

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

## ALLin™ Mega HS HiFi DNA Polymerase

CAT.#	SIZE	COMPONENTS	COMPONENT COMPOSITION
HLE0401	100 u	100 u - ALLin™ Mega HS HiFi DNA Polymerase, 2 u/μl 1,5 ml - 5X ALLin™ Mega HS HiFi Buffer	Enzyme in storage buffer.
HLE0405	500 u	5x 100 u - ALLin™ Mega HS HiFi DNA Polymerase, 2 u/μl 5x 1,5 ml - 5X ALLin™ Mega HS HiFi Buffer	1X ALLin™ Mega HS HiFi Buffer contains 1 mM dNTP, 3 mM MgCl <sub>2</sub> , enhancers, stabilizers.

Storage In the dark at -20°C.

### APPLICATIONS

- Sequencing, including NGS library preparation
- Hot start PCR, multiplexing
- Fast high-fidelity PCR (up to 100 x *Taq*)
- Long PCR up to 20 kb
- Amplification of complex (GC/AT rich) templates
- Blunt-end cloning and other applications

### PRODUCT DETAILS

Derived from our HiFi polymerase, the highQu ALLin™ Mega HS HiFi DNA Polymerase provides much lower error rate PCR with a 100 higher fidelity compared to *Taq*. Compared to Mega HiFi, this hot start enzyme version allows for even higher sensitivity and specificity of PCR as well as for a room temperature reaction setup, and is excellent choice for multiplex reactions. The ALLin™ Mega HS HiFi DNA Polymerase is engineered to be much faster and to generate higher yield of long PCR products up to 20 kb from complex GC-rich templates. Therefore the ALLin™ Mega HS HiFi DNA Polymerase is an excellent choice for longer and very complex PCR applications where the highest fidelity is demanded. It is an enzyme of choice for cloning and all kind of sequencing applications including NGS. Generated blunt-ended PCR products are suitable for ligation into blunt vectors. To increase ligation efficiency, the use of HighEnd™ Repair Kit (HER0101) is recommended.

### PROTOCOL

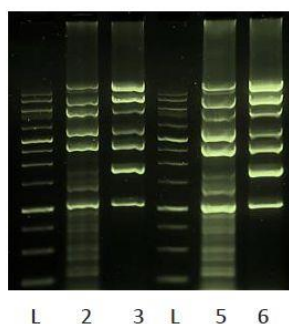
- Take typical measures to prevent PCR cross over contamination, keep your bench clean, wear gloves, use sterile tubes and filter pipet tips.
- Include a no-template control and positive control in parallel.
- Thaw and keep reagents on ice. Mix very well before use. Mixing of the buffer is very important for the final yield!
- For complex, GC rich templates, use 99-100°C denaturation temperature, it might help to increase the yield.
- For established PCRs, try two-step cycling protocol with a combined annealing-denaturation step of 70°C (68°C to 75°C).
- Run an annealing temperature gradient (2°C increments) from 60°C to 66°C to choose the best conditions.
- The longer the amplicon, the longer the extension time: depending on the complexity of the template, perform extension from 10 to 30 sec/kb. Longer extension for more complex templates is needed. For multiplexing, start with extension time needed for the longest fragment.

### BENEFITS

- Hot start enzyme for increased sensitivity and great multiplex results
- Fast, high yield PCR with the fidelity 100x higher than *Taq*
- Up to 20 kb long PCR even from complex templates
- Increased processivity for faster amplification and higher yield
- High thermostability for better denaturation of GC rich templates
- Best choice for NGS library prep. and other sequencing applications
- 5X ALLin™ Buffer includes optimal Mg<sup>2+</sup> and dNTP amount

### PERFORMANCE

For maximum convenience, use 2X ALLin™ Mega HS HiFi Red Mastermix (HLM0501) and ALLin™ Mega HS HiFi Mastermix (HLM0401).



High sensitivity multiplex PCR results achieved with **Allin™ Mega HS HiFi DNA Polymerase**

*Gel analysis of multiplex PCR reactions - compared to competitor enzyme (2; 5), the Allin™ Mega HS HiFi DNA Polymerase (3; 6) gives more specific multiplex result.*

- ✓ Prepare a 50 μl reaction:

Rev. & For. Primers	To 0.2 - 0.6 μM each (~2μl of 10 μM)
cDNA Template <i>or</i>	<100 ng <i>or</i>
gDNA Template	10 - 200 ng
5X ALLin™ Mega HS HiFi Buffer	10 μl
Water (PCR Water WAT0110)	to 49,5 μl
ALLin™ Mega HS HiFi DNA Polymerase, 2 u/μl	0.5 μl

- ✓ Mix gently, avoid bubbles.

- ✓ Place into the instrument set like:

Initial denaturation	1 cycle: 95°C - 1 min
Denaturation	25-35 cycles: 95°C - 15 sec
Annealing	25-35 cycles: 60-66°C - 15 sec
Extension	25-35 cycles: 72°C - 30 sec (30 sec/kb)

- ✓ Store probes for short time on ice, for long at -20°C.

IN VITRO RESEARCH USE ONLY

### ORDERING

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