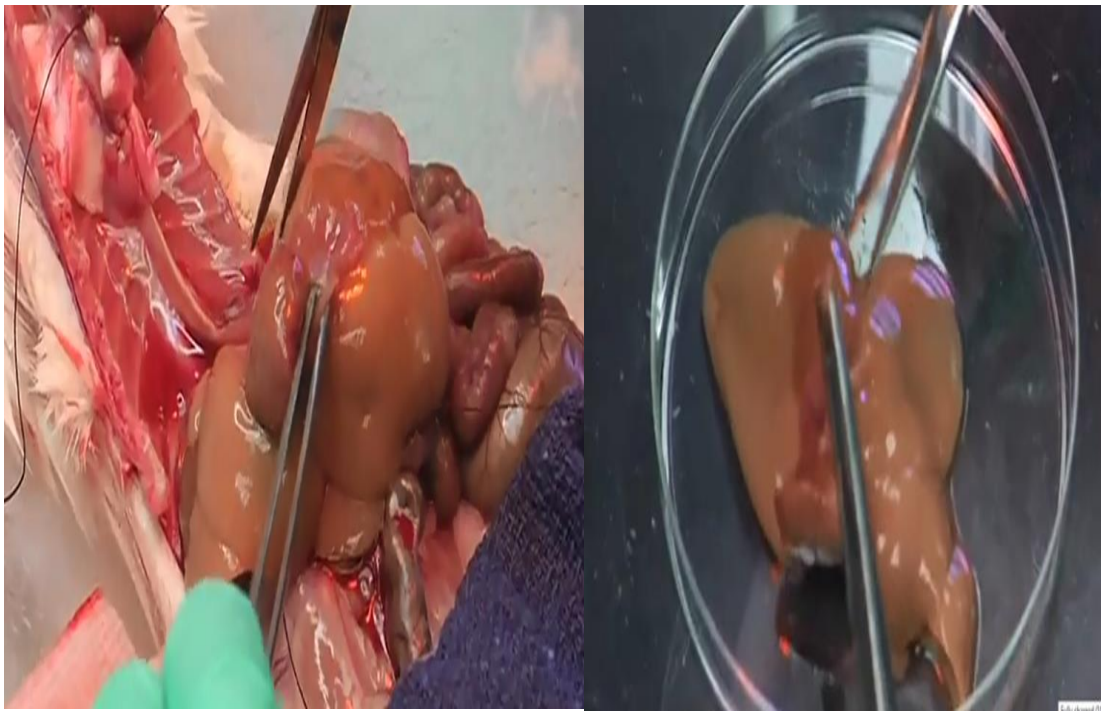
to the portal vein cannula and initiated the infusion in situ at a low flow rate (10 ml/min) with pre-warmed (37 °C) perfusion Buffer I. The liver should be instantly begun to blanch. Once successful cannulation was confirmed, a cut was made at IVC to allow efflux. The chest of the animal was cut. A second cannula connected to a soft tube was placed into the vena cava above the liver in order to enable a recirculating system; the cannula was fixed with a clamp.The perfusion solution was switched to Perfusion buffer II plus collagenase II, increase the rate of flow to 25 ml/min. The liver was become pale in color. A non-recirculating mode was started until the perfusion system was completely filled with collagenase solution then a recirculating perfusion mode was changed with collagenase solution for 15 minutes. After collagenase perfusion, liver was begun to look mushy. The liver was dissected free and placed in a pre-chilled sterile beaker with 20 ml collagenase solution then was taken to tissue cell culture hood (Figure 2.7).



(Figure 2.7): Dissociation of the liver and placing it into sterile Petri dish with collagenase solution.