3.2.P.2.3.7.5 Summary of Analytical Procedure Changes – mRNA-1273 Drug Product

Table 61 provides a summary of the analytical procedure changes implemented prior to method validation. The analytical methods for mRNA-1273 Drug Product have been validated and changes between the validated processes and previously used processes are summarized in Table 62.

Changes to the mRNA-1273 Drug Product analytical procedures are described below:

Identity – The method has been revised from sequencing full Open Reading Frame (ORF) [redacted] The full ORF is still sequenced for CX-024414 mRNA. [redacted] to positively confirm the presence of the correct active in the drug product is considered sufficient given the more stringent method is used for CX-024414 mRNA.

In vitro Translation - An analytical procedure (in vitro translation with methionine labelling) has been developed to determine protein expression.

Lipid Identification/Lipid Impurities – The method has been revised to expand the sample curve, change sample injection volume and target concentration, increase the quantitation limit, and revise the system suitability criteria. The revisions are based on method optimization and considered commensurate with increase in scale. The revised method was qualified and found to be equivalent to the previous method.

Table 61: Summary of Analytical Procedure Revisions – mRNA-1273 Drug Product

Test Parameter	Analytical Procedure	Analytical Procedures				Rationale for Change
		PVU (PN 85201) (SPC-0948)		Norwood Scale A (PN 60075) (SPC-1063)	Catalent Scale A/B (PN 60073) (SPC-1128)	
		Version 2.0 [a]	Version 3.0	Version 1.0/ Version 2.0	Version 1.0/ Version 2.0/	
Appearance	Visual Inspection	SOP-0278 Version 4.0	SOP-0278 Version 4.0	SOP-0278 Version 4.0/ Version 5.0	SOP-0278 Version 5.0	Added instructions for preparation of opalescence reagents, updated clarity assessment instructions and reporting, and made administrative changes.
RNA Content	Anion Exchange HPLC	SOP-0235 Version 6.0	SOP-0235 Version 6.0	SOP-0235 Version 6.0	SOP-0235 Version 6.0	No change
Identity	Reverse Transcription Sanger Sequencing	SOP-0492 Version 3.0/ Version 4.0	SOP-0492 Version 4.0	SOP-0544 Version 3.0	SOP-0544 Version 3.0	Change at Phase 3 lots from sequencing full ORF [redacted] In Phase 3 process, full sequence is confirmed at CX-024414 mRNA release.
Purity Product-related Impurities	RP-HPLC	SOP-0383 Version 1.0	SOP-0383 Version 1.0	SOP-0383 Version 2.0	SOP-0383 Version 2.0	No change (revisions for clarity)
% RNA Encapsulation	Fluorescence	SOP-0298 Version 4.0	SOP-0298 Version 4.0	SOP-0298 Version 5.0	SOP-0298 Version 5.0	No change (revisions for clarity)
Protein Expression	In Vitro Translation Methionine Labelling	N/A [a]	SOP-0937 Version 2.0	SOP-0937 Version 2.0	SOP-0937 Version 2.0	Addition of test parameter to ensure SISPQ.
pH	USP <791>	USP <791>	USP <791>	USP <791>	USP <791>	No change
Osmolality	USP <785>	USP <785>	USP <785>	USP <785>	USP <785>	No change
Particle Size Polydispersity	Dynamic Light Scattering	SOP-0107 Version 3.0	SOP-0107 Version 3.0	SOP-0107 Version 3.0	SOP-0107 Version 3.0	No change
Lipid Identification: SM-102 Lipid Identification: Cholesterol Lipid Identification: DSPC Lipid Identification: PEG2000-DMG Lipid Content: SM-102 Lipid Content: Cholesterol Lipid Content: DSPC Lipid Content: PEG2000-DMG Lipid Content: Lipid Impurities	UPLC-CAD	SOP-0502 Version 3.0	SOP-0502 Version 3.0	SOP-0502 Version 4.0/5.0	SOP-0502 Version 5.0	Version 4.0 revisions based on method optimization commensurate with increase in scale. Version 5.0 revisions: No changes to method. Updated vendors/catalog numbers, added mcg/vial calculation.
Particulate Matter [redacted]	USP <788> Method 2	USP <788> Method 2	USP <788> Method 2	USP <788> Method 2	USP <788> Method 2	No change
Container Content	USP <697>	N/A [a]	N/A [a]	USP <697>	USP <697>	Addition of test parameter to ensure SISPQ.
Bacterial Endotoxins	USP <85> Ph. Eur. 2.6.14	USP <85> Ph. Eur. 2.6.14	USP <85> Ph. Eur. 2.6.14	USP <85> Ph. Eur. 2.6.14	USP <85> Ph. Eur. 2.6.14	No change
Sterility	USP <71> Ph. Eur. 2.6.1	USP <71> Ph. Eur. 2.6.1	USP <71> Ph. Eur. 2.6.1	USP <71> Ph. Eur. 2.6.1	USP <71> Ph. Eur. 2.6.1	No change; sterility testing of PN 60073 will occur at Catalent

Abbreviations: CAD = charged aerosol detector; DSPC = 1,2-Distearoyl-sn-glycero-3-phosphatidylcholine; HPLC = high-performance liquid chromatography; N/A = not applicable; ORF = open reading fame; PVU = personal vaccine unit; SISPQ = safety, identity, strength, purity, and quality; UPLC = ultra-high-performance liquid chromatography

a) Test parameters not present on specification at time of testing. Applicable for lot 8520100101 only.

