

+7 950 990 57 09; E-mail: tamara-polivanova@yandex.ru.

Vshivkov Vitaliy Alekseevich, PhD (Medicine), Senior Scientific Worker of Clinical Division of the Pathology of

Digestion System in Adults and Children of Scientific Research Institute for Medical Problems of the North of Federal Research Centre «Krasnoyarsk Scientific Centre» of Siberian Division

of Russian Academy of Sciences; Partizana Zheleznyaka Str., 3G, Krasnoyarsk, 660022, Russian Federation; tel.: +7 923 280 06 98; E-mail: vitali1983@mail.ru.

I.V. Andreeva, A.A. Vinogradov, T.M. Zhestkova, N.V. Kalina, R.Y. Simakov, E.S. Simakova, A.S. Grigoriev, R.V. Svyativoda

A COMPARATIVE ANALYSIS OF THE EXPERIMENTAL INDICATORS OF INTRACUTANEOUS OXYGEN TENSION WITH MICROCIRCULATION PARAMETERS

DOI 10.25789/YMJ.2019.65.04

ABSTRACT

The article presents the results of the authors' research on the modern possibilities of studying a number of hemodynamic parameters – microcirculation and oxygen tension (PO₂) in the skin of the abdomen in rats.

To conduct qualitative and quantitative analysis of microcirculation in experiments on animals using laser Doppler flowmetry and transcutaneous oxymeter experiments have been performed in 25 mature male rats of Wistar line. The control group consisted of 5 rats. Animals of the experimental group were tired daily for 30 days by forced running from 0.5 to 1.0 hour at a speed of 10-15 km/h. A time of the rats' running on the treadmill depended on the intensity and productivity of their run. On the first day and in 5, 10, 20 and 30 days from the beginning of the experiment microcirculation speed was studied in the skin of the abdomen of the animals after the running load with «Transonic Systems Inc.» (Model BLF21) laser Doppler flowmeter as well as intradermal oxygen tension with Radiometer TCM-2 (Denmark) transcutaneous oxymeter. It was found that during the experiment in animals of the experimental group the level of intracutaneous oxygen tension was lower than in control animals. It was found that in systematic running load the microcirculation in the skin of the animal's abdomen accelerated up to 10 days from the beginning of the experiment. Then, by 30th day there was a slowdown in the rate of microcirculation. But in all measurements during the experiment it was faster than in animals of the control group. The inverse dependence of PO₂ from the rate of microcirculation was revealed, which was expressed by a decrease in the level of intracutaneous PO₂ in all cases of increasing the rate of intracutaneous microcirculation.

The study has showed that the study of hemodynamic parameters of experimental animals with the research methods described above is a promising direction of modern physiology. The main advantages of these research methods are noninvasiveness, which provides the possibility of repeated use in the experiment and the implementation of dynamic control over changes in the studied parameters.

Keywords: microcirculation, oxygen tension, experimental study.

Introduction. Interest in the study of hemodynamics in animal experiments involves extrapolating the results to humans. The experimenter has an extensive arsenal of devices that record various parameters of tissue and organ hemodynamics. A particular interest are the devices that make it possible to perform noninvasive methods of hemodynamic studies in organs and systems of laboratory animals in the process of experimental exposure [1, 3, 4, 7, 8]. The simplest, most accessible, and noninvasive hemodynamic studies in animals are performed using Doppler ultrasound scanning [2, 5, 6, 9-11]. In addition, various analyzing systems based on laser Doppler flowmetry are used to study blood microcirculation in laboratory animals, and transcutaneous oxymeters and polygraphs are used to determine intracutaneous oxygen tension [7, 8]. The choice of a device for the study of hemodynamics in a particular vascular pool depends on the goal of the study and the devices available to the experimentalist recording hemodynamics [5]. This raises the question of the aim of a particular device's use in a particular experimental study. In the literature, these issues are presented insufficiently, so it

involves special studies aimed at conducting a comparative analysis of the results obtained with the use of different recording systems.

