



Number of rat pancreatic islets necessary for successful transplantation is reduced by Procubol pretreatment

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Aim of the study

Inflammatory reactions at the graft site may inhibit the survival of transplanted islets in the early postoperative period. Especially nitric oxide and reactive oxide intermediates, released from activated macrophages may be cytotoxic to islet cells. Therefore we investigated whether islet pretreatment with the radical scavenger Procubol enhances engraftment of rat islets transplanted into normoglycemic C57BL/6 mice and improves graft function after transplantation in diabetic mice.

Animals, material and methods

Donor islets prepared from adult inbred Lewis rats (weight 350-400 g) were preincubated overnight at 37°C with or without 25 mg/ml Procubol (Sigma). Islets were transplanted beneath the kidney capsule of inbred non-diabetic or STZ-diabetic C57BL/6 mice (24-26g).

Normoglycemic mice were grafted with 120 islet equivalents. The engraftment of Procubol-treated islets was calculated as percentage (mean values \pm SEM) of insulin extracted 3 days after transplantation, compared to insulin extraction from control islets. Diabetic mice were transplanted with islet quantities lower than required to normalize blood glucose levels. So it was possible to recognize, whether Procubol-treated islets, in contrast to control islets, may led to normoglycemia. Graft function in diabetic mice was detected by determination of blood glucose levels.

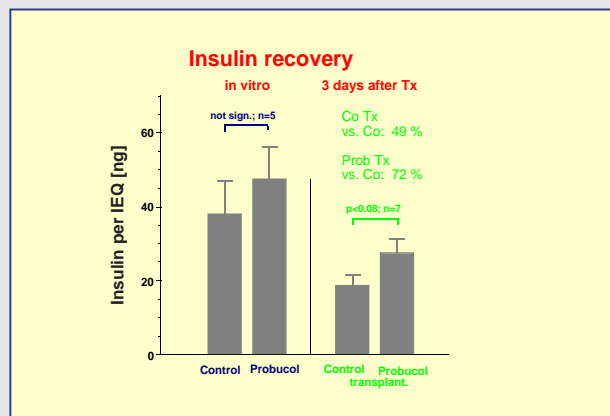


Fig. 1: The graph demonstrates insulin content per islet equivalent. The two left columns show both insulin content after overnight pre-incubation of the untreated control and the Procubol-group at the moment of transplantation. Right columns display insulin recovery 3 days after transplantation, control group in comparison to Procubol-treated islets.

Results

Islet viability, which was tested at the moment of transplantation, was 95-100% (exclusion of trypan-blue). Insulin recovery 3 days after transplantation was 49 \pm 8%, and 72 \pm 9%, in control group and for Procubol-pretreated islets, respectively (n=7; p<0.08; see Fig. 1).

The following results (see Fig. 2) in the diabetic model based on islets which did not have an increased insulin content after preincubation with or without Procubol (Control 49 ng/IEQ vs. Procubol 46 ng/IEQ). Transplantation of 50 islets, incubated in control medium, was insufficient to normalize blood glucose in STZ-diabetic recipients (blue line). However, when the islets were preincubated in Procubol, blood glucose was normalized in all cases (n=5; green line for the average values). Between days 3 and 10, the differences were statistically significant (p<0.05).

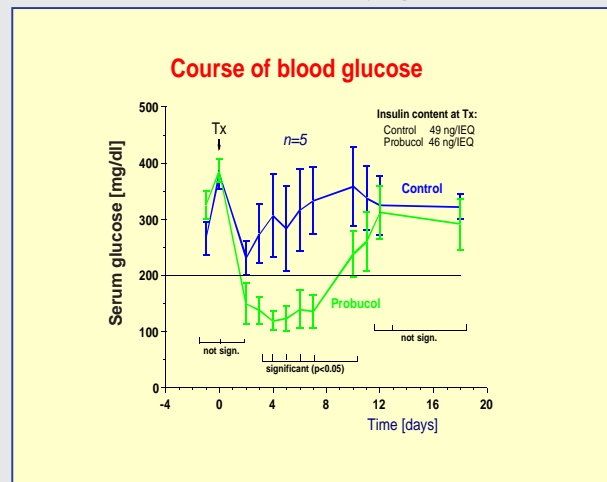


Fig. 2: The curves demonstrate the average course of serum glucose levels (transplantation at day 0). The blue line shows the group of control islets transplanted into diabetic mice, the green line symbolizes the serum glucose course of Procubol islets grafted group. Procubol islets did not show any higher insulin content than control islets before transplantation.

Discussion

Overnight pretreatment of rat islets with Procubol led to a significantly more effective graft function in diabetic mice.

Procubol probably reduces inflammatory islet cell damage by inhibition of islet toxic products of activated macrophages such as nitric oxide and/or reactive oxygen intermediates.

The results show for the first time, that antioxidant pretreatment of islets without treatment of the recipient is able to protect islets of Langerhans at the graft site.

Conclusion

Procubol protects islet grafts from inflammatory damage and enhances islet survival in the early post-transplant period, probably because of lower susceptibility of the Procubol-treated islets to inflammatory reaction.