

IPEN-DOC- 4206

A COMPARATIVE ASSESSMENT OF IMMUNIZATION PROCEDURES FOR DEVELOPMENT OF ANTIPROINSULIN ANTISERA FOR RADIOIMMUNOASSAY (RIA).

Nascimento M.; Borghi V.C.; Bellini M.H.; \*Wajchenberg B.L.; Mesquita C.H.

IPEN-CNEN/SP & \*FMUSP; BRAZIL

Two schedules of immunization were employed for developing antiproinsulin antisera for RIA. Biosynthetic human proinsulin-hPI (Elli Lilly, USA), was injected subcutaneously in guinea pigs in multiple sites. In the 1st schedule was used 50 µg of hPI and the booster injections were administered 4 weeks after the primary immunization and then at 3-week intervals. In the 2nd schedule was used 250 µg of hPI and boosters were done 7, 9 and 18 weeks later. The antibodies titer were testing throughout the immunization period. Three weeks after the 1st booster, total blood was taken from all animals and the antisera were evaluated for cross-reactivity with biosynthetic human insulin and C-peptide from Lilly. As the antisera were not sufficiently specific for hPI they were purified and assessed for kinetic of precipitation and avidity.

Both immunization schedules gave comparable responses. Titers rose to their maximum in about 8 to 9 weeks. The antisera generated by the use of 50 µg of hPI presented higher cross-reactivity with insulin while the reactivity with C-peptide was of the same order in both antiserum groups. The avidity was very variable in the two groups and the three most sensitive antisera required 24h at 40C for achieving maximum binding with the 125I-hPI. However, only one antiserum (from 1st group) was suitable for the RIA. This study emphasizes the difficulties of making valid comparisons between different immunization procedures, especially in the cases when highest avidity is required.

IX Congreso Argentino Biología y  
Medicina Nuclear

IV jornadas del Cono Sur de  
ALASBIMN

14-18 Octubre 1991