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Evaluation of CZ-resin vials for packaging protein-based parenteral formulations

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Abstract

Due to the fact that some proteins have a tendency to bind to glass surfaces, plastic CZ-resin vials were evaluated as an alternative material to glass vials for packaging protein-based parenteral formulations. Physico-chemical tests including protein binding, extractable evaluation, oxygen permeation, light transmission and moisture loss were performed. Data show that two proteins (A and B) were found to bind to USP type I glass but not to CZ-resin. The CZ-resin vials passed all USP test specifications for extractables (organic extractable, non-volatile residue and residue on ignition). The oxygen permeation rate (79.06 cm^3 mm/m^2 24 h atm) was consistent with that reported by the vendor (67 cm^3 mm/m^2 24 h atm). The value obtained for light transmission, which was also found to be consistent with that reported by the vendor, shows that these vials offer no protection from light. The average moisture loss from 2 cm^3 vials filled with water was gravimetrically determined to be 0.04 mg/day/vial when the vials were stored at 40 °C/75% relative humidity (RH). Assuming a 1 cm^3 product fill, this corresponds to approximately a 3% loss over a 2-year period. However, moisture loss was found to be negligible at the typical storage condition of 5 °C for protein formulations. The physico-chemical tests indicate that CZ-resin vial is a suitable candidate for packaging parenteral formulations since it shows low moisture loss at typical storage condition of 5 °C, and does not leach out extractables. However, it should not be used for light-sensitive and oxygen-sensitive parenteral formulations. For proteins A and B, the CZ-resin vial is a viable alternate to the use of glass vials since it offered significantly less protein binding. Protein binding in general, should be evaluated on a case by case basis, since it may vary for different proteins and under different formulation conditions.

Keywords: CZ-resin vial; Plastic vial; Protein adsorption; Ethanol extractables; Oxygen permeation

1. Introduction

Most liquid parenteral formulations are packaged in glass vials, which are impermeable to gases. It is well established that some protein drug substances have a tendency to bind significantly to the pharmaceutical container surface. The protein-surface adsorption is complex, involving electrostatic and/or hydrophobic interactions, and is influenced by the physical state of the container surface, the formulation solution environment and the unique structural aspects of the protein. Surface variations in the protein, arising from conformational and compositional differences, make it difficult to make a generalized correlation between the amount of protein adsorbed and its molecular mass or isoelectric point (Burke et al., 1992). Protein loss must, therefore, be measured with the formulated drug substance, at the required dosage strength and fill-volume, using the desired vial and stopper. Formulation overages may then be used to compensate for drug loss.

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The use of overages is not only expensive, but may not be acceptable from a regulatory perspective. Alternate approaches, such as pre-treatment of the vials, incorporation of an inert protein to saturate the glass, or the use of excipients to reduce surface interaction, such as surfactants, carbohydrates, and amino acids, have all been used (Duncan et al., 1995; Suelter and DeLuca, 1983; Wang and Hanson, 1988). Sometimes, attempts to decrease drug loss have had limited success.

An alternative packaging material that demonstrates minimum protein binding is therefore desirable. One such type of material is CZ-resin (Daikyo, Seiko, Ltd.), which is a high molecular weight plastic material. This study presents data that indicate that vials made from this resin showed no adsorption affinity for the proteins studied. These vials must be further examined with respect to potential interactions with the formulation, ease of machinability during production, and physico-chemical characteristics as a primary packaging component.

This paper describes the results of the physico-chemical evaluation of the CZ-resin vial as a potential primary component for the packaging of protein-based, parenteral formulations. The tests performed were protein binding, extractables (organic, non-volatile residue, residue on ignition), light transmission, moisture loss and oxygen permeation. Since these vials are composed of plastic, they are permeable to gases. A previous study (Qadry et al., 1999) indicated that CZ-resin vials may be inappropriate for packaging oxygen-sensitive formulations even in the presence of a nitrogen-filled headspace.

