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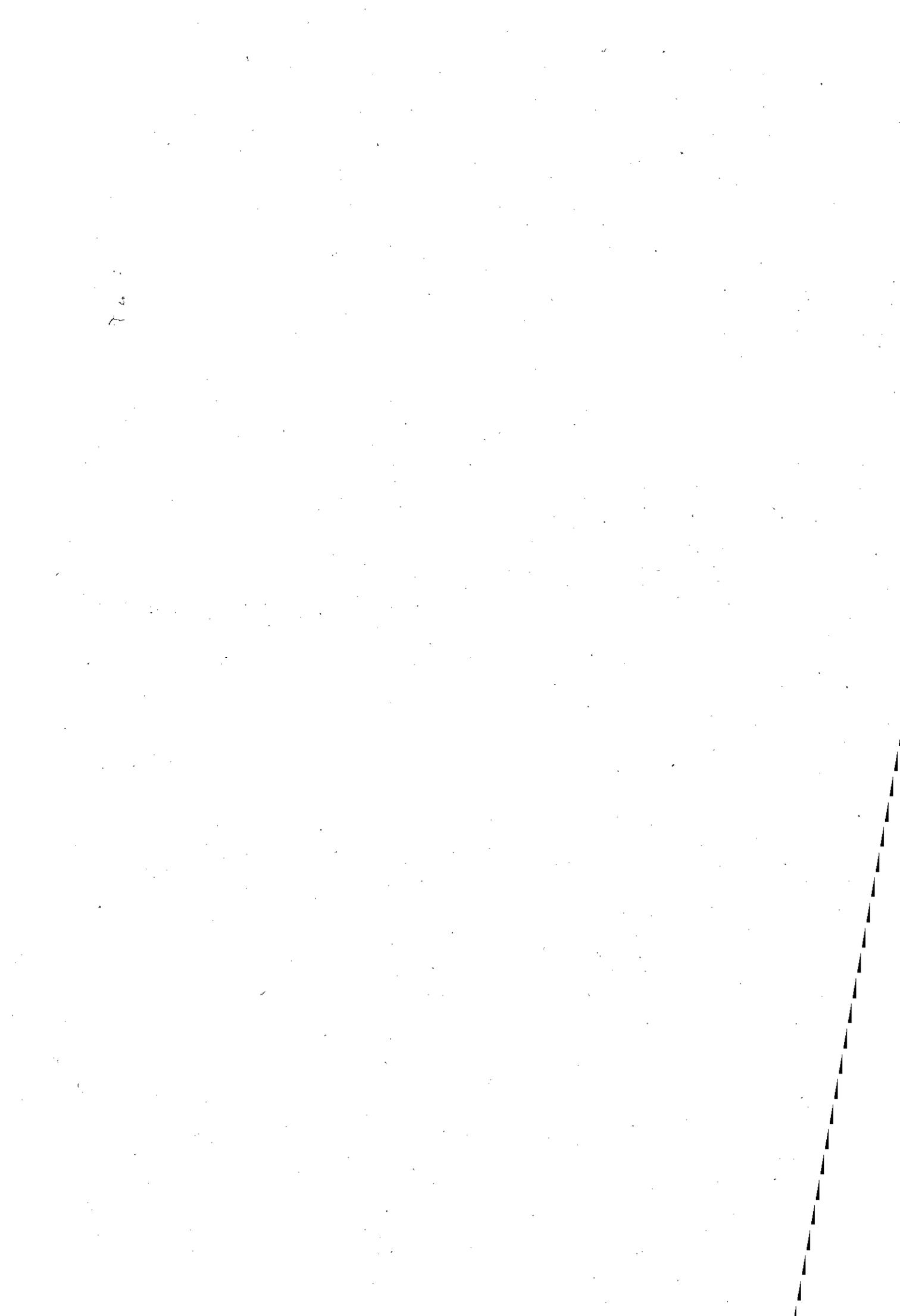
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**Human Body Composition:
Measurement and Relationship with Exercise,
Dietary Intakes and Cardiovascular Risk Factors**

By

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A Doctoral Thesis

Submitted in partial fulfilment of the requirements for the award of Doctor of
Philosophy of the Loughborough University of Technology.

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Abstract

This thesis describes studies related to human body composition, concentrating upon methodology of measurement, and a study on the influence of brisk walking programme upon healthy, previously sedentary middle-aged men.

In chapter 1, the principles of the techniques used for measurement of body composition in this thesis are discussed. The limitations and potential sources of error associated with each are discussed. The response of body composition to exercise, and the relationship of this response to changes in cardiovascular risk factors are considered. General methods are described in chapter 2.

Techniques suitable for measurement of body composition in "field" conditions are evaluated in chapters 3 and 4. Near infra-red interactance was found to under-estimate fatness, to an increasing extent with increasing fatness. Bio-electrical impedance estimates of body composition from different sets of prediction equations from the literature differed significantly. Most overestimated fatness, to an increasing extent with increasing fatness.

In chapter 5 techniques for measurement of subcutaneous adipose tissue are evaluated by comparison with A-mode ultrasound. Skinfold thicknesses were better correlated with subcutaneous adipose tissue thickness than were interactance data.

Chapters 6 and 7 describe a year-long study on the effects of a brisk walking programme on healthy, previously sedentary middle-aged men. Volunteers were randomly allocated to walking or control groups (n = 42 and 23 respectively). Brisk walking for on average 27 minutes per day was not found to influence body composition, although significant changes in lower limb skinfold thicknesses were observed. The relationship of changes in blood pressure and blood concentrations of total cholesterol, lipoprotein-cholesterol subfractions and triglycerides with changes in body composition and fat distribution is examined.

Energy intake did not change during the study, despite the expected increase in energy expenditure, and lack of change in body composition. Changes in dietary cholesterol and fatty acid intakes during the year are described, and related to changes in cardiovascular risk factors.

In conclusion, newer field techniques were not found to be a better predictor of body composition than skinfold thicknesses. Participation in the walking programme did not significantly influence body composition or energy intake.

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I am extremely grateful to all the subjects who volunteered for the studies described in this thesis for being so patient and generous with their time. Thanks must go particularly to the men in the brisk walking study, who met the large commitments of the study despite their busy lifestyles, and in some cases having to travel large distances.

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Publications

Brooke-Wavell K, Jones PRM, Norgan NG, Stensel D, Hardman AE. Influence of a twelve month programme of brisk walking on body composition of previously sedentary middle-aged men. *Ann Hum Biol* 1992; 19: 213A-214A.

Stensel DJ, Hardman AE, Brooke-Wavell K, Jones PRM, Norgan NG. The influence of brisk walking on endurance fitness in previously sedentary, middle-aged men. *J Physiol* 1992;446:123P.

Stensel D, Hardman A, Vallance D, Jones P, Norgan N, Brooke-Wavell K, Winder A. The influence of brisk walking on serum lipoprotein variables in previously sedentary, asymptomatic, middle-aged men. *Clin Sci* 1992;82(3):16P-17P.

Table of Contents

Abstract.....	ii
Acknowledgements.....	iii
Publications.....	iv
Table of Contents.....	v
List of Tables.....	xi
List of Figures.....	xvii

Chapter 1

Introduction.....	1
The two component model of body composition.....	1
Measurement of body composition.....	2
Hydrostatic weighing.....	2
Skinfold thickness.....	4
Near infra-red interactance.....	6
Bioelectrical impedance analysis.....	9
Weight and height.....	12
Measurement of subcutaneous adipose tissue and muscle thickness.....	14
Ultrasound.....	14
Skinfold thickness.....	16
Assessment of fat distribution.....	17
Circumferences and circumference ratios.....	17
Skinfold thicknesses and skinfold ratios.....	18
Association of risk of cardiovascular disease with body composition, fat distribution, dietary intakes, physical activity and plasma lipids and lipoproteins.....	20
Risk factors for cardiovascular disease.....	20
Plasma lipids and lipoproteins.....	20
Body composition.....	22
Fat distribution.....	23
Diet.....	24
Blood Pressure.....	27
Physical Activity.....	27
Summary.....	28

Chapter 2

General Methods	30
Informed consent.....	30
Anthropometry	31
Circumferences.....	31
Skinfold thicknesses.....	32
Bony diameters.....	33
Body Composition.....	35
Hydrostatic weighing.....	35
Skinfold thickness	37
Near Infra-red Interactance.....	38
Bioelectrical impedance.....	40
Ultrasound.....	42
Exercise testing.....	42
Submaximal incremental treadmill test.....	42
Grade-Lactate treadmill test	43
Endurance walk	43
One mile track walk.....	43
Collection of expired air.....	43
Analysis of oxygen and carbon dioxide	44
Analysis of blood lactate	44
Blood biochemistry	44
Blood pressure	45
Statistics.....	45

Chapter 3

Comparison of near infra-red interactance and skinfold thickness with hydrostatic weighing.....	46
Introduction.....	46
Methods	47
Subjects	47
Procedures.....	47
Results.....	48
Comparison of means of body composition techniques.....	48
Regression Analysis.....	49
Further analysis	49
Regression of near infra-red interactance with skinfold thickness	52

Correlation of optical density data with %fat	53
Contribution of variables in near infra-red interactance prediction equation to variance in body fatness.....	54
Correlation of anthropometric variables with body density	54
Discussion	56
Estimation of body composition by near infra-red interactance.....	56
Estimation of body composition from skinfold thicknesses	57
Association of anthropometric variables with body density.....	57
Conclusions.....	57

Chapter 4

Comparison of three "field" techniques for measurement of body composition: skinfold thickness, near infra-red interactance and bioelectrical impedance.....	59
Introduction.....	59
Methods	61
Subjects	61
Procedure	61
Standard Error of Measurement of Body Composition Techniques.....	62
Influence of strenuous exercise on body composition measurements.....	62
Results.....	63
Comparison of mean body composition by three techniques	63
Correlation of bioelectrical impedance and near infra-red interactance measures of body composition with those by skinfold thickness.....	64
Further analysis	65
Correlations of optical densities with body composition	67
Effect of exercise on estimation of body composition	71
Discussion	72
Bioelectrical impedance.....	72
Near infra-red interactance	73
Comparison of bioelectrical impedance, near infra-red interactance, and skinfold thickness	75
Conclusions.....	78

Chapter 5

Assessment of subcutaneous adipose tissue thickness: comparison of near infra-red interactance and skinfold thickness with ultrasound	79
Introduction.....	79
Methods	81
Results.....	83
Comparison of mean ultrasound and skinfold measures of subcutaneous adipose tissue	83
Correlations of skinfold thicknesses with ultrasound subcutaneous adipose tissue thickness measurements.....	85
Skinfold compressability.....	86
Near infra-red interactance	89
Correlation of optical densities with ultrasound subcutaneous adipose tissue thickness	91
Correlation of optical densities with skinfold thickness	92
Muscle-bone interface depth	93
Contribution of muscle thickness to optical densities	94
Discussion	95
Agreement of skinfold thicknesses with subcutaneous adipose tissue thickness	95
Agreement of near infra-red interactance with subcutaneous adipose tissue thickness	95
Conclusions.....	97

Chapter 6

The influence of brisk walking upon body composition and fat distribution of middle aged men, and associations with cardiovascular risk factors.....	98
Introduction.....	98
Methods	100
Subject recruitment.....	100
Criteria for acceptance into the study	100
Screening.....	100
Outline of study	102
The Walking programme.....	103
Test battery.....	103
Coherence.....	104
Compliance	105

Results.....	107
Influence of walking programme on maximal oxygen uptake and oxygen consumption at reference blood lactate concentration.....	107
Influence of walking programme on body composition and fat distribution.....	108
Levels of other cardiovascular risk factors during the walking programme.....	120
Discussion	126
Influence of walking programme on body composition.....	126
Influence of walking programme on fat distribution.....	127
Association of body composition with cardiovascular risk factors	127
Conclusions.....	129

Chapter 7

Influence of programme of brisk walking on dietary intakes of middle aged men, and associations with cardiovascular risk factors.....	130
Introduction.....	130
Methods.....	132
Weighed inventories.....	132
Analysis.....	133
Results.....	134
Dietary intakes at baseline.....	134
Energy intake during walking programme.....	135
Seasonality of energy intake.....	135
Composition of diet.....	136
Fatty acid and Cholesterol intakes.....	138
Association of dietary intakes with levels of cardiovascular risk factors at baseline	139
Association of changes in diet with changes in cardiovascular risk factors	141
Discussion	142
Compliance	142
Dietary intakes at baseline.....	142
Influence of the walking programme on energy intake.....	143
Seasonality of energy intake.....	143
Composition of the diet during the walking programme	144

Associations of dietary intakes with cardiovascular risk factors	144
Conclusions.....	145
 Chapter 8	
Discussion	146
“Field” methods for measurement of human body composition.....	147
Influence of a year-long programme of brisk walking.....	150
 Bibliography.....	156
 Appendix I	
Statement of informed consent	183
Appendix II	
Leaflet for recruitment of subjects for brisk walking study	184
Appendix III	
Description of brisk walking study given to potential volunteers	185
Appendix IV	
Instructions and forms for recording of weighed inventories.....	189
Instruction sheet.....	189
Example Recording Sheet.....	191
Weighed Inventory Recording Sheet	192

List of Tables

Table 1.1	Reported estimates of fat-free density.....	3
Table 2.1	Standard error of measurement of anthropometric variables	34
Table 3.1	Characteristics of middle-aged men and women runners.....	47
Table 3.2	Mean, standard deviation and range of body composition estimates by hydrostatic weighing (HW), skinfold thickness (SKF), and near infra-red interactance (NIRI)	48
Table 3.3	Regression of skinfold thickness (SKF) and near infra-red interactance (NIRI) estimates of body composition with those from hydrostatic weighing (HW): correlation coefficient (r) and standard error of estimate of regression (SEE).....	49
Table 3.4	Bland and Altman analysis: Mean and standard deviation of difference between body composition estimates by skinfold thickness/near infra-red interactance and hydrostatic weighing	51
Table 3.5	Bland and Altman analysis: Correlation coefficient and slope of regression line fitted to plot of mean of two methods against difference between methods.....	52
Table 3.6	Correlation coefficient and standard error of estimation of regression of skinfold thickness (SKF) with near infra-red interactance (NIRI).....	52
Table 3.7	Correlation of optical densities at 940 and 950 nm with % fat by hydrostatic weighing	53
Table 3.8	Correlation of anthropometric variables with body density in middle-aged men.....	55
Table 4.1	Characteristics of subjects.....	61

Table 4.2	Mean and one standard deviation of %fat, fat free mass, and fat mass of 27 male and 27 female young adults by skinfold thickness (SKF), bioelectrical impedance (BIA) and near infrared interactance (NIRI)	64
Table 4.3	Correlation and standard error of estimation of estimates of % fat, fat free mass, and fat mass by bioelectrical impedance (BIA) and near infra-red interactance (NIRI) with those from skinfold thickness.....	65
Table 4.4	Mean and standard deviation of difference between estimates of body composition by bioelectrical impedance/near infra-red interactance techniques and those by skinfold thickness.....	66
Table 4.5	Relation of the difference between body composition method and skinfold thickness with the mean of the method and skinfold thickness: correlation coefficient (r) and slope	67
Table 4.6	Correlation of optical density at five sites with body composition by skinfold thickness.....	68
Table 4.7	Correlation of optical densities and skinfold thickness at biceps site with body fatness (bioelectrical impedance)	69
Table 4.8	Correlation of near infra-red optical densities at biceps (B), triceps (TR), subscapular (SS), suprailiac (SI) and anterior thigh (TH) sites with body composition variables by bioelectrical impedance analysis	70
Table 4.9	Mean, standard deviation and range of % fat of 10 men by skinfold thickness (SKF), bioelectrical impedance (BIA) and near infrared interactance (NIRI), before and after a circuit training session	71
Table 5.1	Standard error of measurement of skinfold thickness, ultrasound depths, and near infra-red interactance.....	82

Table 5.2	Mean, standard deviation, and range of skinfold thicknesses and ultrasound adipose tissue muscle interface depth	83
Table 5.3	Correlation of skinfold thicknesses with ultrasound measurements of subcutaneous adipose tissue thickness.....	85
Table 5.4	Compressability of skinfolds.....	86
Table 5.5	Analysis of variance of differences between sites and sex in skinfold compressability.....	88
Table 5.6	Analysis of variance of differences between individuals in skinfold compressability	88
Table 5.7	Mean and standard deviation of optical densities at 940 and 950 nm.....	89
Table 5.8	Correlation of optical density at 940 nm with optical density at 950 nm.....	91
Table 5.9	Correlation of near infra-red optical densities with ultrasound measurements of subcutaneous adipose tissue thickness.....	92
Table 5.10	Correlation of near infra-red optical densities with skinfold thicknesses	93
Table 5.11	Mean, standard deviation, and range of muscle-bone interface depths.....	93
Table 5.12	Correlation coefficients of multiple regression of fat depth and muscle depth by ultrasound with optical densities.....	94
Table 6.1	Timescale of testing the three cohorts in the brisk walking study.....	102
Table 6.2	Indices of fat distribution used in brisk walking study.....	104
Table 6.3	Characteristics of Walkers and Controls before walking study	105

Table 6.4	Reported time spent walking during each quarter of walking study.....	106
Table 6.5	Maximal oxygen uptake and oxygen consumption at 2mmol/l blood lactate of walkers and controls at baseline and 3, 6 and 12 months after the start of the walking programme	107
Table 6.6	Weight of walkers and controls before and at 3, 6 and 12 months after the start of the walking programme	108
Table 6.7	Analysis of variance with repeated measures of body weight of walkers and controls	108
Table 6.8	Percentage body fat, fat mass and fat-free mass of walkers and controls before and at 3, 6 and 12 months after the start of the walking programme, by hydrostatic weighing (HW) and skinfold thickness (SKF)	109
Table 6.9	Weight and % fat of 12 men during a second year of involvement in the brisk walking study	111
Table 6.10	Waist hip ratio of walkers and controls before and at 3, 6 and 12 months after the start of walking programme	113
Table 6.11	Skinfold thickness ratios at baseline and 3, 6 and 12 months after start of walking programme	115
Table 6.12	Cross-sectional area of muscle and bone at mid-thigh and maximal calf sites in walkers and controls	116
Table 6.13	Principal component analysis of skinfold thicknesses: factor loadings.....	117
Table 6.14	Correlation of changes in body composition and fat distribution with amount of brisk walking performed and changes in fitness	118
Table 6.15	Correlation of body composition and fat distribution indices at baseline with cardiovascular risk factors.....	122

Table 6.16 Correlation of changes in body composition and fat distribution with changes in cardiovascular risk factors.....	125
Table 7.1 Mean dietary intakes of walkers and controls at baseline (compared with national average).....	134
Table 7.2 Energy intake (MJ), weight (kg) and % fat of walkers and controls at baseline and at 3, 6 and 12 months after start of walking programme	136
Table 7.3 Correlation of dietary intakes with cardiovascular risk factors at baseline	140
Table 7.4 Correlation of changes in dietary intakes with changes in cardiovascular risk factors.....	141

List of Figures

Figure 1.1: Diagram of near infra red spectra of water and fat	7
Figure 1.2: Diagram of oscilloscope display during ultrasound measurement of a single interface.....	15
Figure 2.1: Measurement of body composition by hydrostatic weighing	35
Figure 2.2: Diagram of instrument used for measurement of body composition by near infra-red interactance	38
Figure 2.3: Measurement of body composition by bioelectrical impedance.....	41
Figure 3.1: Bland and Altman plot comparing % fat by near infra-red interactance (NIRI) with % fat by hydrostatic weighing (HW).....	50
Figure 5.1: Mean skinfold thickness and subcutaneous adipose tissue thickness at five sites in men and women.....	84
Figure 5.2: Mean skinfold compressability at five sites in men and women.....	87
Figure 5.3: Mean optical densities at five sites in men and women	90
Figure 6.1: Outline of walking study	101
Figure 6.2: Mean body circumferences at five sites during walking programme	112
Figure 6.3: Mean skinfold thicknesses at eight sites during walking programme	114
Figure 6.4: Levels of cardiovascular risk factors during walking programme.....	121
Figure 7.1: Proportion of energy from protein, carbohydrate, fat and alcohol during the walking programme	137

Chapter 1

Introduction

Body composition has been associated with a number of chronic diseases, including coronary heart disease (Seidell 1989). Mortality rates from coronary heart disease in the UK are amongst the highest in the world: the disease is responsible for the death of one man in 11 before the age of 65 (National Forum for Coronary Heart Disease Prevention, 1988). The alarming incidence of coronary heart disease in affluent societies has emphasised the importance of measurement of body composition, for research purposes as well as identification of individuals at risk. The more traditional application of body composition in assessment of nutritional status also remains important, particularly in developing countries.

These applications of body composition often require that measurements are made on large numbers of people. For this to be possible, measurement is likely to be required to be fast, cheap, socially acceptable, and often performed in "field" conditions. Many techniques for measurement of body composition will thus be inappropriate for these requirements.

Techniques suitable for field or large scale studies are evaluated in several populations in the first studies of this thesis. These results are then applied in a controlled, randomised, study of the influence of a year-long programme of brisk walking on healthy, previously sedentary middle-aged men. Responses of body composition, fat distribution and dietary intakes during this program are examined, and related to changes in cardiovascular risk factors.

The two component model of body composition

The study of human body composition involves the quantification of the anatomical or chemical components of the body. Anatomical components are those which may be isolated by dissection, such as adipose tissue, muscle, bone and viscera, whereas identification of chemical components involves the measurement of constituents such as fat, protein, and mineral.

The two component model of body composition involves division of the body into the fat mass and the fat-free mass (Keys and Brozek, 1953). The fat mass consists of the fat stored in subcutaneous, intra-thoracic, intra-abdominal, inter- and intra-muscular depots as well as the "essential lipids" included in central nervous system and bone marrow. The fat-free mass is the mass of all other chemical constituents. This division is distinct from the anatomical division of the body into the adipose tissue mass and lean body mass (Behnke et al., 1942): the adipose tissue mass also includes protein constituents of adipose tissue, whilst essential lipids not contained in adipose tissue are

included in the lean body mass. The percentage of body weight as fat (% fat) is commonly calculated, to allow some adjustment of fatness for body size.

Measurement of body composition

Interest in the measurement of the composition of the human body was revived with the work of Behnke in the 1940s. Since then many techniques have been developed. Some of the newer techniques, such as neutron activation analysis, photon absorptiometry, magnetic resonance imaging, potassium counting and total body electrical conductivity, which have been included in the reviews of Lukaski (1987) and Shephard (1990), require expensive or specialised equipment, which is not widely available. This introduction is confined to the "field" methods used in this thesis, as well as hydrostatic weighing, which is used as a reference against which the other methods are compared. Throughout, only the measurement of healthy, young to middle-aged adults is discussed, as different considerations arise in the measurement of children, the sick and elderly. The description concentrates on the general principles of the techniques, as well the limitations and sources of error associated with each. Practical considerations such as availability, expense, portability, speed of measurement, social acceptability, and requirements for observer training are also considered.

Hydrostatic weighing

Principle

Hydrostatic weighing was one of the first techniques for evaluation of human body composition. The approach was first widely applied by Behnke (1942), and refined by Goldman and Buskirk (1961) and Akers and Buskirk (1969).

The density of the body can be estimated using hydrostatic weighing. The difference in density of fat and fat-free mass is employed to estimate the fat content of the body. The difference between underwater weight and weight in air is used to calculate the weight (and hence volume) of water displaced by the body. The volume of water displaced will be equal to body volume. After adjustment of body volume for the amount of air in the lungs, the density of the body can be calculated from mass/volume.

Procedure

The weight of the body underwater is determined by submerging the subject in a tank of water, sitting in a chair or cradle which is attached to a load cell transducer, from which the underwater weight can be determined.

Measurement is generally made with the subject breathing down to residual volume (although some investigators suggest measurement is made at total lung

capacity). The breath is held until a stable reading is obtained. The weight underwater is corrected by subtracting the weight of the weighing apparatus. The volume of water displaced (which will be equal to the volume of the body) is calculated by subtracting the underwater weight from the weight in air, and dividing by the density of water (which is predicted from the temperature of the water).

The volume of air in the lungs is either predicted from height and age or vital capacity, or simultaneously measured, by rebreathing of marker gas until steady state is achieved. The volume of gas in the lungs is subtracted from the total body volume. The density of the body is calculated by dividing the weight by the corrected volume.

The % body fat can be calculated from body density using the approximation proposed by Keys and Brozek (1953) that the density of body fat at body temperature is 900 kg/m³, whilst that of the fat-free mass is 1100 kg/m³. So % body fat can be calculated according to the equation of Siri (1961):

$$\% \text{ fat} = (4950/\text{density} - 4.5) \times 100$$

Sources of error:

Biological variability will occur in the density of the fat mass and the fat-free mass. The lipid content of adipose tissue varies, causing a standard deviation due to biological variation of 0.5 % fat (Siri, 1961). More importantly the proportions and densities of the constituents of fat-free mass are subject to biological variation: the water content (Siri, 1961), proportion of protein and mineral, and density of mineral (Bakker and Struikenkamp, 1977) may vary.

This biological variation can mean that in some subjects, the density of the fat-free mass may deviate considerably from the generally applied value of 1100 kg/m³. Some reported values for the density of the fat-free mass are shown in Table 1.1. After considering these factors, Lohman (1984) estimated the error of estimating fat content from body density to be ± 3 % fat, in sedentary young men.

Table 1.1
Reported estimates of fat-free density

Fat free density (kg/m ³)	Reference
1057-1189	Werdein and Kyle (1960)
1062	Norris et al. (1963)
1087-1105	Womersley et al. (1976)
1082-1105	Heymsfield et al. (1989)

Population differences in the density of the fat-free mass have also been reported. It is lower in the obese (Womersley et al., 1976), children, women and the elderly (Lohman et al., 1984), and higher in blacks (Schutte et al., 1984) and athletes (MacDougal et al., 1983).

Sources of technical error include the estimation of the residual volume. Systematic differences in estimates of body fat from different prediction or measurement techniques have been observed (Forsyth et al. 1988). Errors may arise from the trapping of alveolar air in the lungs during forced expiration, or the solution of the marker gas in the lungs.

The presence of gas in the intestines, or air trapped in the clothing, may affect the measurement. The intestinal gas content may be in the region of 50 - 300 ml (Bedell et al. 1956). However Durnin and Satwanti (1982) found consumption of carbonated drink prior to measurement to change the estimate of fatness by only about 1.5 % fat, whilst consumption of a meal prior to measurement made a difference of 1 % fat.

General considerations

Despite the possible sources of error discussed above, hydrostatic weighing is still commonly used as a "gold standard" in evaluation of other techniques, as it relies upon applying assumptions to a direct measurement of the body, whilst other indirect methods (such as bioelectrical impedance, near infra-red interactance and skinfold thickness) only predict the value obtained by a direct technique.

Hydrostatic weighing is therefore used as a reference technique against which other techniques are compared in chapter 3. It is also used for assessment of the body composition of middle-aged men during involvement in a brisk walking programme in chapter 6.

Skinfold thickness

Principle

The estimation of body fatness from skinfold thickness relies upon the relationship between the quantity of subcutaneous fat at particular sites, and the total body fatness. The relationship between subcutaneous and total fatness is curvilinear (Allen 1956), requiring transformation of skinfold data.

A large number of prediction equations have been developed relating the skinfold thickness at particular sites with body fatness determined hydrostatically or by other methods. The equations of Durnin and Womersley (1974), are age and sex specific, and predict body density from the logarithm of the sum of four skinfold thicknesses (biceps, triceps, subscapular and suprailiac). The equations of Jackson and Pollock (1978) for men use a quadratic equation including the sum of seven skinfold thicknesses (chest,

axilla, triceps, subscapular, abdomen, thigh, suprailiac), age, and wrist and forearm circumferences. The Jackson et al. (1980) equations for women are again quadratic, including the sum of triceps, thigh, and suprailiac skinfolds, age, and gluteal circumference.

Sources of error

The importance of the methodology in generating the prediction equations has been stressed by a number of authors (Katch and Katch 1980; Lohman 1981; Jackson 1984; Slaughter et al. 1984; Norgan and Ferro-Luzzi 1985; Shephard 1990). Recommendations have been that data should be transformed to take account of curvilinearity. Skinfold data should be summed prior to analysis, since a high degree of inter-correlation is observed. A large sample should be used containing a wide range of fatness, age, and sex. The equations should then be cross-validated on a second "validation" sample, since equations tend to be specific to the sample in which they are generated.

Lohman (1981) identified the sources of error in the estimation of body density from skinfold thickness to consist of the following: inter-individual differences in the distribution of subcutaneous fat; inter-individual differences in the ratio of total subcutaneous fat to total body fat; and technical error in the measurement of skinfolds. He estimated the sizes of these errors to be 1.8, 2.5, and 0.5 % fat respectively, giving a total error of 3.3 % fat. This theoretical error is slightly lower than the standard error in predicting hydrostatically determined body fat from skinfold thickness observed by Durnin and Womersley (1974): standard errors of the estimation of % fat were 3.5 and 5 for women and men respectively.

Population differences in the proportion of fat situated subcutaneously, and the compressability of skinfolds (discussed below) have been reported. Differences exist between different ages, sexes and levels of fatness (Edwards et al., 1955; Allen et al., 1956; Durnin and Womersley, 1974). Moreover ethnic differences have been observed (Jones et al., 1976). Some equations validated on the general population have been found to be unsuitable for the measurement of athletes (Thorland et al., 1984).

The measurement of skinfold thickness is particularly liable to inter-observer differences (Womersley and Durnin, 1973, Burkinshaw et al., 1973), with differences in the percentage of body weight as fat being up to 6% in untrained observers. This stresses the need for training of observers.

The type of caliper used has been reported to affect predicted fatness (Lohman et al., 1984), whilst the pressure exerted by the calipers may affect the measurement (Brozek and Kinzey, 1960). A standard pressure of 10 ± 2 g/mm² has been recommended (Edwards et al., 1955).

The length of time for which the skinfold is compressed will also affect the measurement. It has been recommended that the calipers are applied for two (Edwards et al., 1955) or four (Becque et al., 1986) seconds.

Evaluation

The ability of skinfold thicknesses to predict body density has been evaluated by many investigators, and discussed in a number of reviews (Lohman, 1981; Coward et al., 1988). Provided observers are trained, and prediction equations are well validated and suitable for the population being studied, skinfold thicknesses have been found to be a reasonably good predictor of body density, with a standard error of estimate of 91 kg/m³ in samples of mixed age and sex, and 70 kg/m³ in young men (Lohman, 1981).

General considerations

Providing that observers are well-trained, measurement sites and methodologies are standardised, and appropriately validated equations suitable to the population being studied are used, skinfold thicknesses remain an important tool for assessment of human body composition. Measurement is cheap, fast, and does not require much subject co-operation. Moreover the size and portability and speed of measurement makes the technique particularly suitable for field studies, where many other techniques would be impractical.

Estimates of body composition from skinfold thickness in groups of middle-aged men and women athletes are compared with hydrostatic weighing in chapter 3. Skinfold thicknesses are also compared with other field techniques in chapter 4. The use of skinfold thicknesses in measurement of subcutaneous adipose tissue thickness is evaluated in chapter 5. In chapter 6, skinfold thicknesses are used for assessment of changes in body composition and fat distribution in middle-aged men participating in a programme of brisk walking.

