

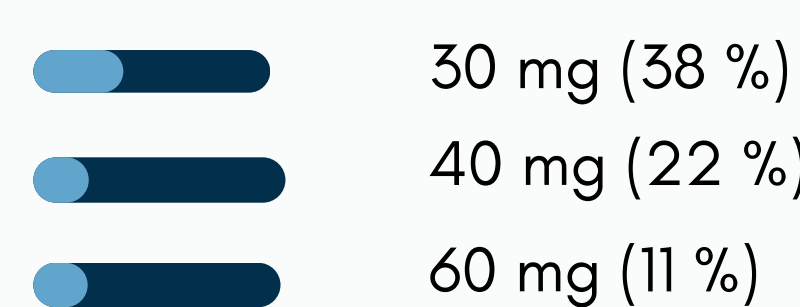
Background and importance



Pentobarbital sodium (PTB) is a barbiturate used for pediatric sedation, without existing marketed specialties.

→ Hospital preparations: PTB suppositories are produced

2018 - 2023: batches revealed significant non-compliance issues, particularly with uniformity of content (26 %)



Aim and objectives



Optimise paediatrics sedation by developing a new formulation (pentobarbital rectal solution) and determining its stability.

Materials and methods

Developing and validating a stability-indicating High-Performance Liquid Chromatography (HPLC) method (according to ICH Q2 guidelines)



Column: Waters XSelect @ HSS T3 (100 x 2.1 mm ; 2.5 μm)

Mobile phase water (pH 3) and acetonitrile (65:35 v/v) in isocratic elution (flow rate 0.4 mL/min)



Quantification was performed using UV detection analyzed at 210 nm.

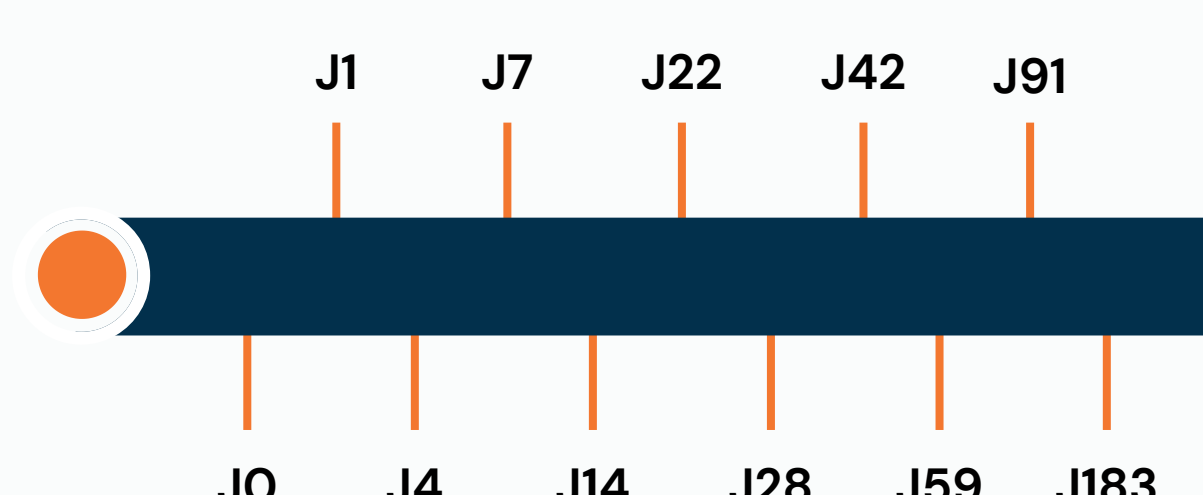


Forced degradation tests under acidic, alkaline, oxidative, and thermal stress conditions were conducted (3h, 4h, 5h, 6h, 24h, 48h)



Stability study: An intra-rectal PTB solution in a sterile water formulation was prepared. 3 batches were prepared, packaged in amber glass vials and syringes (2 pieces; 1.2 mL and 2.4 mL) at 2-8°C and 25°C/60% relative-humidity.