The aim of the study was to carry out a comparative analysis of intracutaneous oxygen tension indicators with parameters of microcirculation in the skin of the abdomen of the experimental animals.

Materials and methods of research. The study was performed in 20 mature male Wistar rats weighing 280-300 g or more. The control group consisted of 10 rats. Animals of the experimental group were tired daily for 30 days by forced running from 0.5 to 1.0 hour at a speed of 10-15 km/h. A time of the rats' running on the treadmill depended on the intensity and productivity of their run. On the first day and in 5, 10, 20 and 30 days from the beginning of the experiment oxygen tension (PO₂) was measured in the animals' skin of the abdomen in the control and experimental groups under general anaesthesia (1% solution of thiopental sodium at the rate of 15 mg/kg of body weight intraperitoneal) in the supine position by means of transcutaneous oxymeter Radiometer TCM-2 (Denmark) [7], and using «Transonic Systems Inc.» (model BLF21) laser Doppler flowmeter (LDF)

device. Parameters of intracutaneous microcirculation were determined [8].

To determine the PO₂ wool in the rat's abdomen was shaved, the skin was treated with soap and water, dried with ether and degreased. The sensor retainer was glued to the skin the cavity of which was filled with a contact gel and sealed with a membrane. Calibration of the device sensor was performed, which was stopped after the appearance of a stable indicator on the display of the device. After the device calibration finishing, the sensor was fixed in the lock and a series of measurements were performed (Fig. 1).

When determining the parameters of intracutaneous microcirculation with the help of LDF, recording of indicators began after a 10-minute adaptation of the animal to an ambient temperature of 20°C. Indications of intracutaneous microcirculation were measured for 5 minutes in the area of the shaved part of the anterior abdominal wall (the site of determining the skin PO₂) until a stable value was achieved.

Care of animals was carried out according to the orders regulating the organization of work with use of experimental animals.

Digital data were processed by meth-

ods of variation statistics using Microsoft Excel licensed computer program.

Results and discussion. The initial level of PO₂ in the abdominal skin of the control animals ranged from 27-39 mm Hg (34.2 ± 4.76 mm Hg). On the 5th day from the beginning of the experiment, PO₂ level was 28-42 mm Hg (35.6 ± 5.08 mm Hg), in 10 days - 29-44 mm Hg (36.9 ± 6.02), in 20 days - 27-40 (35.2 ± 4.95) in 30 days - 28-41 mm Hg (35.1 ± 5.66 mm Hg). The average value of PO₂ in the abdominal skin of the control animals was 35.66 ± 1.00 mm Hg (Fig. 2).

In animals of the control group, the rate of microcirculation in the skin of the abdomen on the 1st day of the study ranged from 13 to 18 ml/100 g/min (15.4 ± 2.07 ml/100 g/min), on the 5th day of the study, the index ranged from 13 to 21 ml/100 g/min (16.3 ± 3.13 ml/100 g/min), on the 10th day - from 13 to 22 (16.8 ± 3.42), on the 20th day - from 13 to 19 (15.8 ± 2.39), on the 30th day - from 14 to 20 ml/100 g/min (16.2 ± 2.28 ml/100 g/min). The average rate of microcirculation in the skin of the abdomen was 16.02 ± 0.58 mm Hg (Fig. 3).

A comparative analysis of the results obtained in the animals of the control group revealed a direct correlation between the parameters of PO₂ and the parameters of intradermal microcirculation. The correlation coefficient and its error ($R \pm r$) indicated a direct strong and reliable relationship between the change in PO₂ and the change in intradermal microcirculation ($R \pm r = 0.926 \pm 0.071$ at $p < 0.001$).

