

Novel Specimen Preparation:

Microwave Fixation

Cryo-preparation



Immunocytochemical Localization

Microwave Fixation

Started in 1970

Advantages:

- Reduces processing times ~90% over bench or mechanical methods
- Reduces extraction of tissue components

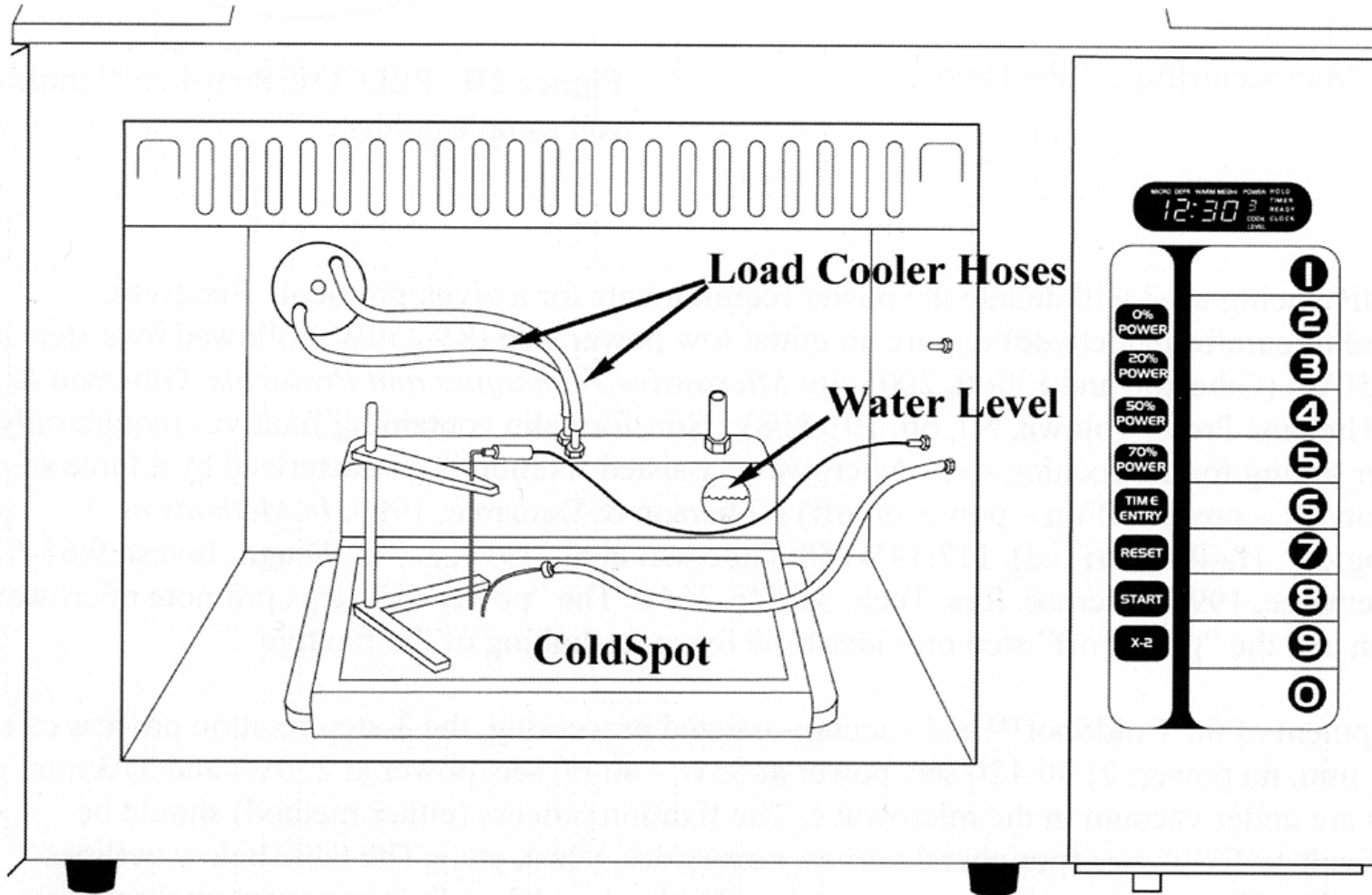
Problems:

- Uneven energy field resulting in hot & cold spots
- Significant energy exposure to tissue

Solutions:

- Coldspot
- Use low wattage reducing:
Tissue microwave exposure
Specimen heating by ~10°C.

