

**SELECTIVE MODULATION WITH LIPOSOMES CONTAINING Der.P1
LABELLED WITH MONOCLONAL ANTIBODIES**

G.P. ROMBI * , G. PIU °

* ISTITUTO DI RADIOLOGIA - UNIVERSITA' DI CAGLIARI
DIRETTORE : PROF. V. RACUGNO
° CENTRO DI ALLERGOLOGIA ED IMMUNOLOGIA CLINICA
DIRETTORE : DR. G. PIU

The use of biologically compatible vehicles, such as liposomes, can be very selective when the monoclonal specific antibodies are chemically coupled with some lipidic components of liposomic artificial membranes. This possible approach should permit a good modulation of the immune system by selective targeting of cells involved in the allergic reactions. The aim of this study is the selective stimulation of PBL, T and B cells in culture by exposition to liposomes containing Der.P1. Liposome preparation was carried out by removing detergent using a dialytical membrane to obtain LUV carriers (size 0.06 -0.08 μ m) .

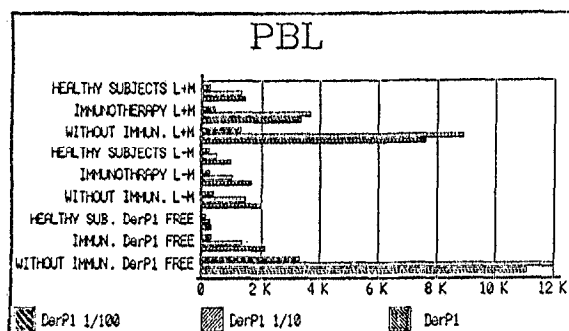
- Liposome composition was DPC 10 μ MOL , cholesterol 5 μ MOL, DPE modified by SPDP 0.6 μ MOL, MAB HT27 and HB31 (Fab')₂ modified by SPDP at final concentrations of 200 μ gr/ml. The concentration of Der.P1 was 0.5-5-50 μ gr/ml. Cell culture was performed by isolation of PBL or by removing T or B cells in a coated plate (microbiological grade).

The cells were cultured in RPMI 1640 medium and exposed for 4 Hrs to the MAB labelled liposomes, liposomes containing Der.P1 without MAB and Der.P1 free in the culture medium. Cells stimulation was measured by adding 3HTdR (activity 2 mCi per mMOL) or by static cytometry after 24 Hrs. Specific IgG and IgE production in the culture medium was also measured. - 5 healthy donors, 5 allergic subjects submitted to immunotherapy and 5 allergic subjects never submitted to immunotherapy have been studied. The biological effects of Der.P1 contained in liposomes are very different than using Der.P1 free in the culture medium. No labelled liposomes are much less active than MAB-labelled liposomes (this fact suggests that the liposome contents intake may be modulated by MAB-receptor interaction). B cells alone are not very responsive. The specific IgG secretion is higher than IgE secretion when the cells are stimulated by liposomes. On the contrary when Der.P1 free is used the IgE secretion is higher.

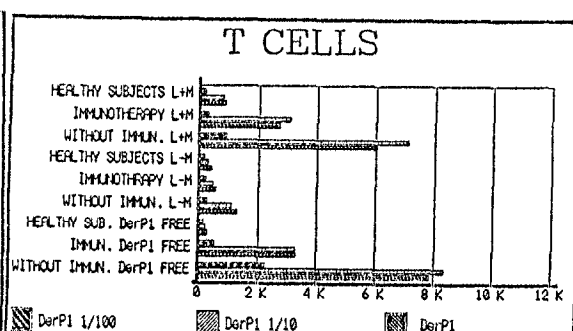
The results obtained show the opportunity of modulating the cellular response in order to lead antibodies production versus specific IgG secretion abolishing the IgE one. Our data show, besides, a strong efficacy of antibodies monoclonal labelled liposomes in making selective the response, as for the use of not MAB-labelled vehicles or free antigen, by activating the chosen cells.

In conclusion this experimental approach can be used to enhance or modulate the specific response to the allergenic stimuli.

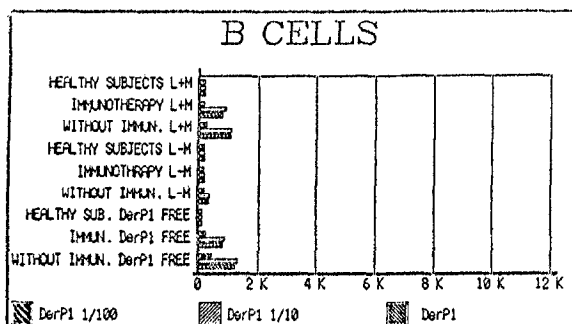
The use in "vivo" of MAB-labelled liposomes as specific artificial carriers is now possible.



GRAPH. 1



GRAPH. 2



GRAPH. 3

GRAPH. 1 - PBL: response to the antigenic stimuli according to the liposome type or free antigen.

GRAPH. 2 - T CELLS: response to the allergenic stimuli according to the liposome type or free antigen.

GRAPH. 3 - B CELLS: response to the allergenic stimuli according to the liposome type or free antigen.

BIBLIOGRAFIA

1. Gregoriadis A., Neerunjun E.D. : Homing of liposomes to target cells. *Biochem. Biophys. Res. Commun.* 65,537-544,1975.
2. Leserman L.D., Weinstein J.N., Blumenthal R., Terry W.D. : Receptor-mediated endocytosis of antibody-opsonized liposomes by tumor cells. *Proc. Natl. Accad. Sci. USA* 77,4089-4093,1980.
3. Leserman L.D., Barbet J., Kourilsky F.: Targeting to fluorescent liposomes covalently coupled with monoclonal antibody or protein A. *Nature*, 288,602-604,1980.
4. Leserman L.D., Macky P., Barbet J.: Cell-specific drug transfer from liposomes bearing monoclonal antibodies. *Nature*, 293,226-228,1981.
5. Loughrey H.C., Choi L.S., Cullis P.R., Bally M.B. : Optimized procedures for the coupling of proteins to liposomes. *Journal of immunological methods* 132,25-35,1990.
6. Rombi G.P., Cossu F., Carboni G. : Selectivity and effectiveness of Nitroimidazolic Radiosensitizers encapsulated in liposomes marked with monoclonal antibodies. 6° Annual Meeting ESTRO (European Society for Therapeutic Radiology and Oncology), Lisbona 25-28 maggio 1987. (p.297 abstr.).
7. Rombi G., Cossu F., Onorato D., Tuveri V.: Liposomi marcati con anticorpi monoclonali e/o incorporanti molecole ferromagnetiche: possibilit  di un loro uso come veicoli artificiali di radioisotopi e farmaci. *Suppl. Tumori*,73 (suppl.4),197,1987.
8. Widder K.J., Senyei A., Sears B.: Experimental methods in cancer therapeutics. *J.Pharm.Sci.* 71,379-387,1982.