Customer Service Report CS0202

Technical Documentation OmniflexPlus

Edition 8

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1 Description

Datwyler's OmniflexPlus coated closures are bromobutyl vial stoppers that are covered with a fluorinated polymer coating. The coating is applied via a proprietary spray-coating process.

The bromobutyl rubber substrate used is FM259/0, colour dark gray, a rubber formulation with silicate filler and an unconventional curing system.

The fluorinated polymer coating provides a chemical inertness comparable with that of PTFE (Poly-TetraFluoroEthylene or Teflon®), ensuring a high degree of compatibility with a broad range of pharmaceutical products.

Since products are totally coated, the trimming edge is covered as well and additional siliconization during the final treatment becomes redundant. The latter is possible thanks to the low friction coefficient of the coated closures and the absence of sticking tendency, also after sterilization. These features and the fact that OmniflexPlus products are uniquely manufactured in Datwyler's FirstLine™ production facility[1], result in a clearly reduced visible and subvisible particle burden.

Datwyler's FirstLine production facility avails of multiple manufacturing lines for the production of OmniflexPlus closures. For contingency reasons these manufacturing lines are available in two segregated areas of the FirstLine building that are remote from each other.

FM259/0, the substrate rubber for OmniflexPlus products is a modern formulated bromobutyl with high chemical purity. Combined with the barrier coating, an extremely low extractables and leachables profile is guaranteed.

OmniflexPlus products are not made with natural rubber latex, dry natural rubber latex or any of its derivatives. They are not made using ingredients of animal origin and do not generate known suspicious chemicals like nitrosamines and 2-mercaptobenzothiazole (MCBT). OmniflexPlus products are unrelated to BSE/TSE risks. Obviously, these products comply with all major pharmacopeia for pharmaceutical rubber.

The flexibility of the OmniflexPlus coating maintains an uncompromised seal integrity of the vial – closure combination. The coring and reseal characteristics of OmniflexPlus coated closures are excellent.

OmniflexPlus coated closures can be gamma sterilized with limited influence on chemical and physical properties.

[1] FirstLine is a brand-new state-of-the-art production unit within the Datwyler Pharma Packaging group where the strictest product specifications for microbiological and particulate cleanliness can be guaranteed.

The OmniflexPlus coating is applied to a selected range of vial stoppers[2], including serum[3] and lyophilization[4] designs.

The thickness of the coating layer varies with the exact location on the stopper and is product design dependent. It typically is between 10 and 20 µm. Coating thickness is reproducible from batch to batch.

[2] For pre-filled syringe plungers, Datwyler offers a different product named Omniflex Coated Plungers ('OCP', 'OmniflexCP'). Omniflex Coated Plungers are not in the scope of this document.

[3] For a selective list of product drawings, see attachment.

[4] For lyophilization stoppers, Datwyler at present puts forward its most recent generation of coated stoppers that are marketed under the name Omniflex3G.

2 Typical compound ingredients

2.1 Natural Rubber Latex

In line with current market expectations, OmniflexPlus products are not made with natural rubber latex, dry natural rubber latex or any of its derivatives.

2.2 Nitrosamines

Nitrosamines are residues of specific curing systems. These residues are in a number of cases known to be carcinogenic.

OmniflexPlus products are formulated without making use of ingredients that potentially give rise to the formation of nitrosamines.

2.3 MCBT

2-mercaptobenzothiazole (2-MBT, 2-MCBT, MCBT) is a rubber chemical that is associated with a health risk.

OmniflexPlus products do not contain 2-mercaptobenzothiazole (MCBT) or any of its derivatives in their composition.

2.4 BSE/TSE

Rubber compounds may contain components that are of animal origin. Most frequently it concerns fatty acids, fatty acid salts or esters that are either present as active components or as additives to active components.

OmniflexPlus does not use components of animal origin. OmniflexPlus products are in full compliance with the European Pharmacopoeia 5.2.8., "Minimizing the risk of transmitting Animal Spongiform Encephalopathy Agents via medicinal products ".

(TSE = Transmissible Spongiform Encephalopathy; BSE = Bovine Spongiform Encephalopathy)

2.5 Heavy Metals

Both US and European legislation impose measures in order to prevent or reduce the impact of packaging and packaging waste on the environment.

OmniflexPlus products fulfill the European Community Guideline 94/62/EC for heavy metals in packaging materials.

This directive states that packaging components shall not contain more than 100 ppm of Lead (Pb), Cadmium (Cd), Mercury (Hg) and Chromium (VI) (Cr).

EC Guideline 94/62/EC imposes the same requirements as the US CONEG regulation ('Toxics in Packaging Clearinghouse (TPCH)' as established by the Coalition of North-eastern Governors (CONEG) in 1992). OmniflexPlus products thus also fulfill the CONEG requirements.

2.6 GMO (Genetically Modified Organisms)

OmniflexPlus products are not made with ingredients that are derived from Genetically Modified Organisms (GMO).

2.7 Bisphenol A (BPA)

Bisphenol A (BPA) and BPA related substances may be present in certain plastic materials. They are associated with health hazards.

Datwyler confirms that OmniflexPlus products do not use Bisphenol A or the following BPA related substances in their composition:

- Polycarbonate
- Polyether sulfone
- Polycarbonate/siloxane co-polymer
- Biostable polyurethanes
- Epoxy resin
- Bisphenol A diglycidylether methacrylate (BIS-GMA)
- Bisphenol A diglycidylether (BADGE)
- Bisphenol A dimethacrylate (BIS-DMA)
- Ethoxylated bisphenol A diacrylates.

3 Shelf life

The shelf life of OmniflexPlus products stored in the original packaging under the ambient storage conditions as described in the ISO 2230, “Rubber Products – Guideline for storage”, is 2 years after packing date.

Hereafter, based on the indications given in the ISO 2230, an additional shelf life of 5 years can be considered.

Compatibility with the drug must be ascertained by the user.

4 Physical properties

4.1 Identification properties

The following tests are used to identify the rubber formulation following typical rubber technology standards.

The physical properties shown in the table below are taken for the rubber substrate compound FM259/0 without the additional fluoropolymer coating.

For technical reasons, these properties are not determined on OmniflexPlus (=FM259/0 in its coated version).

Physical properties – FM259/0

Hardness	°Shore A	ISO 7619-1 (1 second indentation)	39 ± 5
Density	g/cm³	ISO 2781	1.191 ± 0.025

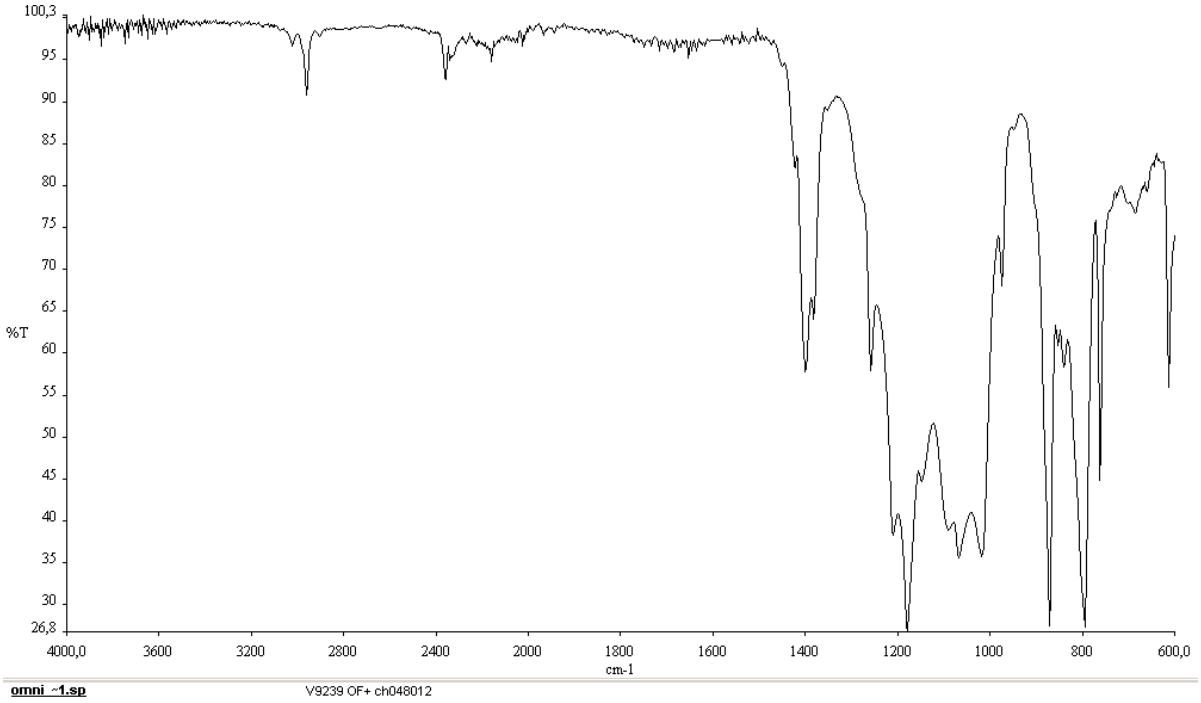
Ash	%	Internal Method(s): Calcination 4h@700°C	31.0 ± 2.0
Compression Set	%	ISO 815-1	max. 18
Tensile Strength	N/mm²	ISO 37	min. 6

4.2 ATR-FTIR scans

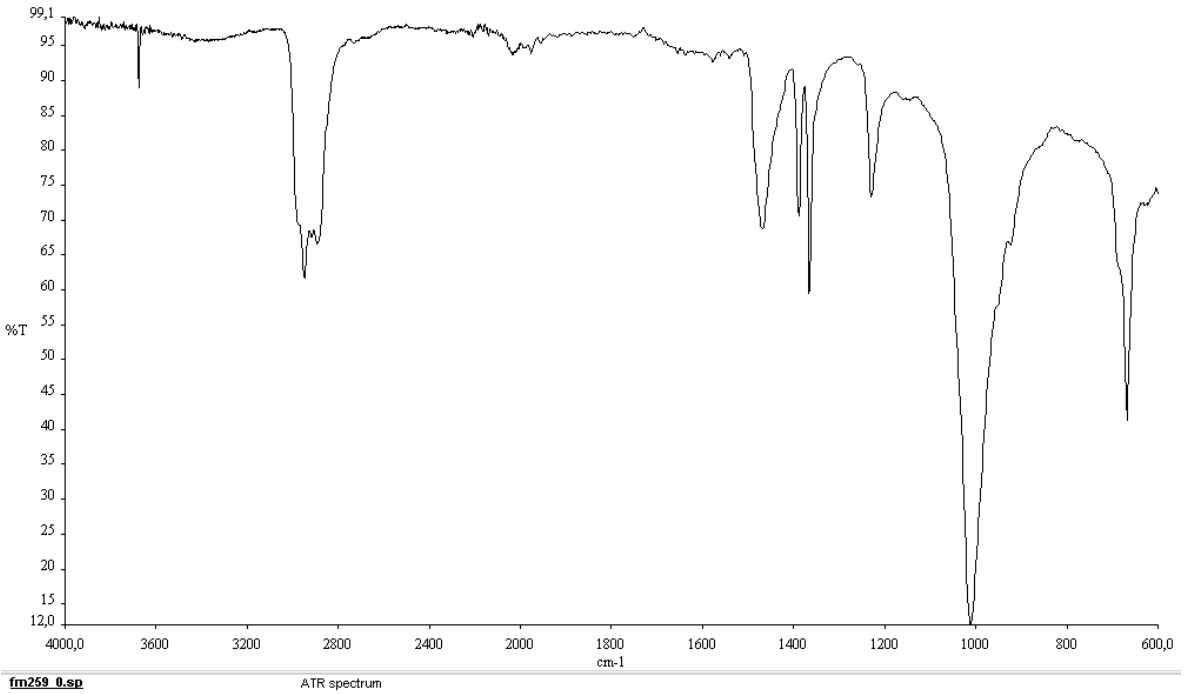
To distinguish between coated and non-coated rubber, one can make use of an ATR-FTIR (Attenuated Total Reflection –Fourier Transform InfraRed) scan from the product surface and that of a cut product. The former reveals the spectrum of the coating, while the latter gives the spectrum of the substrate rubber.

The next figures show a reference spectrum of both surfaces.

FTIR surface scan of OmniflexPlus (coated FM259/0)



FTIR surface scan of FM259/0 (uncoated rubber – cut edge)


99,1
95
90
85
80
75
70
65
60
55
%T
50
45
40
35
30
25
20
15
12,0
4000,0
3600
3200
2800
2400
2000
1800
1600
1400
1200
1000
800
600,0
cm-1
fm259_0.sp
ATR spectrum

4.3 Permeability

Oxygen and water vapour transmission rates are measured as per ASTM D-3985 (oxygen) and ASTM F-1249 (water vapour). They measure the permeability of a material to oxygen viz. water vapour.

To be able to obtain permeability data on rubber compounds, a special mould, yielding a thin rubber slab, is used to prepare test pieces.

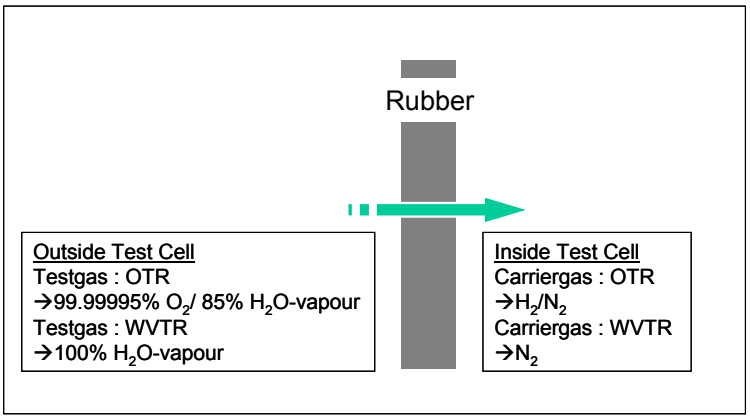
These rubber slabs, with a certain thickness (±1.3mm), are preconditioned at 23°C and 50% RH prior to the actual measurement.

For the test itself, a rubber slab in FM259/0 without coating that matches 50 cm² is cut and clamped in a double chamber test cell, acting as a barrier between both chambers.

The transmission rate is recorded once the system is at steady-state.

Permeability on OmniflexPlus is not determined for technical reasons. The coating itself does not act as a barrier to O_2 and H_2O and hence the below given properties for FM259/0 are to be considered relevant for OmniflexPlus as well.

Schematic principle of a transmission rate measurement



4.3.1 Water Vapour Transmission Rate (WVTR)

Tested slabs: FM259/0 slabs with thickness 1.33mm

Equipment: MOCON, Permatran-W 3/31 MG-module

Conditions: 38 °C ; 100% relative humidity ; 100% flow N_2

	WVTR in g/m².24h, 100% RH / 38°C
FM259/0	0.03

4.3.2 Oxygen Transmission Rate (OTR)

Tested slabs: FM259/0 slabs with thickness 1.27 mm

Equipment: MOCON, Oxtran 2/20 ML-module

Conditions: 38 °C ; 85 % relative humidity ; 100 % O_2

	OTR in cc/m².24h, 85% RH / 38°C, 100 % O_2
FM259/0	77

5 Chemical properties

5.1 Pharmacopeial data

5.1.1 Pharm.Eur.3.2.9. / USP <381> data

A revised version of USP <381>, has come into force on May 1, 2009. Sample preparation, test description and the 2-tier acceptance criteria were largely harmonized with Pharm. Eur. 3.2.9.

Furthermore, USP <381> stipulates that the physicochemical testing of coated products has to be performed on both the rubber substrate and on the coated form.

The tables below summarize USP <381> and Pharm. Eur. 3.2.9. chemical testing on the FM259/0 substrate rubber and on OmniflexPlus respectively.

Pharm. Eur. 3.2.9 / USP <381> data, chemical part – FM259/0 substrate

Characteristic		Amount tested	Units	Limit	Typical Value	
Appearance of solution	Turbidity	Sol. S	NTU	Type I: 6.0 *(*)* Type II: 18.0 *(*)*		0.2
	Colour	Sol. S		See test procedure		pass
Acidity or alkalinity		Sol. S (20 ml)	ml 0.01M HCl ml 0.01M NaOH	0.8 0.3	Blank 0.03 EP USP	 0.03 0.03 0.00
Absorbance		Sol. S	A_{max}220-360 nm	Type I: 0.2 Type II: 4.0		0.01
Reducing substances		Sol. S (20 ml)	ml 0.002M $KMnO_4$	Type I: 3.0 Type II: 7.0		0.1
Extractable heavy metals		Sol. S	ppm Pb^{2+}	2	EP USP	<2 <2
Extractable zinc		Sol. S	ppm Zn^{2+}	5.0		<0.01

Ammonium	Sol. S	ppm NH_4^+	2		<2
Residue on evaporation (only for EP)	Sol. S (50 ml)	mg	Type I: 2.0 Type II: 4.0		0.0
Volatile sulphides	20 cm²	mg S^{2-}	0.02		<0.02

* *By definition corresponding with reference suspensions II and III respectively.*

(Data recorded for batch 30091037)

Pharm. Eur. 3.2.9 / USP <381> data, chemical part – OmniflexPlus

Characteristic		Amount tested	Units	Limit	Typical Value	
Appearance of solution	Turbidity	Sol. S	NTU	Type I: 6.0 *(*)* Type II: 18.0 *(*)*		0.1
	Colour	Sol. S		See test procedure		pass
Acidity or alkalinity		Sol. S (20 ml)	ml 0.01M HCl ml 0.01M NaOH	0.8 0.3	Blank 0.03	0.03
					EP	0.03
					USP	0.00
Absorbance		Sol. S	A_{max}220-360 nm	Type I: 0.2 Type II: 4.0		0.01
Reducing substances		Sol. S (20 ml)	ml 0.002M $KMnO_4$	Type I: 3.0 Type II: 7.0		0.1
Extractable heavy metals		Sol. S	ppm Pb^{2+}	2	EP	<2
					USP	<2
Extractable zinc		Sol. S	ppm Zn^{2+}	5.0		<0.01
Ammonium		Sol. S	ppm NH_4^+	2		<2
Residue on evaporation (only for EP)		Sol. S (50 ml)	mg	Type I: 2.0 Type II: 4.0		0.0
Volatile sulphides		20 cm²	mg S^{2-}	0.02		<0.02

* *By definition corresponding with reference suspensions II and III respectively.*

(Data recorded for batch 30098904)

For OmniflexPlus products Datwyler guarantees a shelf life period of 2 years after the packing date.

