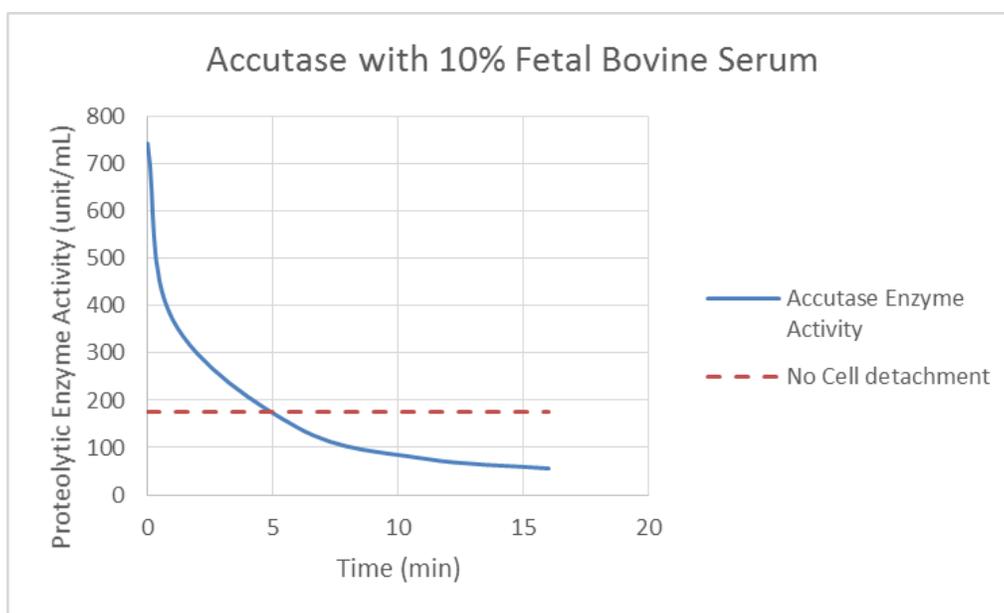




Inactivating Accutase with Fetal Bovine Serum

Normally, Accutase, cell detachment solution does not have to be neutralized following cell passaging due to its gentle nature. In addition, Accutase is inactivated after 45 minutes at 37C. If this method of neutralization does not work for your cells, or if you want to neutralize Accutase in a shorter time period, the addition of serum containing media will accomplish this task. Below is an experiment we conducted to demonstrate the effects of 10% Fetal Bovine Serum on the proteolytic enzyme activity of Accutase.

By diluting a sample of Accutase by 10% with fetal bovine serum, an environment similar to that in cell culture was achieved. The enzyme activity of this diluted sample was measured over time. It was observed that following introduction of the serum to the Accutase, the enzymatic activity decreased. Over time, the activity fell below that of the required level for cell detachment. (Proteolytic enzyme activity must be above 175 units per mL for cells to detach properly.) After about 6 minutes of serum interaction, the enzyme activity of Accutase was well below this level. Serum therefore effectively inhibits Accutase enzyme activity. The following graph contains the data collected in this experiment.



In this graph, the blue line represents the proteolytic enzyme activity of Accutase with 10% fetal bovine serum measured over time. Time zero represents Accutase prior to serum addition. The red dotted line represents the level of activity which will not detach cells.

Conclusion: Accutase will be inactivated 5 minutes following the addition of serum to cells. Please note that this time will vary depending on serum concentration, however, inactivation will still occur.



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