Functional Evaluation and Characterization of a Newly Developed Silicone Oil-Free Prefillable Syringe System

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ABSTRACT: The functionality of a newly developed silicone oil-free (SOF) syringe system, of which the plunger stopper is coated by a novel coating technology (i-coating™), was assessed. By scanning electron microscopy observations and other analysis, it was confirmed that the plunger stopper surface was uniformly covered with the designed chemical composition. A microflow imaging analysis showed that the SOF system drastically reduced both silicone oil (SO) doplets and oil-induced aggregations in a model protein formulation, whereas a large number of subvisible particles and protein aggregations were formed when a SO system was used. Satisfactory container closure integrity (CCI) was confirmed by means of dye and microorganism penetration studies. Furthermore, no significant difference between the break loose and gliding forces was observed in the former, and stability studies revealed that the SOF system could perfectly show the aging independence in break loose force observed in the SO system. The results suggest that the introduced novel SOF system has a great potential and represents an alternative that can achieve very low subvisible particles, secure CCI, and the absence of a break loose force. In particular, no risk of SO-induced aggregation can bring additional value in the highly sensitive biotech drug market. © 2014 Wiley Periodicals, Inc. and the American Pharmacists Association J Pharm Sci 103:1520–1528, 2014

Keywords: protein aggregation; surface chemistry; particle size; stability; imaging analysis; prefilled syringe; physical stability; silicone oil; gliding force; container closure integrity

INTRODUCTION

The recently increased interest in prefilled syringes (PFS) is largely driven by many advantages against traditional ampoules and vials, such as allowing quick and accurate dosing, minimizing dosing errors, reducing the risk of biological contamination, enhanced convenience and ease of use, preventing of overfill, and so on. On increasing number of available biological drugs, the demand for the use of PFS has considerably increased in recent years.

Silicone oil (SO) is widely used as a lubricant in syringes to ensure a smooth gliding behavior of the plunger inside the barrel. Furthermore, siliconization effectively prevents the plunger stoppers from sticking together during the material shipping and manufacturing process. It is also important to minimize the mechanical force when the plunger stoppers are inserted into a barrel using a mechanical stoppering machine. Siliconization is therefore essential to the process capability of the SO-lubricated syringe. However, it is known that the use of SO-coated syringes leads to difficulties in case of some silicone-sensitive drugs, such as protein-based drugs. For example, several reports in the 1980s implicated that the ingress of SO from disposable syringes caused aggregation of human insulin.[1–5] Researchers continue to seek how SO can accelerate the intermolecular interactions of proteins, and stability studies revealed that the oil could negatively affect the chemical and physical stability of the protein formulation, thereby causing a loss of soluble proteins.[6,7] Additional works have been performed to assess the effects of ionic strength and the type of surfactants that can be used to minimize the risk of a SO-induced aggregation.[8–10]

Silicone oil-induced aggregation has now become one of the most frequently discussed subjects in the field of PFS, particularly for developers of highly sensitive biotech drugs.[6,11,12] Once SO detaches from the inner wall of the barrel, oil droplets are formed that contribute to the population of subvisible particles. The levels of visible and subvisible particles have become critical also to the quality of drug products in PFS.[13–15] To mitigate the risk, the reduction of the SO quantity may be one of the options to be considered, but this may not be adequate because siliconization is essential to ensure the functionality of the syringe over the shelf life of the product, and even if the quantity of SO is reduced, the risk of SO-induced aggregation cannot be eliminated completely.

Under such circumstances described above, the demand for the SO-free (SOF) system has been rising, and various technologies have been proposed so far.[16] Most of them are coating technologies in which the surface of the barrel and/or plunger stopper is coated with a lubricious, compatible, and physicochemically inert material. In response to this unmet need, Terumo created an innovative coating technology, which resulted in the successful introduction of an SOF system for polymer-based PFS in 2005 within the Japanese market that is still supplied in the market today.

Through continued research and development, Terumo has introduced the next generation of SOF coating technology called i-coating™ for use on plunger stoppers. By using this

technology, novel SOF system for prefillable syringes can be accomplished.

The purpose of the present study is to characterize the functionality of this novel SOF system by comparing it with current SO system. In this paper, the coated surface was characterized by means of scanning electron microscopic (SEM) observations, FTIR analysis, and X-ray photoelectron spectroscopy (XPS), and the influences on the generation of subvisible particles, container closure integrity (CCI), and break loose gliding forces as important attributes of PFS were assessed.

