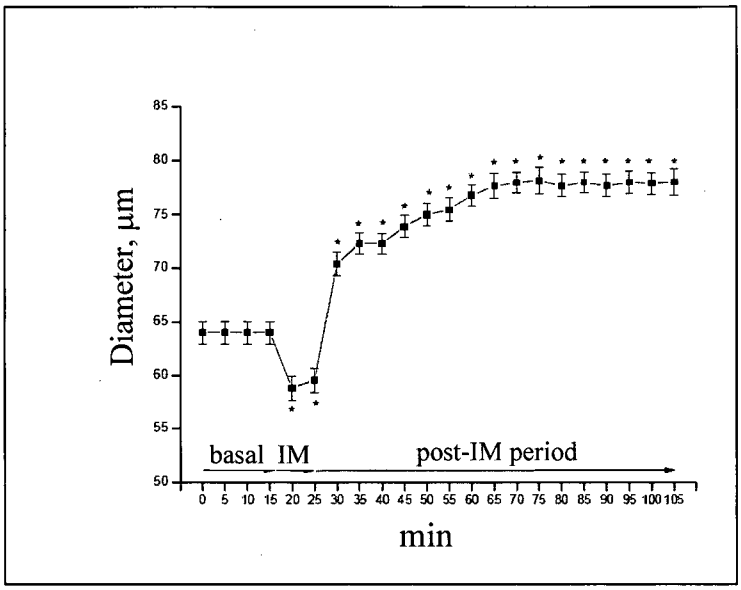


Fig. 21B

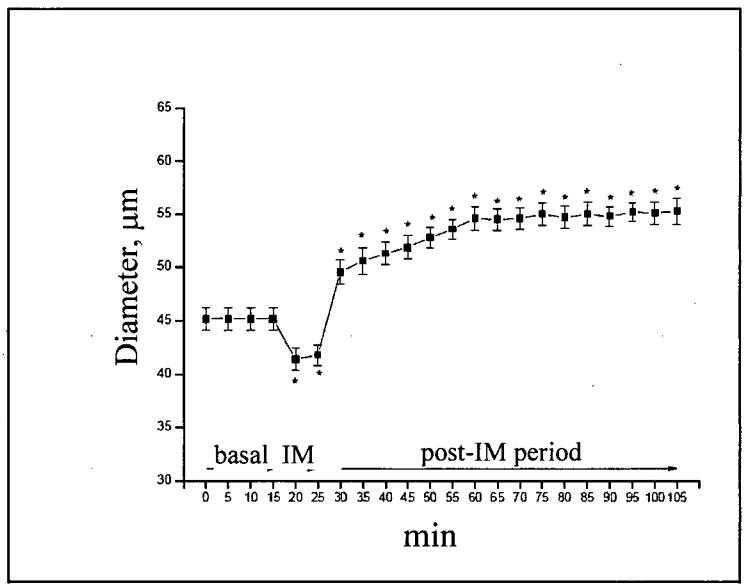


Fig. 21C

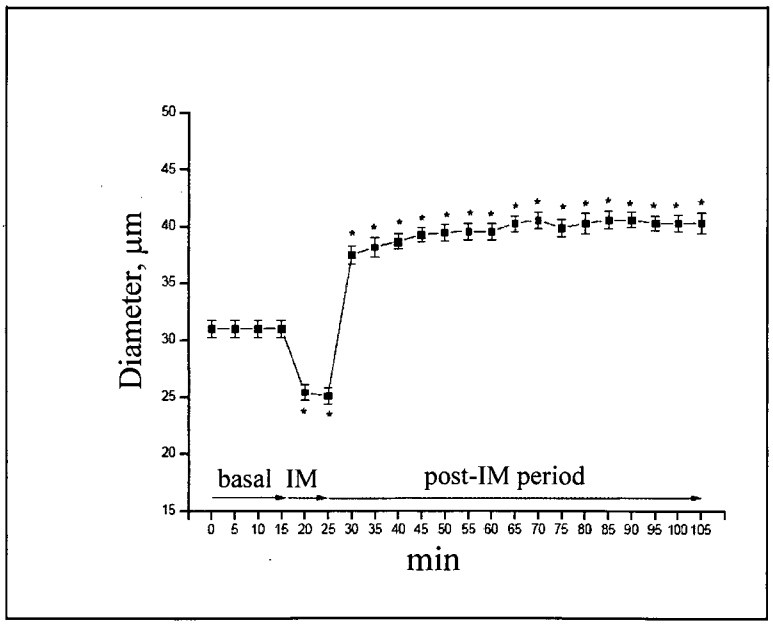


Fig. 21D