Near infra-red interactance

Principle

The measurement of near infra-red interactance involves irradiating a sample with radiation in the near infra-red spectrum. The interactance is the proportion of energy transmitted that returns to the detector. The interactance depends on the distance through which the radiation travels, as well as the physical properties of the substance irradiated, obeying the Beer-Lambert law:

$$\text{Interactance (I)} = I / I_0 = 10^{-kcL}$$

or

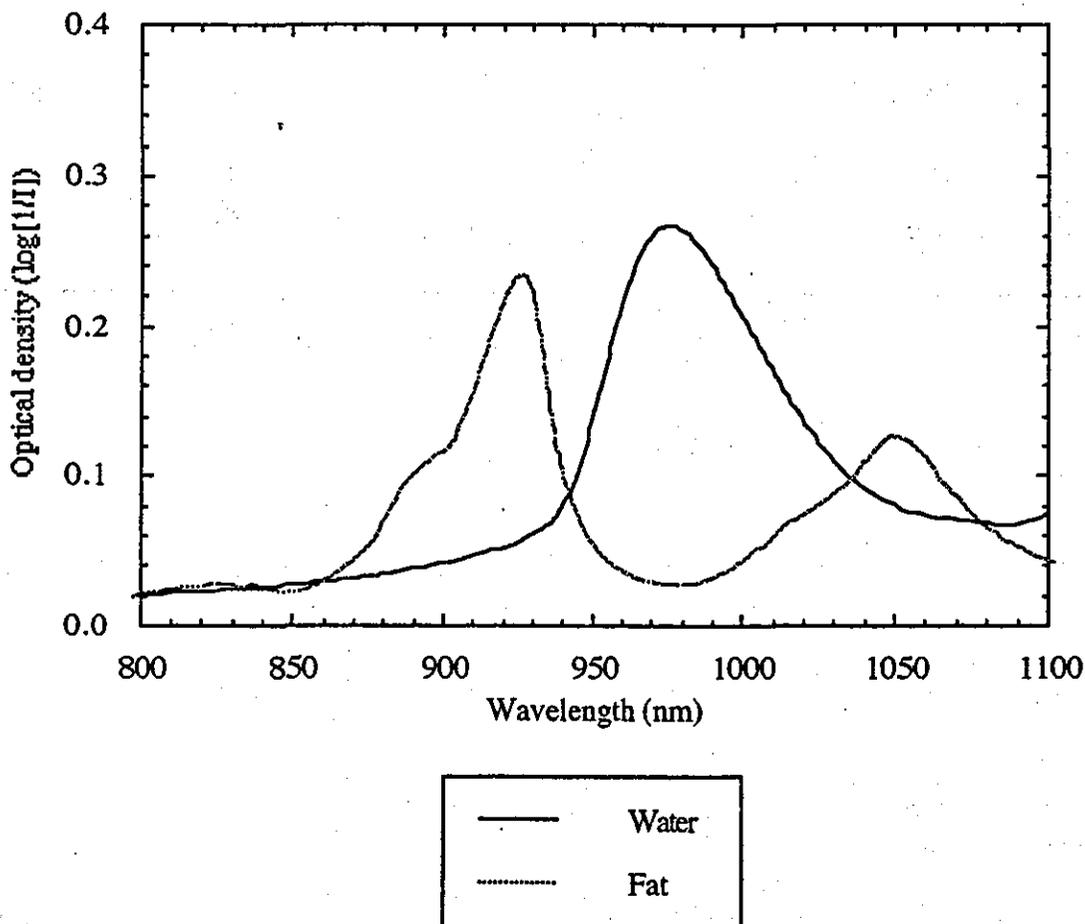
$$\log_{10} (1/I) = kcL$$

where I is the intensity of radiation emerging, I_0 is the intensity transmitted, k is the molar absorption coefficient, c is the molar concentration, and L is the path length through which the radiation travels. $\log_{10}(I_0/I)$ is referred to as the optical density.

For any one homogeneous substance, the concentration and absorption coefficient will be constant, so the interactance will be proportional to the distance the radiation travels. The absorption coefficient is dependent not only upon the nature of the substance, but also upon the wavelength of transmitted radiation. Each substance will have its own absorption spectrum, with characteristic peaks. For example, at 930 nm, there is a peak for fat but not for water, and at 970 nm a peak for water and a trough for fat (Figure 1.1).

Figure 1.1

Diagram of near infra red spectra of water and fat



Adapted from Conway et al. (1984)

Because the near infra-red absorption spectrum is dependent upon the sample composition, near infra-red spectroscopy has been used in analysis of foodstuffs, for

example in measurement of the oil, water and moisture content of grains. Near infra-red estimation of fat and water content of pork and beef have been found to agree well with results from extraction and drying (Norris, 1983; 1984).

Differences in interactance of fat and water can be exploited in measurement of human body composition: adipose tissue being high in fat, and muscle having high water content. The relative absorptions at two wavelengths, at one of which there is greater absorption for fat, and the other greater absorption for muscle, can be used to obtain information about relative proportions of fat and muscle. The radiation used is non-ionising and of low intensity, so measurement is harmless.

Evaluation

Conway et al. (1984) obtained infrared scans for 121 subjects (male and female), using a computerised spectrophotometer, scanning from 700 to 1100 nm. Measurements were made at five sites: biceps, triceps, subscapular, suprailiac, and thigh, and the interactance from all sites averaged. Percentage body fat was also measured by deuterium oxide dilution, skinfold thicknesses (using the equations of Durnin and Womersley, 1974), and for 68 subjects hydrostatic weighing. A prediction equation was developed from the relationship between $\log 1/I$ at two wavelengths in the near infra-red spectrum, and total body fat from deuterium oxide dilution in 53 subjects, and tested on the remaining 68 subjects. Correlation coefficients between near infra-red estimates of body fat and other methods were high: from 0.82 to 0.94, with standard error of estimate ranging from 3.0 to 4.4 percent body fat.

Following these results a commercial instrument was developed: Futrex 5000 (Futrex Inc). The manufacturers have calibrated each machine against their own hydrostatic weighing measurements, to allow estimation of % body fat from optical density readings, and weight, height, sex and activity level.

The estimation of % fat by Futrex 5000 has been assessed by Elia, Parkinson and Diaz (1990) by comparison with hydrostatic weighing in 34 adults. Standard errors of estimate were 3.1 % fat for men and 4.3 for women. These values were slightly lower in men but higher in women than those for estimation of body fat by skinfold thickness (3.7 and 2.5 % fat respectively). However high correlations could be explained by the wide range of body composition of the subjects: methods based on body mass index or weight and height were observed also to have similar standard errors of estimate. Futrex was also found to underestimate % fat, to an increasing amount with increased fatness.

General considerations

Although the technique has not been widely validated, it provides a rapid assessment of body composition, is portable, does not require extensive observer training, and measurement is socially acceptable. So after more validation it may find a place in large-scale surveys, or field studies.

The technique is evaluated in this thesis in four populations: middle-aged men and female athletes in chapter 3, and younger men and women in chapter 4. To further determine the sources of error of the technique, the relationship between reactance and subcutaneous adipose tissue thickness and muscle thickness is examined in chapter 5, by comparison with ultrasound.

Bioelectrical impedance analysis

Principle

The assessment of body composition by bioelectrical impedance depends on the different impedances to an electrical current of the fat-free mass and the fat mass. The fat free mass contains most of the electrolytes and body fluids, which are involved in conduction, so the impedance of the body can be used to estimate the fat free mass.

The impedance of a conductor is related to its length and cross sectional area:

$$Z = rL / A$$

where Z is impedance (ohms), r is volume resistivity in ohm cm, L is the length of the conductor (cm), and A its area (cm²). Multiplying by L/L :

$$Z = rL^2 / AL$$

where AL will be equal to the volume V :

$$Z = rL^2 / V, \text{ or } V = rL^2 / Z$$

This relationship between impedance and volume can be applied to the body, by assuming that the body consists of a series of connecting cylinders.

Procedure

Electrodes are placed on the dorsal surfaces of the hands and feet, proximal to metacarpal phalangeal and metatarsal phalangeal joints, and also between the distal prominences of the radius and ulna and between medial and lateral malleoli at the ankle. Four electrodes are used to minimise the contact impedance. A current of 800 μ A at 50

kHz is applied at the distal electrodes, and the voltage drop at the proximal electrodes measured. Regression equations have been developed relating total body water (measured by D₂O dilution) to impedance measurements (usually as ht^2/Z). As the total body water forms a relatively constant proportion of fat-free mass, fat free mass can be calculated from total body water, and fat mass calculated from the difference between body weight and fat free mass.

Evaluation

Nyboer first applied electrical impedance to biological functions in 1959, and Thomasset in 1962 first used bioelectrical impedance as an index of total body water. Hoffer (1969) observed a correlation between total body water and impedance of 0.84 and 0.91 in normal subjects and patients. The correlation was improved to 0.92 and 0.93 between total body water and Ht^2/Z . Measurements of bioelectrical impedance were not used in evaluation of human body composition until 1985 when Nyboer et al. developed a tetrapolar electrode method.

Several instruments have now been manufactured to measure body impedance. An even larger number of equations have been developed to estimate body composition from impedance. These have been generated by multiple regression in various populations, by comparison with techniques such as deuterium oxide dilution or hydrostatic weighing. When compared to densitometry, the standard errors of estimate of percent body fat calculated using various instruments and equations have been reported to vary widely, between 2.7 (Lukaski et al., 1986) and 6.1 (Segal et al., 1985).

The use of BIA to assess changes in body composition has been evaluated by Deurenberg et al. (1989) who found that they were unable to detect changes in body water and glycogen content with BIA. However Kushner et al. (1990) found BIA to be a better predictor of changes in body water and fat free mass compared to deuterium oxide dilution than anthropometry. Gray et al. (1988) found ht^2/Z to be well correlated with changes in body water during fasting.

Sources of error

In estimating body composition from impedance, it is assumed that the body is cylindrical. As shown in the equations above, the impedance will be proportional to the length of the cylinder, and inversely proportional to its cross sectional area. So the arm, which is relatively long and thin, will have a much greater impedance than the trunk, which is shorter and broader.

As the technique is so highly dependent upon the composition of the limbs, inter-individual differences in distribution of body components are extremely likely to

contribute to the error of the technique. Baumgartner et al. (1986; 1989) have addressed this limitation by measuring the impedance of the body segments separately.

As the impedance of the body is dependent upon the body water content, factors which affect hydration state are likely to affect impedance. The magnitude of change in impedance was examined by Khaled et al. (1988) who found ingestion of 1.2 - 1.8 l water to increase impedance by approximately 15%, and 1.5 to 2 hours strenuous jogging (causing a decrease in weight of 1-2 kg) to decrease impedance by about 13%. Similarly, Deurenberg et al. (1988) found that consumption of beef (but not normal) tea significantly lowered impedance, as did strenuous (but not moderate) exercise. For this reason, factors which may affect hydration state are generally controlled, with subjects restricted from consuming food, drinks, and exercising before measurement. The technique may therefore be unsuitable in some disease states where body water content is substantially altered, such as oedema, kidney disease, or illnesses causing dehydration.

Other factors which may affect impedance are positioning of electrodes: displacement of electrodes by 1 cm may produce a change in impedance of 2.1 % body fat (Elsen et al., 1986), and extremes of temperature: an increase in skin temperature has been found to decrease impedance (Caton et al., 1988). Significant differences between different instruments have also been reported (Deurenberg et al., 1989).

General considerations

Bioelectrical impedance is portable, fast and relatively cheap. It does not require extensive observer training, and is, according to some reports, extremely accurate. However the review above highlights the importance of controlling factors such as electrode placement, ambient temperature, and hydration state. This may diminish the usefulness of the technique in field studies, where ambient temperature may be impossible to control. Controlling hydration state requires subject cooperation, and may not be possible in disease states. It may also make the technique less suitable for athletes and heavy manual workers.

To attempt to explain the reason for the wide variation in the reported accuracy of the technique, a comparison of some of the many different equations for estimating body composition from impedance is carried out in chapter 4. The effect of exercise preceding measurement is also examined.

Weight and height

General Principles

Measurements of weight and height do not actually provide information on body composition. However, they are included here as they have long been applied to provide information on whether populations and individuals are overweight compared to reference standards. They have also been applied in most epidemiological studies, because of the ease of measurement.

Several general principles have been applied in the assessment of body weight. The most straightforward is the comparison of weights and heights of a malnourished population to detect the prevalence of wasting (low weight for age) or stunting (low height for age).

The second is the comparison of body weight with tables of average or "ideal" weight for height. The relative weight (or percentage of ideal weight for height) can then be calculated. Because of the large influence of bone mass on total weight, these tables often provide data for small, medium and large frame sizes (Metropolitan Life Insurance Company 1983).

The third method is the comparison of various weight/height ratios with "ideal" values. The most commonly used is the body mass index (BMI), which was developed by Quetelet (1836). This is calculated as

$$\text{BMI} = \frac{\text{weight (kg)}}{\text{height (m)}^2}$$

Various other ratios have also been developed using different powers of weight and height (e. g. Abdel Malek et al., 1985).

Sources of error

The major drawback of using weight and height is that they do not provide any information on the composition of the body. An individual with a large amount of lean tissue may be overweight for their height despite having a low fat content. Individuals with the same weight and height may therefore have differing percentages of body fat.

The identification of an ideal may also be problematic. The most commonly used standard is the level at which lowest mortality is observed. However this level seems to increase with age (Andres, 1985). Low body weight may be associated with heavy cigarette consumption (Howell, 1971). This may increase the incidence of smoking-related mortality at lower body weights, possibly increasing the ideal. Secular trends

may also influence the ideal, for example an increased level of exercise participation has increased the lean mass, hence increasing the ideal (Garrison et al., 1983).

The technical errors associated with measurement of weight and height are generally considerably smaller than those for measurement of body composition (Lohman et al., 1988). However failure to standardise for clothing worn when measuring body weight, or the height of shoes when measuring height, may cause error.

Biological variation may occur in both weight and height. Variations in hydration and abdominal contents may cause short term fluctuations in weight (Durmin 1961). Conditions such as kyphosis or narrowing of inter-vertebral disks may affect height.

General considerations

Measurement of weight and height can be performed accurately, quickly, cheaply, and with great ease. Measurement is socially acceptable, and minimal observer training is required. An extremely large body of information exists for comparison. For these reasons weight and height are extremely suitable for use in large-scale studies.

However, if information on body composition is required, for example when examining the relationship between obesity and disease, additional measurements would be desirable, because of the limitations discussed above.

Measurement of subcutaneous adipose tissue and muscle thickness

Near infra-red interactance provides an estimate of body composition from the adipose tissue and muscle composition at a particular site. The error of the technique will depend in part on how well interactance measures the composition at this site. To assess this, near infra-red interactance measurements are compared with ultrasound measurements of subcutaneous adipose tissue thickness (SCATT) and muscle thickness in chapter 5.

The use of skinfold thicknesses in the assessment of body composition and fat distribution also depends on the agreement of skinfold thickness with SCATT. To assess in more detail the relative accuracy of the two techniques, skinfold thicknesses are also compared with ultrasound measurements of SCATT. This will allow determination of the relative accuracy of near infra-red interactance and skinfold thicknesses in measuring SCATT.

The principles and sources of error of estimating of SCATT by ultrasound and skinfold thickness are discussed below. The use of near infra-red interactance in measurement of SCATT has not previously been assessed, so no information is available on the associated errors. The principle and procedure have been described above, so near infra-red interactance is not discussed again here.

Ultrasound

Principle

Ultrasound consists of sound waves of frequencies greater than 20,000 Hz (above the audible range). Frequencies of 2 to 10 MHz are generally used for characterisation of human tissues. The ultrasonic waves are produced by applying an alternating field to a piezo-electric crystal, which is housed in a transducer. In A-mode ultrasound, the transducer is applied to the skin, coupled with gel. Some of the ultrasonic waves are reflected from interfaces between tissues, such as the fascia between subcutaneous adipose tissue and muscle. The reflected waves are returned to the transducer, where they strike the piezo-electric crystal, and are converted to a voltage which can be amplified and displayed on a cathode ray oscilloscope. Firing constant pulses of ultrasound more than twenty times per second will produce a constant trace on the display.

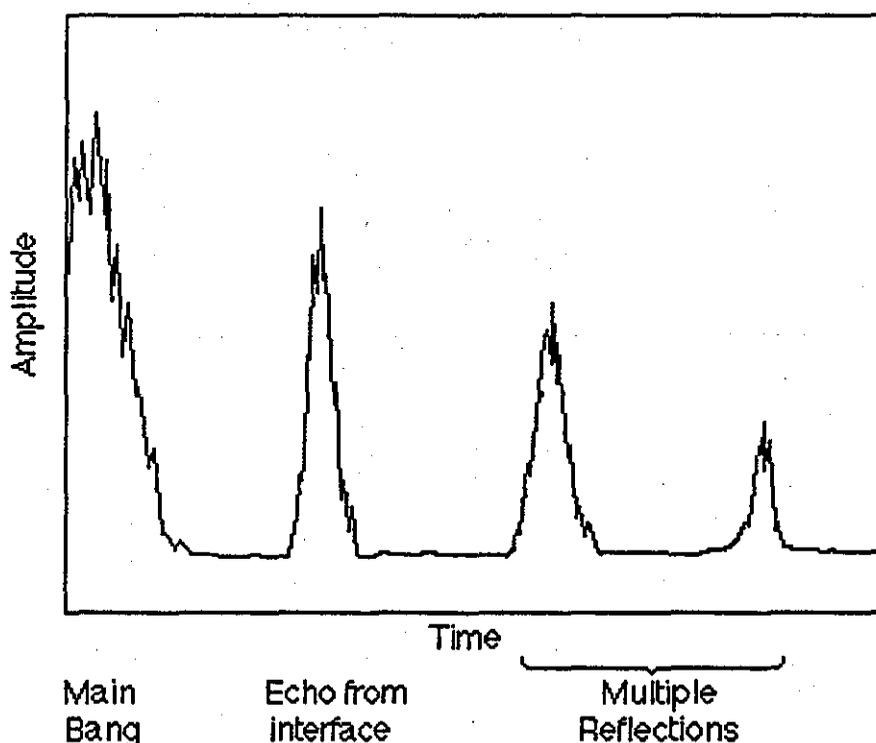
By assuming the velocity of ultrasound in soft tissue to be 1540 m/s (a compromise between the velocity in adipose tissue of 1450 m/s and that in muscle of 1580 m/s), the time taken for the echo to return can be converted to the distance the wave has travelled, which will be twice the distance of the interface from the transducer. A schematic representation of the oscilloscope display from a single reflective surface is

shown in Figure 1.2. The display shows the distance of the interface from which an ultrasonic echo has been reflected, against the amplitude of the echo. The initial peak, or "main bang" is produced by the piezo-electric material. The next peak corresponds to the reflection from an interface. The following peaks are produced by multiple reflections from the same interface. The amplitude of these peaks will decrease, as the amplitude of the echo decreases exponentially with the distance the wave has travelled.

Ultrasound has been used in the assessment of SCATT and muscle thickness by several authors, including Alsmeyer et al. (1963); Haymes et al. (1976) and Borkan et al. (1982).

Figure 1.2

Diagram of oscilloscope display during ultrasound measurement of a single interface



Sources of error

Because of multiple reflections, difficulties may arise in misinterpretation of a multiple reflection of the dermis/adipose tissue interface (which is usually lost within the main bang) as the subcutaneous adipose tissue/muscle interface. Another cause of misinterpretation of the trace is the presence of layers of fascia within the subcutaneous adipose tissue, which has been reported at some sites (Haymes et al. 1976). Some loss in amplitude may occur if the transducer is not perpendicular to the reflecting interface, causing additional difficulties in interpretation.

Skinfold thickness

Principle

Skinfold thicknesses provide a rapid assessment of subcutaneous fat thickness at particular sites. The measurement provided is that of a compressed, double fold of subcutaneous adipose tissue. This can be adjusted if the compressability of the skinfold is known, to calculate the SCATT.

The compressability of the skinfold can be quantified by comparison with other methods, such as ultrasound, needle puncture or roentgegrams, which provide an uncompressed value for the subcutaneous adipose tissue thickness. The compressability is then expressed as a percentage of the uncompressed value:

$$\% \text{ compressability} = \frac{\text{Uncompressed thickness} - (\text{skinfold thickness} / 2)}{\text{Uncompressed thickness}} \times 100$$

Sources of error

Variation in compressability of skinfolds is a potential source of error. Skinfold compressability has been reported to vary according to age (Brozek and Kinzey 1960), sex and site (Lee and Ng 1965, Himes et al. 1979). Moreover significant individual differences in skinfold thickness have been observed by Himes et al. (1979). Values have been reported to range between 10 to 40 %. These differences in skinfold compressability may have implications for the use of skinfold thicknesses in assessment of distribution of adipose tissue.

The technical error and inter-observer differences in measurement of skinfold thicknesses were discussed above. These will also contribute to the error of assessment of SCATT from skinfold thicknesses.

Assessment of fat distribution

The distribution of body fat, as well as the quantity, has important health implications as discussed below. A high proportion of intra-abdominal fat or central fat has been linked with a number of diseases (Bjorntorp, 1988). These findings highlight the need for measurement of fat distribution (particularly characterisation of intra-abdominal fat) as well as body composition.

Techniques such as computerised tomography or radiography allow assessment of internal as well as subcutaneous fat, and so may be extremely useful in the characterisation of fat distribution. However these techniques are not widely available, are expensive, or involve exposing the subject to ionising radiation, so their use in large studies is limited. Computed tomography has revealed an association between central and intra-abdominal fatness (Ashwell et al. 1985), validating the use of anthropometric measures of central fatness for description of fat distribution. The anthropometric techniques used in this thesis for the measurement of fat distribution are discussed below.

Circumferences and circumference ratios

General Principles

Circumference measurements provide information on body size at a particular level. However they will be largely dependent upon body build and frame size. By calculating the ratio of two circumferences, the influence of varying body builds and frame sizes can, to some extent, be accounted for. The waist circumference will vary depending upon the accumulation of intra-abdominal fat. So by calculating a ratio of waist to hip or thigh circumferences, a measure of intra-abdominal fatness can be obtained.

The waist hip ratio and waist thigh ratio have been widely applied. They have been found to be highly associated with incidence of disease, independently from total fatness (Lapidus et al. 1984; Larsson et al. 1984).

Sources of Error

The technical error of measuring circumferences is reasonably low. The intra-observer variability of about 2% (Bray et al. 1978) is considerably lower than that for skinfold thickness.

Large differences have occurred in the level at which circumferences are measured. This is particularly true for waist circumference. Both minimal (Kissebah et al. 1982) and maximal (Lanska et al. 1985) circumferences have been measured. Sites have been identified relative to surface landmarks such as the level of the umbilicus (Larsson et al. 1984) and relative to skeletal landmarks, such as midway between costal

margin and iliac crest (Lapidus et al. 1984; Jones et al. 1986). It has been suggested that skeletal landmarks should be preferred (Shephard, 1990) although they may be difficult to locate in obese subjects. Sometimes a minimal or maximal circumference may not exist, and the level of the umbilicus may vary, so skeletal landmarks provide greater standardisability. However, for the hip circumference, the maximal circumference is generally the easiest to locate.

Circumference ratios tend to be fatness dependent (Garn et al. 1982; 1988). Some standardisation for body fatness may therefore be necessary to examine independent effects of fatness and fat distribution.

General considerations

The measurement of body circumferences is cheap, fast, and does not require extensive observer training. Circumference ratios have been strongly implicated as risk factors for disease.

For these reasons the waist hip ratio is measured in chapter 6, to provide a measure of changes in fat distribution during an exercise program, and to determine whether these changes in fat distribution are related to changes in cardiovascular risk factors.

Skinfold thicknesses and skinfold ratios

General Principles

Skinfold thicknesses allow assessment of the quantity of subcutaneous adipose tissue at particular sites. As the amount of central fat has been associated with the amount of intra-abdominal fat, either central skinfold thicknesses, or ratios of central to peripheral skinfold thicknesses will be useful indicators of fat distribution.

Several skinfold thicknesses and skinfold ratios have been applied to characterise fat distribution. Donahue et al. (1986) used subscapular skinfold thickness as an indicator of central fatness. The ratio of subscapular to thigh skinfolds has been applied by Blair et al. (1984).

Mueller and Stallones (1981) found that the best indication of central fatness was given by the ratio of subscapular to thigh skinfolds. Kaplowitz et al. (1987) also suggested the use of the ratio of subscapular plus triceps to triceps skinfolds.

Another approach has been a factor analysis of skinfolds at several sites (for example Mueller and Reid 1979), enabling a summary of the distribution of subcutaneous fat in the body. The first factor identified is a general fatness factor. Subsequent factors may describe central versus peripheral or upper versus lower distributions of fat.

Sources of error

As discussed above, considerable inter-observer variation in measurement of skinfold thicknesses has been reported. Differences in skinfold compressability (also discussed above) may contribute to inter-individual or age or sex related differences in the relation of skinfolds or skinfold ratios to fat distribution. Skinfold ratios, like circumference ratios, may be fatness dependent (Garn et al. 1982).

General Considerations

Skinfold thicknesses are quick and cheap to measure. However they are subject to inter-observer variation, and require considerable observer training. In some cultures, measurement of some sites may not be socially acceptable. They are however extremely appropriate for field or large-scale studies.

The skinfold ratios above are used to describe fat distribution in chapter 6. Skinfold thicknesses are also measured at several other sites, and a factor analysis performed, to provide a more complete description of fat distribution. These measures of fat distribution are used to examine the influence of a programme of brisk walking on fat distribution, and the associations between fat distribution and other cardiovascular risk factors during this programme.

Association of risk of cardiovascular disease with body composition, fat distribution, dietary intakes, physical activity and plasma lipids and lipoproteins

Risk factors for cardiovascular disease

The incidence of coronary heart disease (CHD) in Britain has not decreased greatly in recent years, despite the identification of a number of factors which are associated with mortality rates (National Forum for Coronary Heart Disease Prevention, 1988). Levels of these risk factors have also remained constant (Mann et al. 1988). In the United States and other countries, a decrease in CHD mortality has been achieved, and this decrease is associated with changes in risk factors (Stamler 1985).

A risk factor has been defined as: "a factor that is statistically associated with an increased risk of developing a defined disease in a prospective study" (Bjorntorp, 1988). However, the association of a risk factor with a disease does not necessarily imply a cause-effect relationship.

Some risk factors for CHD can not be altered. These include sex (with higher CHD rates in men than women), family history (Perkins, 1986) and age. However several factors can be influenced. These include: smoking; serum cholesterol and lipoprotein levels; diet; blood pressure; body fatness and fat distribution; level of physical activity and type A behaviour.

In chapters 6 and 7 a study of the influence of a year-long programme of brisk walking on cardiovascular risk factors is described. The emphasis is on the association of body composition, fat distribution, and dietary intakes with other risk factors. The evidence relating to the identification of the risk factors considered in this study, and the inter-relationships of these risk factors, are discussed below.

Plasma lipids and lipoproteins

A number of studies have identified plasma lipids and lipoproteins (particularly plasma cholesterol and lipoprotein cholesterol subfractions) as being strongly associated with the risk of developing coronary heart disease. The evidence for these associations is discussed below. Following these findings, changes in plasma lipid levels have been used in longitudinal studies, to examine the effect of changing other risk factors (such as body composition, diet, or exercise level) upon risk of CHD. Plasma lipid levels are used in chapters 6 and 7, to allow examination of the effects of a year-long programme of brisk walking in healthy, asymptomatic middle-aged men. They are also related to body composition, fat distribution and dietary intakes.

Total Cholesterol

Differences between populations in median total cholesterol (TC) are highly correlated with national CHD death rates (Keys 1980, Rose 1982) and extent of atherosclerosis (Scrimshaw and Guzman 1968). Many investigators have also observed association between TC levels and risk of CHD in individuals within populations (Rickert et al. 1968; Pooling Project Research Group 1978; Keys 1980; Rose and Shipley 1980; Ducitimiere et al. 1980; Holme et al. 1981; Italian National Research Council 1982; Goldbourt et al. 1985; Stamler et al. 1986). However this association has not been observed in populations with low mean TC levels (Goldbourt 1987), and the lowest range of TC is not always associated with lowest cardiovascular mortality (Goldbourt et al. 1985).

TC has also been related to degree of atherosclerosis at autopsy in men (Rickert et al. 1968; Rhoads et al. 1978; Holme et al. 1981; Sorlie et al. 1981; Reed et al. 1987). In the Framingham Study, extent of atherosclerosis was correlated with TC measured 1, 5 and 9 years before death in men, whilst in women only TC measured 9 years before death correlated significantly (Feinleib et al. 1979).

Total cholesterol thus appears to be a strong predictor of both incidence and extent of atherosclerotic disease, although the relationship may be weaker at lower levels of TC.

Lipoprotein-cholesterol subfractions and triglycerides

Lipoproteins consist of variable proportions of lipids and proteins, and are responsible for the transport of triglycerides and cholesterol. They are classified according to their density. High density lipoproteins (HDL) contain approximately equal amounts of protein and lipid. Low density lipoproteins (LDL) contain less protein than HDL and carry most of the plasma cholesterol. Very low density lipoproteins (VLDL) contain an even smaller proportion of protein, and consist largely of triglyceride.

LDL-C has been positively associated with incidence of cardiovascular disease (Medalie et al. 1973; Gordon et al. 1977; Watkins et al. 1986). Reduction in LDL-C has been associated with reduction of risk of cardiovascular disease (Lipid Research Clinics Program 1984).

HDL-C has been inversely associated with the incidence of CHD (Medalie et al. 1973; Gordon et al. 1977; Miller et al. 1977; Enger et al. 1979; Goldbourt et al. 1985; Castelli et al. 1986; Watkins et al. 1986). Increases in HDL-C have been associated with a decrease in risk of CHD (Castelli et al. 1986; Gordon et al. 1986). HDL-C has also been found to be negatively correlated with the degree of atherosclerosis at autopsy (Holme et al. 1981). However, other studies have failed to find an association between HDL-C and CHD death rates (Keys et al. 1984; Pocock et al. 1986; Levy and Klimov

1987). The failure to find an association in these studies is perhaps due to the confounding influence of LDL-C and TC. Castelli et al. (1983) found the ratio of TC to HDL-C to be strongly associated with risk of CHD (as was the ratio of LDL-C to HDL-C in women only).

Fasting triglyceride (Tg) and very low density lipoprotein (VLDL) levels have been found to be positively associated with increased incidence of cardiovascular disease, although most studies have found this association not to be significant after adjustment for other risk factors (Hulley et al. 1980; Carlson and Bottiger 1981; Aberg et al. 1985; Wallace and Anderson 1987). Aberg et al. (1985) and Gordon et al. (1977 and 1981) have reported weak independent associations between Tg and VLDL levels and arterial disease in women. Tg and VLDL concentrations thus do not seem to have the independent predictive power of HDL-C and LDL-C, although they do seem to be associated with other risk factors: an interactive effect of Tg and TC was suggested by Cambien et al. (1986).

Apolipoproteins

Apolipoproteins (or apoproteins) are the protein constituents of lipoproteins. Apolipoprotein A (ApoA) occurs mostly in HDL, whilst Apo B occurs largely in LDL, but also comprises about 35% of VLDL protein. Apo C and apo E are major constituents of VLDL, but small amounts are also found in HDL, as is Apo D.

Apo B levels have been observed to be higher, and Apo A lower in subjects with cardiovascular disease (Onitiri and Jover 1980; Riesin et al. 1980; Sniderman et al. 1980; Fager et al. 1980, 1981; Whayne et al. 1981; Franceschini et al. 1982; DeBacker et al. 1982; Maciejko et al. 1983; Pilger et al. 1983; Naito 1985; Wallace and Anderson 1987). However, several of these investigators found lipoproteins to be better predictors of disease than apolipoproteins.

Lipoprotein (a)

A relationship between serum lp(a) levels and coronary disease has been reported (Berg et al. 1974; Dahlén et al. 1975; 1976).

Body composition

Overweight has been associated with increased cardiovascular mortality rates (Ashley and Kannel 1974; Rabkin et al. 1977; Lew and Garfinkel 1979; Hubert et al. 1983, Donahue et al. 1987). Waaler (1984) reported a J-shaped relationship between weight and BMI and mortality, with lowest mortality at a BMI of 23 kg/m². He estimated that at optimal BMI, mortality would be reduced by 15%. However mortality

was found to be inversely associated with height, so differences in height may explain some of this relationship.