Compliance of FM259/0 (substrate for OmniflexPlus) and of OmniflexPlus itself with the chemical requirements of Pharm.Eur. 3.2.9 and USP <381> after 2 years shelf life is demonstrated in the tables below.

Pharm. Eur. 3.2.9 / USP <381> data, chemical part – FM259/0 substrate

Characteristic		Amount tested	Units	Limit	Typical Value	
Appearance of solution	Turbidity	Sol. S	NTU	Type I: 6.0 *(*)* Type II: 18.0 *(*)*		0.4
	Colour	Sol. S		See test procedure		pass
Acidity or alkalinity		Sol. S (20 ml)	ml 0.01M HCl ml 0.01M NaOH	0.8 0.3	Blank 0.03 EP USP	0.03 0.06 0.03
Absorbance		Sol. S	A_{max}220-360 nm	Type I: 0.2 Type II: 4.0		0.01
Reducing substances		Sol. S (20 ml)	ml 0.002M $KMnO_4$	Type I: 3.0 Type II: 7.0		0.4
Extractable heavy metals		Sol. S	ppm Pb^{2+}	2	EP USP	<2 <2
Extractable zinc		Sol. S	ppm Zn^{2+}	5.0		<0.01
Ammonium		Sol. S	ppm NH_4^+	2		<2
Residue on evaporation (only for EP)		Sol. S (50 ml)	mg	Type I: 2.0 Type II: 4.0		0.0
Volatile sulphides		20 cm²	mg S^{2-}	0.02		<0.02

* *By definition corresponding with reference suspensions II and III respectively.*

(Data recorded for batch 30030181 after 2 years shelf life)

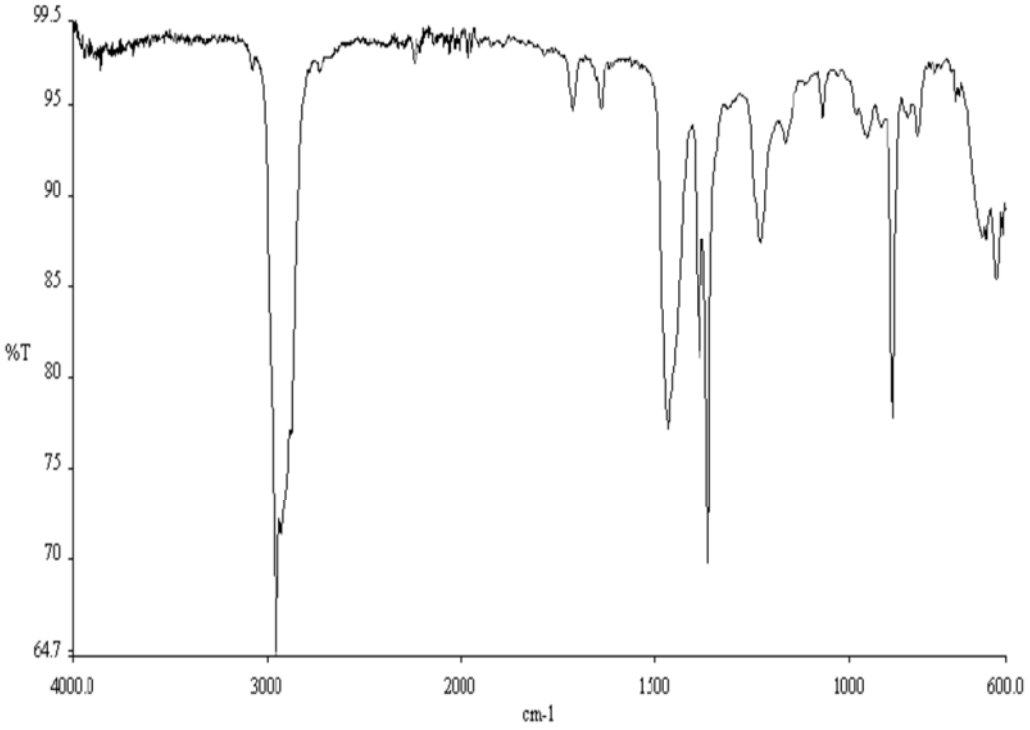
Pharm. Eur. 3.2.9 / USP <381> data, chemical part – OmniflexPlus

Characteristic		Amount tested	Units	Limit	Typical Value	
Appearance of solution	Turbidity	Sol. S	NTU	Type I: 6.0 *(*)* Type II: 18.0 *(*)*		0.2
	Colour	Sol. S		See test procedure		pass
Acidity or alkalinity		Sol. S (20 ml)	ml 0.01M HCl ml 0.01M NaOH	0.8 0.3	Blank 0.06	0.06
					EP	0.06
					USP	0.00
Absorbance		Sol. S	A_{max}220-360 nm	Type I: 0.2 Type II: 4.0		0.01
Reducing substances		Sol. S (20 ml)	ml 0.002M $KMnO_4$	Type I: 3.0 Type II: 7.0		0.1
Extractable heavy metals		Sol. S	ppm Pb^{2+}	2	EP	<2
					USP	<2
Extractable zinc		Sol. S	ppm Zn^{2+}	5.0		<0.01
Ammonium		Sol. S	ppm NH_4^+	2		<2
Residue on evaporation (only for EP)		Sol. S (50 ml)	mg	Type I: 2.0 Type II: 4.0		0.1
Volatile sulphides		20 cm²	mg S^{2-}	0.02		<0.02

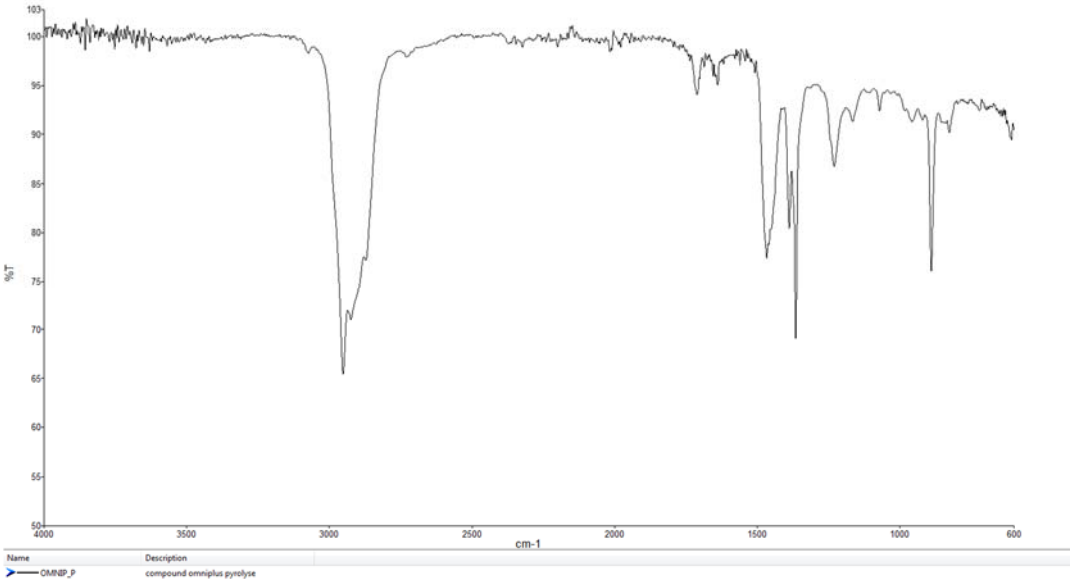
* *By definition corresponding with reference suspensions II and III respectively.*

(Data recorded for batch 30038105 after 2 years shelf life)

Typical IR-spectrum (4000-625cm^{-1}) of a pyrolysate (acc. to Pharm. Eur. 3.2.9.)
- FM259/0 substrate



Typical IR-spectrum (4000-625cm^{-1}) of a pyrolysate (acc. to Pharm. Eur. 3.2.9.)
- OmniflexPlus



Typical UV-spectrum of an aqueous extract (acc. to Pharm. Eur. 3.2.9.) - FM259/0 substrate

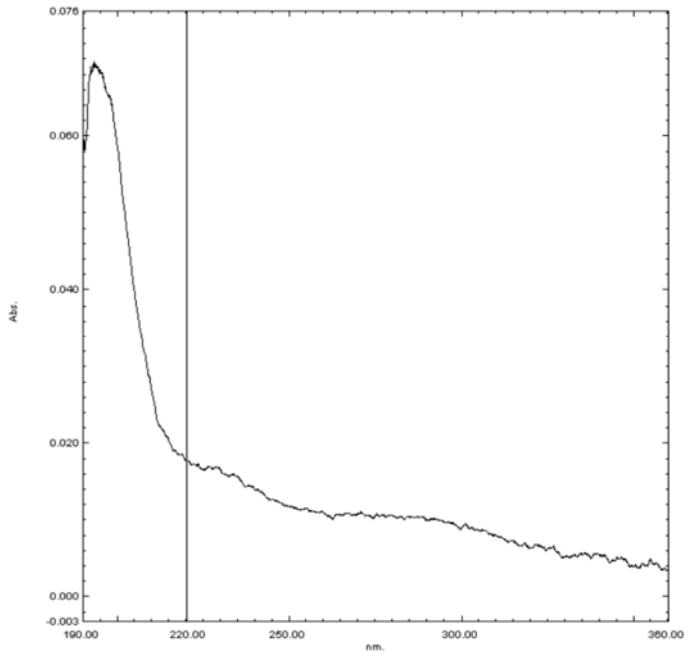
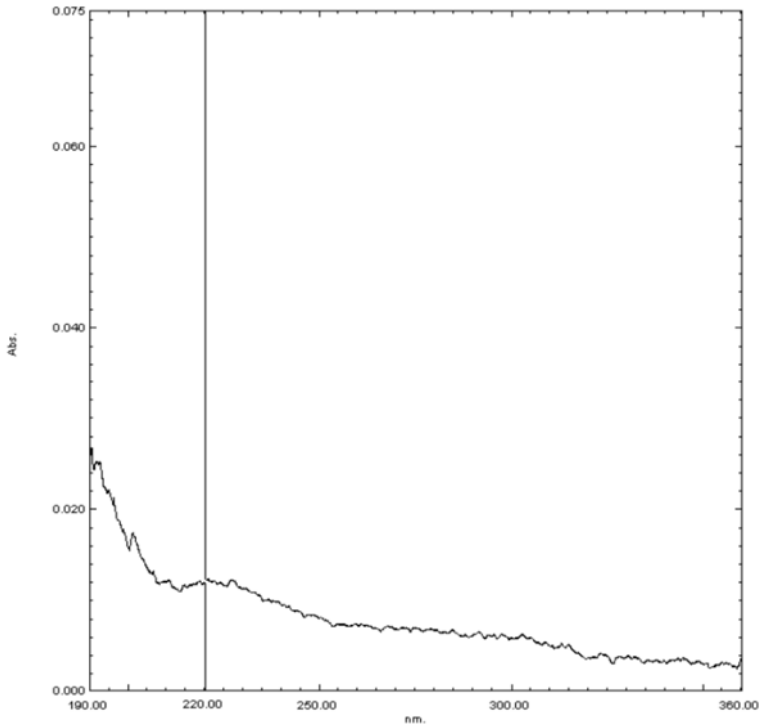


Figure : Typical UV-spectrum of an aqueous extract (acc. to the Pharm. Eur. 3.2.9.) - OmniflexPlus



5.1.2 Japanese Pharmacopeia 7.03

Results for OmniflexPlus, tested according to the physicochemical part of the Japanese Pharmacopeia, chapter 7.03, “Test for Rubber Closures for Aqueous Infusions”, valid version, are given in the table below.

30 g of rubber sample is autoclaved in 300 g distilled water for 60 min. at 121°C.

Due to the peculiar definition of the sample preparation (by rubber mass and not by rubber surface), results are closure design dependent (surface/volume ratio dependency). Results in the table are given for a 13 mm serum stopper (V9239) with a volume of 0.10cm³, a weight of 0.46 g and a total surface of 4.6 cm² . Larger stopper designs expectedly will show results that are at least as good, since the surface/volume ratio of such stoppers is smaller (less rubber surface in contact with the extraction medium).

For the design given, OmniflexPlus complies with the extractable substances part of the Pharm. Jap. 7.03.

JP 7.03 – ‘Extractable Substances’ – OmniflexPlus (V9239)

CRITERIUM	AMOUNT TESTED	UNITS	LIMITS	RESULTS
Appearance (430-650 nm)	10 mm cuvet	%T at 430 nm %T at 650 nm	99.0 % T 99.0 % T	99.6 100.0
Foam test	5 ml	-	foam disap. < 3 min.	pass
pH	20 ml	pH units	difference with blank: max. 1.0 (*)	-0.1
Reducing substances	100 ml	ml 0.002 M $KMnO_4$	2.0	0.4
Evaporation residue	100 ml	mg	2.0	0.0
UV absorb. (220-350 nm)	10 mm cuvet	absorbance	0.2	0.02
Zinc	10 ml	ppm Zn^{2+}	1 ppm	< 0.01

(*) “-“ means more acidic than blank; “+” means more alkaline than blank

(Data recorded for batch 30024100)

The results of other parts of Pharm. Jap. 7.03 carried out on the same batch were as follows :

JP 7.03 – Cadmium, Lead, Acute systemic toxicity, Pyrogen test, Hemolysis test – OmniflexPlus (V9239)

Test	Limit	Result
Cadmium	5 ppm	< 0.006 ppm
Lead	5 ppm	0.8 ppm
Acute systemic toxicity	Limit test	Pass
Pyrogen test	Limit test	Pass
Hemolysis test	Limit test	Pass

(Data recorded for batch 30024100)

5.2 ISO 8871-1

The requirements of the ISO 8871-1, “Elastomeric parts for parenterals and for devices for pharmaceutical use – Part 1: Extractables in aqueous autoclavates”, are identical to those set down in Pharm. Eur. 3.2.9.

Results for Pharm. Eur. 3.2.9. are given in an earlier part of this section.

5.3 Extractable information

A detailed extractables report on OmniflexPlus involving solvents of various polarities, is available on request.

Because of their confidential nature, the results of this extractables study are not given in full detail in this report. They will be available only after conclusion of specific agreements. Please contact your Datwyler sales representative.

6 Functional properties

6.1 Penetrability

Test according to Pharm. Eur. 3.2.9 / USP <381>[5]

Principle
The force needed to completely pierce an elastomeric closure is measured using a universal force tester.

Sample preparation:
10 closures (20 mm serum, type V9048, compound FM259/0, OmniflexPlus coated, batch 748009) are pretreated as described in Pharm.Eur. 3.2.9 / USP <381>:

- The closures are placed in a glass container, covered with distilled water and boiled for 5 minutes. They are rinsed 5 times with cold distilled water.
- The washed and rinsed closures are placed in a conical flask and covered with distilled water (2 ml per cm² of rubber surface area).
- The closures are autoclaved at 121°C for 30 minutes.
- The water is decanted and the closures are allowed to dry at room temperature.

Procedure:

- 10 clean, standard 20 mm vials are filled with water to their nominal volume.
- The filled vials are closed with the rubber closures under test and capped with an aluminium seal.
- The closures are pierced with a 0.8 mm external diameter hypodermic needle and the force required to pierce the closure is recorded by means of a suitable dynamometer.
- The piercing force should not exceed 10 N.

Test results:

OmniflexPlus coated 20 mm serum closures[6] comply with the requirements of Ph. Eur. 3.2.9 and of USP <381> for penetrability (penetration force less than 10 N).

The piercing force observed does not exceed 1.5 N.

6.2 Fragmentation (coring) [7]

6.2.1 Test according to Pharm. Eur. 3.2.9 / USP <381>

Principle

Elastomeric closures for injection vials are pierced with an injection needle. Elastomeric fragments which have been caused by piercing are collected on a filter and counted.

Sample preparation:

[5] For functional testing (penetrability, fragmentation, self-sealing) Pharm. Eur. 3.2.9 and USP <381> since May 1, 2009 follow the same procedure and have the same limit values.

[6] For results on other OmniflexPlus closure designs, please contact your Datwyler Sales Representative.

[7] Both Pharm. Eur. 3.2.9 and USP <381> use the term 'fragmentation'. The term 'coring' however is a synonym that is often used.

36 closures [8] (20 mm serum, type V9048, compound FM259/0, OmniflexPlus coated, batch 748009) are pretreated as described in Pharm.Eur. 3.2.9 / USP <381> (see above for description).

Procedure:

- 3 x 12 clean, standard 20 mm vials are filled with 5 ml of water.
- The vials are closed with the closures to be examined, secured with an aluminium seal and allowed to stand for 16 hours.
- A clean syringe is filled with distilled water .
- A hypodermic needle with external diameter 0.8 mm (21 G) is fitted to the syringe.
- The first closure is pierced, 1 ml of water is injected and 1ml of air is removed.
- This operation is done 4 times for each closure with one and the same needle. The closure is pierced at a different site each time
- A new needle is taken for each subsequent new closure.
- The liquid from 12 vials is filtered over a filter with a pore size of 0.5 µm.
- The fragments on the filter are counted by the unaided eye.
- The total number of fragments per filter (resulting from 12 x 4 = 48 piercings) may not exceed five.
- For each closure sample, 3 filters are prepared.