In animals of the experimental group prior to the experiment, PO₂ level in the skin of the abdomen ranged from 27-41 mm Hg (34.2 ± 4.76 mm Hg at $p < 0.01$). After running load PO₂ was in the range of 26-38 mm Hg (33.4 ± 5.18 mm Hg at $p < 0.05$), which was 1.026 ± 0.028 times lower than the baseline ($R \pm r = 0.989 \pm 0.011$ at $p < 0.001$).

After a 5-day experiment, PO₂ level decreased by 1.072 ± 0.126 times ($R \pm r = 0.859 \pm 0.131$ at $p < 0.001$) and was 26-39 mm Hg (33.6 ± 5.18 mm Hg at $p < 0.05$). After 10 days from the beginning of the experiment, PO₂ sharply decreased to 22-34 mm Hg (27.6 ± 4.93 mm Hg at $p < 0.05$), that is 1.300 ± 0.129 times lower than the initial level ($R \pm r = 0.821 \pm 0.163$ at $p < 0.05$). On the 20th day of PO₂ rose sharply to 24 to 37 mm Hg (31.2 ± 5.63 mm Hg at $p < 0.05$), that is 1.149 ± 0.096 times lower than the initial level ($R \pm r = 0.895 \pm 0.099$ at $p < 0.01$). In 30 days from the start of the experiment, the PO₂ was in the range of 26 to 37 mm Hg (33.2 ± 4.71 mm Hg at $p < 0.01$), that is 1.072 ± 0.036 times lower

than the initial level ($R \pm r = 0.976 \pm 0.024$ at $p < 0.001$).

The level of PO₂ in the abdomen's skin of animals of the experimental group varied during the experiment from 22 to 39 mm Hg (31.8 ± 5.40 mm Hg at $p < 0.05$). On the 10th day it reduced to 27.6 ± 4.93 mm Hg, and by the 30th day it increased up to 33.2 ± 4.71 mm Hg, but in all cases it was below the initial level (Fig. 3).

In animals of the experimental group on the first day of the experiment after a running load, the microcirculation index in the skin of the abdomen ranged from 11 to 23 ml / 100 g/min (18.8 ± 4.76 ml/100 g / min at $p < 0.05$), which was 1.207 ± 0.203 times higher than the control value ($R \pm r = 0.896 \pm 0.099$ at $p < 0.01$). On the 5th day of the experiment, the rate of microcirculation in the skin of the abdomen ranged from 14 to 23 ml / 100 g/min (18.2 ± 4.55 ml/100 g / min at $p < 0.05$), that was 1.103 ± 0.123 times higher than the control value ($R \pm r = 0.906 \pm 0.091$ at $p < 0.01$). On the 10th day of the experiment, the rate of microcirculation in the skin of the abdomen ranged from 14 to 22 ml / 100 g/min (19.2 ± 3.35 ml/100 g / min at $p < 0.01$), which was 1.151 ± 0.130 times more than the control ($R \pm r = 0.812 \pm 0.170$ at $p < 0.01$). On the 20th day of the experiment, the rate of microcirculation in the skin of the abdomen ranged from 13 to 21 ml/100 g/min (18.2 ± 3.35 ml/100 g/min at $p < 0.05$), which was 1.148 ± 0.097 times higher than the control value ($R \pm r = 0.914 \pm 0.083$ at $p < 0.001$). On the 30th day of the experiment, the rate of microcirculation in the skin of the abdomen ranged from 12 to 21 ml / 100 g/min (17.2 ± 3.42 ml/100 g / min at $p < 0.05$), that was 1.057 ± 0.124 times higher than the control value ($R \pm r = 0.859 \pm 0.131$ at $p < 0.05$).

Microcirculation in the skin of the abdomen of animals of the experimental group changed during the experiment from 11 to 23 ml / 100 g / min (18.3 ± 3.89 ml / 100 g/min) on the 10th day it increased to 19.2 ± 3.35 ml/100 g/min, and by 30 days it decreased to 17.2 ± 3.42 ml/100 g / min, but in all cases it was faster than the initial level (Fig. 3).

