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INSTITUTE OF THEORETICAL AND EXPERIMENTAL BIOPHYSICS
PERFTORAN PHARMACEUTICAL COMPANY (RUSSIA)

PREPRINT

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# PERFLUOROCARBON EMULSION, STABILIZED BY PROXANOL

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This edition represents the material on perfluorocarbon emulsions being produced in the Institute of theoretical and experimental biophysics Russian Academy of Sciences and Perftoran Pharmaceutical Company (Russia) for medicobiological application.



There are two alternative directions in the production of artificial gas transfer blood substitutes. The first one means the use of perfluorocarbon emulsions as a gas-carrying substitute and the second is aimed at the utilization of modified hemoglobin as a gas carrier.

These two directions have much in common in the ideas but essentially differ in practice. The use of modified hemoglobin excreted from human erythrocytes turned to be more effective owing to their gas transfer properties but at the same time since these preparations are toxic, their application is still problematic. Specialists have not yet get the necessary control over the responses that modify complex and fragile hemoglobin molecule. Moreover, a technology of blood treatment, its purification, hemoglobin excretion and appropriate hemoglobin modification are problematic.

Until quite recently there is a preconceived opinion as to the utilization of perfluorocarbon emulsions in medicine. A sceptical attitude to this unusual class of blood substitutes has been formed by sluggishness of thinking and conservatism of medical men. The Institute of theoretical and experimental biophysics RAS (ITEB RAS) is among the first in Russia who deals with this problem.

ITEB RAS and Perftoran Pharmaceutical Company conduct their research in the following directions:

I. investigation of the cardioplegic composition to perform the operation on the open heart on the basis of the Ftorem emulsion (perfluorodecalin/perfluorotributilamine, PFD/PFTBA in 7:3 proportion).

II. study of the perfusion composition in order to preserve excreted organs on the basis of the Perfusol emulsion (perfluoromethylcyclohexylpiperidine, PFMCP).

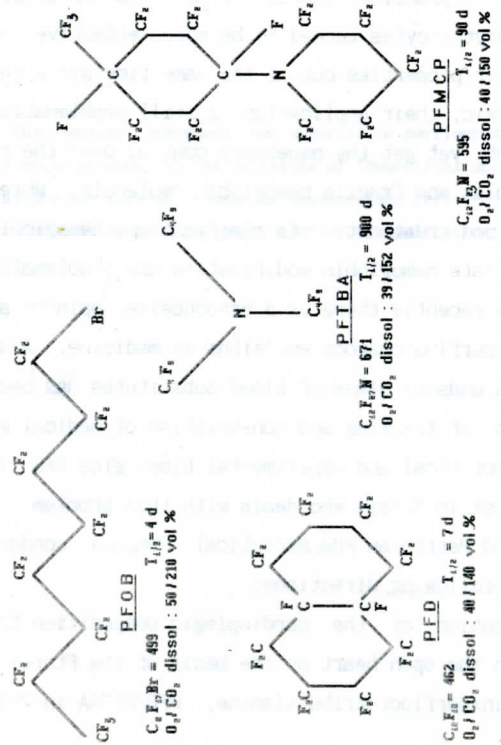


**Fig. 1. Chemical structure of some Perfluorocarbons**



III. examination of plasma substitute with gas transfer ability on the basis of the Perftoran emulsion (perfluorodecalin/perfluoromethylcyclohexylpiperidine, PFD/PFMCP).

IV. scrutiny of the fluorocontrast agents for diagnosis on the basis of the VIMM-RC emulsion (perfluorooctylbromide/perfluoromethylcyclohexylpiperidine, PFOB/ PFMCP).

Figure 1 depicts a chemical structure of the perfluoroorganic compounds being used.

I. The first direction - the elaboration of the cardioplegic composition to perform the operation on the open heart - has been crowned with establishment of the Ftorem emulsion in 1981 (Table 1). This emulsion has the raised stability.

