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CFTR TaqMan[™] Assay Controls

Catalog Number A30421

Pub. No. MAN0014633 Rev. A.0

Note: For safety and biohazard guidelines, refer to the "Safety" appendix in the *Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) Genotyping Experiments User Guide* (Pub. no. MAN0014405). Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

Product information

The CFTR TaqMan[™] Assay Controls contains 18 tubes of artificial template. Each tube contains a heterozygous mutant allele for 10 to 14 different CFTR assays.

Controls can optionally be included in CFTR TaqMan[™] SNP genotyping experiments. Review "CFTR Controls Assay Index" on page 6 to determine the controls appropriate for your experiment.

Note: The CFTR controls are heterozygous. To demonstrate allelic discrimination between the heterozygous mutant control and homozygous wild type samples, you must also run a homozygous wild type gDNA control.

Use this document in conjunction with the *Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) Genotyping Experiments User Guide* (Pub. no. MAN0014405).

Required materials

Unless otherwise indicated, all materials are available through **thermofisher.com**. MLS: Fisher Scientific

(www.fisherscientific.com) or other major laboratory supplier.

Item	Source				
For OpenArray [™] plate or single-tube format					
CFTR TaqMan™ Assay Controls	A30421				
Homozygous wild type gDNA control (50 ng/uL)	403062 or equivalent				
No Template Control UltraPure [™] DNase/RNase-Free Distilled Water	10977-015				
Centrifuge capable of spinning deep well plates at 200 × <i>g</i> or greater	MLS				
Fisher Scientific™ Analog Vortex Mixer	Fisher Scientific™ 02-215-365				
ТЕ, рН 8.0	AM9849				
PCR reaction mix preparation: Plates and film <i>or</i> tubes:					

Item	Source		
MicroAmp [™] Fast Optical 96-Well Reaction Plate with Barcode, 0.1 mL	4346906		
MicroAmp [™] Optical 96-Well Reaction Plate	4306737		
MicroAmp [™] Clear Adhesive Film	4306311		
Nonstick, RNase-free Microfuge Tubes, 0.5 mL	AM12350		
Additional materials for OpenArray™ plate for	mat		
TaqMan [™] OpenArray [™] Genotyping Master Mix (5 mL)	4404846		
OpenArray™ 384-Well Sample Plates (microplates), black	4482221		
OpenArray™ plate with CFTR assays	Custom ordered		
Biomek Seal and Sample Foil Lids	Beckman Coulter 538619		
OpenArray™ AccuFill™ System Tips	4458107		
QuantStudio [™] 12K Flex OpenArray [™] Accessories Kit (contains the items needed to assemble up to 10 plates: 12 lids and plugs, 12 immersion fluid syringes, and 2 carriers)	4469576		
Instrument and software: • OpenArray [™] Sample Tracker Software • QuantStudio [™] 12K Flex OpenArray [™] Plate F • QuantStudio [™] 12K Flex instrument with Op (AccuFill [™] System)	Press 2.0. enArray™ block		
Additional materials for single-tube format			
TaqMan™ Genotyping Master Mix (1 × 1 mL)	4371353		
TaqMan [™] Genotyping Master Mix (1 × 10 mL)	4371355		
TaqMan™ Genotyping Master Mix (1 × 50 mL)	4371357		
TaqMan [™] Genotyping Master Mix (2 × 10 mL)	4381656		
TaqMan™ Genotyping Master Mix (2 × 50 mL)	4381657		
CFTR assays, 20X Assay Working Stock	Custom ordered		
MicroAmp [™] Optical Reaction Plates, any of the f	ollowing:		
MicroAmp [™] Fast Optical 96-Well Reaction Plate with Barcode, 0.1 mL	4346906		
Either of the following: • UltraPure [™] DNase/RNase-Free Distilled Water	10977-015		

• TE, pH 8.0 (1X TE buffer)

AM9849

Item	Source				
MicroAmp [™] Optical 96-Well Reaction Plate with Barcode	4306737				
MicroAmp [™] Optical 384-Well Reaction Plate with Barcode	4309849				
MicroAmp [™] Optical Adhesive Film	4360954				
Instruments and software required:					
QuantStudio [™] 12K Flex instrument or thermal cy	ycler				

Prepare and run CFTR controls: OpenArray[™] plate experiments

Prepare control PCR reactions: OpenArray[™] plate format

- 1. Thoroughly thaw the CFTR and gDNA control tubes.
- 2. Vortex the tubes to mix.
- **3.** In a 96-well Fast or standard plate or in non-stick 0.5 mL tubes, prepare a 1:10 dilution of each of the 18 CFTR controls in 1X TE buffer.

