

3.2.P.3.4. PROCESS STEPS HOLD TIMES – LNP PRODUCTION AND DRUG PRODUCT FORMULATION - TRIS/SUCROSE [PUURS]

3.2.P.3.4.1. Process Hold Times

During process development, studies were performed to determine acceptable hold times for drug substance and bulk drug product during the drug product manufacturing process. The hold times were confirmed during process validation studies.

[Table 3.2.P.3.4-1](#) lists the hold times of the drug product in-process materials and steps during LNP production and bulk drug product formulation.

Table 3.2.P.3.4-1. LNP Production and Bulk Drug Product Formulation Process Hold Times

Material or In-Process Hold Description ^a	Process Steps	Target Hold Time
Drug substance thaw	Controlled thaw equipment: Time drug substance in 4.2 1st ind containers is thawed 4.2 1st ind	4.2 1st ind
	Controlled room temperature thaw: Time drug substance in 4.2 1st ind containers is thawed 4.2 1st ind	
Drug substance post thaw	Maximum time that thawed BNT162b2 drug substance can be held 4.2 1st ind in the 4.2 1st ind container including addition of DS to the vessel up to the point of dilution.	
Drug substance post dilution 4.2 1st ind	Time from addition of 4.2 1st ind to drug substance until end of LNP formation step.	
Organic phase post mix 4.2 1st ind	Time from end of organic phase mixing until end of LNP formation.	
Citrate buffer 4.2 1st ind	Total hold time for citrate buffer post addition of 4.2 1st ind to be used for LNP formulation or concentration/buffer exchange step.	
4.2 1st ind Tris buffer 4.2 1st ind	Total hold time for Tris buffer post addition 4.2 1st ind to be used for buffer exchange/concentration, filtration and concentration adjustment steps.	
	Total hold time for Tris buffer from start of in-line dilution to be used for buffer exchange/concentration, filtration and concentration adjustment steps.	
4.2 1st ind sucrose, 4.2 1st ind mM Tris solution 4.2 1st ind	Total hold time for sucrose/Tris solution post addition 4.2 1st ind to be used for concentration adjustment step.	
4.2 1st ind sucrose, 4.2 1st ind mM Tris solution 4.2 1st ind	Total hold time for sucrose/Tris solution post addition 4.2 1st ind to be used for concentration adjustment step.	
Time of LNP formation	Time from start of mixing aqueous and organic phases at 4.2 1st ind until start of 4.2 1st ind step including collection and hold 4.2 1st ind.	
Time of 4.2 1st ind filtration unit operation 4.2 1st ind	Time from start of 4.2 1st ind operation to end of bioburden reduction filtration 4.2 1st ind	
Post bioburden reduction filtration 4.2 1st ind	Time post bioburden reduction filtration until end of sterile filtration or freezing of DPI 4.2 1st ind until the start of concentration adjustment and cryoprotectant addition.	

a. Temperature values 15-25 °C represent room temperature

b. 4.2 1st ind

c. 4.2 1st ind

d. 4.2 1st ind

e. 4.2 1st ind

Abbreviations: RT = Room temperature; 4.2 1st ind; LNP = Lipid nanoparticle; 4.2 1st ind