

2025 NEFU_China

Restriction endonuclease reactions

ABSTRACT

This protocol is used to having restriction endonuclease reactions based on endonuclease by NEB.

1. Add the following reagents to a PCR tube (e.g. 50µl) on ice.

	A	B
1	Restriction Enzyme	10 units is sufficient, generally 1 µl is used
2	DNA	1µl
3	10 x NEBuffer	5µl
4	ddH ₂ O	To 50µl

2. Incubate at 37°C in water bath for 2 hour.
3. Stop the reaction by heat inactivation. Temperature and time for inactivation depends on the enzyme.
4. Use electrophoresis to confirm if correct construct was present.