

The Effects of Freeze-Thaw Cycling on the Stability of the Adalimumab Biosimilar SB5



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Conclusions

- SB5 was stable in the immediate pack (nude pre-filled syringe) when exposed to multiple freeze-thaw cycles.
- These results may help hospital pharmacists to assess the impact of temperature excursions during shipment or storage on the product quality of SB5.

Introduction

- Temperature excursions may occur during manufacturing, storage, distribution and clinical trials.
- Limited data are available to hospital pharmacists to support decision making following temperature excursions.

Objectives

- The purpose of this stability study was to evaluate the impact of high and low temperature conditions over a short period on the adalimumab biosimilar SB5.

Methods

Temperature exposure

- SB5 drug product (DP) was exposed to extreme temperature cycling conditions with a total of three cycles equating to 144 hours at 30 ± 2°C/65 ± 5% relative humidity and 144 hours at -5 ± 3°C (Table 1).

Table 1. Short-term temperature cycling stability study design for SB5 DP

	Storage conditions	Storage time
Cycle 1		
	30 ± 2°C/65 ± 5% RH	48 hours
	-5 ± 3°C	48 hours
Cycle 2		
	30 ± 2°C/65 ± 5% RH	48 hours
	-5 ± 3°C	48 hours
Cycle 3		
	30 ± 2°C/65 ± 5% RH	48 hours
	-5 ± 3°C	48 hours

RH, relative humidity

Assessments

- Samples were analyzed using a variety of validated methods for appearance, pH, protein concentration, container closure integrity, impurities, charge variants, oxidation, endotoxin, particulates and biological activity.

