



Precipio, Inc.

Instructions for Use

**NRAS Exon 4 (117) Mutation Analysis using ICE COLD-PCR
for Detection with High Resolution Melting**

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Manufacturer

This Kit was manufactured by Precipio, Inc. at 8813 F Street, Omaha, NE 68127, USA.
USA Telephone 1-203-787-7888

Intended Use

For professional use only. Precipio's ICEme Kit offerings are *in vitro* diagnostic assays that enrich clinically actionable somatic mutations in a target of interest. This kit is specifically designed to enrich for mutations in Exon 4 (codon 117) of the NRAS gene that are of clinical relevance. This kit is designed to be used in a clinical diagnostic laboratory by suitably trained personnel testing DNA extracted from formalin-fixed paraffin embedded tissues or from plasma.















Reagent Preparation

All reagents supplied with this kit are ready to use. Some components will need to be thawed, vortexed and/or spun in a microcentrifuge before use; check details in Assay Procedure below. Reagents do need to be combined to produce Master Mixes and reaction mixtures; full details are given in the procedure below.

Kit Components and Storage Conditions

The NRAS Exon 4 (117) Kit contains the components to perform ICE COLD-PCR amplification for mutation enrichment as well as High Resolution Melting for 24 total samples with controls. Enough reagents, including controls, are present to accommodate 4 batches of 6 samples. Recommended storage conditions are listed in **Table 1**.

Table 1: Kit components with recommended storage conditions.

Reagent, ICE COLD-PCR	Lid Color	Color of Label	Number of Tubes	Volume Total (μL)	Storage (°C)
10X Primer/RS-Oligo Mix	Clear 	Blue 	1	290	-20
2X Polymerase Master Mix	Clear 	Green 	4	360	-20
Wild-Type Control	Clear 	White 	1	66	-20
1% K117N Control	Clear 	Red 	1	66	-20
Reagent, HRM	Lid Color	Color of Label	Number of Tubes	Volume Total (μL)	Storage (°C)
HRM Primer Mix	Clear 	Yellow 	1	60	-20
2X HRM Master Mix	Black 	White 	1	960	-20
Sanger Sequencing Primer, 10 μM*	Clear 	Orange 	1	60	-20

* Can be used for uni-directional Sanger Sequencing confirmation.

This kit was designed specifically to enrich the following mutations:

1. c.351G>T; p.K117N
2. c.351G>C; p.K117N

Reagents Required but not Supplied

1. Molecular Biology Grade Water: Thermo Fisher Catalog # AM9937