Manson et al. (1987) reviewed the association of overweight with risk of death. The association was strongest in large studies, and those with follow up of more than 15 years. They commented that some studies failed to adjust for other risk factors. Some investigators have not found obesity or overweight to be independent predictors after adjustment for other factors (Keys et al., 1972; Pooling Project Research Group, 1978; Solberg and Strong, 1983).

Another factor that may decrease the strength of the association between body composition and CHD is that most studies have measured weight or BMI, rather than fat content. This point is illustrated by the study of Segal et al. (1987), who found increased levels of cardiovascular risk factors in obese men, but not in overweight lean men.

Examination of the effects of changes in body weight has yielded similarly inconsistent results. A recent study showed that men whose weight increased more than 15 % had an increased incidence of CHD, even after adjustment for other risk factors (Wannamethee and Shaper, 1990). Abraham et al. (1980); Noppa (1980) and Shapiro et al. (1969) also found an association between weight gain and cardiovascular disease, although Barrett-Connor (1985); Heyden et al. (1971) and Hsu et al. (1977) found no such association. Moderate increases in weight over many years seem beneficial (Avons et al., 1983; Rhoads and Kagan, 1983), and the BMI associated with lowest mortality seems to increase with age (Andres, 1985).

Body composition does seem to be related to other factors associated with cardiovascular disease, such as blood pressure (Kannel et al., 1967); HDL-C (Garrison et al., 1980; Rhoads et al., 1976); LDL-C transport (Miettinen, 1971; Nestel et al., 1973) and VLDL production (Bennion and Grundy, 1978). Ashley and Kannel (1974), Borkan et al. (1986) and Noppa (1980) reported weight gain to be associated with increased blood pressure and blood lipid concentrations.

So it seems that even if body composition does not have a direct effect on incidence of CHD, it is associated with other risk factors. So body composition still has an important place in the etiology of CHD.

Fat distribution

The distribution of body fat was first observed to be associated with chronic disease in the 1950s. Vague (1956) reported that an android distribution of fat was associated with increased risk of atherosclerosis and diabetes. Accumulation of abdominal fat (as evidenced by high ratio of waist to hip circumferences) was found to be a predictor of CHD in Swedish population studies (Larsson et al., 1984; Lapidus et al., 1984). This association was independent of other risk factors, including obesity.

Other investigators have supported the association of centralized fatness (also using indices such as subscapular skinfold thickness, ratio of abdominal to suprailiac or subscapular to thigh skinfolds) with CHD (Stokes et al., 1985; Donahue et al., 1987; Bouchard, 1988), serum lipid levels (Albrink and Meigs, 1964; Despres et al., 1985; Kissebah et al., 1985) and hypertension (Kalkhoff et al., 1983; Blair et al., 1983; Williams et al., 1987).

The indices used to characterise fat distribution above have certain limitations. Firstly, they are fatness dependent (Garn et al., 1982, 1987; Mueller et al., 1986). Secondly, they do not directly measure intra-abdominal fatness. However, comparison with computed tomography has confirmed the relationship of anthropometric indices with quantity of intra-abdominal fat (Seidell et al., 1987).

Diet

Energy Intake

A lower energy intake has been observed in subjects who develop cardiovascular disease (Gordon et al., 1981; Willett and Stampfer, 1986). Lapidus et al. (1986) also observed energy intake to be independently inversely associated with death from all causes and cardiovascular risk factors. Increased levels of physical activity could contribute to increased energy intakes, and hence have a role in reduction of CHD incidence.

A reduction in food intake has been associated with decrease in triglycerides and initial reduction in total cholesterol (Henry et al., 1986).

Dietary fats and lipids

Dietary fatty acids are classified as saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs) or polyunsaturated fatty acids (PUFAs) according to the number of double bonds in the alkyl chain. SFAs contain a straight alkyl chain, i.e. no double bond. Short and medium chain SFAs are found in milk, palm fat and coconut oil, whilst longer chain SFAs are found predominantly in meat, butter fat, and some vegetable oils. MUFAs contain one double bond, whilst PUFAs contain more than one double bond.

The first indications that the prevalence of cardiovascular disease was associated with intakes of fat and cholesterol were observed in population studies between the 1930s and 1950s (reviewed by Keys, 1957; 1963). Since then many further studies have been conducted.

In the Seven Countries Study, high correlations between SFA intake and coronary heart disease incidence and mortality were observed (Keys, 1980). PUFA

intake was not associated with cardiovascular disease, but there was little variation between the populations studied in PUFA intake.

Results from intra-population studies have been less consistent. The percentage of energy obtained from SFAs has been positively associated with cardiovascular disease in some studies (Garcia-Palmieri et al., 1980; McGee et al., 1984; Kushi et al., 1985). However this association was not significant in other studies (Medalie et al., 1973; Garcia Palmieri et al., 1980; Shekelle et al., 1981; Kromhaut and de Lezenne Coulander, 1984). The percentage of energy from PUFAs was negatively associated with risk of coronary disease in the Western Electric Study (Shekelle et al., 1981), but not in the other studies above. Similarly, the P:S ratio (ratio of PUFA to SFA) was found to be inversely associated with incidence of coronary disease by Morris et al. (1977) but not by Medalie et al. (1973). The inconclusive results of these studies may be due to limited variation in intakes, and difficulties in classification of habitual fat intakes and disease symptoms. The effect of family history on CHD may also be a confounding factor.

Mean population serum cholesterol concentrations have also been associated with percentage of energy from fats and SFAs. However, as observed with incidence of CHD, studies of individuals within populations have often failed to show consistent associations, perhaps due to large variations in intrinsic TC levels between individuals (Stallones et al., 1983).

The difficulty of inter-individual variation in incidence of CHD and plasma lipid levels has been overcome in longitudinal studies examining the effect of changing habitual diet. The effect of increasing PUFA intake whilst keeping total fat intake constant was examined by Dayton et al. (1968). The incidence of fatal atherosclerosis was significantly reduced in the experimental group. A diet low in SFAs and cholesterol was associated with significantly lower mean TC and incidence of coronary heart disease than normal mental hospital diet (Turpenien et al. 1979). PUFA intake was found to influence TC levels, with the change occurring principally in the LDL-C subfraction.

Keys et al. (1965) and Hegsted (1986) found the observed change in TC to be correlated with change in SFA and cholesterol intakes, and inversely correlated with change in PUFA intake. They produced regression equations to predict the change in TC that would be expected based on the change in these intakes. The inclusion of MUFA intake did not add to the predictive power of these equations. Many studies have supported these findings. The subject has been reviewed by McNamara (1987) and Grundy (1987). Considerable individual differences have been observed, particularly in response to changes in cholesterol intake (Keys 1965b; Grundy and Vega 1988). The

effect of dietary cholesterol upon plasma cholesterol levels is discussed separately below.

Whilst many earlier studies have considered MUFAs to be neutral as regards risk of cardiovascular disease, Mensink and Katan (1987) suggested a beneficial role for MUFAs. A diet rich in complex carbohydrates was found to decrease both TC and HDL-C, whilst a diet rich in olive oil also decreased TC, but did not influence HDL levels. This finding is consistent with the association of MUFA intake with increased catabolism of LDL (International Collaborative Study Group, 1986).

Dietary cholesterol

Mean cholesterol intake has been associated with population CHD mortality rates (Stamler et al., 1972; Armstrong et al., 1975) and within population mortality rates after adjustment for other risk factors (Shekelle et al., 1981; Kromhaut and de Lezenne Coulander, 1984; Mc Gee et al., 1985; Kushi et al., 1985).

The association between dietary and plasma cholesterol within populations has often been concealed, because of the great individual variation in TC (Jacobs et al., 1979). However increases in dietary cholesterol are associated with increases in TC (Beveridge et al., 1960; Connor et al., 1961; Mattson et al., 1972). This increase in TC is due mostly to an increase in the LDL-C subfraction (Gordon et al., 1982) and Apo B (Appelbaum-Bowden et al., 1979). The effect of decreasing cholesterol intake has been less consistent (Glueck et al., 1986). Individual differences in response have also been observed (Katan et al., 1986), with some subjects consistently responding to a greater extent than others.

Alcohol Intake

Alcohol intake has been associated with plasma HDL (Gordon et al., 1981) and Apo A (Camargo et al., 1985) in cross-sectional studies. Increasing alcohol intake has been found to increase HDL (Barrett-Connor and Suarez, 1982). However this effect was not observed in runners, whose HDL levels were already high (Hartung et al., 1983).

The effect of alcohol consumption upon the incidence of CHD is more controversial. Both abstention and heavy consumption of alcohol have been associated with increased incidence of CHD, whilst moderate alcohol consumption had a protective effect (Moore and Pearson, 1986). However Shaper et al. (1987) found no significant association between alcohol intake and ischaemic heart disease after adjusting for other risk factors. Alcohol intake therefore does not seem to have as strong an association with CHD as other dietary components.

Blood Pressure

Blood pressure is a well established risk factor for CHD (Pooling Project Research Group, 1978; Keys, 1980; Gordon and Kannel, 1972; Stamler et al., 1980), although it is a less powerful predictor of CHD than TC (Winkelstein et al., 1975). It has also been associated with several other risk factors: including body composition and fat distribution (both discussed above) and diet.

The effect of sodium intake on blood pressure has been extensively studied. Populations with low salt intakes appear to have low prevalence of hypertension (Page, 1979). However studies of the association of salt intake with blood pressure have been inconclusive. There seem to be individual differences in response to salt intake (Bittle et al., 1985), with generally greater response in hypertensives (Weinberger et al., 1986); whites over 40 years and blacks (Luft et al., 1979).

The association of potassium with blood pressure is clearer. Hypertension has been associated with low potassium diets, and inversely associated with high potassium diets (Kromhout et al., 1985). The role of calcium in reducing hypertension has also become of interest in recent years, although conflicting results have often been obtained.

Cross-sectional studies have shown those who consume low amounts of alcohol to have lower mean blood pressure (Criqui, 1987; Shaper et al., 1987). The effect of alcohol on blood pressure may be due to recent intake rather than a sustained effect (Maheswaran et al., 1987).

Physical Activity

High levels of both occupational and leisure-time physical activity have been reported to be associated with reduced risk of cardiovascular mortality (Paffenbarger and Hale, 1975; Morris et al., 1980; Siscovick et al., 1984; Paffenbarger et al., 1986; Ekelund et al., 1988). However no association between population CHD mortality rates and habitual occupational activity was observed in the Seven Countries Study (Keys, 1970; 1980).

Physical activity has been found to increase HDL-C concentrations by some investigators (Hartung et al., 1980; Huttenen et al., 1979; Wood and Haskell, 1979; Leon et al., 1979) but not others (Nye et al., 1981; Brownell et al., 1982).

Williams et al. (1983) and Wood et al. (1983) found that changes in HDL-C during an exercise programme were inversely correlated with changes in body weight. Wood (1988) investigated the relative effects of caloric restriction and exercise on plasma lipids, and found no significant difference between the increase in HDL-C and decrease in TG of dieters and exercisers. These results suggest that the exercise-induced changes in HDL-C may be mediated by changes in body composition.

Another explanation that has been proposed to account for the inconsistent results of the effects of exercise training studies on HDL-C is that some threshold level of exercise intensity or duration exists, below which no response occurs (Williams et al. 1982; Wood et al. 1983; Hartung et al. 1983).

Summary

The incidence of coronary heart disease has increased markedly in the past 50 years (National Forum for Coronary Heart Disease Prevention, 1988). The incidence of obesity has also increased over this period (Royal College of Physicians Working Party, 1983). Several factors have been identified as increasing risk of CHD, amongst them obesity. However, in order to identify those at risk, or to examine the effect of various preventative strategies, an appropriate method of measurement of body composition is required. This needs to be sufficiently accurate, yet cheap, fast, socially acceptable and practical enough to be used on large numbers of subjects, often at field locations rather than in a laboratory.

In chapters 3 and 4 of this thesis, field methods are evaluated as to the extent to which they fulfil these requirements. The methods examined are measurement of skinfold thicknesses, which have been in use many years, and often evaluated; and two newer techniques: near infra-red interactance and bioelectrical impedance.

Very little evaluation of near infra-red interactance has been carried out, despite the convenience of the technique. Near infra-red interactance measurements are compared with hydrostatic weighing and skinfold thickness in middle-aged men and female athletes in chapter 3, and in younger men and women in chapter 4.

Bioelectrical impedance has been more extensively examined, but evaluations have yielded conflicting results. To attempt to determine the possible reasons for these conflicting results, bioelectrical impedance is compared with other body composition techniques in chapter 4.

Skinfold thickness and near infra-red interactance involve measurement at discrete sites of the body. The composition of the body as a whole is then estimated from the thickness of subcutaneous adipose tissue at these sites. To further evaluate these techniques, their accuracy in assessment of subcutaneous adipose tissue thickness is evaluated. Measurements from the techniques are compared with ultrasound values in chapter 5.

Body composition and anthropometric indicators of fat distribution are applied in chapter 6, in a study of the influence of a year-long programme of brisk walking on levels of cardiovascular risk factors in healthy, previously sedentary middle-aged men. It has been suggested that the response of high-density lipoprotein cholesterol to exercise may be mediated by changes in body composition. Apart from being

cardiovascular risk factors themselves, body composition and fat distribution are also related to several other risk factors, such as blood pressure and blood lipid levels. For this reason the monitoring of body composition and fat distribution in such a study is important, to determine the relative contributions of changes in body composition, fat distribution and activity to changes in levels of other risk factors.

Diet has also been associated with incidence of cardiovascular disease, and with other cardiovascular risk factors. So, it is also important to monitor diet during the walking programme, as changes in the composition of the diet may confound any changes induced by exercise. The association of dietary intakes during the programme with other cardiovascular risk factors is described in chapter 7. Changes in energy intake are also examined with reference to changes in energy expenditure and energy stores, to provide information on the possible influence of the walking programme on energy balance.

Chapter 2

General Methods

The methods used throughout this thesis are described in this chapter. Where the procedures used in a particular chapter depart from the description below, this will be detailed in the relevant chapter.

Exercise testing and blood biochemistry were measured by David Stensel, in the department of Physical Education and Sports Science of Loughborough University. The procedures adopted in these tests are described in more detail elsewhere (Stensel, in preparation), but a brief description is given below.

Informed consent

Before agreeing to participate in any study, subjects were given a written description of all procedures and equipment to be used, and any potential risks involved. They were given an opportunity to discuss the commitments involved. They were made aware that they were free to leave the study at any time they chose, without giving an explanation. On agreeing to participate in the study they were asked to sign a statement of informed consent (Appendix I).

Height

Height was measured on a Holtain suitcase model stadiometer. The subject stood against the stadiometer so that the heels, buttocks and scapulae were in contact with the backboard, with heels together and in contact with the ground. The head was positioned in the Frankfort Plane, with the line between the lower orbits of the eyes and the external auditory meatii perpendicular to the board. A weight was placed on the headboard to flatten the hair, and overcome friction. The subject was instructed to breathe in, and stand as tall as possible. Slight traction was applied under the mastoid processes and occipital bone, to counteract diurnal variation.

Body Weight

Body weight was recorded after voiding, on Seiko electronic scales (CMS Weighing Equipment, London). These read up to 150 kg to the nearest 50 g. Measurement was generally made with subjects wearing swimming costumes, light shorts or T-shirts. In the walking study, where changes in body weight were assessed, subjects wore only swimming costume or light underclothes. However, body weights for use in exercise tests were measured with subjects dressed in the clothes to be worn during the test.

Anthropometry

The measurements below were made to describe the amount and distribution of body fat. The standard error of measurement (Smeas) of anthropometric measurements was calculated by repeating measurements in 15 middle aged men. The Smeas for each variable is given in Table 2.1.

Circumferences

Circumferences were measured using a Fibron tape (CMS Weighing Equipment, London). All measurements were made with the tape tautened so it made firm contact with the skin, but tissue was not compressed. Upper arm, thigh and calf circumferences were measured on the left side of the body, except in one subject whose left calf was reduced following an injury to the calcaneal tendon.

Upper arm circumference

A mark was made at the mid-point between the acromion and olecranon, with the arm flexed at 90 degrees. Measurement was made horizontally at the level of this mark, with the arm relaxed and hanging beside the body.

Chest circumference

This was measured horizontally, at nipple-level. The arms were raised to allow the tape to be passed around the trunk, and lowered once it was in position. The measurement was taken during normal light respiration, in mid-inspiration, with the shoulders relaxed.

Waist circumference

The mid-point between the costal margin and iliac crest was located and marked. Measurement was made horizontally at this level, with abdominal muscles relaxed.

Hip circumference

Hip circumference was measured with the subject's heels together, and feet at an angle of approximately fifteen degrees. The maximal circumference was identified by raising and lowering the tape. The tape was checked to ensure that it was horizontal at all points.

Thigh circumference

The mid-point between the mid-inguinal point and the proximal border of the patella was located, with the subject sitting so that the thigh was flexed at 90 degrees. This point was marked. Measurement was made at the level of this mark with the subject standing, with weight on the right leg, and the left leg relaxed. The tape was held at right angles to the long axis of the limb.

Calf circumference

Measurement was made with the subject sitting, with the lower leg relaxed, and not in contact with the floor. The tape was lowered and raised until the maximum circumference was found. A mark was then made on the medial aspect of the calf at this level for measurement of medial calf skinfold.

Skinfold thicknesses

Skinfold thicknesses were measured using Holtain Tanner/Whitehouse skinfold calipers (Holtain Ltd., Crymych, U.K.). The calipers were rectangular ended, and exerted a constant pressure of 10 g/mm². The pressure exerted was calibrated by clamping the caliper handle, and hanging known weight from the jaws, until the minimum weight necessary to open the jaws was calculated. If necessary the holding spring was tightened or slackened and the process repeated until the correct pressure was achieved.

All skinfold thicknesses were measured on the left side of the body, except in one subject where a scar prevented measurement of abdominal skinfold on the left, so measurement was taken on the right. Skinfolts were lifted with the thumb and fingers of the left hand one centimetre from the skinfold site, so that the two layers of skin at the point of measurement were parallel. Calipers were applied, and the reading to the nearest 0.1 mm taken after two seconds. This was repeated, and the mean of the two measurements calculated.

Biceps skinfold

The mid-point between the acromion and the olecranon was located with the arm flexed at 90 degrees. Measurement was made at this level, on the anterior aspect of the arm, over the belly of the biceps muscle. During measurement, the arm was relaxed by the side, with the hand supinated.

Triceps skinfold

Triceps skinfold was measured at the same level as the biceps skinfold, but on the posterior aspect of the arm, above the olecranon process. The arm was kept relaxed by the side during measurement.

Subscapular skinfold

Subscapular skinfold was measured with the subject standing, with shoulders relaxed, and arms by the sides. Skinfold thickness was measured vertically 1cm below the inferior angle of the scapula.

Suprailiac skinfold

Suprailiac skinfold was measured vertically, above the iliac crest, on the mid-axillary line.

Mid-axillary skinfold

Mid-axillary skinfold was also measured vertically, on the mid-axillary line, but at the level of the xiphoid process.

Abdomen skinfold

A marking was made five centimetres to the left of, and at the level of, the umbilicus. Skinfold thickness was then measured vertically at the site of this marking.

Anterior thigh skinfold

Measurement was made with the subject standing with the weight on the right leg, left leg relaxed. The skinfold was taken vertically, on the anterior aspect of the thigh, at the level of the mid-thigh marking (midway between mid-inguinal point and proximal border of patella, with thigh flexed at 90 degrees).

Medial calf skinfold

Measurement was made with the subject sitting, with the lower leg relaxed and not in contact with the floor. The skinfold thickness was measured vertically, at the level of the maximal circumference, on the medial aspect of the calf.

Bony diameters

Three diameters were measured, to provide information on frame size. Bi-acromial and bi-iliac diameters were measured using an anthropometer, and bi-epicondylar humerus using a bicondylar vernier (Harpenden). Anthropometer blades were applied firmly, to compress soft tissue.

Bi-acromial diameter

The distance between the acromion processes was measured with the subject's shoulders relaxed.

Bi-iliac diameter

The distance between the lateral borders of the iliac crests was measured.

Bi-epicondylar humerus

The distance between the medial and lateral epicondyles of humerus was measured, with the upper arm held at shoulder level, and elbow bent at 90°.

Table 2.1

Standard error of measurement (Smeas) of anthropometric variables

Measurement	Smeas
Weight (kg)	.01
Height (cm)	.13
Circumferences (cm)	
Arm	.19
Chest	.43
Waist	.59
Hip	.39
Thigh	.39
Calf	.24
Skinfold Thicknesses (mm)	
Biceps	.26
Triceps	.36
Subscapular	.41
Suprailiac	.53
Mid-axillary	.51
Abdomen	.32
Anterior Thigh	.44
Medial Calf	.33
Bony diameters (cm)	
Biacromial	.38
Biiliac	.13
Bi-epicondylar humerus	.04

Body Composition

Hydrostatic weighing

Equipment

The equipment used for hydrostatic weighing was that described by Jones and Norgan (1974). Lung residual volume was measured using a three breath nitrogen dilution technique (Rahn et al. 1949; Durmin & Rahaman 1967).

A cylindrical water tank was equipped with thermostatically controlled heaters. The tank had water inlet and outlet valves, for filling and emptying. A pump was fitted to the tank, to pass the water through a filter. Steps and supports were fitted to the outside and inside of the tank, to facilitate entry and exit. The water in the tank was slightly chlorinated, to prevent transmission of infection, although the water in the tank was generally changed after measurement of 3-4 subjects.

A plastic chair was suspended in the water from a hoist, which was connected to a strain-gauge dynamometer and transducer, which was in turn suspended from the ceiling. A digital voltameter was attached to allow reading of underwater weight.

Snorkels and nose-clips were used to allow respiration underwater. 4 litre rubber anaesthetic bladders filled with 100% oxygen were used to measure residual volume. These were fitted with a 3-way tap, which was tooled to allow a good, airtight fit with the end of the snorkel.

A computer terminal connected to the university mainframe computer was available to allow calculation of body density and percentage fat. The equipment used is shown in Figure 2.1.

Figure 2.1

Measurement of body composition by hydrostatic weighing



Procedure

The water was heated to a temperature of 35 to 36° C, and the heaters electrically isolated before measurement. The pump was also turned off 10 to 15 minutes before measurement, to allow the water to settle. The digital voltameter was calibrated by attaching a known weight to the chair suspension ropes.

Measurement was made after voiding, with the subject wearing a skin-tight swimming costume. The subject was asked to shower before measurement, to reduce skin surface oils and loose hair. The subject then stepped through a chlorinated foot bath, and was guided up the steps and into the tank.

The subject was directed to position himself centrally in the chair, and attach a seat-belt to ensure that he remained in contact with the chair. The feet were positioned on a footrest attached to the chair, to ensure that they did not come into contact with the floor during measurement. A snorkel was attached to the head with an adjustable strip of elastic, and a nose-clip fitted. The subject was asked to try to expire through the snorkel with the end blocked, to ensure that there was no escape of air. The chair was then raised or lowered until the water level reached the chin.

Subjects were then familiarised with the procedures. They were asked to submerge themselves briefly by bending forward from the waist. The purpose for this was threefold: firstly to accustom the subject to submersion; secondly to ensure that complete submersion was possible without the subject, chair or footrest coming into contact with the walls or floor of the tank; and thirdly to make sure that the water level was not too high. Then the subject was asked to submerge completely, practice total expiration through the snorkel, and lift a finger to indicate end expiration. Care was taken to ensure that submersion was gradual, so that the water would not be disturbed and so cause the digital voltameter reading to take longer to reach a plateau. Finally the complete procedure was practised: the subject submerged and expired maximally as before. He remained submerged and held his breath until the digital voltameter reading became stable, and the underwater weight was read. Then the procedure for measuring residual volume was practised: a bladder containing 3 litres of air was attached to the end of the snorkel, the tap opened, and the subject asked to take three deep breaths in and out of the bladder, over a period of 9 seconds.

Once the subject was ready, actual measurement was made as above, except that the bladder contained 3 litres of 100% oxygen, and underwater weight was recorded. The temperature of water in the tank was measured to calculate water density, and the weight of chair and apparatus without the subject measured, to correct the underwater weight.

Measurement was repeated until values agreeing to within 1 % body fat were obtained. An interval of at least 8 minutes was allowed between each measurement, to allow the concentration of oxygen in the lungs to return to normal values.

Calculation

The contents of the bladder were analysed for oxygen and carbon dioxide content, as described below. Nitrogen concentration was calculated by subtraction. Residual volume (RV) was then calculated as follows:

$$RV (l) = \left\{ V(l) \times \frac{[F_1N - n]}{FN - F_1N} \times BTPS \right\} - DS$$

where V is the volume of the bladder (3l); FN is the alveolar nitrogen concentration (80%), n is the nitrogen content of the bladder before rebreathing (0.5%); F₁N is the nitrogen content of the bladder after rebreathing; BTPS is the barometric pressure and temperature correction factor; and DS is the dead space of 3-way valve and snorkel (0.21 l).

The body density (Db) can then be calculated:

$$Db (kg/m^3) = \frac{Ma}{\{ [Ma - Mw] \div Dw \} - RV (l)} \times 1000$$

Where Ma is the mass of the body in air (kg); Mw is the mass of the body in water (kg); and Dw is the density of water (kg/l).

Percentage body fat was calculated from the equation of Siri (1961) given in chapter 1.

Skinfold thickness

The logarithm (base 10) of the sum of biceps, triceps, subscapular and suprailiac skinfolds was calculated, to estimate body density using the age and sex specific equations of Durnin and Womersley (1974). Percentage body fat was then calculated using the equation of Siri (1961).

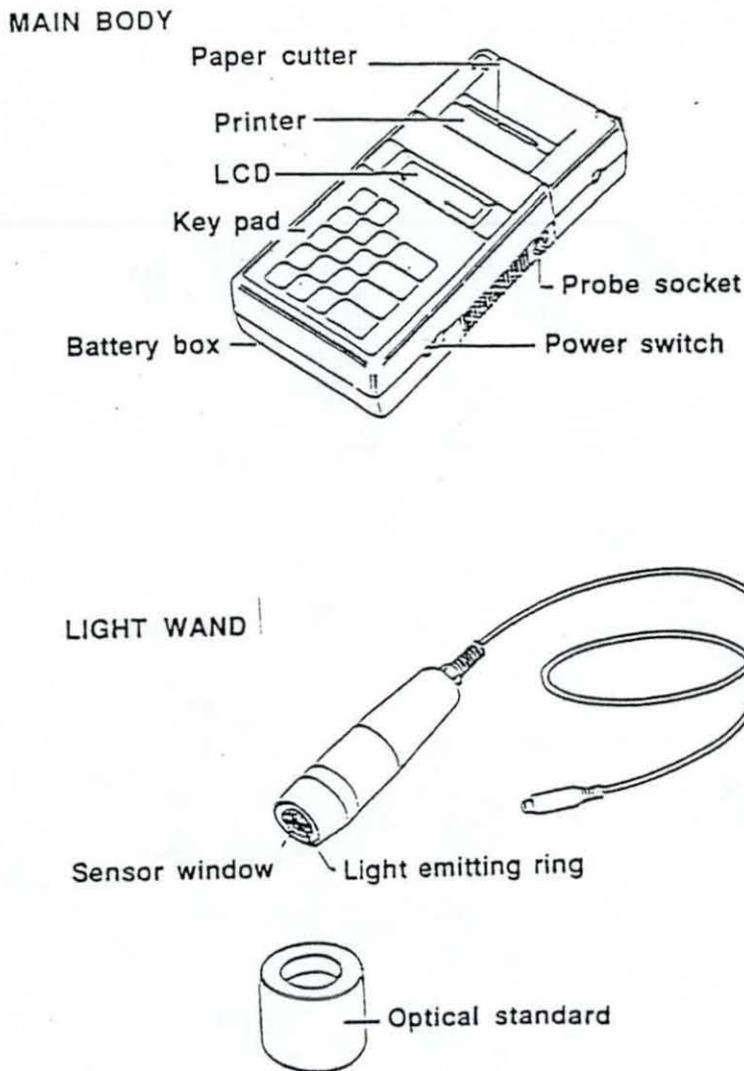
Near Infra-red Interactance

Equipment

Near infra-red interactance was measured using the Futrex 5000 Body Composition Meter (Self Care Products Ltd, Rybar House, Amersham, Bucks, UK). A light wand contained four infra-red emitting diodes, two emitting radiation at a wavelength of 940 nm, and two at 950 nm. These illuminated a circular diffusing ring, which allowed radiation to be emitted evenly. In the centre was a silicon detector which measured the intensity of light re-emitted. The light wand was attached to a Hitachi microprocessor from which the optical density readings ($\log I/I$) were read. An optical standard was provided for calibration. The equipment used is shown in Figure 2.2.

Figure 2.2

Diagram of instrument for measurement of near infra-red interactance (Futrex-5000)



Procedure

Measurement was made in accordance with the procedures described in the Futrex 5000 User's Manual. The light wand was held firmly at the site of the biceps skinfold thickness measurement, with the upper arm relaxed, resting on a table with the upper arm at an angle of 45 degrees, and hand supinated. A light shield was used to prevent ambient light reaching the near infra-red detectors. Optical densities at 940 nm (Od1) and 950 nm (Od2) were recorded.

Values were corrected by measurement of an optical standard, to allow for electronic drift. This correction amounted to up to 10% of the total optical density value in some cases. However this made a difference of less than 1% body fat, as the corrections of the two wavelengths were highly correlated and so tended to counteract each other.

Calculation

Percentage body fat was calculated from measurements at the biceps site, using equations supplied by the manufacturer, which were:

$$\begin{aligned} \% \text{Fat} = & 61 - 65(\text{Od2}) - 141.4(\text{sex}/100) + 0.045(\text{Weight lb}) - 0.243(\text{Height in}) \\ & + 35.9(\text{Od1}) - 121.1(\text{Ex}/100) \end{aligned}$$

This was rearranged to allow metric measurements to be used to:

$$\begin{aligned} \% \text{Fat} = & 61 - 65(\text{Od2}) - 1.414(\text{sex}) + 0.0992(\text{Weight kg}) - 9.567(\text{Height m}) + \\ & 35.9(\text{Od1}) - 1.211(\text{Ex}) \end{aligned}$$

where Od1 and Od2 are optical density measurements at 940 and 950 nm respectively, and sex is coded as -1 for women and 1 for men. Ex is the exercise level which was calculated as described by the manufacturer: scores of 2 (sedentary), 5 (moderate), or 8 (heavy) corresponded to less than 15 minutes, 30 to 60 minutes and over 60 minutes exercise per day respectively. All exercise of an intensity equivalent to or greater than that of brisk walking was included. Where the amount of exercise had varied in the weeks or months preceding the test, an average over the preceding three months was estimated.

Bioelectrical impedance

Equipment

Bioelectrical impedance was measured with the E-Z Comp 7000 Body Composition Meter (Fitness Concepts, Inc.). This consisted of a micro-processing unit, which controlled the application of a 800 μ A, 50 kHz current through leads which were attached via crocodile clips to the distal electrodes. The current returning through proximal electrodes was returned to the unit through similar leads, and the voltage drop and hence impedance was calculated. The unit also allowed entry of subject information, and calculated body composition using pre-programmed equations. The electrodes used were supplied by the manufacturer.

Procedure

Measurements were made according to manufacturers instructions. Four electrodes were used to minimise the contact impedance. The skin was cleansed with alcohol prior to application of electrodes. Electrodes were placed on the dorsal surfaces of the hand and foot, proximal to metacarpal phalangeal and metatarsal phalangeal joints, and also between the distal prominences of the radius and ulna and between medial and lateral malleoli at the ankle. The subject lay supine on a clean dry mat, with limbs slightly abducted such that there was no contact between legs, or between the arms and the body. The position of the subject during measurement is shown in Figure 2.3.

The sex, height, weight and age of the subject were entered into the instrument. Impedance, and body composition estimates calculated from equations of Fitness Concepts Inc. (**BIA**) programmed into the machine were recorded. These equations were not supplied by the manufacturer.