2. Materials and methods

2.1. Materials

CZ-resin vials in the 2 cm^3 size with a capacity of 2.6 cm^3 were obtained from Daikyo, Seiko, Ltd., The West Company, Lionville, PA, USA. These vials were washed with water for injection and sterilized before use. For the adsorption experiments, 2 cm^3 glass vials, made of type I glass, which were supplied by Schott West, were washed with water for injection and depyrogenated. Daikyo 713/B2-40 fluororesin coated stoppers (13 mm) were used for the study. The molecular weights of protein A and protein B used for adsorption experiments were approximately 75 and 20 kDa, respectively.

2.2. Adsorption experiments

Two protein formulations (protein A and protein B) were filtered through a sterile 0.2 μm PVDF filter and aseptically filled into 2 cm^3 vials of CZ-resin and 2 cm^3 glass vials (1 cm^3 fill). Vials were gently rotated in a horizontal position at 5 °C (approximately 4 rpm) to maximize adsorption. The adsorption of these proteins to glass vials is a rapid process. Therefore, the samples were analyzed by reverse phase HPLC method after 1 day. A long-term study was also set up to observe the protein A adsorption to the CZ-resin vial at 5 °C. Samples were taken at 1, 3, 6, 9, 12, 15 and 18 months.

2.3. Extractables testing

The samples were prepared in the following manner:

- The number of vials required to contribute a total internal surface area of 120 cm^2 was calculated, as per the USP Section ⟨661⟩ guidelines.
- Sixteen vials were used for GC/FID organic extractables and 40 vials were used for non-volatile residue/residue on ignition.
- Each vial was filled with ethanol (twice distilled) up to the neck (approximately 2.6 cm^3).
- The vials were placed in a 250 cm^3 glass Erlenmeyer flask and covered with a glass stopper to minimize evaporation. The flask was placed in a preheated oven at 70 °C for 24 h.
- A total of 100 cm^3 of ethanol (twice distilled) contained in a glass Erlenmeyer flask was treated in the same manner to act as the control solution for extractables using gas chromatography with flame ionization detector (GC/FID).
- Another 100 cm^3 of ethanol (twice distilled) contained in a glass Erlenmeyer flask was treated in the same manner to act as the control solution for non-volatile residue and residue on ignition test.

2.3.1. GC/FID organic extractables

A composite solution of ethanol taken from the 16 vials and an equivalent volume taken from the control

solution were both concentrated at room temperature to 5 cm^3 under a stream of nitrogen. A 0.4 μl injection from each sample was analyzed by gas chromatography with flame ionization detector (GC/FID).

2.3.2. Non-volatile residue and residue on ignition

This test was performed according to the USP Section ⟨661⟩ guidelines. A composite solution of ethanol taken from 40 vials was transferred from the CZ vials to a tared crucible and the volatile matter was evaporated on a steam bath. Similarly, an equivalent amount of the control solution was evaporated in a second crucible. The crucibles from the control and the sample ethanol extract were dried at 105 °C for 1 h and their weights were compared quantitatively.

The crucibles containing the non-volatile residue from the CZ vials and the control solution would be used for Residue on Ignition determination, as described in USP ⟨281⟩, if the non-volatile residue obtained is more than 5 mg.

2.4. Light transmission

The diffused light transmission for a vial cut into two along its length was measured according to the USP ⟨661⟩ guidelines. A Cary-1 UV-Vis spectrophotometer equipped with a spectro-sphere was used to measure the light transmission of the CZ material in the region of 290–850 nm.

2.5. Moisture loss

Twenty CZ-resin vials were filled completely with water to represent 100% RH inside the vial. The vials were sealed (crimped) using 13 mm Daikyo 713 stoppers. The vials were stored at a 40 °C/75% RH. Weight loss per vial was recorded on a monthly basis and the average loss per day (mg/vial) was calculated.

Another study was performed where six CZ-resin vials were taken, filled with 1 cm^3 phosphate buffer, sealed (crimped) using Daikyo 713 stoppers and stored at 5 °C. These vials were weighed periodically and the average loss (mg/day/vial) was calculated.

