A new, heavier-than-water silicone oil: a solution of perfluorohexyloctane in polydimethylsiloxane

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PURPOSE. *To prepare and explore new solutions of semifluorinated alkane in silicone oil, which have a specific gravity slightly higher than silicone oil and vitreous fluid (referred to in the following as heavier-than-water silicone oils (HWSs), and to investigate,* in vitro, *whether HWSs can be used to plug retina holes, while allowing dehydration of the subretinal space.*
METHODS. *HWS solutions were prepared with silicone oil 5000 and perfluorohexyloctane (F6H8). The stability was investigated under different conditions. The viscosity was determined by means of a capillary viscometer. The surface and interface tension were measured using the ring method.*
RESULTS. *HWSs are insoluble in an aqueous medium. Densiron®68 (HWS 1.06) is a transparent homogeneous liquid which is slightly heavier (1.06 g/cm^3) than water and has a refractive index close to that of vitreous liquid. Densiron®68 (HWS 1.06) has a low viscosity (1480 mPas) and interface tension (40.82 mN/m), making it an effective tamponade in the surgical treatment of an inferior detached retina. In addition, the interfaces between Densiron®68 and other perfluorocarbon liquids are clearly visible. However, the interface layer between Densiron®68 and water is not clear. Finally, all HWSs are stable over the long term at ambient temperatures, as well as physically and thermally resistant.*
CONCLUSIONS. *Due to its physiochemical properties, Densiron®68 could meet the requirements for a heavier-than-water tamponade. (Eur J Ophthalmol 2005; 15: 627-37)*

KEY WORDS. *F6H8®, Perfluorohexyloctane, Semifluorinated alkane, Densiron®68, Heavier-than-water silicone oil (HWS), Storage stability, Spreading coefficient*

INTRODUCTION

Perfluorocarbon liquids (PFCLs) and silicone oils are established intraoperative tools in vitreoretinal surgery (1-3). The high specific gravity of PFCLs is likely to cause mechanical retinal damage when applied as a long-term vitreous substitute. Therefore, a new class of compounds has been developed; namely, semifluorinated alkanes (RFRH) with a relatively low specific gravity compared to PFCLs (4-7). Perfluorohexyloctane (F6H8®), one of the semifluorinated alkanes (RFRHs), consists of a perfluoro-

carbon (RF) and a hydrocarbon segment (RH). F6H8® is used as a temporary tamponade in the treatment of complicated retina detachments and as an intraoperative tool in retinal surgery (8-10).

To date, there has been no single tamponade agent that fulfills the requirements for a tamponade to simultaneously treat superior and inferior retinal detachments (11). Interest in combining two substances with different specific gravities to tamponade the whole retina is increasing. Silicone oil is useful as a tamponade in the case of superior retinal detachments, and perfluorocarbon in the case of
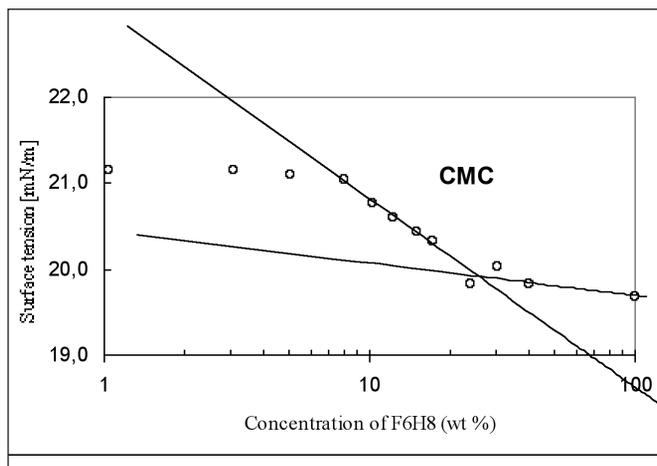
Fig. 1 - *Surface tensions of heavier-than-water silicone oils (HWSs) as a function of the F6H8 concentration.*

inferior retinal detachments. If the two could be used together, they would simultaneously act as a tamponade and a mechanical support for both portions of the retina. Because of their immiscibility and the wide difference in the specific gravities of these two liquids, silicone oil and PFCLs could be injected into the eye chamber without mixing (referred to in the following as double filling) (12). However, because of the incompatible side effects of PFCLs in animal studies, they are not recommended as long-term tamponades (13).