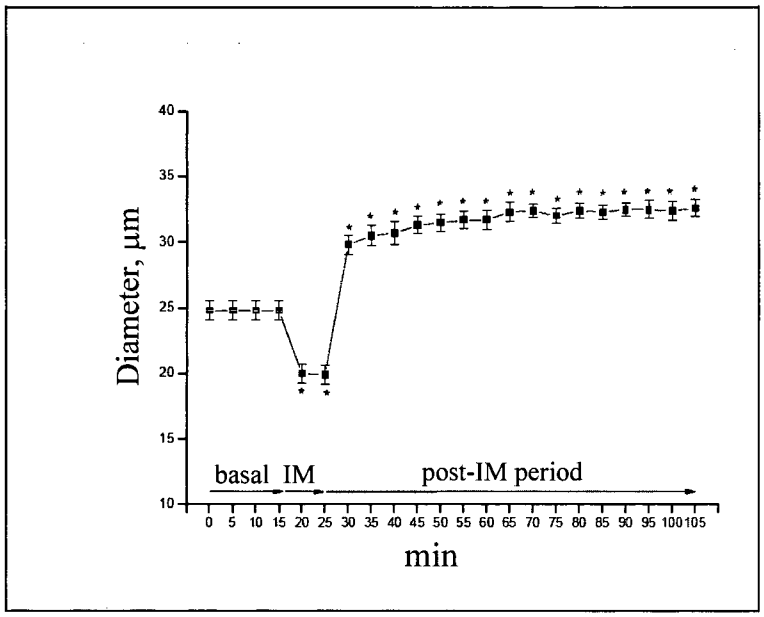


Fig. 21E

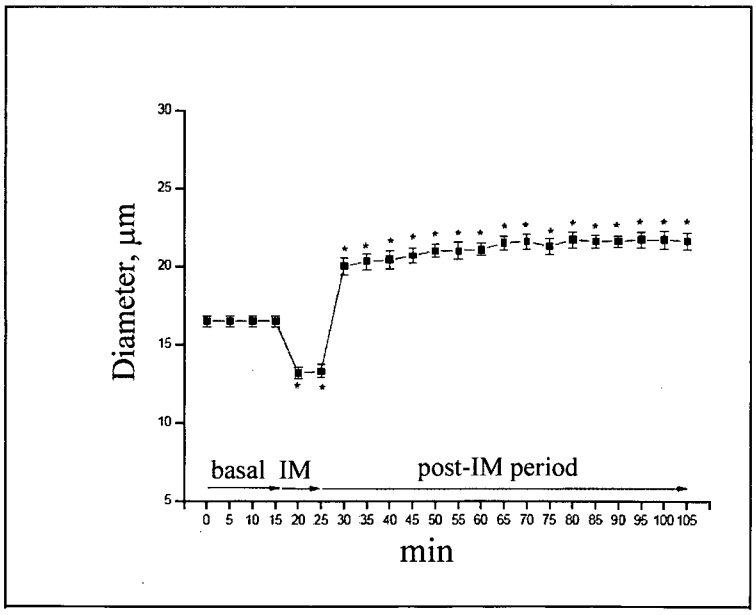


Fig. 22

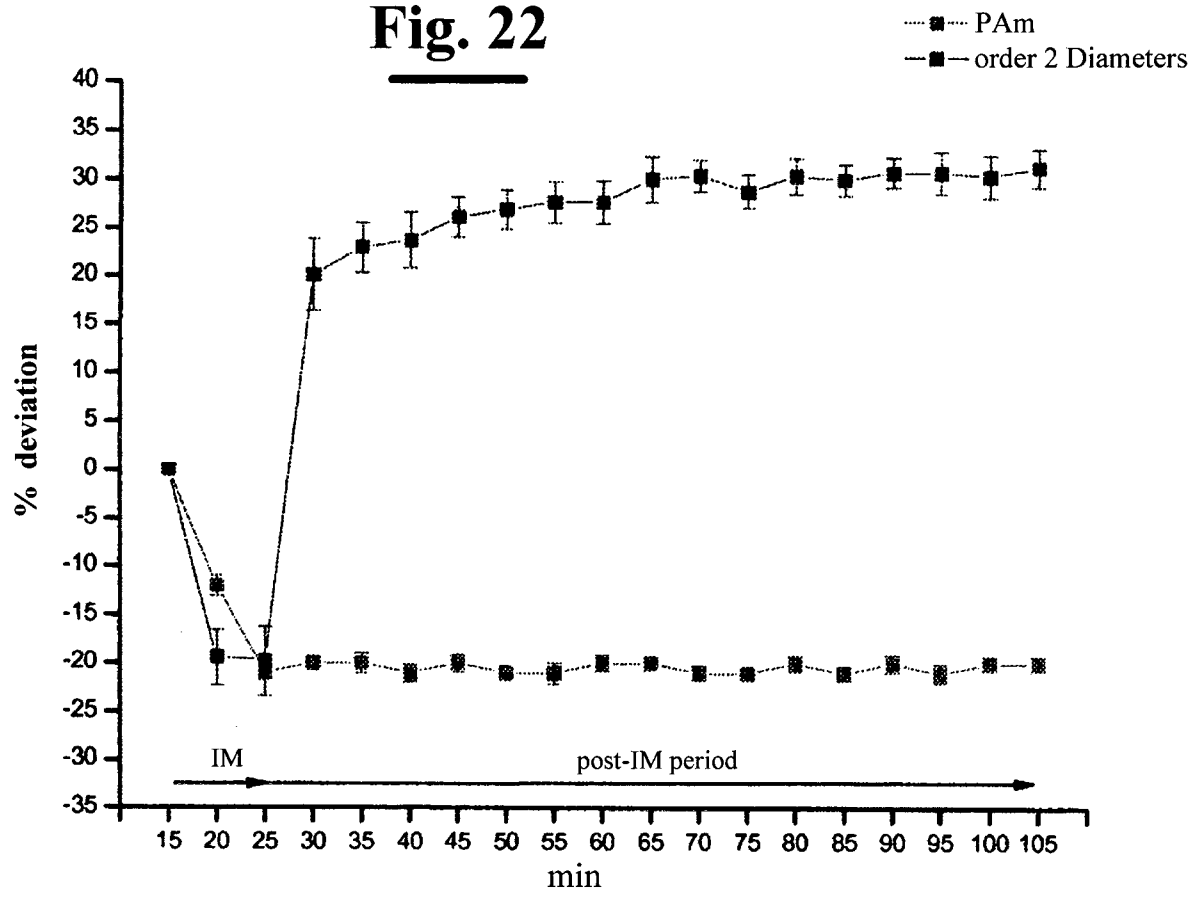

PAm
order 2 Diameters