2.6. Oxygen permeation

Oxygen permeation was performed by Mocon Testing Services (Minneapolis, MN) as per the ASTM Method D-3985. Four different vials were tested. The specified test conditions were ambient oxygen (21%) on the outside of the vial at ambient pressure (760 mmHg) and 0% oxygen on the inside of the vial at 23 °C and ambient humidity.

3. Results and discussion

Following were the results obtained for various experiments performed.

3.1. Adsorption experiments

As shown in Table 1, the adsorption of protein A and B to glass vials was compared to CZ-resin vials (plastic). The protein A formulation lost about 16% of its protein while protein B lost 15.2 % when they were rotated in glass vials at 5 °C for 1 day. An insignificant amount of loss was observed in CZ-resin vials (3% for protein A and 5.3% for protein B).

Table 2 shows the change in concentration of protein A with time when stored at 5 °C. As can be seen from Table 2, the concentration of protein A varies in-

Table 1
Comparison of adsorption of protein to glass versus CZ-resin vial at 5 °C after 1 day

Protein type	Initial amount (μg/cm^3)	CZ-resin vial		Glass vial	
		Amount (μg/cm^3)	Loss (%)	Amount (μg/cm^3)	Loss (%)
A	10.0	9.7	3.0	8.4	16.0
B	12.2	11.5	5.3	10.3	15.2

Table 2
Adsorption of protein A to CZ-resin vial at 5 °C over extended shelf life

Time (months)	Concentration of protein A in the vial (μg/cm^3)
Initial	46.1
1	46.0
3	45.9
6	45.5
9	46.3
12	45.5
15	46.0
18	47.0