In an attempt to overcome these disadvantages, PFCLs were replaced with F6H8® to produce a double filling tamponade. The combination of two "CE-certified" tamponade agents, silicone oil and F6H8, for double filling has been widely studied by a multicenter study group called GRIDOR (Italian Group for Double Filling) (14-16). F6H8
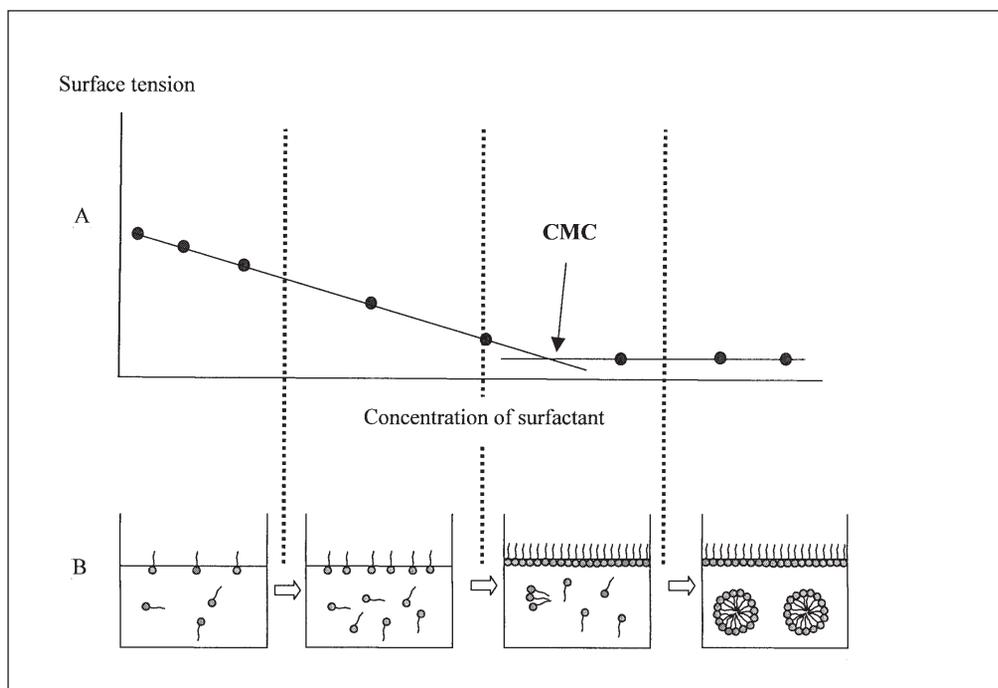

Fig. 2 - *Relationship between surface tension and micelle formation as a function of the surfactant concentration. Surface tension versus surfactant concentration* **(A)**. *Critical micelle concentration is determined as the point of intersection of the two lines. Aggregation process of surfactants* **(B)**. *(BJO 2004; 88: 692-6 with permission from BMJ Publishing Group)*

and silicone oil are normally soluble in one another. However, since F6H8 and silicone oils are immiscible with each other unless an external force, such as stirring or sonification, is applied, this binary system can be used to exploit the tamponing qualities of both substances and to tamponade the whole retina. It has been reported that neither intraocular emulsification nor damage to membranes is caused by the binary system of silicone oil and

F6H8 and that, in particular, the double tamponade is useful for reattaching the retina, which avoids inferior recurrences (11).

Due to its hydrocarbon segment (RH) in the molecule, F6H8® has application potential to dissolve lipophilic substances and silicone oils. F6H8® has already been investigated as a biocompatible solvent for silicone oil adhesions on intraocular lenses (17) and is recommended as a
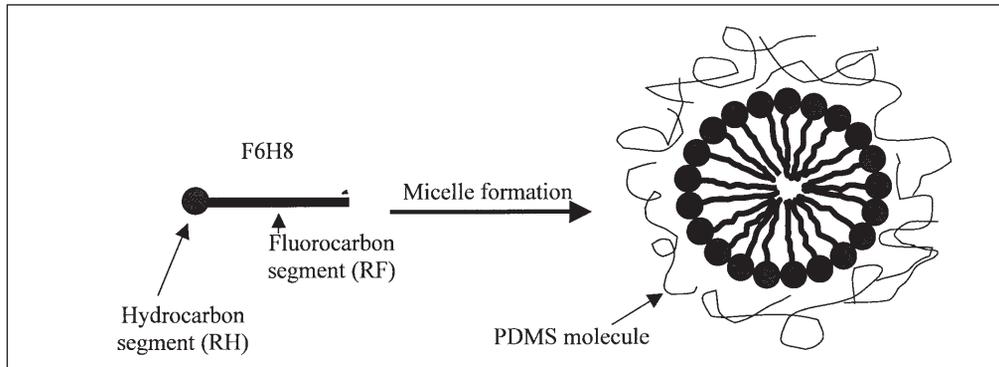
Fig. 3 - *Micelle formation in F6H8/PDMS system. (from BJO 2004; 88: 692-6 with permission from BMJ Publishing Group)*

"wash out" solution after silicone oil removal.

Meinert has developed and patented (18) solutions of semifluorinated alkane in silicone oil that are homogeneous and have a higher specific gravity than water (HWSs). HWSs, consisting of F6H8 and silicone oil 5000, provide an effective long-term tamponade for the lower retinal sectors (18). In a previous article, Meinert and Roy reported on the solubility of semifluorinated alkanes (RFRH) in silicone oils and the important physical properties of HWSs (4). Several studies have demonstrated the possibility of HWSs as long-term endotamponade agents (5, 17-23).

The in vitro study presented had the following objectives:

1. To prepare "heavier-than-water silicone oil" (HWS) solutions with a slightly higher specific gravity than vitreous liquid to tampon the inferior retina in cases of complicated retinal detachment, and with a relatively low viscosity compared to silicone oil 5000.
2. To study their surface and interface tensions.
3. To evaluate the stability of HWSs under different conditions.
4. To study the spreading behavior of HWSs over water.