PELCO Microwave Oven



Min, Max, and Ave. Temperature rise with 40 sec Microwave Exposure and 4 Wattages (70,221,410,550w)

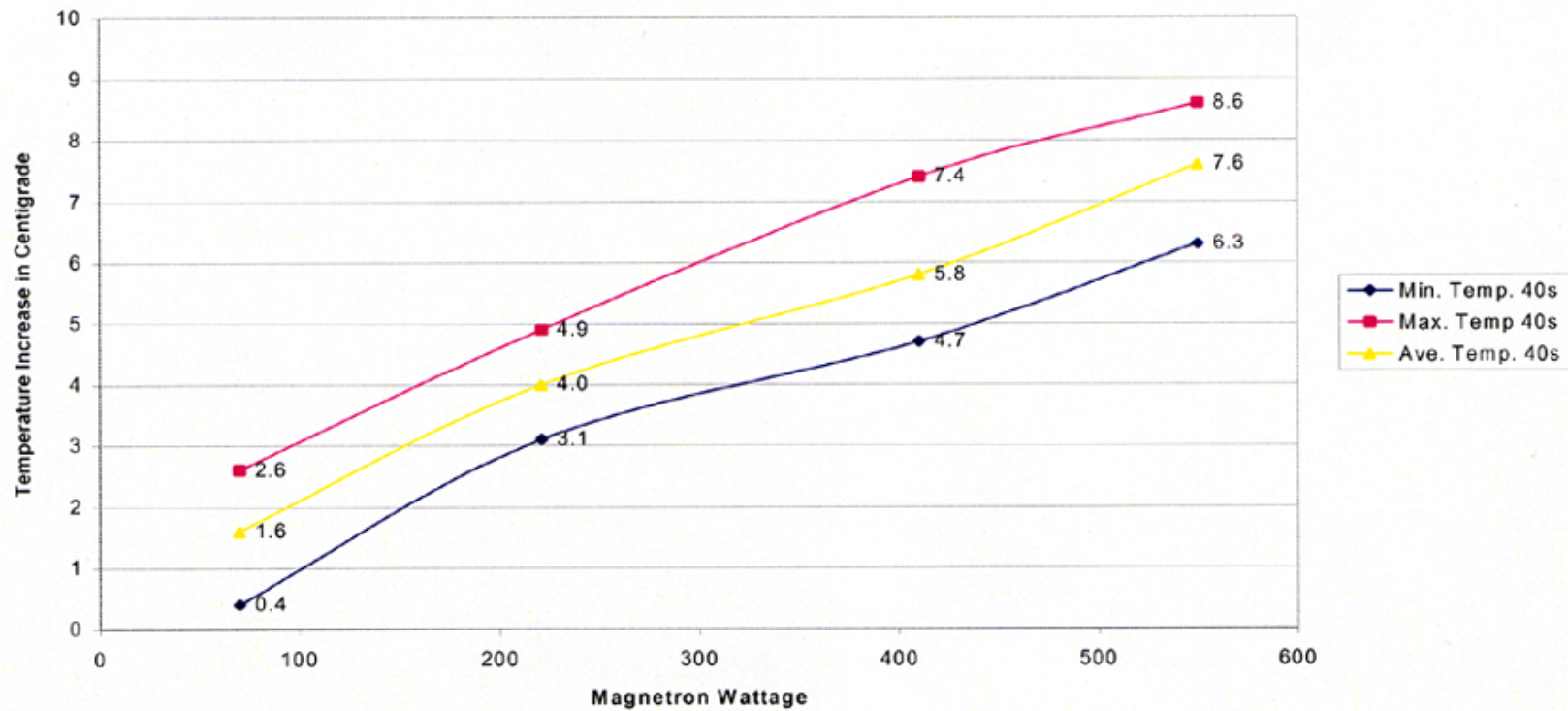


TABLE 1

Min, Max, and Ave. Temperature rise with 80 sec Microwave Exposure and 4 Wattages (70,221,410,550w)