MATERIALS AND METHODS

Materials

PLAJEX^{TM} syringe [Polymer based prefillable syringe, 1 mL long staked needle (27G)] (SOF syringe system) was provided by Terumo Corporation (Tokyo, Japan), which is a newly developed prefillable syringe with a polymer barrel made of cyclic olefin polymer and a butyl rubber plunger stopper coated by use of the i-coating^{TM} technology (a proprietary coating). The corresponding PLAJEX^{TM} syringe lubricated by SO with an uncoated plunger stopper (SO system for comparison) was also provided by Terumo Corporation. A butyl rubber sheet of 1 mm in thickness was obtained from Asahi Rubber Inc. (Saitama, Japan). Using the i-coating^{TM} technology, the coated butyl rubber sheet was provided by Terumo Corporation. As a reference sample, SO-coated butyl rubber sheet was also provided by Terumo Corporation. L-Asparaginase (Asp) was purchased from Prospec-Tany TechnoGene Ltd. (Rehovot, Israel); human IgG was purchased from Equitech-Bio Inc. (Kerrville, Texas, USA); albumin from bovine serum (BSA) and lysozyme from chicken egg white (Lyso) were purchased from Sigma–Aldrich Company LLC., (St. Louis, MO, USA), respectively. Triptic soy agar and triptic soy broth (TSB) were purchased from Becton, Dickinson and Company (Franklin lakes, NJ, USA). Isotonic sodium chloride solution and commercially available saline solution of Japanese Pharmacopoeia grade (JP grade) were obtained from Terumo Corporation. Water for injection (20 mL) was JP grade and purchased from Otsuka Pharmaceutical Company Ltd., (Tokyo, Japan). Crystal violet and all buffer salts (sodium phosphate monobasic, sodium phosphate dibasic, and sodium chloride) were purchased from Kanto Chemical Corporation (Tokyo, Japan). All other chemicals and reagents were of analytical grade.

Scanning Electron Microscopy

Scanning electron microscopic observations were performed using a microscope model KEYENCE VE8800 (Tokyo, Japan) to assess the integrity of the coating layer of the plunger stopper. The surface and the cross-section of the coated and uncoated plunger stoppers were observed with magnifications of ×300 or ×1000. To produce a cross-section, a sample was cut vertically by a razor. After platinum was sputtered, the top surfaces and cross-sections of the samples were observed at an acceleration voltage of 1.7 kV.

Fourier Transform-Infrared Spectroscopy

The top surface of the uncoated and coated plunger stopper was analyzed using an FTIR spectrometer (Spectrum100; PerkinElmer, Wellesley, Massachusetts) equipped with a universal attenuated total reflectance (ATR) sampling accessory.

ATR–FTIR spectra were recorded at a resolution of 4 cm^{-1} and 16 scans per specimen were averaged in a wavenumber range from 650 to 4000 cm^{-1}.

X-Ray Photoelectron Spectroscopy

The top surface of the coated plunger stopper was analyzed by means of XPS (Axis-NOVA, Kratos, Manchester, UK) using monochromated Al-Kα (1486.6 eV) X-ray radiation. The X-ray tube was operated at 15 kV and 20 mA. The take-off angle between the sample plane and the axis of the analyzer was 90°. A pass energy of 80 eV was chosen for the acquisition of a survey spectrum (0–1000 eV). The pressure was maintained below 10^{-8} Pa during the measurements.

Protein Aggregation Study

Ten thousand units of Asp were dissolved in 2 mL of water for injection (JP grade). After complete dissolution, the syringes were filled with 1.0 mL of the Asp solution and stopped with plunger stoppers leaving a predetermined headspace of 0.4 mL. The syringes were also filled with 1.0 mL of pure water for injection as reference for the protein aggregation study. After filling, the syringes were shaken in a shaker (SR-I; TAITEC Company Ltd., Saitama, Japan) for 30 min at 250 rpm at room temperature. Immediately after shaking, aliquots of the sample solution were drawn out through a needle by actuation of the plunger stopper and analyzed for particle concentration by micro-flow imaging (MFI). DPA4200 (Brightwell Technologies Inc., Ottawa, Canada) was used for the measurement of subvisible particles. The number of subvisible particles whose equivalent circular diameter is larger than 5 μm was analyzed in this study. Moreover, to obtain information about particle shape, an aspect ratio (AR) filter was employed to distinguish circular (AR ≥ 0.85) and noncircular particles (AR < 0.85). Here, particles having AR ≥ 0.85 and AR < 0.85 are regarded as SO droplets and SO-induced protein aggregates, respectively, according to previous studies.[17,18] In addition, IgG, BSA, and Lyso were dissolved at 1 mg/mL in a buffer (10 mM Na$_2$HPO$_4$, 130 mM NaCl, pH 7.2) and used.[6] Subvisible particles of these protein solutions were also measured in the same manner as described above.

Dye Penetration Study

To assess the CCI of the syringes, a dye penetration study was conducted. One milliliter of water was filled into the syringes and the headspace of the syringes was adjusted to predetermined distances (0.5, 0.75, 1.0, and 1.35 mm) by pushing the plunger stopper down. A small amount (0.15 mL) of 1.0% crystal violet aqueous solution was then placed into the space between the plunger stopper and the flange. The sample syringes were vertically placed in a decompressed chamber (adjusted at −19.6 kPa with respect to normal atmospheric pressure). After 2 h, the water in the syringes was carefully drawn out through a needle and photometrically analyzed to determine the dye concentration using a spectrophotometer SHIMADZU UV-2450 (Kyoto, Japan).

Microorganism Penetration Study

The study was performed according to the method proposed by the pharmaceutical inspection convention.[19] In addition to the intact PLAJEX^{TM} syringe, a PLAJEX^{TM} syringe with a pinhole (size of about 8 μm) on the pathway of the barrel was

used as a positive control. All the syringe samples were sterilized in an autoclave. The TSB culture medium was filled into the sterilized syringes by aseptic manipulation. All the samples were cultured at $31 \pm 1°C$ for 7 days. After confirmation of sterility, they were immersed into a bacterial broth (10^6 cfu/mL) in a pressurized chamber ($+0.02$ MPa above normal atmospheric pressure) for 30 min and, then, were taken and the barrel surfaces were washed by water for injection and 70% ethanol. The pinhole of the positive control was closed by glue. After that, the samples were incubated at $31 \pm 1°C$ for 14 days. The occurrence of microbial growth was judged by visual observation.