Table 1

Cardioplegic perfluorocarbon emulsion - FTOREM

| COMPONENTS : | | |
|---|---|---|
| PFTBA | - | 6.0 g |
| PFD | - | 14.0 g |
| Proxanol | - | 3.0 g |
| NaCl * | - | 0.8 g |
| KCl | - | 0.11 g |
| $MgCl_2$ | - | 0.012 g |
| $CaCl_2$ | - | 0.004 g |
| $NaH_2PO_4$ | - | 0.02 g |
| $NaHCO_3$ | - | 0.13 g |
| Glucose | - | 0.2 g |
| $H_2O$ | - | 100 ml |

PROPERTY :

| | | |
|---|---|---|
| $F^-$ | - | $< 10^{-5}$M |
| Av. diameter | - | 0.07-0.011 μm |
| Osmolarite | - | 340-360 mOsm |
| Viscosity | - | 2.5 sP |
| pH | - | 7.4-7.8 |
| Vol. $\%O_2$ | - | 7.0 vol% |
| Vol. $\%CO_2$ | - | 60 vol% |

* component of salt composition can be varied and complemented

II. The second one - the development of the perfusion composition to preserve the excreted organs - has completed by the production of the Perfusol emulsion in 1982 (Table 2). This second generation perfluorocarbon has a cyclic structure and nitrogen heteroatom. Its emulsions have a sufficient stability and been cleared from the body (Table 7). Using this preparation (diluted two times) a 24 h conservation, at + 4°C, of the donor heart has been successfully carried out in the experiments with its subsequent heterotopic transplantation to recipient and assessment of functional activity for 6 hours.

III. In 1984 Pharmaceutical Committee of the Russian Ministry of Health (analogous to FDA, USA) has approved the clinical test of the plasma substitute with Perftoran gas transfer ability produced in 1982 under the supervision of Prof. Beloyartzev (Table 3). By a number of reasons the test of preparation has been suspended and then resumed again in 1992. Indications to study Perftoran as a plasma substitute with $O_2$ and $CO_2$-carrying ability which is anti-



Table 2

Perfusion perfluorocarbon emulsions - PERFUSOL

COMPONENTS :

| | | |
|---|---|---|
| PFMCP | - | 10.0 g |
| Proxanol | - | 3.0 g |
| HES | - | 3.0 g |
| NaCl * | - | 0.8 g |
| KCl | - | 0.039 g |
| $MgCl_2$ | - | 0.019 g |
| $CaCl_2$ | - | 0.028 g |
| $NaH_2PO_4$ | - | 0.02 g |
| $NaHCl_3$ | - | 0.13 g |
| Glucose | - | 0.2 g |
| $H_2O$ | - | 100 ml |

PROPERTY :

| | | |
|---|---|---|
| $F^-$ | - | $< 10^{-5}$M |
| Av. diameter | - | 0.07-0.11 μm |
| Onc. pressure | - | 360-380 mm $H_2O$ |
| Osmolarite | - | 340-360 mOsm |
| Viscosity | - | 2.5 sP |
| pH | - | 7.4-7.6 |
| Vol. $\%O_2$ | - | 7.0 vol% |
| Vol. $\%CO_2$ | - | 60 vol% |

* component of salt composition can be varied and complemented

Table 3

"ARTIFICIAL BLOOD" - PERFTORAN

COMPONENTS :

| | | |
|---|---|---|
| PFMCP | - | 6.5 g |
| PFD | - | 13.0 g |
| Proxanol | - | 4.0 g |
| NaCl | - | 0.6 g |
| KCl | - | 0.039 g |
| $MgCl_2$ | - | 0.019 g |
| $NaH_2PO_4$ | - | 0.02 g |
| $NaHCO_3$ | - | 0.13 g |
| Glucose | - | 0.2 g |
| $H_2O$ | - | 100 ml |

PROPERTY :

| | | |
|---|---|---|
| $F^-$ | < | $10^{-5}$M |
| Av. diameter | - | 0.07-0.15 μm |
| Osmolarity | - | 280-340 mOsm |
| Viscosity | - | 2.5 sP |
| pH | - | 7.4-7.6 |
| Vol. % $O_2$ | - | 7.0 vol% |
| Vol. % $CO_2$ | - | 60 vol% |

shock, antiishemia and cardioprotective remedy at:

