

## **IS THE PHAGOCYtic FUNCTION OF CANCER PATIENTS IMPAIRED?**

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### ABSTRACT

Very little is known about impaired functions of the mononuclear phagocytes in cancer patients and hence their diagnostic and/or prognostic significance. Phagocytes play a key role in host defenses associated with increased susceptibility to infections. The monocyte and granulocyte phagocytic function of 24 cancer patients and 26 normal controls was determined using Flow Cytometry. The test allowed the quantitative determination of leucocyte phagocytosis (ingestion of bacteria) using fluorescein-labeled opsonised bacteria. The study demonstrated that cancer patients have a significant decrease in the absolute number of monocytes and phagocytic activity. Could this be used as an indicator of the extent or activity of the disease? Further investigation needs to be carried out.

### INTRODUCTION

The phagocytic system includes circulating polymorphonuclear granulocytes and mononuclear leucocytes as well as fixed macrophages.

Decreased capacity of host defence has been associated with specific cellular abnormalities with syndromes of recurrent or chronic infections.

The phagocytic process can be separated into:

1. Chemotaxis (migration of phagocytes to inflammatory sites)
2. Attachment of particles to the cell surface of phagocytes
3. Ingestion (phagocytosis)
4. Intracellular killing (oxygen-dependent or oxygen-independent)

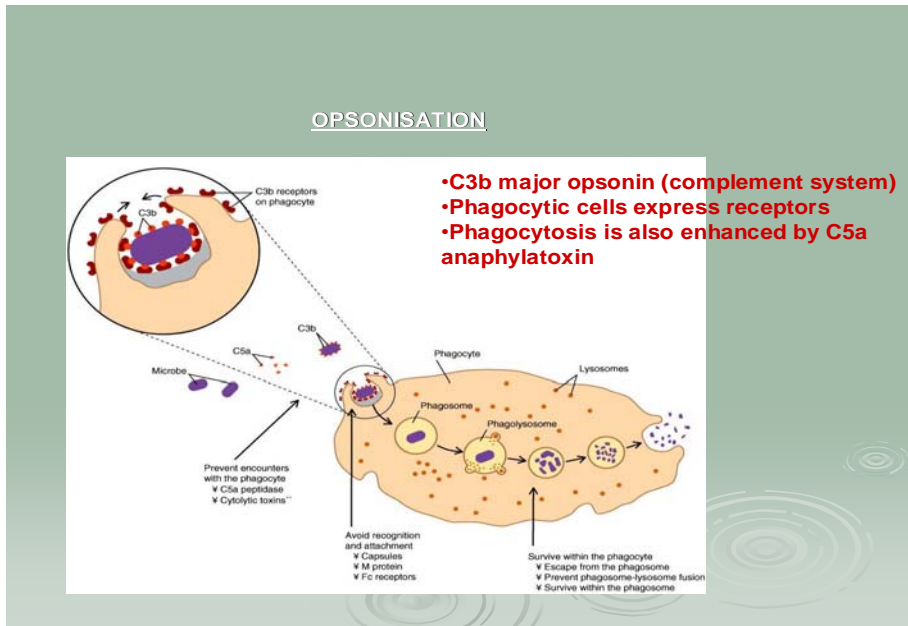
The purpose of this pilot investigation was to study:

- (i) the phagocytic activity of peripheral blood monocytes from patients with Cancer and
- (ii) to investigate any association between phagocytosis and intracellular killing by monocytes in different types of cancer.

### OPSONISATION

Antibody and complement, function as opsonins that bind antigen macrophages and enhance phagocytosis. C3b is the major opsonin of the complement system. (C4b, C3bi, also have opsonisation activity).

C3 activation amplifies coating of antigens by C3b. Phagocytic cells express complement receptors CR1 CR3 CR4 that bind C3b C4 C3bi. When antigen is coated with C3b during complement activation, it binds to cells with receptors therefore phagocytosis starts.



## METHODS

The study was done at the Immunology and Immunogenetics Laboratory Archbishop Makarios III Hospital Nicosia. Heparinised whole blood samples were taken from 24 female Cancer patients, 26 normal controls, (11 female and 9 male) and 49 Thalassaemia major patients (26 female and 23 male), all randomly selected.

Phagocytosis was performed by Flow Cytometry. Quantitative determination of leukocyte phagocytosis (ingestion of bacteria) was performed using fluorescein-labeled opsonised bacteria.

The percentage of phagocytes which have ingested bacteria and their activity (number of bacteria per cell) was measured using FITC-labelled opsonised E.coli bacteria at both 37° C and 0°C.

Quenching solution discriminated between attachment and internalization of bacteria. DNA staining excluded aggregation artifacts of bacteria or cells.