As the technique is dependent upon the water content of the body, factors which may alter body water content were avoided. Subjects were required to avoid strenuous exercise and consumption of alcohol in the 24 hours preceding measurement, and consumption of food, coffee or tea during the 4 hours immediately preceding measurement.

Calculation

Body composition was also calculated from impedance from the equations given below:

Lukaski et al. (1985)

$$\text{Men : FFM} = 0.827 (ht^2 \div Z) + 5.214$$

$$\text{Women: FFM} = 0.821 (ht^2 \div Z) + 4.917$$

(**BIA-L**)

Segal et al. (1985)

Men: $Db = 1.554 - 0.0841 (wt \times Z \div ht^2)$

Women: $Db = 1.1113 - 0.0556 (wt \times Z \div ht^2)$ (BIA-S)

Segal et al. (1988)

Men: $FFM = 6.636 \times 10^4 ht^2 - 0.02117 Z + 0.6285 wt - 0.1238 age + 9.333$

Women: $FFM = 6.4602 \times 10^4 ht^2 - 0.01397 Z + 0.42087 wt + 10.43485$ (BIA-S2)

Deurenberg et al. (1991)

$FFM = 0.340 (ht^2 \div Z) + 0.1534 ht + 0.273 wt - 0.127 age + 4.56 sex - 6.48$

(BIA-D)

Where FFM is fat-free mass (kg); Db is body density (kg/l); Z is impedance (Ω); ht is height (cm); wt is weight (kg); and sex is coded as 1 for men, 0 for women.

Figure 2.3

Measurement of body composition by bioelectrical impedance



Ultrasound

Equipment

Ultrasonic measurements were made using a 5 MHz transducer with an Ekoline 20A Ultrasonoscope (Smith Kline Instruments, Inc., Sunnyvale, CA) in A-scan mode. The transducer was enclosed in a surround with a diameter of 5 cm, as described by Jones et al. (1986), which minimised the amount of movement of the transducer, and assisted in maintaining the transducer perpendicular to the interface being measured. Aquasonic gel was applied to the transducer to couple it to the dermis.

Procedure

Measurements were made with the subject lying to minimise movement and to allow greater muscular relaxation. The echo corresponding to the fat-muscle interface was located at each site. The correct identification of interfaces was facilitated by the use of an oscilloscope display showing echo amplitude. The time taken for the echo to return was converted to distance using the average velocity of ultrasound in soft tissue: 1540 m/s. A foot control was used to obtain a printout of the oscilloscope once the correct echo had been identified, from which the depth was read using X-ray measuring calipers.

Exercise testing

Four exercise tests were conducted. A submaximal, incremental treadmill test was used to estimate oxygen uptake, and establish the treadmill inclines necessary for subsequent tests. A grade-lactate test established the oxygen consumption at a reference lactate concentration. An endurance test examined blood lactate concentrations during sustained exercise. A one mile track test allowed assessment of brisk walking pace.

Before treadmill tests, on a separate visit, subjects were allowed to become familiarised with treadmill walking, until subject and investigator were satisfied that walking was normal, and that the subject was accustomed to treadmill walking.

Submaximal incremental treadmill test

Subjects walked at a constant speed of 4.8 or 5.6 kph (depending upon performance during treadmill familiarisation) for four four-minute stages. During the first stage the incline of the treadmill was 0 or 3°. The incline was increased by 3° at the beginning of each subsequent stage. Expired air was collected during the last minute of each stage, and heart rate was continuously monitored.

Maximum heart rate was estimated from $210 - (0.65 \times \text{age})$ (Astrand 1960). A linear regression equation was fitted to the mean heart rates and rates of oxygen

consumption at each sampling period. This was used to predict oxygen consumption at maximum heart rate. This was taken to be maximal oxygen uptake, despite the lower accuracy of predicted values of maximal oxygen uptake, as the subjects (middle-aged men) were not accustomed to strenuous physical exercise, so a maximal test may be unwise.

A regression between treadmill incline and oxygen consumption was also performed. This was used to calculate the treadmill inclines required to elicit 50%, 60%, 70% and 80% of VO_2max , to be used in subsequent tests.

Grade-Lactate treadmill test

This test again involved four four-minute stages: this time the incline of the treadmill was set to elicit 50%, 60%, 70% and 80% of VO_2max respectively at each stage. The treadmill inclines necessary were calculated from the previous test.

During the last minute of each stage, expired air was collected and heart rate recorded. At the end of each stage a thumb-prick sample of capillary blood was collected, for analysis of blood lactate concentration.

A regression between oxygen uptake and blood lactate concentration was performed, to calculate the oxygen uptake at 2 mmol/l blood lactate. Maximal oxygen uptake was predicted as above.

Endurance walk

This test involved the subject walking at an incline sufficient to elicit 70% of VO_2max for 20 minutes. Expired air was collected during the last minute of each quarter of the test. Heart rate was recorded during each expired air collection. A sample of blood was taken for analysis of blood lactate at the end of each expired air collection.

One mile track walk

Subjects were instructed to walk a distance of 1600 m at a brisk pace (one which could be maintained for 30-45 minutes). The time taken to complete the distance was recorded to assess brisk walking pace, and heart rate was recorded throughout the test.

Collection of expired air

Oxygen uptake and carbon dioxide production during exercise were determined by the collection of samples of expired air. Subjects wore a nose-clip, and a mouthpiece connected via lightweight tubing to a 150 litre Douglas bag. A three-way valve attached to the neck of the Douglas bag was used to open and close the bag.

Analysis of oxygen and carbon dioxide

Oxygen content of expired air was measured using a paramagnetic oxygen analyser (Taylor: Servomex model 570A) and carbon dioxide measured using an infra-red analyser (Lira: Mines Safety Appliances Ltd. Model 3030). Both analysers were calibrated before measurement using certified reference gases, which were themselves calibrated against a "gold standard" reference gas whenever they were changed.

The amount of air used for analysis was measured using a flowmeter, and the remaining air evacuated through a Harvard dry gas meter. Temperature of the expired air sample was measured during evacuation, and used with pressure to correct volumes to standard temperature and pressure.

Analysis of blood lactate

Thumb prick samples of blood were collected into micro-pipettes, and deproteinised in 200 μ l 2.5 % perchloric acid. They were then frozen at - 70 ° C until analysis. Blood lactate concentration was measured on a fluorimeter, using an enzymatic technique.

Blood biochemistry

Two fasting blood samples were obtained at each testing period, on separate days no more than five days apart, to allow for day-to-day variation. Samples were then divided up for analysis.

Haemoglobin concentration was measured photometrically, using a cyanmethemoglobin technique, in a commercially available kit (Boehringer Mannheim, U.K. Ltd). Haematocrit was measured by centrifuging a heparinized micro-pipette of blood, and using a micro-haematocrit reader (Hawksley).

Total cholesterol and triglyceride concentrations were measured using commercially available kits (Boehringer Mannheim, U.K. Ltd). HDL-C was isolated using a manganese chloride/sodium heparin precipitation. It was then measured using the total cholesterol assay above. VLDL-C was estimated from triglyceride concentration, and LDL-C estimated from subtracting HDL-C and VLDL-C from TC, using the equation of Friedewald et al. (1972).

Apoproteins A-I and B were determined by immunoprecipitin analysis using commercially available kits (Atlantic Antibodies, Inc. U.S.A.). Lp(a) concentration was also determined using a commercially available kit (Immuno Ltd.) using an enzyme immunoassay.

Blood pressure

Blood pressure was measured using a random zero sphygmomanometer (Hawksley). The procedures suggested by Elliott & Stamler (1988) were followed.

Subjects sat down for at least five minutes before measurement, with the legs uncrossed. The "zero reading" was set randomly before each measurement. The cuff was placed around the right arm. The brachial artery was located by palpitation of the cubital fossa until a pulse was found. The stethoscope was then held over the brachial artery, and the cuff inflated. The cuff was then deflated until the pulse was first heard, and the sphygmomanometer reading recorded. The cuff was deflated further until the pulse could no longer be heard, at which point the sphygmomanometer reading was recorded again. The cuff was then completely deflated, and the zero reading recorded. This was subtracted from the first two readings to calculate systolic and diastolic blood pressures.

The measurement was repeated, and the mean of the two measurements was taken. However if the blood pressure readings were inconsistent, a third measurement was made.

Statistics

The standard error of measurement (Smeas) was calculated as the standard deviation of the differences between repeated measurements, divided by the square root of 2. All other analyses (Students t-test, Pearson product moment correlation coefficients, regression analysis and analysis of variance) were made using Statview (Brainpower Inc.) on an Apple Macintosh personal computer, as was principal components analysis, using an orthogonal varimax technique.

Skewed data was logarithmically transformed prior to analysis. Prior to correlation and linear regression analyses, bivariate plots were examined, to ensure that relationships were linear and not curvilinear.

Chapter 3

Comparison of near Infra-red interactance and skinfold thickness with hydrostatic weighing

Introduction

Near infra-red interactance is a relatively new technique for the assessment of body composition. It has not been widely evaluated, so it was evaluated here by comparison with hydrostatic weighing, a commonly accepted "gold standard" in body composition studies. Two groups of subjects were studied: middle-aged men and female runners. The primary concern was the accuracy of the technique. Practical considerations on the suitability of the technique for "field" studies were also considered.

The estimation of body fatness by near infra-red interactance involves the use of prediction equations (Chapter 2). These equations include weight and height, as well as anthropometric data. Weight and height are likely to explain a large proportion of the variance in body fatness, so to determine the relative contribution of interactance data to the estimate of fatness, the contribution of interactance to these equations is examined.

Measurement of skinfold thicknesses is currently the most widely used technique for such field studies. They were also measured here to allow a comparison of their performance with that of near infra-red interactance. This comparison concentrated mainly upon the relative accuracy of the two techniques, but other factors were also considered, as in some studies a slight decrease in accuracy may be acceptable if compensated for by other advantages.

A large number of anthropometric measurements have been used in the estimation of body composition. The sites and measurements found to best predict body composition have varied considerably between studies: many of which have been reviewed by Lohman (1981). A number of other anthropometric variables were therefore measured in this study, to determine which were best associated with body composition in this sample. This comparison was performed only with the data for the middle-aged men, as the number of female runners was not sufficient to allow multivariate analysis.

Methods

Subjects

Subjects were 63 middle-aged men who volunteered for the brisk walking study described in chapters 6 and 7; and 13 female competitive athletes: 12 runners and a triathlete, who ran on average 48 ± 22 km per week in the six months prior to measurement. Details are shown in table 3.1.

Table 3.1

Characteristics of middle-aged men and women runners

	Middle-aged men n=63	Women runners n=13
Age (years)	51.8 ± 5.3 (43.4 - 61.6)	36.6 ± 6.6 (22.7 - 47.1)
Height (m)	1.766 ± 0.067 (1.585 - 1.943)	1.682 ± 0.068 (1.594 - 1.805)
Weight (kg)	78.9 ± 10.8 (49.3 - 120.1)	55.7 ± 5.5 (47.6 - 66.3)
Body Mass Index (kg/m ²)	25.2 ± 3.0 (17.3 - 35.3)	19.7 ± 1.9 (15.8 - 22.2)

Procedures

Subjects reported to the laboratory having avoided heavy meals and consumption of carbonated drinks. They were asked to void and change into swimming costumes. Body weight, height, anthropometry and near infra-red interactance and hydrostatic weighing were measured as described in chapter 2.

The "exercise level" used in the equations for calculation of body composition from near infra-red interactance was calculated from the average physical activity over the previous three months, from activity diaries which had been kept by both groups. Of the middle-aged men, 17 were coded as "moderate", and the remaining 46 were coded as "light". Of the female athletes, 11 were coded as "moderate", and 2 who were suffering from injury coded as "sedentary". It is interesting to note that this coding system did not differentiate between athletes performing intense activity, running up to 20 km per day and men walking up to 5 km per day.

Results

Comparison of means of body composition techniques

Descriptive statistics for estimates of body composition by hydrostatic weighing, skinfold thickness, and near infra-red interactance are shown in Table 3.2. Mean % fat by hydrostatic weighing was 28.4 ± 5.8 for middle-aged men, and 20.6 ± 6.0 for female athletes. Estimates of % fat by skinfold thickness were 3.3 lower in the men, but 2.0 higher in the women, than these values. Near infra-red interactance underestimated % fat relative to hydrostatic weighing, by 9.6 in the men, and 4.8 in women. These differences were all significant ($P < 0.001$ in men, $P < 0.05$ in women).

The underestimation by near infra-red interactance was larger than that of 3.5 % fat observed by Elia et al. (1990) in 34 men and women aged 18-40.

Table 3.2

Mean, standard deviation and range of body composition estimates by hydrostatic weighing (HW), skinfold thickness (SKF), and near infra-red interactance (NIRI)

	HW	SKF	NIRI
Middle-aged Men n=63	%fat 28.4 ± 5.8 (16.6 - 43.6)	25.1 ± 4.7 *** (13.5 - 39.2)	18.8 ± 4.0 *** (6.0 - 27.3)
	fat-free mass (kg) 56.2 ± 7.3 (40.2 - 76.4)	58.7 ± 6.2 *** (42.7 - 75.0)	63.8 ± 7.2 *** (46.3 - 87.3)
	fat mass (kg) 22.6 ± 6.7 (9.1 - 51.5)	20.1 ± 6.0 *** (6.6 - 47.0)	15.0 ± 4.6 *** (3.0 - 32.8)
Women runners n=13	%fat 20.6 ± 6.0 (11.8 - 28.6)	22.6 ± 5.2 * (12.3 - 30.0)	15.8 ± 7.7 ** (1.8 - 24.1)
	fat-free mass (kg) 44.1 ± 4.4 (39.2 - 55.4)	42.9 ± 3.8 * (38.8 - 51.7)	46.6 ± 3.7 ** (41.1 - 55.2)
	fat mass (kg) 11.6 ± 3.8 (6.0 - 17.3)	12.7 ± 3.5 * (5.9 - 16.9)	9.0 ± 4.6 ** (0.9 - 13.6)

Significance of difference from hydrostatic weighing by paired t-test:

*** $P < 0.001$ ** $P < 0.01$ * $P < 0.05$

Regression Analysis

Table 3.3 shows correlation coefficients and standard errors of estimation of the regression of skinfold thickness and near infra-red interactance with hydrostatic weighing. Correlation coefficients ranged from 0.58 to 0.91. In both the men and women, better agreement was observed for skinfold thickness than near infra-red interactance. Correlation coefficients were higher in the women than the men.

The standard error of estimation for % fat by skinfold thickness was 4.6 for the men and 3.1 for the women. These values are similar to those reported by Durin and Womersley (1974), but values for the middle-aged men were higher than those reported in young adults by Lohman (1981).

The standard error of estimation for % fat by near infra-red interactance was higher than that for skinfold thickness in the men (4.8) and the women (4.3). These values are slightly higher than those observed by Elia et al. (1990) in males and females aged 18-40.

Table 3.3

Regression of skinfold thickness (SKF) and near infra-red interactance (NIRI) estimates of body composition with those from hydrostatic weighing (HW): correlation coefficient (r) and standard error of estimate of regression (SEE)

Group	Variable	SKF		NIRI	
		r	SEE	r	SEE
Middle-aged men n=63	% fat	.63	4.55	.58	4.78
	fat-free mass	.87	3.64	.84	3.98
	fat mass	.84	3.62	.80	4.06
Women athletes n=13	% fat	.83	3.06	.85	4.27
	fat-free mass	.91	1.65	.87	1.91
	fat mass	.88	1.77	.88	2.23

Further analysis

Regression analysis has some drawbacks when used to compare two methods. Firstly the regression is highly dependent upon sample size, and outlying points have a great influence upon the regression. Secondly, when comparing two techniques, it is preferable that the slope of the regression is approximately 1. By fitting a regression line whose slope deviates greatly from unity, the variance will be reduced, and lower standard errors of estimate will be obtained.

For the regression of % fat by hydrostatic weighing with % fat by skinfold thickness and near infra-red interactance above, slopes of 0.8 and 0.8 respectively were observed in the men, and 0.9 and 0.7 respectively in the women. So a slope of 1 was not observed here, producing more favourable results. The regression analysis is included to allow comparison with other workers' results, but the analysis proposed by Bland and Altman (1986) was also performed to provide a more realistic evaluation of the data.

The Bland and Altman analysis involves plotting the mean of the two techniques against the difference between them (Figure 3.1). The mean difference between the two methods estimates the extent of a consistent bias i. e. over- or under-estimation, and the standard deviation represents the variation around this mean.

Figure 3.1
Bland and Altman plot comparing % fat by near infra-red interactance (NIRI) with % fat by hydrostatic weighing (HW)

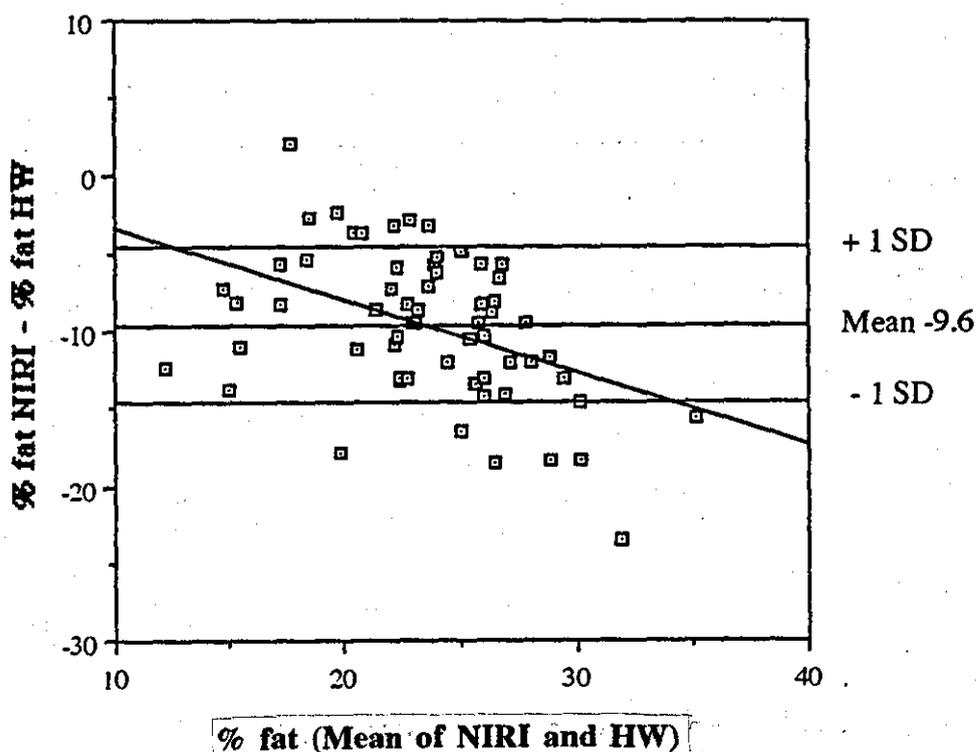


Table 3.4 shows the mean and standard deviation of the differences between the methods. It is apparent that there was a consistent underestimation by near infra-red interactance, whilst skinfold thickness underestimated fatness in the men, and slightly overestimated in the women. The standard deviations confirm the findings of the regression analysis: that error is greater for near infra-red interactance than skinfold thickness; and greater in the men than the women.

Table 3.4

Bland and Altman analysis: Mean and standard deviation of difference between body composition estimates by skinfold thickness/near infra-red interactance and hydrostatic weighing

Group	Variable	SKF-HW		NIRI-HW	
		mean	SD	mean	SD
Middle-aged men n=63	% fat	-3.3	4.6	-9.6	4.8
	fat-free mass	2.5	3.6	7.6	4.1
	fat mass	-2.5	3.6	-7.6	4.1
Women athletes n=13	% fat	2.0	3.4	-4.8	4.1
	fat-free mass	-1.1	1.9	2.6	2.1
	fat mass	1.1	1.9	-2.6	2.1

The Bland and Altman analysis was also used to determine whether the bias is consistent across the whole range of measurement, or whether the bias increased or decreased with increasing values. A regression line was fitted to the Bland and Altman plot (figure 3.1), with the slope indicating the extent of change in bias with increasing values, and the correlation coefficient being a measure of the significance of the relationship. The slope and correlation coefficients for regression lines of plots of the mean of the two techniques against the difference between the two techniques are shown in Table 3.5.

In the men a greater underestimation with increasing fatness was observed for both skinfold thickness and near infra-red interactance, indicated by a negative slope. The extent of this decrease was greater for near infra-red interactance than skinfold thickness, with the slope being nearly twice as steep.

For the women a similar trend towards greater overestimation at increasing values was observed, which was again greater for near infra-red interactance than skinfold thickness. However the slopes were smaller than those for the men, and not significant, possibly because of the small sample size in this group.

Table 3.5

Bland and Altman analysis: Correlation coefficient and slope of regression line fitted to plot of mean of two methods against difference between methods

Group	Variable	SKF-HW		NIRI-HW	
		r	slope	r	slope
Middle-aged men n=63	% fat	.27 *	-.26	.43 ***	-.46
	fat-free mass	.32 *	-.18	.01	-.01
	fat mass	.18	-.11	.54 ***	-.42
Women athletes n=13	% fat	.25	-.15	.41	.26
	fat-free mass	.35	-.16	.31	-.17
	fat mass	.17	-.09	.34	.18

Significance of regression: *** P<0.001 ** P<0.01 * P<0.05

Regression of near infra-red interactance with skinfold thickness

The correlations of skinfold thickness and near infra-red interactance with hydrostatic weighing in table 3.6 tended to be similar, although lower for near infra-red interactance than skinfold thickness. This could be because the two are correlated with each other, which seems plausible as both depend on logarithms of measurements dependent upon the quantity of subcutaneous fat. To determine whether near infra-red interactance agreed better with skinfold thickness than with hydrostatic weighing, near infra-red interactance was correlated with skinfold thickness. Correlation coefficients and standard errors of estimation are shown in table 3.6.

Table 3.6

Correlation coefficient and standard error of estimation of regression of skinfold thickness (SKF) with near infra-red interactance (NIRI)

Group	Variable	r	SEE
Middle-aged men n=63	% fat	.67	3.54
	fat-free mass	.90	2.68
	fat mass	.86	3.11
Women athletes n=13	% fat	.87	2.66
	fat-free mass	.86	2.00
	fat mass	.91	1.53

Skinfold thickness and near infra-red interactance correlated better with each other than with hydrostatic weighing (Table 3.3). This could be because both techniques depend on the subcutaneous adipose tissue thickness. Differences in the proportion of total fat situated subcutaneously will account for some of the difference when NIRI or skinfold thickness are compared with hydrostatic weighing, but will not be involved when NIRI is compared with skinfold thickness.

It must also be considered that the hydrostatic weighing values themselves are not "true" values. This technique itself has a considerable number of associated sources of error, as described in chapter 1. So when comparing two techniques, it is impossible to determine exactly how much of the difference between them is due to errors in the "reference" technique.

Correlation of optical density data with %fat

The equation used to estimate % fat from near infra-red interactance contains six variables: weight, height, gender and activity level as well as the two optical density measurements. It is possible that very little of the variance is explained by optical densities. To determine the extent of agreement between optical densities and fatness, correlation coefficients of optical densities at 940 and 950 nm with % fat by hydrostatic weighing were calculated, and are shown in Table 3.7.

Table 3.7

Correlation of optical densities at 940 and 950 nm with % fat by hydrostatic weighing

	Od 940	Od 950
Middle-aged men	-.57	-.58
Female athletes	-.81	-.82

Optical densities were negatively correlated with fatness. The correlations of the two wavelengths were similar. Correlations were higher in the women than the men, but in both groups correlations were similar to those of % fat (NIRI) with % fat by hydrostatic weighing.

One cause for the lower correlations observed in the men could be their greater age. A more central distribution of body fat with age has been reported, possibly making the composition at the biceps site a less reliable predictor of total body composition with increasing age.

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Contribution of variables in near infra-red interactance prediction equation to variance in body fatness

The correlation of optical densities with body fatness was not much lower than that of the estimate of body fatness from the near infra-red interactance prediction equations. However, these prediction equations also include weight and height, and as weight and height are themselves well correlated with fatness, would be expected to improve the agreement. This finding prompted an evaluation of the relative contribution of the variables in the near infra-red interactance prediction equations. In a multiple regression, weight, height and activity level accounted for 35 % of the variance in body fatness. The addition of Od2 to the regression increased the proportion of variance explained to 45 %. The addition of Od1 did not increase this value further.

So it seems that including one optical density measurement in addition to the other variables can provide more information on body fatness than the other variables alone, whilst the addition of a second optical density measurement does not provide any further information.

Correlation of anthropometric variables with body density

Table 3.9 shows correlation coefficients of skinfold, circumference and diameter data with body density. The best single predictor of body density in this group of middle-aged men was the biceps skinfold, which was better correlated with body density than the predicted % fat from skinfold thickness and NIRI. The waist circumference was the next best correlated variable. In general limb skinfold thicknesses were better correlated with body density than trunk skinfolds, whilst trunk circumferences were better correlated than limb circumferences.

A stepwise regression was performed to determine which combination of anthropometric variables best predicted body density in this sample. Stepwise regression has its limitations, in that entering a number of variables is likely to produce a regression highly specific for the sample studied, which may not apply to the population as a whole. Also some variables will tend to be highly correlated, so should strictly be combined before analysis. However the purpose of the analysis was to determine which variables best explained body density in this sample, for which this was the most convenient method.

Five variables were entered into the regression, which accounted for 65 % of the variance in body density. Variables in order of entry were: biceps skinfold, abdomen skinfold, anterior thigh skinfold, waist circumference and chest circumference. The coefficients of abdomen skinfold and chest circumference were positive, indicating that higher values of these variables would produce higher density values and hence lower

fatness. Abdomen and anterior thigh skinfold thicknesses were both separately poorly correlated with body density, and also more difficult to measure than skinfolds at other sites: reliable measurements of each were not obtained for one subject.

Table 3.8

Correlation of anthropometric variables with body density in middle-aged men (ranked according to correlation)

Measurement	Site	Correlation
Skinfold Thicknesses *	Biceps	-.68
	Medial Calf	-.59
	Triceps	-.57
	Mid-axillary	-.53
	Suprailiac	-.49
	Subscapular	-.48
	Anterior Thigh	-.45
	Abdomen	-.25
Circumferences *	Waist	-.60
	Hip	-.45
	Chest	-.42
	Thigh	-.38
	Upper arm	-.37
	Calf	-.33
Diameters	Bi-iliac	-.19
	Bi-acromial	-.06
	Bi-epicondylar humerus	-.01

* Logarithmically transformed

Discussion

For both skinfold thickness and near infra-red interactance, better agreement with hydrostatic weighing was observed in the female runners than in the middle-aged men. This could be due to the more homogeneous nature of the group of runners. Another possibility is that age-related changes in fat distribution, as reported by Enzi et al. (1986), are responsible for reducing the agreement in older subjects.

Estimation of body composition by near infra-red interactance

Near infra-red interactance was an extremely convenient tool to use. Measurement was faster than that of skinfold thicknesses, and preferred by subjects. Operation was simple, and minimal observer training would be required. The instrument is portable, and whilst more expensive than skinfold calipers, is cheaper than a number of other techniques. These characteristics would seem to make it an ideal tool for field studies.

Near infra-red interactance was found to be a worse predictor of % fat determined by hydrostatic weighing than skinfold thickness, with larger standard errors of estimation. It underestimated fatness, and this underestimation increased with increasing fatness.

It is possible that the underestimation by near infra-red interactance is due to characteristics of the prediction equation used. The prediction equation was developed by regression with hydrostatic weighing by the manufacturers of the Futrex 5000. However details of the number or characteristics of the subjects in the validation group are not available. The poor performance of the prediction equation could be due to population differences, or differences in methodology of hydrostatic weighing or measurement of near infra-red interactance (although manufacturer's instructions were followed). It is possible that better agreement could be seen from development of a prediction equation more suitable to the groups studied, possibly including measurements at more sites.

The correlation of raw near infra-red interactance data with hydrostatic weighing yielded only slightly poorer results than that of skinfold thickness with hydrostatic weighing, suggesting that a reasonable relationship between interactance and fatness might exist. The raw optical density data were correlated as well with hydrostatic weighing as estimates from the equation also containing height, weight, sex and activity level. The addition of one optical density measurement provided more information on body fatness than could be gained from weight, height and activity level alone, although a second optical density measurement provided no further information.

Estimation of body composition from skinfold thicknesses

The agreement of skinfold thickness with hydrostatic weighing was found to be similar to the standard errors reported by Durnin and Womersley (1974), but higher than those reported in young men by Lohman (1981). The increase in bias with increasing fatness observed in the men by the Bland and Altman analysis suggests that skinfold thickness will overestimate fatness in lean subjects, and underestimate in the obese.

Association of anthropometric variables with body density

Of the anthropometric variables studied, the highest correlations with body density were seen with biceps skinfold, then waist circumference. The best combination of variables was: biceps, abdomen and anterior thigh skinfolds, and waist and chest circumferences. Skinfold thicknesses at three of the sites reported by Lohman (1981) to be particularly representative with body density in young adults: subscapular, abdomen, and thigh, were found to have poorest correlations in the group of middle-aged men.

Biceps skinfold was found to be better correlated with body density than % fat (SKF) and % fat (NIRI). This seems to suggest that biceps skinfold would be a better predictor of body density than the sum of four skinfolds and interactance!

The standard error of estimate of body density was lower for biceps skinfold (0.0092) than for the sum of four skinfolds (0.0102). This is in contrast to results of Durnin and Womersley (1974) in men of similar age range, where the standard error of estimate of body density was greater for log biceps skinfold than for log sum of four skinfolds. This difference in the standard error could be caused by the lower standard error of measurement of biceps skinfold than of the other skinfolds measured (Table 2.1). So the reason that biceps skinfold was found to be the best predictor of body density could be that it was measured most reliably in this study. These findings would support the choice of the biceps site in the assessment of body composition. However as this finding is contrary to the reports of other workers, as discussed above, it would seem that the finding is likely to be specific to this sample.

Conclusions

In summary, near infra-red interactance was found measure body composition less accurately than skinfold thicknesses. Near infra-red interactance underestimated fatness: the mean underestimation in the middle-aged men was nearly 10 % fat, so use of this technique could result in misclassification of body fatness. This underestimation was greater at higher levels of fatness. However this may be because the equations used to estimate body composition were inappropriate to this sample.

Skinfold thicknesses performed better than near infra-red interactance throughout. The observed error relative to hydrostatic weighing was similar to that reported in the literature. Biceps skinfold was found to be the single best anthropometric predictor of body density in the sample of middle-aged men.

Chapter 4

Comparison of three "field" techniques for measurement of body composition: skinfold thickness, near Infra-red interactance and bioelectrical impedance

Introduction

In the previous study near infra-red interactance was found to underestimate fatness in middle aged men. A smaller underestimation was observed in a group of female runners, and the correlation with other methods was higher. However the latter group was relatively small. This study involves measurement in younger men and women, to allow examination of whether a consistent underestimation of fatness is again observed, and whether despite this underestimation a better agreement with other methods is obtained as with the female group, and reported by Elia et al. (1990).

In this study near infra-red interactance is compared with two other field techniques. The most well established method used in this study was measurement of skinfold thickness. Whilst the commonly accepted "gold standard" of hydrostatic weighing would have been preferable for an evaluation of the techniques, this involves considerably more time, subject co-operation and technical resources. Any discrepancy between the values obtained from skinfold thickness and those that may have been obtained from hydrostatic weighing would be expected, on the basis of the previous chapter and the work of Elia et al. (1990), to be considerably smaller than the underestimation of fatness observed with near infra-red interactance.

The estimation of body composition from near infra-red interactance depends on measurement at the biceps site only. However, measurement at one site only will mean that inter-individual differences in fat distribution will have a great influence on estimates of body composition. Measurement of skinfold thicknesses at several sites has been reported to provide more accurate estimation of fatness than measurement at one site only (Lohman et al., 1981). In this study, measurement of interactance was also made at four other sites, to determine whether measurement at multiple sites could potentially increase the predictive power of near infra-red interactance.