Diluted controls are stable for up to 1 month at 4°C.

- **4.** Seal the plate with MicroAmp[™] Clear Adhesive Film or cap the tubes.
- 5. Vortex to ensure thorough mixing, then centrifuge.
- 6. Mix the 2X Master Mix gently. Do not invert the bottle.
- 7. Vortex the Assay Working Stock, then centrifuge briefly.
- 8. For each OpenArray[™] plate to be amplified, pipette components into the 384-well OpenArray[™] plate:

Component	Volume
TaqMan™ OpenArray™ Genotyping Master Mix	2.5 µL
 One of the following: UltraPure[™] DNase/RNase-Free Distilled Water (NTCs) 1:10 dilution of the CFTR control^[1] gDNA control 	2.5 µL
Total reaction volume	5.0 µL

^[1] Add only one control to each well.

- **9.** Seal the plate with an aluminum foil seal, remove the foil flap, mark the edges of the filled 4 × 12 area with a pen, then score the foil along those lines. Do not remove the foil from the scored area at this time.
- **10.** Vortex the sealed plate for 5 seconds, then centrifuge the plate for 1 minute at 1,000 rpm. Do not vortex at too vigorous a setting, to avoid excessive bubble formation.

Set up the AccuFill[™] instrument

IMPORTANT! Before proceeding, check the tip expiration date (shown on the outer box that contains the trays of tips). Do not use tips that exceed the expiration date.

- 1. In the OpenArray[™] AccuFill[™] software, click **Setup and Load**.
- 2. In the Setup Load Information window, verify that the Use Sample Integration checkbox is selected.
- **3.** Click **Browse** to the right of Sample Plate, then select the 384well sample plate .csv file that was generated with Sample Tracker Software.
- Click Browse to the right of the plate holder position corresponding to the OpenArray™ of interest, then select the .spf file corresponding to the desired OpenArray™ plate.
- **5.** Click the corresponding 4 × 12 area of the 384-well plate, then click **Next** to open the Setup Deck window.
- 6. Ensure that:
 - Tip boxes are loaded in the AccuFill[™] instrument in the displayed configuration.
 - Lids are removed from the tip boxes.
 - The waste bin in the instrument is emptied.
- 7. In the Setup Deck window:
 - a. Click the **The tips are configured as shown above** checkbox.
 - b. Click the The Waste Bin is empty checkbox.

Transfer reactions to the OpenArray[™] plate in the AccuFill[™] instrument

- Prepare the items needed to seal the OpenArray[™] plates (next section), because the OpenArray[™] plate must be sealed promptly after being loaded with the reactions (this section).
 - **a.** Ensure that the QuantStudio[™] OpenArray[™] Plate Press is ready.
 - b. Gather and remove from its packaging an OpenArray[™] Lid, plug, syringe with OpenArray[™] Immersion Fluid, and syringe tip.
 - **c.** Attach the syringe tip to the syringe and carefully push some of the fluid through the tip to remove air bubbles, then lay the syringe aside.
- 2. Remove the OpenArray[™] plate from its sleeve and place it in the Plate Holder of the AccuFill[™] instrument.

Ensure that the bar code on the OpenArray^m plate is facing left and the serial number is facing right.

- **3.** Using forceps, peel off the foil over the 384-well plate area being used.
- 4. Close the instrument door.

- 5. In the AccuFill[™] software Setup Deck window:
 - a. Click the **The OpenArray Plate is in the Plate Holder** checkbox.
 - **b.** Click the **Remove foil from the highlighted section of the Sample Plate** checkbox.
 - c. Click Load.
- As soon as the Remove OpenArray Plate window appears (after ~90 seconds), open the instrument door, and remove the loaded OpenArray[™] plate.
- 7. Proceed immediately to seal the OpenArray[™] plate (next section).