Dynamic Friction Coefficient Study

The dynamic friction coefficient was analyzed using a continuous-loading surface property tester (TRIBOGEAR; Shinto Scientific Company Ltd., Tokyo, Japan). A steel ball with 10 mm in diameter was put on the surface of the sample sheet (40×40 mm^2 in size) and rolled on the surface along a distance of 20 mm at a speed of 3.0 mm/s at room temperature to measure the dynamic friction coefficient. In accordance with previous studies, the amount of SO on the sheet was adjusted such that it was equivalent to the amount that is realistically present in a prefillable syringe (0.7 mg/syringe).[16,20,21]

Gliding Force Measurement

Gliding forces were measured using a universal testing machine (EZtest; SHIMADZU corporation, Kyoto, Japan). Each

syringe (the SO and the SOF system) was filled with 1.2 mL of water for injection. Then, the distal end of the syringe and the proximal end of the plunger were attached to the fixing portions of the tester. The plunger was moved downward at room temperature and the force required for gliding the plunger stopper and plunger were recorded. The stroke distance and the crosshead speed were set at 30 mm and 200 mm/min, respectively. The maximum gliding force between 0 and 5 mm of stroke distance was defined as "break loose force."

For the stability studies, the water-filled syringes (the SO and the SOF system) were stored at 4°C, 25°C, and 40°C for predetermined periods of time (0, 2, 4, 6, 8, 12, and 24 weeks). The break loose force was determined for each sample syringe.

RESULTS AND DISCUSSION

SEM Observation

The plunger stoppers coated by i-coating™ were observed by SEM to verify the integrity of the coating. The surface and cross-sectional images of uncoated plunger stoppers were relatively rough (Figs. 1a and 1b), whereas a quite smooth surface was observed in case of the coated plunger stopper (Fig. 1c). In addition, as shown in Figure 1d, the coated layer was quite uniform with a thickness of about 5–10 µm. By this SEM observation, it was confirmed that plunger stoppers provided with the SOF system are uniformly coated without any defects by means of i-coating™.
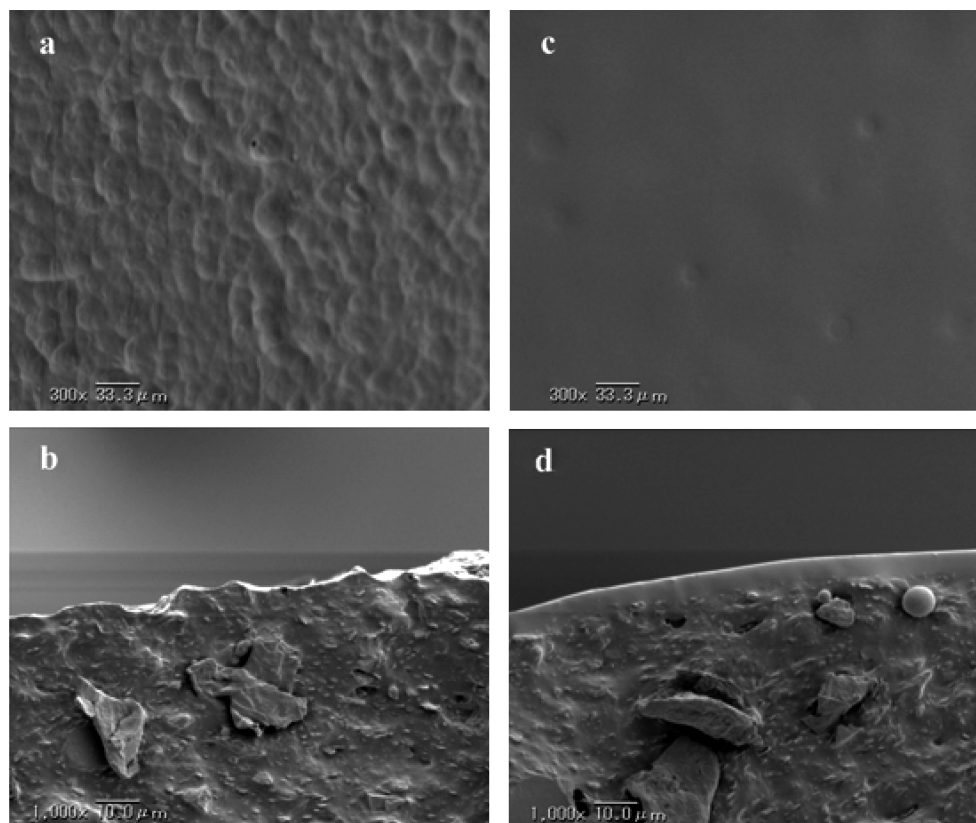