Body composition was also measured by bioelectrical impedance. This technique, while still relatively new, has been more widely assessed than near infra-red interactance. Some workers have found bioelectrical impedance to have better agreement with hydrostatic weighing than anthropometry and have recommended the use of bioelectrical impedance measurements for clinical situations (Lukaski et al.,

1986). Considerably worse agreement with other techniques has been observed by others (e. g. Segal et al., 1985). The use of different equations to estimate body composition from impedance could contribute to this discrepancy. Several sets of regression equations have been published, and some instruments are programmed with equations devised by their manufacturers. The simplest estimate fat-free mass from height squared/impedance. Others also include variables such as weight or age. Most are either gender specific or include a constant term for gender. The equations of Segal et al. (1988) are fatness specific, with different equations for lean and obese subjects, possibly accounting for differences in the composition of the fat-free mass in obese subjects.

In this study estimates of body composition from impedance using several sets of equations are compared, to determine whether the diversity of reports of the accuracy of the technique can be accounted for by the use of different equations by different observers. The sets of equations used are described in chapter 2. The choice of sets of equations used for evaluation was made on two bases. The first was suitability for young, normal-weight Caucasian adults, so equations developed for use in the obese, children, elderly or other ethnic groups were not considered. The second was to choose equations containing a variety of variables, as equations similar to each other are likely to have similar performance.

All sets of equations used were sex-specific, except those of Deurenberg et al., 1991 (BIA-D) which include a constant term for gender. Those of Lukaski et al., (BIA-L) estimate fat-free mass from height squared/impedance. In equations of Segal et al., 1985 (BIA-S) a term for weight is added to estimate body density. The fatness-specific equations of Segal et al., 1988 (BIA-S2) use separate equations for lean and obese subjects. In this sample only the "lean" equations were employed. These include a term for age for men, but not for women, in addition to the square of height, impedance and weight. Body composition estimates from equations programmed into the Fitness Concepts instrument used in this study were also included (BIA). However these equations have not been published.

Because impedance is dependent upon the water content of the body, factors such as exercise which affect the hydration state have been found to influence estimates of body composition from bioelectrical impedance (Khaled et al. 1988; Deurenberg et al. 1988). The effects of exercise-induced changes in the amount or distribution of body fluids on estimates of body composition by skinfold thickness, bioelectrical impedance and near infra-red interactance were examined in this study by repeating measurements after an exercise session. A circuit training session was chosen, as this includes both aerobic exercise and local exercise, and so may be expected to induce changes in both the amount and distribution of body fluids.

Methods

Subjects

Subjects were 27 male and 27 female young adults, who were largely undergraduates of Loughborough University. They were recruited through the distribution of leaflets, and word of mouth. Details of their age, weight, height, body mass index and fatness are shown in Table 4.1. The subjects in this sample were taller, lighter, and leaner than adults of similar age in the general population (OPCS, 1990).

Table 4.1 Characteristics of subjects
Mean \pm standard deviation (range)

	Women n=27	Men n=27
Age (years)	22.7 \pm 3.1 (19.8 - 32.6)	24.7 \pm 3.0 (20.0 - 30.5)
Body Weight (kg)	58.3 \pm 7.4 (47.0 - 74.4)	70.6 \pm 7.5 (59.6 - 85.7)
Height (m)	1.67 \pm 0.05 (1.54 - 1.80)	1.78 \pm 0.06 (1.68 - 1.89)
Body Mass Index (weight/height ²)	20.7 \pm 2.2 (17.0-25.0)	22.1 \pm 2.0 (18.3 - 26.0)
% body fat (skinfold thickness)	22.3 \pm 5.9 (11.9 - 31.4)	14.0 \pm 4.5 (6.9 - 23.6)

Procedure

Subjects were required to avoid strenuous exercise or consumption of alcohol for 24 hours preceding measurement, and to avoid consumption of food, tea or coffee for four hours before measurement. Measurement was made with subjects wearing light shorts and T-shirts, or underclothes. Body weight and height, biceps, triceps, subscapular, suprailiac and anterior thigh skinfold thicknesses, bioelectrical impedance and near infra-red interactance were measured as described in chapter 2. Near infra-red interactance measurements were also made at triceps, subscapular and suprailiac and anterior thigh sites.

The code for exercise level for use with the near infra-red equations for prediction of percent body fat, was assigned according to the reported average

physical activity level during the previous three months. Of the women, 6 were coded as "light", 15 "moderate", and 6 "heavy" activity level. The numbers of men in each category were 4, 12 and 11 respectively.

Standard Error of Measurement of Body Composition Techniques

All measurements were repeated on a group of 8 subjects, who returned to the laboratory within one week of the original measurement, at the same time of day. The standard errors of measurement of % fat by skinfold thickness, bioelectrical impedance and near infra-red interactance were found to be 0.61, 1.19 and 0.63 respectively.

Influence of strenuous exercise on body composition measurements

Subjects

Subjects were 10 men who were recruited at circuit training sessions at Loughborough University. They were aged 19 to 28 years (mean 22.4). Mean height was 1.76 ± 0.04 m, weight 72.7 ± 6.5 kg.

Procedure

Subjects reported to the laboratory before the exercise session having refrained from consuming alcoholic beverages and strenuous exercise for 24 hours and consuming food or drink for four hours. Measurements of weight, height, skinfold thickness at four standard sites, near infra-red interactance at biceps site and bioelectrical impedance were made as described above. All measurements were repeated when subjects returned to the laboratory after the exercise session, which lasted on average 53.3 minutes. All sites and electrode positions were marked, to ensure that differences in measurement site or electrode position would not contribute to any potential changes in estimates of body composition.

Results

Comparison of mean body composition by three techniques

The mean %fat, fat-free mass and fat mass by each technique for men and women are shown in Table 4.2. Mean % fat by skinfold thickness for women and men was 22.3 and 14.0 respectively.

Mean bioelectrical impedance estimates of fat content were generally found to be higher than those from skinfold thicknesses: up to 4.5 %fat higher in men and 3.5 % fat higher in women. The greatest overestimation was by the equations of Deurenberg et al. - BIA-D. For women all BIA predictions except those of Segal et al. (BIA-S2) were significantly different by paired t-test from those by skinfold thickness. In men the difference in means was only significant for predictions of fatness by equations of Fitness Concepts (BIA), and Deurenberg et al. (BIA-D). Mean values from the different equations were also in some cases significantly different from each other.

Percent fat by NIRI was significantly lower than that by SKF and BIA: means of 16.4 and 6.6 % fat for women and men indicate underestimation by 5.9 and 7.4 % fat respectively relative to skinfold thickness.

Table 4.2: Mean and one standard deviation of %fat, fat free mass, and fat mass of 27 male and 27 female young adults by skinfold thickness (SKF), bioelectrical impedance (BIA) and near infrared interactance (NIRI)

	Method	%fat		Fat-free mass (kg)		Fat mass (kg)	
		mean	SD	mean	SD	mean	SD
Women	SKF	22.3	4.2	45.1	4.4	13.2	3.8
	BIA	25.1 ***	5.9	43.4 **	4.7	14.9 **	4.9
	BIA-L	24.2 *	6.0	43.9 *	4.5	14.3 *	4.9
	BIA-S	24.0 **	2.3	44.2 *	5.0	14.0 *	2.8
	BIA-S2	22.9	3.0	44.8	4.2	13.5	3.3
	BIA-D	26.8 ***	3.8	42.4 ***	4.0	15.8 ***	4.0
	NIRI	16.4 ***	5.0	48.5 ***	4.9	9.8 ***	3.9
Men	SKF	14.0	4.5	60.6	6.0	10.0	3.7
	BIA	16.1 *	6.2	59.1 *	6.5	11.5 *	5.0
	BIA-L	15.0	6.2	59.8	6.4	10.8	5.0
	BIA-S	14.9	2.9	60.0	6.2	10.6	2.6
	BIA-S2	12.8	1.9	61.5	6.2	9.1	1.9
	BIA-D	17.5 ***	3.7	58.1 ***	5.0	12.5 ***	3.6
	NIRI	6.6 ***	5.9	65.8 ***	6.9	4.8 ***	4.6

Significance of difference from skinfolds: *P<0.05 **P<0.01 *** P<0.001

Correlation of bioelectrical impedance and near infra-red interactance measures of body composition with those by skinfold thickness

Statistics of the correlation of bioelectrical impedance with skinfold thickness are shown in Table 4.3. Correlation coefficients ranged from 0.62 to 0.82, standard error of estimate from 2.57 to 3.63 % fat. The agreement between the two methods was greater in women, as indicated by higher correlation coefficients and lower standard errors of estimate.

Standard errors of estimate of bioelectrical impedance were lower than many of those reported in the literature. This could be partly due to the relatively homogeneous nature of the sample. There was also very little difference between the equations used in the standard error of estimation.

Although near infra-red interactance underestimated mean fatness, the correlation of near infra-red interactance with skinfold thickness was better than that

of bioelectrical impedance, and standard errors of estimation were lower. These were about 1% body fat lower than those reported by Elia et al. (1990).

Table 4.3: Correlation and standard error of estimation of estimates of % fat, fat free mass, and fat mass by bioelectrical impedance (BIA) and near infra-red interactance (NIRI) with those from skinfold thickness

	SKF	% fat		Fat-free mass (kg)		Fat mass (kg)	
		r	SEE	r	SEE	r	SEE
Women n=27	BIA	0.76	2.81	0.87	2.25	0.88	1.83
	BIA-L	0.78	2.73	0.87	2.25	0.89	1.77
	BIA-S	0.74	2.92	0.95	1.43	0.92	1.49
	BIA-S2	0.81	2.57	0.94	1.48	0.93	1.47
	BIA-D	0.82	2.52	0.95	1.47	0.93	1.42
	NIRI	0.83	2.41	0.94	1.52	0.91	1.64
Men n=27	BIA	0.70	3.31	0.87	2.99	0.77	2.40
	BIA-L	0.71	3.27	0.87	2.99	0.78	2.37
	BIA-S	0.69	3.36	0.93	2.21	0.80	2.27
	BIA-S2	0.62	3.63	0.91	2.51	0.76	2.45
	BIA-D	0.68	3.37	0.92	2.38	0.79	2.32
	NIRI	0.73	3.14	0.91	2.60	0.77	2.40

Further analysis

As in the previous chapter, further analysis was carried out, because of the drawbacks of regression analysis. The analysis of Bland and Altman (1986) was performed: the difference between % fat by each technique and skinfold thickness was plotted against the mean of the technique and skinfold thickness. Mean difference between methods and standard deviation of this difference are shown in Table 4.4.

This analysis did show considerable differences between the various bioelectrical impedance equations. The greatest overestimation of fatness was from the equations of Deurenberg et al. (1991), however the standard deviation of the difference from skinfold thickness was one of the lowest. The greatest standard deviation was observed for the equation of Fitness Concepts, and that of Lukaski et al. (1985), which contained only the height squared/ impedance term.

Table 4.4: Mean and standard deviation of difference between estimates of body composition by bioelectrical impedance/near infrared interactance techniques and those by skinfold thickness

		% fat		Fat-free mass (kg)		Fat mass (kg)	
		mean	SD	mean	SD	mean	SD
Women n=27	BIA	2.85	3.92	-1.69	2.34	1.69	2.34
	BIA-L	1.90	3.83	-1.15	2.31	1.15	2.31
	BIA-S	1.69	1.83	-0.86	1.66	0.86	1.66
	BIA-S2	0.57	2.55	-0.31	1.46	0.31	1.46
	BIA-D	4.54	2.49	-2.64	1.45	2.64	1.45
	NIRI	-5.90	2.79	3.42	1.65	-3.42	1.65
Men n=27	BIA	2.15	4.47	-1.53	3.20	1.53	3.20
	BIA-L	1.07	4.39	-0.79	3.14	0.79	3.14
	BIA-S	0.91	3.30	-0.57	2.25	0.57	2.25
	BIA-S2	-1.15	3.68	0.89	2.56	-0.89	2.56
	BIA-D	3.51	3.35	-2.51	2.38	2.51	2.38
	NIRI	-7.40	4.01	5.21	2.94	-5.21	2.94

The correlation coefficient and slope of the regression line fitted to the above data are shown in Table 4.5. A highly significant slope was observed in predictions by the equations of Segal et al. (1985), with overestimation of fatness in lean subjects, and increasing underestimation with increasing fatness. This will decrease the range of the measurement and explain the low standard deviation of fatness observed by this technique in Table 4.2. This is despite the relatively good agreement observed in the mean, and correlation statistics. However it may be expected that in a sample with a greater number of subjects at the upper end of the range of fatness, a greater discrepancy would be observed: the slope of -0.66 in women indicating that an increase of 10% fat would result in an underestimation greater by 6.6 % fat.

Increasing underestimation of fatness with increasing levels of fatness was observed for men but not for women by the equations of Segal et al. (1988), which were sex-specific. The best agreement by the Bland and Altman analysis was observed for the equations of Deurenberg et al: (1991), which were associated with the greatest difference in means.

For NIRI the standard deviation of differences indicated the agreement with skinfold thickness to be intermediate between the various bioelectrical impedance values. There was a trend towards increasing overestimation with increasing fatness, but this was not significant. This is in contrast with the results from the previous

chapter, where there was a highly significant trend of increasing underestimation with increasing fatness.

Table 4.5: Relation of the difference between body composition method and skinfold thickness with the mean of the method and skinfold thickness: correlation coefficient (r) and slope

		% fat		Fat-free mass (kg)		Fat mass (kg)	
		r	slope	r	slope	r	slope
Women n=27	BIA	.48 *	0.39	.10	0.06	.47 *	0.26
	BIA-L	.50 **	0.39	.04	0.02	.49 **	0.27
	BIA-S	.69 ***	-0.66	.39 *	0.14	.66 ***	-0.34
	BIA-S2	.52	-0.39	.11	-0.04	.34	-0.14
	BIA-D	.19	-0.12	.27	-0.09	.12	0.04
	NIRI	.29	0.18	.28	0.10	.02	0.01
Men n=27	BIA	.41 *	0.37	.17	0.09	.44 *	0.34
	BIA-L	.41 *	0.36	.12	0.06	.44 *	0.34
	BIA-S	.53 **	-0.51	.09	0.04	.50 **	-0.38
	BIA-S2	.78 ***	-0.98	.07	0.03	.73 ***	-0.71
	BIA-D	.28	-0.25	.41 *	-0.18	.03	-0.02
	NIRI	.37	0.30	.32	0.15	.34	0.25

Significance of regression * P<0.05 ** P<0.01 *** P<0.001

Correlations of optical densities with body composition

The near infra-red interactance estimate of % body fat is derived from equations including not only interactance data, but also weight, height, sex, and activity level, so a question arises as to how much of the variance is explained by the optical density data. When optical density data was added to a regression containing other variables, the proportion of variance explained by the regression increased from 63 % to 86 %. However this increase in the amount of variance accounted for was largely due to the first of the variables entered, and the second contributed very little, as the two optical density measurements were found to be very highly correlated ($r=0.99$ for both men and women).

The correlation of optical densities with body composition from skinfold thickness is shown in table 4.6. Highest negative correlations (>0.79) were observed between optical density values at the biceps site with percent body fat by skinfold thickness, with correlation with absolute fat mass being slightly lower. The

correlation of just one optical density measurement with % fat by skinfold thickness was found to be similar to or greater than that of the near infra-red prediction of % fat, from equations also containing terms for weight, height, "activity level" and gender. The interactance data from the biceps site were found to be considerably better correlated with % fat than those from other sites.

Table 4.6

Correlation of optical density at five sites with body composition by skinfold thickness

		% fat		Fat-free mass (kg)		Fat mass (kg)	
		F	M	F	M	F	M
Biceps	Od1	-.83	-.80	-.19	.04	-.76	-.81
	Od2	-.85	-.79	-.15	.07	-.76	-.80
Triceps	Od1	-.41	-.68	-.01	.21	-.34	-.64
	Od2	-.58	-.69	-.08	.24	-.50	-.64
Subscapular	Od1	-.41	-.35	-.02	.33	-.36	-.28
	Od2	-.55	-.53	-.01	.19	-.47	-.49
Suprailiac	Od1	-.51	-.33	-.04	.09	-.44	-.31
	Od2	-.67	-.52	-.10	.02	-.60	-.49
Anterior Thigh	Od1	-.14	-.65	.03	.18	-.10	-.62
	Od2	-.37	-.69	.25	.13	-.21	-.66

In the previous chapter biceps skinfold was found to be better correlated with fatness by hydrostatic weighing than optical density measurements at the biceps site. To determine whether biceps skinfold was again better correlated with fatness, the correlations of biceps skinfold and optical densities at the biceps site with % fat from bioelectrical impedance were compared (Table 4.7). Bioelectrical impedance was used as the reference technique here, as biceps skinfold is used in the estimation of fatness from skinfold thickness, and so would be expected to be highly correlated.

Biceps skinfold was found to be better correlated with body fatness than optical densities in women but not in men. However, the correlations of the two techniques are similar, suggesting that optical densities at the biceps site are no better correlated with fatness than skinfold thickness at the biceps site.

The optical densities were correlated less well with body fatness from bioelectrical impedance than with body fatness from skinfold thickness (Table 4.6).

Table 4.7

Correlation of optical densities and skinfold thickness at biceps site with body fatness (bioelectrical impedance)

	Men	Women
Od1	-.64	-.67
Od2	-.64	-.70
SKF	.61	.73

The final step was to determine whether any combination of interactance data at different wavelengths and several sites, could improve the correlation with fatness, above that observed with measurement at a single wavelength at biceps site only. Table 4.8 shows the correlation of a selection of interactance data with body composition estimates by skinfold thickness.

Conway et al. (1984) found a ratio of measurements at two wavelengths to be a good predictor of fatness. However, in this study, the use of a ratio of two wavelengths was not found to produce higher correlations with fatness than the use of one wavelength alone.

The correlation of interactance measurements at a number of sites with fatness was in all cases lower than that of measurement at biceps site alone. So measurement at biceps site only seemed to be a better predictor of fatness than measurement at a number of sites, even though measurement at a number of sites would be expected to improve the correlation.

Table 4.8.

Correlation of near infra-red optical densities at biceps (B), triceps (TR), subscapular (SS), suprailiac (SI) and anterior thigh (TH) sites with body composition variables by bioelectrical impedance analysis

Site		% fat		fat-free mass (kg)		fat mass (kg)	
		F	M	F	M	F	M
B	Od1/Od2	.74	.36	-.06	-.26	.64	.33
TR	Od1/Od2	.14	.29	-.02	-.28	.17	.25
SS	Od1/Od2	.20	.44	-.23	.04	.18	.48
SI	Od1/Od2	.42	.22	.06	-.10	.40	.23
TH	Od1/Od2	.58	.24	-.07	.04	.53	.23
B&TR&SS&SI&TH	Od1	-.46	-.64	.05	.26	-.48	-.63
	Od2	-.58	-.67	-.18	.26	-.59	-.67
B&TR&SS&SI	Od1	-.49	-.66	-.07	-.27	-.52	-.66
	Od2	-.58	-.68	-.04	-.28	-.60	-.68
B&TR	Od1	-.61	-.67	-.10	.18	-.62	-.68
	Od2	-.67	-.67	-.07	.22	-.67	-.68
B&SS	Od1	-.59	-.63	-.07	.20	-.62	-.64
	Od2	-.66	-.67	-.04	.20	-.66	-.68
B&SI	Od1	-.59	-.68	-.09	.20	-.63	-.69
	Od2	-.66	-.68	-.04	.23	-.67	-.69
B&TH	Od1	-.63	-.63	-.11	.15	-.62	-.65
	Od2	-.71	-.66	-.07	.16	-.68	-.68
B&TR&SS	Od1	-.54	-.64	-.02	.23	-.56	-.64
	Od2	-.62	-.67	.05	.24	-.63	-.67
B&SS&SI	Od1	-.52	-.65	-.08	.25	-.56	-.65
	Od2	-.60	-.68	-.05	.25	-.62	-.68
B&SS&TH	Od1	-.54	-.60	-.11	.20	-.55	-.61
	Od2	-.65	-.66	-.07	.20	-.65	-.68
B&SI&TH	Od1	-.54	-.65	-.04	.21	-.56	-.66
	Od2	-.65	-.67	-.02	.22	-.64	-.68

Effect of exercise on estimation of body composition

The means of weight and % fat by each method before and after exercise are shown in table 4.9. A significant ($P < 0.001$) decrease in weight was observed, the mean weight loss being 0.62 kg (range 0.3 - 1.1 kg). A lower estimate of % fat after exercise was observed for each method. However for skinfold thickness and near infra-red interactance this difference was small and not significant. The bioelectrical impedance estimate of % fat was consistently lower after exercise, the mean decrease being 2.6 ± 1.6 % fat, with a maximum decrease of 5.1 % fat. The magnitude of this difference is considerably greater than the standard error of measurement of this technique (1.2 % fat).

Table 4.9

Mean, standard deviation and range of % fat of 10 men by skinfold thickness (SKF), bioelectrical impedance (BIA) and near infrared interactance (NIRI), before and after a circuit training session

	Pre-exercise	Post-exercise
SKF	12.1 ± 2.2 (9.9 - 17.3)	11.9 ± 2.4 (9.2 - 17.8)
BIA	11.1 ± 3.5 (5.9 - 17.1)	8.5 ± 3.9 *** (3.0 - 14.7)
NIRI	7.4 ± 2.9 (2.8 - 11.9)	6.9 ± 3.1 (0.5 - 11.0)

*** Significantly different from Pre-exercise $P < 0.001$

The mean impedance decreased from 432 to 424 Ω , a decrease of 1.8 %. This is less than that observed by Khaled et al. (1988), who found strenuous jogging for 1.5 to 2 hours to cause a decrease in weight of 1 to 2 kg, and a decrease in impedance of about 13 %. However a 5 minute period of bicycle ergometer work was found by Garby et al. (1990) to produce small and non significant changes.

Discussion

There was consistently better agreement between methods in women than in men, as observed in the previous chapter, However in this case the age of men and women was similar, as was body mass index, and the range of body composition. So the possible reasons for this are not apparent.

Bioelectrical impedance

Bioelectrical impedance was found to give higher mean estimates of fatness than skinfold thicknesses. This is consistent with the findings of Elia et al. (1990) who found % fat by bioelectrical impedance to be an average of 3 % fat higher than values from hydrostatic weighing, and 5 % fat higher than those from skinfold thickness but in contrast to those of deCossio et al. (1986) who found results for bioelectrical impedance to be lower than those from hydrostatic weighing and skinfold thicknesses. However different instruments were used by these two groups, so the discrepancy could be accounted for by the findings of Deurenberg et al. (1989), that different instruments used for estimation of body fatness from impedance produced different means.

The different sets of equations evaluated were found to produce significant differences in mean estimated fatness. The standard errors of estimation of the different bioelectrical impedance equations employed were found to be low, and remarkably similar. However the analysis of Bland and Altman (1986) revealed differences in performance of the different equations. Some showed a significant trend to increasing overestimation or underestimation at increasing levels of fatness, whilst this trend was considerably weaker for others.

Of the different equations employed, means of % fat from the Segal et al. (1985 and 1988 - BIA-S and BIA-S2) equations were found to be closer to those from skinfold thickness. These equations also seemed to agree best under regression analysis, with high correlation coefficients, and low standard errors of estimate. However from the Bland and Altman analysis it was apparent that this technique tended to increasingly underestimate fatness at higher levels of fatness. This explains the low standard deviation and range of % fat estimates by this technique. The subjects in this study were all in the lean to normal range of fatness, but if this bias persisted in a sample with a wider range of fatness, then the higher standard errors of estimation reported by Segal et al. (1985) may be explained.

Overestimation at high levels of fatness and underestimation at lower levels were also observed by Hodgdon and Fitzgerald (1987), who estimated % body fat from equations supplied by the manufacturer of their device. However they

postulated that the discrepancy between impedance and underwater weighing estimates of fatness at extremes of fatness could be due to violation of the assumption of constant density of fat-free mass used in the estimation of body fat from body density. However Gray et al. (1989) found an overestimation of fat-free mass and hence underestimation of % body fat from impedance predictions using the equations of Segal et al. (1988) in very obese subjects compared to underwater weighing.

Bioelectrical impedance estimates of body fatness were found to be significantly lower after strenuous exercise. One factor likely to be involved in the change in impedance produced by exercise is a change in body water content. Impedance is observed to decrease with presumed dehydration after exercise, and increase after ingestion of fluids. The decrease in impedance would cause an increase in estimates of fat-free mass, and so a decrease in estimates of fat content.

Exercise may also affect the skin temperature, contact resistance and cutaneous blood flow. Caton et al. (1988) found an increase in skin temperature from 24.1 ± 1.8 °C to 33.4 ± 1.4 °C caused by a change in ambient temperature from 14.4 to 35.0 °C to decrease impedance by 35 Ω , and estimates of % fat by 3 % fat. This change in skin temperature is probably considerably greater than that observed in this study, where a period of approximately 20 minutes elapsed between the end of the exercise period and the measurement of impedance, and so difference in skin temperature could probably not entirely explain the observed change in impedance. Beckett et al. (1991) found a decrease in skin blood flow of 36% to produce a 3.5 Ω change in impedance, which produced a change of only 0.4 % body fat. The contact resistance may also change with alterations in skin temperature, or as a result of sweating. However the four-electrode technique of measuring impedance should theoretically minimise the effect of contact impedance on body impedance measurements.

Another factor which may be involved is the redistribution of body fluids. As discussed in chapter 1, the impedance of the arm will make up a disproportionately large proportion of total impedance, because of its relatively low cross-sectional area. So exercise causing an increase in the proportion of body fluids in the arm could change the observed whole body impedance even without any change in the total quantity of body fluid.

Near infra-red interactance

Near infra-red interactance was again found to underestimate fatness, by 5.9 % fat in women, and 7.2 % fat in men. These values were intermediate between those of the middle-aged men and female athletes in the previous chapter. The underestimation was greater in both groups of men than in both groups of women.

Elia et al. (1990) found % fat from near infra-red interactance to be only about 1 lower than skinfold thickness, and 2.5 lower than densitometry.

The correlation of near infra-red estimates of fatness with those from skinfold thickness were found to be relatively high, 0.83 for women and 0.73 for men compared with that of 0.61 in middle aged men, but lower than that of 0.9 in female athletes. So the correlations appear to be consistently lower in men than women. This could not be explained in terms of the range, as the range of fatness observed in the middle-aged men was greater than that in any other group. However a poorer correlation in older subjects could be explained by changes in fat distribution with age. The proportion of fat is situated subcutaneously has been reported to be significantly inversely correlated with age (Enzi et al. 1986), but no allowance for age is made in the prediction equations.

In the younger men and women in this chapter, there was no evidence of increasing underestimation with increasing levels of fatness, as observed in the previous chapter. This might be explained by the fact that the comparison in this chapter was with skinfold thickness. In the previous chapter, some trend for increasing underestimation with increasing fatness was also observed with skinfold thickness relative to hydrostatic weighing. So using skinfold thickness as the reference technique might have obscured an effect which might have been observed if hydrostatic weighing had been used as the reference. However the trend for skinfold thickness was only observed in the middle-aged men and not the female athletes, and was considerably smaller than that for near infra-red interactance. So it seems unlikely that the change in the reference technique could have totally obscured the trend.

Optical densities at the biceps site were well correlated with body fatness in both the men and women in this study. This correlation was similar to that observed in female athletes in the previous chapter, and considerably better than that observed in the middle-aged men. Optical densities at the biceps site were found to be equally or better correlated with fatness than that at any other site or combination of sites. This is consistent with the findings of Conway and Norris (1985). It was also found to be as well correlated with fatness as the estimate of fatness from prediction equations containing a number of other variables in addition to optical density data. However a single measurement of skinfold thickness was found to be as well correlated with fatness by bioelectrical impedance as the optical density data. The prediction of body composition was not improved by employing a ratio of the two optical density measurements, or any combination of measurements at several sites. So optical densities do not appear to be a better predictor of body composition than measurement of skinfold thickness at a single site.

Conway and Norris (1985) reported good agreement of near infra-red interactance with hydrostatic weighing using a scanning spectrophotometer. This allowed a wide range of near infra-red wavelengths to be used. So it is possible that with better instrumentation, the accuracy of near infra-red interactance could be greatly improved.

Comparison of bioelectrical impedance, near infra-red interactance, and skinfold thickness

The standard errors of estimation between techniques observed here are mostly low. The reason for standard errors of estimation and correlation coefficients being low could be the relative homogeneity of this group, or the large influence outlying points may have on correlation statistics. However it is possible that bioelectrical impedance and near infra-red interactance are better correlated with skinfold thickness than with the fat content of the body itself. This seems particularly likely to be the case for near infra-red interactance, where the method depends on the logarithm of a measure dependent upon the quantity of subcutaneous fat. For bioelectrical impedance this explanation does not seem so likely, as in this case the measurement is proportional to the fat-free mass of the body, and the quantity of subcutaneous fat at any site will not affect the measurement. However, as discussed in chapter 1, the impedance of the arm makes up a high proportion of the total body impedance, and two of the four skinfold sites are also on the arm (although these are often the smallest of the four sites). So it could be argued that as both methods are dependent to a large extent upon the composition of the arm, there may be a higher correlation between these two methods than there would be if this were not so.

Practical considerations

The potential advantages of bioelectrical impedance and near infra-red interactance over skinfold thickness as "field" techniques are speed and ease of measurement; greater social acceptability; and less inter-observer variation and observer training.

In this study, near infra-red interactance was found to be the least time consuming technique, and skinfold thickness the most time-consuming. The near infra-red interactance and bioelectrical impedance instruments used both included a processor which allowed immediate calculation of body composition using pre-programmed equations, which may be an advantage in terms of time and convenience in some situations. The latter techniques did not generally require clothing to be changed, which may also reduce the time taken for measurement, and is more socially acceptable.

The degree of inter-observer variation may often be important, particularly in measurement of skinfold thicknesses. Womersley and Durnin (1973) reported maximum inter-observer differences in estimation of body composition from skinfold thickness between two experienced and one inexperienced investigators to amount to 5% body fat in men, and 6% in women.

Inter-observer variation in measurement of body composition by near infra-red interactance is likely to be due to factors such as location of site, pressure with which light wand is applied, or the degree of relaxation of the biceps brachii. Jebb et al. (1991) compared the inter-observer variability in assessment of body fatness by skinfold thickness and near infra-red interactance, and found the coefficients of variation to be 4.6 and 4.2 % respectively: similar but slightly greater for skinfold thickness. However, measurement was made by experienced observers. It seems likely that for inexperienced observers this value would increase, particularly for skinfold thickness.

Elsen et al. (1986) examined inter-observer variation in measurement of impedance and found that one observer obtained consistently higher impedance values even when skin marks were used to aid location of sites: average inter-observer difference was $4.8 \pm 2.8 \Omega$ with marks, $4.2 \pm 6.8 \Omega$ without. This would probably amount to a difference in the estimate of body composition of about 1 % body fat. The cause of this inter-observer variation could be the placement of electrodes, which was also examined by the authors. The displacement of electrodes by 1 cm at wrist or ankle was found to produce a 2.1% change in impedance, 2 cm displacement 4.1% change, which would result in an error in estimation of body composition in the order of 3 % body fat.

The standard error of measurement was greater for bioelectrical impedance than for other methods. Repeat measurements were made on separate days, so the observed error will include biological variation as well as measurement error. It is therefore possible that the larger standard error of measurement is due to greater biological variation, which may be brought about by changes in hydration state and ambient temperature. This is despite conditions being controlled in that subjects were required to restrain from consuming food or drink prior to measurement, from exercising for twelve hours before measurement, and consuming alcohol for twenty four hours before measurement, and measurements were made at the same time of day to minimise the effects of diurnal variation.

Factors which may alter the impedance, such as exercise, consumption of food or liquid, ambient temperature, and possibly disease which may alter hydration state, and oedema, are likely to be particularly important in field studies.

