GENETIC FACTORS AFFECTING IMMUNOLOGICAL RESPONSES IN AGING INDIVIDUALS

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It is now widely accepted that the immunological status of elderly individuals is altered as compared to that of younger individuals. Immunological phenomena have wide-ranging implications including the susceptibility and responses of elderly individuals to infections, the responses to immunoprophylaxis, the occurrence of auto-immune phenomena and the increased frequency of malignant conditions. Some believe that old age is the commonest cause of immunodeficiency and others have gone so far as to propose that alterations in immunological mechanisms are the underlying causes of aging. Many have chosen to study age-related immunological changes in their own right without addressing the question of whether they are the causes or the consequences of aging.

It is very difficult to discriminate between age-related changes and changes which are secondary to underlying diseases which are common in old age[1]. Nevertheless it has been convincingly shown, by comparing healthy elderly with chronically ill elderly individuals, that the observed age-related changes in the immune system are real[2].

Although there have been conflicting reports, many studies suggest that a decline in the number of circulating lymphocytes occurs after middle age, usually affecting the T-lymphocytes[3]. It has been reported that an increase in the proportion of cells expressing surface marker for helper (T4) and suppressor (T8) lymphocytes occurs in old age but there is no increase in lymphocytes expressing pan-T cell markers[4].

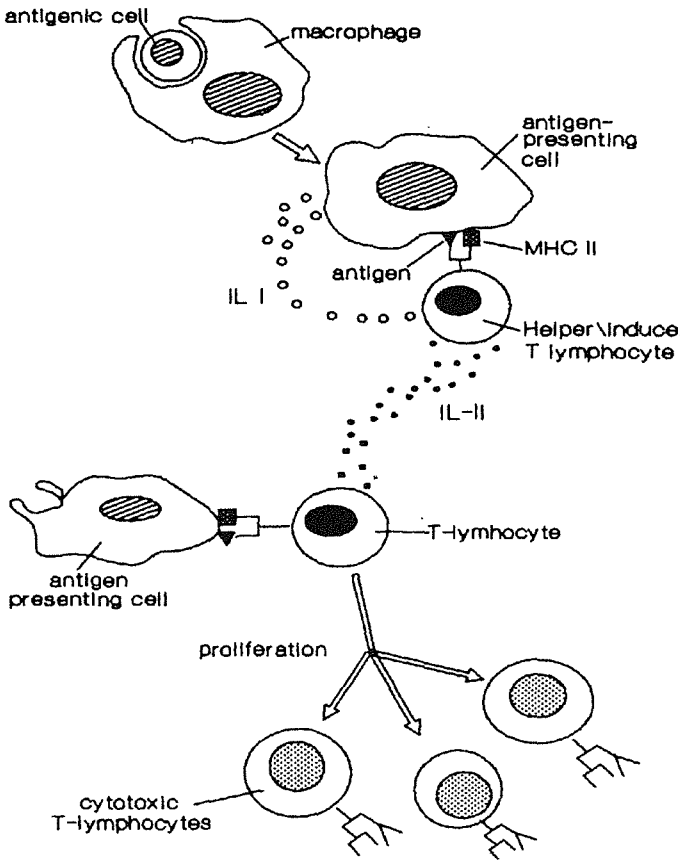
A decline in T-cell mediated responses is usually observed in old age. This includes, for example, delayed skin hypersensitivity to common test antigens and particularly a decrease in primary reactions in response to antigens to which individuals had not been previously exposed such as dinitrochlorobenzene, DNCB, which was used in most tests[3].

Circulating levels of natural antibodies to blood group antigens usually decrease with age. Although total immunoglobulins show no age-related changes there is a tendency for an increase in IgG and IgA and a decrease in IgM[5]. Immunological responses to vaccine preparations are less marked in old age although they may still confer adequate levels of immunological protection[6,7]. These and other observations suggestive of decreased humoral immunity might appear to be in conflict with the finding that circulating B-lymphocytes do not decrease with age. They are also apparently contradictory to the increased prevalence of auto-antibodies and benign monoclonal gammopathies with increasing age[8,9].

It is now appreciated that T- and B- lymphocytes functions are not necessarily related to their absolute or relative numbers. The impaired antibody production by B cells and impaired T-lymphocyte mediated responses can both be explained on the basis of altered responsiveness or activity of T4 inducer - helper and T8 suppressor lymphocytes and the complex network in which the various cells of the immune system are inter-related.