% deviation
IM
post-IM period
min

Fig. 23

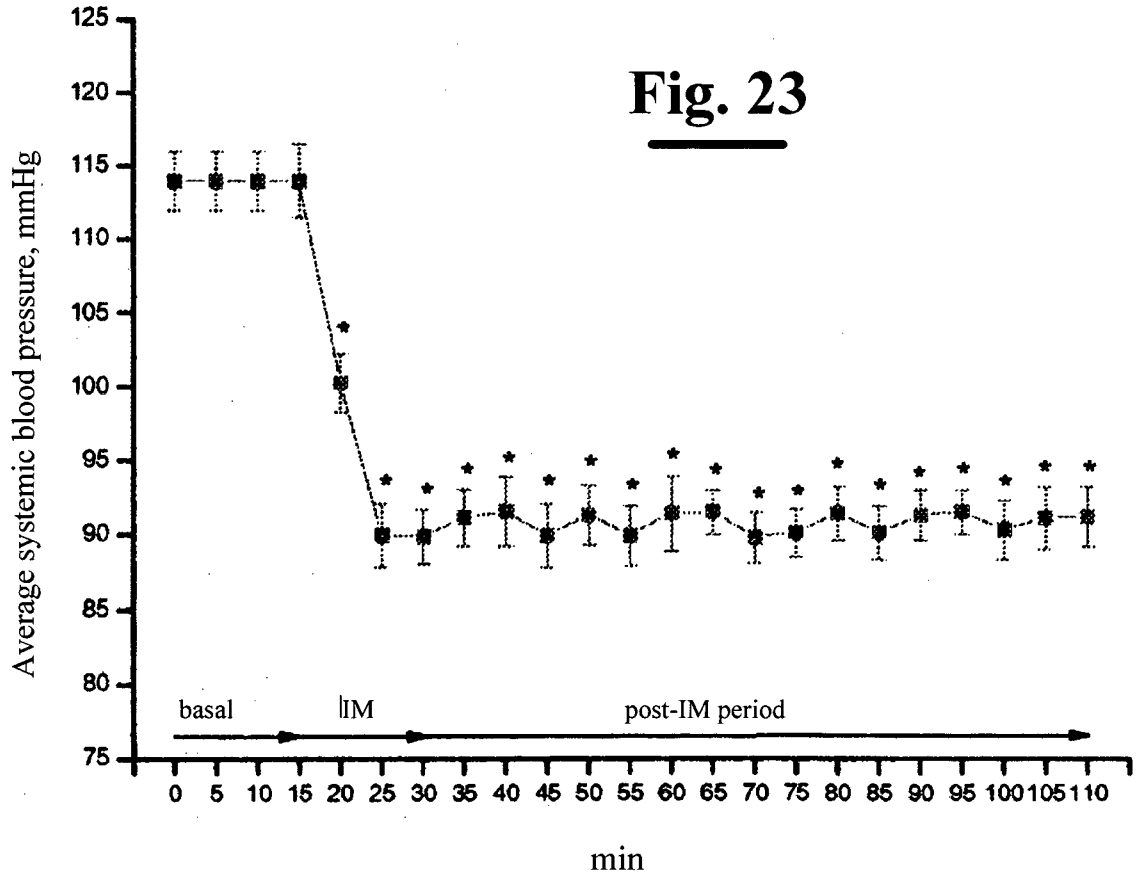

Average systemic blood pressure, mmHg
basal
IM
post-IM period
min