Changes in impedance induced by consumption of food or drink, or variations in hydration state during the menstrual cycle have generally been reported to be small. Gleichauf and Roe (1989) found significant changes in weight, impedance and fat-free mass during the menstrual cycle, but these changes were generally too small to be of biological significance. However they suggested that larger changes in weight during the menstrual cycle may be a confounding influence. Chumlea et al. (1987) found no significant association of difference between impedance and underwater weighing estimates of body composition with physiological variables such as time of day, interval since previous meal or drink, and changes during the menstrual cycle and oral contraceptive usage. Rising et al. (1991) found no significant effect of intake of 700 ml of fluid or consumption of breakfast on body composition determined by bioelectrical impedance. Elsen et al. (1986) found no significant difference between ingestion of 750 to 1000 ml of electrolyte solution and water, and no significant effect of extraction of 500 ml blood on impedance measurements. However Khaled et al. (1988) found ingestion of 1.2 to 1.8 litres of fluid low in electrolytes to increase impedance by approximately 15%, which may amount to a difference in the order of 10 % body fat.

The effect of exercise on measurements of impedance may decrease the convenience of the technique in some groups, such as athletes, or heavy manual workers, where inaccurate estimates of body composition may be obtained unless subjects are prevented from exercising before measurement. The changes in ambient temperature employed by Caton et al. (1988) were relatively large, but still within the range which may be observed in some laboratories, and greater extremes of temperature may be observed in field studies.

The controlling of the above mentioned conditions necessitates that body composition measurement must be prearranged, and may be intrusive into subjects' lifestyles. In this study, avoiding the consumption of alcohol before measurement was the condition which was most unpopular with some of the students, and in two cases appointments had to be re-arranged after this condition was broken. Some subjects also found the avoidance of food for four hours before measurement difficult! Some of the athletes who were considering the study on the influence of strenuous exercise on body composition measurements withdrew because they did not wish to alter their training schedules.

Conclusions

The controlling of the conditions discussed above may make bioelectrical impedance less suitable for field studies. The discrepancy of the reported accuracy of the technique may be due in part to the use of different prediction equations. In addition, factors such as ambient temperature, electrode placement, and the stringency with which confounding factors are controlled may play a part. This highlights the need for standardising the methodology, and validating the prediction equations used.

Near infra-red interactance measurements of body composition may be appropriate where accuracy of measurements is not imperative. Accuracy might be improved if appropriately validated equations or more accurate instrumentation were available. Its advantages are that the minimum of time and subject co-operation are required, it is socially acceptable, and requires little observer training.

Measurement of skinfold thickness may often be the most convenient technique in field studies. Its major disadvantage is the larger inter-observer variation than other techniques, and measurement may be less socially acceptable. However on the basis of the discussion above, it still seems to be likely to be the most accurate of the three techniques, so long as observers are well trained, and the inter-observer reliability is studied if necessary.

CHAPTER 5

Assessment of subcutaneous adipose tissue thickness: comparison of near Infra-red interactance and skinfold thickness with ultrasound

Introduction

In the previous chapter the ability of a measurement of near infra-red interactance at a single site to predict body composition was evaluated, and optical densities at the biceps site alone were found to be almost as good a predictor of total body composition as equations also including height, weight and activity level. The optical densities were found to be better correlated with estimates of body composition from skinfold thickness, than those from hydrostatic weighing or bioelectrical impedance.

The optical density is dependent upon the fat and water content of the site measured. The optical density is the logarithm of the inverse of interactance, which will be equivalent to the negative of the log of interactance. So if the interactance is dependent upon subcutaneous adipose tissue thickness (SCATT), this would account for negative correlations with a value of percent fat which is proportional to the logarithm of skinfold thicknesses.

To determine the relationship between the near infra-red interactance and SCATT, interactance measurements were compared with ultrasound measurements of SCATT. Ultrasound measurements of muscle thickness were also made, to allow determination of whether interactance also depends on muscle thickness.

According to the Futrex Research Manual (Futrex Inc), the absorbance for fat is greater for Od1 than Od2, whilst for water the absorbance is greater for Od2 than Od1. So both optical densities were recorded, to allow determination of whether a regression of the two optical densities could be produced to predict SCATT or muscle thickness. Conway and Norris (1987) found a ratio of two measurements to be correlated with fatness, so the ratio of the two was also assessed.

In previous chapters, skinfold thicknesses were found to be better associated with body fatness than interactance. They are measured in this chapter also, to determine whether this better agreement is due to better estimation of SCATT.

This comparison also allowed an evaluation of skinfold compressability. The major source of error in estimating SCATT from skinfold thickness is that calipers compress the fold of dermis and adipose tissue. Although the pressure applied by the skinfold calipers is constant (by convention 10 g/mm² - Edwards et al. 1955) the degree to which skinfolds are compressed (compressability) may vary. Skinfold compressability has been reported to vary according to age (Brozek and Kinzey

1960), sex and site (Lee and Ng 1965, Himes et al. 1979). Moreover significant individual differences in skinfold compressability have been observed by Himes et al. (1979). Values have been reported to range between 10 to 40 %. Such differences in skinfold compressability have important implications in assessment of body fatness and fat distribution by skinfold thickness. In this study the skinfold compressability was assessed, and site, sex, and individual differences evaluated.

A-scan ultrasound was used to provide an estimate of thickness of a single, uncompressed layer of adipose tissue. Ultrasound measurements of subcutaneous adipose tissue have been found to be highly correlated with SCATT by electrical conductivity (Booth et al. 1966); needle puncture measurements (Bullen et al. 1965) and soft tissue radiographs (Hawes et al. 1972), although difficulties may occur in identifying the fat muscle interface. The use of ultrasound also enabled measurement of the depth of the muscle-bone interface, and hence calculation of the muscle thickness at some sites. This allows evaluation of whether the optical density also depends on the quantity of muscle at the measurement site.

Measurements by all techniques in this chapter include the dermis as well as subcutaneous adipose tissue, as when ultrasound measurement was made the dermis adipose tissue interface was sometimes concealed within the main bang, and difficult to identify. It would also be difficult to correct optical density data for dermis thickness, so for ease of measurement, the dermis was included in all measurements. So when subcutaneous adipose tissue thickness is referred to in the text, this is in reality subcutaneous adipose tissue plus dermis thickness.

Methods

Subjects

Subjects were the 27 male and 27 female adults who volunteered for the study described in the previous chapter, whose characteristics are shown in table 4.1.

Procedure

Measurements of skinfold thickness, optical densities, and ultrasound measurements of the depth of the fat-muscle interface were made at the biceps, triceps, subscapular, suprailiac and anterior thigh sites, as described in chapter 2. Each site was marked with water soluble pen, to ensure that measurements by each technique were made at the same location.

Ultrasound measurements of muscle depth were made at biceps, triceps and anterior thigh sites. They were not made at subscapular and suprailiac sites, as it was not found to be possible to locate a muscle bone interface at these sites. Adipose tissue muscle interface depth was subtracted from muscle bone interface depth to yield muscle thickness.

Skinfold compressability

Percent compressability of skinfolds was calculated as:

$$\text{Compressability} = \frac{\{ \text{US} - (0.5 \times \text{SKF}) \} \times 100}{\text{US}}$$

Where US is the ultrasound measurement of adipose tissue muscle interface depth (mm) and SKF is the thickness of a double fold as measured with skinfold calipers (mm).

Standard error of measurement of skinfold thickness, ultrasound, and near infra-red interactance

The standard errors of measurement (Smeas) of skinfold thickness, ultrasound estimates of fat muscle and bone muscle interface depths and near infra-red interactance optical density values are shown in Table 5.1. Smeas for skinfold thickness was within or below the range of 0.5 to 1.0 mm reported by Lohman (1981). Smeas was lower for skinfold thickness than for ultrasound, consistent with results of Borkan et al. (1982) that intraobserver reliability was greater for skinfold thickness.

The standard error of measurement of the optical densities is difficult to compare directly. However the Smeas of Od1 and Od2 was approximately 50% and 40% respectively of the standard deviation of the sample, whilst the corresponding

values for skinfold thickness and ultrasound were approximately 15 % and 35 %. So the standard error of measurement of the interactance data seems to be greater relative to the sample variation than that of the other methods.

Table 5.1

Standard error of measurement of skinfold thickness, ultrasound depths, and near infra-red interactance

	Skinfold thickness (mm)	Ultrasound fat muscle interface depth (mm)	Ultrasound bone muscle interface depth (mm)	NIRI Optical density 940 nm	NIRI Optical density 950 nm
Biceps	0.54	0.92	2.45	0.0317	0.0261
Triceps	0.74	1.50	7.93	0.0327	0.0379
Subscapular	0.39	0.59	-	0.0252	0.0267
Suprailiac	0.86	1.18	-	0.0279	0.0230
Ant. Thigh	0.27	0.69	3.71	0.0231	0.0241

Results

Comparison of mean ultrasound and skinfold measures of subcutaneous adipose tissue

The mean, standard deviation and range of skinfold thickness and depth of adipose tissue muscle interface by ultrasound are shown in table 5.2 and figure 5.1. Skinfold thickness was smallest at the biceps site, and largest at anterior thigh for women, and suprailiac for men. Adipose tissue thickness by ultrasound was again smallest at biceps site, and largest at suprailiac for both men and women. Mean skinfold and ultrasound values were significantly higher in women than men at biceps, triceps, and anterior thigh sites ($P < 0.01$). Conversely, suprailiac skinfold was higher in men ($P < 0.05$).

Table 5.2

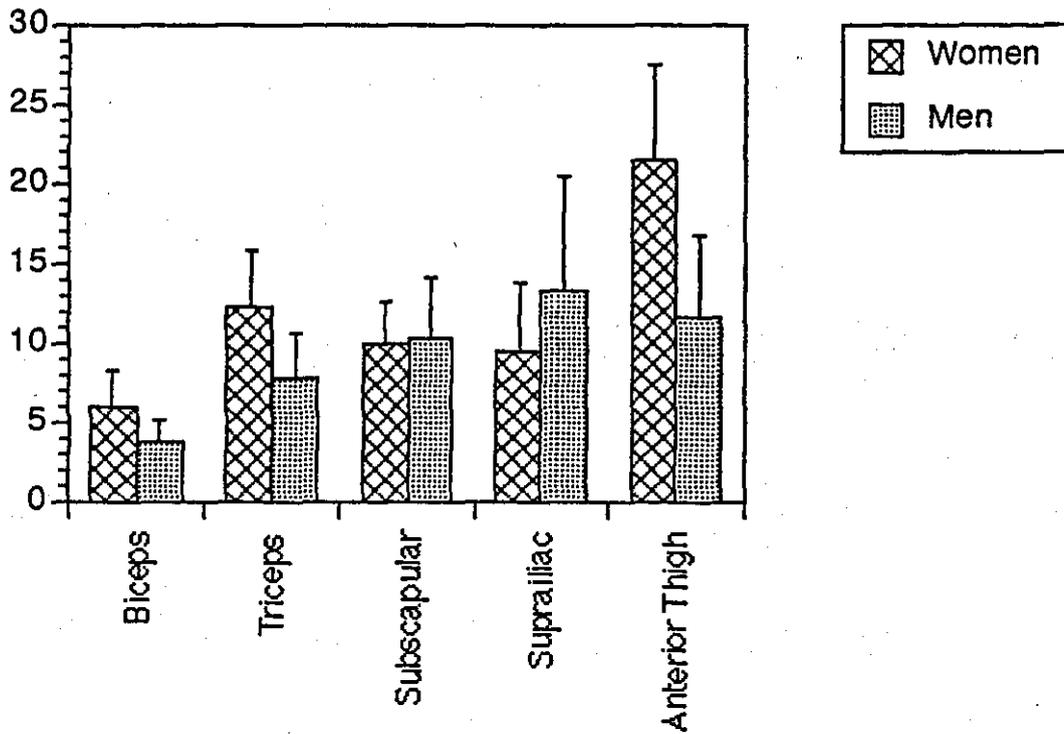
Mean, standard deviation, and range of skinfold thicknesses and ultrasound adipose tissue muscle interface depth

	Skinfold thickness (mm)		Ultrasound Subcutaneous adipose tissue thickness (mm)	
	Women	Men	Women	Men
Biceps	5.9 ± 2.4 (2.1 - 11.6)	3.8 ± 1.3*** (2.3 - 7.3)	4.1 ± 1.7 (2.1 - 8.4)	3.0 ± 1.0** (1.9 - 5.5)
Triceps	12.3 ± 3.5 (5.2 - 21.2)	7.8 ± 2.8*** (4.3 - 14.3)	8.2 ± 2.3 (4.0 - 13.3)	5.4 ± 2.1*** (2.1 - 11.8)
Subscapular	10.0 ± 2.6 (6.1 - 17.4)	10.3 ± 3.8 (6.0 - 20.4)	6.4 ± 1.4 (3.6 - 9.7)	6.0 ± 2.0 (2.1 - 10.7)
Suprailiac	9.5 ± 4.2 (3.6 - 23.6)	13.3 ± 7.1* (5.3 - 33.5)	11.5 ± 4.4 (2.7 - 19.8)	11.6 ± 5.4 (3.1 - 23.2)
Anterior Thigh	21.5 ± 6.0 (8.7 - 34.4)	11.6 ± 5.2*** (5.4 - 30.1)	10.3 ± 3.3 (4.6 - 15.8)	5.7 ± 2.4*** (2.9 - 13.9)

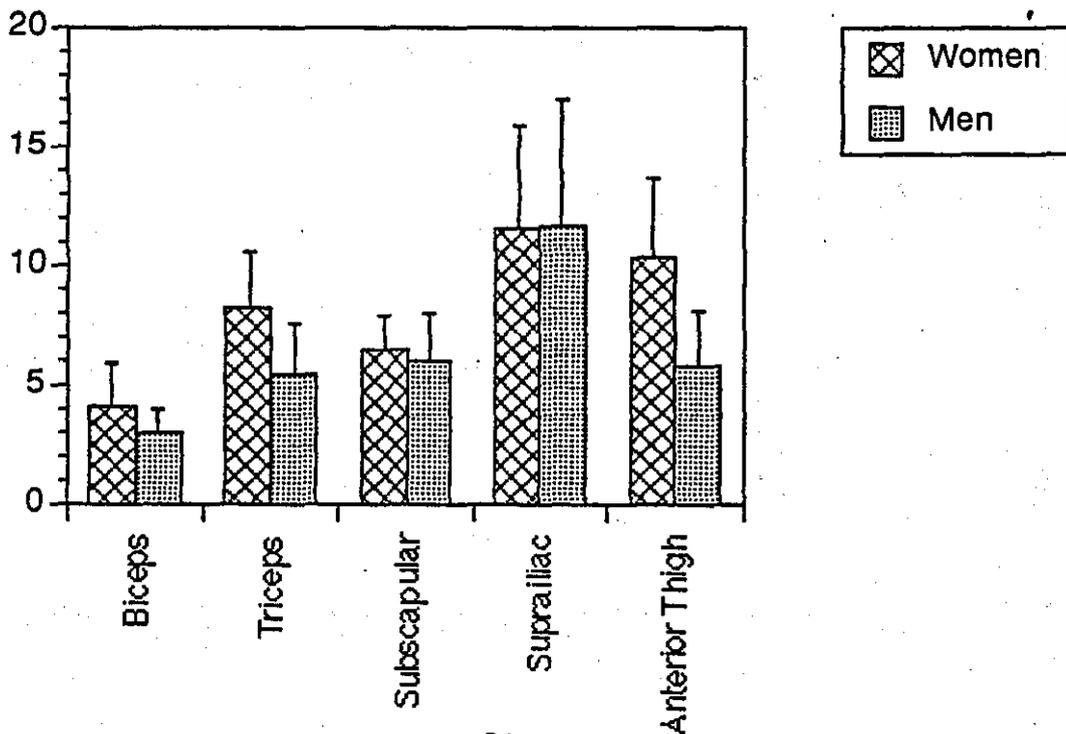
*** $P < 0.001$ ** $P < 0.01$ * $P < 0.05$ significantly different from women

Figure 5.1

Mean and one standard deviation of skinfold thicknesses at five sites (mm)



Mean and one standard deviation of subcutaneous adipose tissue thickness by ultrasound at five sites (mm)



Correlations of skinfold thicknesses with ultrasound subcutaneous adipose tissue thickness measurements

The correlation coefficients of skinfold thicknesses with adipose tissue muscle interface depths by ultrasound at each site are given in Table 5.3. These ranged from 0.56 to 0.92. Higher correlations were observed in men than women at the biceps site. The lowest correlation coefficients were observed at the biceps site for men, and at triceps and thigh sites for women. Lower correlations at these sites could be due to variation in compressability of skinfolds, or difficulties in interpretation of ultrasound echoes. At the biceps site in men the quantity of subcutaneous adipose tissue was sometimes very small, and it was found difficult to distinguish the echo corresponding to the dermis adipose tissue interface from the adipose tissue muscle interface. At the triceps site in women, multiple interfaces have been reported by Haymes et al. (1976), although these were more frequently observed at suprailiac site. The low correlation observed at the anterior thigh site could be due to difficulties in lifting the skinfold: a reliable measurement could not be obtained in four women.

Table 5.3

Correlation of skinfold thicknesses with ultrasound measurements of subcutaneous adipose tissue thickness

	Women	Men
Biceps	.83	.74
Triceps	.56	.89
Subscapular	.70	.80
Suprailiac	.78	.84
Anterior Thigh	.61	.92

Haymes et al. (1976) reported better correlation between skinfold and ultrasound measurement of subcutaneous adipose tissue in women than men. Correlation coefficients at suprailiac, subscapular and triceps sites were lower in men (0.59 - 0.76), but higher in women (0.85 - 0.92) than those observed in this study, as was that of 0.8 reported for both men and women by Bullen et al. (1965). The high correlation in women in these studies could be due to the large range of subcutaneous adipose tissue thickness: standard deviations being two to three times greater in the sample of Haymes et al. than those observed in this study. Fanelli et al.

(1984) and Borkan et al. (1982) observed correlation coefficients between the two methods to be lower than those in this study. Higher correlation coefficients were observed with direct measurement of cadavers by Lee and Ng (1965).

The agreement between the two methods in this study thus seems to generally be accordance with reported values in the literature. However, correlation coefficients are not ideal tools for comparisons between studies, as they are influenced by sample size and range, and the presence of outlying points. They were used here, because other statistics were generally not reported by the authors above.

Skinfold compressability

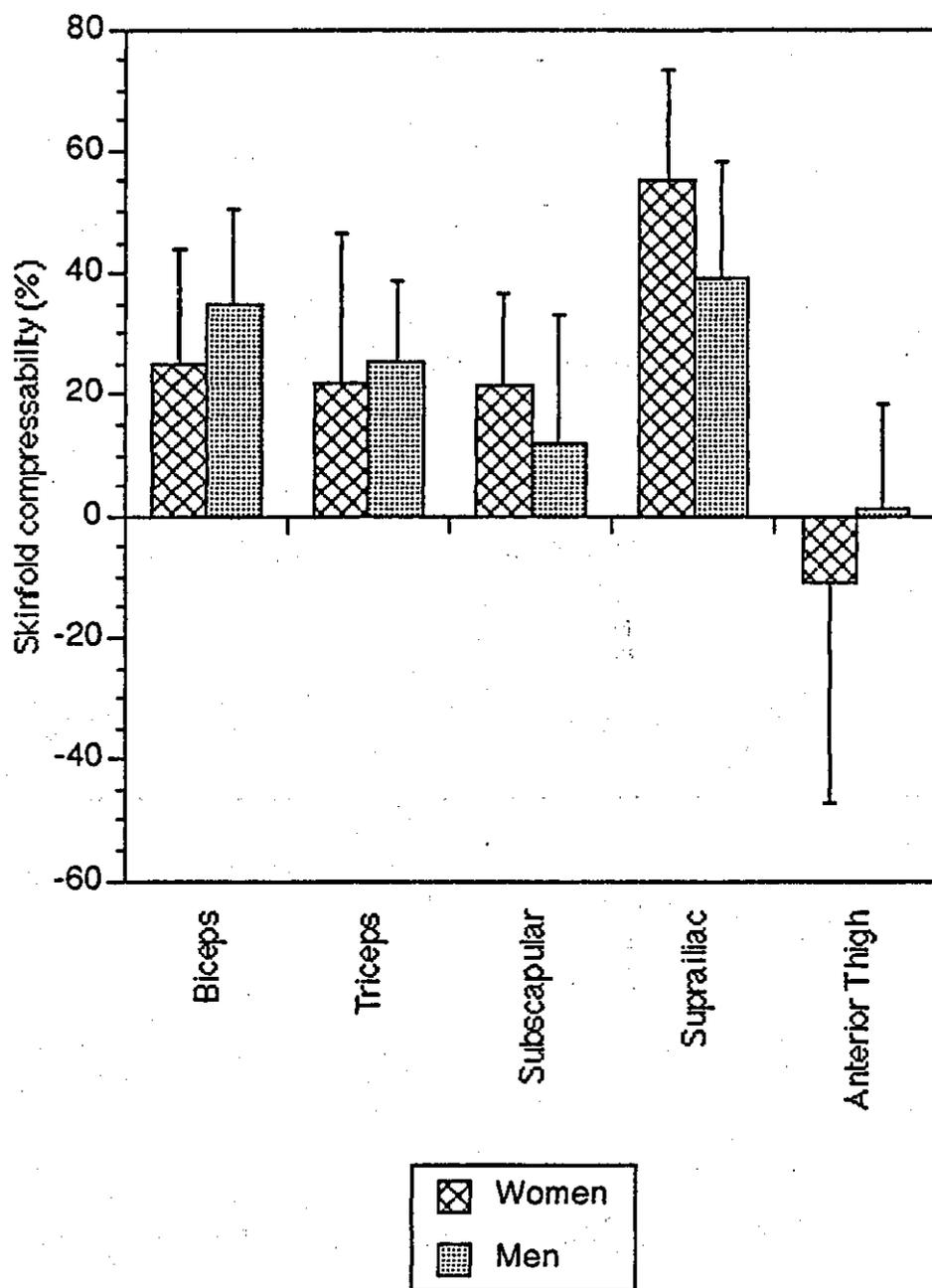
Percent compressability of skinfolds is shown in table 5.4, and Figure 5.2. Compressability was greatest at the suprailiac site, and least at anterior thigh site, for both men and women. At the anterior thigh site a negative compressability was observed, indicating that the skinfold thickness was more than twice the subcutaneous adipose tissue plus dermis thickness observed by ultrasound. Variation in compressability as assessed by standard deviation was considerably greater at this site in women than at any other site. Other sites where variation in compressability was greatest were triceps in women, and subscapular and suprailiac in men. These all tended to be sites where skinfold thickness was largest.

Table 5.4

Compressability of skinfolds (%): mean and standard deviation

	Women	Men
Biceps	24.8 ± 19.1	34.9 ± 15.6
Triceps	21.9 ± 24.8	25.2 ± 13.6
Subscapular	21.7 ± 15.0	11.9 ± 21.2
Suprailiac	55.4 ± 18.0	39.1 ± 19.0
Anterior Thigh	-11.0 ± 36.2	1.0 ± 17.5

Figure 5.2
Skinfold compressability of women and men at five sites
Mean and standard deviation



Two-factor analysis of variance revealed no significant difference between women and men in skinfold compressability, but a significant difference between sites ($P<0.001$) and a significant interaction between site and sex ($P=0.001$). (Table 5.5) No significant differences among individuals were observed (Table 5.6), as reported by Himes et al. (1979) for men.

Table 5.5
Analysis of variance of differences between sites and sex in skinfold compressability

Variable	df	Sum of squares	Mean Square	F
Sex (A)	1	1.1	1.1	0.003
Site (B)	4	74117.4	18529.4	43.488 ***
Interaction (AB)	4	8097.6	2024.4	4.751 **
Residual	252	107371.0	426.1	
Total	261	189587.1	726.4	

*** $P<0.001$ ** $P<0.01$

Table 5.6
Analysis of variance of differences between individuals in skinfold compressability

Variable	df	Sum of squares	Mean Square	F
Individual	53	22543.7	425.3	0.531
Residual	208	166535.8	800.7	
Total	261	189079.5		

Mean compressibilities at sites other than anterior thigh ranged from 20-55% for women, and 10-40% for men. These values agree with those of 10-40% observed by Fanelli et al. (1984), and 30% by Garn and Gorman (1956), both in young men. Compressibilities at the triceps site were lower than those observed by Bullen et al. (1965) in men and women of wide age range (33 and 39% respectively), but higher than the 16% observed by Brozek and Mori (1958) in middle aged men.

The negative compressability at the thigh site in women is due largely to two large skinfold values causing two extremely negative compressability values. If these two outliers are removed the compressability increases to approximately 0 %, and the

correlation of thigh skinfold with ultrasound values increases to 0.93. Whilst difficulties were experienced in measurement of thigh skinfold in women in some cases due to the tautness of the skin, and a reliable measurement could not be obtained for four women, there seemed to be no reason to exclude these two outlying cases where a repeatable measurement was obtained.

Near infra-red interactance

Mean optical density measurements at 940 and 950 nm (Od1 and Od2 respectively), and the ratio of these measurements, are shown in Table 5.7, and figure 5.3. Od2 was significantly higher than Od1 at all sites measured.

Table 5.7

Mean and standard deviation of optical densities at 940 and 950 nm

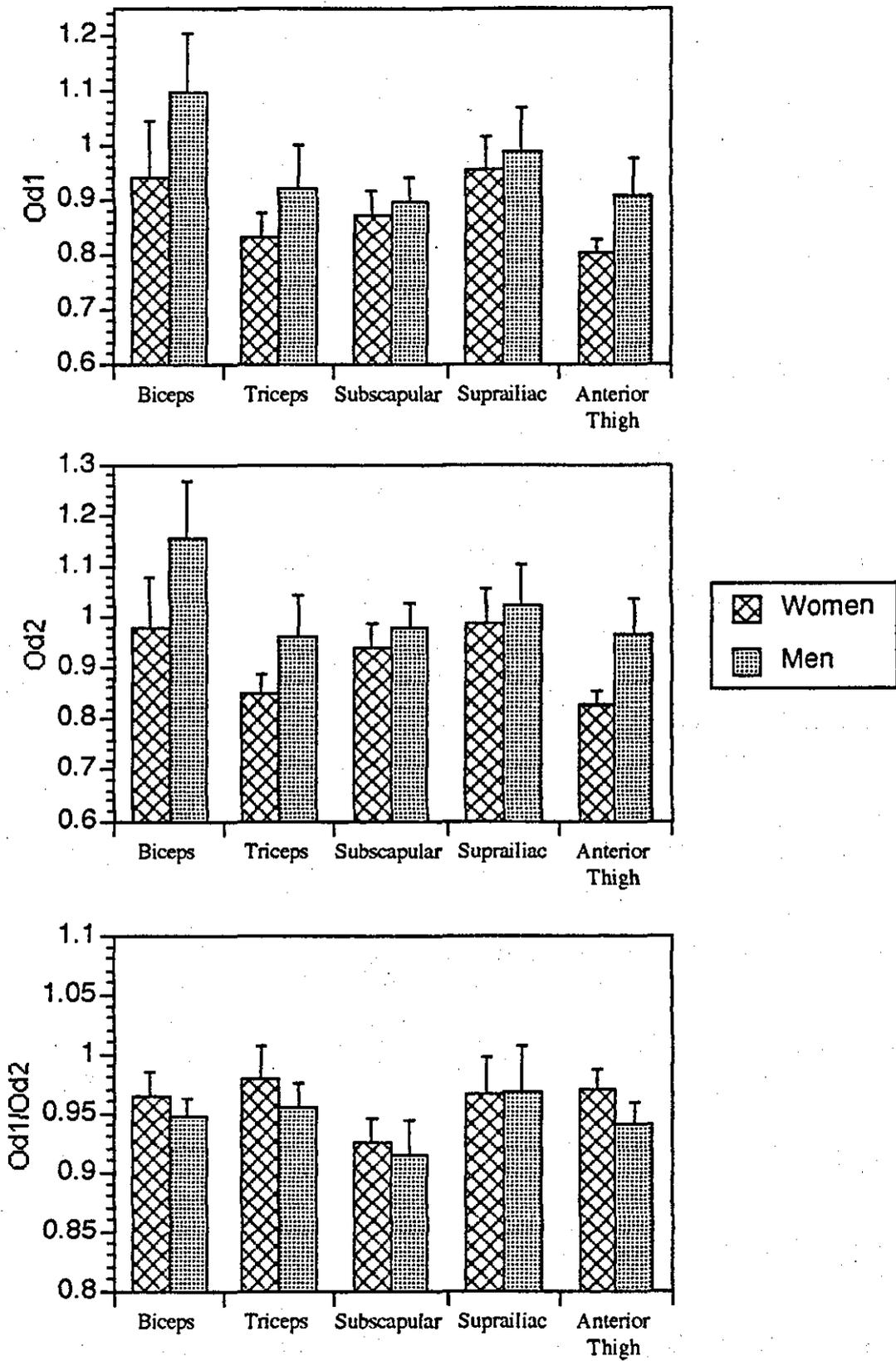
	Od 940		Od 950		Od1/Od2	
	Women	Men	Women	Men	Women	Men
Biceps	0.940 ± 0.103	1.094** ± 0.110	0.977 ± 0.103	1.156** ± 0.124	0.964 ± 0.020	0.947** ± 0.014
Triceps	0.830 ± 0.046	0.918** ± 0.082	0.849 ± 0.040	0.963** ± 0.094	0.979 ± 0.028	0.955** ± 0.020
Sub- scapular	0.870 ± 0.047	0.895* ± 0.045	0.940 ± 0.047	0.980** ± 0.055	0.925 ± 0.020	0.914 ± 0.030
Supra- iliac	0.955 ± 0.059	0.989* ± 0.080	0.989 ± 0.068	1.023 ± 0.085	0.966 ± 0.875	0.968 ± 0.038
Anterior Thigh	0.802 ± 0.023	0.908** ± 0.067	0.828 ± 0.026	0.966** ± 0.071	0.970 ± 0.016	0.940** ± 0.020

** P < 0.01 * P < 0.05 significantly different from women

Od1 was significantly higher for men than women at all sites measured. Od2 was significantly higher in men at all sites except suprailiac, and the ratio Od1/Od2 significantly lower in men at all sites except for subscapular and suprailiac. This is in general accordance with results for skinfold thickness and ultrasound, where values for men were also significantly lower than those for women at biceps, triceps and thigh sites.

Significant differences between sites were observed by analysis of variance. Optical densities (Od1 and Od2) were significantly higher at biceps than all other sites for men. For women values at triceps and thigh sites were significantly lower than those at biceps and suprailiac sites. The ratio of the two values (Od1/Od2) was lower at subscapular than all other sites (P < 0.05, Scheffe test).

Figure 5.3
Od1, Od2 and Od1/Od2 at five sites in men and women: mean and SD



The two optical densities were well correlated with each other ($r=0.85-0.99$, Table 5.8), especially at the biceps site. According to the manufacturers (Futrex Research Manual, Futrex Inc.) the difference between the two wavelengths is of primary concern in measurement of body composition. So it seems surprising that correlations of the two are so high, especially at the biceps site where measurement is recommended. With correlations of 0.99 between the two variables, very little extra information would be expected to be obtained from measurement of a second variable.

Table 5.8

Correlation of optical density at 940 nm with optical density at 950 nm

	Women	Men
Biceps	.99	.99
Triceps	.85	.98
Subscapular	.92	.86
Suprailiac	.87	.89
Ant. Thigh	.85	.96

To determine whether the use of both optical density variables could improve the prediction of SCATT, a stepwise regression was performed with SCATT (ultrasound) as independent, and Od1 and Od2 as dependent variables. This was repeated for each site for men and women. However, only one optical density variable was ever entered into the regression. This indicated no additional information on SCATT could be gained by using both optical density variables rather than just the one. This was probably because of the degree of inter-correlation of the two variables, as discussed above.