Note: For best results, seal the OpenArray[™] plate within 90 seconds of completion of loading, to prevent evaporation.

Seal the OpenArray[™] plate

IMPORTANT! Handle the OpenArray[™] plate and case only by the edges throughout this procedure.

 Place the filled OpenArray[™] plate in the QuantStudio[™] 12K Flex OpenArray[™] Plate Press 2.0.

Ensure that the bar code is facing left and the serial number is facing right.

 Remove the clear plastic sheets from the top and the bottom of the lid, remove the red protective film around the edge of the OpenArray[™] lid, then seat the lid on the OpenArray[™] case in the plate press.



- (1) Protective film (remove)
- 2 Adhesive
- ③ Protective film (remove)
- ④ Notched end (align with serial number)
- **3.** Engage the press mechanism until the green flashing light changes to a steady green light (~20 seconds).
- **4.** Disengage the press, then remove the OpenArray[™] case.

5. While holding the OpenArray[™] case by the edges, insert the prepared syringe tip into the port in the case and carefully inject immersion fluid until the case is filled.



It is important that the syringe tip is in front of the array when filling the case with immersion fluid.

Minimize creation of air bubbles when you dispense the fluid; one small air bubble in the case is acceptable.

- 6. Continuing to hold the case vertically, remove the syringe tip, insert the screw end of the OpenArray[™] Plug into the port and rotate clockwise until the black handle breaks off.
- 7. Clean the case with a laboratory wipe that has been thoroughly sprayed with ethanol, then dry the case with a clean laboratory wipe.

Run the OpenArray[™] plate(s) on the QuantStudio[™] 12K Flex instrument

On the instrument touchscreen, touch to extend the loading arm, and place the OpenArray[™] plate(s) on the plate adapter.

Ensure that the plate barcode and serial number are facing the front of the instrument.

- **2.** Touch **a** to retract the loading arm.
- 3. On the Home screen of the QuantStudio[™] 12K Flex Software, select **Run > OpenArray**.
- 4. In the Select Instrument pane, select the QuantStudio[™] instrument.
- Click Get Plate IDs to import the barcode(s) of the OpenArray[™] plate(s).

Once the OpenArray[™] serial numbers appear, the loaded .spf files corresponding to each plate should appear in the Setup File field.

If not, click **Browse**, then select the correct loaded .spf file from the Loaded SPF folder.

- 6. (*Optional*) If desired, click **Browse** to change the QuantStudio[™] Experiment File Location.
- **7.** (*Optional*) If desired, change the software-determined Experiment File Name.

8. On the instrument touchscreen, touch Start Run.

The instrument pauses at 41 or 42 seconds prior to the end of the run. Wait for the system to complete the run before opening the .eds file.

9. Transfer the .eds file from the instrument to an accessible location for analysis in Genotyper Software.

Prepare and run CFTR controls: single-tube experiments

Prepare control PCR reactions: Single tube

- 1. Thoroughly thaw the CFTR and gDNA control tubes.
- 2. Vortex the tubes to mix.
- **3.** In a 96-well Fast or standard plate or in non-stick 0.5 mL tubes, prepare a 1:100 dilution of each of the 18 CFTR controls in 1X TE buffer.

Diluted controls are stable for up to 1 month at 4°C.

- **4.** Calculate the number of reactions needed for each assay. Include at least 2 NTCs for each assay.
- **5.** Calculate the volume of each component needed. Include extra reactions to compensate for the volume loss that occurs during pipetting.
- 6. Mix the 2X Master Mix gently. Do not invert the bottle.
- 7. Vortex the Assay Working Stock, then centrifuge briefly.
- 8. Pipette the following volumes per reaction into a 96-well, Fast 96-well, or 384-well plate:

Component	384-Well Plate	96-Well Fast Plate	96-Well Plate
TaqMan [™] Genotyping Master Mix	2.50 µL	5.00 µL	12.50 µL
CFTR TaqMan [™] SNP Genotyping Assay	0.25 µL	0.50 µL	1.25 µL
UltraPure™ DNase/RNase-Free Distilled Water	0.25 µL	0.50 µL	1.25 μL
One of the following: • UltraPure™ DNase/RNase- Free Distilled Water (NTCs) • 1:100 dilution CFTR control ^[1] • gDNA control	2.00 µL	4.00 µL	8.00 µL
Total reaction volume	5.00 µL	10.00 µL	25.00 µL

^[1] Add only one control to each well.

