Acta Paediatrica Hungarica 29 (1–2), pp. 55–57 (1988–89)

Antigen presentation by monocytes of CGD-patients and by phenylbutazone treated monocytes of normal peripheral blood

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Activation of T helper cells by protein antigens requires presentation of so-called processed antigen by accessory cells in conjunction with class II MHC molecules. In order to study the role of oxygen metabolism in antigen-processing and/or presentation we used monocytes of patients with chronic granulomatous disease (CGD) as a model to study possible relationships. Furthermore, peripheral blood monocytes of normal donors were treated with phenylbutazone to mimic the results obtained with CGD monocytes. The results suggest that the generation of oxygen radicals contributes to the processing of protein antigens and likewise to effective antigen presentation.

Materials and Methods

Adherent cells (monocytes) from patients with CGD and from normal donors were isolated from ficoll isolated mononuclear cell suspensions by adherence to plastic. Adherent cells of normal donors were treated with phenylbutazone (100–1000 μg/ml; 30 min. at 37 °C) to mimic the CGD condition [5]. Antigen presentation by adherent cells was studied in two systems: adherent cells were either cultured with peripheral blood T and B cells and the T cell dependent antigen ovalbumin (OA) or adherent cells were cultured with anamnestic antigens like tetanus toxoid or Candida albicans to induce T cell proliferation. In the former assay activation with OA induces resting B cells to differentiate into OA specific IgM plaque forming cells (PFCs see ref. 1).

Results and Discussion

We previously showed that in normal T and B cells activated with OA which is presented to the T cells by adherent cells optimal PFC responses are induced at doses of 1–3 μg OA/ml. At higher concentrations of OA, T suppressor cell activity will prevail over T helper activity [1]. As indicated in table I adherent cells from CGD patients required extremely high OA (i.e. 100 μg/ml) concentrations to activate the T helper cells operative in inducing OA-specific PFCs. In earlier experiments we showed that adherent cells of CGD pa-

TABLE I

CGD adherent cells determine the altered PFC/OA-dose relationship

Co-culture	PFC/10^6 cells		
	3 μg OA	30 μg OA	100 μg OA
PBL contr + 5% AC contr	1.100	320	80
PBL pat + 5% AC pat	42	457	935
PBL mother + 5% AC pat	83	540	1.079

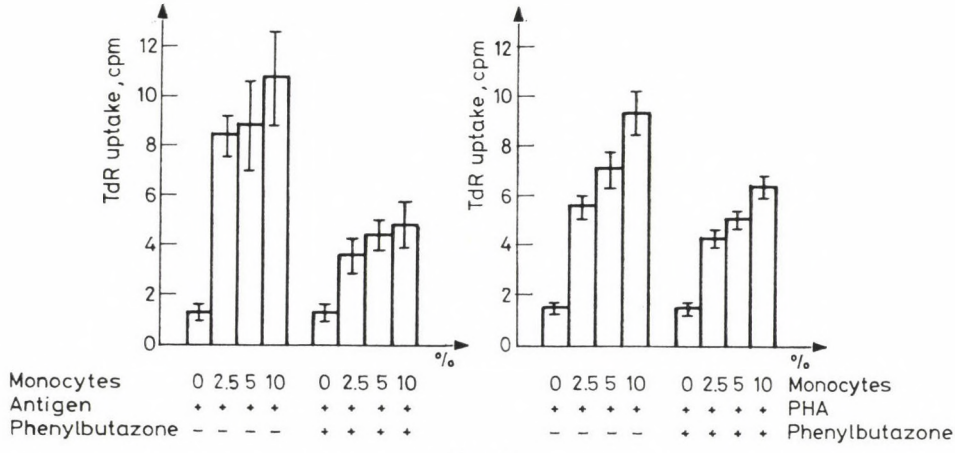

FIG. 1

tients, pulsed with OA for 2 hours and washed and stripped for residual membrane bound OA by pronase treatment, were defective in re-expressing processed antigen [3]. Treatment of adherent cells from normal donors with phenylbutazone which affects the generation of oxygen radicals [4, 5], mimicked completely the results obtained with CGD adherent cells [3].

The effect of phenylbutazone treatment on adherent cells of normal donors used in a T cell proliferation assay is indicated in Fig. 1. The results indicate a significant decrease of antigen-induced T cell proliferation in the presence of phenylbutazone treated adherent cells. The effect is not significant in mitogen-induced T cell proliferation. Recently it has been suggested that oxygen radicals modulate the expression of MHC class II molecules: unlike normal adherent cells, CGD cells do not down regulate class II MHC expression when cultured in vitro for 24 to 48 hours [2]. We compared class II MHC expression on non-treated and phenylbutazone treated cells and found no significant differences using several monoclonal antibodies with different specificities (data not shown).

On basis of our results we conclude that CGD adherent cells are at least partially defective in re-expressing processed protein antigens. This result

strongly suggests that oxygen radicals contribute to the processing of protein antigens. Impairment of oxygen radical generation may likewise affect or modulate antigen presentation. This assumption does not imply that specific humoral or cellular immunity is impaired in CGD patients, since oxygen radical independent antigen processing pathways may exist as well.

References

1. Ballieux RE, Heijnen CJ, Uytdehaag F, Zegers BJM. Regulation of B cell activity in man: role of T cells. Immunol Rev 45, 3, 1979
2. Gruner S, Volk H-D, Falck P, von Baehr R. The influence of phagocytic stimuli on the expression of HLA-DR antigens: role of reactive oxygen intermediates. Eur J Immunol 16, 212, 1986
3. Heijnen CJ, van der Meer JWM, Zegers BJM. Altered antigen presentation in the induction of the in vitro antigen-specific T helper cell function in patients with chronic granulomatous disease. Clin Exp Immunol 66, 111, 1986
4. Oyanagui Y. Inhibition of superoxide anion production in macrophages by anti-inflammatory drugs. Biochem Pharmac 25, 1473, 1976
5. Strauss RR, Paul BB, Sbarra AJ. Effect of phenylbutazone on phagocytosis and intracellular killing by guinea pig polymorphonuclear leucocytes. J Bacteriol 96, 1982, 1968