Figure 1. SEM micrographs of the surface and cross-section of the plunger stoppers coated by i-coating™. (a) Top surface of an uncoated plunger stopper ($\times$300), (b) cross-section of an uncoated plunger stopper ($\times$1000), (c) top surface of a coated plunger stopper ($\times$300), and (d) cross-section of a coated plunger stopper ($\times$1000).
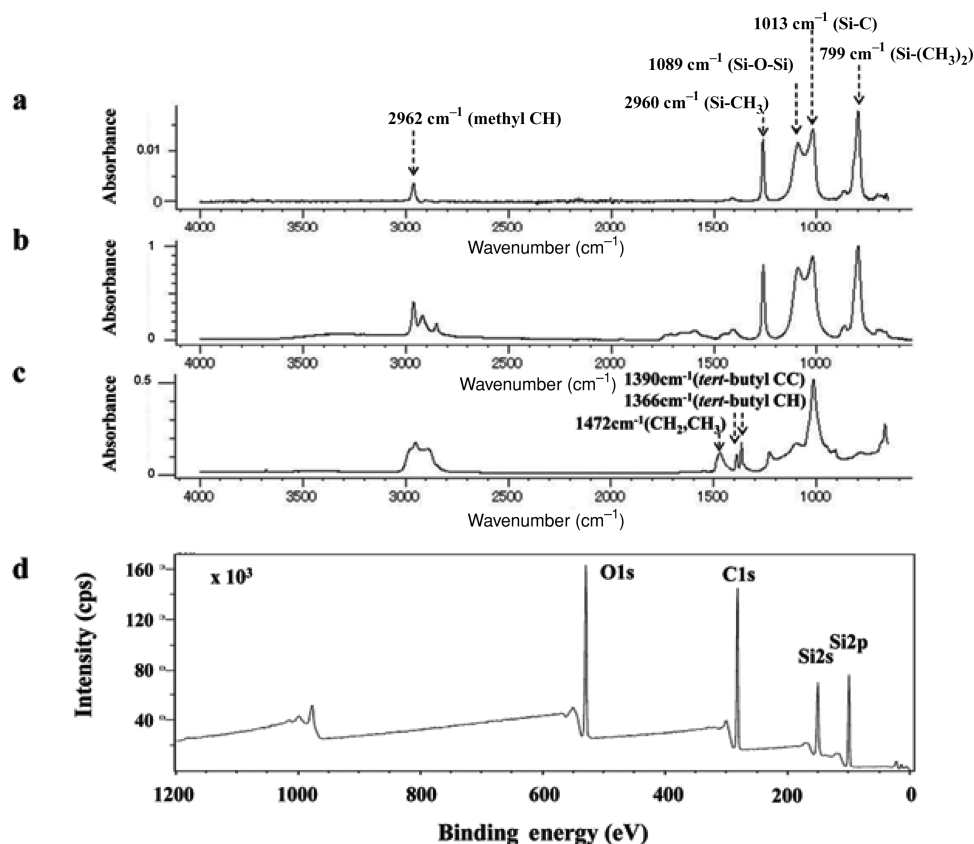
Figure 2. FTIR spectrum of (a) the surface of the coated plunger stopper, (b) reference spectrum of silicone, and (c) the surface of the uncoated plunger stopper. The peaks between (a) and (b) at 2962 cm^{-1} (methyl CH), 1260 cm^{-1} (Si–CH$_3$), 1089 cm^{-1} (Si–O–Si), 1013 cm^{-1} (Si–C), and 799 cm^{-1} [Si–(CH$_3$)$_2$] coincided well with those of the reference spectrum. (d) XPS survey spectrum, measured at the top surface of the coated plunger stopper.

FTIR and XPS Analysis

Figure 2a shows a FTIR spectrum of the surface of a coated plunger stopper. The peaks observed around 2962 cm^{-1} (methyl CH), 1260 cm^{-1} (Si–CH$_3$), 1089 cm^{-1} (Si–O–Si), 1013 cm^{-1} (Si–C), and 799 cm^{-1} [Si–(CH$_3$)$_2$] well coincided with those of the FTIR reference spectrum (Fig. 2b), clearly indicating that the surface layer of the plunger stopper was formed by the silicone-based resin, as intended. As a control, Figure 2c shows the FTIR spectrum of the surface of an uncoated plunger stopper. As seen in Figure 2c, butyl rubber-related peaks were observed around 1472 cm^{-1} (CH$_2$, CH$_3$), 1390 cm^{-1} (*tert*-butyl CC), and 1366 cm^{-1} (*tert*-butyl CH). In contrast, these distinctive peaks were not observed in Figure 2a. This FTIR spectra comparison also helped confirm that the plunger stoppers equipped in the SOF system were uniformly coated by silicone-based resin without exposure of the uncoated butyl rubber surface.

Figure 2d shows the XPS survey spectrum of the top surface of the coated plunger stopper. As evident, signals corresponding to O1s, C1s, Si2s, and Si2p were observed. The quantitative ratio of these elements was found to be C(1s):O(1s):Si(2p) = 1.00:0.44:0.46, quite close to the theoretical ratio (1.00:0.50:0.50) calculated from the composition of the silicone-based resin of i-coating^{TM}. Thus, FTIR and XPS analysis showed in mutual agreement that the surface of the coated plunger stopper was certainly covered by silicone-based resin employing the i-coating^{TM} technology.

Protein Aggregation

A comparative study was conducted to assess the difference in protein aggregation between the SO and the SOF system by means of a MFI technique, according to previous studies.[20,21] In this study, Asp aqueous solution (Asp sol.), IgG aqueous solution (IgG sol.), BSA aqueous solution (BSA sol.), and Lyso aqueous solution (Lyso sol.) were used as silicone-sensitive model protein solutions, and the sample syringes were placed under shaking condition. Another set of syringes was filled with water for injection (JP) as reference. The number of particles with different shape (AR $\geq$ 0.85) and (AR $<$ 0.85) are compared in Figures 3a and 3b, respectively.

