3.2.P.3.4. IN-PROCESS MONITORING AND CONTROL – LIPID NANOPARTICLE (LNP) PRODUCTION AND BULK DRUG PRODUCT FORMULATION - TRIS/SUCROSE [PUURS]

This section provides the in-process controls (relevant process parameters and in-process testing for control [IPT-C]) with their acceptable ranges/acceptance criteria for the LNP production and Tris/Sucrose bulk drug product formulation manufacturing operations. This section also includes the specifications for 4.2 1st ind Tris buffer, 4.2 1st ind from external suppliers.

3.2.P.3.4.1. Dilution of Drug Substance

The process controls and set-points for dilution of drug substance are provided in Table 3.2.P.3.4-1.

Table 3.2.P.3.4-1. Process Controls for Dilution of Drug Substance

Process Control	Category	Controlled Setpoint
4.2 1st ind		

3.2.P.3.4.2. 4.2 1st ind Tris Buffer, 4.2 1st ind from External Suppliers

The 4.2 1st ind Tris buffer, 4.2 1st ind from external suppliers must meet the specifications described in Table 3.2.P.3.4-2. The 4.2 1st ind Tris buffer, 4.2 1st ind is tested for identity by or on behalf of the drug product manufacturer, and the remaining tests may be accepted based on the supplier's certificate of analysis.

Table 3.2.P.3.4-2. Specifications for 4.2 1st ind Tris Buffer, 4.2 1st ind from External Suppliers

Test	Analytical Methods	Acceptance Criteria
Identity[a]	Ph. Eur. 2.2.24	Sample spectrum is consistent with appropriate reference spectrum
Bioburden	USP <61>, Ph. Eur. 2.6.12	4.2 1st ind
Endotoxin	USP <85>, Ph. Eur. 2.6.14	
pH	USP <791>, Ph. Eur. 2.2.3	

a. Test performed by or on behalf of the drug product manufacturer.

3.2.P.3.4.3. Buffer Formulation

The formulation buffers are tested for pH. Process controls and acceptance criteria are provided in Table 3.2.P.3.4-3.

Table 3.2.P.3.4-3. Process Controls for Buffer Formulation

Process Control	Category	Acceptance Criteria
4.2 1st ind		
4.2 1st ind		

3.2.P.3.4.4. Preparation of Organic Phase

The process controls and acceptable ranges for preparation of the organic phase are provided in Table 3.2.P.3.4-4.

Table 3.2.P.3.4-4. Process Controls for Preparation of the Organic Phase

Process Control	Category	Acceptable Range
4.2 1st ind		
4.2 1st ind		

3.2.P.3.4.5. Preparation of Sucrose, Tris Solution

Concentration adjustment of the filtered bulk can be performed in a one-step or a two-step dilution process. A buffer solution containing 4.2 1st ind Sucrose, 4.2 1st ind Tris can be used in both one-step and two-step dilution processes. A buffer solution containing 4.2 1st ind sucrose, 4.2 1st ind Tris is used only in a two-step dilution process.

3.2.P.3.4.5.1. Preparation of 4.2 1st ind Sucrose, 4.2 1st ind Tris Solution

The process control and acceptance criterion for preparation of the 4.2 1st ind Sucrose, 4.2 1st ind Tris solution are provided in Table 3.2.P.3.4-5.

Table 3.2.P.3.4-5. Process Controls for Preparation of 4.2 1st ind Sucrose, 4.2 1st ind Tris Solution

Process Control	Category	Acceptance Criteria
4.2 1st ind		

3.2.P.3.4.5.2. Preparation of 4.2 1st ind Sucrose, 4.2 1st ind Tris Solution

The process control and acceptance criterion for preparation of the 4.2 1st ind Sucrose, 4.2 1st ind Tris solution are provided in Table 3.2.P.3.4-6.

Table 3.2.P.3.4-6. Process Controls for Preparation of 4.2 1st ind Sucrose, 4.2 1st ind Tris Solution

Process Control	Category	Acceptance Criteria
4.2 1st ind		

3.2.P.3.4.6. Lipid Nanoparticle Formation and Stabilization

The process controls and acceptable ranges for lipid nanoparticle formation and stabilization are provided in Table 3.2.P.3.4-7.

Table 3.2.P.3.4-7. Process Controls for Formation and Stabilization of LNPs

Process Control	Category	Acceptable Range
4.2 1st ind		
4.2 1st ind		

3.2.P.3.4.7. Buffer Exchange and Concentration

The process controls and acceptable ranges for concentration, buffer exchange and filtration are provided in Table 3.2.P.3.4-8.

Table 3.2.P.3.4-8. Process Controls for Buffer Exchange and Concentration

Process Control	Category	Target Set-point/Acceptable Range
4.2 1st ind		

3.2.P.3.4.8. Concentration Adjustment and Addition of Cryoprotectant

4.2 1st ind

3.2.P.3.4.8.1. Concentration Adjustment and Addition of Cryoprotectant for a Target Batch Size Range of 4.2 1st ind RNA or 4.2 1st ind RNA 4.2 1st ind

The process controls and acceptable ranges/acceptance criteria for concentration adjustment and addition of cryoprotectant are provided in Table 3.2.P.3.4-9.

Table 3.2.P.3.4-9. Process Controls for Concentration Adjustment and Addition of Cryoprotectant

Process Control	Category	Acceptable Range/Acceptance Criteria
4.2 1st ind		

3.2.P.3.4.8.2. Concentration Adjustment and Addition of Cryoprotectant for a Target Batch Size Range of 4.2 1st ind RNA (4.2 1st ind)

3.2.P.3.4.8.2.1. 4.2 1st ind

4.2 1st ind

The process controls and acceptable ranges/acceptance criteria for concentration adjustment 4.2 1st ind are provided in Table 3.2.P.3.4-10.

Table 3.2.P.3.4-10. Process Controls for First Concentration Adjustment Step to 0.5 mg/ml

Process Control	Category	Acceptable Range/Acceptance Criteria
4.2 1st ind		

3.2.P.3.4.8.2.2. 4.2 1st ind

4.2 1st ind

The process controls and acceptable ranges/acceptance criteria for concentration adjustment 4.2 1st ind are provided in Table 3.2.P.3.4-11.

Table 3.2.P.3.4-11. Process Controls for Second Concentration Adjustment Step to 0.1 mg/ml

Process Control	Category	Acceptable Range/Acceptance Criteria
4.2 1st ind		

3.2.P.3.4.8.2.3. Optional Filling into Flexible Freeze Thaw (FFT) Bags

The process parameters for the DPI are outlined in Table 3.2.P.3.4-12.

Table 3.2.P.3.4-12. Process Parameters For Drug Product Intermediate

Process Parameters	Acceptable Range
Storage temperature DPI	-90 to -60 °C[a]

a. The drug product intermediate shelf life at -90 to -60 °C is described in Section 3.2.P.3.4 Controls of Critical Steps and Intermediates-Drug Product Intermediate 0.5 mg/mL -Stability Summary – Tris/Sucrose .

Further processing of the DPI in flexible containers is described in Section 3.2.P.3.3 Description of Manufacturing Process and Process Controls - Fill Finish - Tris-Sucrose.