Perform PCR (single-tube assays)

Follow the instructions in the instrument user guide for this procedure.

1. Set the following thermal cycling parameters:

Step Temperature		Duration	Cycles	
Activate	95°C	10 minutes	HOLD	
Denature 95°C		15 seconds	40	
Anneal/ Extend	60°C	1 minute	40	

If you are using a real-time PCR instrument, select **Standard** mode in thermal cycler settings.

IMPORTANT! Do not use Fast Mode thermal cycling conditions with TaqMan[™] Genotyping Master Mix, even when using Fast plates, blocks, or thermal cyclers.

2. Set the following reaction volumes:

MicroAmp [™] Optical Reaction Plate	Reaction volume
384-well	5.00 µL
Fast 96-well	10.00 µL
96-well	25.00 µL

3. Load the plate and start the run.

Analyze CFTR controls

Analyze the control assays in the TaqMan[™] Genotyper Software

- 1. Create a study, then upload the completed CFTR control genotyping experiment EDS files into the study.
- 2. Check the Analysis Settings are set to default.

Analysis Setting	s					
믿 Set the defa	ult analysis settings.					
Call Settings	Control Identifiers	QC Settings				
Study-Level S	ettings					
Call Method: @	Autocalling 💿 Classif	ication Scheme	2			
	Multiplate Analysis:	V	Use Hardy-Weinberg for Analysis:			_
	Protect Manual Calls:	V	Use Positive Controls for Analysis:	V	Baseline for Real-time Data:	V
Lise Reference	Panels for Autocalling:		Heterozygote:	Allow	Cycle Number:	

3. Click Analyze.

Each CFTR assay allele discrimination plot displays data points for all CFTR Controls run in the experiment.

In this example (obtained using the QuantStudioTM 12K Flex software), the data points marked with a green dot are for the CFTR Control designed to the heterozygous mutant for this assay. The data points marked with a red dot are wild type gDNA samples. The data points marked with x are from non-specific activity for the CFTR Controls that are not designed specifically for this assay.



- **4.** Identify the corresponding control for each CFTR assay (see "CFTR Controls Assay Index" on page 6).
- **5.** In the Results Table, select all control samples that *are not* associated with the assay.

6. Right-click, then select **Omit** to hide these samples in the data plot.

Resu	lts	Ref	erence Samples	Pop	ulation S	tatistics					
	View 🗸		Group by 🗸	E B	ookmai	rk 🗸 🧮	, Tag 🗸 🛔	GT Chang	e Call 🗸		
#			Sample ID		₹1	Call	Manual	Quality	VIC®	F	
33			CFTR Control 14		A/A			0.00	12.929		
34			CFTR Control 14		A/-			0.00	35.852		
35			CFTR Control 13		-/-			0.20	78.128		
36			CFTR Control 13		No Ar	mplificatio		0.84	198.856		
37			CFTR Control 12		Under	inplificatio		0.69	184.589		-
38			CFTR Control 12		Under	termined		0.00	53.761		
39			CFTR Control 11		Possit	ole Kare Al	lele	0.00	36.108		
40			CFTR Control 11		Clear	Manual C	all	0.99	81.564		
41			CFTR Control 10		Omit			0.00	41.460		Ε
42			CFTR Control 10		Un-or	nit		0.00	55.705		
43			CFTR Control 1		Tag fo	or Ref Pan	el	0.39	2,884.600	2,	
44			CFTR Control 1		Unsta	a Selected		0.45	1,603.208	1,	
45			CFTR Control 1			y selected		0.46	3,143.828	2,	ſ
46			CEPH gDNA		Set Bo	okmarks		0.98	566.726	з,	Ŧ
	•		111		Select	All Bookn	narks			Þ.	

