

## > 2X Master HotStart

| <i>cat. no.</i>    | <i>amount</i> | <i>note</i>                       |
|--------------------|---------------|-----------------------------------|
| STS-HXMMixW 200*   | 5ml           | 2X Hot Start XtraWhite Master Mix |
| STS-HXMMixW 1000*  | 25ml          | 2X Hot Start XtraWhite Master Mix |
| STS-HXMMixRTL 200  | 5ml           | 2X Hot Start XtraRTL Master Mix   |
| STS-HXMMixRTL 1000 | 25ml          | 2X Hot Start XtraRTL Master Mix   |

\*2X Hot Start XtraWhite Master Mix is supplied with appropriate quantity of 6X Loading Dye

2X Hot Start XtraWhite Master Mix and 2X Hot Start XtraRTL Master Mix (Genespin proprietary formulation), are two premixed, ready-to-use solution containing Hot Taq Pol, dNTPs and MgCl<sub>2</sub> in a Reaction Buffer optimized for use in PCR amplification of targets present in low copy number and to avoid amplification of non-specific products. Both buffers contain 3.0mM magnesium, PCR enhancers and thickening agents (vortex thoroughly prior to use).

Both Master Mixes allow for specific PCR amplification by keeping the enzyme inactive until the temperature reaches approximately 40°C, while also reducing samples preparation time as well as risk of contamination from multiple pipetting steps.

2X Hot Start XtraRTL Master Mix contains Orange G dye that allows gel loading and electrophoresis of the sample directly from the PCR\* tube, without further manipulation. 2X Hot Start XtraWhite Master Mix is supplied with appropriate quantity of 6X Loading Dye. The Orange G dye migrates at the same rate as a duplex DNA fragment of approximately 50 Kbp and does not interfere with DNA migration when it is used as a loading dye for agarose gel electrophoresis.

### FOR RESEARCH USE ONLY

#### SHIPPING

Shipped in green ice.

#### STORAGE

Store at -20C°. Avoid freeze/thaw cycles.

#### SHELF LIFE

12 months

#### FORM

Liquid

#### CONCENTRATION

2X conc.

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### Assay Set-Up:

Before starting, vortex all components thoroughly to ensure homogeneity.

Prepare a premix for the number of assays you need according to the following protocol:

| component    | stock conc. | final conc.  | 30ul reaction |
|--------------|-------------|--------------|---------------|
| 2X Hot Start | 2X          | 1X           | 15.0ul        |
| primers      | 1ug/ul each | 50ng/ul each | 2ul each      |
| DNA Template | -           | -            | 10-20ng       |
| MG Water     | -           | -            | up to 30ul    |

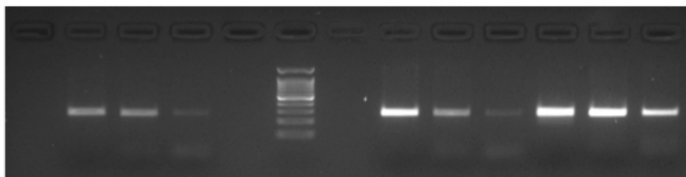
### Cycling conditions:

Spin down the tubes/plate briefly to remove bubbles and place them into the cycler.

|               |         |        |        |
|---------------|---------|--------|--------|
| denaturation  | 95°C    | 5 min  | 1x     |
| denaturation  | 95°C    | 30 sec | 20-35x |
| annealing (1) | 50-68°C | 30 sec |        |
| extension (2) | 72°C    | 30sec  |        |

1) The annealing temperature depends on the melting temperature of the primers used.

2) The elongation time depends on the length of the fragments to be amplified. A time of 1 min/kb is recommended.



2X Competitor

2X Hot Start  
MMix GeneSpin

2X Hot Start  
XtraWhite Master Mix

Comparisons of the amplification efficiency of new **2X Hot Start XtraWhite** and old **GeneSpin 2X HMMix** versus Competitor 2X PCR Master Mix were performed using serial dilutions (1:2, 1:20, 1:200) of human genomic DNA and specific primers corresponding to a centromeric region (SatCen11). New **2X Hot Start XtraWhite** shows higher efficiency in amplifying the centromeric region than old GeneSpin 2X HMMix and the Competitor 2x PCR Master Mix.