DETECTION OF SINGLE CELLS PRODUCING ANTI-GAMMA GLOBULIN ANTIBODIES IN VITRO

BY

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SUMMARY

The technique of localized hemolysis in liquid was successfully adapted for the detection of single cells producing anti-BGG antibodies. The system was characterized and the optimal conditions established for use of the assay to study the cellular response in rabbits immunized with gamma globulin.

To summarize the findings:

1. Coupling of BGG to SRBC with ECDI produces indicator cells which can detect specific anti-BGG PFC. The sensitivity of the BGG-SRBC is determined by the concentration of reactants used in the coupling solution. To couple 0.5 ml of packed SRBC, 600 mg of BGG and 500 mg of ECDI is recommended.
2. BGG elutes from BGG-SRBC upon standing. These indicator cells, therefore, must be washed immediately prior to use.
3. Fetal calf serum in the assay media decreased the recovery of anti-BGG plaques. It was found that 1% human serum albumin could be used in place of FCS to protect the spleen cells from aggregation and the indicator cells from spontaneous lysis without inhibiting the anti-BGG PFC counts.

4. The anti-BGG PFC detected with BGG-SRBC could be inhibited with soluble BGG. The inhibition was dose-dependent; amounts of BGG in excess of 0.1 mg/ml were 100% inhibitory for the anti-BGG PFC.
5. In our hands, the liquid and agar systems were equally able to detect anti-BGG PFC.
6. Rabbits, immunized with BGG were found to have from 25 to 1,000 PFC/10^8 spleen cells, producing specific anti-BGG antibodies.