3.8. Determination the capacity of insulin metabolism in liver and evaluation of the inhibitory effect of bacitracin on insulin degradation in isolated hepatocytes

Normal or diabetic isolated hepatocytes were incubated for various periods of time at 37 C° with 100 nM insulin or 200 nM insulin with or without different concentrations of bacitracin (300 µM, 1 mM).

In normal isolated hepatocytes, firstly the addition of 100 nM insulin to intact monolayers (15 min), insulin was significantly degraded by the cells in comparing with insulin blank ($p < 0.05$). Insulin degradation significantly decreased over time ($p < 0.05$). Degradation of insulin in cells was inhibited by bacitracin in two concentrations (300 µM, 1 mM). Almost 80% of inhibition was obtained. (Figure 3.26)

As shown in (Figure 3.27), there was no degradation of 100 nM insulin in diabetic isolated hepatocytes. Insulin level was significantly increased in streptozotocin-intoxicated hepatocytes when compared with insulin blank ($p < 0.05$). In the presence of bacitracin (300 µM, 1 mM), insulin level significantly decreased and returned to the normal level ($p < 0.05$).

Significant degradation of 200 nM insulin during 60 min incubation with normal hepatocytes at 37 C° is shown by the second column of (Figure 3.28) ($p < 0.05$). Bacitracin (300 µM), produced at least 70% decrease in degradation. 1 mM of bacitracin was decreased the degradation and having the greatest effect (90% inhibition) ($p < 0.05$). (Figure 3.29), it indicates that some reactivity of insulin (200 nM) occurs in isolated diabetic hepatocytes and thus bacitracin can exert an inhibitory effect of insulin reactivity.