Fig. 24A

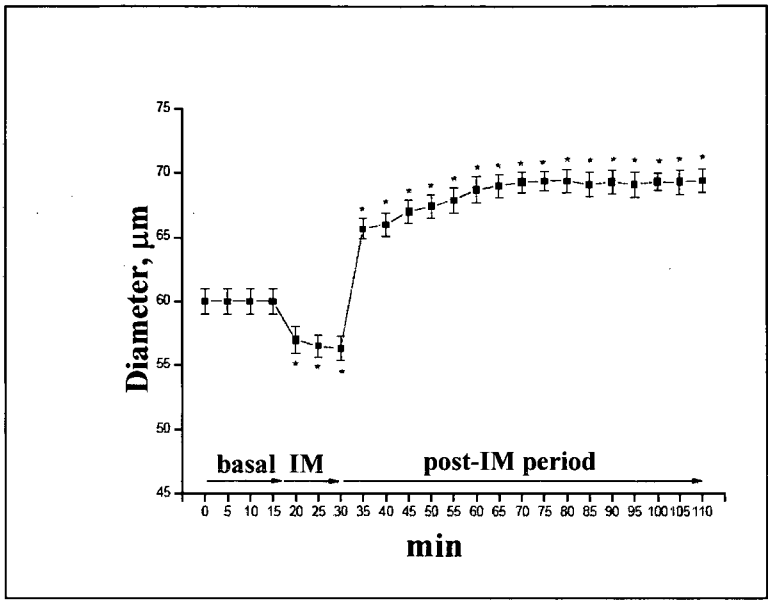


Fig. 24B

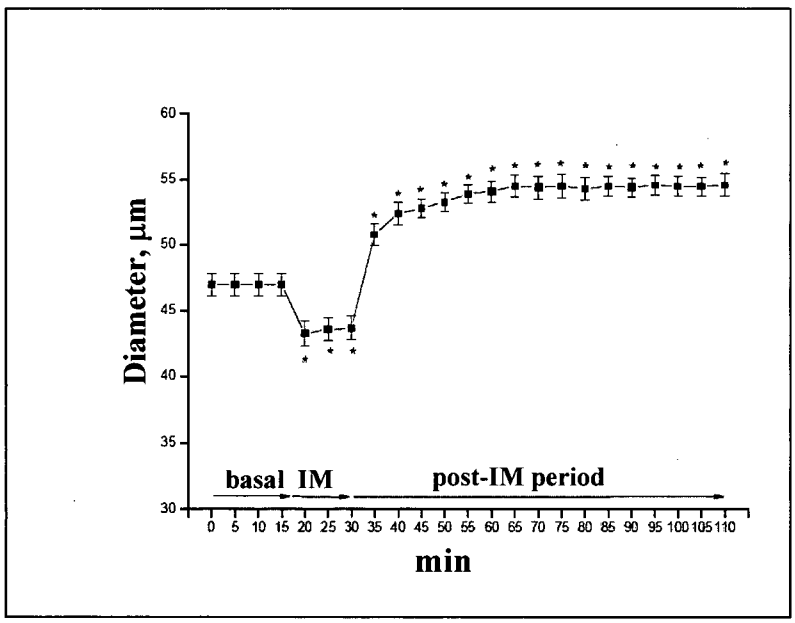


Fig. 24C

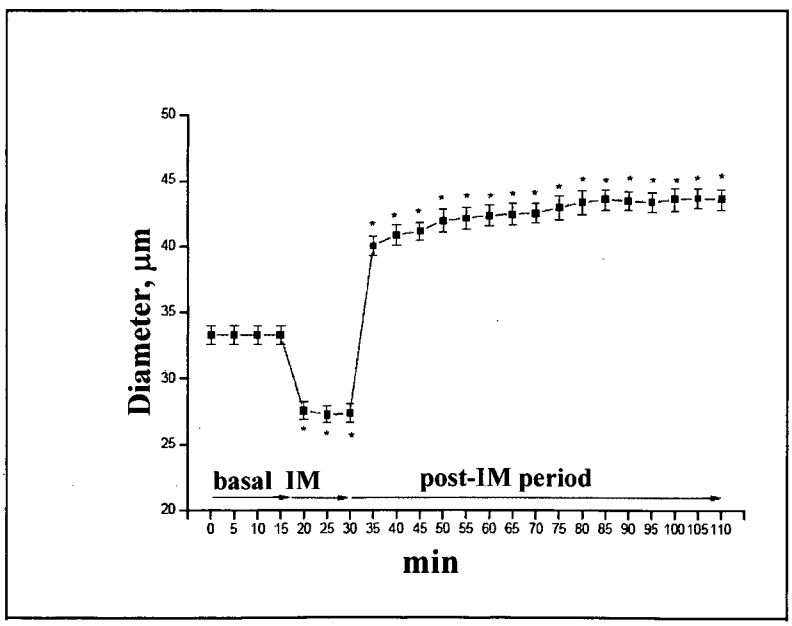


Fig. 24D