Table 62: Summary of Analytical Procedure Revisions – mRNA-1273 Drug Product Validated Procedures

Test Parameter		Analytical Procedure	Analytical Procedures: Catalent Scale A/B (PN 60073) (SPC-1128) Qualified Procedures	Analytical Procedures: Catalent Scale A/B (PN 60073) (SPC-1128) Validated Methods	Description of Change
Appearance		Visual Inspection	SOP-0278 Version 5.0	SOP-0278 Version 5.0	No change. The method evaluates the appearance of samples in accordance with USP<631>, Ph. Eur 2.2.1, and Ph. Eur. 2.9.20.
RNA Content		Anion Exchange HPLC	SOP-0235 Version 6.0	SOP-0999 Version 1.0	1. Removed weighing requirement for standards and sample prep after dilution. 2. Check standard changed from [redacted]
Identity		Reverse Transcription Sanger Sequencing	SOP-0544 Version 3.0	SOP-1032 Version 1.0	Addition of positive control and positive control system suitability criteria, removal of intermediate PCR step.
Purity		RP-HPLC	SOP-0383 Version 2.0	SOP-0996 Version 1.0	1. Removed weighing requirement for standards and sample prep after dilution. 2. Check standard changed from [redacted]
Product-related Impurities					
% RNA Encapsulation		Fluorescence (RiboGreen) [redacted]	SOP-0298 Version 5.0	SOP-1000 Version 1.0	
In Vitro Translation		In Vitro Translation Methionine Labelling	SOP-0937 Version 2.0	SOP-0937 Version 2.0	No significant changes.
pH		USP <791>	USP <791>	USP <791>	No change
Osmolality		USP <785>	USP <785>	USP <785>	No Change
Particle Size		Dynamic Light Scattering	SOP-0107 Version 3.0	SOP-0998 Version 1.0	No significant changes
Polydispersity					
Lipid Identification	SM-102	UPLC-CAD	SOP-0502 Version 5.0	SOP-1001 Version 1.0	Previous methods combined to contain sample preparations for [redacted], mRNA-1273 LNP, and mRNA-1273 DP. Sensitivity solution target [redacted] (to align with updated QL). QL updated to [redacted] in previous methods.
	Cholesterol				
	DSPC				
	PEG2000-DMG				
Lipid Content	SM-102				
	Cholesterol				
	DSPC				
	PEG2000-DMG				
	Lipid Impurities				
Particulate Matter	≥ 25 µm	USP <788> Method 2	USP <788> Method 2	USP <788> Method 2	No Change
	≥ 10 µm				
Container Content		USP <697>	USP <697>	USP <697>	
Bacterial Endotoxins		USP <85> Ph. Eur. 2.6.14	USP <85> Ph. Eur. 2.6.14	USP <85> Ph. Eur. 2.6.14	
Sterility		USP <71> Ph. Eur. 2.6.1	USP <71> Ph. Eur. 2.6.1	USP <71> Ph. Eur. 2.6.1	

3.2.P.2.3.7.6 Comparability – mRNA-1273 Drug Product

3.2.P.2.3.7.6.1 Comparability – mRNA-1273 Drug Product Batch Analyses

Comparative batch analyses are provided in Table 63 to Table 66.

Testing results for key quality attributes are similar and within specification for both Phase 1/2 (PVU Scale), Norwood Vanrx initial Scale A, Norwood Vanrx Scale A PPQ, and Catalent Scale A PPQ and Catalent Scale B.

Table 63: Comparative Batch Analyses – mRNA-1273 Drug Product (PVU Scale)

Test Parameter		Analytical Procedure	Acceptance Criteria		PVU Scale (PN 85201)			
					8520100101	8520100102	8520100103	8520100104
Appearance		Visual Inspection	Color	White to off-white dispersion.	White to off-white dispersion.	White to off-white dispersion.	White to off-white dispersion.	White to off-white dispersion.
			Particulates	May contain visible, white or translucent product-related particles	Essentially free of particulates	Essentially free of particulates	Essentially free of particulates	Essentially free of particulates
RNA Content		AEX-HPLC	[redacted]					
Identity		Reverse Transcription Sanger Sequencing	Sequence matches 100% of the coding region		Conforms	Conforms	Conforms	Conforms
Purity		RP-HPLC	[redacted]					
Product-Related Impurities			Report % area for each impurity group (IG)	IG1(pre-main peak area)	[redacted]			
				IG2(post-main peak area)				
				IG3(mRNA-adduct species)				
% RNA Encapsulation		Fluorescence	≥ 70%					
Protein Expression		In Vitro Translation Methionine Labelling	[redacted]		N/A [a]			
pH		USP <791>						
Osmolality		USP <785>						
Particle Size		Dynamic Light Scattering						
Polydispersity			Report result					
Lipid Identification	SM-102		Matches RT of reference		Conforms	Conforms	Conforms	Conforms
	Cholesterol		Matches RT of reference		Conforms	Conforms	Conforms	Conforms
	DSPC		Matches RT of reference		Conforms	Conforms	Conforms	Conforms
	PEG2000-DMG		Matches RT of reference		Conforms	Conforms	Conforms	Conforms
Lipid Content	SM-102	UPLC-CAD	[redacted]					
	Cholesterol							
	DSPC							
	PEG2000-DMG							
	Lipid Impurities						RRT / % area	RRT / % area
			Individual Impurities	Report %area and RRT	ND	ND	[redacted] / < LOQ	[redacted] / < LOQ
							[redacted] / < LOQ	[redacted] / < LOQ
							[redacted] / < LOQ	[redacted] / < LOQ
							[redacted] / < LOQ	[redacted] / < LOQ
							N/A / N/A	[redacted] / < LOQ
			Total Impurities	Report % area	ND	ND	< LOQ	< LOQ
Particulate Matter	[redacted]	USP <788> Method 2	[redacted]					
Container Content		USP <697>			N/A [a]	N/A [a]	N/A [a]	N/A [a]
Bacterial Endotoxins		USP <85> Ph. Eur. 2.6.14	[redacted]					
Sterility		USP <71>	No Growth		No Growth	No Growth	No Growth	No Growth