Test results:

OmniflexPlus coated 20 mm serum closures[9] comply with the requirements of Ph. Eur. 3.2.9 and of USP <381> for coring (less than 5 fragments per 48 piercings).

6.2.2 Test according to proprietary procedures

6.2.2.1 *Multiple piercing with 21G needles*

The same Pharm. Eur. 3.2.9 / USP <381> procedure with 3 x 12 vials as in the previous paragraph was followed with one exception : each vial (stopper) is pierced 25 times, using a new needle every 5 piercings. The number of penetrations per stopper thus is considerably increased from 4 punctures per stopper to 25.

Test results :

Table : coring results after multiple piercing with 21G needles

Sample		**# fragments per 300 piercings or per 12 vials**
FM259/0 V9048 20 mm serum OmniflexPlus Batch 30032050	Filter 1 – Vials 1-12	2
	Filter 2 – Vials 13-24	1

8 Pharm. Eur. 3.2.9 and USP <381> prescribe 12 closures. Testing in this case was carried out in triplicate.

9 For results on other OmniflexPlus closure designs, please contact your Datwyler Sales Representative.

	Filter 3 – Vials 25-36	1

6.2.2.2 *Multiple piercing with 18G needles*

Another test according to a modified Pharm. Eur. 3.2.9 / USP <381> procedure, this time however with 3 x 20 stoppers, with 18G instead of 21G needles and with 5 instead of 4 piercings per stopper, was performed. The number of stoppers in the test, the size of the needle and the number of penetrations thus each were increased in comparison with the pharmacopeial procedure.

The outer viz. inner diameter of an 18G needle are 1.270 mm and 0.838 mm respectively, while for the 21G needle these dimensions are 0.813 mm and 0.495 mm respectively. An 18G needle thus is considerably larger than a 21G.

Further details of the test are as outlined below.

Sample preparation :

60 closures (20 mm serum, type V9048, compound FM259/0, OmniflexPlus coated, batch 905017) are pretreated as follows:

- The closures are placed in a glass container and rinsed 3 times with distilled water at 80 °C.
- The rinsed (wet) stoppers are transferred to a small Ready-for-Sterilization (RfS®) bag, which is then sealed.
- The RfS bag is autoclaved during 30 min. at 121°C.
- After autoclaving, the RfS bag is dried overnight at 70 °C.
- The closures are kept in the sealed RfS bag until use.

Procedure:

- 3 x 20 standard, clean vials are filled with 5 ml of water.
- The vials are closed with the closures to be tested and secured with an aluminium seal.
- A clean syringe is filled with distilled water .
- A hypodermic needle with an external diameter of 1.2 mm (18 G) is fitted to the syringe.
- Each closure is pierced 5 times with the same needle and at the fifth piercing 5 ml of water is injected in the vial. A new needle is used for each subsequent closure.
- The liquid from 20 vials is filtered over a filter with a pore size of 0.5 µm.
- The fragments on the filter are counted with the unaided eye.
- For each rubber closure type, 3 filters are prepared.
- No fragments should be found on the filter (resulting from 20 x 5 =100 piercings) .

Test results:

OmniflexPlus coated 20 mm serum closures in the above described testing and of the described batch generated less than 5 fragments per 20 x 5 = 100 penetrations.

Note:

The same test was carried out on stoppers of the same batch that had not been subjected to the above described sample preparation, but instead were gamma sterilized at 34 kGy.

Also in this case less than 5 fragments per 100 penetrations were noted.

6.3 Self-sealing

6.3.1 Test according to Pharm. Eur. 3.2.9 / USP <381>

Principle

Elastomeric closures for injection vials are pierced several times with an injection needle and examined for

leakage forced by a pressure differential across the closure.

Sample preparation:
30 closures [10] (20 mm serum, type V9048, compound FM259/0, OmniflexPlus coated, batch 748009), are pretreated as described in Pharm.Eur. 3.2.9 / USP <381> (see above for description).

Procedure:

- 30 clean, standard 20 mm vials are filled with water to their nominal volume.
- The filled vials are closed with the rubber closures under test and capped with an aluminium seal.
- Each closure is pierced ten times with a 0.8 mm external diameter (21 G) hypodermic needle, taking care to pierce the closure at a different spot each time.
- A new needle is used for each closure.
- The vials are then immersed upright in a 0.1% aqueous solution of methylene blue, in an enclosure in which the pressure is reduced to 0.73 bar for 10 minutes.
- After restoration of atmospheric pressure, the vials are left immersed for another 30 minutes.
- The vials are rinsed and the contents are checked for any blue discoloration.
- None of the vials may show any discoloration.

Test results:

OmniflexPlus coated 20 mm serum closures[11] comply with the requirements of Ph. Eur. 3.2.9 and of USP <381> for self-sealing. No discoloration was observed.

6.3.2 Tests according to proprietary procedures

6.3.2.1 *Multiple piercing with 21G needles*

The same Pharm. Eur. 3.2.9 / USP <381> procedure with 3 x 10 vials as in the previous paragraph was followed with one exception : each stopper is pierced 25 times, using a new needle every 5 piercings. The number of penetrations per stopper thus is considerably increased from 10 punctures per stopper to 25.

[10] Pharm. Eur. 3.2.9 and USP <381> prescribe 10 closures. Testing in this case was carried out in triplicate.

[11] For results on other OmniflexPlus closure designs, please contact your Datwyler Sales Representative.

Test results :

Table : self-sealing results after multiple piercing with 21G needles

Sample		# vials with colored solution per 10 vials
FM259/0 V9048 20 mm serum OmniflexPlus Batch 30032050	Series 1 - Vials 1-10	0 / 10
	Series 2 - Vials 11-20	0 / 10
	Series 3 - Vials 21-30	0 / 10

6.3.2.2 *Single penetration with 18G needle followed by pressurization*

This proprietary procedure involves penetration of the stopper with an 18G needle, followed by storage of the vials at 50 °C whereby an internal overpressure is created.

Sample preparation:

20 closures (20 mm serum, type V9048, compound FM259/0, OmniflexPlus coated, batch 905017) are pretreated as described sub 6.2.2.2 (rinsing with water at 80 °C, autoclaving 30 mins/121 °C in an RfS bag, followed by overnight drying at 70 °C).

Procedure:

- 20 clean, standard 20 mm vials are filled with a 0.1% aqueous methylene blue solution.
- The filled vials are closed with the rubber closures under test and capped with an aluminium seal.
- Each closure is pierced with an 18 G hypodermic needle, fitted to a syringe, while the vial is in upright position. 5 ml of air is injected and then 5 ml of methylene blue solution is removed.
- The closure is cleaned in the piercing zone to remove all traces of the dye.
- The vials are placed in horizontal position in an oven at 50°C whereby the methylene blue solution must be in contact with the closure.
- After 24 h the vials are checked for dye leakage at the piercing spot.
- No leakage of dye should be observed.

Test results:

OmniflexPlus coated 20 mm serum closures of the above described batch comply with the self-sealing requirement as put forward in this test. No leakage of dye was observed.

Note:

The same test was carried out on stoppers of the same batch that had not been subjected to the above described sample preparation, but instead were gamma sterilized at 34 kGy.

Also here no leakage of dye was observed.

6.4 Closure/vial seal integrity

At the time the testing described in this section was carried out, there was no pharmacopeia or international standard that offered an uncomplicated method for testing of closure/vial seal integrity[12].

Therefore this type of testing was carried out in according to three different proprietary procedures that were obtained from customers.

6.4.1 Proprietary procedure #1 ('French Pharmacopeia test')

In this test the vial-closure combination is tested for the penetration of methylene blue under the influence of a pressure differential. Penetration of the dye should not occur.

Sample preparation:
15 closures (20 mm serum, type V9048, compound FM259/0, OmniflexPlus coated, batch 748009) are pretreated as described sub 6.2.2.2 (rinsing with water at 80 °C, autoclaving 30 mins/121 °C in an RfS bag, followed by overnight drying at 70 °C).

Procedure[13]:
- 15 vials are half filled with distilled water.
- The filled vials are closed with the rubber closures under test and capped with an aluminium seal.
- The vials are autoclaved during 30 min. at 121°C and allowed to cool down to room temperature.
- The vials are then placed in inverted position in a methylene blue solution in a container in which the pressure is reduced to 0.2 bar.
- After one hour the pressure in the container is quickly returned to atmospheric pressure.
- 30 minutes later the vials are checked visually for any blue coloration of the contents.
- There should be no vials with a blue discoloration of the contents.

Test results:

OmniflexPlus coated 20 mm serum closures of the above described batch complied with the requirement of this test. No discoloration was observed in any case.

Note:

The same test was carried out on stoppers of the same batch that had not been subjected to the above described sample preparation, but instead were gamma sterilized at 34 kGy.

Also here no leakage of dye was observed.

[12] At present such method is offered in ISO 8871-5.

[13] The procedure as described was the same as in the French Pharmacopeia at the time the testing was conducted. It shows analogy with the procedure currently included in ISO 8871-5, however is still different on a number of points. ISO 8871-5 works with a sample size of 10 closures. It foresees filling of the vials to nominal volume and no autoclaving after filling, stoppering and capping. Pressure reduction in ISO8871-5 is with 27 kPa and not to 0.2 bar (absolute).

6.4.2 Proprietary procedure #2 ('phenol-saline leak test')

In this test the vial-closure combination is tested for leakage of a phenol –sodium chloride solution under the influence of a pressure differential. The weight loss of the vials should not exceed 1 mg.

Phenol is added to the saline solution with the intention to act as a surfactant that facilitates ingression of the solution in the closure/vial interface.

Sample preparation:
10 closures (20 mm serum, type V9048, compound FM259/0, OmniflexPlus coated, batch 748009) are pretreated as described sub 6.2.2.2 (rinsing with water at 80 °C, autoclaving 30 mins/121 °C in an RfS bag, followed by overnight drying at 70 °C).

Procedure:
- A phenol – saline solution is prepared by dissolving 9.0 g NaCl and 4.0 g phenol in 1 liter of distilled water.
- 10 vials are labeled with the sample reference and numbered .
- The vials are filled with 6.0 ml of the phenol – saline solution and the neck of each vial is carefully wiped to remove any spilled liquid.
- The filled vials are closed with the rubber closures under test and capped with an aluminum seal.
- Each filled vial is weighed (0.1 mg accuracy)
- The vials are then kept in inverted position for 16 hours in an enclosure wherein the pressure is lowered to 30-40 mm Hg.
- After this time, the pressure is returned to atmospheric and the vials are weighed again. Any weight change is recorded.
- The weight difference should not exceed 1 mg.
-

Test results:

OmniflexPlus coated 20 mm serum closures of the above described batch complied with the requirement of this test. No weight losses > 1 mg were observed.

Note:

The same test was carried out on stoppers of the same batch that had not been subjected to the above described sample preparation, but instead were gamma sterilized at 34 kGy.

Also here no weight losses > 1 mg were observed.

6.4.3 Proprietary procedure #3 ('CO_2 seal integrity test')

In this test, vials are filled with an amount of solid CO_2 which, after passing to the gaseous state, creates an internal overpressure of ca. 1.5 bar. The vials are stored at room temperature for a period of up to three months. Any leakage of CO_2 gas is detected by weighing the vials over time.[14]

Sample preparation:
20 closures are pretreated as described sub 6.2.2.2 (rinsing with water at 80 °C, autoclaving 30 mins/121 °C in an RfS bag, followed by overnight drying at 70 °C).

[14] Apart from leaking through the closure/vial interface, on the longer run carbon dioxide may also permeate through the stopper and 'leak' in this way.

Closures tested:

1. 20 mm closures V9048 , compound FM259/0, OmniflexPlus coated, batch 748009
2. 20 mm closures V9048 , compound FM259/0, not coated, batch 803918
3. 20 mm closures PTFE (Teflon®) coated

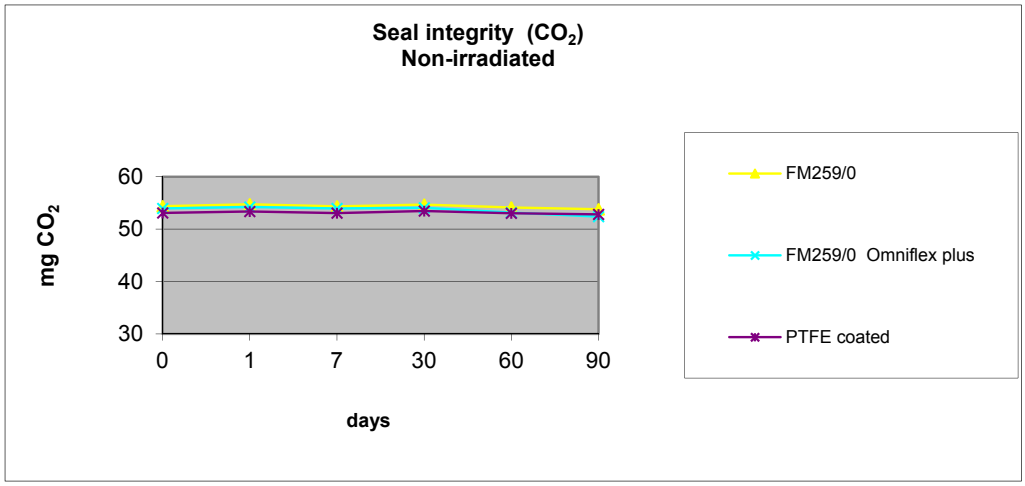
Procedure:

- For each closure type, standard 20 mm vials with a volume of 18 ml are filled with 53 ± 5 mg of solid carbon dioxyde (dry ice).
- Each vial is immediately closed with the closure type under test and capped with an aluminium seal.
- Allow the dry ice to sublimate and weigh the vials with an accuracy of 0.1 mg. Discard any vials which contain an amount of CO_2 which deviates more than 10% from the target of 53 mg.
- Repeat above procedure until 10 vials are obtained with the correct amount of CO_2 .
- The vials are stored at room temperature and weighed after 1, 7, 30, 60 and 90 days (accuracy 0.1 mg).

The change in weight of vials is illustrated in the table and figure below.

Table : weight change (with non-irradiated closures)

Storage time (days)						
Closure type	**0**	**1**	**7**	**30**	**60**	**90**
FM259/0	54.37	54.75	54.35	54.69	54.15	53.79
FM259/0 OmniflexPlus	53.95	54.24	53.96	54.06	53.16	52.40
PTFE coat	53.11	53.38	53.06	53.46	53.03	52.83

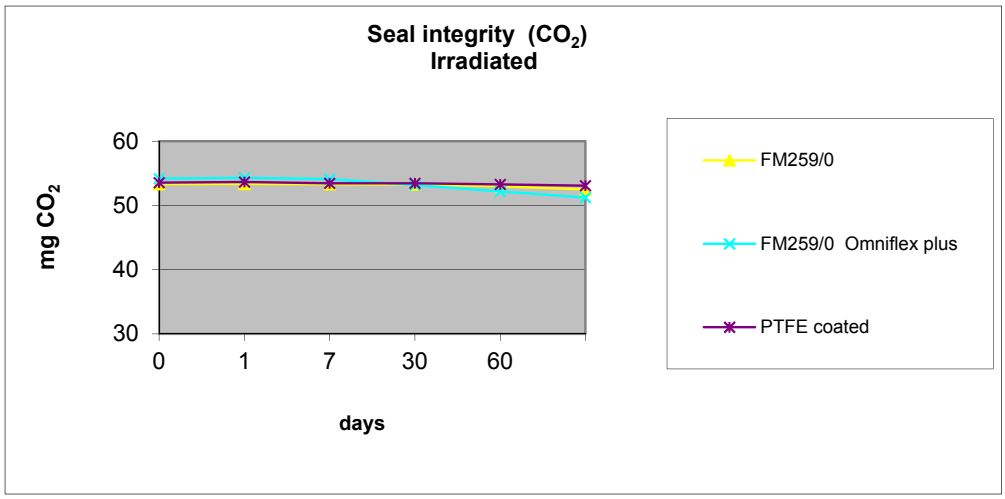


The same test was carried out on stoppers of the same batch that had not been subjected to the above described sample preparation, but instead were gamma sterilized at 34 kGy.

The change in weight of vials, closed with gamma- irradiated closures is shown in the table and figure immediately below.

Table : weight change (with closures irradiated @ 34 kGy)

Storage time (days)						
Closure type	**0**	**1**	**7**	**30**	**60**	**90**
FM259/0	53.31	53.35	53.28	53.24	52.96	52.49
FM259/0 OmniflexPlus	54.17	54.29	54.09	53.16	52.16	51.22
PTFE coated	53.55	53.65	53.45	53.43	53.26	53.06



6.5 Influence of low temperature storage on functional properties

In view of more and more drugs being involved in low temperature shipping and/or storage, the impact of low temperature on functional properties of OmniflexPlus stoppers was studied.

The study was conducted in 2 parts.

One part concerned storage at -78 °C. 20 mm serum V9048 OmniflexPlus stoppers were the subject of the study.

The second part concerned storage at 5 °C and at -23 °C. 20 mm serum V9048 OmniflexPlus stoppers as well as 20 mm lyophilization V9154 OmniflexPlus stoppers were included.

It was verified whether after low temperature storage the requirements for the functional properties as per Pharm. Eur. 3.2.9 / USP <381> and per ISO 8871-5 were still met.

Stopper/vial combinations that were stored at room temperature for the same period, were included in the test as controls.

Functional properties were recorded after low temperature storage and after the stopper/vial combinations had come back to room temperature.

