Tenderness of meat is an important quality attributes for consumers. Calpastatin activity and sarcomere length are some of the methods used to measure meat tenderness. Calpastatin inhibits calpain, which is an enzyme responsible for proteolysis of the muscle protein structure. Therefore, high calpastatin relates to tougher meat. Muscles with longer sarcomere lengths have shown lower resistance to shear force. Lower shear force, more tender the meat. Previous studies might have been detecting fragments of inactive calpastatin. In order to investigate the degradation of calpastatin mechanism an experiment was conducted using three different methodologies to measure calpastatin activity or quantity over a period of 180 total days. Longissimus dorsi samples from between the 12th and 13th rib of the beef carcass (n = 12) were extracted at 0 hour postmortem. These samples were assayed for calpastatin by the traditional method, ELISA and Western Blot at day 0, 90, and 180. Calpastatin activity for the traditional method decreased from a maximum of 3.3 on day 0 to a minimum of 0.1 on day 180. The ELISA and Western Blot analysis did not show results for any of the days. The sarcomere length study we compared sarcomere length of ten Longissimus dorsi samples from each specie: beef, pork and lamb carcasses using three different preparation methods: fresh, frozen conventionally (frozen), and frozen in liquid nitrogen and powdered (powdered). There was no difference between preparation methods (P < 0.05), but between animals. Powdering samples seemed to be beneficial as far as time and material are concerned.