- acute and chronic hypovolemia (wound, hemorrhagic, burn and infectious toxic shock, brain injury, operative and postoperative olygohemia);



- microcirculatory and peripheral circulatory disturbance; change in tissue and respiratory metabolism (pyo-septic state, infections, cerebral circulatory disturbance, fat embolism);
- antiischemia protection of donor organs (preliminary preparation of a donor and a recipient);
- cardioplegia (the use of artificial circulation in the preparation);
- regional and local applications (regional perfusion, washing of suppurative wounds, peritoneal and other cavities).

Cancerogenicity, mutanogenicity, immunomodulation and embryotoxicity do not feature the Perftoran emulsion.

Storage conditions are of great significance to the perfluorocarbon emulsions stabilized by proxanol. Prolonged preservation and change therewith of the average particle size determine the further application of the emulsion in medicobiological practice. The change of the emulsion diameter depends upon many factors including surfactants, the method of formulation and the degree of emulsion freezing. Data analysis shows the Perftoran-type emulsion being stored without freezing for nearly one month has a tendency to enlarge the average particle size (Fig. 2). Here, the most acceptable storage conditions are at + 4°C. Keeping these conditions the emulsion enlarges significantly slower and its size does not exceed the limits installed. When a storage time of the emulsion is prolonged up to two years at - 18°C, there are no sharp changes in the average particle size of the emulsion. And intermediate values of the average size fits in the range of the initial and terminal value of the mean diameter (Fig. 3).

Besides multiple freezing and thawing do not lead to the essential changes in the average particle size (Table 4).

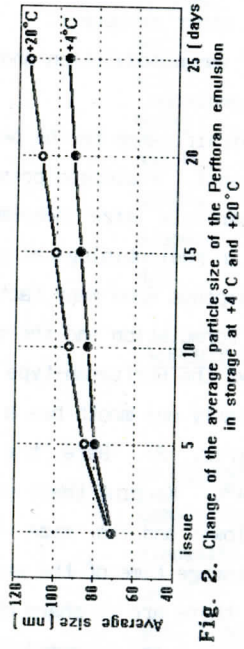


Fig. 2. Change of the average particle size of the Perftoran emulsion in storage at +4°C and +20°C

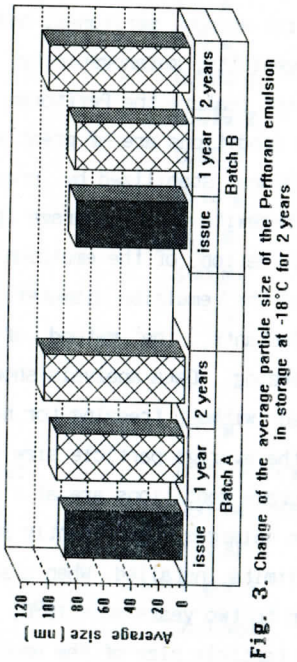


Fig. 3. Change of the average particle size of the Perftoran emulsion in storage at -18°C for 2 years



Table 4

Dependence of average size particle of Perftoran emulsion on frequent freezing and thawing ( μm )

| Number preparation | Number of freezing and thawing operation (-18°C) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | init. | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
| 71 | 0.07 | 0.078 | 0.084 | 0.085 | 0.090 | 0.090 | 0.089 | 0.090 |
| 72 | 0.07 | 0.072 | 0.071 | 0.078 | 0.081 | 0.083 | 0.084 | 0.085 |
| 73 | 0.07 | 0.073 | 0.075 | 0.081 | 0.083 | 0.085 | 0.086 | 0.087 |

It is known that when the fraction of the particles is more than 0.4 μm in size, the acute toxicity is rapidly increased. At present it is recognized that the most acceptable size is the emulsion particles with the mean diameter of 0.1 μm. Above particles are much smaller than erythrocytes (7-8 μm) but their number considerably exceeds the number of erythrocytes. It provides several hundreds times larger area of the surface for respiratory metabolism compared to blood cells. Figure 5 demonstrates a typical distribution of the emulsion according to size obtained by electronic microscope. Electrostatic electrography of the Perftoran emulsion has been procured by the method of negative contrast. Particle distribution (Fig. 4.) has been obtained by the method of SdFFF (Sedimentation Field Flow Fractionation) and Alliance Pharm. Corp. (USA) has kindly provided it.

