英裔老年人群的身體組成與淋巴細胞亞單位間的關係

Body composition and lymphocyte subsets in an Anglo-Celtic elderly population

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Aging is accompanied by changes in body composition. Furthermore, with aging, immunocompetence is decreased. Insufficient studies are available on the relationships between body composition (fat, bone and non-bone lean mass) and immune function. Aside from the possible adverse effects on immune function of undernutrition, it has been reported that there is an increased incidence of respiratory tract infections and postoperative sepsis in the obese. Further investigation is required as to whether this observation could be attributed to impairment of immune function.

The aim of the present study is to investigate the association between body composition and lymphocyte subsets in apparently healthy elderly people.

One hundred and forty-three free-living elderly and representatively sampled (67 men and 76 women), aged 67 – 86 years, were studied. Body composition was measured using anthropometry and bioelectrical impedance analysis. A subset of 30 elderly subjects (14 men and 16 women) had their lymphocyte subsets measured. Lymphocyte subsets (CD3, CD4, CD8 and CD19) were assayed using an EPICS 752 flowcytometer.

Using partial Spearman correlation analysis, it was found that in elderly men, there were negative correlations between any of body mass index (BMI), total body fat (using BIA), fat-free mass (using anthropometry) with CD8 count (r=-0.56, P=0.04; r=-0.71, P=0.006; and r=-0.63, P=0.02, respectively). On the other hand, in elderly women, there were positive correlations between total body fat (using BIA) with any of CD3, CD4 and CD19 counts (r=0.53, P=0.04; r=0.63, P=0.01; and r=0.53, P=0.04, respectively).

These findings suggest that although lean mass and immunocompetence decline with age, in apparently healthy elderly population, there are likely to be protective mechanisms which could prevent deterioration of immunocompetence caused by changes of body composition, and that these might be gender dependent. Further studies are required to clarify the interplay between body composition, gender and immuncoompetence in the elderly.

Introduction

Body composition is one of the various biological indices which predicts disease occurrence. For example, many studies provide evidence that obesity is associated with diabetes mellitus, particularly non-insulin-dependent diabetes mellitus (NIDDM)[1]. Low and high body mass indices (BMI) are associated with greater total mortality[2].

Aging is accompanied by changes in body composition. With ageing, lean body mass is reduced, while body fat mass is increased[3,4]. These changes in body composition in the elderly are compounded by the fact that immunocompetence in the elderly is often decreased compared to their younger counterparts[5]. However, there are few data on the prevalence of immunodeficiency in the elderly. On current evidence, it could be expected that the elderly are at high risk of infectious diseases. Indeed, infectious diseases are represented amongst the five major causes of death in an elderly population in an industrialized society[6].

Malnutrition may play a role in the development of immunodeficiency in elderly populations[5]. In different studies, nutritional supplementation has improved the immunocompetence and decreased proneness to infection in elderly individuals[5,7,8].

There are insufficient studies to observe the relationships between body composition and immunocompetence. Retrospective studies report an increased incidence of respiratory tract infections and postoperative sepsis in the obese[9]. It was not resolved as to whether the infections were caused by mechanical disadvantage[10], micronutrient deficiencies[11], immune function impairment, or an interaction among these in the obese.

The aim of the present study is to identify associations between a more detailed assessment of body composition and lymphocyte subsets in the elderly. This study may help explain the pathogenesis of immune-mediated diseases in elderly populations.

Subjects and methods

Subjects
Using telephone directory listings[12], 143 apparently healthy and free-living elderly subjects of Anglo-Celtic ancestry (67 men and 76 women), aged 67 – 86 years, were randomly selected from an area of five postcodes in the South-Eastern region of the Melbourne Metropolitan Area; these

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postcodes, surrounding the current geographical epicentre of Melbourne, were also characteristic of the greater Melbourne Statistical Division. No attempt was made to select subjects on the basis of body habitus, weight change, or general health status. A subset of 30 apparently healthy elderly subjects (14 men and 16 women) had their lymphocyte subsets measured. This study was approved by the Human Ethics Committee of Monash Medical Centre.

Body composition measurements

Anthropometry. With the subjects wearing light clothes and no shoes, body weight was measured to the nearest 0.1 kg, and height to the nearest 0.1 cm. Four skinfold thicknesses (triceps, biceps, suprailiac, and subscapular) were twice measured with the same Harpenden caliper to the nearest 0.2 mm and were averaged for use in later analyses. Mid-upper-arm, waist (umilical level) and hip (maximal gluteal level) circumferences were measured with a non-elastic measuring tape to the nearest 0.5 cm. The waist-to-hip circumference ratio (WHR) was calculated to measure abdominal fat distribution. Body mass index (BMI) was calculated as body weight (BWT) in kilograms divided by height (HT) in metres, squared. Percent body fat was estimated using the Durnin and Womersley tables for age 50 years and over, since they do not provide for older age groups[13]. Total body fat (TBF-A) was calculated by multiplying per cent body fat by BWT. Fat-free mass (FFM-A) was obtained by subtracting TBF from BWT.