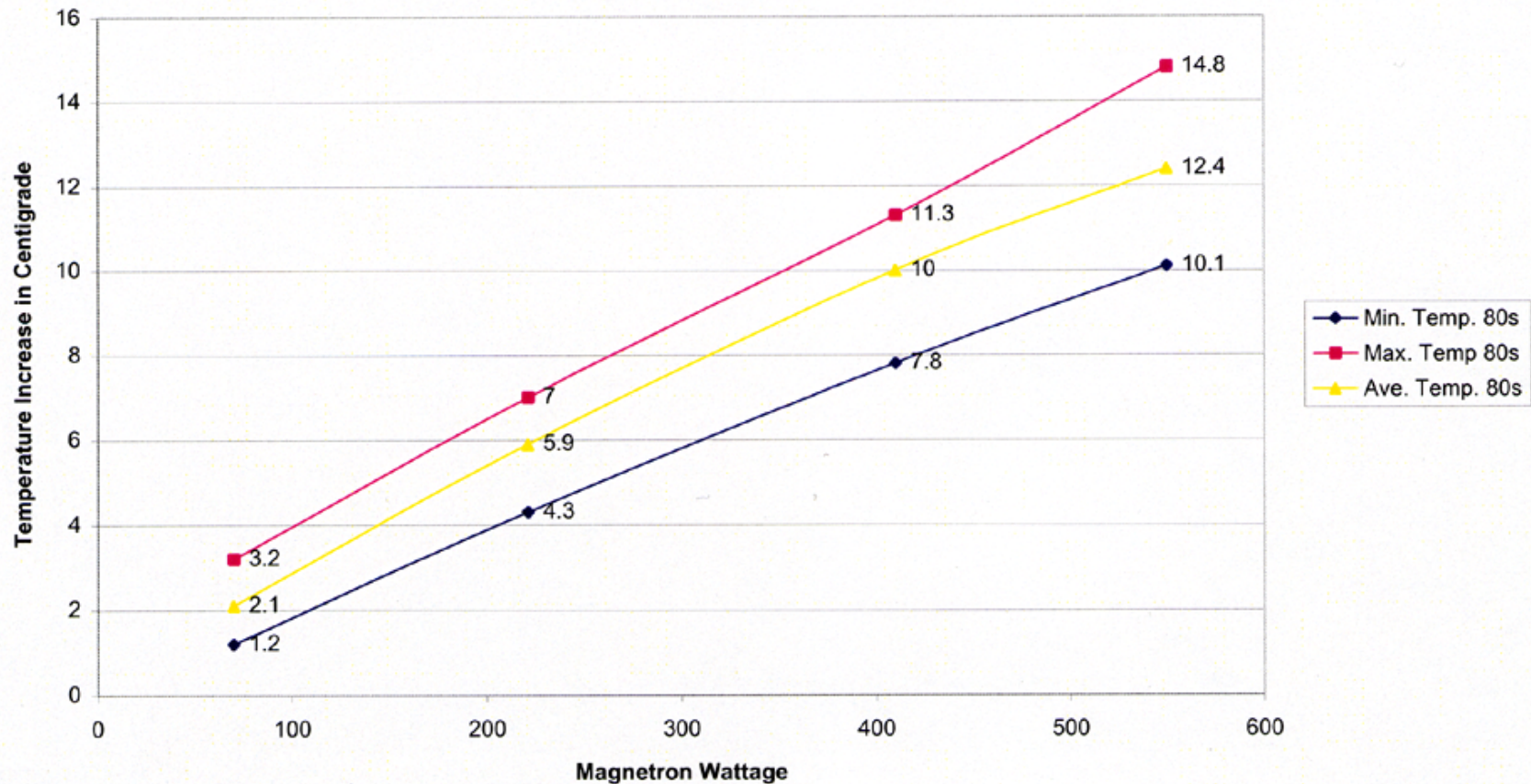
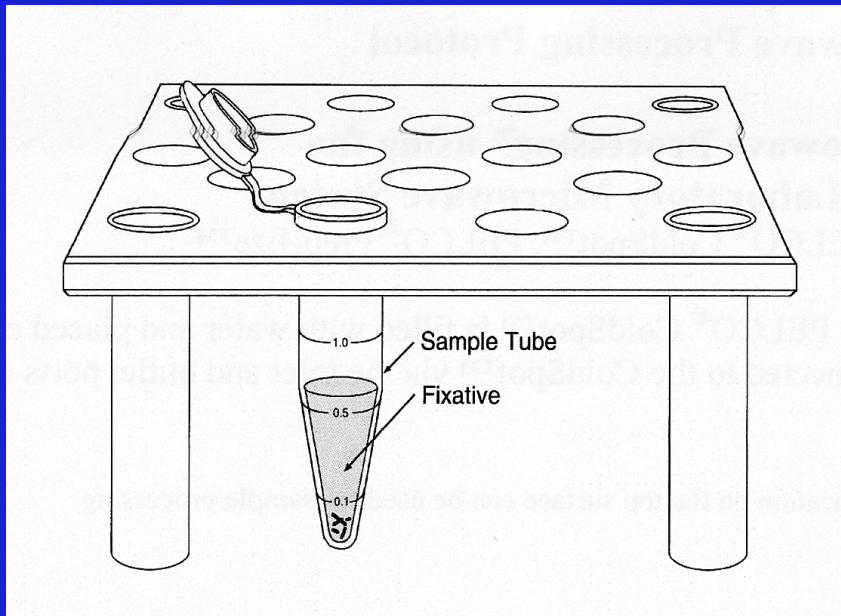