Abbreviations: AEX = anion exchange chromatography; CAD = charged aerosol detector; DSPC = 1,2-Distearoyl-sn-glycero-3-phosphatidylcholine ; EU = endotoxin units; HPLC = high-pressure liquid chromatography; LOQ = limit of quantitation; ND = not detected; RP = reverse phase;RT = retention time; RRT = relative retention time; UPLC = ultra-high-performance liquid chromatography
a) Test parameters not present on specification at time of testing.

Table 64: Comparative Batch Analyses – mRNA-1273 Drug Product Norwood Vanrx (Scale A)

Test Parameter		Analytical Procedure	Acceptance Criteria		Norwood Vanrx Scale A					
					6007520001		6007520002		6007520003	
Appearance		Visual Inspection	White to off-white dispersion. May contain visible, white or translucent product-related particles		White to off-white dispersion. Essentially free of particulates.		White to off-white dispersion. Essentially free of particulates.		White to off-white dispersion. Essentially free of particulates.	
RNA Content		AEX-HPLC	[redacted] (Target: 0.20 mg/mL)		[redacted]					
Identity		Reverse Transcription Sanger Sequencing	Sequence matches description		Conforms		Conforms		Conforms	
Purity		RP-HPLC	[redacted]							
Product-Related Impurities		RP-HPLC	Report % area for each impurity group (IG):	IG1 (pre-main peak area)	[redacted]					
				IG 2 (post-main peak area)						
				IG 3 (mRNA-adduct species)						
% RNA Encapsulation		Fluorescence	[redacted]							
Protein Expression		In Vitro Translation								
pH		USP <791>								
Osmolality		USP <785>								
Particle Size		Dynamic Light Scattering								
Polydispersity		Dynamic Light Scattering	Report result							
Lipid Identification	SM-102	UPLC-CAD	Matches retention time of reference		Conforms		Conforms		Conforms	
	Cholesterol	UPLC-CAD	Matches retention time of reference		Conforms		Conforms		Conforms	
	DSPC	UPLC-CAD	Matches retention time of reference		Conforms		Conforms		Conforms	
	PEG2000-DMG	UPLC-CAD	Matches retention time of reference		Conforms		Conforms		Conforms	
Lipid Content	SM-102	UPLC-CAD	[redacted]							
	Cholesterol	UPLC-CAD								
	DSPC	UPLC-CAD								
	PEG2000-DMG	UPLC-CAD								
	Lipid Impurities	UPLC-CAD	Individual Impurities	Report % area and RRT	RRT	% Area	RRT	% Area	RRT	% Area
					[redacted]					
							N/A	N/A	N/A	N/A
							N/A	N/A	N/A	N/A
			Total Impurities	Report % area						
Particulate Matter	[redacted]	USP <788> Method 2								
Container Content		USP <697>								
Bacterial Endotoxins		USP <85> Ph. Eur. 2.6.14								
Sterility		USP <71>	No Growth		No Growth		No Growth		No Growth	

Abbreviations: AEX = anion exchange chromatography; CAD = charged aerosol detector; CFU = colony-forming unit(s); DLS = dynamic light scattering; DSPC = 1,2-Distearoyl-sn-glycero-3-phosphatidylcholine; EU = endotoxin unit(s); HPLC = high-pressure liquid chromatography; RP = reverse phase; RRT = relative retention time; UPLC = ultra-high-performance liquid chromatography

Table 65: Comparative Batch Analyses – mRNA-1273 Drug Product (Norwood Vanrx Scale A PPQ)