→ Visual appearance, PTB concentration, pH, and osmolality were evaluated



Concentration in each batch: 25 mg/mL

Results

Method validation

Linearity	Coefficient of determination R^2 ($R^2 = 0.9994 > 0.98$) Cochrane test (homogeneous variances) Slope significantly different from 0 (Student test)
Precision (CV < 5%)	Repeatability: CV = 0.333% Fidelity: CV = 1.14%
Accuracy (CV < 5%)	Control 1: CV = 1.33% Control 2: CV = 0.78% Control 3: CV = 1.02%

Chromatographic parameters

Retention time (min)	3.073
Retention factor (k)	2.84
Efficiency: theoretical plate number (N)	7830
Asymmetry factor (As)	1.11

Injection repeatability: **CV = 0.08% (< 2%)**



Forced degradation

- Degradation observed in H_2O_2 3% within 3 hours at room temperature.
- Degradation observed in NaOH 1N from 3 hours at room temperature.
- No degradation observed in HCl 1N after 48 hours at room temperature.
- At 80°C after 48 hours, formation of slightly coeluted impurity.

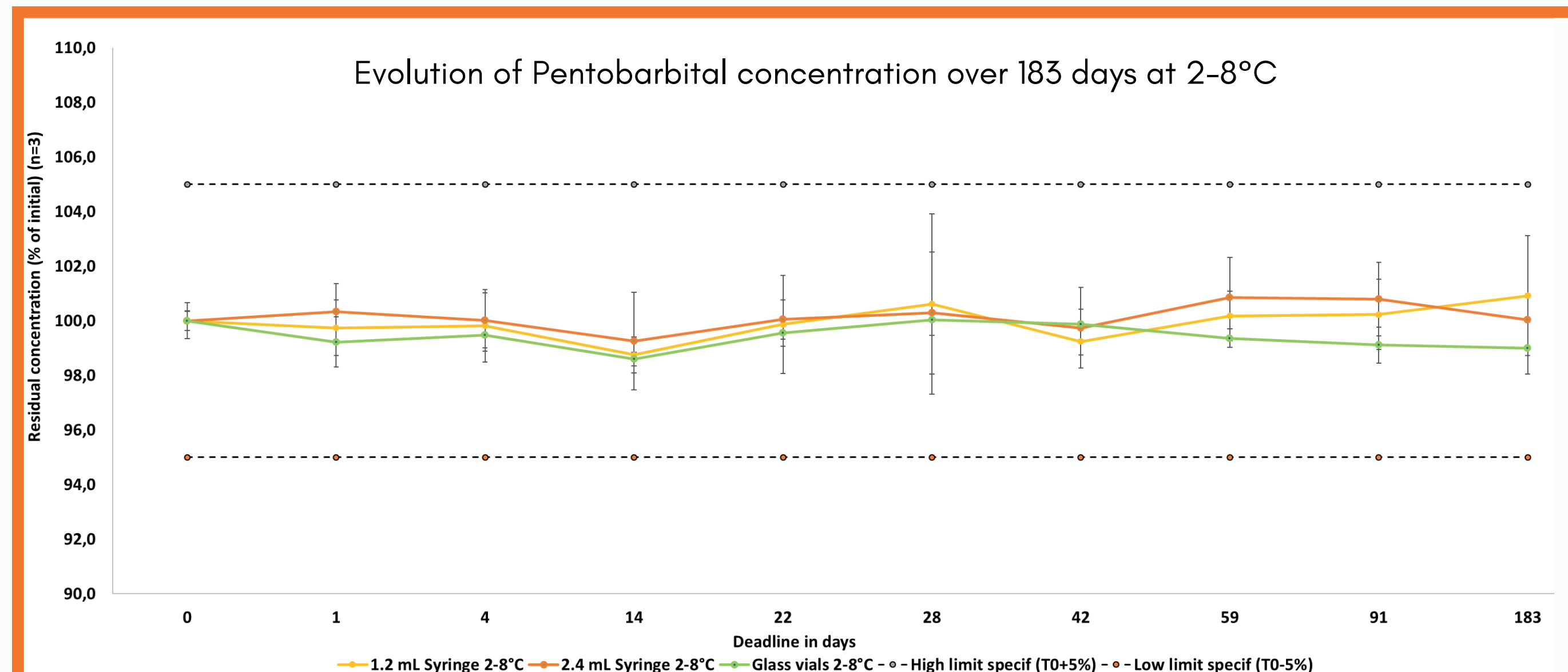
The method showed **good repeatability, resolution, specificity, precision, accuracy, and linearity** from 75 to 175 μg/mL.