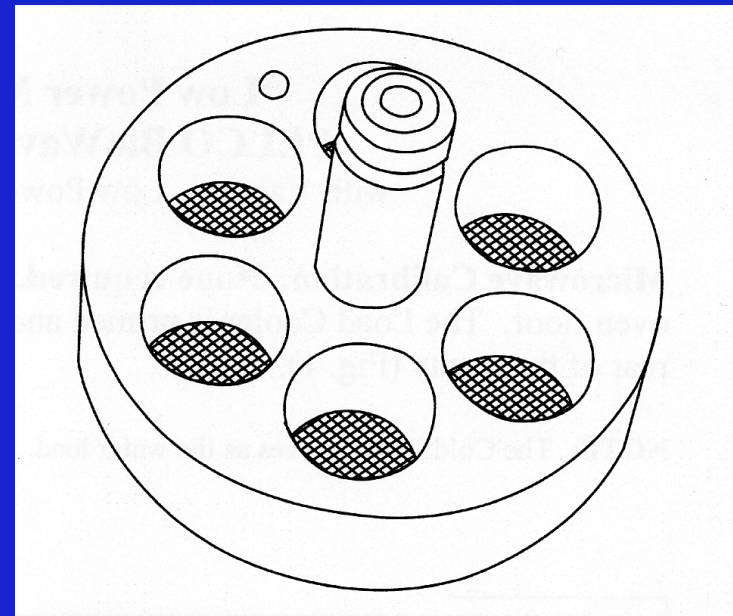
TABLE 2

Microcentrifuge Tube Holder



PELCO Prep-Eze™

Multi-well Holder



Sample Protocol for Plant Tissue

FIX: 1min off – 40 sec on(p1) – 3min off-40sec on (p2)–3min off VAC

Rinse: 2 x 40sec (p1)

OsO₄: 1min off – 40 sec on(p1) – 3min off VAC

Note: can use 2% OsO₄ in Cacod. or use reduced OsO₄

Dehydrate: 40sec (p1)

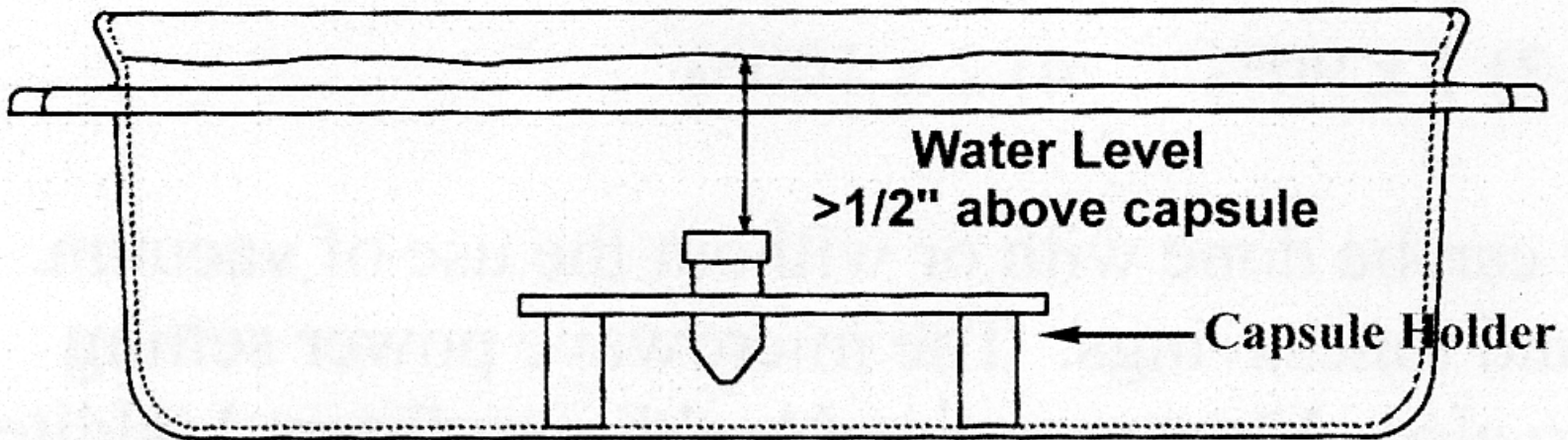
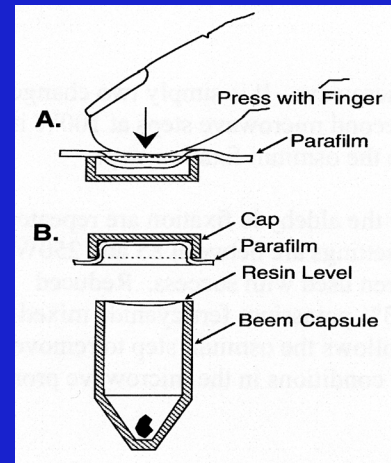
- a) 1 x 50%
- b) 1 x 70%
- c) 1 x 90%
- d) 2 x 100%