Test Parameter		Analytical Procedure	Acceptance Criteria		Norwood Vanrx Scale A PPQ					
					6007520004		6007520005		6007520006	
Appearance		Visual Inspection	White to off-white dispersion. May contain visible, white or translucent product-related particulates		White to off-white dispersion. Essentially free of particulates.		White to off-white dispersion. Essentially free of particulates.		White to off-white dispersion. Essentially free of particulates.	
RNA Content		AEX-HPLC	[redacted] (Target: 0. 20 mg/mL)		[redacted]					
Identity		Reverse Transcription Sanger Sequencing	Sequence matches description		Conforms		Conforms		Conforms	
Purity		RP-HPLC	[redacted]		[redacted]					
Product-Related Impurities			Report % area for each impurity group (IG):	IG1 (pre-main peak area)	[redacted]					
				IG 2 (post-main peak area)						
				IG 3 (mRNA-adduct species)						
% RNA Encapsulation		Fluorescence	[redacted]							
Protein Expression		In Vitro Translation								
pH		USP <791>								
Osmolality		USP <785>								
Particle Size		Dynamic Light Scattering								
Polydispersity										
Lipid Identification	SM-102	UPLC-CAD	Matches retention time of reference		Conforms		Conforms		Conforms	
	Cholesterol		Matches retention time of reference		Conforms		Conforms		Conforms	
	DSPC		Matches retention time of reference		Conforms		Conforms		Conforms	
	PEG2000-DMG		Matches retention time of reference		Conforms		Conforms		Conforms	
Lipid Content	SM-102		[redacted]							
	Cholesterol									
	DSPC									
	PEG2000-DMG									
	Lipid Impurities		Individual Impurities	Report % area and RRT	**RRT**	**% Area**	**RRT**	**% Area**	**RRT**	**% Area**
					[redacted]					
			Total Impurities	Report % area						
Particulate Matter	[redacted]	USP <788> Method 2	[redacted]							
Container Content		USP <697>								
Bacterial Endotoxins		USP <85> Ph. Eur. 2.6.14								
Sterility		USP <71>	No Growth		No Growth		No Growth		No Growth	

Abbreviations: AEX = anion exchange chromatography; CAD = charged aerosol detector; CFU = colony-forming unit(s); DLS = dynamic light scattering; DSPC = 1,2-Distearoyl-sn-glycero-3-phosphatidylcholine; EU = endotoxin unit(s); HPLC = high-pressure liquid chromatography; PPQ = process performance qualification; RP = reverse phase; RRT = relative retention time; UPLC = ultra-high-performance liquid chromatography

Table 66: Comparative Batch Analyses – mRNA-1273 Drug Product Catalent (Scale A PPQ)

Test Parameter		Analytical Procedure	Acceptance Criteria	Catalent Scale A PPQ 6007320001 (057G20)	Catalent Scale A PPQ 6007320002 (062G20)	Catalent Scale A PPQ 6007320003 (001H20)
Appearance		Visual Inspection	White to off-white dispersion. May contain visible, white or translucent product-related particulates	White to off-white dispersion. Essentially free of particulates	White to off-white dispersion. Essentially free of particulates	White to off-white dispersion. Essentially free of particulates
RNA Content		AEX-HPLC	[redacted] (Target: 0. 20 mg/mL)	[redacted]	[redacted]	[redacted]
Identity		Reverse Transcription Sanger Sequencing	Sequence matches description	Conforms	Conforms	Conforms
Purity		RP-HPLC	[redacted]	[redacted]	[redacted]	[redacted]
Product-Related Impurities		RP-HPLC	Report % area for each impurity group: Impurity Group 1 (pre-main peak area)	[redacted]	[redacted]	[redacted]
			Impurity Group 2 (post-main peak area)			
			Impurity Group 3 (mRNA-adduct species)			
% RNA Encapsulation		Fluorescence	[redacted]	[redacted]	[redacted]	[redacted]
Protein Expression		In Vitro Translation				
pH		USP <791>				
Osmolality		USP <785>				
Particle Size		Dynamic Light Scattering				
Polydispersity						
Lipid Identification	SM-102	UPLC-CAD	Matches retention time of reference	Conforms	Conforms	Conforms
	Cholesterol		Matches retention time of reference	Conforms	Conforms	Conforms
	DSPC		Matches retention time of reference	Conforms	Conforms	Conforms
	PEG2000-DMG		Matches retention time of reference	Conforms	Conforms	Conforms
Lipid Content	SM-102	UPLC-CAD	[redacted]	[redacted]	[redacted]	[redacted]
	Cholesterol					
	DSPC					
	PEG2000-DMG					
	Lipid Impurities			RRT / %Area	RRT / %Area	RRT / %Area
			Individual Impurities: [redacted] area (Report RRT)	[redacted]	[redacted]	[redacted]
			Total Impurities: [redacted] area			
Particulate Matter	[redacted]	USP <788> Method 2	[redacted]	[redacted]	[redacted]	[redacted]
Container Content		USP <697>				
Bacterial Endotoxins		USP <85> Ph. Eur. 2.6.14				
Sterility		USP <71>	No Growth	No Growth	No Growth	No Growth

Abbreviations: AEX = anion exchange chromatography; CAD = charged aerosol detector; CFU = colony-forming unit(s); DLS = dynamic light scattering; DSPC = 1,2-Distearoyl-sn-glycero-3-phosphatidylcholine; EU = endotoxin unit(s); HPLC = high-pressure liquid chromatography; PPQ = process performance qualification; RP = reverse phase; RRT = relative retention time; UPLC = ultra-high-performance liquid chromatography

Table 67: Comparative Batch Analyses – mRNA-1273 Drug Product Catalent (Scale B)

