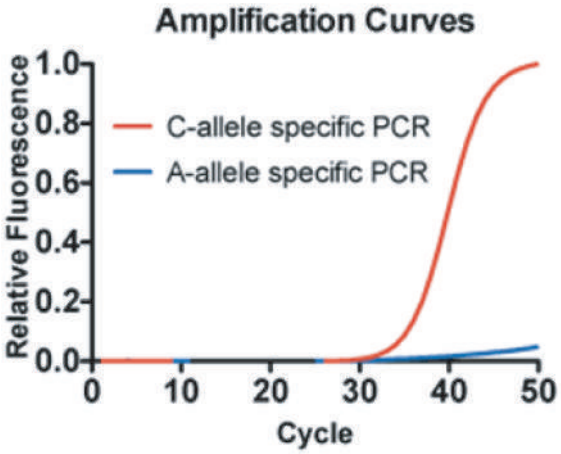
# Application note for HiDi DNA polymerase (#9001) and HiDi Taq DNA polymerase (#9201)

**SNP genotyping example by allele-specific PCR:** SNP: rs72921001, A/C, chromosome 11

target sequence: 5'...TCTTGATCCGAGGCCTACAG**C/A**TTTTGATCCCTTCTCTATCCC...3'
C-allele primer: 5'-GAATGGGATAGAGAAGGGATCAAAA**G**-3'
A-allele primer: 5'-GAATGGGATAGAGAAGGGATCAAAA**T**-3'

reverse primer: 5'-CTGCTGCTTGAAAATGGATTGTG-3'

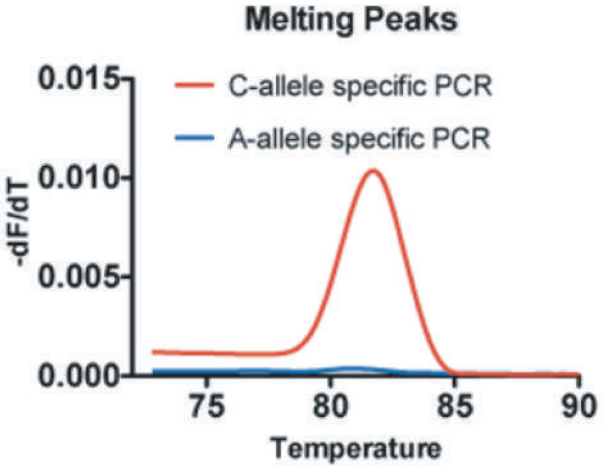
## HiDi DNA polymerase (#9001) with GreenDye (#2000)



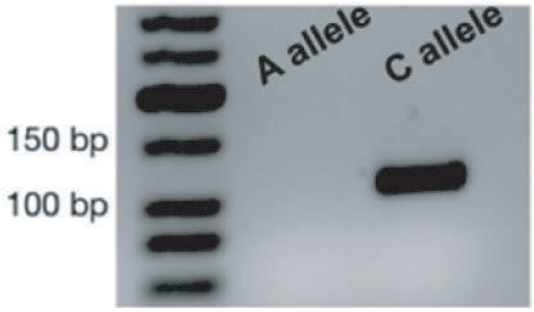

Example data of allele-specific PCR.
Reactions were performed from 1 ng/µl of HeLa gDNA in the presence of a realtime dye (GreenDye, #2000), indicating the amplification of the C-allele specific primer only. The A-allele specific primer is discriminated, thus not amplified up to 50 cycles.

PCR protocol:
95°C - 3 min (initial denaturation)
95°C - 10 sec
57°C - 15 sec
72°C - 30 sec (50 cycles)

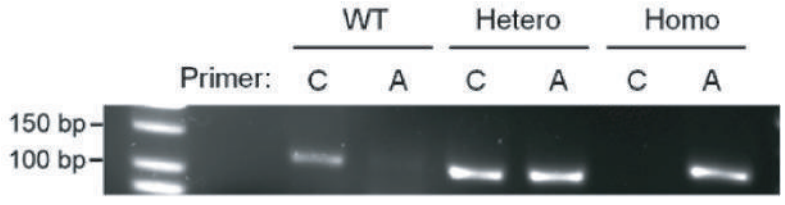
**red curve - PCR with C-allele specific primer**
**blue curve - PCR with A-allele specific primer**




Example data of melting curves.
A melting peak indicates the amplification of the C-allele specific primer. The A-allele specific primer is discriminated and does not result in a melting peak.




Agarose gel of PCRs.
Specific amplicon can be detect from reactions with HeLa gDNA with C-allele specific primers.




Genotyping of SNP (rs72921001) from reference human genome samples.
Samples from C-allele homozygote and A-allele homozygote show only an amplicon from C-allele specific and A-allele specific primer, respectively.
Samples from heterozygote shows PCR products amplified from both primers

HiDi and HiDi Taq DNA polymerase are based on scientific results, which are published in the open access and peer-reviewed Journal PLOS ONE, 9(5), e96640:
Drum M, Kranaster R, Ewald C, Blasczyk R, Marx A (2014)
„Variants of a Thermus aquaticus DNA Polymerase with Increased Selectivity for Applications in Allele- and Methylation-Specific Amplification."