Bio-electrical impedance analysis. Resistance and reactance were measured by a four-terminal impedance analyser (RJL-systems, Detroit) while the subjects were supine with arms and legs abducted, and not touching the body. Two current electrodes were placed, one each on the dorsal surfaces of the right hand and of the foot, at the distal metacarpals and metatarsals, respectively. Two detector electrodes were placed, one each at the right pisiform prominence of the wrist and between the medial and lateral malleoli at the right ankle. The resistive component of body impedance between the right wrist and right ankle was then measured to the nearest Ohm. Fat-free mass (FFM-BIA) was estimated using the Lukaski equation[14]. Total body fat (TBF-BIA) was then obtained.

Lymphocyte subset analysis

Venous blood samples were obtained in EDTA-treated tubes early in the morning, after an overnight fast. Blood specimens were set up on the same day of collection. 100 µL of whole blood was taken and placed in a Wasserman tube. The following FITC and PE conjugated antibody combinations were used: G1+G1 (negative control by isotype matched), CD45+CD14, CD3+CD4, CD3+CD8, CD2+CD19 (Coulter). Ten microlitres of antibody was added to each labelled tube, mixed with a Coulter QPREP machine and then left covered for 60 min at room temperature. Cells were lysed and fixed using the 35-s cycle of QPREP. The specimens were then stored in the dark, overnight at 4 °C. On the next day, cell suspensions were transferred into appropriately labelled Eppendorf tubes (with caps on) and spun at 1200 rpm for 5 min. The supernatant was removed and the cells were resuspended in 300 µl PBS/1% BSA/0.01% NaAz and then 2% paraformaldehyde was added. Enumeration of lymphocyte subsets was done using an EPICS 752 flowcytometer (Coulter). Total lymphocyte count was used to obtain the absolute counts of CD3 (total T-cells), CD4 (T-helper cells),

CD8 (T-suppressor cells) and CD19 (B-cells).

Statistical analysis

Results are expressed as mean ± SD. Covariance and non-parametric Spearman correlation analyses using SAS software were performed[15].

Results

There were no differences in age or BMI between men and women. As expected, gender differences in WHR, TBF and FFM were significant (Table 1).

Table 1. Body composition characteristics of the subjects, by gender.

Variable	Men (n=67)	Women (n=76)	P
Age (year)	72.4 ± 4.2	72.6 ± 5.8	NS
BMI (kg/m^2)	25.9 ± 3.3	26.2 ± 3.4	NS
WHR	0.97 ± 0.05	0.86 ± 0.06	<0.0001
TBF - A (kg)	19.3 ± 5.7	24.2 ± 6.0	<0.0001
TBF-BIA (kg)	16.8 ± 6.2	20.2 ± 5.6	<0.001
FFM - A (kg)	56.1 ± 7.3	42.1 ± 6.1	<0.0001
FFM-BIA (kg)	58.3 ± 6.1	45.6 ± 7.8	<0.0001

mean ± SD.
TBF - A= total body fat by anthropometry.
TBF - BIA= total body fat by BIA.
NS= not significant.
FFM-A= fat-free mass by anthropometry.
FFM-BIA= fat-free mass by BIA.

There were no significant gender differences in age, CD3, CD4, CD8 and CD19 counts (Table 2).

Table 2. Lymphocyte subsets of 30 elderly subjects, by gender.

Variable	Men (n=14)	Women (n=16)	P
Age (year)	73.3 ± 4.6	74.5 ± 6.2	NS
CD3 (x10^6/l)	1002 ± 379	1062 ± 447	NS
CD4 (x10^6/l)	722 ± 237	722 ± 328	NS
CD8 (x10^6/l)	398 ± 173	434 ± 289	NS
CD19 (x10^6/l)	155 ± 69	175 ± 65	NS

mean ± SD.
NS= not significant.

In elderly men, there were negative correlations between any of BMI, TBF-BIA, FFM-A with CD8 count (r=0.56, P=0.04; r=0.71, P=0.006; and r=0.63, P=0.02, respectively). On the other hand, in elderly women, there were positive correlations between TBF-BIA with any of CD3, CD4 and CD19 counts (r=0.53, P=0.04; r=0.63, P=0.01; and r=0.53, P=0.04, respectively) (Table 3).