As already mentioned perfluoromethylcyclohexylpiperidine has been introduced in the Perftoran to enhance a stability. It allows without additional surfactants (egg yolk phospholipids) to increase a stability of this preparation at room temperature to one month. It is well known that heat sterilization sharply deteriorates physicochemical emulsion properties, namely the average particle size enlarges that unfavourable influence all biological effects of perfluorocarbon emulsion including blood rheology and microcircula-

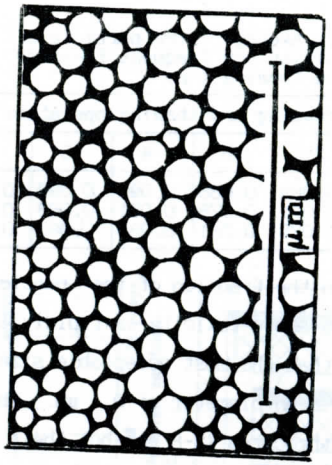


Fig. 5. Electrostatic electrography of the particle of Perftoran obtained by negative contrast method on electronic microscope

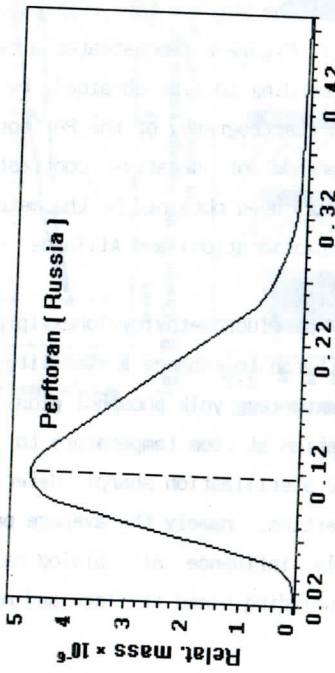


Fig. 4. Particle distribution of the Peftoran emulsion according to size



tion. Just from this point of view the Perftoran emulsion does not expose to heat sterilization compared to the components of Perftoran and technological devices which go through heat sterilization. Perftoran is prepared under aseptic conditions that are always confirmed by the test for sterility (Table 5). As is known hormone and other preparations that are widely applied in medicine are prepared on the same conditions but they cannot go through heat sterilization. That is why the particle distribution of the Perftoran emulsion is much preferable than of the foreign analogous (Table 6).

Tables 7 and 8 show data on the excretion of the Perftoran emulsion from organs after intravenous administration. Since one of the perfluorocarbons of Perftoran is perfluorodecalin with the period of halfexcretion, 7 days, the excretion of another perfluorocarbon, i.e perfluoromethylcyclohexylpiperidine, from the body is of the main interest. Presented data witness that when the Perftoran emulsion is being administered plethoric (10 ml/kg) or during blood substitution (50-60%), the whole excretion of perfluoromethylcyclohexylpiperidine is 18-24 months (Table 7). Data on excretion of perfluorocarbons and proxanol from blood channel and sarcoplasmic membranes of cardiocytes are also of great interest. The latter factor, probably, determines membranotropic effects of the Perftoran emulsion (Table 9).

Surfactants such as block-sopolimers of ethylene and propylene oxide with different molecular mass, proxanol have been used as stabilizers of the emulsions (Table 10). According to our and literature data the emulsions stabilized by proxanol have many positive properties of proxanol:

- lowering of blood viscosity due to the proxanol influence on regular blood elements;

Table 5

System of preclinical testing of Perftoran

| Physico-chemical research (studies) | Biological research (studies) |
|---|---|
| 1. Average size of particles<br>2. Content of PFCs<br>3. Content of Proxanol<br>4. Concentration of F-ions<br>5. pH | 1. Sterility<br>2. Pyrogenic effects<br>3. The complement-activating effects<br>4. Cardiotoxicity |