Test Parameter		Analytical Procedure	Acceptance Criteria			Catalent Scale B 6007320005 (011J20)	
Appearance		Visual Inspection	White to off-white dispersion. May contain visible, white or translucent product-related particulates			White to off-white dispersion, Essentially free of particulate	
RNA Content		AEX-HPLC	[redacted] (Target: 0. 20 mg/mL)			[redacted]	
Identity		Reverse Transcription Sanger Sequencing	Sequence matches description			Conforms	
Purity		RP-HPLC	[redacted]				
Product-related Impurities			Report % area for each impurity group:	Impurity Group 1 (pre-main peak area)		[redacted]	
				Impurity Group 2 (post-main peak area)			
				Impurity Group 3 (mRNA-adduct species)			
% RNA Encapsulation		Fluorescence	[redacted]				
Potency		In Vitro Translation					
pH		USP <791>					
Osmolality		USP <785>					
Particle Size		Dynamic Light Scattering					
Polydispersity							
Lipid Identification	SM-102	UPLC-CAD	Matches retention time of reference			Conforms	
	Cholesterol		Matches retention time of reference			Conforms	
	DSPC		Matches retention time of reference			Conforms	
	PEG2000-DMG		Matches retention time of reference			Conforms	
Lipid Content	SM-102		[redacted]				
	Cholesterol						
	DSPC						
	PEG2000-DMG						
	Lipid Impurities					RRT	%
			Individual Impurities	[redacted] area (Report RRT)		[redacted]	
			Total Impurities	[redacted] area			
Particulate Matter	[redacted]	USP <788> Method 2	[redacted]				
Container Content		USP <697>					
Bacterial Endotoxins		USP <85> Ph. Eur. 2.6.14					
Sterility		USP <71>	No Growth			No Growth	

Abbreviations: AEX = anion exchange chromatography; CAD = charged aerosol detector; CFU = colony-forming unit(s); DLS = dynamic light scattering; DSPC = 1,2-Distearoyl-sn-glycero-3-phosphatidylcholine; EU = endotoxin unit(s); HPLC = high-pressure liquid chromatography; PPQ = process performance qualification; RP = reverse phase; RRT = relative retention time; UPLC = ultra-high-performance liquid chromatography

3.2.P.2.3.7.6.2 Scale A to Scale B Comparability

Comparability was evaluated using relevant technical information to demonstrate that the changes to the mRNA-1273 Drug Product manufacturing process do not have an adverse impact on the quality, safety, and efficacy of the mRNA-1273 Drug Product and that the pre- and post-change product are comparable. The goal of comparability is to demonstrate that the quality attributes of the pre-change and post-change materials are highly similar and that the existing knowledge is sufficiently predictive to ensure that any differences in quality attributes have no adverse impact upon safety or efficacy of the mRNA-1273 Drug Product according to ICH Guideline Q5E: Comparability of Biotechnological/Biological Products Subject to Changes in Their Manufacturing Process. Analytical comparability was assessed by 1) release, 2) stability when available, and 3) extended characterization testing, against pre-defined acceptance criteria. Forced degradation will be evaluated in the next comparability phase to support clear definition of post-licensure stability comparability requirements. In-process controls and process parameters were also evaluated against expected ranges to support the comparability demonstration.

Moderna developed a robust manufacturing process and analytical comparability assessment strategy to support site and scale changes. Comparability is governed by protocol DPAD-PRO-0431 which addresses comparability for CX-024414 mRNA, [redacted], and mRNA-1273 LNP as well as mRNA-1273 Drug Product. Each material will be assessed independently using a modular approach and reported in Section 3.2.S.2.6 {CX-024414 mRNA}, [redacted], and Section 3.2.S.2.6 {mRNA-1273 LNP}.

The four phases of comparability are planned as described in Table 68. To date, only Phase 1 and Phase 2 of comparability are complete and reported in this section. Changes in manufacturing scale and site are listed in Table 69 and the lots listed in Table 70.

The following elements were included in the comparability study:

1. Description and justification of process changes including sites, scales, raw materials, process equipment and evaluation of process performance with respect to critical process parameters (CPPs) and in-process controls (IPCs) (Section P.2.3.7.6.2.1).
2. Statistical evaluation of comparability of release testing results (Section P.2.3.7.6.2.3).

Extended analytical characterization testing was not performed for mRNA-1273 Drug Product as part of comparability studies because the mRNA-1273 Drug Product characteristics are the same as the mRNA-1273 LNP. The results of the extended characterization testing for mRNA-1273 LNP conformed to the comparability expected range and are presented in Section 3.2.S.2.6 {mRNA-1273 LNP}.

Table 68: Four Phases of Comparability

Comparability Phase	mRNA-1273 Drug Product	Manufacturing Site
Phase 1- Initial Baseline	Development and clinical supply	ModernaTX, Inc (Norwood, MA)
	[redacted] vials	ModernaTX, Inc (Norwood, MA)
	[redacted] vials	Catalent (Bloomington, IN)
Phase 2- Preliminary Scale B	[redacted] vials	Catalent (Bloomington, IN)
Phase 3- Scale B Comparability	[redacted] vials	Catalent (Bloomington, IN)
Phase 4- Formal Comparability	Develop post-licensure comparability protocols	Catalent (Bloomington, IN)