Stability study

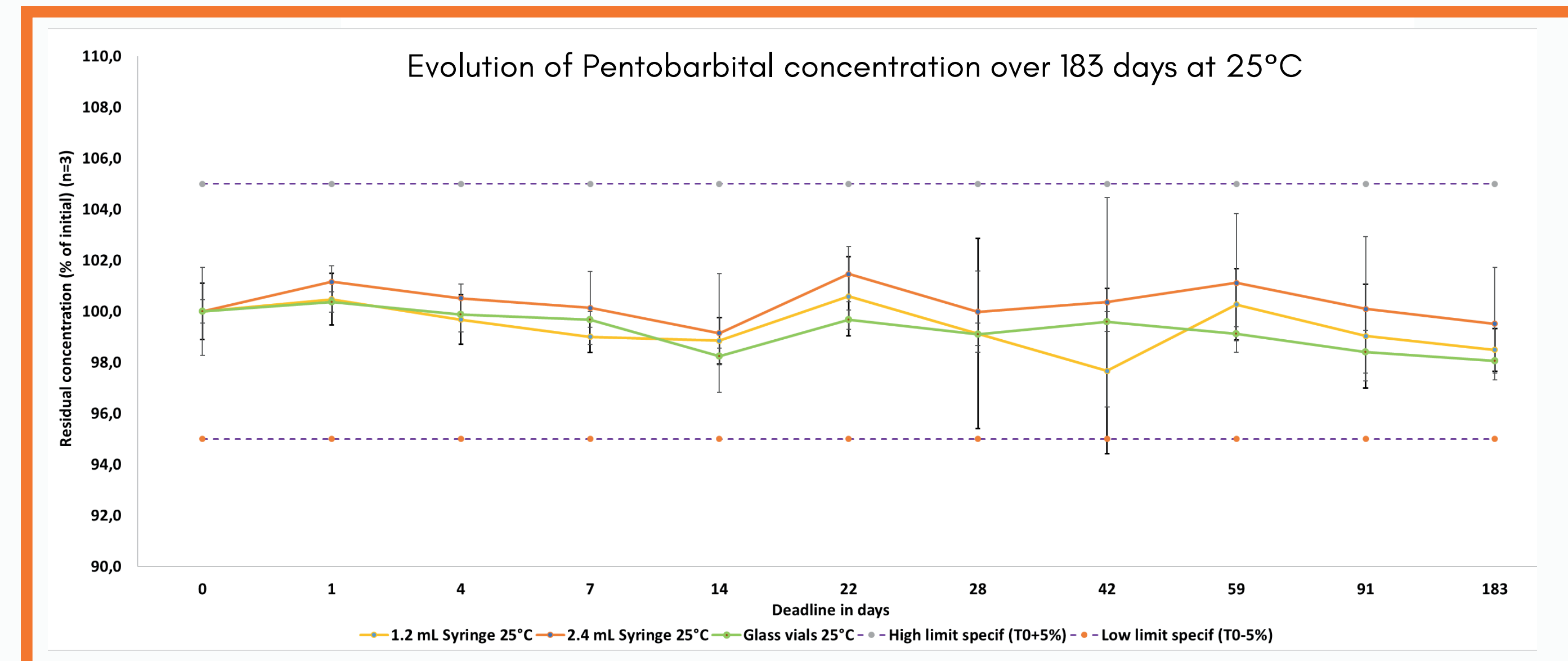