Table 6

Particle size distribution of Perftoran* and 20% Fluosol-DA emulsions according to size (Vorobyev S. 1993, Mitsuno T. 1981)

| Particle diameter (μm) | Perftoran | Fluosol-DA 20% |
|---|---|---|
| less than 0.1 | 85.2 % | 39.2 % |
| 0.1-0.2 | 14.4 % | 53.0 % |
| 0.2-0.3 | 0.4 % | 5.9 % |
| more than 0.3 | - | 1.9 % |
| average value | 0.07 | 0.118 |

* particle distribution has been determined by negative contrast method on electrone microscope

Table 7

Excretion of PFMCP from organs

| Conditions of administration | Time after administration | Content of PFMCP in rat organs summary in % to total dose of administered PFOC |
|---|---|---|
| blood substitution by Perftoran, 50-60% | 3 days | 3.2±1.92 |
| | 14 days | 8.1±1.99 |
| | 1 month | 1.5±1.74 |
| | 2 month | 9.5±0.32 |
| | 6 month | 2.9±0.21 |
| | 8 month | 2.1 |
| | 13 month | 1.25 |
| | 18 month | 0.5 |
| | 24 month | traces |
| Plethoric administration Perftoran in dose of 10 ml/kg | 7 days | 18.8 |
| | 1 month | 10.0 |
| | 3 month | 5.4 |
| | 6 month | 2.4 |
| | 12 month | 1.1 |
| | 18 month | 0.85 |
| | 24 month | traces |



Table 8

Content of PFMCP in rat tissues after 50-60% of blood substitution by Perftoran

| Organs | mg/g tissue | | | | | |
|---|---|---|---|---|---|---|
| | 14 days | 1 mont. | 6 mont. | 13 mont. | 18 mont. | 24 mont. |
| Liver | 6.7 | 3.42 | 1.22 | 0.14 | traces | not trac |
| Spleen | 1.4 | 6.4 | 3.9 | 0.14 | traces | not trac |
| Marrow | 9.8 | traces* | traces | not trac | not trac | not trac |
| Lymhpogangl | not trac | 0.48 | traces | not trac | not trac | not trac |

* traces - less than 0.05 mg/g tissue

Table 9

Excretion of PFD/PFMCP and Proxanol from blood vessels and sarcoplasmic in percentage from total dose of PFOC and Proxanol administered with Perftoran emulsion, data are obtained by analysing intergral intensity of $^{19}$F-NMR spectrum and by spectrophotometry

| Conditions of administrat. | Time after administr. (houres) | Summary content of PFOC in blood (%) | Proxanol content in blood (%) | PFOC content in membrans (mkg/mg protein) |
|---|---|---|---|---|
| Plethoric administrat. Perftoran in dose of 20 ml/kg | initial | 100 | 100 | - |
| | 1 | 95 | 20 | 2.1 |
| | 2 | - | 1.5 | - |
| | 4 | - | 1.0 | - |
| | 6 | - | 0.5 | 4.1 |
| | 12 | 70 | 0.3 | 6.0 |
| | 24 | 50* | 0.3 | 10.5 |
| | 72 | 20 | not traced | 5.2 |

* value nearing the period of halfexcretion

- diminution of erythrocytes aggregation contributing to better microcirculation;

- increase of erythrocytes resistance to osmotic and acid hemolysis;

- decreasing of coronary and vascular resistance;

- development of ventricular ejection;

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| less than 0.1 | 85.2 % | 39.2 % |
| 0.1-0.2 | 14.4 % | 53.0 % |
| 0.2-0.3 | 0.4 % | 5.9 % |
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| Marrow | 9.8 | traces* | traces | not trac | not trac | not trac |
| Lymhpogangl | not trac | 0.48 | traces | not trac | not trac | not trac |