Table 69: Summary of mRNA-1273 DP Manufacturing Process Scales and Sites

Scale A		Scale B	
Site	**Nominal Batch Scale**	**Site**	**Nominal Batch Scale**
ModernaTX, Inc. (Norwood, MA)	[redacted] multiple dose vials (Not for Commercial Use) PN 60075	Catalent Biologics, LLC (Bloomington, IN)	Up to [redacted] multiple dose vials (For Emergency Use Authorization and/or Commercial Use) PN 60073
Catalent Biologics, LLC (Bloomington, IN)	[redacted] multiple dose vials (For Emergency Use Authorization and/or Commercial Use) PN 60073		

Table 70: Genealogy of Clinical Lots for Comparability Demonstration

CX-024414 mRNA Lot	[redacted]	mRNA-1273 LNP Lot	mRNA-1273 Drug Product Lot	Use
8410000101		N/A	8520100101	Phase 1 (DMID Protocol 20-0003)
8410000102			8520100102	Phase 2 (Study P201)
8410000103			8520100103	
8410000104			8520100104	
4007220002		5006820002	6007520001	
			6007520002	Phase 3 (Study P301)
			6007520003	
4007220003		5006820003	6007520004	
			6007520005	
4007220003 4007220004		5006820003 5006820004	6007520006	
			6007320001 (Catalent Lot 057G20)	Intended for Clinical/EUA/ Commercial Use
4007220003		5006920006	6007320002 (Catalent Lot 062G20)	
4007220004 4007220005		5006920007	6007320003 (Catalent Lot 001H20)	
4007220002 4007220004		5007320002	6007320005 (Catalent Lot 011J20)	Intended for Clinical/EUA/ Commercial Use
4007220003 4007220005		5007320004		

3.2.P.2.3.7.6.2.1 Description and Justification of Process Changes

The mRNA-1273 Drug Product manufacturing process was scaled-up at ModernaTX, Inc. from personal vaccine unit (PVU) scale to [redacted]-vial scale to provide clinical material. The process

was then transferred to Catalent where it was scaled up to [redacted]-vial scale and [redacted]-vial scale to provide clinical and commercial material. One [redacted] vial lot was used to assess the initial comparability of the [redacted]-vial scale against the baseline expected ranges. A detailed comparison of the clinical manufacturing processes used to date is provided in Table 71.

While analytical method development has been performed concurrently with process development, no analytical method changes have been implemented that impact the comparison of data generated from the tests for the purpose of comparability assessment. Analytical method changes are summarized in Table 61. Full process and analytical development history are described in Section 3.2.P.2.3.7.5.

Table 71: Comparison of Process and Drug Product Presentation Changes mRNA-1273 Drug Product - PVU Process vs Scale A Process (Norwood) vs Scale B Process (Catalent)

Step	Parameter	PVU Process	Scale A Process Norwood (PN 60075)	Scale A Process Catalent (PN 60073)	Scale B Process Catalent (PN 60073)	Comparability Assessment
N/A	Batch Process	Integrated batch from mRNA transcription to mixing [redacted] to mRNA 1273 formulation	mRNA-1273 LNP is supplied in [redacted] bags. DP process includes compounding of mRNA-1273 LNP, sterile filtration, filling into vials, and freezing	mRNA-1273 LNP is supplied in [redacted] bags. DP process includes compounding of mRNA-1273 LNP, sterile filtration, filling into vials, and freezing	mRNA-1273 LNP is supplied in [redacted] bags. DP process includes compounding of mRNA-1273 LNP, sterile filtration, filling into vials, and freezing	Meet demand
N/A	Manufacturing Scale	N/A[a]	[redacted]			Meet demand
N/A	Batch Scale (Nominal)	N/A[a]				Meet demand
Thawing	N/A	N/A; mRNA-1273 LNP is not frozen prior to fill/finish	Bulk Thawing	Passive Thawing	Active Thawing	Decrease processing time for thaw and increase control of LNP thaw
Dilution	DP Target Concentration	0.5 mg/mL	0.20 mg/mL	0.20 mg/mL	0.20 mg/mL	To enable 100 mcg per 0.5 mL dose
Fill	Fill Volume	0.6 mL	5.0 mL	6.3 mL	6.3 mL	Larger fill volume to enable multiple 0.5 mL doses per vial
	Container Closure	Schott 2R Vial with 13 mm PLASCAP	Ompi 10R vial with 20 mm PLASCAP	Ompi 10R vial with traditional stopper and aluminum seal closure	Ompi 10R vial with traditional stopper and aluminum seal closure	Larger vial to enable larger fill volume. Vial sealing system changed to standard serum stopper and crimp seal to accommodate manufacturing at larger scales. Product contact surface material of stopper is unchanged.
Packaging / Labeling	Timing relevant to Fill and Storage	Post-vial inspection Prior to storage.	Flexible **Option 1** Packaged/labeled prior to storage. **Option 2** Packaged and stored. Thawed for labeling. Placed back into storage	Packaged and stored. Thawed for labeling. Placed back into storage	Flexible **Option 1** Packaged/labeled prior to storage. **Option 2** Packaged and stored. Thawed for labeling. Placed back into storage	To allow for flexibility in labeling and packaging.
Visual Inspection	N/A	Manual	Manual	Manual	Semi-Automated Visual Inspection (SAVI)	Alternatives to manual inspection to decrease processing times and enable large scale manufacturing
Freezing	Conditioning	None	None	Initial pre-conditioning freezing at [redacted]	Initial pre-conditioning freezing at [redacted]	To ensure completion of freezing process prior to -20°C storage
Storage	Temperature	-70°C (-60°C to -90°C)	-70°C (-60°C to -90°C)	-20°C (-15°C to -25°C)	-20°C (-15°C to -25°C)	Intended long-term storage condition for commercial presentation

Abbreviations: DP = drug product; LNP = lipid nanoparticle; N/A = not applicable
a) The PVU scale did not have a designated manufacturing scale or nominal batch size.