Results

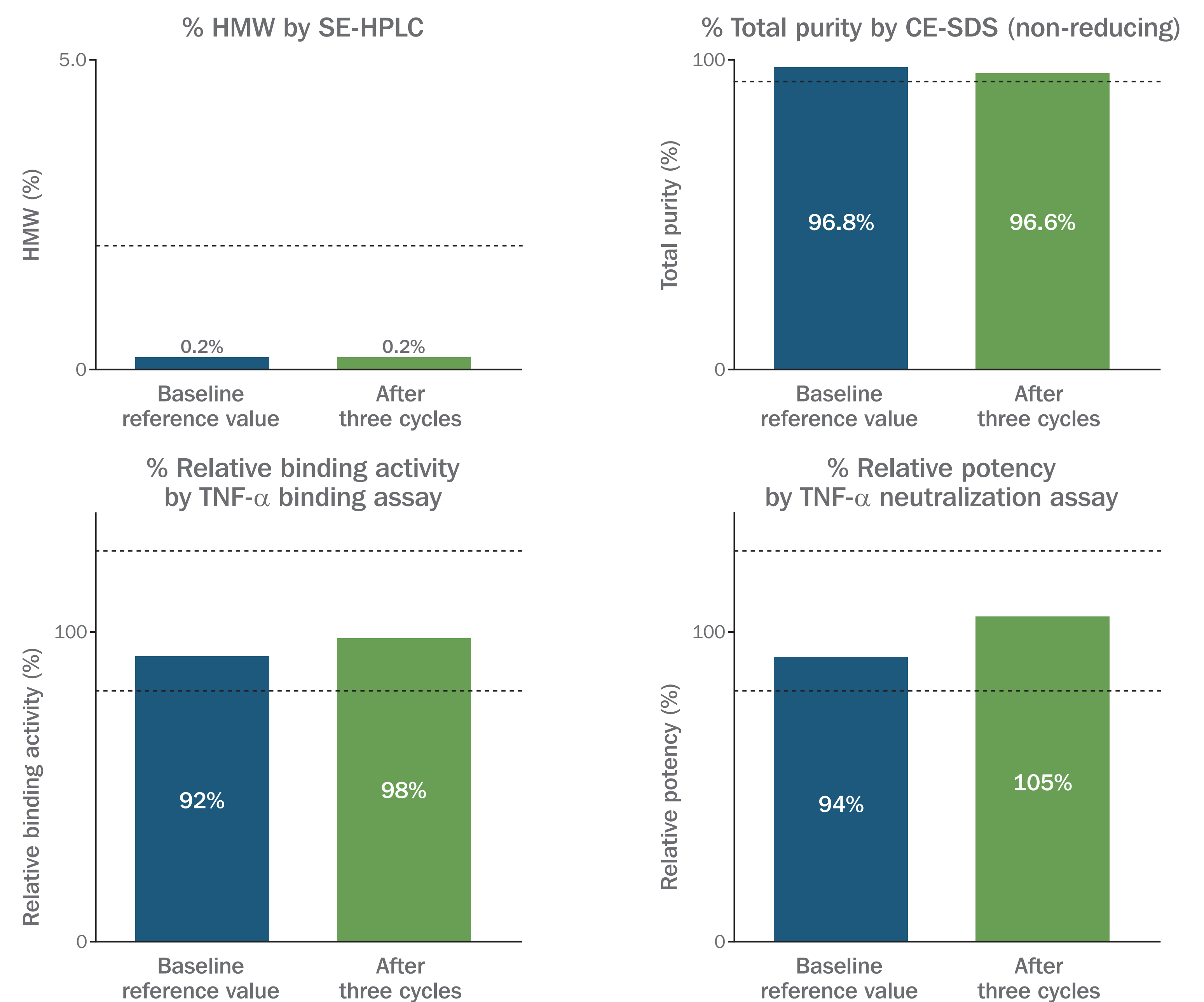
Critical quality attributes

- There were no apparent changes in critical quality attributes between baseline and following three temperature cycles (Figure 1).
- All results met the stability acceptance criteria for the four critical quality attributes.

Other

- Table 2 shows appearance, including colour, clarity, visible particle, pH, protein concentration, oxidation level, charge variant, endotoxin, container closure integrity and particulates, after three temperature cycles versus baseline.
- The results showed no apparent changes and/or met the stability acceptance criteria for each product quality attribute over the three temperature cycles.

Figure 1. Short-term temperature cycling result of SB5 DP across four critical quality attributes



----- Stability acceptance criteria.

CE-SDS: capillary electrophoresis-sodium dodecyl sulfate; HMW: high-molecular-weight species; SE-HPLC, size exclusion-high performance liquid chromatography; TNF- α , tumour necrosis factor- α .

Table 2. Test results of SB5 drug product at baseline and following three temperature cycles

Category	Test item	Baseline reference value	Temperature cycle 3
General test	Appearance: Colour	Colourless	B8 ≤ Sample < B7
	Appearance: Clarity	18 NTU	17 NTU
	Appearance: Visual particulates	Practically free from particles	Practically free from particles
	pH	5.3	5.3
Quantity test	Protein concentration (A ₂₈₀) (mg/mL)	51.6	49.7
Purity and impurities	SE-HPLC	% HMW impurities	0.2
	CE-SDS (non-reducing)	% Total purity	96.8
		% Single highest impurity	2.1
	icIEF	% Isoelectric point of main peak	8.6
		% Acidic	21.8
% Main		67.1	
Biological activity	Competitive binding assay to TNF- α by FRET	% Binding activity relative to reference standard	92
		% Potency relative to reference standard	94
	TNF- α neutralization assay by NF- κ B reporter gene	% Binding activity relative to reference standard	98
		% Potency relative to reference standard	105
Safety	Particulates ^a	Particle ≥10 μ m: particles/syringe	1521
		Particle ≥25 μ m: particles/syringe	15
	Endotoxin (EU/mL)	<5	
	Container closure integrity	NS	All sample syringes tested negative for visible signs of dye incursion
Additional tests	CEX-HPLC	% Acidic	23.5
		% Main	67.2
		% Basic	9.3
	Oxidation	% Heavy chain Met34	0.6
		% Heavy chain Met83	0.3
		% Heavy chain Met256	5.8
		% Heavy chain Met432	ND
		% Light chain Met4	0.2
	Particulates	Particle ≥2 μ m: particles/syringe	12217
		Particle ≥5 μ m: particles/syringe	6257
Particle ≥8 μ m: particles/syringe		2476	

^aThe acceptance criteria of particulate matter are 'Particle ≥10 μ m: ≤6000/syringe' and 'Particle ≥25 μ m: ≤600/syringe' according to Ph. Eur. 2.9.19/<USP 788>

CE-SDS, capillary electrophoresis-sodium dodecyl sulfate; CEX-HPLC, cation exchange-high performance liquid chromatography; DP, drug product; EU/mL, endotoxin units per millilitre; FRET, fluorescence resonance energy transfer; HMW, high-molecular-weight species; HPLC, high performance liquid chromatography; icIEF, imaged capillary isoelectric focusing; ND, not detected; NF- κ B, nuclear factor kappa-light-chain-enhancer of activated B cells; NS, not scheduled; NTU, nephelometric turbidity unit; SE-HPLC, size exclusion-high performance liquid chromatography; TNF- α , tumour necrosis factor- α .