As seen in Figure 3a, in case of water-filled syringes, a remarkable number of circularly shaped subvisible particles (AR $\geq$ 0.85, regarded as oil droplets) was detected in the SO system (average 963/mL), whereas much less or even a negligible amount was found in the SOF system (2/mL), suggesting that the proposed SOF system contributes a significantly smaller number ($p < 0.001$, $n = 10$) of subvisible particles than the SO system. It is known that subvisible particles could be formed by the ingress of SO to the medium from the siliconized plunger stopper or barrel.[6,11,12] The observed phenomenon, therefore, may be caused by the same reason. When protein formulation was filled in syringes (Asp sol.), the number of subvisible particles (AR $\geq$ 0.85) drastically increased in the SO system (average 12,099/mL), whereas almost no change was observed in the SOF
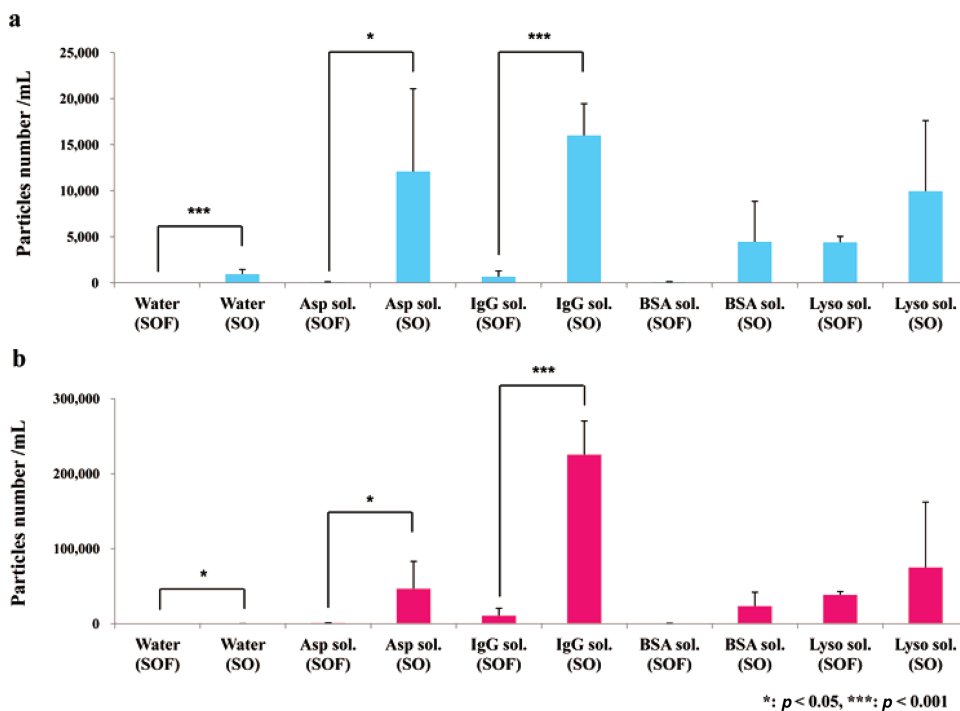
Figure 3. Comparison of the numbers of subvisible particles generated after filling water for injection (water), model protein solutions such as asparaginase (Asp sol.), human IgG sol., bovine serum albumin (BSA sol.), and lysozyme (Lyso sol.) into the SO and the SOF system, determined by MFI analysis. (a) AR < 0.85, (b) AR < 0.85. Data represent the mean values and corresponding standard deviations ($n = 10$ for water, $n = 5$ for model proteins).

system (average 73/mL), suggesting that the protein solution may accelerate the ingress of SO by some interfacial interaction or, that the ingressed oil subsequently forms protein–silicone-oil aggregates ($p < 0.05$, $n = 5$). The same tendencies were also observed in other model proteins (IgG sol., BSA sol., and Lyso sol.).

Figure 3b presents a comparison of the measurement result for noncircular-shaped particles (AR < 0.85, regarded as protein aggregates) between the SO and the SOF system. Analogous results were obtained in the two cases of water-filled and protein-solution-filled syringes (Asp sol.). Namely, when the SO system was used for protein formulation, an extremely large number of particles was formed (average 46,659/mL), whereas significantly fewer were generated in the SOF system (average 897/mL) ($p < 0.05$, $n = 5$). From the results of a previous study, BSA was believed to be a positive control as it showed the most dramatic SO-induced aggregation at pH 7.2.[6] On the other hand, Lyso was expected to be a negative control as no particular change was observed at pH 7.2. Because of the large error bar, we cannot discuss all of the details of this result. However, BSA showed a SO-induced aggregation even though the t value was >0.05 (p value was 0.051 in case of AR < 0.85) and Lyso showed a lesser impact of SO (p value was 0.40 in case of AR < 0.85). In the Lyso sample, relatively higher particle counts were observed even in the SOF system. To obtain further information, an additional study was conducted with noncoated plunger stopper and syringe. As a result, after shaking samples, the particle counts increased even in the non-SO condition (data not shown). This result suggests that the high particle counts were not caused by SO but rather physical stress itself. Overall, the same tendencies were observed in all model proteins. Therefore, we assume that it can be minimized through a reduction

in the content of SO in protein pharmaceutical formulations. It is interesting that a large number of circular particles (AR ≥ 0.85) was observed in the water filled SO system (average 963/mL), whereas the number of noncircular particles (AR < 0.85) was quite small (average 2/mL).