Correlation of optical densities with ultrasound subcutaneous adipose tissue thickness

Table 5.9 shows the correlation of near infra-red interactance data with subcutaneous adipose tissue plus dermis thickness by ultrasound. Ultrasound values were skewed, and so were logarithmically transformed: however correlation coefficients were similar to those from the untransformed data. Od1 and Od2 were better correlated with ultrasound than the ratio of the two measurements. In women the agreement was considerably better at the biceps than at any other site. For men, the correlation observed at the biceps site was better than that at suprailiac, thigh, and, for Od1 and Od1/Od2, suprailiac sites. The correlation at other sites except suprailiac was better than in women (0.51-0.69). However, even at sites where

correlation was reasonably good, coefficients were still lower than those between skinfold thickness and ultrasound.

Table 5.9

Correlation of near infra-red optical densities with ultrasound measurements of subcutaneous adipose tissue thickness

	Od1		Od2		Od1/Od2	
	Women	Men	Women	Men	Women	Men
Biceps	-.79	-.67	-.77	-.68	.56	.40
Triceps	-.21	-.67	-.35	-.69	.16	.42
Subscapular	-.17	-.54	-.24	-.68	.13	.27
Suprailiac	-.41	-.21	-.66	-.37	.55	.34
Ant. Thigh	-.23	-.51	-.04	-.57	.49	.22

Correlation of optical densities with skinfold thickness

Correlations of optical densities with skinfold thicknesses (Table 5.10) were considerably higher than those of optical densities with ultrasound, with coefficients at the biceps site being -0.86 to -0.90. The ratio Od1/Od2 again agreed worse with skinfold thickness, and correlations of Od2 were generally lower than those of Od1.

Correlation at the anterior thigh site in women was poorer than at other sites, as was observed on comparison with ultrasound: this could again be explained by the wide variation of skinfold compressability observed at this site.

The reason for the better correlation of optical densities with skinfold thickness than with ultrasound could be due to the larger standard error of estimate of ultrasound. However ultrasound measures only the distance of an interface: both skinfold thicknesses and optical densities may be influenced by differences of a qualitative nature, such as the protein or water content of the tissue, or quantity of underlying muscle.

Table 5.10**Correlation of near infra-red optical densities with skinfold thicknesses**

	Od1		Od2		Od1/Od2	
	Women	Men	Women	Men	Women	Men
Biceps	-.90	-.86	-.89	-.87	.77	.37
Triceps	-.63	-.75	-.44	-.72	.17	.46
Subscapular	-.60	-.56	-.53	-.36	.06	.43
Suprailiac	-.66	-.59	-.53	-.41	.38	.43
Ant. Thigh	-.32	-.59	-.10	-.49	.49	.44

Muscle-bone interface depth

Mean depth of the muscle-bone interface by ultrasound at the three sites for which this measurement was possible is shown in table 5.11. This measurement, which included dermis, subcutaneous adipose tissue and muscle compartments was found to be significantly greater in men than women ($P < 0.001$). Mean depth was similar at each site: 23.0 - 26.8 mm for women and 31.3 - 32.5 mm for men, although a large range was observed, especially at triceps and anterior thigh sites.

Table 5.11**Mean, standard deviation, and range of muscle-bone interface depths**

	Muscle-bone interface depth (mm)	
	Women	Men
Biceps	25.5 ± 3.7 (18.8-34.3)	32.5 ± 3.7 *** (26.3-40.5)
Triceps	23.0 ± 7.0 (11.2-38.2)	33.4 ± 9.1 *** (12.2-48.6)
Anterior Thigh	26.8 ± 7.1 (12.5-38.9)	31.3 ± 6.6 *** (20.5-46.0)

*** $P < 0.001$ ** $P < 0.01$ * $P < 0.05$ significantly different from women

Contribution of muscle thickness to optical densities

The possible contribution of the quantity of underlying muscle to optical density measurements was examined using a multiple regression of optical density with adipose tissue muscle interface depth and muscle thickness. Regression coefficients for the three sites for which measurement of muscle thickness was possible are shown in Table 5.12. Correlation coefficients were not much larger than correlation coefficients of optical density with ultrasound adipose tissue plus dermis thickness: so the addition of muscle thickness did not greatly improve agreement. The regression coefficient of the muscle term in the regression was only significant ($P < 0.05$) in one case: in the regression of Od2 at the biceps site for women. The muscle term thus seemed to contribute little towards the optical density value.

The near infra-red beam may not penetrate the entire depth of muscle. The manufacturers estimate the maximum depth of penetration of near infra-red in the human body to be 4 cm (Futrex Inc). To allow for incomplete penetration, all depths greater than 40 mm were recoded as 40 mm, and the regression repeated. However the regression was not improved. It therefore seems that interactance is dependent solely upon the subcutaneous adipose tissue thickness, and not upon the amount of underlying muscle.

Table 5.12

Correlation coefficients of multiple regression of fat depth and muscle depth by ultrasound with optical densities

	Od1		Od2		Od1/Od2	
	Women	Men	Women	Men	Women	Men
Biceps	.82	.72	.81	.73	.66	.45
Triceps	.25	.65	.45	.68	.22	.49
Ant. Thigh	.18	.44	.12	.52	.54	.29

Discussion

Agreement of skinfold thicknesses with subcutaneous adipose tissue thickness

The correlation between skinfold thickness and SCATT by ultrasound was found to be in accordance with literature values. Skinfold compressability was found to range from -11 to 55 %. Considerable differences between sites were observed. Despite considerable variation within each site, significant differences between individuals were not found. This would tend to endorse the use of skinfold thicknesses in the measurement of body fatness and fat distribution. If differences between individuals in skinfold thickness had been found (as reported by Himes et al. 1979), this would mean consistent underestimation of SCATT in individuals with more compressible skinfolds.

The effect of age on skinfold compressability was not examined in this study, because the age-range of subjects was relatively narrow. However Brozek and Kinzey (1960) reported significantly different compressability in different age groups, with compressability increasing with age. This has implications for the use of skinfold thicknesses in the assessment of body composition and fat distribution, as it means that skinfold thicknesses would underestimate SCATT in older subjects relative to younger ones. The problem has often been avoided in the use of skinfold thicknesses for assessment of body composition, by the use of age-specific equations, such as those of Durnin and Womersley (1974). However increases in compressability with age may affect the use of skinfold thicknesses for assessment of fat distribution.

Agreement of near infra-red interactance with subcutaneous adipose tissue thickness

Optical densities were generally better correlated with ultrasound subcutaneous adipose tissue thickness in men than women. However the agreement with skinfold thickness was much greater than that with ultrasound. This could be due to the higher standard error of measurement of ultrasound. However this could be an indication of skinfold thickness and near infra-red interactance being dependent upon differences of a qualitative nature, such as the hydration state of the tissue, or the protein content of the dermis. These would not be detected by ultrasound, which provides only information on the depth of a particular interface. Another explanation is that the skinfold thickness and interactance may be partly dependent upon the quantity or quality of underlying muscle. However this seems unlikely, as the addition of muscle thickness to a multiple regression of near infra-red interactance

with ultrasound subcutaneous adipose tissue thickness did not increase the agreement between the two. So it seems apparent that the interactance is dependent only upon subcutaneous adipose tissue: quantity and possibly to some degree other factors such as hydration state.

The ratio of the two optical densities was always found to agree less well with measurements by other techniques than either of the optical density measurements alone. Theoretically, the technique is dependent upon measurements at two wavelengths, with differing absorption coefficients for water and fat, so the use of both wavelengths should increase the information obtained. However, although the optical density at 950 nm was consistently higher than the optical density at 940 nm, measurements at the two sites were highly correlated, explaining the failure of a second optical density measurement to improve the agreement with subcutaneous adipose tissue. It is possible that with the two measurements being relatively close in the absorption spectrum (Figure 1.1), the absorption coefficients did not differ greatly enough to accurately describe the relative proportions of water and fat present. However the use of just one wavelength at the biceps site produced good correlations with ultrasound, and especially skinfold thickness, so the use of just one measurement is sufficient to provide information on subcutaneous fat. But skinfold thickness seems a better predictor of subcutaneous fat: when Tables 5.10 and 5.11 are compared, it is apparent that the correlation of skinfold thickness with ultrasound is greater than the correlation of near infra-red interactance with ultrasound.

The agreement of optical densities with other methods was generally better at the biceps site than at other sites. Possible reasons for this are the greater standard deviation of the measurement at the biceps site, indicating a wider range of measurements, which would be expected to increase the correlation coefficient, or differing dermis thicknesses at different sites affecting the interactance. This observation may explain the failure of inclusion of measurements at other sites in addition to data from the biceps site to improve the agreement with body fatness observed in the previous chapter.

Conclusions

Near infra-red interactance optical densities were correlated with SCATT assessed by other techniques, but not with muscle thickness. Optical densities at the two wavelengths measured were very highly correlated with each other, and no extra information was obtained from using both wavelengths rather than just one. Correlation with SCATT was better at the biceps site than the other sites measured, supporting the choice of this site for assessment of body composition by NIRI.

However, skinfold thicknesses were found to be a better predictor of SCATT than NIRI. No significant inter-individual differences in skinfold compressibility were observed, but there were significant differences between sites.

Chapter 6

The influence of brisk walking upon body composition and fat distribution of middle aged men, and associations with cardiovascular risk factors

Introduction

In this chapter a study of the influence of a year-long programme of brisk walking upon body composition, fat distribution and levels of cardiovascular risk factors in previously sedentary middle-aged men is described. This study was carried out in conjunction with the department of Physical Education and Sports Science at Loughborough University, where exercise testing and blood biochemistry were carried out. The influence of the walking programme on fitness and blood biochemistry is described in more detail by Stensel (in preparation).

This chapter concentrates on the influence of the walking programme on body composition and fat distribution, and the associations of any changes with changes in cardiovascular risk factors. The three initial aims were to examine the influence of a programme of brisk walking on body composition and fat distribution; to determine whether changes in body composition and fat distribution could be partly responsible for changes in other cardiovascular risk factors; and to examine the associations between body composition and fat distribution and other cardiovascular risk factors in the group as a whole.

Studies on the influence of exercise programmes on body composition have been somewhat inconclusive. Sex differences in the response to exercise have been reported (Bjorntorp 1989), so only studies involving men are considered here. Several studies have observed programmes of running or walking to decrease fatness of middle-aged men (e. g. Leon et al., 1979; Kukkonen et al., 1982; Wood et al., 1988), although energy intake decreased in most of these studies, suggesting that the change in fatness was not due to exercise alone. Hagan et al. (1986) observed very little change in weight in men who did not reduce energy intake. In this study, men were instructed not to make conscious changes to their diet, which was monitored and is described in the next chapter. If the men complied with this instruction, the effect of exercise alone on body composition may be examined.

The influence of exercise on fat distribution has only come under scrutiny more recently. A cross-sectional study has shown lower waist to hip ratios in subjects practicing vigorous activities on a regular basis (Tremblay et al, 1990). In addition, involvement in exercise programs (using cycle-ergometer training) has been shown to

preferentially decrease trunk fatness (Despres et al. 1985; 1988; Tremblay et al., 1988). Fat distribution was monitored in this study to determine the effect of brisk walking on fat distribution.

Body composition and fat distribution have been found to be associated with incidence of cardiovascular disease, and with blood lipid levels, as described in chapter 1. As such, they may be a confounding influence when examining the effect of exercise on blood lipids. In addition, Wood et al. (1988) suggested that exercise induced changes in cardiovascular risk factors may be mediated by changes in body composition. So one aim of this study is to determine whether any changes in blood lipids could be due to changes in body composition or fat distribution, rather than exercise, and to determine the relative influences of these factors.

The data also provide an opportunity for both cross-sectional and longitudinal examinations of the associations between blood lipid levels and body composition and fat distribution.

Methods

Subject recruitment

Subjects were recruited by a number of means. Advertisements were placed in newspapers. The study was described on radio. Leaflets were distributed in university departments and doctor's and dentist's surgeries, and during a presentation at a health promotion day at Loughborough Medical Centre (Appendix II).

Meetings were then held at Loughborough University in which the study was described, written descriptions of the procedures used were distributed (Appendix III), and men were able to ask any questions they had. Individual meetings were also arranged if required. Men who expressed an interest in joining the study were screened before acceptance into the study.

Criteria for acceptance into the study

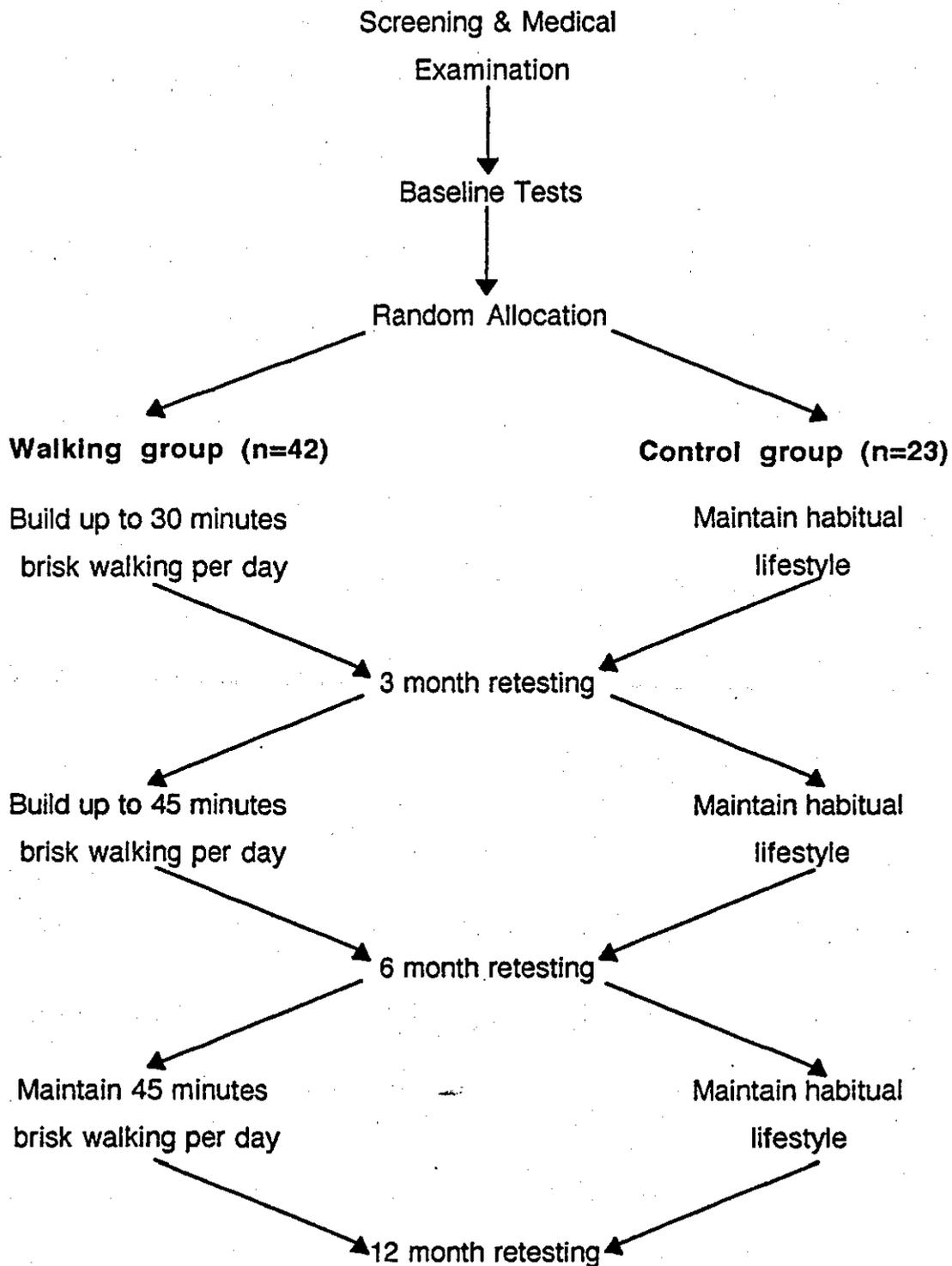
Men were required to fulfil the following criteria to be accepted into the study:

1. Aged 42 to 59 years, as in this age range total cholesterol levels have been reported not to vary with age (Mann et al. 1988).
2. Sedentary, i.e. not employed in a strenuous job or currently involved in a regular physical activity program.
3. Non-smokers or light cigar smokers.
4. Healthy and free of known cardiovascular disease.
5. Plasma total cholesterol less than 6.7 mmol/l.
6. Blood pressure less than 160/95 mmHg.
7. Prepared to be randomly assigned into either an exercise or control group.
8. Prepared to not receive any feedback until the end of the study, to ensure that this could not induce any changes in lifestyle.

Screening

Each man was examined by doctors at Loughborough University Medical Centre. On an initial visit, smoking habits and physical activity level were assessed by means of questionnaires (Stensel, in preparation). Plasma total cholesterol and blood pressure were measured as described in chapter 2. Men who fulfilled all the above criteria were accepted into the study. In addition, men with slightly elevated plasma total cholesterol, but with normal blood pressure, and men with slightly elevated blood pressure but normal total cholesterol were accepted, as the effect of risk factors has been reported to be interactive (Tunstall-Pedoe & Smith, 1990).

Figure 6.1
Outline of Brisk Walking Study



Outline of study

The protocol of the study is shown diagrammatically in Figure 6.1. After screening and acceptance into the study, a baseline set of tests was conducted. The men were then randomly assigned on a 2:1 basis into either walking (n=41) or control (n=22) groups. The number in the walking group was greater to allow an examination of whether a dose-response relationship existed. The men in the walking group took up the exercise programme described below. All men were asked not to make any other changes to their diet or habitual lifestyle. Testing was repeated at 3, 6 and 12 months after the start of the walking programme.

To facilitate testing of this large group of subjects, the group was split into three cohorts, each containing approximately two-thirds walkers and one third controls. The timescale of testing these three cohorts is shown in Table 6.1.

Table 6.1

Timescale of testing the three cohorts in the brisk walking study

Month	Baseline	3 months	6 months	12 months
April	Cohort 1			
May	Cohort 2			
June				
July	Cohort 3	Cohort 1		
August		Cohort 2		
September				
October		Cohort 3	Cohort 1	
November			Cohort 2	
December				
January			Cohort 3	
February				
March				
April				Cohort 1
May				Cohort 2
June				
July				Cohort 3

The study was approved by the University Ethical Committee (reference EAC 88:M4/21.2).

The Walking programme

The walking group were asked to take up brisk walking, which was described as faster than normal pace, but a rate which could be maintained for 30-45 minutes, each walk being continuous and at least 20 minutes long.

Fortnightly targets were set which increased progressively to 420 minutes in the first three months (an average of 30 minutes per day). Targets were then progressively increased to 630 minutes by the end of six months (an average of 45 minutes per day). This level was maintained for the rest of the year. These targets were set to ensure that men were increasing their energy expenditure by more than the 5 MJ per week, reported by Haskell (1986) to be associated with beneficial changes in HDL-C levels.

Fortnightly targets were set to allow men to split up the walking to accommodate it into their lifestyles. The walking could be split up however was most convenient, so it was not necessary to walk every day: missed days could be made up as convenient. However each walk was required to be at least 20 minutes long, and it was stipulated that at least five separate walks per week must be taken from 3 months onwards. The walking was required to be in addition to any habitual walking or other exercise.

The control group were required to maintain their habitual level of exercise.

Test battery

Testing was performed at baseline, and at 3, 6 and 12 months after the start of the walking programme. However, for reasons of time or expense, not all tests were performed at each time interval. The tests performed are described below. The methods used for each test are described in chapter 2.

Body Composition

Body composition was assessed by hydrostatic weighing at baseline and 12 months. However because of the time and technical assistance required for this measurement, it was not performed at 3 and 6 months. Changes in body composition during the year were assessed from measurements of skinfold thicknesses at baseline, 3, 6 and 12 months, as measurement of skinfold thickness was found to probably be the best rapid technique for assessment of body composition in previous chapters.

Fat distribution

Fat distribution was assessed from measurements of body circumferences and skinfold thicknesses. The influence of the walking programme on five body circumferences and 8 skinfold thicknesses was examined. Various ratios of these measurements were also employed, chosen as they have been reported to characterise

central fatness, or be associated with risk of cardiovascular disease. The ratios chosen are summarised in Table 6.2.

A principal components analysis of skinfold thicknesses at eight sites was also performed. Factor scores throughout the year were compared, to summarise changes in skinfold thicknesses.

Table 6.2

Indices of fat distribution used in brisk walking study

Ratio	Abbreviation	Reference
Waist to hip circumferences	WHR	Larsson et al. (1984)
Subscapular to thigh skinfolds	ss/th	Mueller et al.(1984)
Triceps + subscapular to subscapular skinfolds	tr+ss/ss	Kaplowitz et al. (1987)

Dietary analysis

Dietary intakes were assessed at baseline, 6 and 12 months. Measurement was not made at 3 months, to decrease the commitment for subjects. Methods used are described in chapter 7.

Exercise Testing

A submaximal incremental treadmill test was performed at baseline only, to prescribe individual levels for subsequent tests. The grade lactate test, endurance walk and one mile track walk were performed at baseline, 3 months, 6 months and 12 months. Each exercise test was performed on a separate visit to the laboratory.

Blood pressure and blood biochemistry

Fasting total cholesterol, triglyceride, HDL-C, LDL-C, VLDL-C, Apo A, ApoB, haemoglobin and haematocrit levels were determined at baseline, 3, 6 and 12 months, as was resting blood pressure. Lipoprotein(a) was only measured at baseline and 12 months, for reasons of expense.

Coherence

About 100 men expressed an interest in the study, and attended the meetings held. The great majority of these had responded to newspaper or radio advertisements. Of these, some found the commitment involved too great, or were unhappy about one or more of the procedures involved, and withdrew. Two men also withdrew because they were unwilling to accept randomisation, despite the fact that to make the randomisation

more acceptable, an opportunity to participate in the walking programme in the second year of the study was offered to all subjects who were in the control group for the first year. Also, the walking group was twice the size of the control group, which would increase the likelihood of any subject being chosen for the walking group.

79 men continued their interest, and attended screening appointments. 6 were rejected because of ill health or high blood pressure or cholesterol levels. 73 men started the study, although one dropped out when he learned he was in the control group. Of the remaining 72 men, 48 were randomly assigned to the walking group, and 24 to the control group.

Of these 72 men, 7 withdrew during the course of the study: 6 walkers and 1 control. The work commitments of two of the walkers increased, so they found they no longer had the time to continue in the study. The remaining 5 withdrew as a result of ill health. 65 men completed the study. The characteristics of these men are shown in Table 6.3. There were no significant differences between walkers and controls in any of these characteristics.

Table 6.3

Characteristics of Walkers and Controls before walking study

	Walkers n=42	Controls n=23
Age (years)	50.3 ± 5.4 (42.4 - 59.4)	51.6 ± 5.0 (42.6 - 60.6)
Weight (kg)	79.1 ± 9.8 (60.0 - 102.3)	78.2 ± 11.9 (49.2 - 113.9)
Height (m)	1.77 ± 0.07 (1.59 - 1.94)	1.77 ± 0.06 (1.69 - 1.90)
Body Mass Index (WT/HT ²)	25.3 ± 2.6 (19.7-31.3)	24.9 ± 3.1 (17.3 - 33.5)

Compliance

No structured exercise sessions were held in this study: the walking programme was designed to be fitted into subjects' lifestyles. The amount of walking performed was monitored by means of training diaries completed by the walkers. These consisted of sheets on which the amount of walking performed each day was recorded. Each sheet covered one fortnight, so men could compare the amount of walking during that fortnight with the target levels. These sheets were sent to the university monthly. It was stressed to the men that truthful recording was more important than meeting the targets.

The amount of walking reported in each quarter of the study is shown in Table 6.4. Over the year as a whole, the average reported time spent walking was 27.2 ± 9.2 minutes per day (range 10.9 to 46.6).

Table 6.4
Reported time spent walking during each quarter of walking study
(minutes per day)

Time	Mean	Standard deviation
Months 0-3	23.5	9.1
Months 3-6	27.4	10.3
Months 6-9	30.3	11.0
Months 9-12	30.3	11.0

Results

Influence of walking programme on maximal oxygen uptake and oxygen consumption at reference blood lactate concentration

The maximal oxygen uptake and oxygen consumption at 2mmol/l blood lactate are shown in Table 6.5. Analysis of variance revealed significant changes through the year ($P < 0.001$) and significant difference in response between walkers and controls ($P < 0.01$) for both variables.

Table 6.5

Maximal oxygen uptake and oxygen consumption at 2mmol/l blood lactate of walkers and controls at baseline and 3, 6 and 12 months after the start of the walking programme: mean \pm standard deviation

	Month	Walkers	Controls
Maximal oxygen uptake (ml/kg/min)	0	35.9 \pm 4.6	35.2 \pm 5.2
	3	36.8 \pm 4.9	35.4 \pm 5.3
	6	37.0 \pm 5.7	33.6 \pm 5.2
	12	35.0 \pm 4.9	32.2 \pm 4.6
Oxygen uptake at 2 mmol/l lactate	0	21.5 \pm 3.3	20.3 \pm 2.5
	3	24.0 \pm 4.0	21.5 \pm 3.5
	6	24.7 \pm 3.4	20.4 \pm 3.9
	12	22.9 \pm 3.6	18.8 \pm 3.3

Predicted maximal oxygen uptake increased during the first six months in walkers, whilst decreasing in controls. In the final six months, a decrease in both groups was observed. The net effect was a decrease in the controls, and a smaller decrease in walkers. So it seems that participation in the walking programme may have prevented the decrease in maximal oxygen uptake which might otherwise have occurred.

The decrease in maximal oxygen uptake was large, especially in the control group. However, methodological factors such as the use of sub-maximal tests, and small sample size may contribute to this decrease.

Oxygen uptake at 2 mmol/l lactate increased for the first six months in walkers, with a decrease in the final six months. In controls there was an increase in the first three months, followed by a decrease for the final 9 months. The net effect was an increase in walkers and a decrease in controls.

The different responses of walkers and controls firstly confirm the compliance of the walking group, and secondly suggest that the walking was sufficient to bring about some physiological change, or at least to prevent a progressive decline.

Influence of walking programme on body composition and fat distribution

Body weight

The weight of walkers and controls before and at 3, 6 and 12 months after the start of the walking programme is shown in Table 6.6. During the year, the weight of the walkers decreased by an average of 0.2 kg, with the greatest loss in the first three months, and then a progressive increase until the end of the year. The controls also showed a mean loss in weight during the first three months, and an increase until the end of the year, resulting in a mean increase in weight over the year of 0.7 kg.

Table 6.6

Weight of walkers and controls before and at 3, 6 and 12 months after the start of the walking programme (Mean and standard deviation)

	Walkers		Controls	
	Mean	SD	Mean	SD
Baseline	79.1	9.8	78.2	11.9
3 months	78.4	9.5	77.6	11.9
6 months	79.1	9.8	78.8	11.8
12 months	78.9	9.5	78.9	12.8

An analysis of variance was performed to determine whether the response of the walkers (i. e. decrease in weight) differed significantly from the response of the controls (i. e. increase in weight). This analysis of variance is shown in Table 6.7.

Table 6.7

Analysis of variance with repeated measures of body weight of walkers and controls

Variable	df	Sum of squares	Mean Square	F
Group: walker/control (A)	1	71.4	71.4	0.2
Subjects within groups	61	26908.8	441.1	
Repeated measure (B)	3	15.3	5.1	3.2 *
Interaction (AB)	3	7.6	2.5	1.6
BxSubjects within groups	183	292.6	1.6	

* P < 0.05

There was no significant difference between the weight of walkers and controls. The interaction term of the analysis of variance (corresponding to the difference in response between walkers and controls) was also not significant, indicating that the slight decrease in weight of the walkers was not significantly different from the slight increase in the weight of the controls. However the repeated measures term was significant, indicating that the weight of both walkers and controls changed during the year. Post hoc Scheffe tests revealed that the weight at three months was significantly different than weight at other times.

Body composition

Body composition of walkers and controls during the walking programme is shown in Table 6.8. The % fat of both walkers and controls was lower at the end of the year than the beginning of the year. This decrease was greater in the walkers than the controls. Changes during the year can be examined from measurements of skinfold thickness. As with body weight, a trend was observed of a decrease during the first three months, and then a progressive increase until the end of the year.

Table 6.8

Percentage body fat, fat mass and fat-free mass of walkers and controls before and at 3, 6 and 12 months after the start of the walking programme, by hydrostatic weighing (HW) and skinfold thickness (SKF): Mean \pm standard deviation

		Walkers		Controls	
		HW	SKF	HW	SKF
% fat	Baseline	28.7 \pm 5.3	25.3 \pm 4.2	29.5 \pm 7.2	25.4 \pm 5.4
	3 months	-	23.7 \pm 4.1	-	24.2 \pm 5.2
	6 months	-	24.0 \pm 4.3	-	24.6 \pm 5.5
	12 months	27.6 \pm 4.8	24.9 \pm 4.5	29.3 \pm 7.4	25.1 \pm 5.1
fat mass (kg)	Baseline	22.9 \pm 5.8	20.3 \pm 5.1	23.8 \pm 8.4	20.4 \pm 7.3
	3 months	-	18.8 \pm 4.8	-	19.2 \pm 7.2
	6 months	-	19.2 \pm 5.0	-	19.9 \pm 7.3
	12 months	22.0 \pm 5.2	19.9 \pm 5.1	23.9 \pm 8.9	20.4 \pm 7.4
fat-free mass (kg)	Baseline	56.1 \pm 7.0	58.8 \pm 6.0	54.6 \pm 7.9	57.8 \pm 6.0
	3 months	-	59.6 \pm 6.1	-	58.3 \pm 6.0
	6 months	-	59.9 \pm 6.2	-	58.9 \pm 6.0
	12 months	56.7 \pm 6.8	59.0 \pm 6.0	55.3 \pm 8.2	58.6 \pm 6.5

Analysis of variance revealed the same trend for % fat as observed for weight: no significant difference between groups; or significant difference in response between groups, but significant changes during the year ($P < 0.05$). Scheffe tests were performed, which showed that means at 3 months were significantly lower than those at baseline and 12 months, and means at 6 months lower than baseline.

Significant changes during the year in mean fat mass and fat-free mass were observed ($P < 0.05$). The magnitude of this trend was again greater in walkers, but not significantly so. Mean fat mass decreased in the first three months, and increased for the rest of the year, whilst mean fat-free mass increased during the first six months and decreased in the final six months. The weight change over the year was due entirely to an increase in fat-free mass in controls, whilst in walkers, mean fat-free mass increased, whilst mean fat mass decreased, resulting in a net loss of weight.

Seasonality of changes in weight and body composition

The changes in weight and body composition during the year observed above could be due to involvement in the study, which may have made subjects more aware of their diet and health, inducing temporary changes in lifestyle. Another possibility is that these changes are due to a seasonal effect. The baseline set of tests were carried out between April and June, so a decrease in weight occurred in the spring and summer, and a progressive increase during the rest of the year.

To determine whether changes during the year were more likely to be due to involvement in the study or a seasonal effect, the weight and % fat of 12 men who continued in the study for a second year were examined. The hypothesis was that if the same trend was observed, the changes would be likely to be due to a seasonal effect, whilst if no evidence of this trend was observed, the changes in the first year would be likely to be due to temporary lifestyle changes after joining the walking study.

The weight and % fat of this group of 12 men during the two years of study are shown in Table 6.9.

Table 6.9

Weight and % fat of 12 men during a second year of involvement in the brisk walking study: mean and standard deviation

	Weight (kg)	% fat
Baseline	80.0 ± 14.8	25.9 ± 6.7
3 months	80.3 ± 15.0	24.9 ± 6.2
6 months	80.7 ± 14.9	24.1 ± 6.6
12 months	80.7 ± 16.3	25.5 ± 6.1
15 months	80.0 ± 15.9	25.3 ± 6.7
18 months	80.9 ± 16.1	25.7 ± 6.3
24 months	80.6 ± 16.3	26.3 ± 7.4

Changes in mean body weight in the second year were similar in direction and magnitude to those observed in the first year. Changes in % body fat during the first six months of the second year were smaller than those in the first year.