6.5.1 Penetration force acc. Pharm. Eur. 3.2.9./USP <381>

6.5.1.1 *Storage for 1 and 3 months at –78 °C.*

Table : penetration forces after low temperature storage

OmniflexPlus stopper	**Penetration force (in N) (average for 10 stoppers – limit value is 10 N)**			
	1 month storage @		3 months storage @	
	room T	- 78 °C	room T	- 78 °C
20 mm serum V9048 batch 539004	1.69	1.71	2.06	2.00

No negative influence of the above described low temperature storage on penetration forces of the OmniflexPlus stoppers under test is noticed.

In all cases the requirements of Pharm. Eur. 3.2.9 and USP <381> are met for the tested stoppers.

6.5.1.2 *Storage for 1 and 3 months at 5°C and at –23°C*

Table : penetration forces after low temperature storage

OmniflexPlus stopper	**Penetration force (in N) (average for 10 stoppers – limit value is 10 N)**					
	1 month storage @			3 months storage @		
	room T	5 °C	- 23 °C	room T	5 °C	- 23 °C
20 mm serum V9048 batch 452001	1.75	1.66	1.83	1.83	1.81	1.54
20 mm lyo V9154 batch 514013	1.63	1.52	1.59	1.77	1.83	1.80

No negative influence of the above described low temperature storage on penetration forces of the OmniflexPlus stoppers under test is noticed.

In all cases the requirements of Pharm. Eur. 3.2.9 and USP <381> are met for the tested stoppers.

6.5.2 **Fragmentation acc. Pharm. Eur. 3.2.9./USP <381>**

6.5.2.1 *Storage for 1 and 3 months at –78 °C.*

Table : fragmentation after low temperature storage

OmniflexPlus stopper	**Fragmentation (# / 48 piercings – limit value 5)**			
	1 month storage @		3 months storage @	
	room T	- 78 °C	room T	- 78 °C
20 mm serum V9048 batch 539004	3	0	4	2

No negative influence of the above described low temperature storage on fragmentation of the OmniflexPlus stoppers under test is noticed.

In all cases the requirements of Pharm. Eur. 3.2.9 and USP <381> are met for the tested stoppers.

6.5.2.2 *Storage for 1 and 3 months at 5°C and at –23°C*

Table : fragmentation after low temperature storage

OmniflexPlus Stopper	**Fragmentation (# / 48 piercings – limit value 5)**					
	1 month storage @			3 months storage @		
	room T	5 °C	- 23 °C	room T	5 °C	- 23 °C
20 mm serum V9048 batch 452001	0	0	0	0	0	0
20 mm lyo V9154 batch 514013	0	0	0	0	3	0

No negative influence of the above described low temperature storage on fragmentation of the OmniflexPlus stoppers under test is noticed.

In all cases the requirements of Pharm. Eur. 3.2.9 and USP <381> are met for the tested stoppers.

6.5.3 Self-sealing acc. Pharm. Eur. 3.2.9./USP <381>

6.5.3.1 *Storage for 1 and 3 months at –78 °C.*

Table : self-sealing after low temperature storage

OmniflexPlus Stopper	**Self-sealing (# of vials with discoloration – limit value = 0)**			
	1 month storage @		3 months storage @	
	room T	- 78 °C	room T	- 78 °C
20 mm serum V9048 batch 539004	0	0	0	0

Low temperature storage at the above described conditions is found not to affect the self-sealing capacity of the OmniflexPlus stoppers under test.

In all cases the requirements of Pharm. Eur. 3.2.9 and USP <381> are met for the tested stoppers.

6.5.3.2 *Storage for 1 and 3 months at 5°C and at –23°C*

Table : self-sealing after low temperature storage

OmniflexPlus Stopper	**Self-sealing (# of vials with discoloration – limit value = 0)**					
	1 month storage @			3 months storage @		
	room T	5 °C	- 23 °C	room T	5 °C	- 23 °C
20 mm serum V9048 batch 452001	0	0	0	0	0	0
20 mm lyo V9154 batch 514013	0	0	0	0	0	0

Low temperature storage at the above described conditions is found not to affect the self-sealing capacity of the OmniflexPlus stoppers under test.

In all cases the requirements of Pharm. Eur. 3.2.9 and USP <381> are met for the tested stoppers.

6.5.4 Container closure seal integrity acc. ISO 8871-5

Principle:

Vials that are filled with liquid, stoppered and capped are submerged in a coloured solution. The vials are inspected for leakage as a result of a pressure differential across the closure/vial interface.

Sample preparation :

Closures are pretreated as follows:

- The closures are placed in a glass container and rinsed 3 times with distilled water at 80 °C.
- The rinsed (wet) stoppers are transferred to a small Ready-for-Sterilization (RfS®) bag, which is then sealed.
- The RfS bag is autoclaved during 30 min. at 121°C.

- After autoclaving, the RfS bag is dried overnight at 70 °C.
- The closures are kept in the sealed RfS bag until use.

Procedure:
- 10 vials are filled to nominal volume with water.
- The filled vials are closed with the rubber closures under test and capped with an aluminium seal.
- The vials are then completely immersed in a methylene blue solution in a container on which the pressure is reduced with 0.27 kPa.
- After 10 minutes the pressure in the container is restored to atmospheric pressure.
- 30 minutes later the vials are removed from the container and their outside is rinsed with water.
- The vial contents are then inspected for any traces of blue coloration.
- There should be no vials with a blue discoloration of the contents.

6.5.4.1 *Storage for 1 and 3 months at –78 °C.*

Table : container / closure seal integrity after low temperature storage

OmniflexPlus stopper	**Container / closure seal integrity (# of vials with discoloration – limit value = 0)**			
	1 month storage @		3 months storage @	
	room T	- 78 °C	room T	- 78 °C
20 mm serum V9048 batch 539004	0	0	0	0

Low temperature storage at the above described conditions is found not to affect the closure/vial sealing capacity of the OmniflexPlus stoppers under test. In all cases the requirement of ISO 8871-5 is met for the tested stoppers.

6.5.4.2 *Storage for 1 and 3 months at 5°C and at –23°C*

Table : container / closure seal integrity after low temperature storage

OmniflexPlus stopper	**Container / closure seal integrity (# of vials with discoloration – limit value = 0)**					
	1 month storage @			3 months storage @		
	room T	5 °C	- 23 °C	room T	5 °C	- 23 °C
20 mm serum V9048 batch 452001	0	0	n.i.	0	0	n.i.
20 mm lyo V9154 batch 514013	0	0	n.i.	0	0	n.i.

n.i. = not included in this part of the test

Low temperature storage at the above described conditions is found not to affect the closure/vial sealing capacity of the OmniflexPlus stoppers under test. In all cases the requirement of ISO 8871-5 is met for the tested stoppers.

6.6 Influence of steam sterilization and drying on functional and on chemical properties

Steam sterilisation is the most frequently used process for the sterilisation of rubber closures prior to aseptic filling of parenteral drugs. It is known that heat treatments are a stress factor for rubber products that may alter their chemical and functional properties.

A study was carried out to find out the effect of steam sterilization and subsequent drying on OmniflexPlus stoppers. For this study a stopper with a high penetration thickness (3.1 +/- 0.3 mm) was chosen. The high penetration thickness constitutes a worst case for functional properties (penetration force, fragmentation and self-sealing). Should there be any effect of the steam sterilization and drying cycle, then the effect can reasonably be assumed to be more prominent for stoppers with a higher penetration thickness.

20 mm lyophilization stoppers V9172 from batch 30237129, packed in Ready-for-Sterilization bags, were subjected to various steam sterilization and drying cycles according to the scheme below.

Table : various steam sterilization and drying conditions[15]

	Steam sterilization	Drying
A (*)	None	None
B	30 mins @ 121 °C	16 hrs @ 70 °C
C	60 mins @ 121 °C	16 hrs @ 70 °C
C	60 mins @ 121 °C	4 hrs @ 110 °C
E	60 mins @ 121 °C	8 hrs @ 110 °C
F	60 mins @ 121 °C	16 hrs @ 110 °C

(*) This condition corresponds with products in their delivery state from Datwyler.

It was studied whether any of these conditions had an impact on functional properties (penetrability, fragmentation, self-sealing), as well as on chemical properties as per Pharm. Eur. 3.2.9 / USP <381>.

Test results

For the stoppers under test, no effect was found, neither for functional nor for chemical testing.[16]

6.6 Influence of double steam sterilization on functional properties

20 mm serum OmniflexPlus stoppers V9048 from batch 30032050 were taken. Part of the samples was subjected to a double steam sterilization[17] 45 minutes / 121 °C. Another part that served as reference was not subjected to this pretreatment.

Functional properties (penetrability, fragmentation, self-sealing) were tested on the double steam-sterilized samples and on the reference according to Pharm. Eur. 3.2.9 / USP <381>. The results are summarized in the table below.

[15] The most common steam sterilization conditions in industry are 30 minutes / 121 °C.

[16] It is not Datwyler's intention to promote extreme heat treatments. Purpose of the study was to demonstrate that typical heat treatments used for processing rubber closures, or slight exaggerations thereof, do not present a risk on non-compliance. The final responsibility for the validation of steam sterilisation procedures lies with the end user who needs to verify the observations described here using the company's proprietary equipment and procedures.

[17] Multiple sterilization cycles as a rule are not recommended. Double steam sterilization of stoppers that were left over after a filling run however is a practice that occasionally is encountered.

Table : functional properties acc. Pharm. Eur. 3.2.9 / USP <381> after double steam sterilization cycle

Test	Unit	Limit	Result	
			Double steam ster. (45 mins/121 °C)	Reference
Penetrability	N	<= 10 N	1.6	1.6
Fragmentation	-	<= 5 fragments/48 penetrations	1	0
Self-sealing	-	0	0	0

Double steam sterilization 45 mins / 121 °C was found not to affect the functional properties of the OmniflexPlus closures under test. The functional requirements of Pharm. Eur. 3.2.9 / USP <381> were met also after double steam sterilization.

6.7 Influence of long term storage on functional properties

Retention samples of a batch of 20 mm serum V9048 OmniflexPlus stoppers (batch 049005) were retrieved from Datwyler's archive after 5 years of storage at ambient conditions. Functional properties as per Pharm. Eur. 3.2.9 / USP <381> were tested on these samples. Test results are summarized in the table below.

Table : Functional properties acc. Pharm. Eur. 3.2.9 / USP <381> after 5 years storage at ambient conditions

Test	Unit	Limit	Result
Penetrability	N	<= 10 N	1.7
Fragmentation	-	<= 5 fragments/48 penetrations	1
Self-sealing	-	0	0

5 years storage at ambient conditions was found not to affect the functional properties of the OmniflexPlus coated 20 mm serum closures under test. The functional requirements of Pharm. Eur. 3.2.9 / USP <381> were met also after this storage period.

7 Moisture content

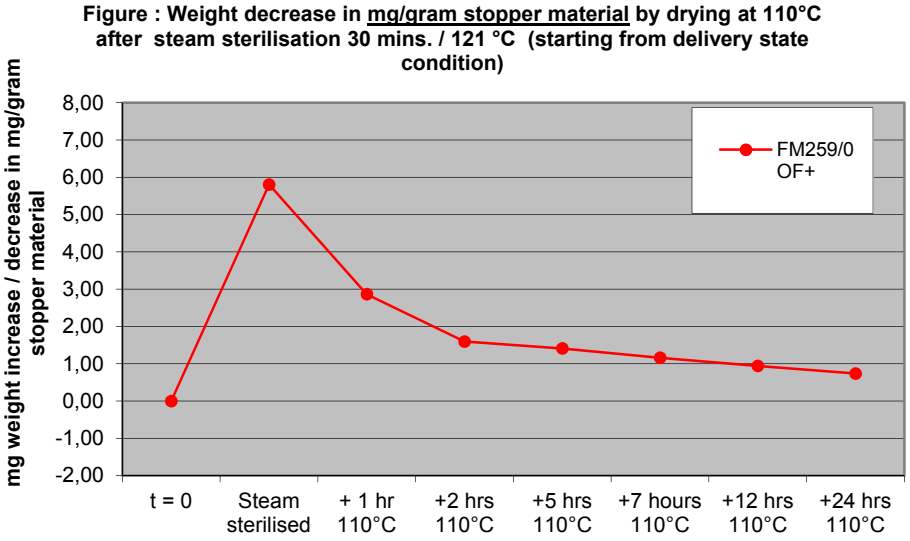
Results for moisture content that are given below are indicative only as the actual moisture content is dependent on numerous factors like stopper design, packaging way, climate, actual steam sterilization and drying equipment, etc.

The typical moisture content of FM259/0 OmniflexPlus stoppers after their final treatment at Datwyler, i.e. after washing, drying and packaging, is in the range of 0.3 w/w% (3 mg water/g rubber)

Additional moisture is absorbed during a steam sterilization step. For 20 mm serum OmniflexPlus stoppers (batch 549024) the moisture content was found to be 0.94 % after steam sterilization for 30 minutes at 121 °C.

The drying curve at 110 °C of the steam sterilized stoppers was recorded. The results are displayed in the figure below.

Figure : Moisture uptake after autoclaving 30min at 121°C and moisture release by drying at 110 °C



8 Pharmaceutical properties

8.1 Compatibility with cephalosporins

Cephalosporin powders are known to be very sensitive to contact with rubber closures and a satisfactory compatibility is difficult to achieve. Upon dissolution of the product with water for injection, a turbidity usually develops which can be attributed, in most cases, to an interaction between the rubber and the product.

The type of cephalosporin, the size of the powder particles, the contact time with the rubber closure, the type of closure material and the storage temperature are all factors which influence the degree of turbidity which eventually develops in the solution.

OmniflexPlus coated 20 mm serum closures were tested in contact with two types of cephalosporins for storage times up to 12 months. In all cases, the turbidity in the reconstituted solutions was very low and nearly independent of the contact time.

8.1.1 Test procedure

Preparation of vials:

Standard 20 mm type I glass vials are washed in a lab washing machine at 90°C, followed by 3 rinsing steps (1x water at room temperature, 1x distilled water at room temperature and 1x distilled water at 90°C).

The washed and rinsed vials are dried in a hot air oven at 150°C (in inverted position) .

The vials are stored in a sealed PE bag until use.

Preparation of closures:

For each closure type and each cephalosporin type, 18 closures are rinsed 3 times with distilled water at 80°C.

The rinsed (wet) closures are transferred to a small RfS bag, which is sealed.

The RfS bag is autoclaved during 30 minutes at 121°C .

After autoclaving, the RfS bag is dried overnight at 70°C.

The closures are further stored in the sealed RfS bag until use.

Closures tested:

20 mm closures V9048 , compound FM259/0, OmniflexPlus coated, batch 748009

20 mm closures V9048 , compound FM259/0, not coated, batch 803918

20 mm closures PTFE (Teflon®) coated

Test procedure:

The vials are filled with 100 mg of the cephalosporin powder, closed with the rubber closure under test, and capped with an aluminium seal. For each rubber closure type and each cephalosporin type, 18 vials are prepared.

The vials are stored in inverted position in a climate chamber at a temperature of 40°C and a relative humidity of 75%. Samples (3 vials per type) are taken for analysis after 1, 2, 3, 6, 9 and 12 months of storage time.

For the test, 3 ml of distilled water is added to the vials for complete dissolution of the cephalosporin powder.

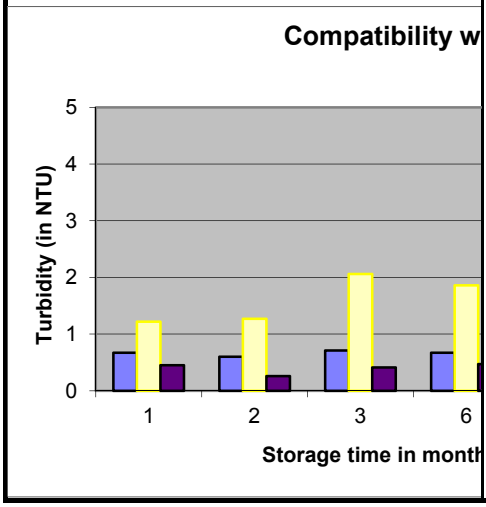
The turbidity of the resulting cephalosporin solutions is measured with a HACH Model 18900 Nephelometer. The results are expressed in NTU (Nephelometric Turbidity Units).

8.1.2 Compatibility with Cefazolin sodium

The turbidity data for the reconstituted solutions after storage times of 1 to 12 months are presented in the table and figure below.

Table : Turbidity (NTU) in Cefazolin-Na solutions

	Contact time (months) at 40°C					
Closure type	**1**	**2**	**3**	**6**	**9**	**12**
PTFE coated	0.67	0.60	0.71	0.67	1.00	0.59
FM259 not coated	1.22	1.27	2.06	1.86	3.30	3.77
FM259 OmniflexPlus	0.45	0.26	0.41	0.47	0.52	0.58

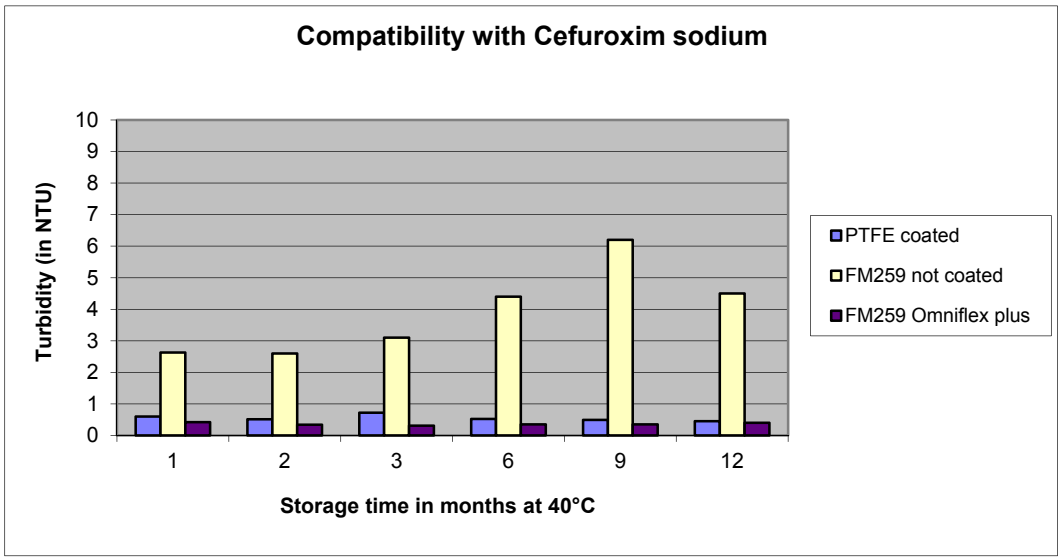


8.1.3 Compatibility with Cefuroxim sodium

The turbidity data for the reconstituted solutions after storage times of 1 to 12 months are presented in the table and figure below.