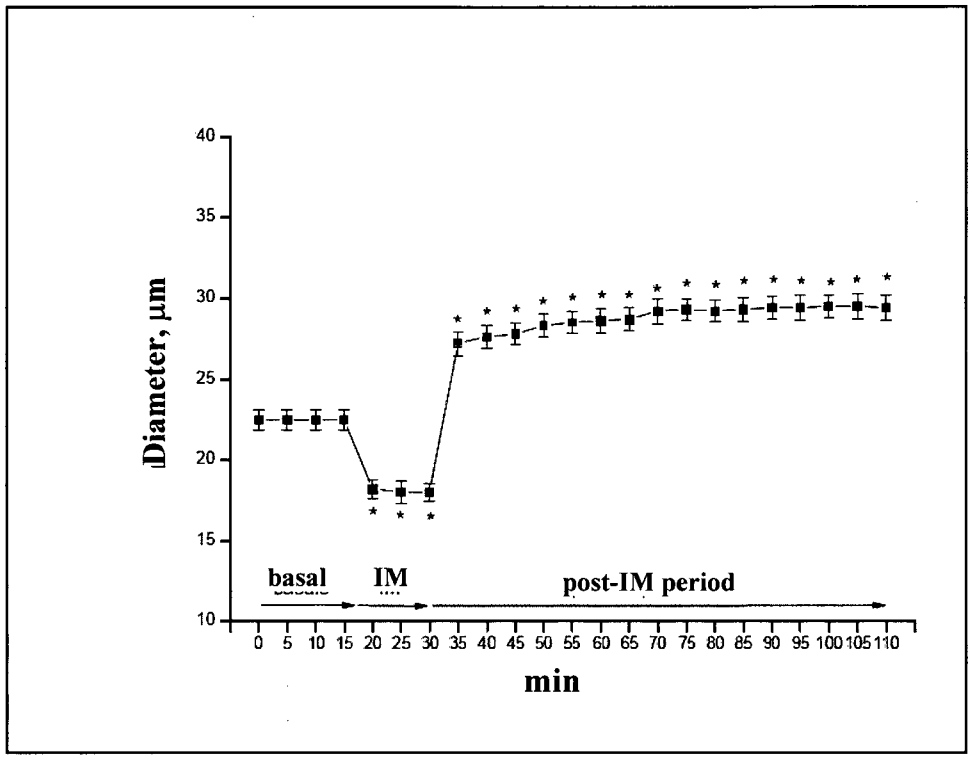


Fig. 24E

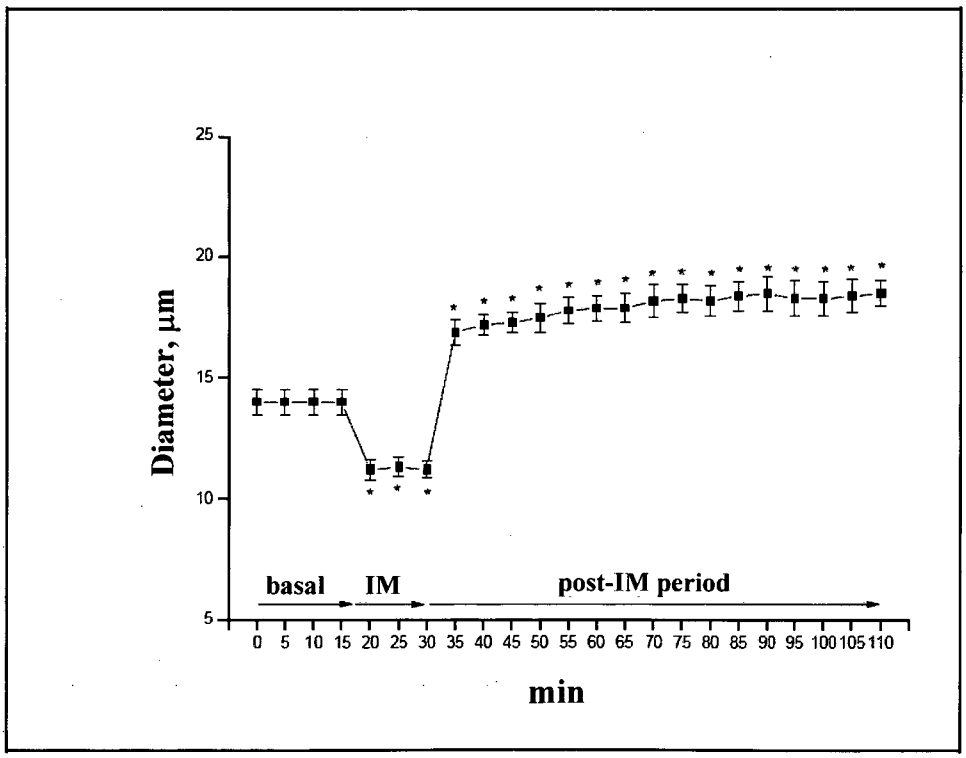


Fig. 25

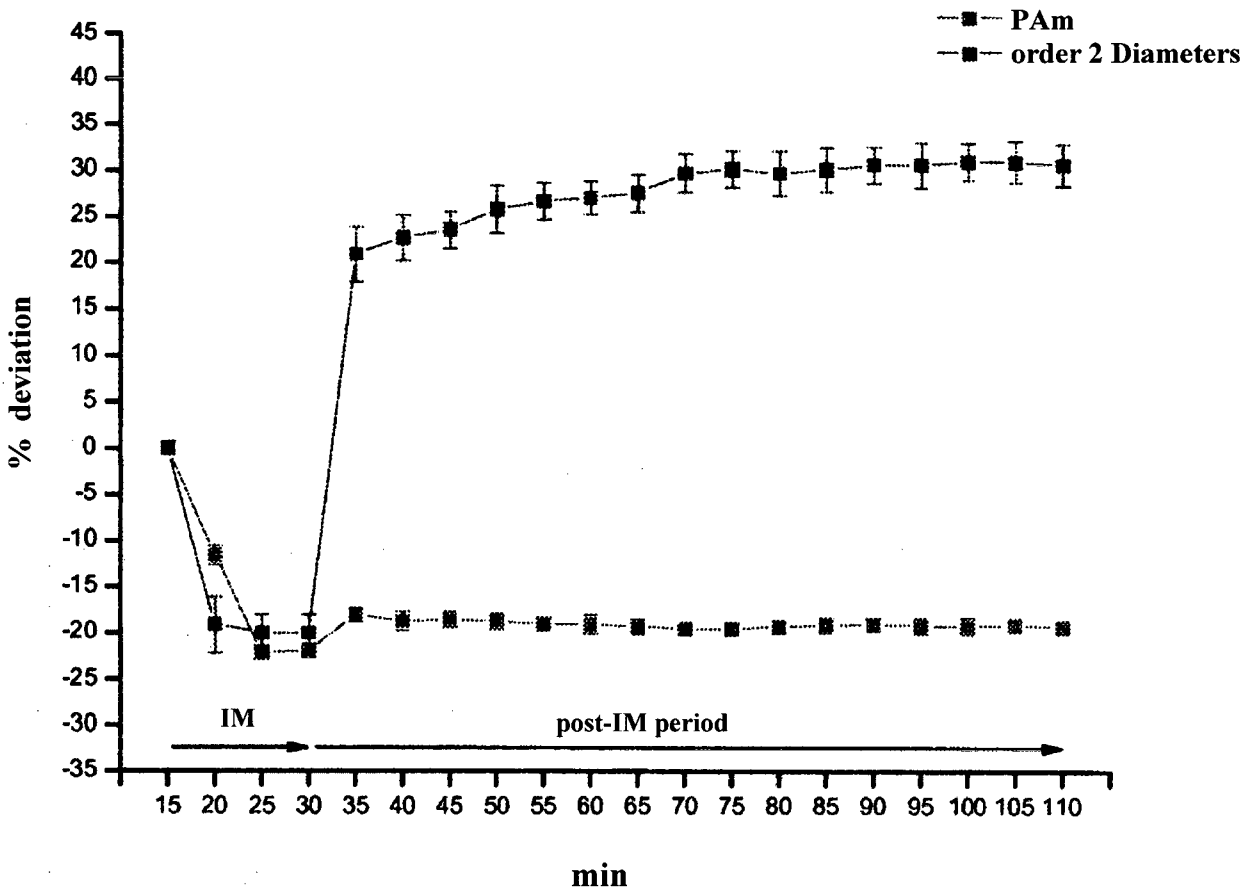

PAm
order 2 Diameters
% deviation
45
40
35
30
25
20
15
10
5
0
-5
-10
-15
-20
-25
-30
-35
IM
post-IM period
15 20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110
min