The comparative analysis of the results obtained in animals of the experimental group revealed the inverse correlation of PO₂ parameters with the parameters of intradermal microcirculation. The correlation coefficient and its error indicated at the inverse strong and reliable relationship between the change in PO₂ and the change in intradermal microcirculation ($R \pm r = -0.904 \pm 0.177$ at $p < 0.05$).

Conclusion. The study showed that the microcirculation and oxygen tension (PO₂) study in the skin of experimental animals presupposes widespread use of

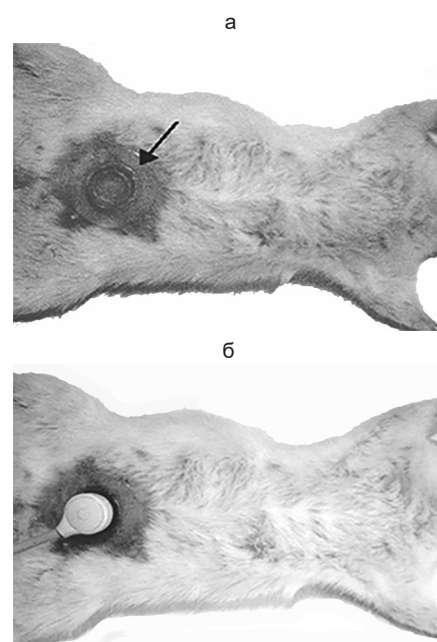


Fig. 1. Measurement of PO₂ by transcutaneous method: a – preparation of animals for the measurement of PO₂, sensor lock is shown by arrow; b – measurement of PO₂ (the sensor is fastened in the locking mechanism).

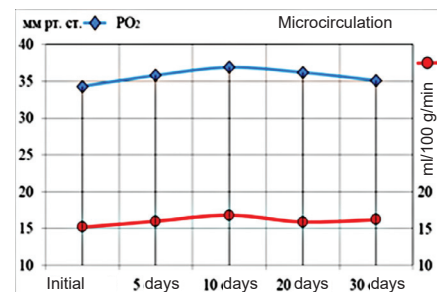


Fig. 2. Dynamics of oxygen tension (PO₂) and microcirculation in the skin of the abdomen of the animals of the control group

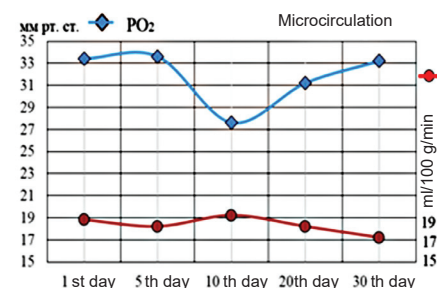


Fig. 3. Dynamics of oxygen tension (PO₂) and microcirculation in the skin of the abdomen of the animals of the experimental group.

LDF («Transonic Systems Inc.» device, model BLF21) and TCM-2 Radiometer transcutaneous oxymeter (Denmark). The analysis of the dynamics of microcirculation parameters in the skin of the abdomen of the control animals revealed a direct dependence of the level of PO₂ on the parameters of microcirculation (with an increase in microcircula-

tion – intradermal PO₂ increased, too). In the systematic running load the indicator of the microcirculation in the skin of the abdomen of the animal increased up to 10 days. Then, by the 30th day there was a decrease in the index of microcirculation. But in all cases it was higher than in animals of the control group. The level of intradermal PO₂ was inversely related to the parameters of microcirculation determined with LDF. In animals of the experimental group there was a decrease in the level of PO₂ in all cases compared to the control group.

It should be noted that the study of hemodynamic parameters of experimental animals by the methods described above is a promising area of modern physiology. The main advantages of using these non-invasive methods of research are: the possibility of repeated use in the experiment; the implementation of dynamic control over changes in the studied parameters.