TABLE I - CHEMICAL AND PHYSICAL PROPERTIES OF F6H8® AND SILURON 5000

	F6H8®	Siluron 5000
Purity (%)	>99	>99
Boiling point (°C)	223	–
Specific gravity (g/cm^3, 25 °C)	1.33	0.97
Viscosity (mPas, 25 °C)	3.44	5250
Refractive index (n_D^{20})	1.343	1.404
Surface tension (mN/m, 25 °C)	19.65	20.80
Interface tension (mN/m, 25 °C)	45.27	43.01
Volatile compounds (%)	–	<0.05 at 200 °C, 24 h

TABLE II - CHARACTERISTICS OF HEAVIER-THAN-WATER SILICONE OILS (HWS$_S$)

	HWSs		
	HWS 1.01	HWS 1.03	HWS 1.06*
F6H8 (wt%^{3})	17.4	24.0	30.5
Specific gravity (g/cm^3)	1.01	1.03	1.06
Viscosity (mPas)	3167	1948	1480
Refractive index ($n(n_D^{20}$)	–	–	1.387

The numbers 1.01, 1.03, and 1.06 after HWS indicate the specific gravities of HWSs
* Densiron® 68 has been approved (CE-certified) in Europe for intraocular use
wt% = Weight percent

METHODS

Substances

The current studies were performed using highly puri-fied implants, Siluron 5000 (silicone oil 5000; Fluoron GmbH, Neu-Ulm, Germany) and perfluorohexyloctane (F6H8®, Fluoron GmbH, Neu-Ulm, Germany), which are CE-certified as liquid implants for ophthalmology. Their properties are listed in Table I.

Experiments

Preparation of HWSs. To prepare HWSs, F6H8 was added dropwise to silicone oil 5000 while stirring, and the mixture stirred until it was completely homogeneous. The concentrations of F6H8 in the HWSs were 17.4, 24.0, and 30.5 weight percent (wt%), respectively, for HWS 1.01, HWS 1.03, and HWS 1.06 (in the following, the numbers 1.01, 1.03, and 1.06 refer to the respective specific gravi-ties of the HWSs). Based on its composition, the prepara-tion HWS 1.06 is registered under the name Densiron® 68.

Viscosity. The viscosities of the HWSs were determined using a semi-automatic Ubbelohde capillary viscometer (capillary type 532 40/IV, Schott).

***In vitro* aspiration experiment using a model eye.** A total of 4 mL of oil were placed in each glass model eye. After incubation at 37 °C for 1 h, the oils were removed by means of an 18-gauge needle using a vacuum pump. The pressure was maintained at 17 mbar (12.8 Torr) during oil aspiration. The oil removal experiment was performed three times for each sample.

Interfacial visibility between Densiron®68 and other implants. To evaluate the visibility of the interfacial layer between two intravitreal substances, PFCLs (perfluorooc-tane, perfluorodecalin), RFRHs (F6H6, F6H8), and water were placed in 10-mL bottles and Densiron®68 was then added to each bottle, avoiding turbulence.

Surface and interface tension measurements. Surface (against air) and interface tension (against water) measure-ments were performed with the Krüss K-12 processor ten-siometer, employing the DuNuöy-ring method.

Solubility of PFCLs in silicone oil 1000 and Densiron®68. In order to determine the solubility of PF-CLs in these oils, 4 mL of Densiron®68 and silicone oil 1000 were placed in each test tube. PFCLs were gradual-ly added to each of the oils while stirring. In addition, three droplets (20 µL) of PFCL were first placed in the glass vials and then 4 mL each of Densiron®68 and sili-cone oil 1000 were added to examine whether PFCL bub-bles dissolve in the oils. These samples were kept at am-bient temperatures, avoiding turbulence.

Storage stability. To test the storage stability, 10 mL of HWS were placed in each glass vial and these were stored at corresponding temperatures. For the short-term storage stability study, HWSs were stored at correspond-ing temperatures. Assessment of the long-term storage stability of HWSs was performed by centrifuging at 13,000 rpm at 20 °C for 1 h using an ultracentrifuge

TABLE III - VISCOSITY AND ASPIRATION RATE OF OILS AT 37 °C

Oils	Viscosity (mPas)	Aspiration rate (mL/min)
Silicone 1000	1020	2.79
Silicone 5000	5250	0.55
Densiron®68	1480	2.39

TABLE IV - VISIBILITY OF THE INTERFACIAL LAYER BETWEEN TWO SUBSTANCES AND TURBIDITY AT THE INTERFACE

	Δn_D^{20}	Interfacial visibility	Turbidity
D68/PFO	0.1170	Good	No
D68/PFD	0.0770	Good	No
D68/water	0.0543	Moderate	No
D68/silicone 5000	0.0170	Poor	No

D68 = Densiron®68; PFO = Perfluorooctane; PFD= Perfluorodecalin; Δn_D^{20}= Difference in the refractive indexes of two substances at 20 °C.

(Beckman, Ti 50.4 rotor). For the thermal stability tests, each HWS was sterilized at 121 °C for 20 minutes and 160 °C for 2 hours. To test the long-term storage stability, HWSs stored for at least 4 years were used.

***In vitro* cytotoxicity assay.** For the cytotoxicity assay of HWSs, human adult retinal pigment epithelial cells (ARPE-19) and mouse cells (L929) were used. This study was carried out at BSL BIOSERVICE Scientific Laboratories GmbH (Munich, Germany) according to the international guideline ISO 10993: HWSs stored for more than 4 years were extracted for 24±2 h with a cell culture medium and the extracts were incubated with each of the cells (L929 and ARPE-19) for 68 to 72 h. The protein content of the individual culture was then analyzed as a measure of cytotoxicity and compared to that of the untreated control samples.

RESULTS

Specific gravity and viscosity

The measured specific gravities and viscosities of oils are listed in Table II. All HWSs used in this study had a specific gravity slightly higher than that of water (between 1.01 and 1.06 g/cm^3). The specific gravities of the HWSs increased in relation to the F6H8 content of the solution. The viscosities of the HWSs ranged between the values for silicone oil 1000 and 5000. The viscosity of Densiron®68 (HWS 1.06) was the lowest (1480 mPas) and close to the value for silicone oil 1000.