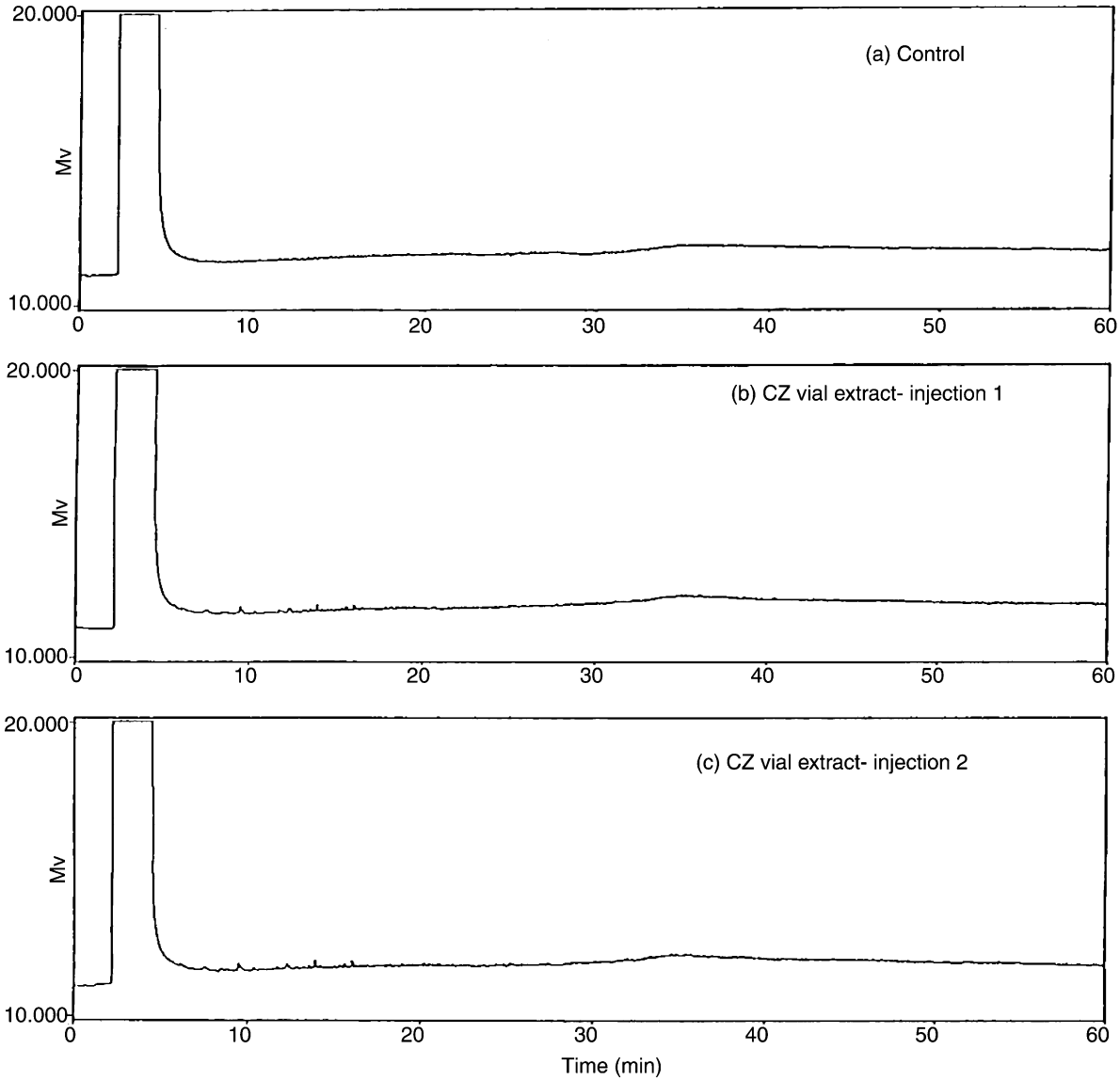

Fig. 1. GC profiles comparing the extractables from CZ-resin vials vs. a glass control.

significantly over an extended shelf life of 18 months when stored at 5 °C. Also, since the rate of moisture loss at 5 °C is negligible (Table 4), moisture loss would have negligible to no effect on the concentration of protein A in the vial at the end of 18 months.

These experiments indicate that CZ-resin has a potential as an alternative material to glass for storing the proteins that adsorb to glass.

3.2. Extractables testing

The gas chromatographic profiles of the vial extract in duplicate and the control solution are shown in Fig. 1. No extractables were detected from CZ vials. The exact material composition of the CZ vial is proprietary information. While many other types of polymers do extract components into alcohol when subject

Table 3
Moisture loss from CZ-resin vials sealed with 13 mm Daikyo 713 stoppers at 40 °C/75% RH

Vial	Rate of moisture loss over 30 days (mg/day/vial)	Rate of moisture loss over 60 days (mg/day/vial)	Rate of moisture loss over 92 days (mg/day/vial)	Rate of moisture loss over 122 days (mg/day/vial)
1	0.038	0.035	0.037	0.038
2	0.040	0.034	0.035	0.039
3	0.036	0.032	0.035	0.038
4	0.044	0.031	0.034	0.037
5	0.046	0.036	0.037	0.039
Average (1–5)	0.041	0.033	0.036	0.038
6		0.031	0.034	0.035
7		0.034	0.036	0.039
8		0.030	0.035	0.036
9		0.029	0.033	0.036
10		0.029	0.034	0.036
Average (1–10)		0.032	0.035	0.037
11			0.036	0.038
12			0.040	0.041
13			0.036	0.040
14			0.039	0.041
15			0.038	0.037
Average (1–15)			0.036	0.038
16				0.037
17				0.037
18				0.038
19				0.039
20				0.036
Overall average	0.041	0.032	0.036	0.038
Overall S.D.	0.004	0.002	0.002	0.002

to 70 °C for 24 h, Fig. 1 demonstrates that material is particularly "clean" and does not leach components. These results are consistent with those reported by the vendor.

Non-volatile residue was not detected. The specification to pass the test as given in USP, the difference in weight between residue and blank not to exceed 15 mg, was met. The test for residue on ignition was not performed since the non-volatile residue was not more than 5 mg.

3.3. Light transmission

The percent transmission at 830 nm (91.5%) was consistent with the value reported in the Daikyo Technical Report (91%). Because these clear vials offer no resistance to light, they should not be used to package light sensitive formulations.