3.2.P.2.3.7.6.2.2 Process Performance

All post-change comparability lots were manufactured with CPPs controlled within the PARs provided in Table 72. CIPCs met the in-process control expected range provided in Table 73. IPCs control charts were reviewed for consistency as part of the comparability demonstration and all results were within the control limits. Microbial control monitoring was performed per the process microbial control strategy to confirm the process comparability and all results were within the criteria established in the mRNA-1273 Drug Product microbial control strategy (Section 3.2.P.3.4). Process hold times were evaluated against the established ranges and no excursions from the proven acceptable range occurred during manufacture, with the exception of cumulative process duration at 2 – 8°C.

During the manufacture of mRNA-1273 Drug Product lot 6007320005 (Lot 011J20) the total allowable maximum cumulative processing duration at 2 – 8°C was exceeded by [redacted] (Table 72). The major product attribute impacted by exceeding the mRNA-1273 Drug Product Cumulative Process Duration is mRNA purity. The mRNA degradation rate in mRNA-1273 Drug Product at 2 – 8°C [redacted]. Therefore, an additional 1% loss in mRNA purity may be expected in the product as a worst-case estimate due to the additional [redacted] 2 – 8°C prior to freezing. The mRNA purity result for lot 6007320005 (Table 75) is [redacted]. As predicted, considering the additional time at 2 – 8°C, the result is [redacted] purity results demonstrated thus far for mRNA-1273 Drug Product manufacture at: PVU scale (Table 63), Norwood Scale A (Table 64), Norwood Scale A PPQ (Table 65), to Catalent Scale A PPQ (Table 66), but meets the comparability acceptance criteria of [redacted] main peak area (Table 75).

Table 72: mRNA-1273 Drug Product Critical Process Parameters

Step	Process Variable	PAR	6007320005 (Catalent Lot 011J20)
Dilution	Dispensed Dilution Buffer weight	[redacted]	
Stoppering and Capping	Crimp Pressure		
Cumulative Processing Duration	Cumulative process time out of refrigeration (TOR, 15 – 25°C)		
Cumulative Processing Duration	Cumulative process duration at 2 – 8°C		
Vial Freezing and Storage	Vial Conditioning Temperature		Conforms

a) Packaging and labeling activities for mRNA-1273 Drug Product at Scale B ([redacted] vials) took longer than anticipated and as a result the cumulative processing duration at 2-8°C exceeded the PAR. The impact to product quality and scale comparability is assessed above in Section 3.2.P.2.3.7.6.2.2.

Table 73: mRNA-1273 Drug Product In-process Controls

Attribute	Sample Point	Acceptable Range	Classification	6007320005 (Catalent Lot 011J20)
pH	Dilution Buffer Preparation	[redacted]	IPC	[redacted]
Osmolality	Dilution Buffer Preparation	[redacted]	IPC	[redacted]
Post-filtration integrity testing	Dilution Buffer Preparation	[redacted]	IPC	[redacted]
Post-filtration Bioburden	Dilution Buffer Preparation	[redacted]	IPC	[redacted]
Post-filtration Endotoxin	Dilution Buffer Preparation	[redacted]	IPC	[redacted]
Filtration Pressure	Clarification Filtration	[redacted]	IPC	[redacted]
Post-Clarification mRNA Concentration	Clarification Filtration	[redacted]	CIPC	[redacted]
Post-Filtration Integrity Testing	Clarification Filtration	[redacted]	IPC	Not Recorded [a]
Filtration Pressure	Sterilizing Filtration	[redacted]	CIPC	[redacted]
Pre-Filtration Integrity Test Value	Sterilizing Filtration	[redacted]	IPC	[redacted]
Pre-Sterilizing Filtration Bioburden	Sterilizing Filtration	[redacted]	CIPC	[redacted]
Post-Filtration Integrity Test Value	Sterilizing Filtration	[redacted]	CIPC	[redacted]
Fill Weight	Filling	[redacted]	CIPC	Conforms
USP<790> Destructive Visual Inspection	Visual Inspection	Per USP<790> Guidance	CIPC	[redacted]

a) No result was recorded for the Post-Filtration Integrity testing for lot 6007320005 (Catalent lot number: 011J20) A deviation has been opened at Catalent for this event, Deviation REC 266233.

3.2.P.2.3.7.6.2.3 Analytical Comparability

3.2.P.2.3.7.6.2.3.1 Statistical Approach for Analytical Comparability

For each comparison of scales for the manufacture of mRNA-1273, a consistent statistical approach was applied to assess comparability for all quantitative lot release results and selected extended characterization results. The main steps were:

1. Plot the results from each scale in order of manufacturing, with the two scales side-by-side. This plot provided a visual assessment of any trends or shifts in the results along with changes in variability.
2. Calculate a [redacted] confidence, [redacted] coverage tolerance interval using results from preliminary baseline with development and clinical supply, including Scale A PPQ lots.

a. Assess the results against the statistical assumption of normality using a normal quantile plot prior to adopting the calculated interval.

b. If the tolerance interval is wider than the specification limits or extended characterization acceptance criteria, which is a common occurrence for small initial data sets, the tighter set of limits will be applied.

