



# AmplideX<sup>®</sup> *FMR1* Controls



**DISCOVER**  
**DEVELOP**  
**DIAGNOSE**

Asuragen is a molecular diagnostics company using genomics to drive better patient management through best-in-class clinical testing solutions.

# AmplideX<sup>®</sup> *FMR1* Controls

## Applications for gDNA Control mixtures:

- Assure amplification of GC rich templates across different size categories of CGG-repeats (NOR, INT, PM, and FM)
- Calibration of mobility of GC-rich PCR products on different platforms (sequence confirmed CGG sizes of alleles up to 116 CGGs)
- Amplification of low levels of FM alleles in presence of NOR and PM alleles (artificial mosaic sample)
- Different methylation states (un-, partially-, and fully methylated alleles) across different size ranges

# AmplideX<sup>®</sup> *FMR1* Controls

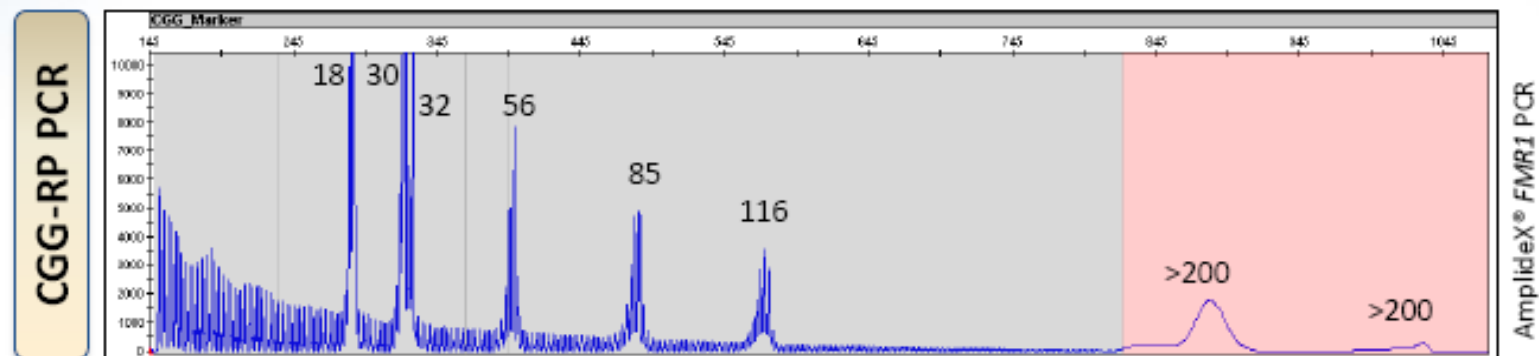
Asuragen proprietary cell line gDNA mixtures containing multiple alleles that can be used as controls in AmplideX<sup>®</sup> *FMR1* PCR and mPCR assays.

## 1. AmplideX<sup>®</sup> PCR Process Control

contains gDNA templates for amplification of 7 alleles used to calibrate instrument sizes and as a routine batch control. 18, 30, 32, 56, 85, 116 and >200 CGGs

- use 2  $\mu$ L in AmplideX<sup>®</sup> PCR, or lab developed *FMR1* PCR
- use 2x4  $\mu$ L in AmplideX<sup>®</sup> mPCR (4 $\mu$ L for each FAM and HEX reaction)

# 1. AmpliX<sup>®</sup> PCR Process Control



# Derivation of Correction Factors Using AmpliDeX<sup>®</sup> PCR Process Control

- Obtain sizes for peaks corresponding to 18, 30, 32, 56, 85 and 116 CGG (in mPCR, use FAM channel).
- Perform linear fit and use slope for mobility correction factor and intercept for size correction factor.
- Determine unknown CGG repeat number by using peak size [bp]:

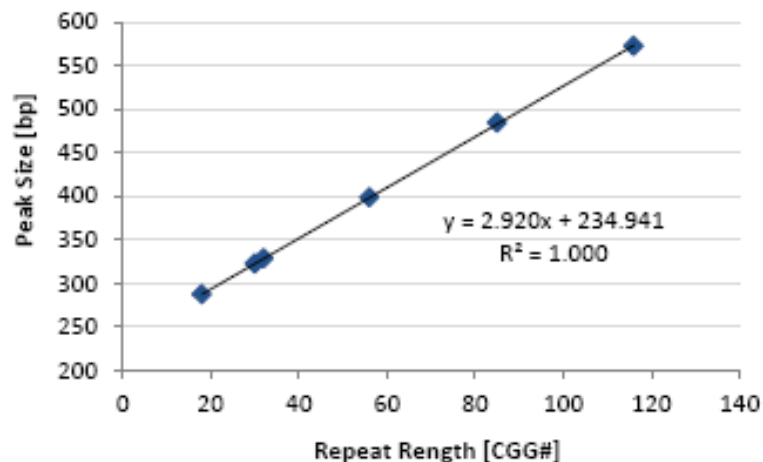
CGG repeats	Size
18	287.21
30	322.43
32	328.28
56	398.45
85	484.49
116	572.68

$C_0$  234.941

$m_0$  2.920

$$CGG_i = \frac{Size_i - c_0}{m_0}$$

AmpliDeX Process Control Fit



Repeat Range	Sizing Accuracy
<70 CGG	+/- 1 CGG
71-120 CGG	+/- 3 CGG
121-200 CGG	+/- 5 CGG
>200	Full mutation