## RESULTS

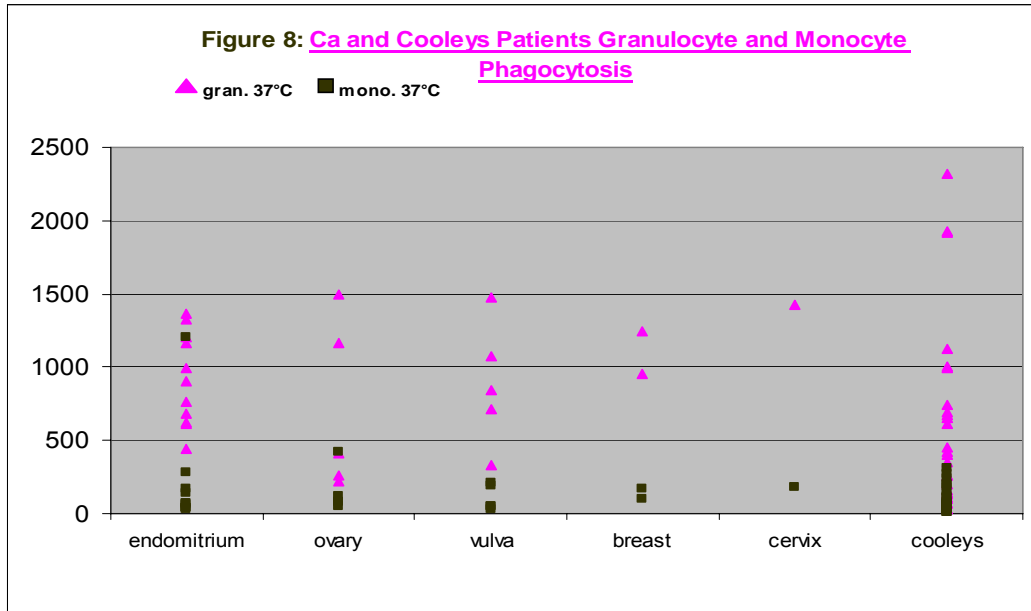
Phagocytosis and the subsequent digestion are multistep and multifactorial. Measurement of the ingestion of bacteria took place under controlled conditions. (0°C and 37°C).

The mean average phagocytic activity of all 24 Cancer patients had mean fluorescence intensity (E.coli/cell) of 880 units of granulocyte activity and 161 units of monocyte activity at 37°C.

The normal control group had a phagocytic activity of mean fluorescence intensity of 1223 units and 321 units respectively.

The Cancer patients were divided into 5 groups according to their diagnosis. Eleven patients had Cancer of the Endometrium, five patients had Cancer of the Ovary, five patients had Cancer of the Vulva, two patients had Breast cancer and one patient had cancer of the Cervix.

The Mean Fluorescence intensity (E.Coli/cell) at 37°C of the eleven patients with cancer of the Endometrium was 916.6 units of granulocyte activity and 197 units of monocyte activity. (Figure 1)



The five patients with Cancer of the Ovary had a Mean Fluorescence intensity (E.Coli/cell) at 37°C of 709.7 units of granulocyte activity and 152.5 units of monocyte activity.

The five patients with Cancer of the Vulva had a Mean Fluorescence intensity (E.Coli/cell) at 37°C of 888.4 units of granulocyte activity and 104.9 units of monocyte activity.

The two patients with Breast cancer had a Mean Fluorescence intensity (E.Coli/cell) at 37°C of 1100 units and the patient with Cervix Cancer had a Mean Fluorescence E.Coli/cell) at 37°C of 1424 units.

## CONCLUSIONS

Our study demonstrated that granulocytes from Cancer patients (endometrium,ovary,vulva,breast,cervix) investigated, showed an increased phagocytic activity for opsonised bacteria.

In comparison, the 35% of Thalassaemia patients had a significantly lower phagocytosis activity. This was shown by the reduction in the flow cytometry fluorescence of the phagocyte-associated bacteria.

Phagocyte fluorescence depended on the relative number of attached and internalised FITC-labelled particles. Low phagocyte fluorescence was in accordance with the increased rate of internalisation.

We have shown that the peak fluorescence of phagocytes from patients with homozygous  $\beta$  Thalassaemia was significantly below that of the normals.

The relative number of attached and internalised FITC-labelled particles in Thalassaemia patients had a mean average for granulocytes of 265 cells and for monocytes of 73 cells. In normal donors the mean average Fluorescence intensity for granulocytes was 1223 cells and for monocytes was 321 cells.

In all Cancer patients studied, except patients with Ovarian cancer, phagocytosis was increased in about 50% independently of the histological type and the clinical stage of the tumour.

The hypothesis that cancer patients have an increased number of surface Fc receptors agrees with the increased activity of phagocytosis in these patients. Leukocyte phagocytosis is critically dependent on the degree of opsonisation of the bacteria as well as on the number of Fc receptors present on the granulocyte cell membrane.

It may be postulated that the increased phagocytic activity observed in all the cancer patients studied with the exception of the patients with the Ovarian cancer, is directly related to the higher density of Fc receptors present on their cell membrane and vice versa.

Either intrinsic differences in the monocyte-macrophage population or serum factors able to modify the monocyte function may be responsible for the increased receptors and phagocytosis.

This may be directly related to the better response of patients with Cancer of the ovary to chemotherapy than radiotherapy.

Further investigation needs to be carried out.