Fig. 26A

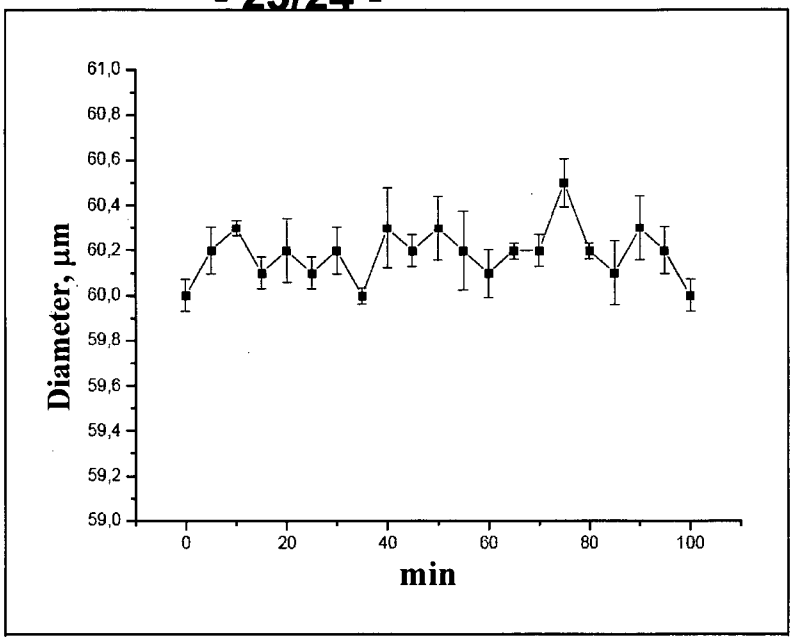


Fig. 26B

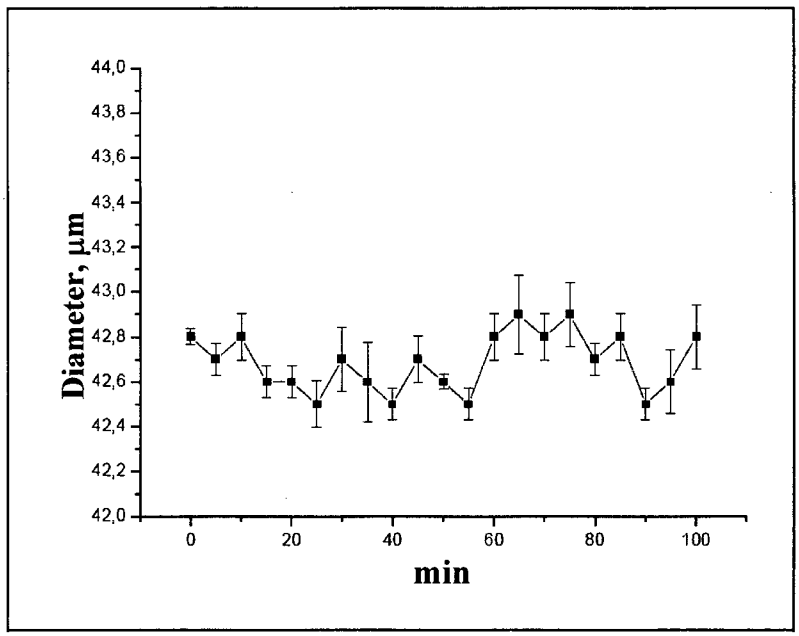


