Abstract #M1012

In vitro Integrity of Two Recombinant Human Monoclonal Antibodies after Instantaneous Injection by DosePro® Delivery Technology

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Abstract

PURPOSE: Subcutaneous self-administration by syringe or autoinjector may require an injection time of 10 seconds or longer. This can be difficult for those with limited dexterity (e.g. rheumatoid arthritis patients). Instantaneous injection (< 1/10th second) may improve usability; however, the impact on the integrity of biologics must be understood. Two IgG mAbs (~150kDa MW) were instantaneously injected by DosePro and evaluated for purity, particulates, conformational changes and activity. Also, effects of storage conditions on mAb sensitivity to instantaneous injection were evaluated.

METHODS: DosePro cartridges were filled with mAb formulation and assembled into DosePro samples. Samples were either injected into air or into buffer to minimize air-liquid interface interactions. Samples were analyzed for purity loss (HPLC), particle formation (Micro-Flow Imaging™), conformational changes (near-UV CD), and potency changes (Bio-Assay) compared to vial controls. To test whether storage in DosePro increased mAb sensitivity to the effects of instantaneous injection, filled cartridges were stored at 5°C (long-term storage condition), 25°C, and 40°C with controls. At specific time points, samples were injected into air and were analyzed. Results were compared against manually expelled DosePro cartridges and glass vials. Volumes delivered by DosePro (target: 0.5mL) were measured gravimetrically.

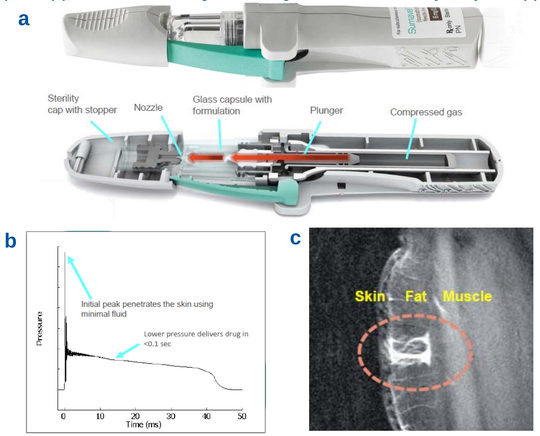
RESULTS: All results were consistent with expected characteristics of the mAb formulations. Instantaneous injection did not induce degradation or aggregation; purity loss was consistent with controls, ranging between 0.8-0.9% after 6 months at 25°C. Particle counts were within normal range of variability for these formulations. Subvisible particles (≥2µm, ≥10µm, and ≥25µm) of both manually expelled and actuated samples were comparable to those of vial controls after 12 months of storage at 5°C. No conformational change was detected by near-UV CD after injection compared to controls at all time points. Potency was ≥90% and similar to controls at all time points. There were no significant differences in stability (injected or manually expelled) versus controls. Delivered volume was 0.499±0.007 mL after 12 months at 5°C.

CONCLUSIONS: All mAb formulations were compatible with the DosePro and displayed the same stability compared to controls. There were no changes in mAb molecular integrity, observable tertiary structure, purity or potency following injection by DosePro technology.

Background

Advances in formulation techniques and technologies have allowed biologics to be formulated at ever higher concentrations, even exceeding 150mg/mL. With these ultra-concentrated formulations, viscosity becomes an important factor, often necessitating either large bore needles or longer injection times. This can be a problem, especially for needle phobic patients or users with manual dexterity limitations, such as those with rheumatoid arthritis. Pressing the plunger rod or holding an autoinjector steady for the required length of time may be painful and/or very difficult. The compressed gas powered DosePro needle-free injection system (Zogenix, Inc.) is capable of delivering highly viscous solutions or suspensions subcutaneously in a fraction of a second. It accomplishes this by forcing the drug product through a laser drilled nozzle, creating a high velocity stream of liquid that pierces the skin (Figure 1). The injection depth is determined by the pressure profile of the compressed gas actuator and the nozzle dimensions[1]. The injection occurs in approximately 45ms, making it ideal for those with limited dexterity or needle phobia[2].

Figure 1: DosePro instantaneous injection delivery system(a), DosePro pressure profile (b), and ultrasound image confirming subcutaneous delivery of injectate (c).



Purpose

- To evaluate the impact of instantaneous injection on the biophysical integrity of two IgG mAbs (~150kDa MW).