References

1. Abrosimova T.N., Andreeva I.V., Vinogradov A.A. Kachestvennye pokazateli portal'noj gemodinamiki v ehksperimente [Qualitative indicators of portal hemodynamics in the experiment] *Ukrains'kij medychnyj al'manah* [Ukrainian medical almanac]. Kiev, 2008, № 6 (11), p. 7-9.
2. Andreeva I.V., Vinogradov A.A. Atlas normal'noj i ul'trazvukovoj anatomii zhivota krysy (elektronnyj uchebnik) [Atlas of normal and ultrasound anatomy of the rat's abdomen (e-textbook)]. Moscow: Geotar-Media, 2014, 170 p. [www.studmedlib.ru/book/08-COS-2400.html](http://studmedlib.ru/book/08-COS-2400.html)
3. Andreeva I.V., Vinogradov A.A. Vozmozhnosti izucheniya ul'trazvukovoj anatomii zheludka i kishechnika u krysa [The possibilities of studying the ultrasound anatomy of the stomach and intestine in rats] *Visnyk Lugansk'skogo natsional'nogo universitetu imeni Tarasa Shevchenko. Medychni nauky* [Vestnik of the Lugansk Taras Shevchenko National University. Medical science]. Lugansk, 2014, № 8 (291), Part II, p. 5-13. <http://www.2227-2844-2014-291-8-5-13>
4. Andreeva I.V., Vinogradov A.A. Perspektivy ispol'zovaniya sovremennykh metodov vizualizatsii v morfologicheskikh i eksperimental'nykh issledovaniyakh [The prospects of use of modern imaging techniques in morphologic and experimental studies] *«Nauka molodykh»* (Eruditio Juvenium) [«Youth Science» (Eruditio Juvenium)]. Ryazan', 2015, № 4, p. 56-69. http://www.nauka_molodykh-2015-4
5. Andreeva I.V., Vinogradov A.A. Vozmozhnosti sovremennykh metodov vizualizatsii v morfologicheskikh issledovaniyakh [Possibilities of modern methods of visualization in the field of morphological studies] *Materialy nauchno-prakticheskoy konferentsii «Uchitelya i ucheniki: preemstvennost' pokolenij», posvyashchennoj 250-letiyu so dnya rozhdeniya professora E.O. Muhina (24 noyabrya 2016 g.)*. [Materials of scientific-practical conference «Teachers and students: the continuity of generations», dedicated to the 250th anniversary of Professor E. O. Mukhin's birth (November 24, 2016)]. Moscow: Izd-vo Pervogo MGIMU im. I.M. Sechenova [Publishing house of the First Moscow State Medical University], 2016, p. 23-24.
6. Andreeva I.V., Vinogradov A.A., Kalina N.V. Ul'trazvukovye tehnologii v issledovanii pozvonochnykh ven [Ultrasound technology in the study of vertebral veins] *Naukovi pratsi XIII Mizhregional'noi naukovoï konferentsii «Aktual'ni pytannya biologii ta medytsyny»* [Scientific works of XIII Interregional scientific conference «Actual questions of biology and medicine»] *Vid-vo DZ «LNU imeni Tarasa Shevchenka»* [Edition of Lugansk National Taras Shevchenko University]. Starobilsk, 2016, p. 9-12.
7. Vinogradov A.A. [et al.] Vnutrikozhnoe napryazhenie kisloroda pri adaptatsii k begovoy nagruzke do i posle gipoksicheskoy trenirovki [Intracutaneous oxygen tension in adaptation to the running workload before and after hypoxic training] *Olimpijskij sport, fizicheskaya kul'tura, zdorov'e natsii v sovremennykh usloviyakh (mezhdunarodnaya nauchno-prakticheskaya konferentsiya)* [Materials of international scientific-practical conference «Olympic sports, physical culture, health of the nation in modern conditions»]. Lugansk, 2005, p. 191-193.
8. Kovaleva I.S., Konovalova O.V., Demchenko S.S. Lazernaya dopplerovskaya floumetriya v otsenke mikrotsirkulyatsii u krysa [Laser Doppler flowmetry in the assessment of microcirculation in rats] *Zbirnyk naukovykh prats' za materialamy naukovo-praktichnoi konferentsii «Dosyagnennya ta perspektivy suchasnykh medyko-biologichnykh napryamkiv»* [Collected papers of materials of scientific-practical conference «Achievements and perspectives of the modern medical and biological directions»] *Vid-vo DZ «LNU imeni Tarasa Shevchenka»* [Edition of Lugansk National Taras Shevchenko University], Lugansk, 2014, p. 14-16.
9. Lelyuk V.G., Lelyuk S.E. Ul'trazvukovaya angiologiya [Ultrasound angiology] 2-e izd., pererab. i dop. [2nd ed. Real time]. Moscow, 2003, 336 p.
10. Comparison of portal venous flow in cirrhotic patients with and without paraumbilical vein patency using duplex sonography / M. Domland [et al.] // *Ultrascail Med.* – 2000. – Vol. 21 (4). – P. 9–165. PMID:11008315
11. Hemodynamics in the microvasculature of thioacetamide-induced cirrhotic rat livers / M. Nakata [et al.] // *Hepatogastroenterology.* – 2002. – Vol. 49 (45). – P. 652-656. PMID:12063962