T-lymphocytes recognise an antigen only if it has previously been taken up by another cell, usually a macrophage, which then presents it as 'processed' antigen on its cell surface in association with the major histocompatibility (MHC Class II) proteins. These antigen-presenting cell release interleukin I which acts as a mediator activating the T4 class of lymphocytes (Figure 1). These in turn secrete interleukin II under whose influence other T cells proliferate into large clones of mature T cells. These include T4 helper-induced cells or T8 suppressor lymphocytes or cytotoxic cells which all display antigen-specific receptors on their surface. These receptors are somewhat similar to immunoglobulin, each having a constant domain and a variable one. They are bound to the plasma membrane. The cytotoxic cells can then interact with the antigen cell while helper and suppressor cells retain regulatory roles.

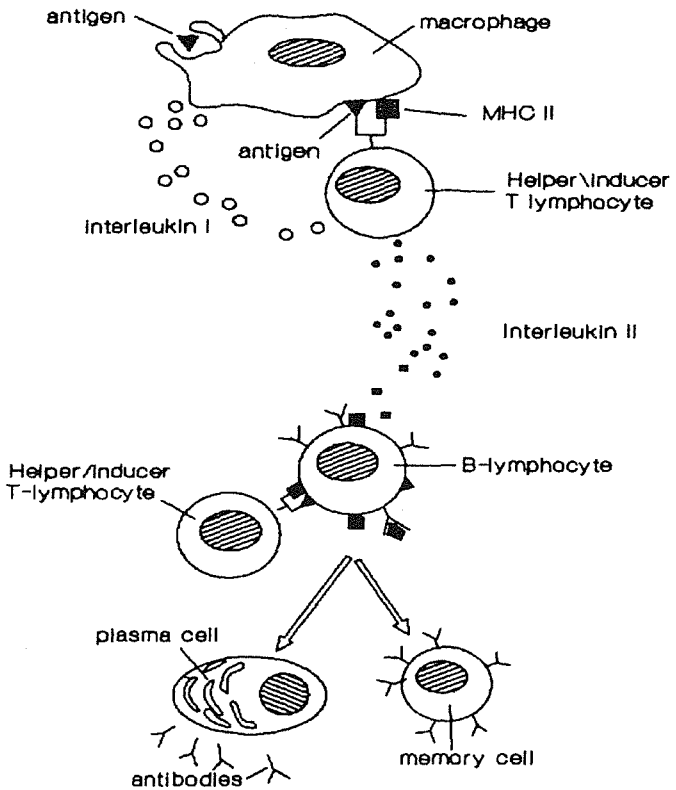
Fig. 1. T-lymphocyte response



Similarly helper-inducer T4 lymphocytes are necessary for B cell hormonal responses (Figure 2). B cells bear immunoglobulin as receptors on their surface; these can recognise and bind freely - circulating antigens but this process is not enough to stimulate proliferation of B cell clones and their subsequent differentiation to antibody producing cells. They also require the collaboration of T4 helper lymphocytes which have interacted with antigen-presenting cells. Stimulated T4 lymphocytes then bind to B cells bearing the same antigen; interleukin 2 (IL 2) is released affecting

the growth of B lymphocytes which settle in B lymphoproliferative zones and form clones of cells which eventually differentiate into antibody-producing plasma cells or transform into memory cells for permanent immunity.

Fig. 2. B-lymphocyte response



Various other interleukins have been described, including IL-3 which is produced by mitogen- or antigen-stimulated T-lymphocytes and is involved in the regulation of growth and differentiation of pluripotent stem cells, leading particularly to the differentiation of myeloid cells. IL-4, IL-5 and IL-6 cause activation and growth of B cells, as well as promoting differentiation of other cells such as eosinophils by IL-5.

Interferon, lymphotoxins and prostaglandins which are produced by lymphocytes and macrophages also have immuno-modulatory or immuno-regulating roles in specific T cell responses. Thus specific immune responses are regulated by a complex network of cellular and humeral interactions. They are controlled by specific mediators and require also the expression of appropriate receptors on the plasma membranes of the responsive cells.