Fig. 26C

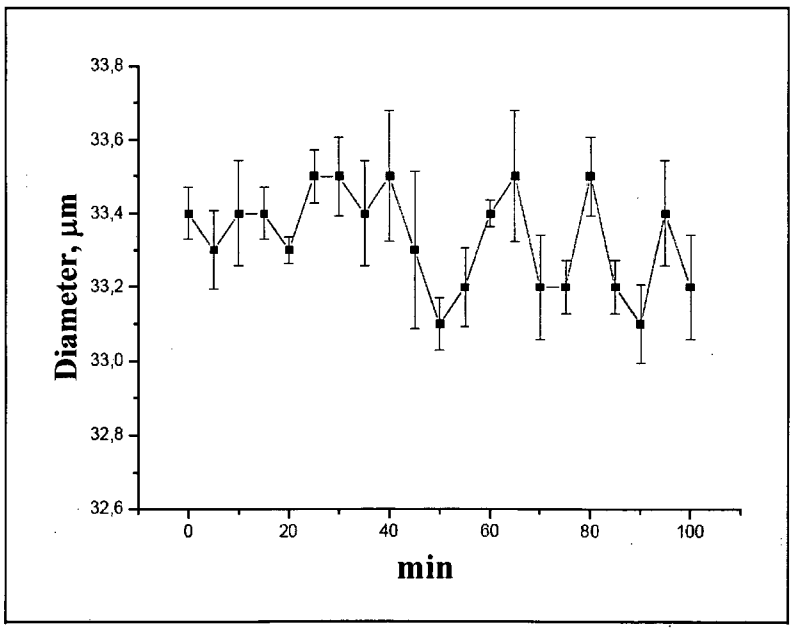


Fig. 26D

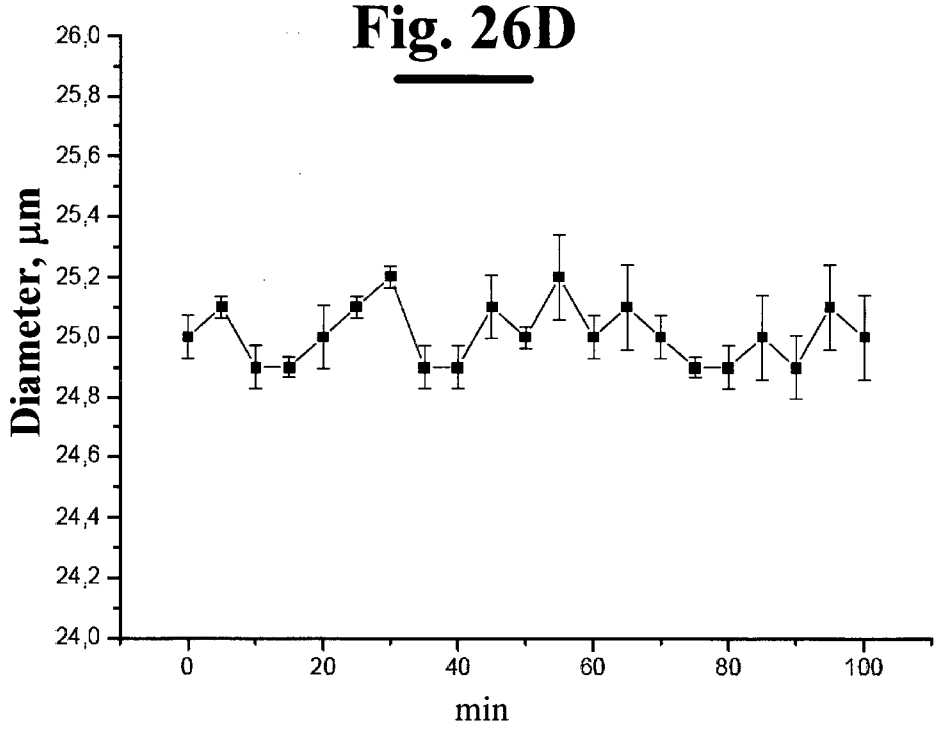


Fig. 26E

