Experiment (7)
Restriction Fragment Length Polymorphism (RFLP)

Materials:

1X TBE buffer

1% agarose gel

Sterile, deionized water, restriction enzyme 10X buffer, Acetylated BSA, DNA sample, EcoRI restriction enzyme.

Protocol:

1. In a sterile tube, assemble the following components in the order listed below.

Component	Volume
Sterile, deionized water	16.3µl
Restriction Enzyme 10X Buffer	2.0µl
Acetylated BSA, 10µg/µl	0.2µl
DNA, 1µg/µl	1.0µl
Mix by pipetting, then add:	
Restriction Enzyme, 10u/µl	0.5µl
Final volume	20µl

2. Mix gently by pipetting, close the tube and centrifuge for a few seconds in a microcentrifuge. Incubate at the enzyme's optimum temperature (37°C) for 1–4 hours.
3. For sample loading, mix 3µl of 6X loading dye with 2µl of DNA sample.
4. Load 5µl of prepared samples into wells and in different well load 5µl of DNA marker
5. Put it onto the apparatus.
6. Apply a continuous voltage of 95V for 50 minutes.

According to: https://worldwide.promega.com/-/media/files/resources/protocols/technical-manuals/101/restriction-enzymes-protocol.pdf?la=en