3. Assess comparability by determining the percentage of post-change values that fall in the comparability range. Comparability is demonstrated if at least [redacted] of the new-scale results fall within the comparability range. This will be applied for Preliminary Scale B Comparability N=2 Scale B; and Scale B site start-up comparability of N=3 PPQ Scale B (new process trains at the same site, same scale will perform N=1 PPQ for comparability).

a. When one or more acceptance criteria are not met, investigate to determine if the initial- and new-scale product is comparable.

b. Since the assessment will be performed for a large number of release and extended characterization tests, an occasional excursion beyond the comparability limit may occur due to random variation (the 1% of results which may fall beyond the limits). Any excursions beyond comparability limits will be considered in the context of the full range of testing performed.

In addition to the clinical lots identified in Table 70, statistical analysis included all representative lots manufactured to date to establish the expected analytical ranges. The additional lots are provided in Table 74.

Table 74: Additional Representative mRNA-1273 Drug Product Lots Used for Statistical Analysis

Lot	Scale	Status	Date of Manufacture	References
DHM-47516	[redacted]	Development	01 Apr 2020	QC-OTH-0181
DHM-47519		Development	01 Apr 2020	QC-OTH-0181
6007520007		GMP Ph3	09 Jul 2020	COA-0475

3.2.P.2.3.7.6.2.4 Release Testing

Release testing of mRNA-1273 Drug Product was performed in accordance with the specification listed in Table 75. All results conformed to both the specification and comparability acceptance criteria.

Table 75: mRNA-1273 Drug Product Release Testing

Test	Analytical Method	Specification Acceptance Criteria	Comparability Acceptance Criteria	6007320005 (Catalent Lot 011J20)
Appearance	Visual	White to off-white dispersion. May contain visible, white or translucent product-related particulates.	White to off-white dispersion. May contain visible, white or translucent product-related particulates.	White to off-white dispersion. Essentially Free of particulates.
RNA content	Anion Exchange HPLC	[redacted]	[redacted]	[redacted]
Identity	Reverse Transcription/ Sanger Sequencing	Sequence matches description	Sequence matches description	Conforms
Purity	RP-HPLC	[redacted]	[redacted]	[redacted]
Product-related impurities	RP-HPLC	Report % area for each impurity group: Impurity Group 1 (pre-main peak area) Impurity Group 2 (post-main peak area) Impurity Group 3 (mRNA-adduct species)	[redacted]	[redacted]
% RNA encapsulation	Fluorescence (RiboGreen)	[redacted]	[redacted]	[redacted]
Cell-free Translation	Cell-free-translation/ methionine labeling	[redacted]	[redacted]	[redacted]
Particle size	Dynamic Light Scattering	[redacted]	[redacted]	[redacted]
Polydispersity	Dynamic Light Scattering	Report result	[redacted]	[redacted]
SM-102	UPLC-CAD	Matches retention time of reference	Matches retention time of reference	Conforms
Cholesterol	UPLC-CAD	Matches retention time of reference	Matches retention time of reference	Conforms
DSPC	UPLC-CAD	Matches retention time of reference	Matches retention time of reference	Conforms
PEG2000-DMG	UPLC-CAD	Matches retention time of reference	Matches retention time of reference	Conforms
SM-102	UPLC-CAD	[redacted]	[redacted]	[redacted]
Cholesterol	UPLC-CAD	[redacted]	[redacted]	[redacted]
DSPC	UPLC-CAD	[redacted]	[redacted]	[redacted]
PEG2000-DMG	UPLC-CAD	[redacted]	[redacted]	[redacted]
Lipid impurities	UPLC-CAD	[redacted]	[redacted]	[redacted]
[redacted]	USP <788> Method 2	[redacted]	[redacted]	[redacted]
pH	USP <791>	[redacted]	[redacted]	[redacted]
Osmolality	USP <785> Freezing Point Depression	[redacted]	[redacted]	[redacted]
Container content	USP <697>	[redacted]	[redacted]	[redacted]
Bacterial endotoxin	USP <85>, Ph. Eur. 2.6.14	[redacted]	[redacted]	[redacted]
Sterility	USP <71>, Ph. Eur. 2.6.1	No growth	No growth	No growth

Abbreviations: kDa = kilodalton; RRT = relative retention time; RT = retention time

a) Calculated tolerance interval was wider than mRNA-1273 DP specification. Therefore, the comparability acceptance criteria are the same as the specification values.
b) Calculated tolerance interval lower limit exceeded the mRNA-1273 DP specification. Therefore, the comparability acceptance criteria lower limit is the same as the specification lower limit.
c) Calculated tolerance interval upper limit exceeded the mRNA-1273 DP specification. Therefore, the comparability acceptance criteria upper limit is the same as the specification upper limit.

3.2.P.2.3.7.6.3 Comparability Conclusion

The results from the Scale B ([redacted] vial) batches of mRNA-1273 Drug Product manufactured at Catalent (Bloomington, IN) demonstrated that the pre-change and post-change manufacturing processes and quality attributes were comparable.