In vitro *aspiration experiment using a model eye*

The mean aspiration rates of oils are listed in Table III. The aspiration rate depended on the viscosity of the used oils. The aspiration time of Densiron®68 was much shorter than that of silicone 5000. There was no significant difference between silicone 1000 and Densiron®68.

Refractive index

Densiron®68 (HWS 1.06), which was selectively investigated in this study, is a homogeneous and transparent liquid and has a refractive index of 1.387 (the refractive index of water and silicone oil is 1.333 and 1.404, respectively).

Interfacial visibility between intravitreal substances

In *in vitro* experiments, the interfacial layers of the two tamponade agents were clearly visible in the case of Densiron®68/PFO and Densiron®68/PDF (Tab. IV), while the interfacial visibility between Densiron®68 and silicone oil was poor. The interfaces of two phases were maintained for at least 12 weeks.

Surface and interfacial tension of HWSs

The results of the study of the surface (against air) and

TABLE V - SURFACE AND INTERFACE TENSIONS OF HEAVIER-THAN-WATER SILICONE OILS (HWS$_S$)

	HWS 1.01	HWS 1.03	HWS 1.06*
Surface tension (mN/m)	19.84	19.43	19.13
Interface tension (mN/m)	56.25	45.43	40.82

*Densiron®68

TABLE VI - COMPARISON OF THE STABILITY OF HEAVIER-THAN-WATER SILICONE OILS (HWSS) DURING STORAGE AT DIFFERENT TEMPERATURES FOR 24 HOURS

Storage temperature (°C)	HWSs		
	HWS 1.01	HWS 1.03	HWS 1.06
0	HT	HT	O
2	HT	HT	HT*
5	HT	HT	HT
10	HT	HT	HT

*HWS became transparent and homogenous (within 30 min) when stored at room temperature
HT = Homogeneous and transparent; O = Opaque

interface tensions (against water) of HWSs are listed in Table V. Compared to the starting substances (F6H8 and silicone oil 5000), the surface tensions of three HWSs, HWS 1.01, HWS 1.03, and HWS 1.06, were lower than that of silicone oil. The interface tensions of HWSs were in the range 40.82 to 56.25 mN/m. Both the surface and interface tensions of the HWSs decreased with increasing specific gravity (Tab. V).

The nature of the F6H8/silicone oil solution

In this study, in order to investigate the nature of binary HWS, the surface tensions of HWSs were measured as a function of the F6H8 concentration. The measured values of surface tensions are plotted against the concentration of F6H8 in Figure 1. The surface tensions decrease continuously from 8 to about 24 wt% with increasing concentration. Above 24 wt%, surface tension remains virtually constant. Critical micelle concentration (CMC) is read off from the intersection of two straight lines, one in the descending part of the curve, the other through the plateau. The CMC of the HWS is about 25 wt%. The existence of CMC indicates that F6H8 aggregates to form micelles in silicone oil.

Stability of HWSs

The stability of HWSs was investigated at different temperatures and varying storage times in this study. First, for the short-term stability test, the samples were stored at temperatures of 0 °C, 2 °C, 5 °C, and 10 °C for 24 hours. The test results are summarized in Table VI. HWS 1.01 and HWS 1.03 were very stable at all storage temperatures. When stored at 0 °C and 2 °C, HWS 1.06 evidenced temporary opaqueness, but did not solidify. HWS 1.06 became spontaneously transparent and homogenous within 30 minutes when it remained at room temperature. Above 5 °C, there was neither phase separation nor

turbidity in HWS 1.06. Furthermore, HWS 1.06 (Densiron®68) was investigated at varying storage intervals and different temperatures. The results in respect to the storage stability of HWS 1.06 are summarized in Table VII. When stored at -18 °C, HWS 1.06 solidified within 1 hour and became slightly opaque. However, this opaqueness disappeared after standing at room temperature. When stored for a long period at -18 °C, phase separation occurred after thawing. The separated two phases were transparent and became homogeneous after a holding time of at least 1 day at room temperature. After storage for 1 to 24 hours at 0 °C, HWS 1.06 was opaque, but had not solidified. HWS 1.06 became homogeneous and transparent after standing at room temperature. These samples stored at 2 °C for over 12 hours became temporarily opaque. These samples became spontaneously homogeneous and transparent within 30 minutes. When stored at 5 °C, no optical change was observed.

In addition, the study of long-term stability was performed with HWSs stored for more than 4 years at room temperature. The results are shown in Table VIII. After this period, neither optical change nor phase separation was observed in any HWSs, even when centrifuged at 13,000 rpm for 1 hour. Moreover, none of the HWSs evidenced phase separation after sterilization at 121 °C for 20 minutes and 165 °C for 2 hours.

Based on GC-MS analysis of the OH endgroup by means of NMR and Karl Fischer titration, there was no time-dependent change in the chemistry of the components. Furthermore, the viscosities of the HWSs were constant over a long storage time (irrespective of the storage time) after reaching the CMC level during preparation of the HWS.