* traces - less than 0.05 mg/g tissue

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| Plethoric administrat. Perftoran in dose of 20 ml/kg | initial | 100 | 100 | - |
| | 1 | 95 | 20 | 2.1 |
| | 2 | - | 1.5 | - |
| | 4 | - | 1.0 | - |
| | 6 | - | 0.5 | 4.1 |
| | 12 | 70 | 0.3 | 6.0 |
| | 24 | 50* | 0.3 | 10.5 |
| | 72 | 20 | not traced | 5.2 |

* value nearing the period of halfexcretion

- diminution of erythrocytes aggregation contributing to better microcirculation;

- increase of erythrocytes resistance to osmotic and acid hemolysis;

- decreasing of coronary and vascular resistance;

- development of ventricular ejection;

- the influence on blood flow in the great vessels, lowering of a resistance in them;
- increase of charge on the surface of cell membranes.

The main drawback of the emulsions stabilized by proxanol is side effects or reactogenicity which is caused as considered by activation of the complement system and connects with surfactants - proxanol or pluronic. Replacing an emulgator (Pluronic F-68) by phospholipids known as good stabilizers of the perfluorocarbon emulsions, foreign authors to a great extent cut the problem on reactogenicity. But at the same time they obtain a preparation without a number of positive properties which are typical for the proxanol emulsions (Table 11).

The activation degree of the complement system has been investigated by indirect assessment method of functional activity of C5a (anaphilatoxin) according to neutropenic response. It is due to intravascular activation of the complement is accompanied by redistributed neurophyl response consisting in momentary but short-time disappearance of neutrophils from blood channel and their aggregation in the vessels of lungs.

It has been established in the experiments that maximum value of neutropenic index is not to be more than 2 cond. units. Lower index, less reactogenicity is observed in the perfluorocarbon emulsions stabilized by proxanol. Perfluorocarbon Perftoran-type emulsions presented in Table 12 have neutropenic index 1.57 and 1.67 cond. units on the average respectively in 5 and 20 min. after intravenous administration of this preparation in the animal body.

The studies have shown that the proxanol solution alone does not have reactogenic properties as well as that one specially treated by high pressure using desintegrator. Apparently, reactogenici-



Table 10

Physicochemical and biological properties of surfactants (proxanol)

| Proxanol | Mol.weight /D/ | Concentr. react.gr.(M/M) | Concentr. hydrophpb.gr.(%) | $LD_{50}$ g/kg |
|---|---|---|---|---|
| F-168/38/ (Russian) | 5700 | 0.24 | 19 | 24.0 |
| F-268/35/ (Russian) | 7200 | 0.48 | 19.5 | 15.0 |
| F-268/1/ (Russian) | 8000 | 0.20 | 19.0 | 14.2 |
| Pluronic F-68 (Serva) | 8300 | 0.20 | 21 | 9.4 |

Table 11

Dependence of the complement activating effects (neutropenic index) on preparation method of PFC-emulsions

| Components | Pressure cond.unit. | T(°C) c.unit. | Neutropynic index (cond.unit.) after 5 min. | after 20 min. |
|---|---|---|---|---|
| Control | - | - | 1.27 | 1.42 |
| Poliglukin | - | - | 1.28 | 1.44 |
| Proxanol | - | - | 1.33 | 1.21 |
| Proxanol | 12.3 | 14.0 | 1.62 | 1.20 |
| Supernatant emulsion | - | - | 1.26 | 1.42 |
| Emulsion | 3.3-7.80 | 7-9 | 1.69 | 1.72 |

ty of Pluronic F-68 is caused by more expressed detergent properties compared to proxanol (Table 10). Here it can be assumed that some phisicochemical changes in the emulsion occur on the other level that is responsible for activation of the complement system. The method of nuclear magnetic resonance has been used in order to reveal this effect. During the experiments it has been established

that the spectrum of the reactogenic emulsion has a signal for CH group which essentially differs from that one of the same group in the areactogenic emulsion. In the reactogenic emulsion a signal is characterized by the appearance of "hard" fraction - wide component and decrease of amplitude of the CH signal. It should be emphasized that the appearance of the wide component is also observed in the areactogenic emulsion but is significantly smaller in respect of amplitude of the narrow component. The ratio of amplitude of the narrow component to the hard fraction is a fraction index that correlates with the neutropenic index (Fig. 6).