Table : Turbidity (NTU) in Cefuroxim-Na solutions

	Contact time (months) at 40°C					
Closure type	**1**	**2**	**3**	**6**	**9**	**12**
PTFE coated	0.60	0.51	0.72	0.52	0.49	0.45
FM259 not coated	2.63	2.60	3.07	4.37	6.23	4.53
FM259 OmniflexPlus	0.42	0.34	0.31	0.35	0.35	0.4



8.1.4 Conclusion

The test results indicate that the OmniflexPlus coated closures involved in the test are very compatible with cephalosporins, compared to non-coated closures. The turbidity which develops in the vials is very low and does not change much with storage time. In this respect OmniflexPlus closures involved in the test are comparable to PTFE (Teflon) coated closures.

8.2 Compatibility with solvents

8.2.1 Compatibility with oils

Closures tested:
1. 20 mm closures V9048 , compound FM259/0, OmniflexPlus coated, batch 748009
2. 20 mm closures V9048 , compound FM259/0, not coated, batch 803918
3. 20 mm closures PTFE (Teflon®) coated

8.2.1.1 Test procedure

Preparation of vials and of closures was the same as in the previous section.

Test procedure:

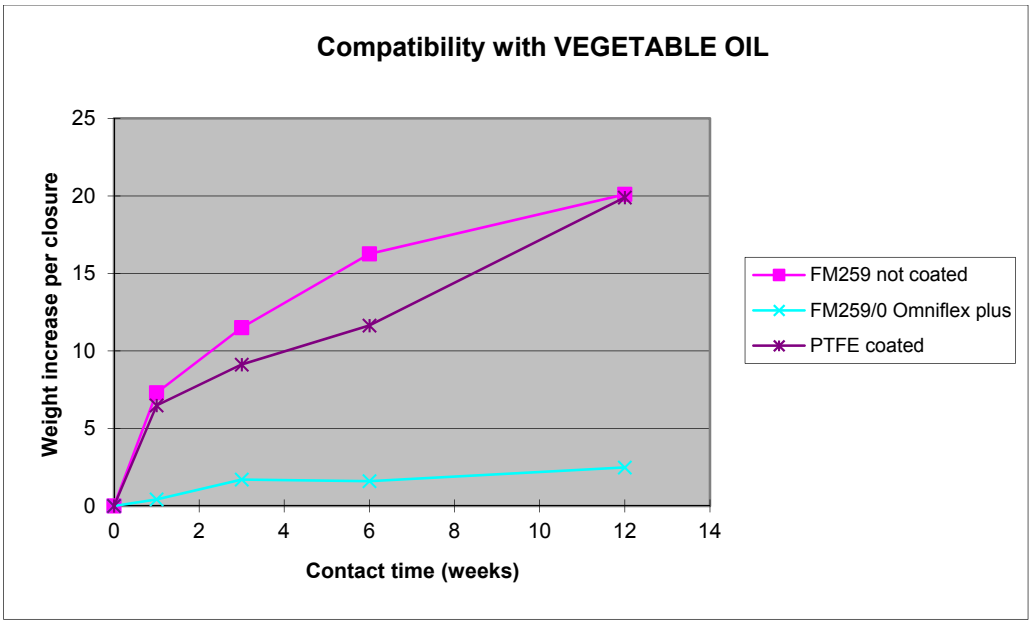
- For each closure sample and solvent, 20 closures are numbered and weighed accurately (0.1 mg accuracy).
- The vials are filled with 1 ml of the solvent and closed with the numbered and weighed closures.
- The vials with vegetable and mineral oil are stored in inverted position at room temperature for up to 12 weeks.
- The vials with ethyloleate/cottonseed oil are stored in inverted position at 40°C for up to 40 days
- For each closure sample, 5 vials are retrieved for tests, after 1, 3, 6 and 12 weeks (1, 6, 20 and 40 days for ethyleoleate/cottonseed oil) and the swelling of the closures is determined gravimetrically (weight increase as a result of solvent absorption).

8.2.1.2 Compatibility with vegetable oil

The data on the absorption of peanut oil by OmniflexPlus coated closures, uncoated and PTFE coated closures are presented in the table and figure below.

Table : Weight increase (mg) of closures in contact with peanut oil

	Contact time (weeks) at room temp.			
Closure type	**1**	**3**	**6**	**12**
PTFE coated	6.5	9.1	11.6	19.9
FM259 not coated	7.3	11.5	16.3	20.1
FM259 OmniflexPlus	0.4	1.7	1.6	2.5



In contact with vegetable oil the OmniflexPlus coated closures involved in the test show considerably less swelling (measured as a weight increase) than uncoated rubber closures.

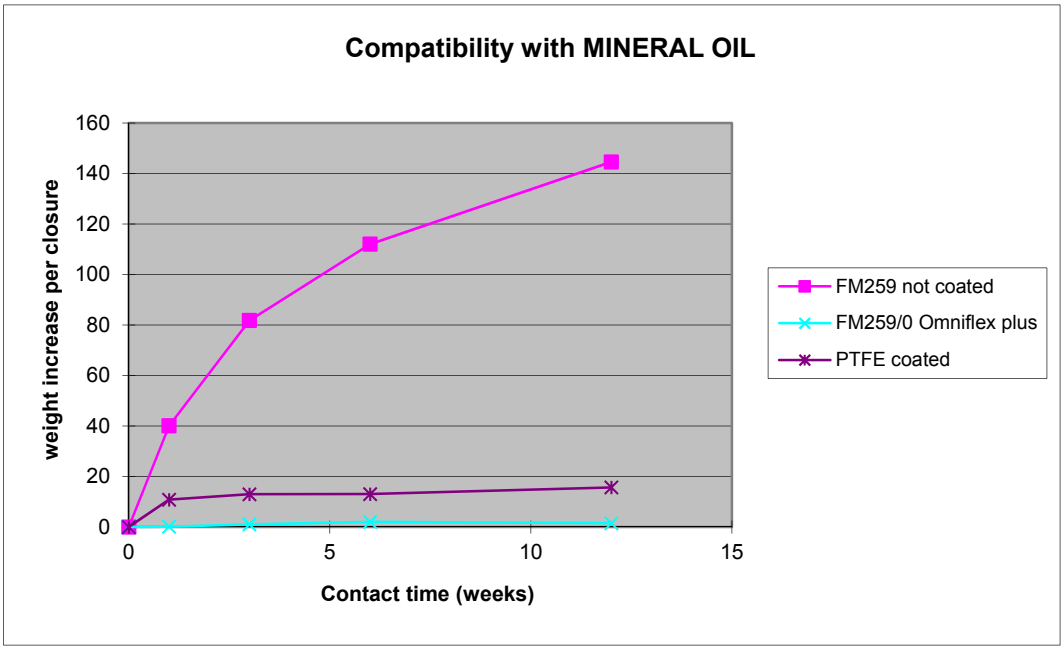
The result can be explained by the barrier effect of the OmniflexPlus coating.

8.2.1.3 Compatibility with mineral oil

The data on the absorption of paraffin oil by OmniflexPlus coated closures, uncoated and PTFE coated closures are presented in the table and figure below.

Table : Weight increase (mg) of closures in contact with paraffin oil

Closure type	Contact time (weeks) at room temp			
	1	3	6	12
PTFE coated	10.8	13	13.1	15.7
FM259 not coated	40.1	81.8	112.0	144.6
FM259 OmniflexPlus	0.2	1.1	2.0	1.5



In contact with mineral oil the OmniflexPlus coated closures involved in the test show considerably less swelling (measured as a weight increase) than uncoated rubber closures.

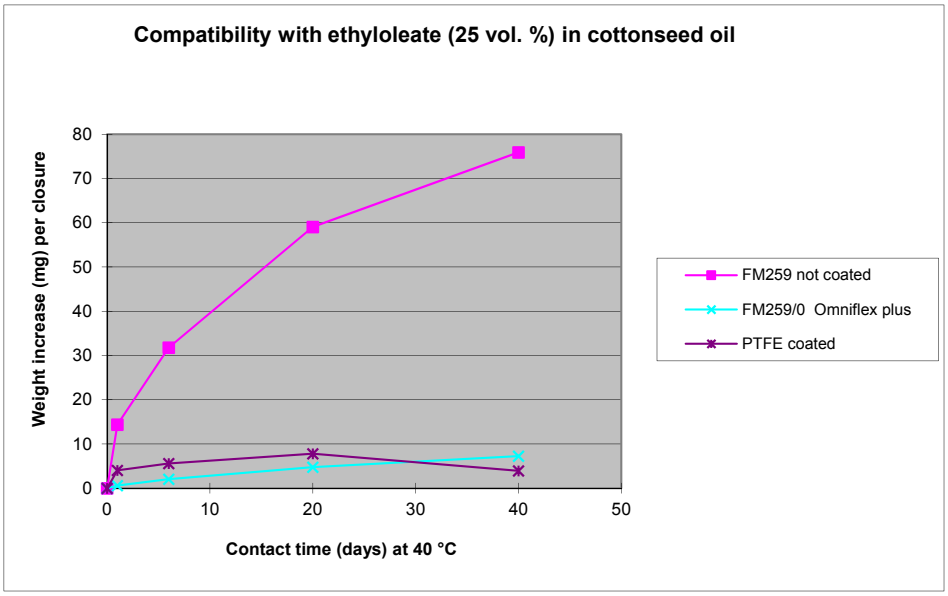
The result can be explained by the barrier effect of the OmniflexPlus coating.

8.2.1.4 Compatibility with ethyloleate (25 vol. %) in cottonseed oil

The data on the absorption of ethyloleate / cottonseed oil by OmniflexPlus coated closures, uncoated and PTFE coated closures are presented in the table and figure below.

Table : Weight increase (mg) of closures in contact with ethyloleate (25 vol.%) in cottonseed oil

Closure type	Contact time (days) at 40°C			
	1	6	20	40
PTFE coated	4.1	5.6	7.8	3.9
FM259 not coated	14.4	31.8	59.1	75.9
FM259 OmniflexPlus	0.6	2.1	4.8	7.3



In contact with ethyloleate / cottonseed oil the OmniflexPlus coated closures involved in the test show considerably less swelling (measured as a weight increase) than uncoated rubber closures.

The result can be explained by the barrier effect of the OmniflexPlus coating.

8.2.2 Compatibility with ethanol (95 vol %)

The compatibility of OmniflexPlus coated closures with 95% ethanol was evaluated by measuring the amount of UV active extractables after autoclaving and after storage at 40°C for up to 16 weeks.

8.2.2.1 Test procedure

Closures tested:
1. 20 mm closures V9048 , compound FM259/0, OmniflexPlus coated, batch 748009
2. 20 mm closures V9048 , compound FM259/0, not coated, batch 803918
3. 20 mm closures PTFE (Teflon®) coated

Preparation of vials, preparation of closures and type of closures tested is as in the previous section.

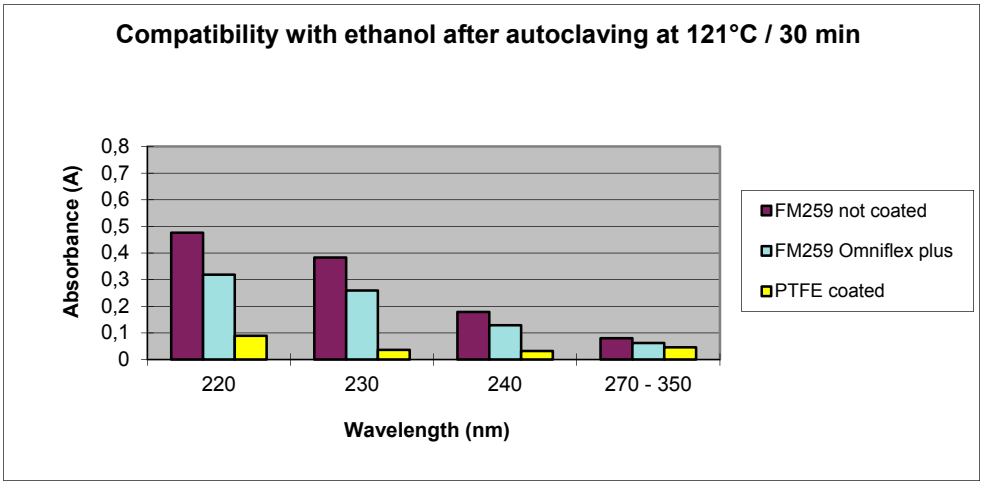
Test procedure:
- For each closure sample, 12 vials are filled with 5 ml ethanol. The vials are closed with the closures under test and capped with an aluminium seal.
- For each closure type, 3 vials are autoclaved in inverted position for 30 minutes at 121°C and allowed to cool to room temperature. The UV absorbance (190 – 350 nm) is measured.
- The other 9 vials are stored in inverted position at 40°C. After 1 , 4 and 16 weeks, the ethanol in 3 vials is tested for UV absorbance.
- The UV absorbance of the test solution is measured against pure ethanol 95% in the reference cell.

8.2.2.2 Compatibility with ethanol 95% after autoclaving

The UV absorbance data at 220, 230, 240 nm and the max. absorbance in the range 270 to 350 nm for ethanol which has been autoclaved in contact with rubber closures are shown in the table and figure below.

Table : UV absorbance (A) of ethanol after autoclaving in contact with rubber closures

Closure type	Wavelength (nm)			
	220	230	240	270 - 350
PTFE coated	0.089	0.036	0.032	0.046
FM259 not coated	0.476	0.383	0.179	0.080
FM259 OmniflexPlus	0.319	0.259	0.129	0.062



8.2.2.3 Compatibility with ethanol 95% at 40°C

The UV absorbance data (A max) in the range of 270 to 350 nm for ethanol that has been in contact with rubber closures for a period of up to 16 weeks, as well as the absorbance at 220 nm in the same conditions, are shown in the following tables and figures.

Table : A max in the range of 270 to 350 nm

Closure type	Contact time at 40°C (weeks)			
	0	1	4	16
PTFE coated	0	0.011	0.038	0.125
			0.137	0.274
			0.053	0.163

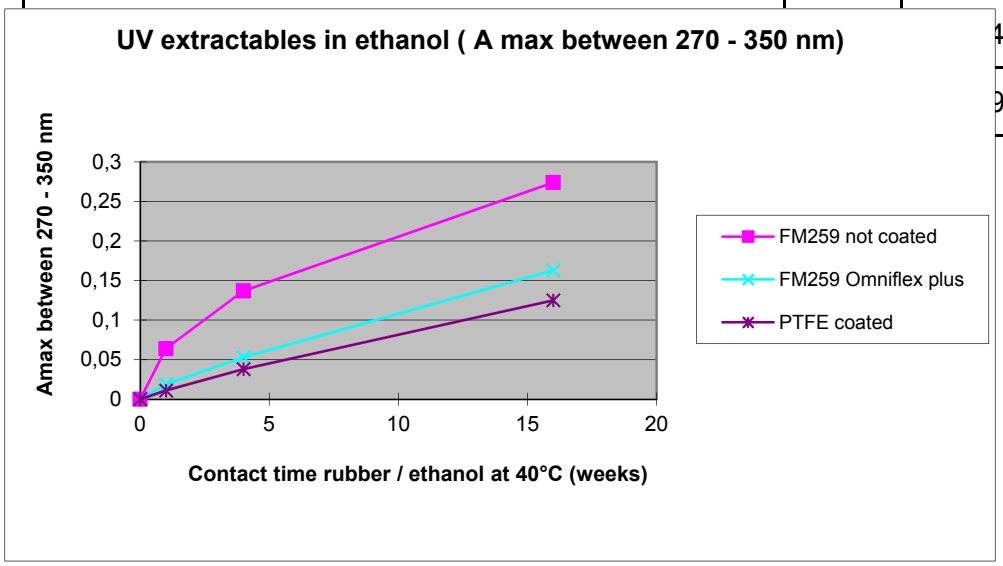
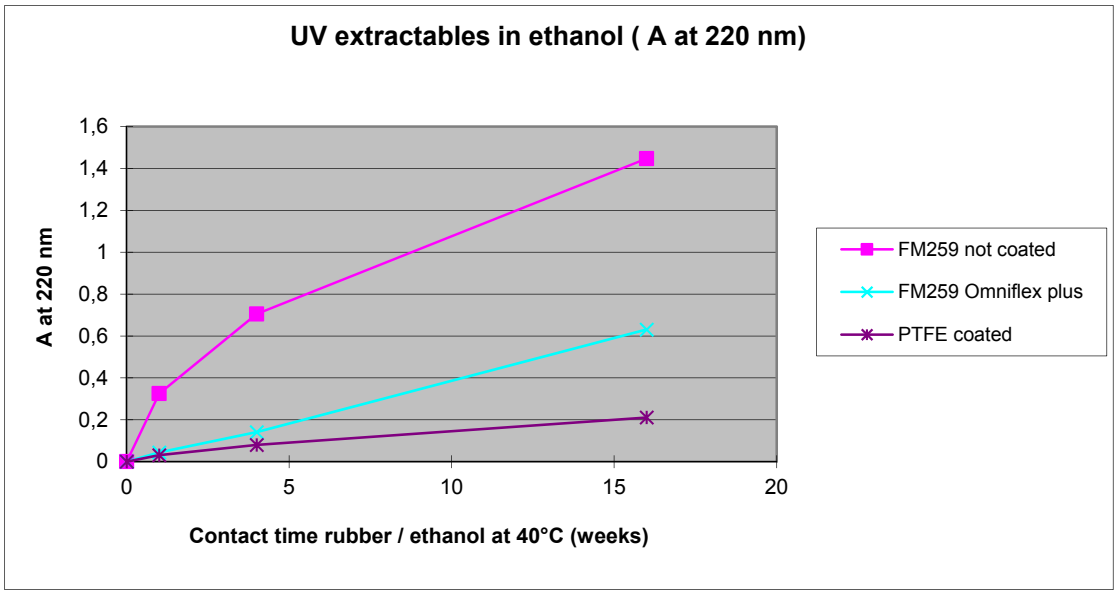


Table : Absorbance at 220 nm

Closure type	Contact time at 40°C (weeks)			
	0	1	4	16
PTFE coated	0	0.031	0.08	0.211
FM259 not coated	0	0.325	0.705	1.447
FM259 OmniflexPlus	0	0.044	0.141	0.631



8.2.2.4 Conclusion

UV absorbance of ethanol 95 % that has been in contact with the OmniflexPlus coated closures involved int the test is lower than that of ethanol 95 % in contact with uncoated FM259.

