

Product Testing Report



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Cas9 Nickase D10A Protein Functional Test

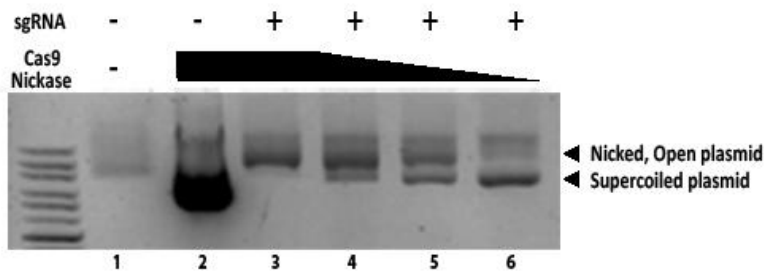
Method

To test the ability of Cas9 Nickase D10A Protein (Cat. No. K032/K132) to generate single strand breaks *in vitro*, Cas9 Nickase D10A Protein was first pre-incubated with sgRNA (200 nM) at 37°C for 30 minutes, and then added to the super-coiled plasmid (250 ng) and continued to incubate for another hour at 37°C. Varying concentration of Nickase (100 nM, 50 nM, 25 nM, 10 nM) was used to examine the dose dependent nicks in the super-coiled plasmid.

Results

Cas9 Nickase D10A Protein nicked the plasmid leading to a more open, circular plasmid, which migrated through the gel slower compared to the super-coiled DNA, in a Cas9-dose dependent manner.

Gel image showing the ability of abm's Cas9 Nickase D10A Protein (Cat. No. K032/K132) to nick supercoiled plasmid to become an open circle.



Legend

- Lane 1: Freeze thawed (nicked) supercoiled plasmid
- Lane 2: 100 nM Cas9 Nickase D10A Protein (no sgRNA)
- Lane 3: 100 nM Cas9 Nickase D10A Protein + sgRNA
- Lane 4: 50 nM Cas9 Nickase D10A Protein + sgRNA
- Lane 5: 25 nM Cas9 Nickase D10A Protein + sgRNA
- Lane 6: 10 nM Cas9 Nickase D10A Protein + sgRNA

Conclusion

abm's Cas9 Nickase D10A Protein (Cat. No. K032/K132) makes single strand breaks efficiently at a concentration of 100 nM and leaves almost no super-coiled plasmid as compared to the lower Cas9 nickase concentrations.