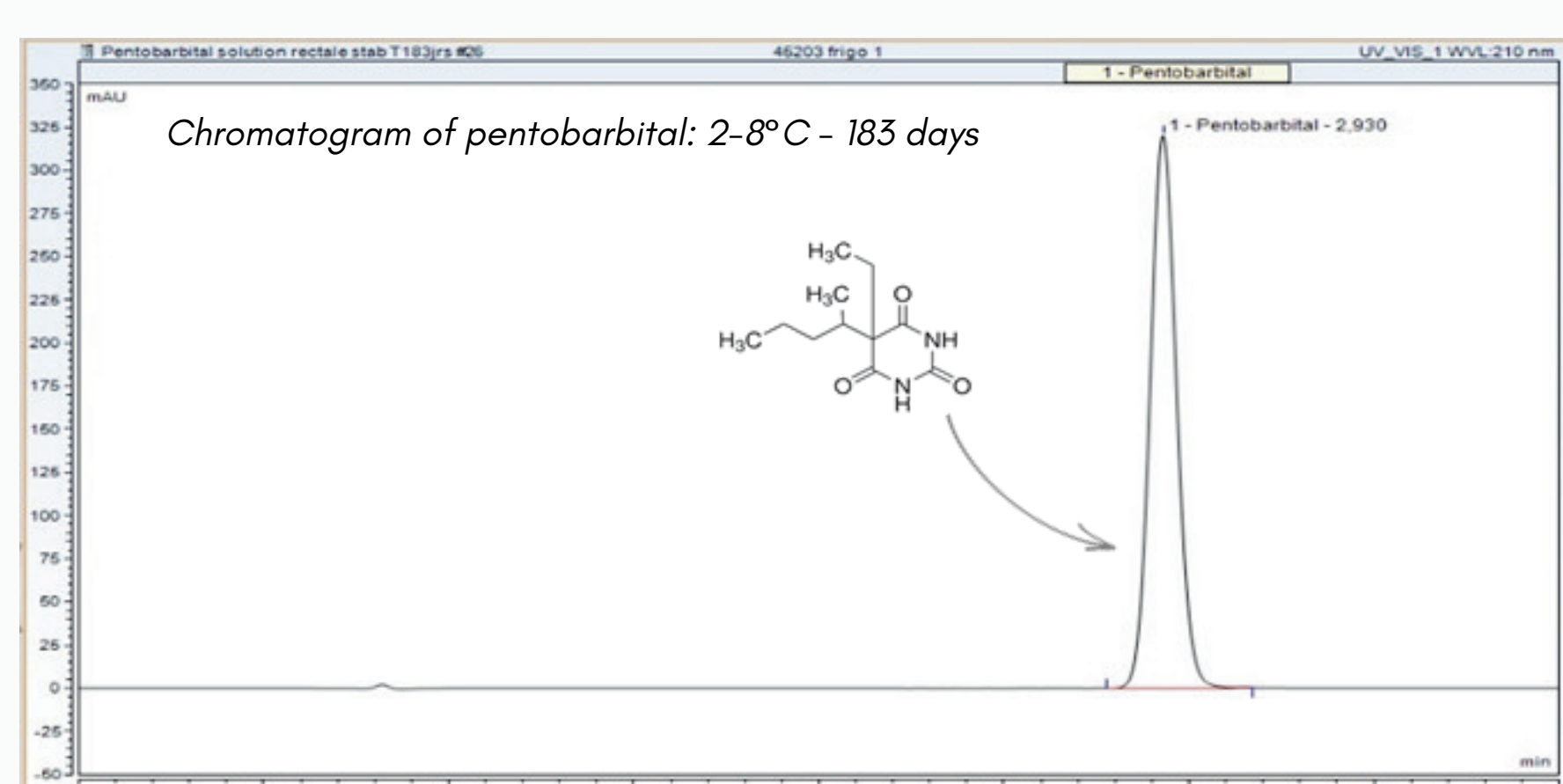