The observed decrease also suggests that SO droplets could have the potential to induce protein aggregation in the various protein solutions.

Although the detailed mechanism of the formation of protein aggregations is still obscure, all the results clearly prove that SO causes the formation of subvisible particles and, hence, that the SOF system based on the i-coating™ technology is superior to the SO system in terms of avoiding the formation of subvisible particles in the syringe.

Container Closure Integrity

The CCI of the SOF system was tested by dye and microbial penetration studies. Figure 4 schematically shows the configuration of the sample syringe used for this study, and the results of the dye penetration test using 1% crystal violet as marker. The syringes with various headspaces were tested to examine the influence of internal pressure. As listed in the table, all samples showed very low absorbance (lower than the detection limit), irrespective of the volume of the headspace, meaning that the penetrated dye amount was almost negligible. Actually, the calculated concentration of crystal violet in the inner, water-filled volume was less than 10^{-5} times that of the outer solution. This result assures a sufficient CCI of the SOF system.

The results of a microorganism penetration study are shown in Figure 5. After 14 days of incubation, the TSB culture medium, filled in the SOF syringes and immersed in a bacterial
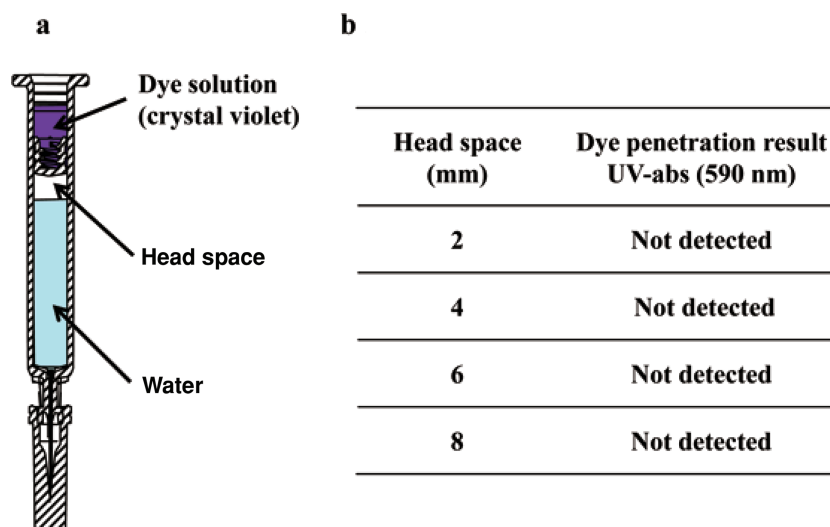
Figure 4. Experimental set up and results of the dye penetration study. (a) Schematic drawing of the configuration of sample syringe, (b) dye concentration (absorbance at 590 nm) penetrated into inner volume across various headspaces. Data represent mean values and corresponding standard deviations ($n = 10$).
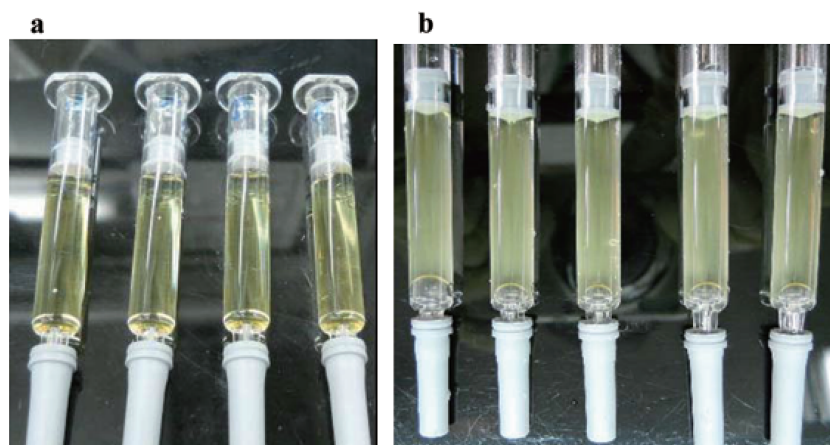

Figure 5. Microorganism penetration study: the syringes filled with TSB culture medium were cultured for 14 days after the immersing treatment into a bacterial broth (10^6 cfu/mL). (a) The SOF system showing all four investigated syringes. The inner solution remained clear without any visual change. (b) Positive control (the SOF syringes with a pinhole on the barrel) of all five investigated syringes. The inner solution became considerably turbid by the invasion and growth of microorganisms.

broth (10^6 cfu/mL), did not show any particular changes in its appearance and the solution inside of the syringes remained clear (Fig. 5a). By contrast, pin-holed SOF syringes (positive control) showed a considerable change in that the medium became turbid (Fig. 5b).

The judgments for the occurrence of microbial growth were 0/4 for the SOF syringe and 5/5 for its positive control. This result indicates that the SOF system did not allow the penetration of microorganisms.

Dynamic Friction-Coefficient Analysis

In order to assess quantitatively the lubricious properties of coated plunger stoppers, the dynamic friction coefficient was determined using a continuous-loading surface property tester. Butyl rubber sheets, pristine, coated by SO and coated by the i-coating™ technology, were used as model sam-

ples replacing the plunger stoppers used in the SO and the SOF system, respectively, because of their identical surface composition.