Finally, the in vitro biocompatibility of HWSs stored for more than 4 years was tested using human adult retinal pigment epithelial cells (ARPE-19) and mouse cells (L929). Table VIII shows the test results for cell cytotoxicity. None

TABLE VII - STORAGE STABILITY OF HEAVIER-THAN-WATER SILICONE OIL (HWS) 1.06 AT DIFFERENT TEMPERATURES

Storage temperature (°C)	Storage time, hr				
	1	3	12	24	Long term
-18	O, S	O, S	O, S	O, S	O, S (38 days)
0	O, F	O, F	O, F	O, F	O, F (4 days)
2	HT, F	HT, F	HT*, F	HT*, F	HT*, F (4 days)
5	HT, F	HT, F	HT, F	HT, F	HT, F (4 days)

*HWS 1.06 became homogeneous and transparent within 30 minutes
O = Opaque (turbid); S = Solidified; F = Fluid; HT = Homogeneous and transparent

of the test samples significantly inhibited cell growth, that is, all HWSs stored for more than 4 years were nontoxic and biocompatible.

Solubility of PFCLs in silicone oil 1000 and Densiron®68

The results regarding the solubility of PFCLs are shown in Table IX. The maximal solubility of PFCLs in silicone oil 1000 was 3.1 vol% and up to 5.1 vol% in Densiron®68, when external forces were applied. These solutions were optically transparent and homogeneous up to the point of maximal solubility. In both cases, an excess of PFCLs resulted in phase separation. These systems became opaque if stirred, or other external forces were applied. Furthermore, excessive PFCLs extracted up to 83 vol% F6H8, as a component of Densiron®68, due to the better solubility of F6H8 in PFCLs than in silicone oil. In in vitro diffusion experiments, droplets of PFD or PFO penetrated Densiron®68 within 24 hours, while PFCLs remain unchanged under silicone oil for up to several months.

DISCUSSION

The significant objective of this study was to investigate whether F6H8 forms highly ordered structures (micelles) in silicone oil, or whether the binary system exists only as a homogeneous and purely physical solution of F6H8 in silicone oil. The phase behavior of solutions of F6H8 in silicone oil has been described for the first time by Meinert (24). To date, there has been no detailed information on the evidence of micelle formation in these systems. The fact that surfactant molecules self-assemble into large aggregates – micelles – in solution can be demonstrated experimentally. Measurement of the surface tension is one of the methods to determine the CMC. Figure 2 illustrates the relationship between the surface tension and micelle formation as a function of the surfactant concentration. The proportion of molecules present on the surface, or as micelles in the bulk of the liquid, chiefly depends on the concentration of the surfactants. At low concentrations surfactants will favor arrangement on the surface (Fig. 2B). As the surface becomes crowded with surfactants, more surfactant molecules will arrange themselves into micelles. Above the CMC, the surface becomes completely loaded with surfactants and any further additions result in the formation of micelles in the solution (Fig. 2B). Semifluorinated alkanes (RFRHs) have the basic amphiphilic diblock architecture required to act as surfactants in binary mixtures with hydrocarbon or fluorocarbon solvents. RFRHs tend to be adsorbed and aggregated in hydrocarbon and fluorocarbon solvents and also in their mixtures (25). The surface tensions of the HWSs measured decrease with an increasing concentration of F6H8 until a breaking point, indicating the CMC, from which point on, the surface tension remains relatively constant (Fig. 1). The existence of micelles of F6H8 in silicone oil has been proven by determining the CMC. Polydimethylsiloxanes (PDMS), as lipophilic compounds, are soluble in lipophilic hydrocarbons but insoluble in lipophobic perfluorocarbons. The solubility of amphiphilic semifluorinated alkanes in PDMS is determined by the lipophilic RH segments of RFRH molecules. Therefore, it must be assumed that the lipophilic RH segments turn towards the macromolecule PDMS to form the outer shell of a micelle, while the lipophobic RF segments are located inside to form a fluorophilic core (Fig. 3). In this study, solutions of perfluorohexyloctane (F6H8) in silicone oil, with a slightly higher specific gravity than silicone oil and vitreous fluid (HWSs), were explored. Oils for use in vitreoretinal surgery should be transparent and have a refractive index close to that of the vitreous fluid. HWS 1.06, with a specific gravity of 1.06 g/cm^3 and a viscosity of 1480 mPas (Densiron®68), was investigated in this study. Densiron®68 is transparent and its refractive index is 1.387 (the refractive index of water and silicone oil are 1.333 and 1.404, respectively).

TABLE VIII - LONG-TERM STABILITY OF HEAVIER-THAN-WATER SILICONE OILS (HWS$_S$)

HWSs	Transparency[*]	Centrifugation[†]	Heat treatment[‡]	Viscosity[§]	Biocompatibility	
					L929 cell	ARPE cell
HWS 1.01	No change	Stable	Stable	No change	Nontoxic	Nontoxic
HWS 1.03	No change	Stable	Stable	No change	Nontoxic	Nontoxic
HWS 1.06	No change	Stable	Stable	No change	Nontoxic	Nontoxic

*HWSs stored for more than 4 years at ambient temperature were homogenous and clear
†HWSs stored for more than 4 years at ambient temperature were centrifuged at 13,000 rpm for 1 hr
‡HWSs stored for more than 4 years at ambient temperature were sterilized at 121 °C for 20 min and at 165 °C for 2 hr
§During storage

Practical experience has shown that silicone oil is disadvantageous as a tamponade in the case of an inferior detached retina, because its specific gravity is less than that of the vitreous fluid (2, 3). A number of perfluorocarbon liquids (PFCLs) have also been investigated for use in intravitreous surgery (1, 2). The high specific gravity of PFCLs makes them useful as a tamponade in the case of an inferior detached retina. The extremely high specific gravity could, however, not only damage the sensitive retinal tissue, but also increase the chance of dispersion of the liquid in the vitreous fluid (9, 13).