Table 12

Changes of neutrophils amount (*10%) and neutropenic index (cond.units) of indirect factor of complement activaiting sistem in rabbit peripheric blood in 5 and 20 minutes after administration of Perftoran emulsion prepared under pressure of 3.3 and 7.8 cond.units.

| Emulsion N | Initial amount neutrophils | After 5 min | | After 20 min | |
|---|---|---|---|---|---|
| | | neutrophils | index | neutrophils | index |
| V-1 | 2842 | 4149 | 0.96 | 1809 | 2.19 |
| V-2 | 5628 | 6663 | 1.20 | 3740 | 2.12 |
| V-3 | 2782 | 2630 | 1.44 | 3150 | 1.25 |
| V-4 | 2345 | 1614 | 2.30 | 3811 | 0.79 |
| V-5 | 2075 | 1922 | 1.51 | 1380 | 2.09 |
| V-6 | 1426 | 1888 | 1.60 | 1833 | 1.60 |
| V-7 | 3560 | 3016 | 1.65 | 2997 | 1.67 |
| V-8 | 1564 | 1063 | 2.06 | 1484 | 1.47 |
| V-9 | 1782 | 1317 | 1.89 | 1654 | 1.51 |
| average value of index | | | 1.57 | - | 1.67 |



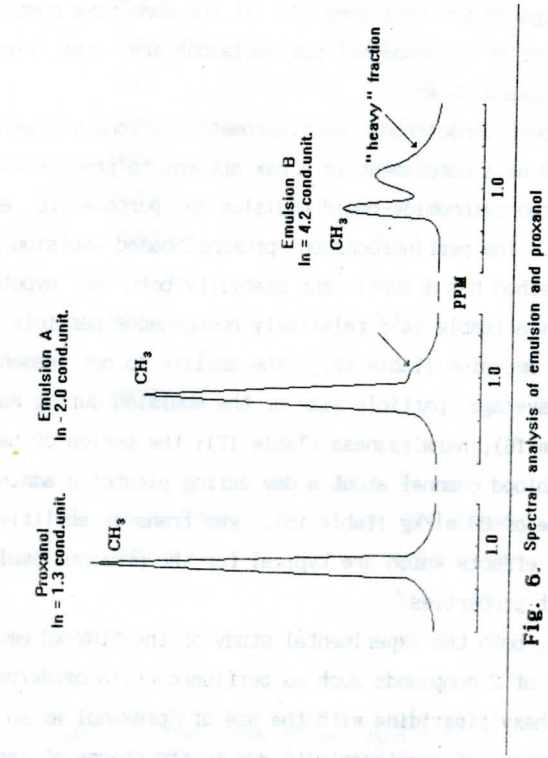


**Fig. 6.** Spectral analysis of emulsion and proxanol

IV. The next stage - the advent of fluorocontrast agents for diagnosis on the basis of the second generation perfluorocarbons (perfluorooctylbromide/perfluoromethylcyclohexylpiperidine) - has completed by the development of the VIMM-RC emulsion (Table 13).

The PFOB (perfluorocontrast compound) - based emulsions stabilize with egg yolk phospholipids since proxanol turned to be a low effective emulgator for this compound. At the same time hemorheological properties of the phospholipid emulsions are less expressed than of the proxanol ones.

Another perfluorocarbon, perfluoromethylcyclohexylpiperidine, has been used as a complement in a new attempt to produce the proxonal perfluorooctylbromide-based emulsion on purpose to enhance its stability. The perfluorocarbon proxanol-based emulsion obtained by this method has a sufficient stability both at hypothermia and normothermia (table 14); relatively homogeneous particle composition of the emulsion (Table 15); the ability do not essentially change the average particle size of the emulsion during multiple thawing (Table 16); harmlessness (Table 17); the period of halfexcretion from blood channel about a day during pleutoric administration in a dose of 20 ml/kg (Table 18); gas transfer abilities; hemorheological effects which are typical for the proxanol emulsions, fluorocontrast properties.