3.4. Moisture loss

The data on moisture loss (for 30, 60, 92 and 122 days) from the CZ vials is shown in Table 3. Vial numbers 1–5 had consistent average weight losses when measured at 30-day intervals up to 122 days. The average moisture loss was consistent between the four groups of vials when measured at 122 days.

While an average loss of 0.04 mg/day/vial may be significant over a 2-year shelf life, these storage conditions are extreme for the protein formulations that would potentially be packaged. Assuming a 1 cm^3 product fill, this corresponds to approximately a 3% loss over a 2-year period. Moisture loss will be lower at typical storage conditions of 5 °C. Therefore, moisture loss was calculated at typical storage condition of 5 °C. The data in Table 4 shows negligible amount of rate of moisture loss at this condition.

Table 4
Moisture loss from CZ-resin vials sealed with 13 mm Daikyo 713 stoppers at 5 °C

Vial number	Rate of moisture loss over 31 days (mg/day/vial)	Rate of moisture loss over 98 days (mg/day/vial)	Rate of moisture loss over 185 days (mg/day/vial)
1	0.000	0.000	0.001
2	0.000	0.000	0.001
3	0.000	0.000	0.001
4	0.000	0.000	0.001
5	0.000	0.000	0.002
6	0.000	0.000	0.002
Average	0.000	0.000	0.001
S.D.	0.000	0.000	0.000

Table 5
Oxygen permeation for CZ vials as expressed in two different units of measure

Vial number	Oxygen permeation rate (cm^3/package 24 h)	Oxygen permeability (cm^3 mm/m^2 24 h atm)
1	0.0122	67.45
2	0.0168	92.88
3	0.0142	78.51
4	0.0140	77.40
Average	0.0143	79.06

3.5. Oxygen permeation

The oxygen permeation rate is shown in Table 5, together with the values converted to the same units as those used by the vendor.

The average value of 79.06 cm^3 mm/m^2 24 h atm was consistent with the 67 cm^3 mm/m^2 24 h atm value reported by the vendor. This value of oxygen permeation through CZ vials determines the potential use of CZ vials to package oxygen-sensitive formulations.

4. Conclusion

The protein binding tests performed on CZ-resin vials and USP type I glass suggest that the CZ-resin vial is a potential candidate for an alternative material to the glass vial because of the low affinity of the proteins to bind to the surface of the CZ-resin vial. Also, the moisture loss test indicates that the percent moisture loss will be lower than 3% for a 1 cm^3 fill volume when the vial is stored at the typical protein storage condition of 5 °C. However, the vial does not offer any resistance to light, and should not be used for light sensitive formulations. Furthermore, a previous study (Qadry et al., 1999) indicates that CZ-resin vials may be inappropriate for packaging oxygen-sensitive formulations even in the presence of a nitrogen-filled headspace. To overcome this problem, CZ-resin vials can be packaged in sealed aluminum pouches and the pouches can be flushed with nitrogen.

References

Burke, C.J., Steadman, B.L., Volkin, D.B., Tsai, P.-K., Bruner, M.W., Middaugh, C.R., 1992. The adsorption of proteins to pharmaceutical container surfaces. Int. J. Pharm. 86, 89–93.

Duncan, M.R., Lee, J.M., Warchol, M.P., 1995. Influence of surfactants upon protein/peptide adsorption to glass and polypropylene. Int. J. Pharm. 120, 179–188.

Qadry, S.S., Roshdy, T.H., Knox, D.E., Phillips, E.M., 1999. Model development for O_2 and N_2 permeation rates through CZ-resin vials. Int. J. Pharm. 188, 173–179.

Suelter, C.H., DeLuca, M., 1983. How to prevent losses of protein by adsorption to glass and plastic. Anal. Biochem. 135, 112–119.

Wang, Y.-C.J., Hanson, M.A., 1988. Parenteral formulations of proteins and peptides: stability and stabilizers. J. Parent. Sci. Technol. 42, S3–S26.