Unlike PFCs, the HWSs used in this study had a slightly higher specific gravity than water (between 1.01 and 1.06 g/cm^3) (Tab. II). The slightly higher specific gravity of HWSs than that of water could theoretically result in an inferior tamponade without mechanical damage to the retina, due to drainage of the subretinal fluid and contact with the internal surface of the vitreous cavity.

As a rule, the duration of removal of the internal tamponade from the eye depends on the viscosity of the oils used. Wolf et al's recent work has provided an indication that, clinically, aspiration of the 3800-cSt heavy silicone oil (Oxane HD) takes approximately 10 minutes (22). To date, there has been no publication that deals with the degree of difficulty of removing Densiron®68 during the intraocular operation. Based on the *in vitro* studies (Tab. III), the low viscosity of Densiron®68 (1480 mPas) could facilitate injection and removal with microsurgical instruments and manipulation of the retinal during vitrectomy. It is well known that low-viscosity PFCs and silicone oils tend to emulsify and disperse in the eye cavity. It has been reported that, in spite of the low viscosity (1480 mPas), Densiron®68 causes neither clinically significant dispersion nor emulsification during the intraocular operation (23). This effect could be explained by the fact that F6H8 molecules form spherical micelles in silicone oil (Figs. 2 and 3). The spherical symmetry of micelles tends to mobilize the oil. Thus, the viscosity of oils is reduced. In addition, F6H8 molecules are surrounded by silicone oil 5000 and are shielded from water. Therefore, Densiron®68 is very stable and may improve resistance to emulsification and dispersion compared to silicone oil 1000 and PFCLs.

The temporary tamponade agent PFCL should allow direct replacement with endotamponades such as Densiron®68 or silicone oil at the end of vitreoretinal surgery. Therefore, the visibility of the meniscus of the substances used is important during tamponade replacement. According to Table IV, the interfacial layer between the endotamponade agent Densiron®68 and tamponade liquids such as PFCLs is clearly visible, due to the differences in the refractive indexes ($\Delta n_D^{20} > 0.07$). In the case of systems consisting of Densiron®68 and silicone oil 1000, or silicone oil 5000, the interface is not clearly visible. Therefore, the direct exchange of Densiron®68 for silicone oils should be avoided.

The surface (against air) and interface tensions (against water) of an internal tamponade agent provide useful information on the effectiveness of tamponing. Fluorocarbons and silicone oils have low surface tension values (ranging from 18 to 20 mN/m). The surface tensions of HWS 1.01, HWS 1.03, and HWS 1.06 (Tab. V) are comparable to both starting substances, F6H8 and silicone oil 5000 (Tab. I). The surface tensions of HWSs decrease as the concentration of F6H8 in HWSs increases. The measurement of surface tension indicates that the intermolecular forces of silicone oil become weak as the concentration of F6H8 increases. As demonstrated in Table V, the interfacial tensions of HWSs also decrease the higher the F6H8 content. It is known that high interfacial tension limits the passage of PFCLs though retinal breaks and increases the effectiveness of the PFCLs as an intraocular tamponade for retinal breaks (26). The link between the physical properties of F6H8, such as the specific

TABLE IX - SOLUBILITY OF PERFLUOROCARBON LIQUIDS IN SILICONE OIL 1000 AND IN DENSIRON®68 AT 37 °C

	Silicone oil 1000	Densiron®68
PFD	3.1 vol %	5.1 vol %
PFO	2.4 vol %	4.1 vol %

PFD = Perfluorodecalin; PFO = Perfluorooctane

TABLE X - CALCULATED SPREADING COEFFICIENTS OF SOME TAMPONADE AGENTS BASED ON MEASURED VALUES OF SURFACE AND INTERFACE TENSIONS

Tamponade agent	Spreading coefficient
HWS 1.01	-4.07
HWS 1.03	7.14
HWS 1.06 (Densiron®68)	12.05
F6H8®	7.08
Silicone oil 5000	8.19
Perfluorodecalin	-4.80
Perfluorooctane	3.00

HWS = Heavier-than-water silicone oils

gravity and the interfacial tension, and the effectiveness of the tamponade has been reported (23, 26-27). Wetterqvist et al (27) studied the behavior of pure F6H8 and a series of HWSs, HWS 1.01, HWS 1.03, and HWS 1.06, in the model eye chamber without indent. The contact area between the HWS and the eye chamber increases with increasing specific gravity of the HWSs. It has been reported that the effectiveness of an internal tamponade agent depends on its ability to make contact with the internal surface of the vitreous cavity (27). In the short term, this is controlled by the agent's specific gravity and interfacial tension. In the long term, the viscosity of the material is critical for maintaining its integrity and thus reducing dispersion.

Another significant characteristic of some tamponade agents is their ability to spread over the entire surface of the eye chamber. The ability of a given substance to cover a surface area can be described by the spreading coefficient (28, 29). The spreading coefficient (S) for non-aqueous liquids, such as hydrocarbons, fluorocarbons, and their analogues, is obtained from the measured values of surface and interfacial tension and calculated using the following equation:

$$S = \delta_{water} - \delta_{oil} - \delta_{water/oil}$$

where δ_{water} is the surface tension of water (72 mN/m), δ_{oil} is the surface tension of oil, and $\delta_{water/oil}$ is the interface tension between water and oil. The calculated spreading coefficients of some tamponade agents are listed in Table X. The higher the spreading coefficient, the wider oil spreads on the surface of the water. As demonstrated in Table X, Densiron®68 (HWS 1.06) has a higher spreading coefficient (S = 12.05) than those of its starting substances, silicone oil 5000 and F6H8 (S = 8.19 and 7.08, respectively). In contrast, HWS 1.01 and perfluorodecalin have a negative spreading coefficient. Thus, HWS 1.06 will spread better than HWS 1.01 and perfluorodecalin over the surface of the water. The spreading coefficients of HWSs are surprising. It was expected that HWSs would have spreading coefficients similar to those of the two initial substances, F6H8 and silicone oil. Instead, the spreading coefficients of HWS 1.01 and HOS 1.06 differed from those of the initial substances (Tab. X). It must be assumed that these unexpected spreading coefficients are related to the nature of HWSs, i.e., are due to the fact that the micelles are formed by the self-assembly of F6H8 in silicone oil (Figs. 2 and 3). The formation of self-assembled aggregates (micelles) in silicone oil largely depends on the F6H8 concentration.

It is well known that the effectiveness of a tamponade agent depends on its specific gravity and viscosity, as well as its surface and interface tension (27). It has also been reported that the ability of a tamponade agent to close breaks depends on the interaction of three phases (27): vitreous fluid, tamponade agent, and retina. In other words, the effectiveness of an internal tamponade agent depends not only on its interaction with water, but also on its ability to contact or wet the retina (30, 31).

Wetterqvist et al have explored the bubble shape and contact behavior of HWSs (provided by Fluoron GmbH, Neu-Ulm, Germany) using a model eye chamber. It was found that HWS 1.06 (Densiron®68) provides much better contact to the inferior chamber wall than HWS 1.01 and HWS 1.03 (27).

Therefore, it must be assumed that Densiron®68 forms a flat-bottomed bubble on the retinal surface, thus maximizing liquid drainage, and provides good contact in the vitreous cavity due to the favorable difference between the specific gravity of Densiron®68 and that of the vitreous fluid and its excellent spreading coefficient.

As a basic study of HWSs, Meinert and Roy examined the solubility of the semifluorinated alkanes F6H6 and F6H8 in silicone oils. The solubility of semifluorinated alkanes (RFRHs) in silicone oil increases with an increasing RH proportion in the RFRH, decreasing silicone oil viscosity, and increasing temperature (4, 18).

In the present study, the behavior of the binary systems of HWSs was studied in relation to temperature and time. In an observation period of more than 4 years at temperatures higher than +2 °C, all HWSs were homogeneous, optical transparent solutions. No alteration took place in any sample (HWS 1.01, HWS 1.03, and HWS 1.06). At temperatures below +2 °C, HWS 1.06 was temporary opacified, but became homogenous and transparent at room temperature. These HWSs were also thermodynamically stable systems under sterile conditions (121 °C and 165 °C).

As already mentioned, according to the chemical analyses (GC-MS and OH endgroup analysis), there are no time-dependent changes in the chemistry of the components of HWSs. Moreover, all HWSs stored for more than 4 years were nontoxic and biocompatible in the in vitro cytotoxicity assay with human adult retinal pigment epithelial cells (ARPE-19) and mouse cells (L929). These test results indicate that HWSs are chemically stable irrespective of the storage time.

Finally, the solubilities of PFD and PFO were examined. According to Table IX, PFCLs are slightly more soluble in Densiron®68 than in silicone oil 1000 when an external force is applied. Up to the maximal solubility, these solutions are optically transparent and homogenous. The further addition of PFCLs causes phase separation in both cases. According to in vitro diffusion experiments, small PFCL bubbles remain unchanged under the silicone oils up to a period of several months, while they are absorbed in Densiron®68 within 24 hours. But it should be mentioned that, when replaced by a long-term endotamponade, PFCLs must be removed from the vitreous cavity as completely as possible.

The results can be summarized as follows:

• F6H8, as an amphiphile, forms micelles in silicone oils.
• Due to its specific gravity (1.06 g/cm^3), which is slightly higher than vitreous liquid, Densiron®68 is suitable as a vitreous tamponade for the inferior retina in the treatment of complicated retina detachment.
• Based on in vitro experiments, the low viscosity of Densiron®68 (1480 mPas) allows safe and easy handling during surgery.

• Densiron®68, with its favorable spreading behavior, allows good wetting of the retina.
• Densiron®68 is not only thermodynamically stable over a wide temperature range, but is also a long-term stable and homogeneous liquid.
• Densiron®68 is non-toxic and biocompatible according to the cytotoxicity test with mouse cells (L929) and human adult retinal pigment epithelium cells (ARPE-19).

Therefore, Densiron®68, with a specific gravity of 1.06 g/cm^3 and a viscosity of 1480 mPas, has promising potential as a long-term endotamponade agent.

All authors have a commercial interest in Densiron®68.

