

Check the product label for actual catalog number, lot and expiry date.

## ALLin™ HS Iso-Colorimetric DNA Amplification Kit

CAT.#	SIZE	COMPONENTS	COMPONENT COMPOSITION
IDK0301	100 r of 25 µl	1.25 ml - ALLin™ HS Iso-Colorimetric Mix, 2X 1 ml - PCR Water	The 1X HS Iso-Colorimetric amplification mix includes a Hot Start Bst Polymerase (large fragment), 3 mM MgSO <sub>4</sub> , 1.6 mM dNTPs, enhancers, stabilizers and an inert dye that changes its color during DNA amplification as the pH is changing.
IDK0305	500 r of 25 µl	5 x 1.25 ml - ALLin™ HS Iso-Colorimetric Mix, 2X 5 x 1 ml - PCR Water	

Storage In the dark at -20°C.

### APPLICATIONS

- Point of care & field colorimetric DNA target detection
- Point of care colorimetric LAMP - loop-mediated amplification
- Isothermal DNA amplification with real-time monitoring
- WGA - whole genome amplification
- RAM - ramification amplification

### PRODUCT DETAILS

The ALLin™ HS Iso-Colorimetric DNA Amplification Kit enables sensitive target DNA molecules detection in real time without the use of any equipment. The kit employs the robust Hot Start Bst polymerase in a master mix with all reaction components and the pH-sensitive inert dye which changes the colour from pale orange to bright yellow as soon as the reaction pH changes with the synthesis of DNA reaching a plateau. The mix detects as little as 3 target DNA molecules in a short time of 20 minutes without the use of any equipment. The 2X master mix includes the high-performance buffer, dNTPs and a recombinant Hot Start Bst Polymerase large fragment having strong 5' - 3' strand displacement activity and efficient 5' - 3' polymerase activity working at 55 - 70°C. The activity of the enzyme is blocked at ambient temperature due to molecular inhibition based hot-start technology, and the polymerase is activated only at 45 - 50°C what reduces non-specific amplification, primer dimer formation and is therefore excellent for ambient reaction setup. The polymerase has neither 5' - 3' nor 3' - 5' exonuclease activity and retains only minor reverse transcription activity. The kit includes PCR water; only templates and primers have to be supplied by the user. ALLin™ HS Iso-Colorimetric DNA Amplification Kit is a tool of choice for such applications like LAMP, WGA, RAM with additional advantages of a room temperature set up, field detection as well as of high-temperature reactions, what makes amplification of complex and GC-rich templates more efficient.

### ISOTHERMAL AMPLIFICATION PROTOCOL EXAMPLE

- Take typical measures to prevent contamination, keep your bench clean, wear gloves, use sterile tubes and filter pipet tips.
- Include a no-template control and positive controls in parallel.
- Thaw and keep reagents on ice. Mix well before use.
- Ambient temperature assembly can be performed with the HS Mix, as the enzyme becomes activated only above 45°C.
- Perform the reaction at 65°C. If needed, optimize the reaction temperature between 55 - 70°C for each template/primers system. Complex templates may require higher temperature.
- Suggested reaction time is 20 - 30 minutes. For some low copy number targets 30 - 60 minutes might be required to observe the color change.
- Design primers with predicted melting temperature of about 60°C.
- Prepare 10X primer mix in water or TE Buffer, for example, for LAMP: 16 µM FIP, 16 µM BIP, 2 µM F3, 2 µM B3, 8 µM LoopF, 8 µM LoopB.

IN VITRO RESEARCH USE ONLY

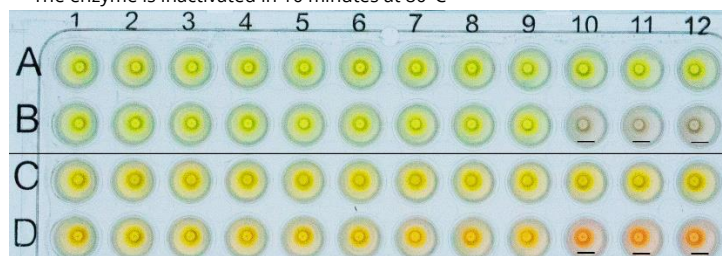
### BENEFITS

- Colorimetric, simple real-life reaction monitoring without the equipment: Orange solution - no DNA; yellow solution - amplified DNA.
- Hot-start function for an ambient reaction setup and minimized background
- Efficient 20 minutes DNA amplification at constant 55-70°C temperature
- Robust on complex templates and crude samples
- Low-copy (<3 molecules) targets detection

### PERFORMANCE

Technical characteristics of Hot Start Bst Polymerase (large fragment):

- The polymerase is blocked at ambient temperature and activated at temperatures above 45°C
- Strong 5' - 3' strand displacement activity
- 5' - 3' polymerase activity
- No 5' - 3' exonuclease activity and No 3' - 5' exonuclease (proofreading) activity
- Minor reverse transcriptase activity
- Optimal amplification temperature is 65°C
- Working temperature range is 55 - 70°C
- Optimal reaction time is 20 min., if needed it can be extended to 30 - 60 minutes
- The enzyme is inactivated in 10 minutes at 80°C



**ALLin™ HS Iso-Colorimetric DNA Amplification Kit (A & B) performs with a higher sensitivity compared to competitor kit (C & D).** 7 serial dilutions of virus ssDNA have been amplified according to protocol, starting with 0.25 ng/25 µl rxn (A1-A3, C1-C3), dilution factor of 10, 3 replicates each. A no-template control was B10-12 & D10-D12. Plates were prepared at room temperature. Reaction carried out for 20 min at 65°C and photographed.

*The use of this product in certain applications may be covered by patents. The user has to analyse all applicable Limited Use Label Licenses and may need licensing for certain cases.*

✓ Prepare a 25 µl reaction:	
ALLin™ HS Iso-Colorimetric Mix, 2X	12.5 µl
10X primer mix (variable, depends on application)	2.5 µl
Template DNA (variable, depends on application)	1 µl
PCR Water (supplied)	to 25 µl
✓ Mix gently, avoid bubbles.	
✓ Place into the thermostat or qPCR instrument to incubate:	
Amplification	65°C - 10 - 30 min temperature can be between 55 - 70°C, time between 20 - 60 min
Optional: Inactivation	80°C - 10 min
Examine and photograph the plates. DNA-containing samples will be bright yellow (see image above A1-12), the no-DNA controls and no-amplification samples will remain darker-orange (above B10-12). Store reactions for short time on ice, for long time at -20°C. High yield amplification product may increase the viscosity of the solution, mix well and dilute if needed for downstream applications.	

### ORDERING

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