Therefore, both the experimental study of the VIMM-RC emulsion on the basis of 2 compounds such as perfluorooctylbromide/perfluoromethylcyclohexylpiperidine with the use of proxanol as an emulgator and decrease of reactogenicity due to the change of technology make it possible to enhance the range of emulsion application from fluorocontrast agent to plasma substituting preparations with gas transfer ability.



Table 13

Contrast perfluorobrom emulsion - "VIMM-RC"

COMPONENTS :

| | | |
|---|---|---|
| PFOB | - | 13.0-26.0 g |
| PFMCP | - | 6.5-13.0 g |
| Proxanol | - | 4.0 g |
| NaCl | - | 0.6 g |
| KCl | - | 0.039 g |
| $MgCl_2$ | - | 0.019 g |
| $NaH_2PO_4$ | - | 0.02 g |
| $NaHCO_3$ | - | 0.13 g |
| Glucose | - | 0.2 g |
| $H_2O$ | - | 100 ml |

PROPERTY :

| | | |
|---|---|---|
| $F^-$ | - | $< 10^{-5}$ M |
| Av. diameter | - | 0.07-0.15 μm |
| Osmolarity | - | 280-340 mOsm |
| Viscosity | - | 2.5-3.5 cPs |
| pH | - | 7.4-7.6 |

Table 14

The change of the average particle size ( μm ) of the VIMM-RC emulsion during storage at +4°C and +20°C

| T(°C) of storage | Time of storage (days) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | issue | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
| + 4°C | 0.076 | 0.077 | 0.076 | 0.076 | 0.080 | 0.086 | 0.092 | 0.096 |
| +20°C | 0.076 | 0.080 | 0.082 | 0.085 | 0.094 | 0.094 | 0.110 | 0.114 |

Table 15

Particle distribution of the VIMM-RC and Perftoran emulsions (method of negative contrast)

| Emulsions | Particle diameter (μm) | | | |
|---|---|---|---|---|
| | less than 0.1 | 0.1 - 0.2 | 0.2 - 0.3 | more than 0.3 |
| VIMM-RC | 77% | 22% | 1.0% | not observed |
| Perftoran | 86.5% | 12.6% | 0.9% | not observed |

Table 16

The change of the average particle size (μm) of the VIMM-RC emulsion at multiple thawing and freezing (-18°C)

| Indicator | A number of thawing/freezing (-18°C) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | issue | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
| aver. particle size (μm) | 0.076 | 0.080 | 0.082 | 0.083 | 0.083 | 0.087 | 0.090 | 0.097 |
| | 0.076 | 0.079 | 0.081 | 0.083 | 0.085 | 0.088 | 0.089 | 0.098 |

Table 17

Acute toxicity of the VIMM-RC emulsion if it is intraperitoneal administration in mice.

| A number of mice in group | Dosage (ml/mouse) | Time of observation ( days ) | | | | | | | Response |
|---|---|---|---|---|---|---|---|---|---|
| | | 2 | 4 | 6 | 8 | 10 | 12 | 14 | |
| n=10 | 0.6 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | no |
| n=10 | 0.8 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | no |
| n=10 | 1.0 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | no |

Table 18

Excretion of PFD/PFOB from blood vessels of rabbit (n=3) in percentage from total dose of PFOC administered with VIMM-RC emulsion, data are obtained by analysing integral intensity of $^{19}$F-NMR spectrum

| Conditions of administration | Time after administration (houres) | Summary content of PFOC (%) |
|---|---|---|
| Plethoric administration in dose of 20 mg/kg | 1 | 97.1 |
| | 3 | 93.7 |
| | 6 | 86.6 |
| | 9 | 81.0 |
| | 21 | 62.0 |
| | 24 | 54.0* |
| | 27 | 44.2 |

* value nearing the period of halfexcretion.



This information material summarizes some directions of the activity in the field of production of proxanol perfluorocarbon compounds-based emulsions in the Institute of theoretical and experimental biophysics RAS and Perftoran Pharmaceutical Company. The authors do not pretend owing to restricted space to put the material completely and acknowledge all colleagues for provided support.