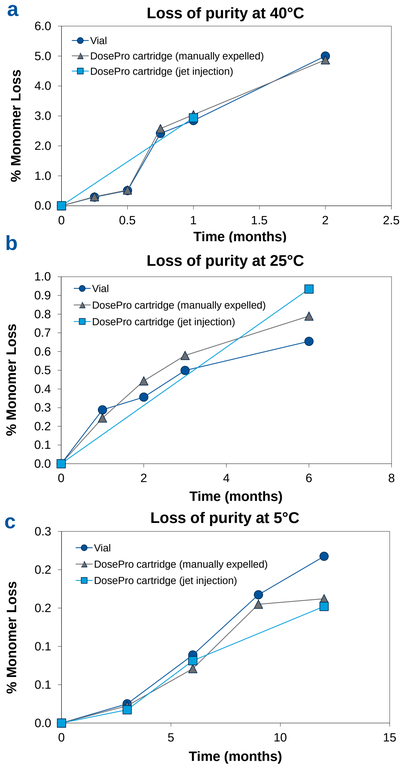
Methods

- DosePro cartridges were filled by hand with two mAb formulations. Vials were hand filled at the same time to serve as controls.
- Filled cartridges were then collected in one of three ways
  - Manually expelled into 15mL conical tube over approximately 5-10 seconds
  - Assembled onto a DosePro compressed gas actuator and instantaneously injected into empty 15mL conical tubes
  - Or, assembled onto a DosePro actuator and instantaneously injected into formulation buffer to minimize air-liquid interfacial interactions
- Collected samples were analyzed using the following analytical techniques **HPLC-SEC** (for monomer loss), **Micro-Flow Imaging** (for particle formation), **Near UV-CD** (for conformational changes), and **Bio-Assay** (for potency changes).
- The effects of storage in the DosePro cartridges were also explored. Samples were stored at 5°C, 25°C, and 40°C and tested at specified time points. Humidity was not controlled as the DosePro cartridge is non-permeable.
- Delivered volume was measured gravimetrically by collecting the injected samples from 5 devices at 0, 6, and 12 months.

Results

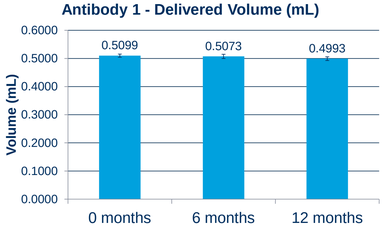
- When measured by HPLC-SEC, there was no significant difference in purity between the vial controls, the manually expelled DosePro cartridges, or the cartridges subjected to instantaneous injection. Purity loss is consistent with expected characteristics of the two mAbs.

Figure 2: Representative purity loss of Antibody 1. No noticeable difference in purity loss between controls and instantaneous injected samples after storage at 40°C (a), 25°C



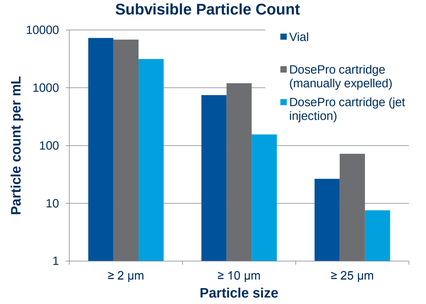
- Gravimetrically measured delivered volume remained consistent over 12 months. Five samples were injected and measured at each time point. Figure 3 shows the average delivered volume and the standard deviation between the 5 samples. Standard deviations for 0, 6, and 12 months were 0.0056, 0.0075, and 0.0067 respectively. All individual samples were within 3.2% of nominal (0.500 mL).

Figure 3: Delivered volume was measured gravimetrically. Results show consistent delivered volume over 12 months



- Subvisible particle counts for both the manually expelled and instantaneous injection samples were comparable to vial controls after 12 months of storage at 5°C. Particles were measured by MFI and counted in the following size bins: ≥2µm, ≥10µm, and ≥25µm. All particle counts were within the acceptable range for the antibodies. Figure 4 shows the particle counts per mL.

Figure 4: A representative subvisible particle count after 12 months storage at 5°C. Particle counts are comparable and within expected range for all samples.



- Molecular conformation of the two tested molecules did not show any signs of change after instantaneous injection when measured by near-UV CD (data not shown).
- Potency of the antibodies were not adversely affected by the instantaneous injection through the DosePro device. By BioAssay, all potency measures were over 90% of expected value, which is within the variability of the assay.
- Instantaneous injection did not noticeably affect pH, osmolality, or protein concentration as measured by spectrophotometer at 280nm (data not shown).

Conclusions

- There were no observed incompatibilities between the mAb formulations tested and the DosePro primary container.
- Instantaneous injection did not result in significant changes in biophysical characteristics when assessed by:
  - Monomer purity loss as measured by HPLC-SEC
  - Subvisible particle counts as measured by MFI
  - Potency as measured by BioAssay
  - Conformational changes as measured by near-UC CD
  - pH, osmolality, or protein concentration
- Storage conditions did not affect the sensitivity of the mAb formulations to instantaneous injection by the DosePro device.
- Delivered volume was consistent over 12 months.

Acknowledgements

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References

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2. Shah U.U., Roberts M., Gul M.O., Tuleu C.,Beresford M.W. Needle-free and microneedle drug delivery in children: A case for disease-modifying antirheumatic drugs (DMARDs). Int. J. of Pharmaceutics. 416:1, 1-11 (2011).