The plot displays traces for all controls that are specific to the assay.



7. Repeat the steps above for each assay.

(Optional) Generate a Reference Panel

For best results, generate a new Reference Panel for each lot of OpenArray[™] plates.

If necessary, first create a study containing data that will be used as reference samples. Set **QuantStudio™ 12K Flex Real-Time PCR System** for Instrument Type, and **Real-time** for Experiment Type in the study properties.

- 1. In TaqMan[™] Genotyper Software, open the study that contains data points to be used as reference samples.
- 2. In the Workflow Menu pane, select Analysis > Results.
- **3.** Click-drag in the scatter plot to select one or more data points.

Select data points for each homozygous genotype (FAM[™] dye-labeled or VIC[™] dye-labeled) and the heterozygous genotype.

4. Right-click in the scatter plot, then select Tag for Ref Panel.

- 5. Repeat the selection and tag steps for additional data points.
- **6.** Confirm that the correct samples have been tagged as reference samples in the Results table.

In the Results tab, **Reference Sample** must be checked in the **View** dropdown list to view the tags.

- **7.** Repeat the data point selection, tag, and confirmation steps for each SNP assay of interest.
- 8. Select File → Save.
- **9.** Select **File Generate Reference Panel**, enter a name for the panel and save the file (.lap) to the desired location.

(*Optional*) Add samples to an existing Reference Panel

During analysis in Genotyper Software, you can add samples to an existing Reference Panel.

The Reference Panel must already be imported into the study containing samples that will be added to the Reference Panel.

- In the scatter plot of the study, select data points for the samples of interest, right-click in the plot, then select Tag for Ref Panel.
- 2. Select File → Save.
- **3.** Select **File Generate Reference Panel**, enter a name for the panel and save the file (.lap) to the desired location.

Target Name	Assay ID	CFTR control number	Target Name	Assay ID	CFTR control number
1259insA	C_201385097_10	CFTR1	405+1G>A	C_151693789_10	CFTR9
2307insA	C_172767551_30	CFTR1	711+5G>A	C_100964812_20	CFTR9
3821delT	C_151693965_20	CFTR1	A455E	C_191740225_10	CFTR9
936delTA	C_151693831_20	CFTR1	G330X	C32295672_20	CFTR9
c.1486T>G	C_203097281_10	CFTR1	1507V	C8853252_20	CFTR9
D579G	C_201390122_10	CFTR1	W1282X	C32545014_20	CFTR9
E1371X	C_201404896_10	CFTR1	1154insTC	C_201380060_10	CFTR10
L927P	C_201395153_10	CFTR1	1898+3A>G	C_201390132_10	CFTR10
M1V	C_201392609_10	CFTR1	405+3A>C	C_100964777_10	CFTR10
Q359K/T360K	C_203006811_10	CFTR1	711+1G>T	C_100964811_10	CFTR10
R117H	C26083773_20	CFTR1	L967S	C8853384_10	CFTR10
R553X	C27861430_20	CFTR1	R764X	C27531535_30	CFTR10
Y1092X[c.3276C>A]	C_26083714C_10	CFTR1	S1255X	C26083699_10	CFTR10
1213delT	C_201380061_10	CFTR2	S466X(c.1397C>G)	C_64676233D_20	CFTR10
1949del84	C_151693911_10	CFTR2	S549N	C32545131_20	CFTR10
2585delT	C_201409799_10	CFTR2	Y1092X(c.3276C>G)	C_26083714D_20	CFTR10
3120G>A	C_151693932_20	CFTR2	2105-2117del13insAGAAA	C_172767549_10	CFTR11
4382delA	C_201404927_10	CFTR2	3171delC	C_151693938_20	CFTR11
444delA	C_151693816_20	CFTR2	Е92К	C_32545261C_10	CFTR11
852del22	C_151693827_10	CFTR2	P205S	C32545236_20	CFTR11
CFTRdele22.23_mu	C99000003A_10	CFTR2	Q552X	C32545126_20	CFTR11
CFTRdele22.23_wt	C99000003B_20	CFTR2	R1162X	C64676246_10	CFTR11
G551D	C32545127_20	CFTR2	R1283M	C32545013_10	CFTR11
P5L	C_191876963_20	CFTR2	R347P	C656878C_30	CFTR11
Q1238X	C32545039_20	CFTR2	S492F	C656861_20	CFTR11

