

## Letter to the Editor

### Advantages of Placenta as a Source for Mass Production of HLA-Antisera

Human Leukocyte Antigens (HLA) of fetus are responsible for induction of HLA antibodies in mother. Exposure to the mismatched paternal HLA antigens in the fetus gives rise to a polyclonal antibody response against paternal HLA antigens. HLA antibodies are present in the maternal circulation of the placenta and are absorbed onto the placenta [1]. The major source of HLA antisera is from sera of multiparous women [2]. Placenta fluid a mixture of serum and tissue fluids has proved to be a valuable source of HLA antisera [3-5]. Selection of source material, i.e., peripheral blood (PB) sera or placenta sera could be justified according to their inherent merits and disadvantages. In this study the two sources are compared to each other in terms of practicability in Iranian parous women.

Placenta and PB samples were collected from 334 Iranian parous women. The placenta processing was carried out mainly as stated [4]. Briefly, placenta fluid was extracted, centrifuged and stored at -20°C. Before dispensing in the microplates, it was centrifuged again. PB sera were stored at -20°C until use. Lymphocytotoxicity test was performed as described [6]. Cells of 101 healthy volunteers were HLA typed by means of commercial antisera. These cells were used for characterization of PB sera and placenta sera. The specificity and sensitivity of antisera were determined through defining r-value [1]. The positive sera having acceptable antisera were tested for presence of Hepatitis B surface antigen and of Human immunodeficiency virus antibody. None of these sera were positive for either of the tests.

The placenta processing method used in this report has several advantages: needs few equipment, minimum handling of placenta for reducing the risk of contamination. Mean serum volume obtained from each placenta was 121 ml. One of the major advantages of placenta over PB is its containment of large volume of sera without bothering the parous women. Forty-six parous women were selected based on presence of lymphocytotoxic antibodies with acceptable reactivity in their PB sera. The reactivities of PB and placenta sera were compared in these parous women. Lymphocytotoxic antibodies were shown in 22 placenta sera (6.59% of total women studied). The specificity and sensitivity (r-value) of the placenta sera in 15 cases were equal to and in 7 cases were lower than those of PB sera. Twenty-four placenta sera did not show lymphocytotoxic antibodies, while their PB sera showed such antibodies. These different reactivities may show that starting screening process directly from placenta is more advisable. Nine PB sera and 3 placenta sera (2.69% and 0.89% of total women studied, respectively) demonstrated the quality of reagent antisera against one HLA specificity with r-value  $\geq 0.875$ . These antisera

reacted with HLA-A2, -B5, -B7, -B22 and -B27. The quality of these antisera are quite comparable with commercial ones, and their acceptable reagent quality were also approved by "Reference Laboratory" of the Ministry of Health, Treatment and Medical Education of Iran. Therefore, placenta as a source for mass production of HLA antisera seems to be more economic and practical than PB.

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#### REFERENCES

1. Zachary, A.A., Murphy, N.B., Smerglia, A.A. and Braun, W.E. (1986) Screening sera for HLA antibodies. In: *Manual of clinical laboratory Immunology* (Rose, N. R., Friedman, H. and Fahey, J.L. eds.), third edition, Washington D.C. pp. 824-834.
2. Johnson, A.H., Katovich Hurley, C. and Hartzman, R.J. (1996) Human leukocyte antigen (HLA): the major histocompatibility complex of human and transplantation Immunology. In: *Clinical diagnosis and management by laboratory methods* (Henry, J.B. ed.), W.B. Saunders, Pennsylvania. pp. 969.
3. Colomb, B.W. (1997) Histocompatibility testing. In: *Medical Immunology* (Stites, D.P., Terr, A.I., Parslow T.G. eds.), Connecticut: Appleton and Lange, pp. 291.
4. Graham, M.L., Simonin, T.B. and Davey, R.J. (1989) Harvesting HLA antibodies from placentas. *Laboratory Medicine, March* 169-171.
5. Middleton, D., Bodmer, J., Heyes, J. et al. (1993) HLA typing reagents: alloantisera and monoclonal antibodies. In: *Histocompatibility testing A Practical Approach* (Dyer, P. and Middleton, D. eds.), Newyork: Oxford University Press Inc. pp. 22.
6. National Institute of Health (1979) NIH lymphocyte microcytotoxicity technique. NIAID manual of tissue typing techniques. Publication No. NIH 80-545. Dept. of Health, Education and Welfare, Atlanta.

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