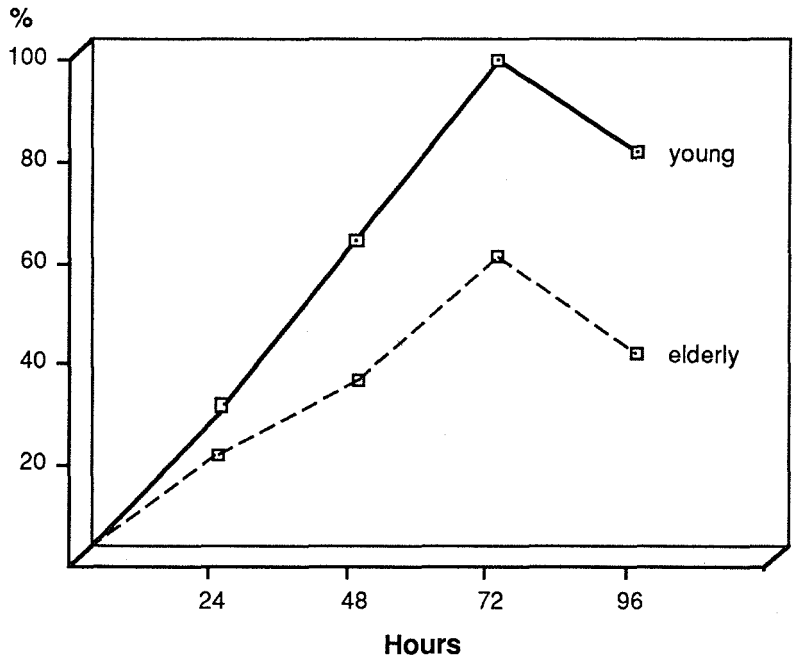
Both mediators and receptors are under director genetic control. The initiation of the immune response following antigen stimulation depends on the synthesis of a protein mediator in an individual cell, which in its turn depends on the processes of DNA transcription and mRNA translation. The cascade of immunological events also depends on the expression of specific surface receptors which are also proteins and depend on the process of transcription and translation. It is clear that the failure of gene expression of a mediator or a receptor could disrupt the whole chain or network of events and the proliferation of cell clones which constitute an immune response.

It is conceivable that the altered immunological responses in aging may be the results of failure of gene expression of a mediator or a receptor.

There is now an increasing body of evidence which implicates age-related genetic changes in the altered functioning of the immune system in elderly individuals. Most of this evidence derives from the behaviour of mitogen-stimulated lymphocytes in vitro.

In such systems it has been shown that the protein synthetic activity of mitogen-stimulated lymphocytes decreases with age. This decrease parallels the rate of lymphocytes proliferation. Figure 3 shows the incorporation of leucine in phytohaemagglutinin (PHA) - stimulated lymphocytes. The peak uptake corresponds to the peak proliferative activity at 72 hours, after which there is a rapid decline. In lymphocytes from elderly individuals (70-80 years of age) the rate of uptake at all stages of culture is much lower than that of lymphocytes from young (23-32 years old) adults[10]. These observations apply to both rats and human subjects.

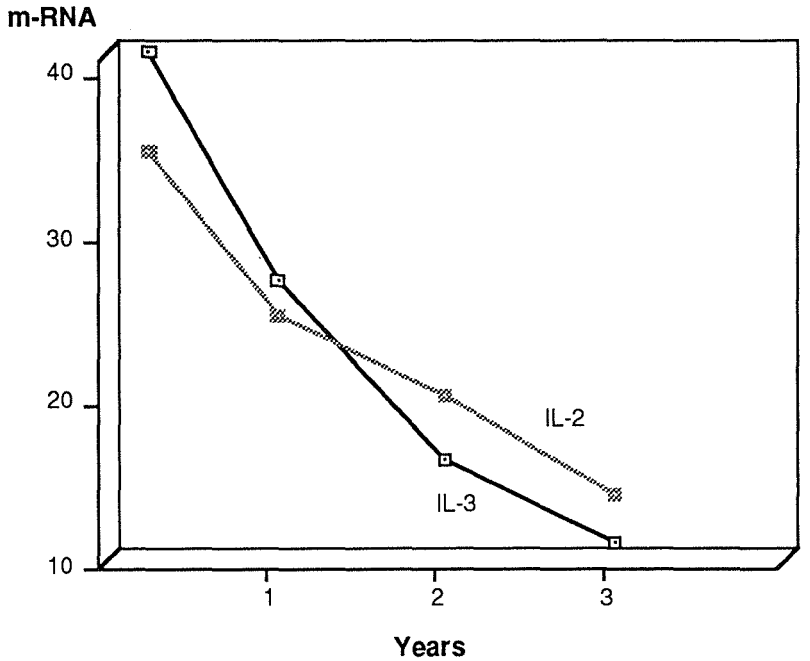
Fig. 3. Incorporation of leucine by PHA-stimulated lymphocytes



It has also been shown that interleukin 2 production in mitogen-stimulated lymphocytes decreases with age (Figure 4). IL-2 in culture supernatant can be estimated by the use of monoclonal antibodies which react specifically with IL-2. Measurement of the rate of incorporation of radioactive valine or leucine into the molecules of IL-2 demonstrates that this decline is the result of decreased synthesis and not increased utilisation or destination[11].