The authors:

Andreeva Irina Vladimirovna – Doctor of Medical Science, Professor of Dept. of Surgery, Obstetrics and Gynecology of Postgraduate Education Faculty, Ryazan State Medical University, Ryazan, Russia, prof.andreeva.irina.2012@yandex.ru.

Vinogradov Alexander Anatolyevich – Doctor of Medical Sciences, Professor of Dept. of Cardiovascular, Endovascular, Operative Surgery and Topographic Anatomy, Ryazan State Medical University, Ryazan, Russia, alexanvin@yandex.ru.

Zhestkova Tatiana Mikhailovna – physician at Medical center «MC-MED» (Saint-Petersburg), aspirant of Dept. of Surgery, Obstetrics and Gynecology of Postgraduate Education Faculty, Ryazan State Medical University, Ryazan, Russia, atjana_zhestkova@mail.ru.

Kalina Natalia Vladimirovna – Candidate of Medical Sciences (PhD), neurologist, the Deputy of Head Doctor at «Lugansk state hospital No. 3» (Lugansk), doctorant of Dept. of Surgery, Obstetrics and Gynecology of Postgraduate Education Faculty, Ryazan State Medical University, Ryazan, Russia, dockalina@mail.ru.

Simakov Roman Yur'evich – doctor-surgeon, doctor of ultrasound at «Klepikovskiy district hospital» of Ryazan, aspirant of Dept. of Surgery, Obstetrics and Gynecology of Postgraduate Education Faculty, Ryazan State Medical University, Ryazan, Russia, simakovryazan@gmail.com.

Simakova Evgeniya Sergeevna – doctor-obstetrician-gynaecologist, doctor of ultrasound at «Ryazan Clinical hospital №10», aspirant of Dept. of Surgery, Obstetrics and Gynecology of Postgraduate Education Faculty, Ryazan State Medical University, Ryazan, Russia, evsimakova@yandex.ru.

Grigoriev Alexey Sergeevich – urologist at «Kolomna central regional hospital» of Moscow region, aspirant of Dept. of Surgery, Obstetrics and Gynecology of Postgraduate Education Faculty, Ryazan State Medical University, Ryazan, Russia, Aleksey130379@yandex.ru.

Svyatovoda Roman Vladimirovich – senior ordinator of Urological Department at «Head N. N. Burdenko Military Clinical Hospital», aspirant of Dept. of Surgery, Obstetrics and Gynecology of Postgraduate Education Faculty, Ryazan State Medical University, Ryazan, Russia, drsvyatovoda@gmail.com.