The result can be explained by the barrier effect of the OmniflexPlus coating.

8.2.3 Compatibility with other solvents

If you have questions on compatibility of OmniflexPlus with other solvents or solvent blends than reported in this document, it is recommended to take contact with your Datwyler sales representative.

Datwyler may avail of non-confidential compatibility data with such solvents or, in cooperation with the customer, can set up a study to generate such data.

Solvent or drug carrier compatibility should be investigated on a case-by-case basis. It has been demonstrated in certain studies that OmniflexPlus closures are not affected by specific individual solvents, but are affected by certain blends of them.

8.3. Compatibility with preservatives

Rubber closures have a tendency to absorb some drug formulation components, to a variable extent depending on their composition. If such components are originally present in only small quantities, this absorption phenomenon may pose a problem.

In this study, the absorption of three common preservatives, methyl paraben, propyl paraben and benzyl alcohol, was measured over time in solutions in contact with coated and non-coated rubber closures.

8.3.1 Methyl- and propyl paraben

8.3.1.1 Test procedure

Preparation of vials:
- Standard 20 mm type I glass vials are washed in a lab washing machine at 90°C, followed by 3 rinsing steps (1x water at room temperature, 1x distilled water at room temperature and 1x distilled water at 90°C).
- The washed and rinsed vials are dried in a hot air oven at 150°C (in inverted position) .
- The vials are stored in a sealed PE-bag until use.

Preparation of closures:
- For each closure type and each paraben type, 20 closures are rinsed 3 times with distilled water at 80°C.
- The rinsed (wet) closures are transferred to a small RfS bag, which is sealed.
- The RfS bag is autoclaved during 30' at 121°C .
- After autoclaving, the RfS bag is dried overnight at 70°C.
- The closures are further stored in the sealed RfS bag until use.

Closures tested:
1. 20 mm closures V9048 , compound FM259/0, OmniflexPlus coated, batch 748009
2. 20 mm closures V9048 , compound FM259/0, not coated, batch 803918

Paraben solutions:
1. Methyl paraben (methagin) : 0.5817 g / l in distilled water.
2. Propyl paraben (propagin) : 0.1245 g / l in distilled water.

The concentrations that were chosen are in a range that is common in industry and were set in cooperation with a customer.

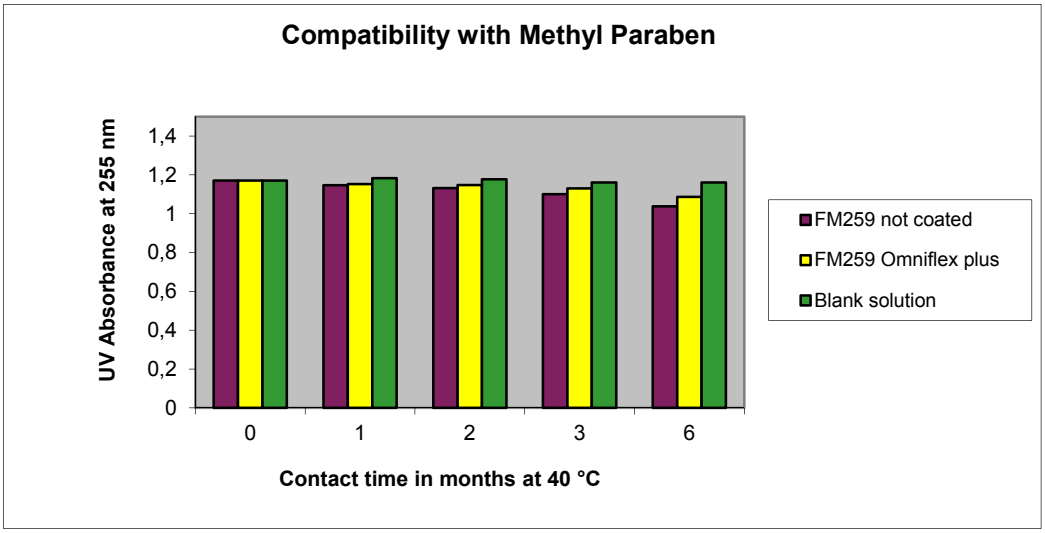
Test procedure:
- For each closure sample and each paraben type, 40 vials are filled with 5 ml of the paraben solution. The vials are closed with the closures under test and capped with an aluminium seal.
- 20 vials are stored in inverted position (test vials) and 20 reference vials are stored upright (solution not in contact with the rubber closure) at 40°C , for a period up to 6 months.
- After 1, 2, 3 and 6 months, 5 test vials and 5 reference vials are retrieved and the concentration of the parabens is measured by means of UV spectrophotometry at 255 nm. For this purpose, the methyl paraben solutions are diluted 1/50, the propyl paraben solutions 1/12.5.

8.3.1.2 Compatibility with methyl paraben

The UV absorbance data of methyl paraben solutions (at 255 nm) in contact with different closure types are shown in the table and figure below, in comparison with solutions which were not in contact with any rubber closure (‘blank solution’).

Table : UV absorbance at 255 nm (methyl paraben)

	Contact time (months) at 40°C .				
	0	**1**	**2**	**3**	**6**
Blank solution	1.171	1.183	1.177	1.161	1.161
FM259 not coated	1.171	1.147	1.132	1.101	1.038
FM259 OmniflexPlus	1.171	1.153	1.148	1.130	1.087



In aqueous solutions of the studied concentration OmniflexPlus stoppers involved in the test absorb slightly less methyl paraben than uncoated closures do.

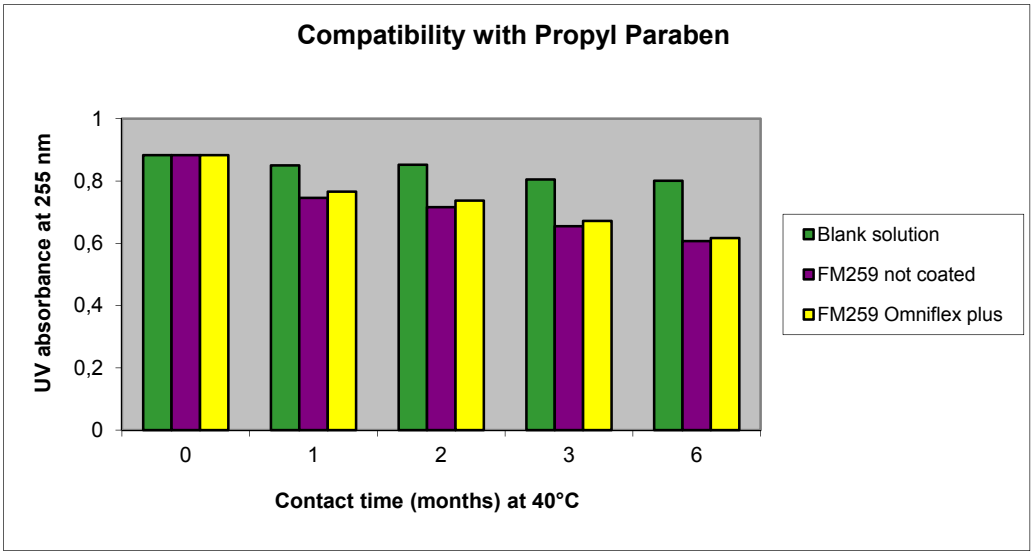
8.3.1.3 Compatibility with propyl paraben

The UV absorbance data of propyl paraben solutions (at 255 nm) in contact with different closure types are shown in the table and figure below, in comparison with solutions which were not in contact with any rubber closure (‘blank solution’).

Table : UV absorbance at 255 nm (propyl paraben)

	Contact time (months) at 40°C .				
	0	**1**	**2**	**3**	**6**
Blank solution	0.883	0.85	0.852	0.805	0.801

FM259 not coated	0.883	0.746	0.716	0.655	0.607
FM259 OmniflexPlus	0.883	0.766	0.737	0.672	0.617



In aqueous solutions of the studied concentration OmniflexPlus stoppers involved in the test absorb slightly less propyl paraben than uncoated closures do.

8.3.2 Benzyl alcohol

8.3.2.1 Test procedure

Preparation of closures:
- For each closure type the needed number of closures is rinsed 3 times with distilled water at 80°C.
- The rinsed (wet) closures are transferred to a small RfS bag, which is sealed.
- The RfS bag is autoclaved during 30 min. at 121°C .
- After autoclaving, the RfS bag is dried overnight at 70°C.
- The closures are further stored in the sealed RfS bag until use.

Closures tested:
1. 20 mm 'PTFE' plug coated closures
2. 20 mm closures V9048 , compound FM259/0, OmniflexPlus coated (batch 347002)
3. 20 mm closures V9048 , compound FM259/0, not coated (batch 348278).

Benzyl alcohol solution:
Benzyl alcohol at 1 w / vol % in distilled water.
The concentration was chosen in a range that is common in industry.

Test procedure:
- For each stopper sample vials are filled with 10 ml of 1% benzyl alcohol.
- The vials are stoppered with the stopper under investigation and are crimped with an aluminum cap.
- The vials are stored in inverted position in a climate chamber at 40 °C / 75% RH (relative humidity).

- After 1, 2, 3 and 6 months three vials per stopper type are taken and are analyzed for residual benzyl alcohol concentration.
  Analysis is performed using HPLC as technique.

8.3.2.2 Compatibility with benzyl alcohol

The change in concentration of an aqueous 1 w/v % benzyl alcohol solution in contact with rubber closures, at 40°C, for a period up to 6 months is shown in the table below.

Table : benzyl alcohol concentrations (w/v %)

Stopper type	Contact time at 40°C (months)			
	1	2	3	6
PTFE coated	1.01	1.01	1.02	1.07
FM259 not coated	0.99	1.00	1.02	1.03
FM259 OmniflexPlus	1.00	1.00	1.03	1.05

Note: concentrations in the table are the average of the 3 measured vials.

At the studied concentration no absorption of benzyl alcohol from aqueous solutions is being observed, neither for OmniflexPlus coated closures, nor for uncoated closures involved in the test.

8.3.3 Benzalkonium chloride

8.3.3.1 Test procedure

Preparation of vials and of closures:
See procedure for benzyl alcohol in previous section

Closures tested:
1. 20 mm 'PTFE' plug coated closures
2. 20 mm closures V9048 , compound FM259/0, OmniflexPlus coated (batch 347002)
3. 20 mm closures V9048 , compound FM259/0, not coated (batch 348278).

Benzalkonium chloride solution:
Benzalkonium chloride at 0.02 w / vol % in distilled water.
The concentration was chosen in a range that is common in industry.

Test procedure:
- For each stopper sample vials are filled with 10 ml of 0.2 % benzalkonium chloride.
- The vials are stoppered with the stopper under investigation and are crimped with an aluminum cap.
- The vials are stored in inverted position in a climate chamber at 40 °C / 75% RH (relative humidity).
- After 1, 2, 3 and 6 months three vials per stopper type are taken and are analyzed for residual benzalkonium chloride concentration.
  Analysis is performed using HPLC as technique.

8.3.3.2 Compatibility with benzalkonium chloride

The change in concentration of an aqueous 0.02 w/v % benzyl alcohol solution in contact with rubber closures, at 40°C, for a period up to 6 months is shown in the table below.

Table : Benzalkonium chloride concentrations (w/v %)

Stopper type	Contact time at 40°C (months)			
	1	2	3	6
PTFE coated	0.019	0.020	0.020	0.019
FM259 not coated	0.020	0.020	0.019	0.020
FM259 OmniflexPlus	0.020	0.020	0.020	0.020

Note: concentrations in the table are the average of the 3 measured vials.

At the studied concentration no absorption of benzalkonium chloride from aqueous solutions is being observed, neither for OmniflexPlus coated closures, nor for uncoated closures involved in the test.

9 Biological properties

9.1 USP <1031>

USP <1031>, “The Biocompatibility of Material used in Drug Containers”, stipulates that the biocompatibility of an elastomeric material is evaluated according to the two stage testing protocol specified in the USP <381>. An elastomeric material that does not meet the requirements of the first-stage testing (in vitro, USP <87>), may qualify as a biocompatible material by passing the second stage testing (in vivo, USP <88>).

No class or type distinction is made between elastomeric materials that meet the requirements of first-stage of testing and those that qualify as biocompatible meeting the second-stage requirements.

9.2 USP <87>

Biological testing (-elution test-) is carried out on a sample of OmniflexPlus 20 mm lyophilization stoppers[18] as per USP <87>, “Biological Reactivity Tests, In Vitro”. OmniflexPlus is proven to be non-cytotoxic. A copy of the report is attached here below.

[18] In view of the nature of USP <87> testing, the results can reasonably be assumed to be transferable to other OmniflexPlus closure geometries.

TOXIKON
Leaders in Life Science and Technology

TEST RESULT REPORT

Project Number:	**TE 09713**	**Study Number:**	**09-B1815-N1**
Sponsor:	**Helvoet Pharma Belgium NV**	**Report Date:**	**01/10/2009**
Contact:	*Mrs. Nadia Nouri*		
Address:	Industrieterrein Kolmen 1519 3570 Alken, Belgium	Date Sample Arrival: Technical Initiation:	23/09/2009 28/09/2009
PO.Number:	PB0903108	Technical Completion:	01/10/2009

Study	**Elution Test - ISO**	Temp/Time	37°C/24 hours
Test Item	V9172 FM259/0 OmniflexPlus Not Irradiated	Ratio	25cm²/20mL
Lot	30140129	Vehicle	MEM-Complete

REFERENCE: According to "ISO 10993-5, 1999: Biological Evaluation of Medical Devices- Part 5:Tests for In Vitro Cytotoxicity." and "USP 32-NF 27, 2009: <87> Biological reactivity test, in vitro." Toxikon Reference: SOP 3.1.2.3, rev. 07

PROCEDURE: The biological reactivity of a mammalian monolayer, L929 mouse fibroblast cell culture, in response to the test item extract was determined. Extracts were prepared at 37±1°C for 24 hours in a humidified atmosphere containing 5±1% carbon dioxide (static). Positive (natural rubber) and negative (silicone) control articles were prepared to verify the proper functioning of the test system. The extracts were sterile filtered. The maintenance medium on the cell cultures is replaced by the extracts of the test item or control article in triplicate and the cultures are subsequently incubated for 48 hours, at 37±1°C, in a humidified atmosphere containing 5±1% carbon dioxide. Biological reactivity was rated on the following scale: Grade 0 (No reactivity); Grade 1 (Slight reactivity), Grade 2 (Mild reactivity), Grade 3 (Moderate reactivity) and Grade 4 (Severe reactivity). The test item is considered non-cytotoxic if none of the cultures exposed to the test item shows greater than mild reactivity (Grade 2).

RESULTS: No reactivity (Grade 0) was exhibited by the cell cultures exposed to the test item at the 48 hours observation. Severe reactivity (Grade 4) was observed for the positive control article. The negative control article showed no signs of reactivity (Grade 0).

CONCLUSION: Based on the evaluation criteria mentioned above, the test item is considered non-cytotoxic.

RECORD STORAGE: All raw data generated in this study will be archived at Toxikon Europe, according to SOP 4.2.8.

AUTHORIZED PERSONNEL

ir. Peter Cornelis
Study Director

Ellen Sacreas
Quality Assurance

9.3 USP <88>

In view of the demonstrated compliance with USP <87>, performing USP <88> testing on OmniflexPlus stoppers is redundant.

Documentation of compliance with USP <88> was though generated at the time of product development. This documentation is however not reproduced in this document.

9.4 ISO 8871-4

ISO 8871-4, “Elastomeric parts for parenterals and for devices for pharmaceutical use – Part 4: Biological requirements and test methods”, specifies biological requirements for bacterial endotoxins, bioburden, cytotoxicity and intracutaneous and systemic toxicity.

The requirements for endotoxins and bioburden are left open and shall be agreed upon between supplier and user.