Visual appearance: unchanged

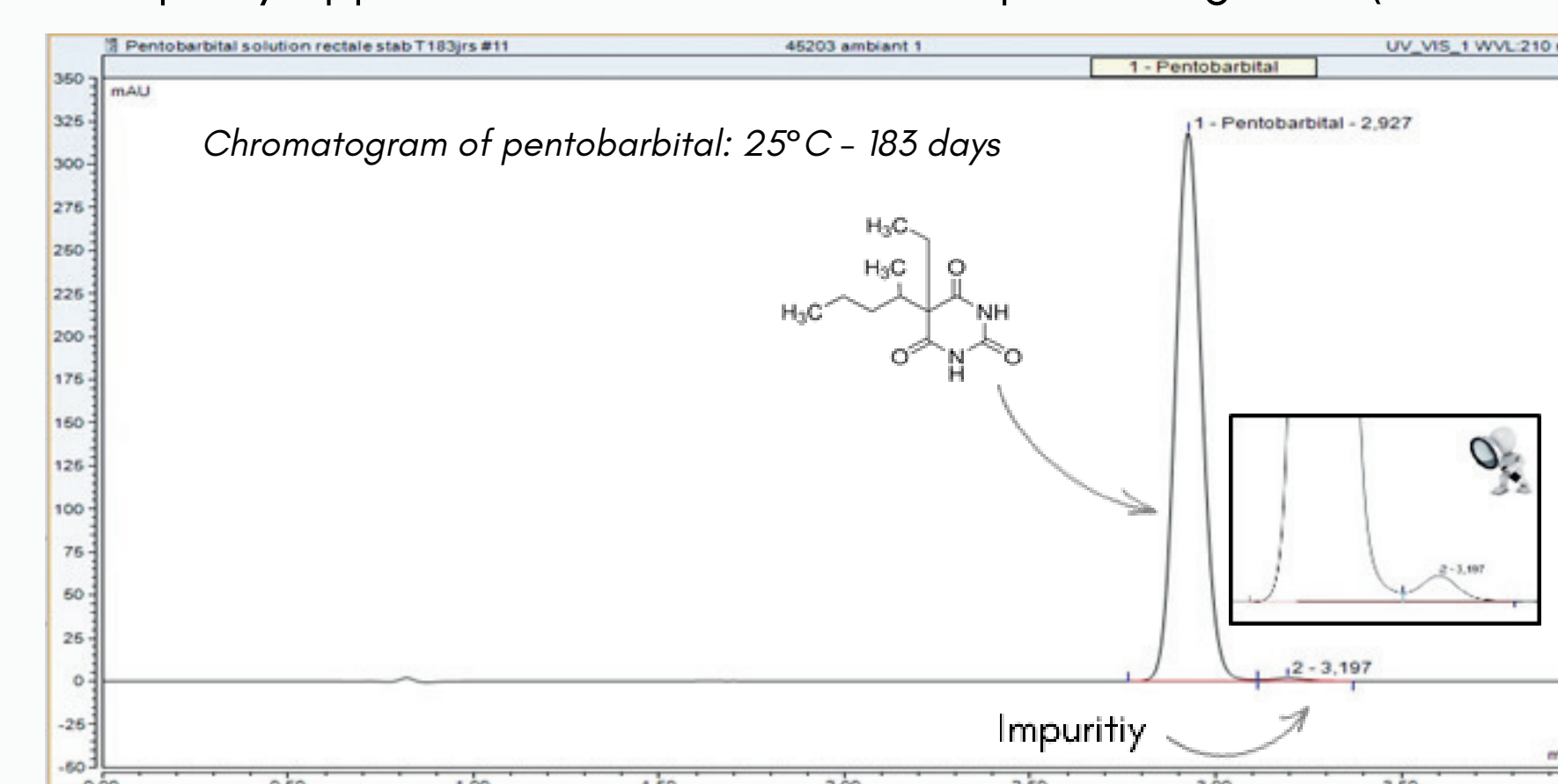
Content assessment:



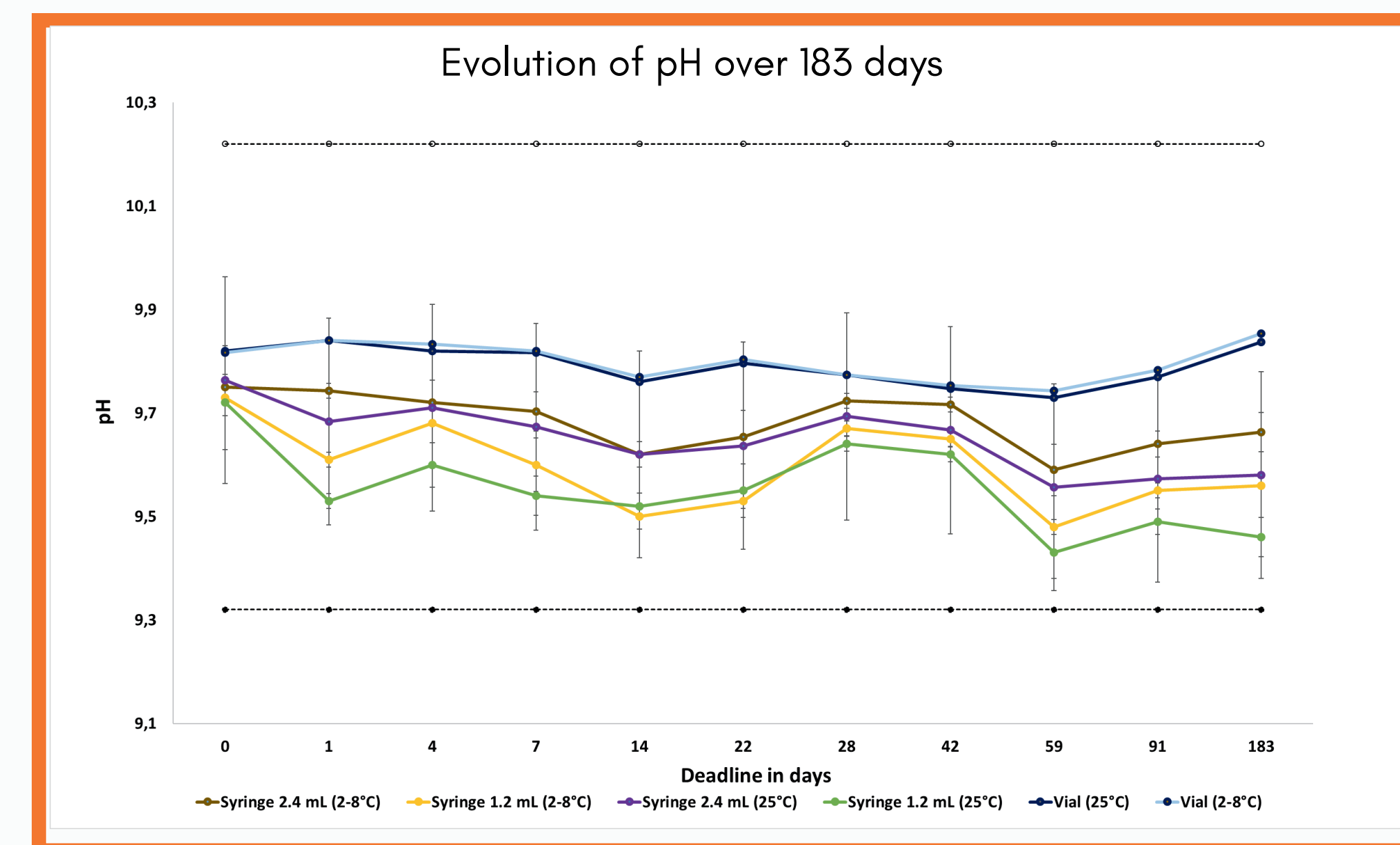
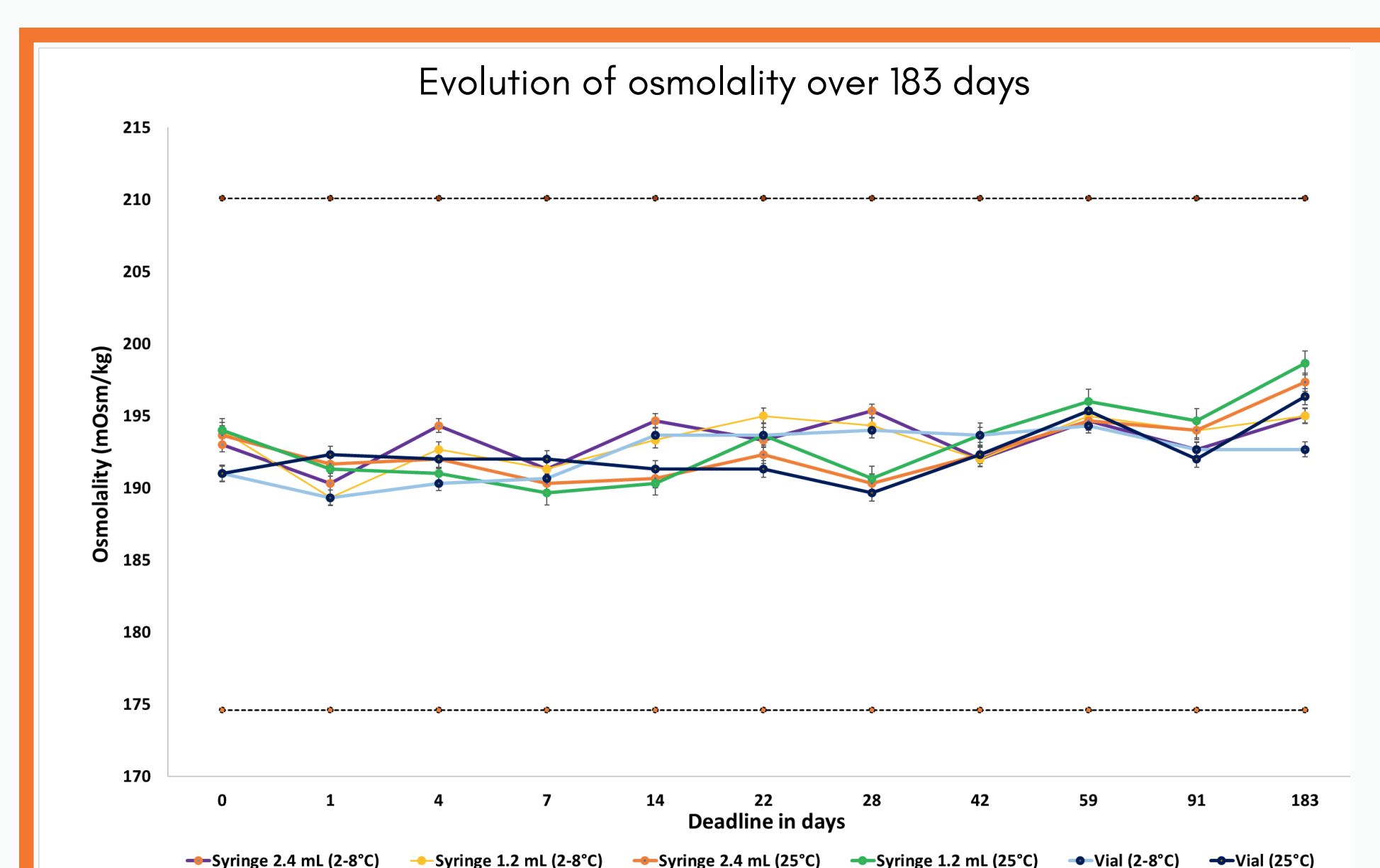
At Day 183, the Pentobarbital concentration at 2-8°C remained above 95% of the initial concentration.



At Day 183, the Pentobarbital concentration at 25°C remained above 95% of the initial concentration, but an impurity appeared from D59 coeluted representing 0.8% (relative-area)



pH and osmolality:



Osmolality and pH remained stable in both conservations.

Discussion et conclusion

Pentobarbital rectal solution can be produced and stored for 183 days at 2-8°C in vials and syringes 2 pieces. The formation of coeluted impurities suggests that storage at room temperature is not recommended. Syringe packaging enables standard doses to be produced, limiting the risk of error and simplifying the administration process.