

AccuStart[™] MarkerSelect PCR SuperMix ROX[™]

Cat. No.	95104-250	Size:	250 x 20-µL reactions (2 x 1.25 mL)
	95104-012		1250 x 20-µL reactions (10 x 1.25 mL)
	95104-05K		5000 x 20-µL reactions (1 x 50 mL)

Store at -20°C protected from light

Description

AccuStart MarkerSelect PCR SuperMix is a 2X concentrated, ready-to-use reaction cocktail for routine PCR amplification of DNA fragments for characterization with allelic discrimination assays. It contains all components, except primers, probes and template. AccuStart MarkerSelect PCR SuperMix simplifies reaction assembly, improves reproducibility, and reduces the risk of contamination. A key component is AccuStart MarkerSelect PCR polymerase, which contains monoclonal antibodies that bind to the polymerase and keep it inactive prior to the initial PCR denaturation step. Upon heat activation the antibodies denature irreversibly enabling specific and efficient primer extension and probe utilization with the convenience of room temperature reaction assembly. AccuStart MarkerSelect mixes also contain additives to enable robust and accurate genotyping from agricultural and environmental samples that might contain PCR inhibitors.

Components

AccuStart MarkerSelectPCR SuperMix

2X reaction buffer containing MgCl₂, dNTPs (dATP, dCTP, dGTP, dTTP), AccuStart Taq DNA Polymerase, ROX reference dye, and stabilizers.

Storage and Stability

AccuStart MarkerSelect PCR SuperMix is stable for 1 year when stored in a constant temperature freezer at -20°C and protected from light. For convenience, it may be stored unfrozen at +2 to +8°C for up to 6 months. No loss of performance was demonstrated after 20 freeze-thaw cycles or 2 months at +20°C.

Reaction Assembly

Component	Volume for 20-µL rxn.	Final Concentration
AccuStart MarkerSelect PCR SuperMix ROX (2X)	10 µL	1X
Primer & Probe	Variable	0.5X to 1X
Nuclease-free water	Variable	
DNA Template	<u>0.5 – 5 μL</u>	Variable
Final Volume (µL)	20 µL	

Note: For smaller reaction volumes (i.e. 5-µL reactions), scale all components proportionally.

Reaction Protocol

Incubate the completed reaction mix in thermal cycler as follows:

Initial denaturation:	95°C, 5 min (up to 10 min for difficult templates)
PCR cycling (20 – 40 cycles:)	95°C, 5 s
	60°C, 30s
Hold	4°C until processed for analysis

Full activation of AccuStart Taq DNA polymerase occurs within 30 seconds at 95°C. Complete denaturation of dsDNA target is important for efficient PCR amplification and may require different initial denaturation times depending on the properties of a given target sequence. Cycling protocol may require adjustment for longer amplicons (>150bp).

Quality Control

Kit components are free of contaminating DNase and RNase and validated in PCR performance tests.

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