Bacterial Endotoxin Testing and bioburden testing are frequently carried out for OmniflexPlus closures that are supplied in Ready-for-Sterilization format. Datwyler’s current specification for bacterial endotoxins viz. bioburden on OmniflexPlus closures produced in Datwyler’s FirstLine plant are < 0.02 EU / cm² of rubber surface area and < 0.05 CFU / cm². For a 20 mm serum stopper this corresponds with < 0.2 EU / closure viz. < 0.5 CFU / closure.

For the toxicity tests, the same approach as in USP <1031> is given in ISO 8871-4, including reference to the USP <87>, in vitro test, for cytotoxicity testing and to the USP <88>, in vivo test, for the intracutaneous and systemic toxicity testing.

10 Particulate cleanliness

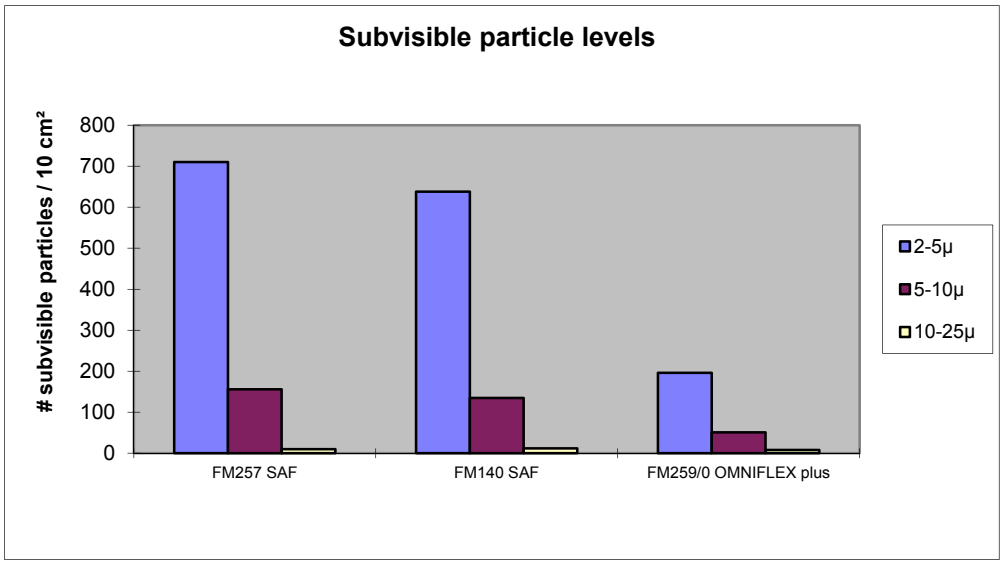
Method

The level of subvisible and visible particles on 20 mm serum OmniflexPlus coated closures was determined using the method described in ISO 8871-3.

The typical numbers of subvisible and visible particles found on OmniflexPlus coated closures (20 mm type V9048), compared with the numbers for non-coated closures in two other rubber compounds (compounds FM257 and FM140) are shown here below. The non-coated closures are siliconized by means of a high-viscosity (30,000 cSt) silicone oil (SAF siliconization process).

10.1 Subvisible particles

According to ISO 8871-3, ten OmniflexPlus coated closures V9048 (100 cm² rubber surface) are placed in an Erlenmeyer flask and submerged in 100 ml particle-free water (< 100 particles larger than 2 µm per 5 ml) . The flask is shaken mechanically for 20 sec. and the fluid is checked for particles by means of an electronic particle counter.



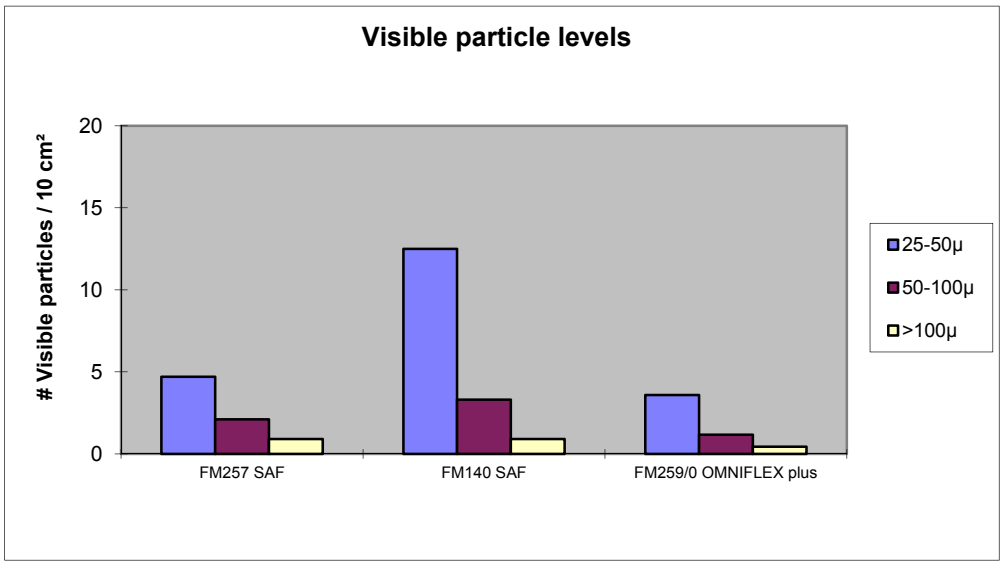
The number of subvisible particles released in ISO 8871-3 testing by OmniflexPlus coated closures is found to be significantly lower than by uncoated, siliconized closures. This finding is confirmed by historical results of finished product testing at Datwyler.
The number of subvisible particles is lower thanks to the absence of a surface siliconization step in the washing process of OmniflexPlus coated closures. Surface silicone is known to be contributing for a large part to subvisible particulate formation by elastomeric closures. OmniflexPlus coated closures do not require such a siliconization step thanks to the properties of the OmniflexPlus coating.

10.2 Visible particles

According to ISO 8871-3, ten OmniflexPlus coated closures V9048 (100 cm² rubber surface) are placed in an Erlenmeyer flask and submerged in 50 ml of a 0.03% solution of the sodium salt of N-methyl-N-oleylmethylaminoethanesulfonic acid[19] in purified water, filtered through a 1.2 µm filter. The flask is shaken mechanically for 20 sec. and the fluid is filtered over a 0.8 µm membrane filter. The process is repeated with another 50 ml of the fluid which is filtered over the same membrane filter.

[19] Detergent as per ISO 8871-3 at the time of testing. Later versions of ISO 8871-3 stipulate Polysorbate 80 as detergent.

The particles on the membrane filter are counted under a microscope at a 50x magnification.



The number of visible particles released in ISO 8871-3 testing by OmniflexPlus coated closures is found to be lower than by uncoated, siliconized closures.

10.3 Impact of transport on particulate cleanliness

The impact of transport on OmniflexPlus closures in Ready-for-Sterilization packaging has been performed on 13 mm serum closures, type V9239. Vibration testing according to ISTA method 1A[20] was chosen to simulate transport.

Two standard Datwyler boxes were filled under production conditions with 4 Ready-for-Sterilization bags containing each 4,000 pieces of 13 mm serum (V9239) OmniflexPlus closures. 3 bags were filled with products from 3 independent production batches (1 bag each), while the fourth bag was filled with products from one of the 3 batches, in order to fill the box in a way that is representative for shipments to customers of the product code in question.

One box has was subjected to transport simulation testing according to ISTA Method 1A, while the other box was kept on storage.

After the transport simulation test, particulate cleanliness in both the visible and subvisible range of the products before and after transport simulation was measured as per ISO 8871-3.

Additionally products after transport simulation were subjected again to visible inspection testing that makes part of Datwyler's finished product testing for this product code.

[20] ISTA = International Safe Transit Association. Method 1A uses fixed displacement vibration.

10.3.1 Visible particulate cleanliness

The results of visible particulate cleanliness testing as per ISO 8871-3 on products before and after transport simulation are given in the tables below.

Table : Results for products before transport simulation

# particles / 10 cm²	Batch number			Int. warning limit (*)
Size range	**448013**	**447020**	**447021**	
25-50 µm	2.7	3.4	1.2	8
50-100 µm	1.7	3.5	0.6	6.0
>100 µm	0.3	1.1	0.3	1.6

(*) internal warning limit

Table : Results for products after transport simulation

# particles / 10 cm²	Batch number			Int. warning limit
Size range	**448013**	**447020**	**447021**	
25-50 µm	5.3	2.3	1.1	8
50-100 µm	3.0	1.8	0.3	6.0
>100 µm	1.1	0.1	0.2	1.6

Conclusion

No clear effect of transport simulation on visible particulate cleanliness is noted for the stoppers under test. Numerical results on products from the same batch may be higher or lower after transport simulation. Such variability is inherent to this type of testing. In all cases results are well below internal warning limits used at Datwyler.

10.3.2 Subvisible particulate cleanliness

The results of subvisible particulate cleanliness testing as per ISO 8871-3 on products before and after transport simulation are given in the tables below.

Table : Results for products before transport simulation

# particles / 10 cm²	Batch number			Int. warning limit (*)
Size range	**448013**	**447020**	**447021**	
2 – 5 µm	105	261	185	600
5 - 10 µm	14	40	32	130
10 - 25 µm	2	2	2	21

(*) internal warning limit

Table : Results for products after transport simulation

# particles / 10 cm²	Batch number			Int. warning limit
Size range	**448013**	**447020**	**447021**	
2 – 5 µm	171	273	146	600
5 – 10 µm	22	51	18	130
10 - 25 µm	1	3	1	21

Conclusion

No clear effect of transport simulation on subvisible particulate cleanliness is noted for the stoppers under test. Numerical results on products from the same batch may be higher or lower after transport simulation. In all cases results are well below internal warning limits used at Datwyler.

10.3.3 Finished product inspection

Part of finished product inspection is the assessment of the number of products with minor cosmetic defects in a statistical sample as per in-house Standard Operating Procedures.

In this particular case 500 products were inspected per bag.

No differences were seen in results for products of the above described batches before and after transport simulation.

11 OmniflexPlus formats

OmniflexPlus products are available in the following formats:

1. **Bulk**
   The products are washed with a validated washing program (FLCO). After washing and drying they are packed in multiple protective polyethylene bags (non-sterilizable).

2. **Ready-for-Sterilization ('RfS')**
   OmniflexPlus products are FLCO washed and dried. Then they are packed in steam-sterilizable bags (RfS bags) and protective non-sterilizable polyethylene overwrapping. Steam sterilization of the products in the RfS bags after removal of the protective overwrapping is performed before aseptic filling.

3. **Ready-To-Use ('RTU')**
   OmniflexPlus products are FLCO washed and dried. Then they are packed in multiple multilayer bags that are compatible with gamma irradiation ('RTU bags'). The products are gamma sterilized at a contractor, using a validated irradiation process.
   OmniflexPlus products are equally available in irradiation compatible Rapid Transfer Port bags.

Supportive technical documentation on FLCO washing, Ready-for-Sterilization and Ready-To-Use is available from Datwyler. Please ask your Datwyler Sales Representative.

12 Compatibility with steam sterilization

12.1 Autoclaving

The testing described in this paragraph illustrates the behavior of OmniflexPlus closures after steam sterilization under exaggerated conditions.

The purpose of the testing is to investigate whether OmniflexPlus closures develop a tendency to stick upon steam sterilization and whether there is any visible damage to the coating layer.

12.1.1 **Autoclave test in bulk**

Test procedure:

Glass beakers with internal diameter of 75 mm were loaded with 100 OmniflexPlus coated 20 mm closures V9048 and with 100 OmniflexPlus coated 20 mm 2-leg lyophilization stoppers each. The total stacking height of the closures is approximately 7 cm for the 20 mm serum stoppers and 9.5 cm for the 20 mm lyo stoppers respectively, which is an exaggeration of the stacking height that may be encountered when sterilizing closures in bulk in stainless steel containers.

The closures were rinsed 3 times with distilled water at room temperature and subsequently autoclaved at 121°C for 30 minutes.

The beaker with the autoclaved closures is stored for 24 hours at room temperature, whereby the closures are kept immobile, after which time the beaker is inverted and the closures are dumped on a flat surface.

The number of closures sticking together and the number of closures with a damaged coating are recorded.

Figure :

OmniflexPlus closures (20 mm lyo) stacked in beaker	**Dried OmniflexPlus closures (20 mm lyo) dumped on table**
	

Test result:

The autoclaved OmniflexPlus coated closures involved in the test were found not to stick together . No closures with damage to the coating were observed. In this respect they are comparable with siliconized closures.

12.1.2 Autoclave test in RfS® bags

Test procedure:

2,500 OmniflexPlus coated 20 mm closures V9048, packaged in a Ready-for-Sterilization (RfS) bag of suitable size, are subjected to an autoclave cycle at 121°C for 45 minutes[21], followed by 90 minutes vacuum drying . Thereafter, the RfS bags are left to cool to room temperature, outside the autoclave.

Also, RfS bags containing 1800 closures each, are autoclaved using the above cycle and then stacked 5-high, during the cooling period outside the autoclave[22].

The closures are transferred to a hopper and checked for twinning and sticking which prohibits passage through the hopper.

Test result:

The autoclaved OmniflexPlus coated closures involved in the test were found not to stick together and passed through the hopper without any problems. No closures with damage to the coating were observed.

12.2 Closure insertion force

In this test, the force required to insert OmniflexPlus coated 20 mm closures V9048 in a standard 20 mm vial is measured, in comparison with uncoated (siliconized) closures. Tests were done on steam sterilized, on gamma irradiated and on untreated samples.

Preparation of closures:

Autoclaved closures:

- For each closure type, 10 closures are rinsed 3 times with distilled water at 80°C.
- The rinsed (wet) closures are transferred to a small RfS bag, which is sealed.
- The RfS bag is autoclaved for 30 minutes at 121°C .
- After autoclaving, the RfS bag is dried overnight at 70°C.
- The closures are further stored in the sealed RfS bag until use.

[21] Typically stoppers are sterilized at 121 °C for 30 minutes.

[22] Stacking was undertaken for the purpose of the experiment only. In order to avoid all risks on clumping and on deformation of stoppers, stacking of bags with steam sterilized and dried stoppers in warm condition shall not be undertaken in industrial practice !

Irradiated closures
- For each closure type, 10 non-pretreated closures are gamma irradiated with a dose of 34 kGy.

Closures tested:
1. 20 mm closures V9048, OmniflexPlus coated, batch 905017
2. 20 mm closures V9048 , compound FM259/0, not coated, siliconized, batch 803918

Test procedure:
- The neck of a standard 20 mm vial is degreased with MIBK (methyl isobutyl ketone).
- A closure is inserted in the vial at a speed of 300 mm/min. by means of a tensile test bench and the maximum force during closure insertion is recorded.
- The same vial is used for all closure samples. Between samples, the vial neck is degreased with MIBK.

Test results:

		Insertion force (N)		
		Untreated	**Steam sterilized (30 mins/121 °C)**	**Irradiated (34 kGy)**
V9048 FM259/0 SAF1 siliconized Batch 803918	Min		9.4	5.4
	Max	n.m.	16.0	11.2
	Avg		**12.0**	**7.4**
V9048 FM259/0 20 mm serum OmniflexPlus Batch 905017	Min	8.4	7.8	7.2
	Max	13.0	12.7	10.7
	Avg	**9.8**	**10.0**	**9.0**

n.m. = not measured

Conclusion

The OmniflexPlus coated 20 mm serum closures involved in the test exhibit similar insertion forces as uncoated, siliconized closures of the same product design and in the same compound.

Insertion forces for the tested OmniflexPlus closures were found not to be significantly affected by sterilization.

12.3 Influence of steam sterilization and drying on performance properties

20 mm lyophilization stoppers V9172 from batch 30237129, packed in Ready-for-Sterilization bags, were subjected to various steam sterilization and drying cycles according to the scheme below (same scheme as under 6.6).

Table : various steam sterilization and drying conditions[23]

	Steam sterilization	Drying
A (*)	None	None
B	30 mins @ 121 °C	16 hrs @ 70 °C
C	60 mins @ 121 °C	16 hrs @ 70 °C
C	60 mins @ 121 °C	4 hrs @ 110 °C
E	60 mins @ 121 °C	8 hrs @ 110 °C
F	60 mins @ 121 °C	16 hrs @ 110 °C

(*) This condition corresponds with products in their delivery state from Datwyler.

It was studied whether any of these treatments has an impact on the following performance characteristics :

- Clumping tendency (stickiness after sterilization and drying)
- Vial insertion force

For the OmniflexPlus closures involved in the test, no effect on either of these properties was found.[24]

13 Compatibility with gamma irradiation

Demonstration of the suitability of OmniflexPlus vial stoppers after gamma irradiation at doses of 25 and of 40 kGy has been a sub-project within Datwyler's larger 'Ready-To-Use' project for rubber closures.

Under the name 'Ready-To-Use' Datwyler supplies rubber closures with a Sterility Assurance Level (SAL) of 10^{-6} . The closures have been rendered sterile by gamma irradiation at an approved contractor. They are supplied to customers with a 2 year shelf life. Extensive product, process and packaging validation based on ISO 11137 formed the backbone of the project.

[23] The most common steam sterilization conditions in industry are 30 minutes / 121 °C.

[24] It is not Datwyler's intention to promote extreme heat treatments. Purpose of the study was to demonstrate that typical heat treatments used for processing rubber closures, or slight exaggerations thereof, do not present a risk on non-performance. The final responsibility for the validation of steam sterilisation procedures lies with the end user who needs to verify the observations described here using the company's proprietary equipment and procedures.

Data packages have been compiled that demonstrate the physico-chemical and functional suitability of the closures during their 2 year shelf life.

For OmniflexPlus vial closures the following material and closure characteristics were part of the study:

1. Physical properties (hardness and compression set)
2. Functional properties (penetration force, fragmentation, self-sealing and sealing capacity as per Pharm. Eur. 3.2.9 / USP <381>)
3. Chemical properties (USP <381> and Pharm. Eur. 3.2.9 compliance)
4. Biological properties (USP <87> compliance)
5. Performance characteristics (insertion behaviour in vials and clumping tendency)

Results for OmniflexPlus vial closures are summarized in a separated Datwyler Customer Service Report. For more details on this study, please contact your Datwyler Sales Representative.

