

**Center for Engineering Plants for Resistance Against Pathogens**  
A NATIONAL SCIENCE FOUNDATION SCIENCE AND TECHNOLOGY CENTER



# **Biotechnology Laboratory**

*Purification of Thymus DNA*

University of California, Davis  
One Shields Ave • Davis, California 95616 • (530) 752-6552



# DNA PURIFICATION

## Sheep Thymus



### Background

The DNA extracted in the laboratory *DNA Extraction from Sheep Thymus* was too impure to analyze further.

The protocol below will help remove the excess proteins from the sample and allow us to run the resulting DNA as part of the *Restriction Enzyme Analysis Laboratory*.

Papain, contained in the meat tenderizer, removes the many proteins bound to and around the DNA. These proteins make the initial extraction very crude and if not removed could inhibit enzyme digest

### Materials

#### *For Each Lab Group*

- One microtube of previously extracted DNA
- 2 disposable pipettes

#### *Common Materials*

- centrifuge
- meat tenderizer solution
- 100% ethanol
- TE buffer

### Advance Preparation

1. Have your students do the *DNA Extraction from Sheep Thymus* Laboratory. Spool a sample of the extracted DNA into a microtube and freeze.
2. Make a supersaturated solution of meat tenderizer (approximately 1 teaspoon per 100 ml). Be sure the meat tenderizer brand you choose contains papain.





## Procedure

1. Remove spooled DNA from the freezer and thaw.
2. Centrifuge your DNA tube for 30 seconds. Pour off the supernatant.
3. Using a disposable pipet, add 1 ml super saturated meat tenderizer solution. Mix by rotating tube end over end. Let sit for 2 min at room temperature.
4. Centrifuge the tube for 30 seconds. Pour off supernatant.
5. Your instructor will add 1 ml 100% ethanol. This gets rid of the papain, the active ingredient in meat tenderizer. Mix again by rotating end over end. Let sit for 2 min.
6. Centrifuge tube for 1 minute and pour off the supernatant.
7. Using a disposable pipet, resuspend the DNA in 0.5 ml TE buffer.
8. Place tube in freezer and let sit overnight.

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🕒 **STOP POINT:** Place tube in freezer until ready to continue with protocol for the restriction enzyme digest of your DNA.

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**Note:** If using Methylene Blue as your staining technique be sure to add twice or even three times as much DNA than indicated in the protocol, with the restriction enzymes and buffer. With that much DNA no water needs to be added as well. Methylene Blue requires a high concentration of DNA in order to be seen.