CFTR Controls Assay Index

Target Name	Assay ID	CFTR control number	Target Name	Assay ID	CFTR control number
S489X	C_201387593_10	CFTR2	W846X (c.2538G>A)	C_201166057_10	CFTR11
W1089X	C26083715_30	CFTR2	1924del7	C_201390137_10	CFTR12
CFTRdele2.3_wt	C99000002B_20	CFTR3	2347delG	C_201392634_10	CFTR12
1548delG	C_201387587_10	CFTR3	711+3A>G	C_201407349_10	CFTR12
1898+5G>T	C_151693907_10	CFTR3	A559T	C27861435_20	CFTR12
2942insT	C_191876972_20	CFTR3	E92X	C_32545261D_20	CFTR12
3849+10kbC>T	C_100965026_20	CFTR3	G1244E	C_201166058_10	CFTR12
CFTRdele2.3_mu	C99000002A_10	CFTR3	1507del	C_151693868_10	CFTR12
G1069R	C_191751848_30	CFTR3	L1077P	C26083717_30	CFTR12
I336K	C_201385076_10	CFTR3	R334L	C_201385074_10	CFTR12
Q39X	C_201385102_10	CFTR3	S945L	C_201395160_10	CFTR12
R560G	C_201390094_10	CFTR3	W401X(c.1202G>A)	C_201385107_10	CFTR12
R709X	C32295645_30	CFTR3	3007delG	C_201395164_10	CFTR13
V520F	C32545166_10	CFTR3	712-1G>T	C_151693825_10	CFTR13
Y122X	C656917_30	CFTR3	E822X	C_201392657_10	CFTR13
1288insTA	C_151693839_10	CFTR4	G480C	C656863_30	CFTR13
1812-1G>A	C_151693905_10	CFTR4	G551S	C32545128_20	CFTR13
2183AA>G	C172767555C_30	CFTR4	G622D	C27536988_20	CFTR13
2789+2insA	C_201395133_10	CFTR4	M1101K	C32295578_10	CFTR13
574delA	C_151693817_20	CFTR4	Q98R	C_201397567_10	CFTR13
5T	C203006801C_10	CFTR4	S1255P	C26083700_20	CFTR13
delF311	C_151693835_20	CFTR4	2043delG	C_151693912_20	CFTR14
E60X	C32295754_20	CFTR4	2622+1G>A	C_172948526_20	CFTR14
N1303K	C32544994_20	CFTR4	3120+1G>A	C_100964958_20	CFTR14
Q525X	C_201387609_10	CFTR4	3905insT	C_172767553_20	CFTR14
R1070Q	C32295591_20	CFTR4	D110H	C656922_20	CFTR14
W1204X(c.3611G>A)	C26083704_30	CFTR4	E1104X	C_201400019_10	CFTR14
1461ins4	C_201387572_10	CFTR5	H199Y	C32295707_20	CFTR14
1677delTA	C_151693870_20	CFTR5	Q493X	C656860_20	CFTR14
2108delA	C_151693913_20	CFTR5	R352Q	C11399128_20	CFTR14
3199del6	C_151693939_20	CFTR5	S549R(c.1645A>C)	C32545132_20	CFTR14
3659delC	C_151693962_20	CFTR5	1717-8G>A	C_191876965_10	CFTR15
9T	C203006801D_20	CFTR5	S364P	C32545211_20	CFTR13
c.4028delG	C_203097285_10	CFTR5	2869insG	C_172767552_10	CFTR15
D836Y	C_191861136_20	CFTR5	3791delC	C_172767557_20	CFTR15
G178R	C11399242_20	CFTR5	4209TGTT>AA	C_203006827_10	CFTR15
P67L	C_200992880_10	CFTR5	457TAT>G	C_172767554_10	CFTR15