Table 3. The Spearman's partial correlation coefficients for relationships between body composition and lymphocyte subsets in 30 elderly subjects, by six.

Body composition	Men lymphocyte subsets				Women Lymphocyte subsets			
	CD3	CD4	CD8	CD19	CD3	CD4	CD8	CD19
BMI	-0.18	0.05	-0.56*	-0.07	0.44	0.46	0.29	0.41
TBF-A	-0.07	0.20	-0.52	0.01	0.32	0.24	0.30	0.38
FFM-A	-0.06	0.19	-0.63*	-0.01	0.35	0.21	0.22	0.06
TBF-BIA	-0.41	0.01	-0.71**	-0.04	0.53*	0.63*	0.34	0.53*
FFM-BIA	0.11	0.23	-0.43	0.05	0.34	0.19	0.24	0.09

*P<0.05; **P<0.01.

Discussion

The analysis presented here is predicated on an assumption

that protein status, which is represented by lean body mass or FFM, is a determinant of immunocompetence (in this case CD4). It has been well documented that there is a close relationship between the body's protein store and various physiologic functions. When more than 20% of body protein has been lost, most physiologic functions are significantly impaired[16]. In the clinical setting, if has been reported that a deficit of total body protein (greater than 20% is associated with more postoperative complications such as the major complications of pneumonia and wound healing[17]. Combined with the paediatric experience[18], it can be concluded that protein undernutrition is strongly associated with immune function impairment.

The elderly represent a unique subset of the population with limited nutritional and physiological reserve capacities. Both cross-sectional and longitudinal studies have demonstrated that there is a linear decline of lean mass with aging[3,4]. The current literature suggests a possible role for cytokines like interleukin-1 (IL-1) and tumor necrosis factor (TNF) in the development of lean body mass reduction in the elderly[19-23]. On the other hand, current immuno-epidemiological data are insufficient to support this view. Whether there is a linear decline of certain lymphocyte subsets such as CD4, with aging, will require an age-related community-based immuno-epidemiological study, preferably longitudinal.

The present study does not provide adequate evidence that FFM is a determinant of immunocompetence. The first possible explanation is that the correlations between body composition, such as BMI, TBF and FFM, and lymphocyte subsets may not be linear. The second possible explanation is that, insofar as body composition is concerned, FFM consists severally of total body water, protein and bone mass[24,25]. Protein and bone mass constitute probably two of the most important nutritional reserves in the elderly[26]. Neither anthropometry nor BIA was able to distinguish these three components. Their interaction with lymphocyte subsets thus cannot be explored extensively. Furthermore, the protein component of FFM also includes lymphoid organs, such as spleen and lymph nodes. The lymphocyte pool in these organs effect blood lymphoctes[27]. It is questionable as to what extent a reduction of FFM in the elderly would contribute to the reduction of lymphoid organ mass, and thereby compromises the blood lymphocyte count.

These preliminary findings provide an indication that fat-

ness has beneficial effects on certain lymphocyte subsets in elderly women, since total body fat had positive correlations with CD3, CD4 and CD19 counts. In elderly men, total body fat had negative correlations with CD8 counts (Fig. 1). The mechanisms which operate to allow these findings are not clear, although clearly gender acts as a differentiating factor. Small sample size in this study also means that judgement should be reserved.

It may be that, in the elderly, each compartment of body composition plays its own role as a determinant of certain lymphocyte subsets, which in turn, can be used as a predictor of morbidity and mortality[28]. Whether overall relationships are linear or unimodal, and of prospective significance to allow opportunities for useful intervention, awaits more definitive studies.

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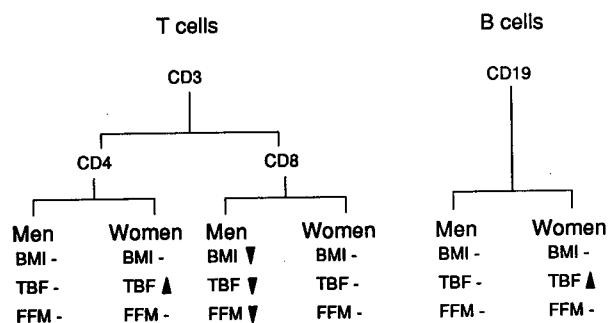
Figure 1. Body compartments, a determinant of lymphocyte status. ▲ = positive effect; - = no effect; ▼ = negative effect. T cell subsets are: CD3 (total T cells), CD4 (T-helper cells) and CD8 (T-suppressor cells).

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