Note: if alcohol is used, change to acetone in last 100%

Resin infiltration: Spurr's with vacuum (p2)

- a) 1:1 resin for 3 min.
- b) 100% resin for 3 min.
- c) 100% resin for 3 min.

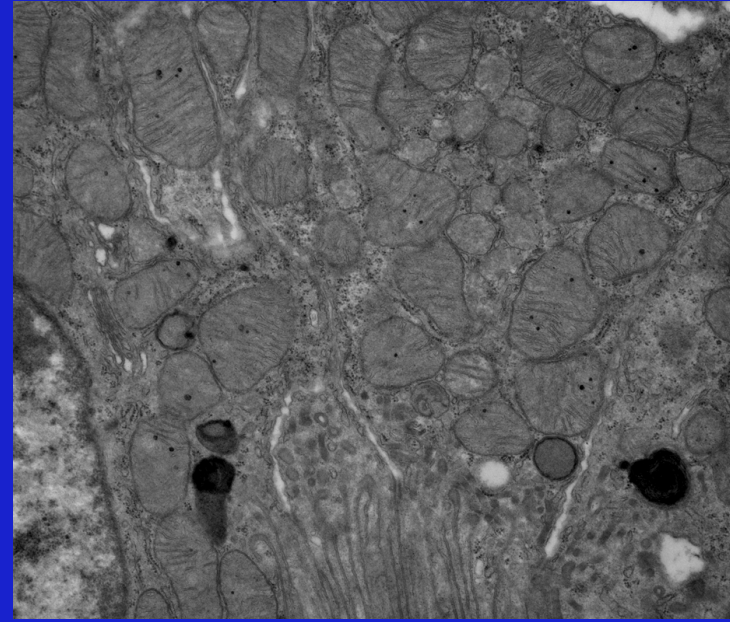
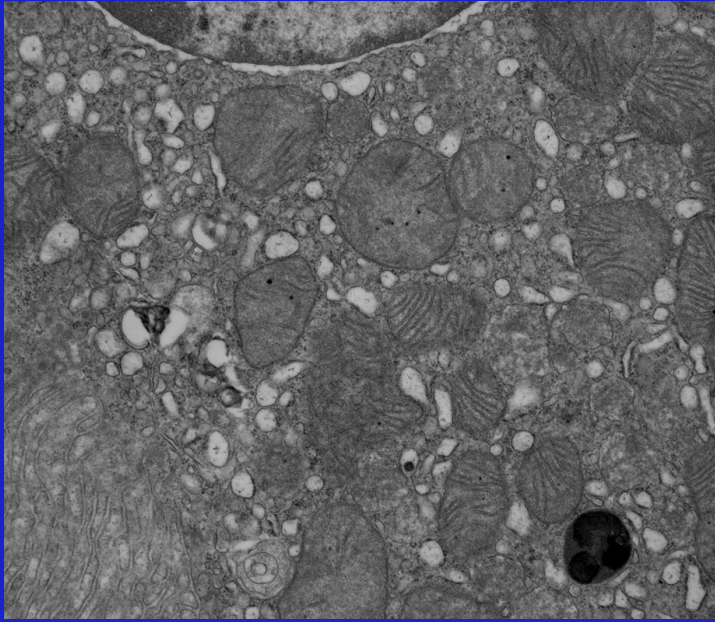
Resin Polymerization



Conventional

Microwave

Kidney



Cardiac
Muscle