Target Name	Assay ID	CFTR control number	Target Name	Assay ID	CFTR control number
R560T	C_27861436C_30	CFTR5	E585X	C_201390130_10	CFTR15
T338I	C656880_30	CFTR5	K710X	C26083748_20	CFTR15
2143delT	C_151693914_30	CFTR6	Q220X	C_201407366_10	CFTR15
4005+1G>A	C_173080716_10	CFTR6	R1066C	C26083724_20	CFTR15
663delT	C_151693822_20	CFTR6	S466X(c.1397C>A)	C_64676233C_10	CFTR15
G542X	C11399026_30	CFTR6	T351S	C656875_20	CFTR15
G85E	C32545286_20	CFTR6	1525-1G>A	C_201387583_10	CFTR16
1506V	C59055679_20	CFTR6	2184delA	C_172767547_20	CFTR16
P1013H	C_191876978_20	CFTR6	2789+5G>A	C_100964951_10	CFTR16
R1158X	C32545051_10	CFTR6	406-1G>A	C_151693815_10	CFTR16
R334W	C656883_30	CFTR6	I1234V	C32545041_10	CFTR16
R851X	C656779_20	CFTR6	L227R	C_201407370_10	CFTR16
W401X(c.1203G>A)	C_201385108_10	CFTR6	Р574Н	C32545116_20	CFTR16
1717-1G>A	C_100964916_20	CFTR7	R1070W	C_191830125_20	CFTR16
1898+1G>A	C_151693906_10	CFTR7	R352W	C_191876959_20	CFTR16
3876delA	C_151693993_20	CFTR7	S549R(c.1647T>G)	C32545130_20	CFTR16
621+1G>T	C_100964806_20	CFTR7	1471delA	C_201387575_10	CFTR17
c.3297C>A	C_203097284_10	CFTR7	2184insA	C_172767550_10	CFTR17
G970R	C_201395170_10	CFTR7	C524X	C32545165_20	CFTR17
L467P	C_172861700_20	CFTR7	G1349D	C26083686_30	CFTR17
L732X	C_201392631_10	CFTR7	L1065P	C26083725_20	CFTR17
R75X	C64676210_20	CFTR7	L206W	C32545235_20	CFTR17
S341P	C_201385079_10	CFTR7	Q890X	C32295630_20	CFTR17
1078delT	C_172767546_20	CFTR8	Q98X	C_201397564_10	CFTR17
1341+1G>A	C_201385109_20	CFTR8	R347H	C656878D_20	CFTR17
3121-1G>A	C_201397573_10	CFTR8	S1196X	C32295560_20	CFTR17
394delTT	C_151693788_20	CFTR8	Y569D	C_201390110_10	CFTR17
D1152H	C32545067_20	CFTR8	1248+1G>A	C_201385092_10	CFTR18
D614G	C_191856000_20	CFTR8	1811+1.6kbA>G	C_201390100_10	CFTR18
E831X	C_201395108_10	CFTR8	2055del9>A	C_172767548_10	CFTR18
F508del	C_151693869_10	CFTR8	935delA	C_151693830_20	CFTR18
R170H	C8853206_30	CFTR8	F508C	C_100964862_10	CFTR18
R560K	C_27861436D_20	CFTR8	L558S	C_191876966_20	CFTR18
S1251N	C32545018_20	CFTR8	Q1313X	C32544992_10	CFTR18
1833delT	C_201390108_10	CFTR9	R1066H	C26083723_20	CFTR18
2183delAA	C172767555D_20	CFTR9	R117C	C27540773_10	CFTR18
2711delT	C_201395120_10	CFTR9	W1204X(c.3612G>A)	C_151693964_20	CFTR18

Target Name	Assay ID	CFTR control number	Target Name	Assay ID	CFTR control number
3272-26A>G	C_100964965_10	CFTR9	W846X (c.2537G>A)	C_201395114_10	CFTR18
3667del4	C_151693963_20	CFTR9	NA	NA	NA

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Table 1 Revision history of Pub. no. MAN0014633

Revision	Date	Description
A.0	25 February 2016	New document

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