Figure 6 presents the comparison of the dynamic friction coefficients determined for the three materials investigated: uncoated butyl rubber sheet, SO-coated butyl rubber sheet, and coated butyl rubber sheet. The purpose of this study was to measure the smoothness of the solid surface property. As evident from Figure 6, the dynamic friction coefficient of the coated butyl rubber sheet was 10 times lower than that of the uncoated butyl rubber sheet ($p < 0.001$, $n = 9$). Observation using SEM (Fig. 1) revealed that the surface of the coated plunger stopper was much smoother than that of the uncoated one. Therefore, it is assumed that the obtained result clearly showed the degree of surface smoothness between uncoated butyl rubber and the sheet coated using the i-coating™ technology.
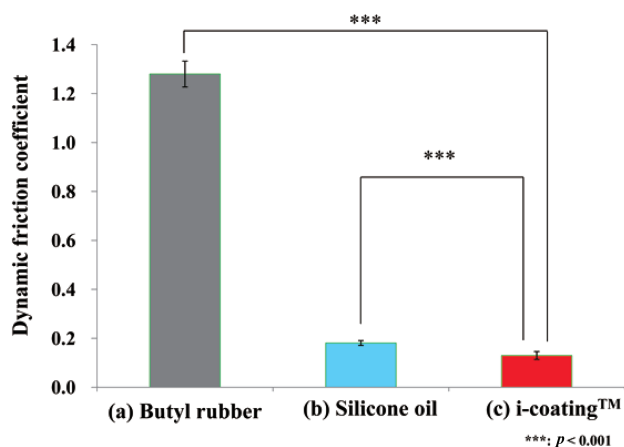
(*n* = 9)	(a) Butyl rubber	(b) Silicone oil	(c) i-coating™
Means	1.28	0.18	0.13
SD	0.0530	0.0098	0.0160

Figure 6. Comparison of the dynamic friction coefficient of (a) a pristine butyl rubber surface, (b) a butyl rubber surface coated by SO, and (c) a butyl rubber surface coated by i-coating™. Each sample sheet (40×40 mm², 1 mm in thickness) was analyzed for the dynamic friction coefficient using a continuous loading surface property tester. Data represent mean values and corresponding standard deviations ($n = 9$).

Surprisingly, it is notable that the dynamic friction coefficient of the sheet coated using the i-coating™ technology was lower than that of the SO-coated butyl rubber sheet ($p < 0.001$, $n = 9$). As mentioned above, this study measured the smoothness of the solid surface property. However, according to a fluid lubricant coating such as SO, it would be difficult to predict the difference between coated butyl rubber sheets with solid lubrication and that of the SO-coated butyl rubber sheet with fluid lubrication as the SO on the surface is easily moved when force is applied. In this measurement, a steel ball was placed vertically on the sheet surface and pulled back horizontally to measure the surface friction. However, in cases of plunger stopper movement within the syringe, a concentric force was generated by the plunger stopper expansion.

Among those differences, it is thought these conclusions have been obtained only by comparing the material physical properties, that is, the apparent dynamic friction coefficients, of the samples. They may not be completely representative of the findings obtained using actual plunger stoppers. Although the value of this study is the ability to simply obtain differences as quantitative values, these results strongly indicate that the application of i-coating™ to a butyl rubber surface drastically decreases the friction resistance of the surface without using SO.

Gliding Force Measurement

A comparative study was conducted to reveal the difference in the gliding force between the SO and the SOF system. To simulate the condition under which syringes are actually used, the specimens were stored at 40°C for 12 weeks in advance. The profiles of the gliding force of the SO and the SOF system are shown in Figure 7.

As seen in Figure 7, both syringe systems provided different gliding force profiles. The SO system showed a maximum
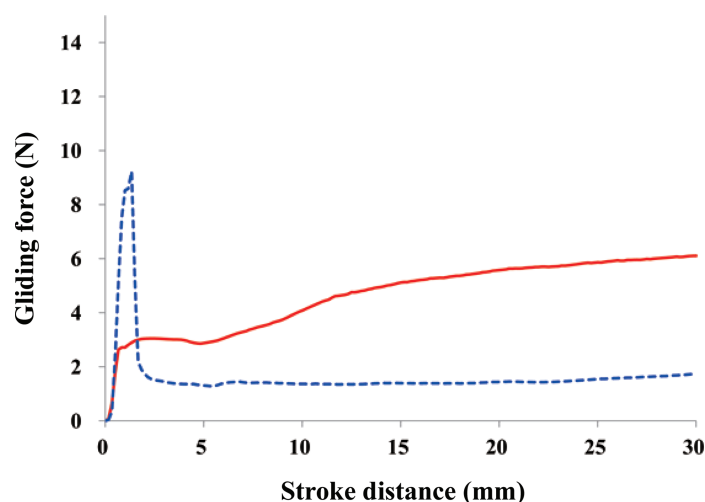

Figure 7. Comparison of the gliding force profiles between the SO system (dotted line) and the SOF system (solid line). The sample syringes were measured after storage for 12 weeks at 40°C, and the data were corrected at a stroke of 200 mm/min.

gliding force of 9 N in the form of a sharp peak at a stroke distance of 3 mm (break loose). The gliding force subsequently reduced to a lower level of about 2 N. On the contrary, no significant difference between the break loose and gliding force was observed in the SOF system. After the initial rise to about 3 N, the gliding force gradually increased to the maximum value of 6 N at a stroke distance of 30 mm.