The levels of mRNA which codes for IL-2 can be measured by using a specific single-stranded cDNA probe. This too shows a decrease which, in 3 year old rats, is about 25% of the levels in lymphocytes from young rats. Interleukin 3 shows a similar decline again to about 25% of the levels in younger animals. These results show that there is an age-related decline in interleukin production which occurs at the transcriptional level and which implies that the age-related change must affect the DNA.

Fig. 4. Levels of IL-2 and IL-3 in aging male rats



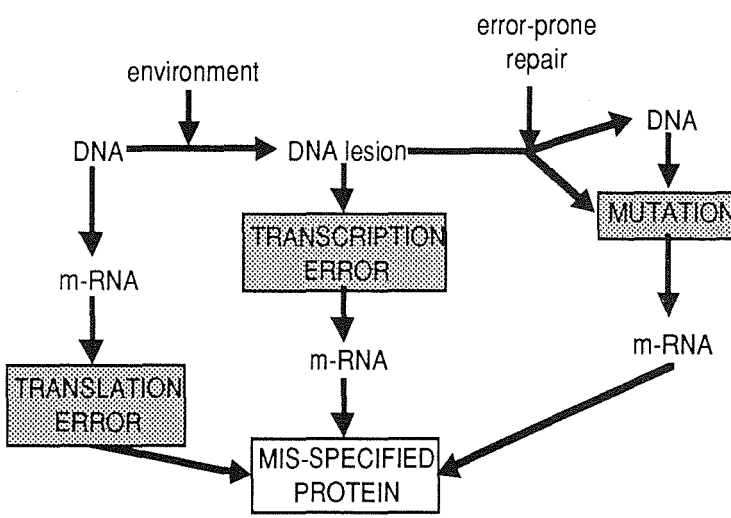
A decline in gene expression does not affect all mediators uniformly. For example it does not affect the complement system which comprises a group of proteins which are mediators responsible for non-specific functions involved in the body's defences, mainly the activation of macrophages and mast cells, opsonization (binding to antigens or antigen-antibody complexes and promoting their phagocytosis) and the cytolysis of cells by natural-killer cells. The complement system is not produced by the cells of the immune system but mostly by hepatocytes and fibroblasts.

The decline in gene expression of IL-2 and IL-3 is a first step which indicates that the fundamental age-related changes in lymphocytes may be genetic changes affecting the expression of mediators and corresponding receptors. Since these are key factors in immune responses their implications could range from a decreased recognition and cytolysis of malignant cells which consequently give rise to neoplasia.

Genetic factors have been strongly proposed as possible key factors in the mechanism of aging. The genetic theory of aging has been considered to be the main rival completing with the immunological theory of aging. It seems, however, that it is possible that these two aspects of aging may be reconciled and that the basic defect in the function of lymphocytes resides in the flow of information from DNA via mRNA to protein.

This process may be disrupted either at the level of DNA and transcription or at the translational level of mRNA (Figure 5). Age-related alterations in DNA are likely to result from repeated exposure to various subtle environmental agents which continually cause DNA damage. This may include a spectrum of changes involving the sequence of bases in the DNA molecule and collectively termed DNA lesions. The DNA of elderly individuals is more susceptible to such lesions. If these lesions happen to occur in a gene which is being transcribed, the mRNA produced will be erroneous, the end result being protein mis-specification; and if this lesion happens to occur in a proliferating cell the resulting clone may be similarly affected.

Fig. 5. Possible genetic damage affecting protein synthesis



However, DNA damage is not necessarily permanent or irreversible. The fate of DNA lesions is determined by the various enzymatic activities collectively termed DNA repair which often leads to complete reversal of the DNA lesion, but may proceed incorrectly in what is termed error-prone repair resulting in a gene mutation. Again this mutation may affect a cell alone. Although transcription and translation proceed normally the intended protein would be mis-specified and therefore abnormally functioning or non-functioning.

The aging individual is possibly more susceptible to error-prone repair. Various techniques are now available for detecting single strand breaks and other specific DNA lesions and also for assessing DNA repair. Such techniques are now opening the way for more critical studies on molecular aspects of aging.

Normally transcribed mRNA can still give rise to protein mis-specification because of translational errors caused by faults in cellular control mechanisms. Perhaps, in the lymphoid system age-related DNA changes are more prominent than in other tissues because of its immense diversity of function and its numerous far-reaching implications.

The molecular age-related changes in the immune system are only beginning to be clearly understood. They are fundamental issues which need to be better elucidated in order to provide better insight into the immunological age-related changes which affect so many other processes whether related to the recognition of hetro-antigens affecting the susceptibility to infections, the failure of recognition of malignant cells which leads to neoplasia or the mis-recognition of self-antigen which underlies many diseases in the elderly.

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