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REFERENCES

1. Chang S, Ozmert E, Zimmerman NJ. Intraoperative perfluorocarbon liquids in the management of proliferative retinopathy. Am J Ophthalmol 1988; 106: 688-74.
2. McCuen BW 2nd, De Juan E, Machemer R. Silicone oil in vitreoretinal surgery. Part I: Surgical techniques. Retina 1985; 5: 189-97.
3. McCuen BW 2nd, De Juan E, Landers MB, Machemer R. Silicone oil in vitreoretinal surgery. Part II: Results and complications. Retina 1985; 5: 198-205.
4. Meinert H, Roy T. Semifluorinated alkanes: a new class of compounds with outstanding properties for utilization in the ophthalmic field. Eur J Ophthalmol 2000; 10: 189-97.
5. Meinert H, Knoblich AR. The use of semifluorinated alkanes in blood-substitutes. Biomat Art Cells Immob Biotech 1993; 21: 583-95.
6. Meinert H. Semifluorierte Alkane und ihre Verwendung. Eur Pat EP 0859751: 2000.
7. Meinert H. Semifluorinated compounds in ophthalmology. J Vitreoretina 1992; 1: 31-5.
8. Zeana D, Becker J, Kuckelkorn R, Kirchhof B. Perfluorohexyloctane as a long-term vitreous tamponade in the experimental animal. Int Ophthalmol 1999; 23: 17-24.
9. Kirchhof B, Wong D, Van Meurs J, et al. Use of perfluorohexyloctane as a long-term internal tamponade agent in complicated retinal detachment surgery. Am J Ophthalmol 2002; 133: 95-101.
10. Hiscott P, Magee RM, Colthurst M, Lois N, Wong D. Clinicopathological correlation of epiretinal membranes and posterior lens opacification following perfluorohexyloctane tamponade. Br J Ophthalmol 2001; 85: 179-83.
11. Herbert E, Stappler T, Wetterqvist C, Williams R, Wong D. Tamponade properties of double-filling with perfluorohexyloctane and silicone oil in a model eye chamber. Graefes Arch Clin Exp Ophthalmol 2004; 242: 250-4.
12. Peyman GA, Conway MD, Soike KF, Clarke LC. Long term vitreous replacements in primates with intravitreal Vitreon or Vitreon plus silicone. Ophthalmic Surg 1991; 2: 657-64.
13. Peyman GA, Schulman JA, Sullivan B. Perfluorocarbon liquids in ophthalmology. Surv Ophthalmol 1995; 9: 375-95.
14. Bottoni F, Arpa P, Vinciguerra P, Zenoni S, De Mofetta V. Combined silicone and fluorosilicone oil tamponade (double filling) in the management of complicated retinal detachment. Ophthalmologica 1992; 204: 77-81.
15. Rizzo S, Vento A, Di Bartolo E, Genovesi-Ebert F. Distacco

di retina traumatico: doppio tamponamento F6H8-PDMS. I Traumi Oculari, Atti del Simposio S.I.E.T.O. Givre, Firenze: 23 Maggio; 2003: 611-9.

16. De Mofetta V, Bottoni F, Arpa P, Vinciguerra P, Zenoni S. The effect of simultaneous internal tamponade on fluid compartmentalization and its relationship to cell proliferation. Retina 1992; 12: 40-5.

17. Langefeld S, Kirchhof B, Meinert H, Roy T, Aretz A, Schrage NF. A new way of removing silicone oil from the surface of silicone intraocular lenses. Graefes Arch Clin Exp Ophthalmol 1999; 237: 201-6.

18. Meinert H. Semifluorierte Alkane und ihre Verwendung. Deutsches Patent DE 195 36 504 (PCT/EP96/03542): 1997.

19. Hoerauf H, Laqua H. Severe emulsification after combined use of a partially fluorinated alkane and silicone oil. Graefes Arch Clin Exp Ophthalmol 2002; 240: 131-6.

20. Hoerauf H, Menz DH, Dresp J, Laqua H. O_{44}-Ein Lösungsmittel für Silikonöladhäsionen auf Intraokularlinsen. Klin Monatsbl Augenheilkd 1999; 214: 71-6.

21. Hoerauf H, Kobuch K, Dresp J, Menz DH. Combined use of partially fluorinated alkanes, perfluorocarbon liquids and silicone oil: an experimental study. Graefes Arch Clin Exp Ophthalmol 2001; 239: 373-81.

22. Wolf S, Schön V, Meier P, Wiedemann P. Silicone oil-RMN3 mixture ("heavy silicone oil") as internal tamponade for complicated retinal detachment. Retina 2003; 23: 335-42.

23. Wong D, Van Meurs JC, Stappler T, et al. A pilot study on the use of a perfluorohexyloctane/silicone oil solution as a heavier-than-water internal tamponade agent. Br J Ophthalmol 2005; 89: 662-5.

24. Meinert H. Semi-fluorinated alkanes and their use. US Pat US 6,262,126: 2001.

25. Blinks BP, Fletcher PDI, Thompson RL. Surfactants properties of semifluorinated alkanes in hydrocarbon and fluorocarbon solvents. Ber Bunsenges Phys Chem 1996; 100: 232-6.

26. Wong D, Lois N. Perfluorocarbons and semifluorinated alkanes. Semin Ophthalmol 2000; 15: 25-35.

27. Wetterqvist C, Wong D, Williams R, Stappler T, Herbert E, Freeburn S. Tamponade efficiency of perfluorohexyloctane and silicone oil solutions in a model eye chamber. Br J Ophthalmol 2004; 88: 692-6.

28. Weers JG, Dellamary LA, TararaTE, Trevino LA, Ranney HM. Fluorochemical solutions for the delivery of lipophilic pharmaceutical agents. US Pat US 5,874,481: 1999.

29. Faithfull NS, Weers JG. Treatment and diagnosis of respiratory disorders using fluorocarbon liquids. US Pat US 5,853,003: 1998.

30. Fawcett IM, Williams RL, Wong D. Contact angles of substances used for internal tamponade in retinal detachment surgery. Graefes Arch Clin Exp Ophthalmol 1994; 232: 438-44.

31. Williams R, Wong D. The influence of explants on the physical efficiency of tamponade agents. Graefes Arch Clin Exp Ophthalmol 1999; 237: 870-4.