The observed difference in the gliding force profile is thought to originate from the difference in the lubrication mechanism of both syringe systems. Namely, in the SO system, as SO is coated onto the surfaces of the barrel and the plunger stopper, the SO on the barrel always provides additional lubricity while the plunger stopper moves down in the barrel. Therefore, the gliding force under steady-state conditions is lower compared with the SOF system. In this case, however, because sufficient SO cannot be supplied at the initial stage, a large gliding force is required as a break loose force until the plunger stopper starts to move. This type of profile is commonly observed in currently available SO system including disposable types of syringes.

On the other hand, the SOF system has a lubricious silicone-based resin that is coated on the plunger stopper instead of SO. Even though the gliding force of the SOF system was a little higher than in the SO system, it is far below the maximum permissible value for actual use. This could be caused by the absence of additional lubricity in the SOF syringes, supplied by SO in the SO system, to reduce the friction force generated during the movement of the plunger stopper. To confirm this, the actual moving plunger stopper shape was observed using a CCD camera. As a result, it was observed that the distance of the two ribs decreased and the width of plunger stopper (between two ribs) increased (data not shown). This indicates that the plunger stopper was compressed and the expansion force increased before the plunger stopper moved. This conclusion is also supported by the observed gradual increase in the gliding force.

In order to examine further differences in the break loose force between the SO and the SOF system, stability studies were performed at 4°C, 25°C, and 40°C for 24 weeks. In this study, the maximum force between 0 and 5 mm of stroke
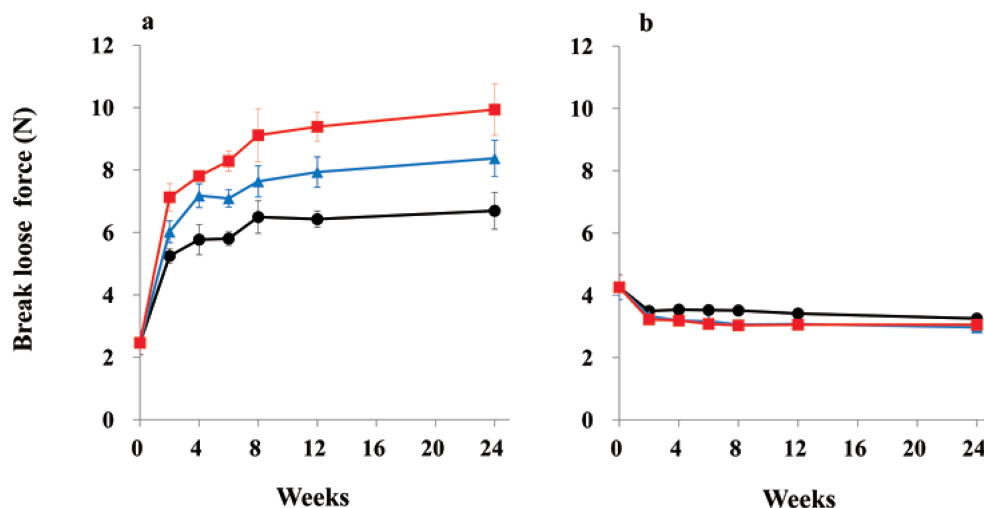
Figure 8.   Break loose force changes of (a) the SO system and (b) the SOF system versus aging time at various temperature; 5°C (●), 25°C (▲), and 40°C (■). Data represent mean values and corresponding standard deviations ($n = 5$).

distance was defined as "break loose force." The results are shown in Figures 8a and 8b.

As seen in Figure 8a, the break loose forces in the SO system gradually increased with the lapse of time and finally reached constant equilibrium values after 8 weeks. These constant values increased on increasing temperature (about 6 N at 5°C, 8 N at 25°C, and 10 N at 40°C). On the other hand, as seen in Figure 8b, no changes were observed in the break loose force of the SOF system, irrespective of storage period and temperature. The constant break loose force value was about 3 N in all investigated cases.

The temperature dependence of the break loose force observed in the SO system could be attributed to the thermal expansion of the plunger stopper. For more details, it is considered that the SO layer located between plunger stopper and barrel is gradually spread out in response to the plunger stopper expansion on aging time, resulting in a thinner SO layer that increases the initial friction (break loose force). The viscosity of the SO may contribute to the observed temperature dependence in various ways. On the other hand, in case of the SOF syringe, in which the plunger stopper is coated chemically with a silicone-based resin, the lubricity of the plunger stopper hardly changes by temperature or aging time, as evidenced by Figure 8b.

CONCLUSIONS

Through a series of comparative studies, it was demonstrated that a newly developed SOF system prevents effectively the formation of subvisible particles including both, SO droplets and oil-induced aggregations. Furthermore, it was confirmed by dye and microorganism penetration studies that the SOF system assures a sufficient CCI. In addition, the SOF system showed no significant difference between the break loose and gliding force and did not change with aging time and temperature. Therefore, the SOF syringes enable to minimize the risk of unexpected rapid release during aspiration and unexpected gliding force change during storage. All these results prove that the newly developed SOF system using the i-coating™

technology has great advantages compared with the current SO system.

In conclusion, the proposed novel SOF system has a great potential and represents a novel alternative that can achieve a very low level of subvisible particles, secure CCI, and the absence of a break loose force. In particular, no risk of SO-induced aggregation and very low level of subvisible particles can bring additional value to the highly sensitive biotech drug market.

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