



Precipio, Inc.

# Instructions for Use

**NRAS Exon 4 (146) Mutation Analysis using ICE COLD-PCR  
Coupled to TaqMan SNP Genotyping Analysis**

## Table of Contents

Manufacturer	2
Reagent Preparation	2
Intended Use	2
Kit Components and Storage Conditions	2
ICE COLD-PCR Coupled to Downstream Analysis	3
Primary Sample Collection, Handling and Storage	3
Notes for this Assay	4
ICE COLD-PCR Protocol	5
Thermal Cycler Program for ICE COLD-PCR	7
Quality Control of ICE COLD-PCR Products (OPTIONAL)	8
Technical Support/Questions	8
Appendix	8

Copyright © 2018 by Precipio, Inc.

All rights reserved. This Instruction manual or any portion thereof may not be reproduced in any form, or transmitted outside of the recipient's organization in any form by any means — electronic, mechanical, photocopy, recording, or otherwise — or used in any manner whatsoever without the express written permission of the Company.

## Manufacturer

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

## 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.









## 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 146) 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.

## Kit Components and Storage Conditions

The NRAS Exon 4 Kit contains the components to perform ICE COLD-PCR amplification for mutation enrichment 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% A146T Control	Clear 	Red 	1	66	-20

This kit was designed specifically to enrich the following mutations:

1. c.436G>A; p.A146T