the needle was assessed by pushing a small amount of liquid out of each needle and resealing the assemblies prior to storage. Small volumes (5–20 µL) were withdrawn from each syringe and assayed for trace metal analysis using inductively coupled plasma mass spectrometry (ICP-MS). In addition, stability of the mAb was compared after storage in vials and prefilled syringes. The main conclusions from these studies were that silicone oil could not lower the potential for corrosion, but the stainless steel needle did not cause significant changes in stability of the mAb in the prefilled syringe (Liu et al., unpublished observations).

Potential problems with tungsten in prefilled syringes

Tungsten pins are used to create the hole for placement of the needle in staked needle prefilled syringes. During the processing with these tungsten pins, tungsten oxide vapor deposits in the narrow volume of the syringe where the needle is attached, and tungsten particles can shed during the storage of the prefilled syringes. Proteins can interact with the tungsten particles and oxides resulting in protein aggregation (Swift, Nashed-Samuel, Liu, Narhi, & Davis, 2007). It has been reported that the aggregation of Epoetin® (EPO) in prefilled syringes is linked to the interaction of tungsten and is a likely cause of immunogenicity (Seidl et al., 2012). Investigations were also carried out to determine the root cause of the interaction of tungsten with proteins (Jiang et al., 2009; Liu et al., 2010). The study by Jiang et al. investigated the relationship between tungsten, visible particles, and protein aggregation. Two different model proteins were incubated with extracts from tungsten pins used in the manufacturing of the prefilled syringes and compared to the proteins incubated with different tungsten solutions which included tungstic acid (H_2WO_4), tungsten trioxide (WO_3), sodium tungstate dihydrate ($Na_2WO_4{\cdot}2H_2O$), and sodium polytungstate ($Na_6O_{39}W_{12}$). The extracts from the tungsten pins were more effective in generating soluble protein aggregates than the tungsten solutions. It was also shown that the effect of tungsten on aggregation was greater at pH below 4 and dependent on tungsten species and concentration. One of the model proteins had a large helical content and it was suggested that electrostatic interactions were mainly responsible for the tungsten–protein interactions. Spectroscopic studies using FTIR and CD showed that the protein aggregates were composed of essentially natively folded proteins, which may explain the highly immunogenic response of these aggregates. Aggregates of natively folded protein often mimic the replicative coat protein surface of a virus (termed "virus-like particles") and generate an immune response that results in effective neutralization of properly folded proteins (Rosenberg, 2006).

Studies using a mAb have also shown an interaction with tungsten resulting in precipitation (Bee, Nelson, Freund, Carpenter, & Randolph, 2009). Specifically, tungsten poly anions below pH 6 precipitated the mAb rapidly within seconds. At pH 5 tungsten levels at about 3 ppm were sufficient to precipitate a mAb at low concentration (0.02 mg/mL). It was also shown as for previous model proteins that the native secondary structure of the mAb was retained in the precipitated mAb. Most importantly, it was concluded that a small number of tungsten particles were sufficient to precipitate the mAb at pH 5.