14 Handling recommendations

It is recommended that OmniflexPlus closures be purchased from Datwyler as a Ready-for-Sterilization (RfS) product in RfS bags (polyethylene/Tyvek bags) or as Ready-To-Use (RTU) product in RTU bags (triple layer PE/EVOH/PE bags).

RfS and RTU closures are supplied already washed and finally rinsed with WFI water to reduce particulate, endotoxin and bioburden levels, and dried to remove surface moisture.

The RfS closures can then be steam sterilized and dried by the user, thus requiring no additional handling of the closures themselves.

RTU closures have already been rendered sterile at SAL = 10^{-6} by gamma irradiation.

If OmniflexPlus closures are purchased in bulk form (non-RfS and non-RTU), therefore requiring processing by the user, the following processing conditions are recommended:

Washing :

The use of detergents or other chemicals is not necessary and should be avoided. If operational procedures include a closure pretreatment, a simple rinse with WFI is sufficient.

Sterilization :

The preferred method for sterilization of OmniflexPlus closures is in flexible, breathable containers, i.e. RfS bags. Other containers are acceptable, provided that steam penetration and moisture transmission rates are high. In order to avoid any risk on stopper clumping and potential effects on the coating, containers which cause the closures to remain in pooled water during the cool down phase should be avoided.

Steam sterilization at 121° C for 30 to 60 minutes is recommended. Sterilization at higher temperatures e.g. 134°C should be avoided. Multiple steam sterilization at 121°C is recommended to be limited to twofold sterilization.

OmniflexPlus closures are not to be sterilized by dry heat.

In case of gamma irradiation by the user, product suitability at maximum dose shall be demonstrated.

Drying:

Closures which will be used on aqueous products may be dried in the autoclave by reducing the pressure while maintaining heat. The closures should be removed from the autoclave as soon as the cycle is completed.

Closures which need to be dried more thoroughly, should be dried in an oven, as quickly as possible after sterilization, at a temperature not exceeding 110 °C. Drying in flexible breathable containers is recommended to hasten the drying process. Drying time will depend on container design, closure design, the number of closures in the container, and the required final moisture level. It should be determined experimentally.

In case of sterilization and drying in RfS bags, the latter should not be stacked while the closures are still warm.

Processing:

Before processing on filling lines, including vibratory bowls, OmniflexPlus stoppers must be sufficiently dried. On rare occasions, processing of large amounts of OmniflexPlus stoppers, that were insufficiently dried after steam sterilization, was found to lead to deposition on certain parts of the filling line of a material that is related to the OmniflexPlus coating.(Residual moisture for the stoppers in question was determined to be at 0.7 %).

After further drying of stoppers of the same batch, no such objectionable conditions were met anymore.

15 Available designs

OmniflexPlus closures are available in various stopper designs. OmniflexPlus closures are put forward for use in serum applications (13 mm and 20 mm injection and 32 mm infusion stopper geometries).

The product drawings of a non-exhaustive list of available product designs can be found in attachment. To check on the availability of other product designs, please contact your Datwyler Sales Representative.

For lyophilization stopper geometries, Datwyler at present recommends use of its third generation coated closures that are marketed under the name Omniflex3G. Omniflex3G was developed when OmniflexPlus was already for a long time in the market. Omniflex3G offers enhanced robustness in terms of container closure integrity *before vial capping*, which is especially appreciated in the case of freeze-drying closures.

16 References

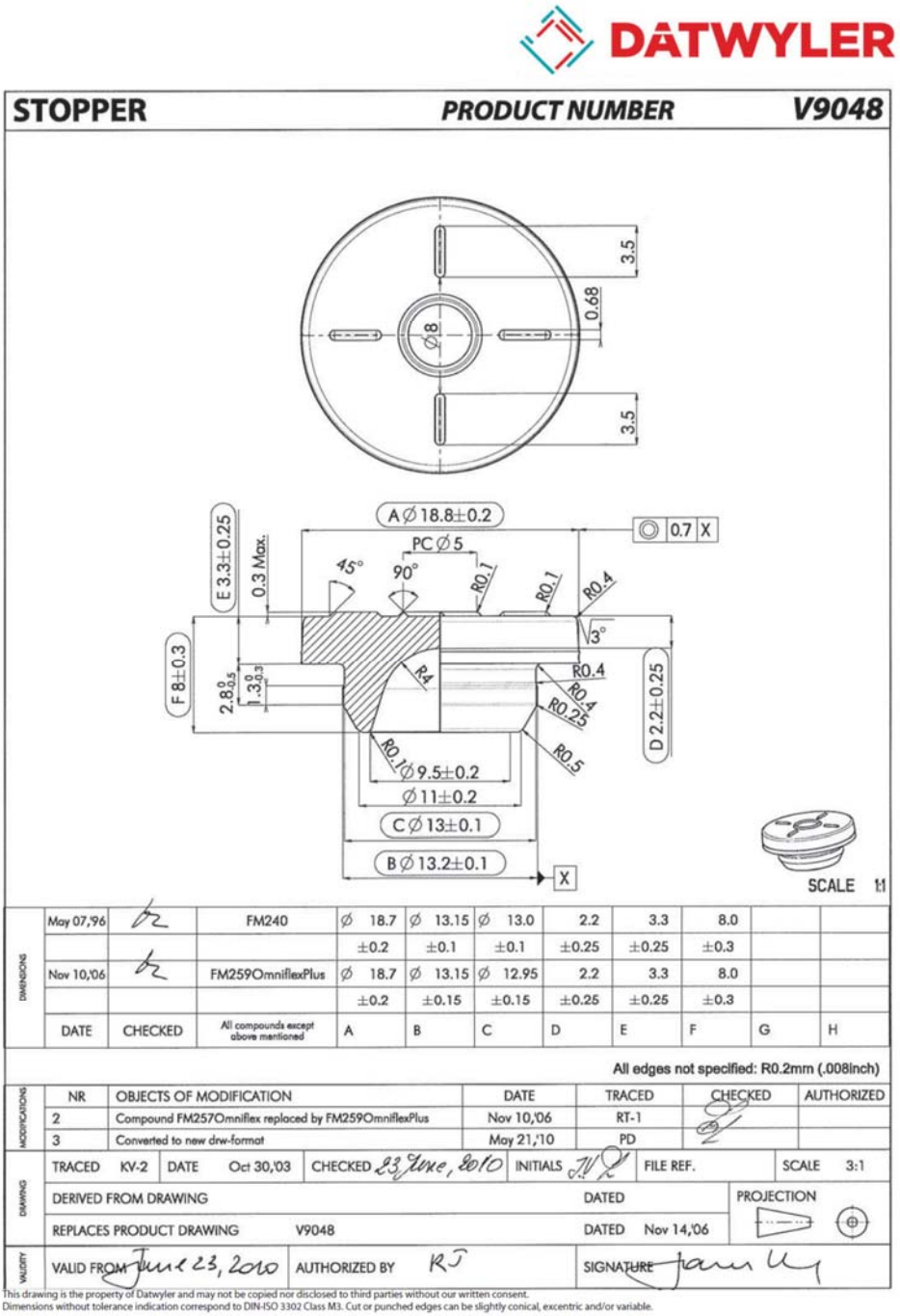
1. *ISO 37 "Rubber, vulcanised or thermoplastic - Determination of tensile stress-strain properties"*
2. *ISO 815 "Rubber vulcanised or thermoplastic - Determination of compression set at ambient, elevated or low temperatures"*
3. *ISO 2230 "Rubber Products – Guideline for storage"*
4. *ISO 2781 "Rubber, vulcanised or thermoplastic – Determination of density"*
5. *ISO 7619-1 "Rubber, vulcanised or thermoplastic – Determination of indentation hardness Part 1 : Durometer method (Shore hardness)"*
6. *ISO 8871-1 "Elastomeric parts for parenterals and for devices for pharmaceutical use – Part 1: Extractables in aqueous autoclavates"*
7. *ISO 8871-3 "Elastomeric parts for parenterals and for devices for pharmaceutical use – Part 3: Determination of released-particle count"*
8. *ISO 8871-5, "Elastomeric parts for parenterals and for devices for pharmaceutical use – Part 5: Functional requirements and testing"*
9. *ASTM D3985 - 05 Standard Test Method for Oxygen Gas Transmission Rate Through Plastic Film and Sheeting Using a Coulometric Sensor*
10. *ASTM F1249 - 06 Standard Test Method for Water Vapor Transmission Rate Through Plastic Film and Sheeting Using a Modulated Infrared Sensor*
11. *Pharm.Eur. 5.2.8. "Minimising the risk of transmitting TSE via medicinal products"*
12. *Pharm.Eur. 3.2.9. "Rubber Closures for Containers for Aqueous Parenteral Preparations, for Powders and for Freeze-Dried Powders."*
13. *USP <381> "Elastomeric Closures for Injection"*
14. *USP <87> "Biological Reactivity Tests, in Vitro"*
15. *USP <88> "Biological Reactivity Tests, in Vivo"*
16. *Pharm. Jap. 7.03 "Test for Rubber Closures for Aqueous Infusions"*
17. *European Directive 94/62/EC on Packaging and Packaging Waste*
18. *Coalition of North Eastern Governors (CONEG) Heavy Metals Legislation*
19. *www.ista.org*
20. *Compound data sheet FM259/0 OmniflexPlus, dated October 14, 2010*
21. *Datwyler Technical Documentation 'OmniflexPlus', Ed. 6, April 2003*
22. *Datwyler report CS0034*
23. *Datwyler report CS0117*
24. *Datwyler R&D report R0243*
25. *Datwyler R&D report R0137*
26. *Datwyler Extractables Report OmniflexPlus*
27. *Datwyler report CS0097*
28. *Datwyler report CS0183*
29. *Datwyler report CS0087*
30. *Datwyler report R0085*
31. *Datwyler report R0027*
32. *Datwyler report CS0052*
33. *Datwyler report R0084*
34. *Datwyler report R0001*
35. *Datwyler report CS0001*
36. *Datwyler report R0210*
37. *Datwyler report CS0112/7*
38. *Datwyler report R0236*

17 History

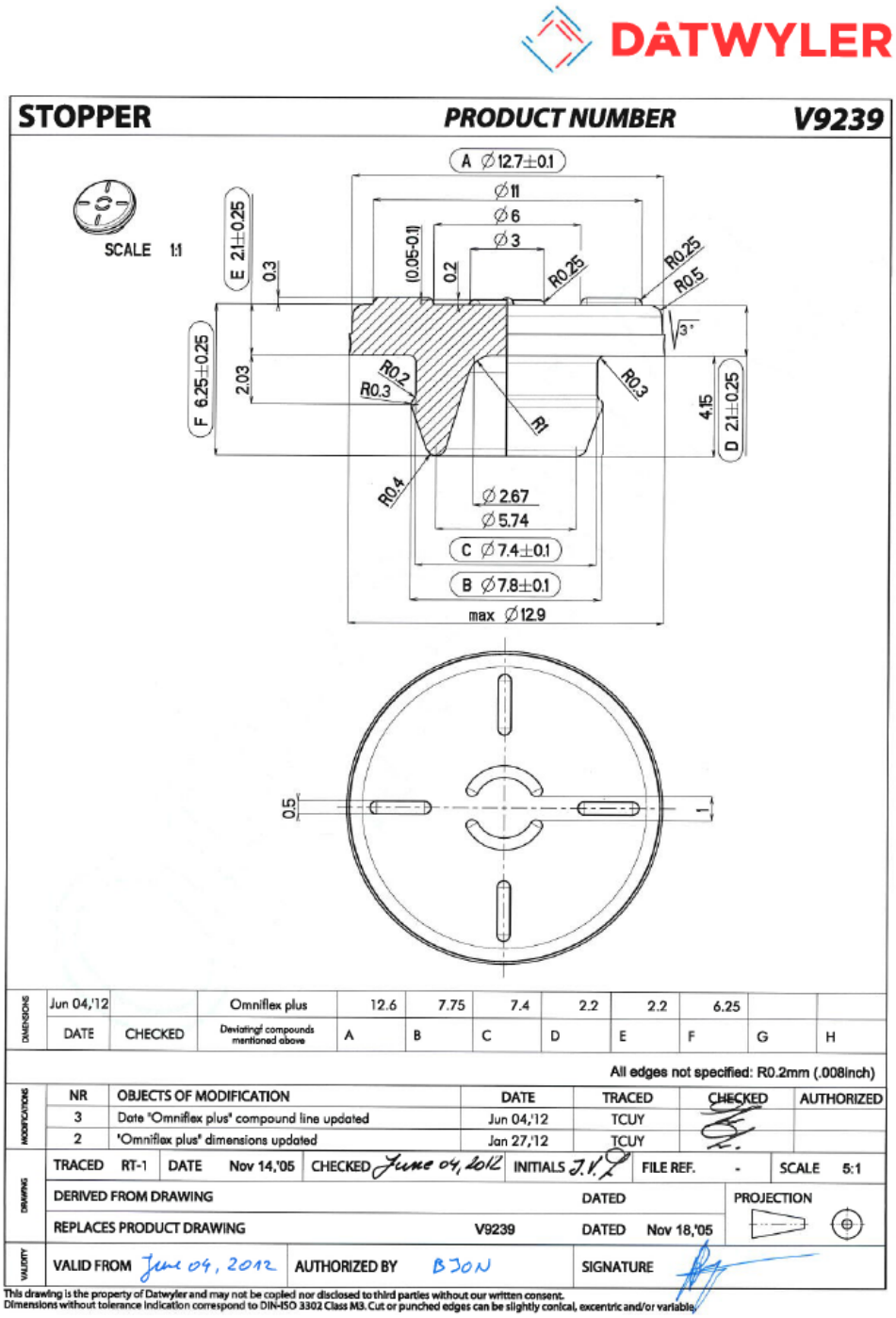
Edition (Issue Date)	*Change (chapter + change)*	*Comment (Rationale)*
7 (February 23, 2012)	Complete overhaul of Ed. 6 (2003)	Update to current state of knowledge and documentation
8 (November 17, 2014)	• Corrected endotoxin load and bioburden expressed per cm² • Exchanged product drawing V9239 for most recent version	• Correction of typos • Update

Attachments

Product drawing V9048



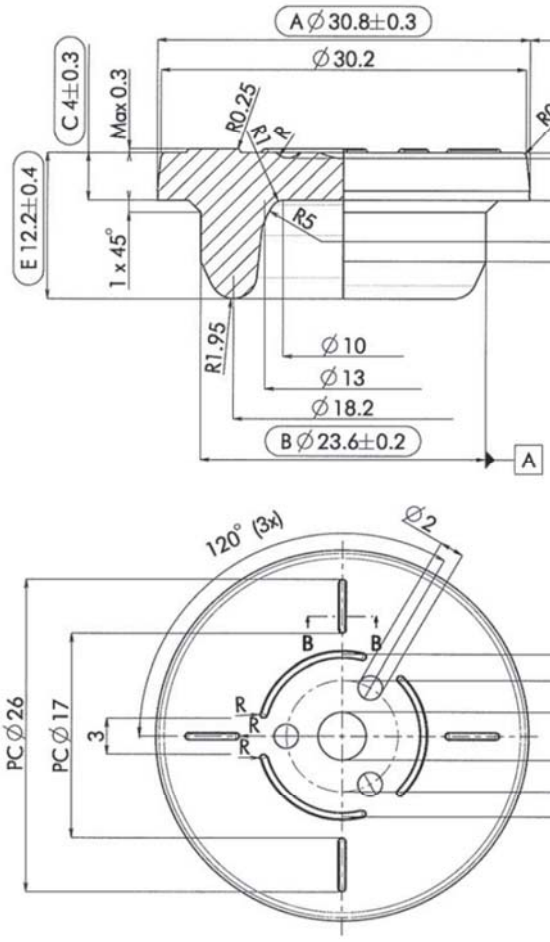
Product drawing V9239

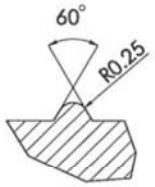


Product drawing V9240

DATWYLER

ISO 32mm INFUSION STOPPER ***PRODUCT NUMBER*** **V9240**





SECTION B-B
SCALE 10:1

SCALE 1:1

	Mar 12,'04		OMNIFLEX / OMNIFLEXPLUS	Ø 30.90	Ø 23.70	4.1	4.0	12.3			
	Jun 28,'01		FM257	Ø 30.90	Ø 23.70	4.0	4.0	12.3			
DIMENSIONS	May 20,'96		FM240	Ø 30.65	Ø 23.50	4.0	4.0	12.2			
	DATE	CHECKED	Deviating compounds	A	B	C	D	E	F	G	H

All edges not specified: R0.2mm (.008inch)

MODIFICATIONS	NR	OBJECTS OF MODIFICATION	DATE	TRACED	CHECKED	AUTHORIZED
	2	Converted to new drw-format	May 21,'10	PD		
	1	Tolerance changed / dimensions for FM257 added	jun 28,'01	KV-2		

DRAWING						
TRACED KV-2	DATE Mar 11,'04	CHECKED 14 June, 2010	INITIALS	FILE REF.	SCALE 2:1	
DERIVED FROM DRAWING				DATED	PROJECTION	
REPLACES PRODUCT DRAWING			V9240	DATED Mar 17,'04		

VALIDITY			
VALID FROM June 23, 2010	AUTHORIZED BY RJ	SIGNATURE	

Dimensions without tolerance indication correspond to DIN-ISO 3302 Class M3. Cut or punched edges can be slightly conical, excentric and/or variable.

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