However, in this sub-group of subjects, changes in body weight and % fat were not significant in either the first or second year. The lack of significance of changes during the year could be due to the small number of subjects studied. As some evidence of a seasonal trend was again observed, at least in weight, a seasonal effect can not be discounted, which may have been significant if a larger number of subjects had been studied. However these changes may have occurred purely by chance, so it is impossible to determine which of the two possible explanations is more likely, or whether a combination of the two effects was the cause of the changes.

Body circumferences

Changes in body circumferences during the year are shown in figure 6.2. Changes in body circumference measures were small: less than 1 cm. On analysis of variance, there was no significant difference in response between walkers and controls. However for all circumferences except arm and thigh, there were significant changes through the year.

Changes during the year in waist hip circumference are shown in table 6.10. Changes through the year in waist-hip circumference were not significant. The changes in waist circumference and waist hip ratio followed a similar trend to that observed for weight and % fat: a decrease during the first three months, and an increase during the rest of the year. The magnitude of this trend was again greater in walkers than controls.

Figure 6.2
Mean body circumferences at five sites during walking programme
percentages of baseline values

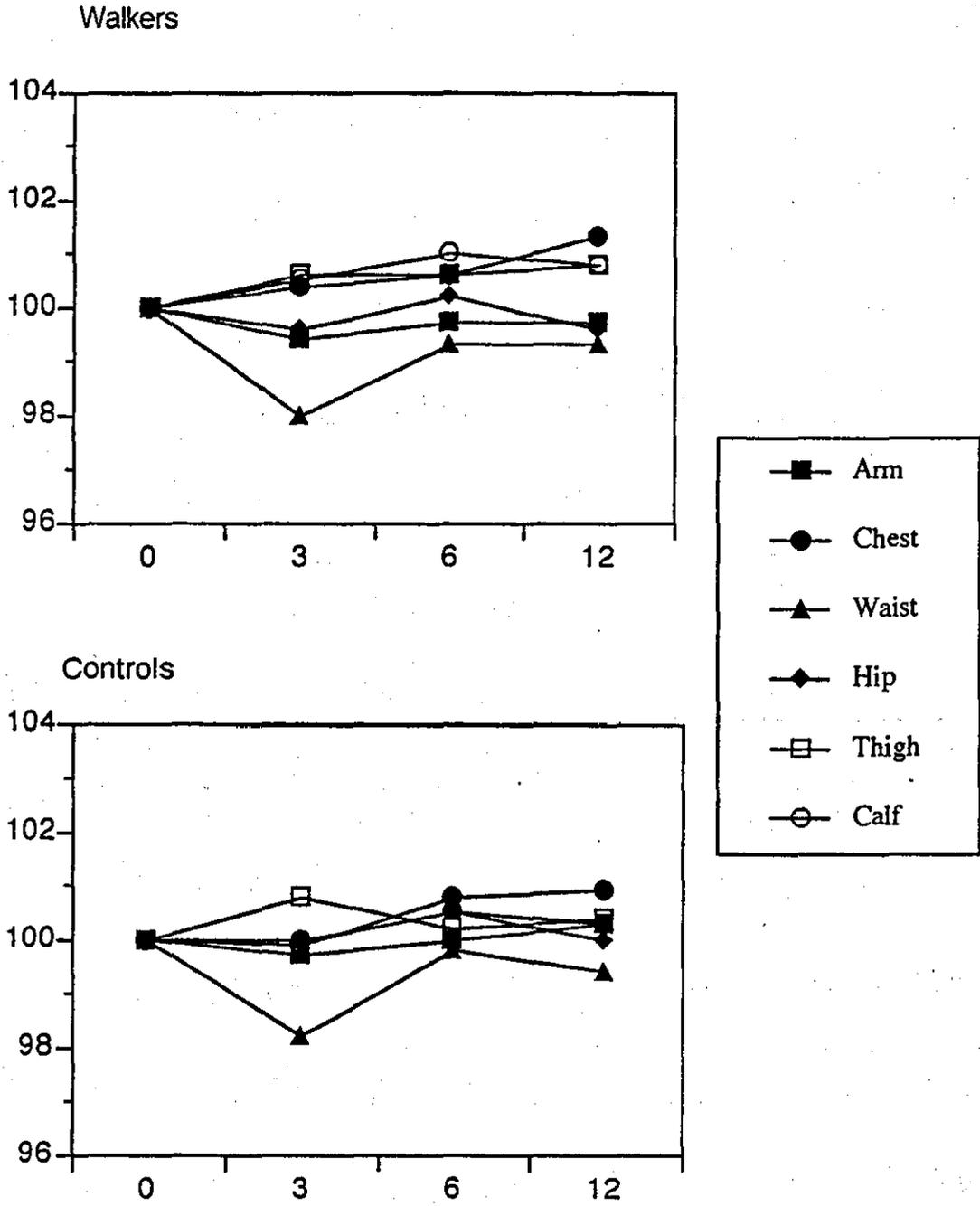


Table 6.10**Waist hip ratio of walkers and controls before and at 3, 6 and 12 months after the start of walking programme**

	Walkers		Controls	
	Mean	SD	Mean	SD
Baseline	.95	.06	.95	.07
3 months	.93	.07	.94	.07
6 months	.94	.06	.95	.07
12 months	.94	.06	.95	.07

Skinfold thicknesses

Changes in mean skinfold thicknesses were proportionately greater than those of circumferences (Figure 6.3). Significant changes through the year were again apparent at all sites except medial calf ($P < 0.01$). The tendency was again towards a decrease in the first three months, and an increase in the final six months.

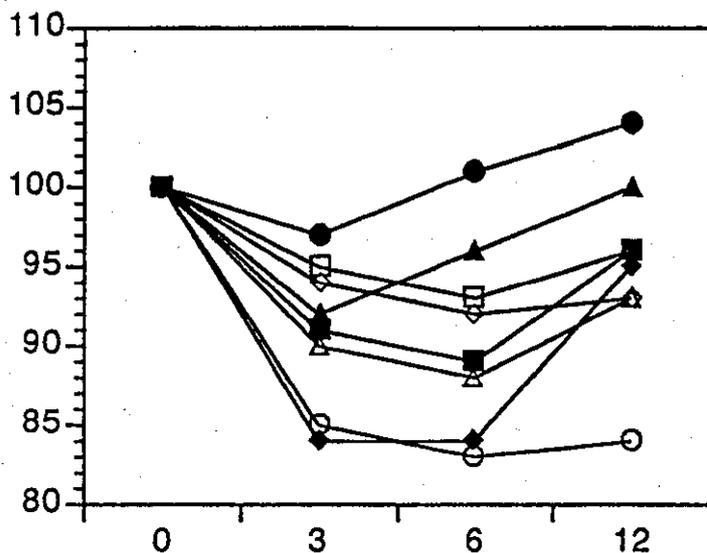
At two sites: anterior thigh and medial calf, the interaction term in the analysis of variance was significant ($P < 0.05$), indicating a significantly different response between walkers and controls. Skinfold thickness at the anterior thigh site decreased in both walkers and controls during the first three months. However the increase during the rest of the year was greater in controls than walkers, resulting in a mean loss of 0.9 mm over the year in walkers, and a mean gain of 0.9 mm in controls. Mean medial calf skinfold thickness decreased by 0.6 cm in walkers.

These decreases in peripheral fatness with very little change in total fatness suggest that the proportion of central fatness may have increased. This means that participation in the walking programme may have actually resulted in a less favourable distribution of body fat.

To determine whether the decrease in anterior thigh and medial calf skinfold thicknesses was dependent upon the amount of walking performed, the correlation between the change in skinfold thicknesses and amount of walking performed was examined. However, no association was observed between change in skinfold thickness and time or distance walked during the year, so there was no evidence of a dose-response relationship.

Figure 6.3
Mean skinfold thickness at eight sites during walking study:
percentages of baseline values

Walkers



Controls

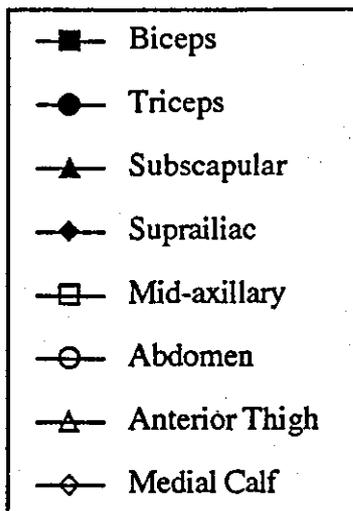
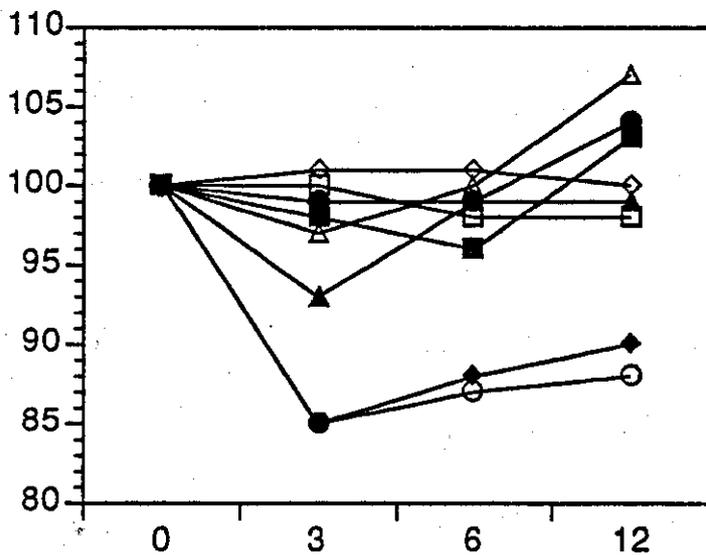


Table 6.11

Skinfold thickness ratios at baseline and 3, 6 and 12 months after start of walking programme

Sites	Month	Walkers		Controls	
		Mean	SD	Mean	SD
ss/th	0	1.37	0.70	1.23	0.48
	3	1.37	0.70	1.18	0.46
	6	1.44	0.67	1.25	0.46
	12	1.41	0.76	1.22	0.44
tr+ss/ss	0	1.74	0.26	1.72	0.20
	3	1.78	0.27	1.76	0.23
	6	1.77	0.24	1.72	0.21
	12	1.79	0.27	1.74	0.20

Mean values of skinfold ratios examined during the year are shown in Table 6.11. Analysis of variance of these ratios revealed that there was no difference between walkers and controls in mean values or response. There were however significant changes through the year in triceps plus subscapular to subscapular ratio (tr+ss/ss). Mean values for this ratio were higher at 3 months than baseline (mostly due to an decrease in subscapular skinfold) and higher at 12 months than baseline (mostly due to an increase in triceps skinfold thickness).

However it must be considered that these ratios have been found to be fatness dependent (Garn et al. 1982; 1988). So the observed trend in tr+ss/ss may be partly due to changes in fatness rather than actual changes in fat distribution. The fatness dependence of these ratios is considered below.

Cross-sectional area of fat and muscle of lower limb

Skinfold thickness at anterior thigh and medial calf sites decreased, with no significant change in thigh and calf circumferences. The possibility that this was due to an increase in cross-sectional area of muscle is examined, by estimating the cross sectional area of muscle and bone of the lower limb, at the levels of thigh and calf circumferences. This was achieved by assuming the limb to be circular, and subcutaneous fat to be evenly distributed around the limb. The radius of the limb (R) was calculated from the circumference (C):

$$R = C/2\pi$$

The thickness of adipose tissue and dermis was estimated from the skinfold thickness. This was adjusted to account for the compressability of the skinfold, as described in chapter 2. Significant differences in skinfold compressability exist between

different sites, and different age-groups, as discussed in the previous chapter. The skinfold compressibilities reported by Martin et al. (1985) were used, as these were determined at the same sites as measured in this study, and their subjects were nearer in age to the men in this study than the young adults studied in the previous chapter. The % compressibility at anterior thigh and medial calf sites was 33.6 and 34.4 respectively.

The adjusted SCATT was subtracted from the radius of the limb to calculate the cross sectional area of muscle and bone (AMB):

$$AMB = \pi \times (R - SCATT)^2$$

Estimated muscle and bone cross sectional areas at mid-thigh and maximal calf sites are shown in Table 6.12. At both sites there was an increase during the year in the walking group: most of this increase occurring during the first three months of the walking programme, with a decrease during the final six months. At the mid-thigh site there was also an increase during the first three months in the control group, with a subsequent decrease to the baseline value, so the response of the walkers was not significantly different from that of the controls. However at the calf site, the mean cross sectional area of muscle and bone of the control group remained relatively constant, and analysis of variance revealed a significantly different response in the walkers relative to the controls ($P < 0.001$).

The increase in calf muscle and bone cross sectional areas in walkers tended to be associated with increase in maximal oxygen uptake, although this trend was not significant ($r=0.27$, $P = 0.08$). Changes in muscle plus bone cross-sectional area at either mid-thigh or maximal calf site were not associated with time or distance walked.

Table 6.12

Cross-sectional area of muscle and bone at mid-thigh and maximal calf sites in walkers and controls (cm²)

Site	Month	Walkers		Controls	
		Mean	SD	Mean	SD
Mid-thigh	0	156.1	26.5	152.4	21.6
	3	161.2	25.6	156.7	20.6
	6	162.0	24.2	153.4	22.1
	12	158.7	23.5	152.3	20.0
Maximal Calf	0	93.9	15.8	93.0	10.1
	3	96.2	13.3	93.0	11.1
	6	97.4	14.0	93.7	9.9
	12	97.1	13.7	92.5	9.7

Principal component analysis of skinfold thicknesses

A principal component analysis was performed to allow a summarisation of skinfold thicknesses at eight sites. Two factors were identified with an eigenvalue greater than 1. The factors accounted for 56 and 18 % of the variance respectively. Factor loadings are shown in Table 6.13. The first factor was associated with general fatness, with high loadings for all skinfold thicknesses, the lowest loading being for anterior thigh skinfold thickness. The second factor seemed to represent extremity versus trunk fatness, with positive loadings for skinfold thicknesses at anterior thigh, medial calf, and triceps sites, and negative loadings at mid-axillary, subscapular, abdomen and suprailiac sites.

Table 6.13

Principal component analysis of skinfold thicknesses: factor loadings

Site	Factor 1	Factor 2
Biceps	.82	.06
Triceps	.79	.35
Subscapular	.78	-.38
Suprailiac	.86	-.27
Mid-axillary	.84	-.42
Abdomen	.67	-.24
Anterior Thigh	.49	.76
Medial Calf	.68	.51

Mean scores of Factor 1 (related to all skinfold thicknesses i. e. general fatness factor) changed significantly through the year ($P < 0.05$) but there was no significant difference between walkers and controls, and the difference in response was not significant. As would be expected, the changes during the year were parallel to changes in % fat: the mean Factor 1 score decreased during the first six months, and increased during the final six months.

Factor 2 scores showed a significantly different response between walkers and controls ($P < 0.05$). In the walkers, Factor 2 scores decreased during the first 3 months, indicating a decrease in extremity skinfold thicknesses relative to trunk skinfold thicknesses, and increased during the rest of the year. In controls, Factor 2 scores increased during the year.

Fatness-dependence of indices of fat distribution

The fatness dependence of the indices used in this study was examined by calculating correlations with % fat (from hydrostatic weighing). The waist hip circumference ratio was found to be highly correlated with body fatness ($r=0.60$; $P<0.001$), as was Factor 2: the peripheral versus central fatness factor from factor analysis ($r=0.42$; $P<0.001$). However none of the skinfold ratios employed were significantly correlated with body fatness.

As waist hip ratio and Factor 2 scores were found to be fatness dependent, the changes in the variables during the year may be due to changes in total fatness rather than fat distribution. As changes in these variables followed the same trend as changes in weight and % fat, this explanation seems increasingly likely.

The ratio of triceps plus subscapular to subscapular skinfolds was not found to be fatness dependent. So the changes in this variable during the year observed above may indicate actual changes in fat distribution, independently of changes in fatness.

Relationship of changes in body composition and fat distribution with changes in fitness

Correlations of changes in body composition and fat distribution with the amount of walking performed and changes in fitness are shown in table 6.14.

Table 6.14

Correlation of changes in body composition and fat distribution with amount of brisk walking performed and changes in fitness

	mean mins/day (walkers)	mean miles/day (walkers)	$\dot{V}O_2$ max (ml/kg/min)	$\dot{V}O_2$ at 2mmol lactate
weight	-.21	-.20	-.23	-.20
BMI	-.20	-.19	-.26*	-.20
%fat	-.16	-.14	.09	-.17
fm(kg)	-.20	-.18	-.02	-.21
ffm(kg)	.01	.01	-.22	-.02
F1	.08	.07	-.23	-.15
F2	-.23	-.24	-.10	-.09
WHR	.11	.10	.02	-.02
ss/th	.13	.13	.04	-.04
tr+ss/ss	.09	.08	.18	-.00

* $P < 0.05$ ** $P < 0.01$ *** $P < 0.001$

Changes in body composition or fat distribution were not related to either the time or distance walked during the year, so no evidence of a dose response relationship was observed. Changes in maximal oxygen uptake ($\dot{V}O_2$ max) were inversely related to changes in BMI, as may be expected, as the maximal oxygen consumption is standardised for body size by dividing by body weight. Changes in $\dot{V}O_{2\text{max}}$ were not related to other parameters of body composition or fat distribution.

The change in oxygen consumption at 2mmol/l lactate (a measure of endurance) was not related to any of the body composition or fat distribution variables studied.

Levels of other cardiovascular risk factors during the walking programme

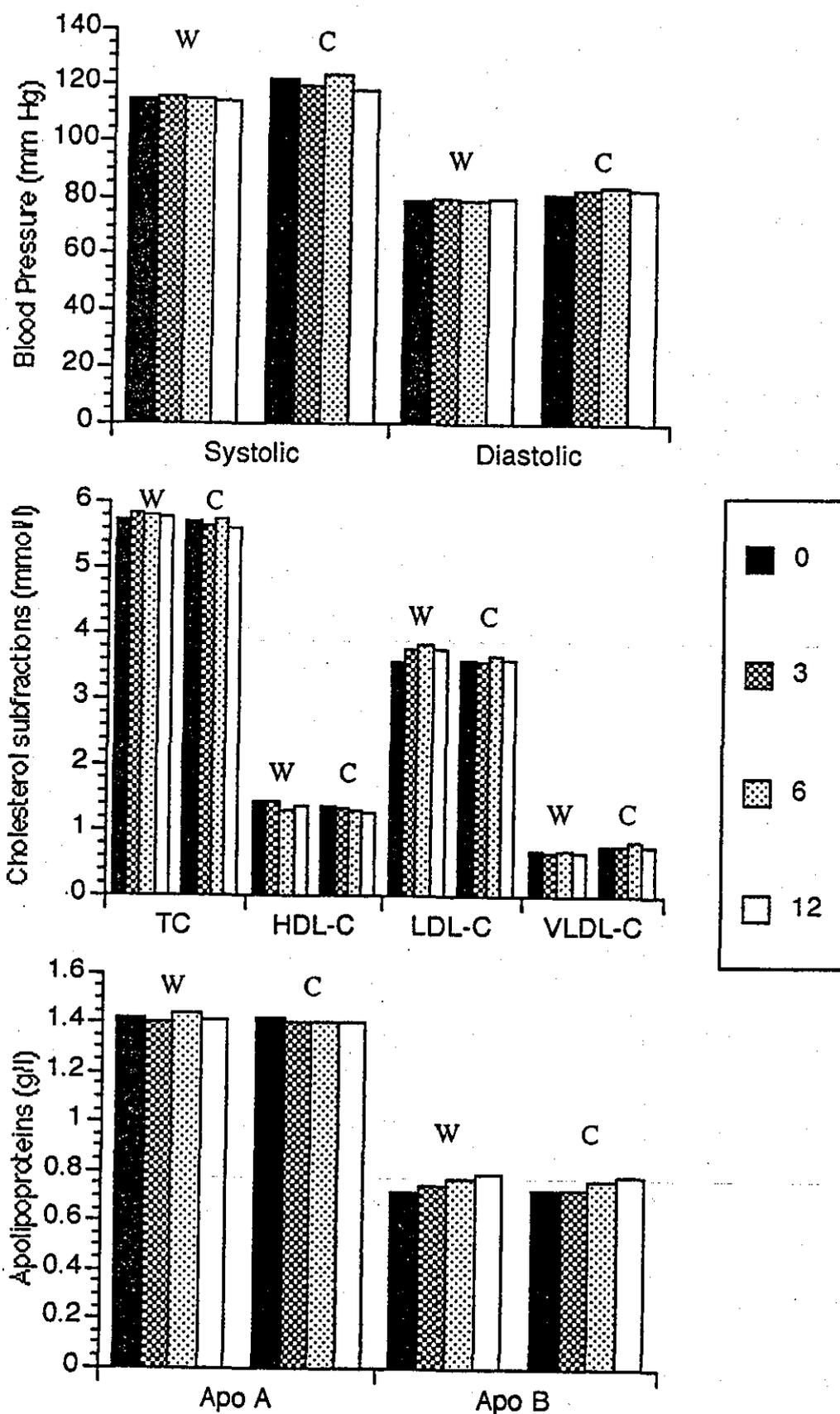
Influence of brisk walking programme

Baseline levels of systolic and diastolic blood pressures, total cholesterol, high density lipoprotein cholesterol and low density cholesterol were compared with average values from a random survey of British adults of similar age (OPCS 1990). Levels of all variables apart from diastolic blood pressure were found to be significantly ($P < 0.001$) lower than the population mean. The reason for this may be twofold: firstly the subjects were self-selected: subjects volunteering for a study such as this are likely to be more interested in issues of health, and may therefore lead "healthier" lifestyles. Secondly, the criteria for accepting subjects into the study excluded subjects with high blood pressure or total cholesterol.

Levels of blood pressure and blood lipids and lipoproteins of walkers and controls throughout the walking study are shown in figure 6.4. There was very little change in these risk factors during the course of the study. On analysis of variance, no significant differences between walkers and controls were observed, and there was no difference in response between the two groups, for any of the above variables. However there were significant changes through the year in HDL-C (which decreased during the year); Apo B, and lp(a) (which increased during the year); and LDL-C and the ratio of TC/HDL (which increased during the first six months and decreased during the final six months).

There was no significant difference in response between walkers and controls in any of the cardiovascular risk factors studied. There was therefore no evidence that participation in the walking programme produced any favourable changes in blood pressure or cholesterol metabolism.

Figure 6.4
Mean blood pressure, cholesterol subfractions and apolipoprotein subfractions in walkers (W) and controls (C), at baseline and 3, 6 and 12 months after the start of the walking programme



Relationship of body composition and fat distribution with other cardiovascular risk factors at baseline

The correlations of various indices of body composition and fat distribution with blood pressure and blood lipid concentrations are shown in Table 6.15. A number of significant associations were observed, despite the sample size here being considerably smaller than those of cross-sectional studies described in chapter 1, and the exclusion of subjects with high blood pressure and total cholesterol concentrations.

Table 6.15
Correlation of body composition and fat distribution indices at baseline with cardiovascular risk factors

	BPS	BPD	TC	HDL	LDL	TC/ HDL	TG	ApoA	ApoB	lp(a)
weight	-.14	.06	-.01	-.35**	-.02	.22	.27*	-.25*	.04	.16
BMI	.05	.14	.11	-.31*	.04	.28*	.40***	-.20	.14	.04
%fat	.14	.04	.19	-.23	.20	.27*	.22	-.24	.28*	-.23
fm(kg)	.03	.04	.15	-.32*	.14	.31*	.30*	-.27*	.23	-.10
ffm(kg)	-.22	.07	-.13	-.21	-.13	.04	.11	-.10	-.13	.28
F1	.16	.10	.16	-.13	.11	.17	.26*	-.05	.07	.03
F2	-.21	-.16	-.16	-.04	-.08	-.13	-.21	-.12	-.10	-.10
WHR	.18	.16	.24	-.27*	.18	.34**	.41***	-.16	.21	-.15
ss/th	.12	.05	.20	-.12	.13	.22	.33**	-.01	.13	.15
tr+ss/ss	-.18	-.08	-.27*	.04	-.18	-.21	-.33**	-.08	-.20	-.16

* P < 0.05 ** P < 0.01 *** P < 0.001

In general, lower amounts of total body fat and central fatness were associated with a more favourable risk factor profile, in agreement with the literature (chapter 1).

Systolic and diastolic blood pressures were not found to be associated with body composition, or indices of fat distribution in this sample.

Total cholesterol (TC) concentration was inversely associated with the ratio of triceps plus subscapular to subscapular skinfold thicknesses (i. e. associated with central or inversely associated with peripheral fatness). TC was not significantly correlated with body weight or fatness.

High density lipoprotein cholesterol (HDL) concentration was inversely correlated with body composition (weight, body mass index and fat mass) and with the ratio of waist to hip circumferences (WHR). However when the partial correlation of WHR with HDL with fat mass held constant was calculated, the association was no

longer significant ($r=-.14$). The apolipoprotein component of HDL (Apo A) was inversely correlated with body composition (weight and fat mass) but not significantly correlated with fat distribution.

Results for the ratio of total to HDL cholesterol were inverse to those for HDL: the ratio was positively correlated with BMI and body fatness, and with waist hip ratio. However the association of waist hip ratio was again not significant after adjustment for body fatness.

Plasma triglyceride concentration (TG) was significantly correlated with most of the body composition and fat distribution variables studied. The association of fat distribution with TG was independent of total fat content. Partial correlations for WHR, ss/th and tr+ss/ss were 0.31, 0.30 and -0.29: all were statistically significant ($P < 0.05$). So TG concentration was associated with both central and peripheral fatness.

Apoprotein B (one of the protein components of LDL and VLDL) was associated with % fat, but not with fat distribution. Lipoprotein (a) was not significantly associated with any of the body composition or fat distribution variables studied.

In summary higher levels of body weight or body fatness were associated with higher concentrations of triglycerides, TC/HDL and Apo B, and lower concentrations of HDL cholesterol and Apo A. A higher proportion of central fat was associated with higher total cholesterol and triglyceride concentrations.

Fat mass did not seem to be a better predictor of blood lipid levels than body weight. Similarly, there did not seem to be any one measure of fat distribution which better predicted blood lipid levels than the others studied.

Relationship of changes in body composition and fat distribution with changes in cardiovascular risk factors

There was no significant difference between walkers and controls in mean changes in blood lipids and blood pressure. However there were significant changes during the year in HDL, LDL, TC/HDL, Apo B and lp (a). To determine whether these changes were associated with changes in body composition or fat distribution, correlation coefficients were calculated which are shown in Table 6.16.

Results were generally similar to those from the cross-sectional data above. Correlation coefficients were generally of similar magnitude to that analysis, despite the fact that in making longitudinal comparisons, the effect of genetic variation is ruled out. However intraindividual changes in the variables studied were considerably smaller than the inter-individual variations being analysed above.

Systolic blood pressure was inversely associated with Factor 2: the extremity versus trunk factor from principal components analysis. So higher systolic blood pressure was associated with a greater proportion of trunk fatness, or lower proportion

of extremity fatness. Diastolic blood pressure was again not associated with any of the variables studied.

Change in total cholesterol was associated with change in body weight and BMI, but not with changes in fat distribution.

Changes in HDL were inversely correlated with changes in fat distribution (waist/hip ratio) but not with changes in body composition. However changes in Apo A and ratio of TC to HDL were not associated with any of the variables studied.

Although at baseline TC and LDL were not associated with body composition or fat distribution, changes in weight and BMI were significantly correlated with changes in TC and LDL, as well as Apo B. None of these changes were correlated with fat distribution changes.

Changes in triglyceride concentration were associated with changes in Factor 1 (subcutaneous fatness factor from principal components analysis), and inversely associated with ratio of triceps plus subscapular to subscapular skinfold thickness. So changes in triglyceride concentration were associated with both total and central fatness.

Changes in lipoprotein (a) concentration were inversely correlated with Factor 1 (general fatness factor) and correlated with changes in fat distribution (ratio of triceps plus subscapular to subscapular skinfold thickness). So change in lp(a) was inversely correlated with both total and central fatness.

In summary, reductions in weight or total body fat were associated with reductions in total cholesterol, LDL cholesterol Apoprotein B concentrations and triglycerides. Reductions in central fat were associated with reductions in systolic blood pressure and triglyceride concentrations, and increases in HDL cholesterol and lipoprotein (a) concentrations.

In this longitudinal analysis, body weight was found to be better correlated with blood lipids than % fat or fat mass. Again there did not seem to be any one index of fat distribution which performed better than the others.

Table 6.16

Correlation of changes in body composition and fat distribution with changes in cardiovascular risk factors

	BPS	BPD	TC	HDL	LDL	TC/ HDL	TG	ApoA	ApoB	lp(a)
weight	.22	.14	.28*	-.09	.27*	-.10	.18	.15	.26*	-.05
BMI	.20	.14	.30*	-.07	.27*	-.10	.21	.16	.27*	-.05
%fat	.06	-.02	-.04	-.04	-.02	-.05	-.05	.06	.08	-.12
fm(kg)	.11	.03	.06	-.05	.07	-.07	.03	.11	.14	-.17
ffm(kg)	.09	.12	.23	-.04	.20	-.02	.16	.04	.10	.16
F1	.19	-.06	.22	-.06	.12	-.17	.40**	.08	.09	-.31*
F2	-.27*	.13	.00	-.07	.08	.14	-.17	-.05	.09	.23
WHR	.01	-.07	-.01	-.28*	.09	-.10	-.04	-.23	-.02	-.13
ab/si	-.15	-.11	-.27*	.07	-.30*	.00	-.01	-.39**	-.24	.23
ss/th	.02	-.01	-.05	.01	-.09	.00	.13	.03	-.07	-.17
tr+ss/ss	.02	-.09	-.19	.16	-.16	-.09	-.27*	-.12	-.17	.30*

* P < 0.05 ** P < 0.01 *** P < 0.001

Discussion

Influence of walking programme on body composition

Participation in the brisk walking programme did not significantly influence body composition, although weight and % fat did decrease during the first three months. The possibility that this is due to an adaptation in energy intake is explored in the next chapter. Alternatively, walkers might have reduced energy expenditure during the rest of the day.

Mean weight decreased slightly in the walkers and increased in controls, although the difference in response was not significant. The increase in controls could be due to a trend to an increase with age, as reported by Montegriffo (1971). If the trend observed in this study persisted for a number of years, it is possible that brisk walking could assist in preventing this secular increase.

A significant decrease in body fat of middle-aged men was observed in a 16 week programme of running (Oscari and Williams, 1968), a year-long running programme (Wood et al. 1983), a 16 week brisk walking programme (Leon et al. 1979) and a 10 week brisk walking programme (Weltman et al. 1980). The first two of these studies involved higher intensity exercise than that in this study, whilst the study of Leon et al. involved greater duration (90 minutes per day), so it is possible that a greater intensity or duration of exercise is necessary to significantly influence body fatness. However, a decrease in energy intake was observed in the latter three studies, which may explain the decrease in fatness.

Body weight and % fat were observed to change significantly during the year, in walkers and controls. These changes could be due to involvement in the study, which may have made subjects more aware of their diet and health, inducing temporary changes in lifestyle. Another possibility is that these changes are due to a seasonal effect. The baseline set of tests were carried out between April and June, so a decrease in weight occurred in the spring and summer, and a progressive increase during the rest of the year. Some subjects involved in the study reported that their weight tended to be lower in the summer, due to changes in appetite, activity, or energy intake. Seasonal changes in weight and body composition have been reported in developing countries, where there are large seasonal variations in activity and availability of food. However in developed countries these influences are greatly diminished. In chapter 7 the diet of the men will be examined, allowing determination of whether a seasonal variation in energy intake occurred which may have caused these changes.

Studying the weight changes of subjects who continued in the study for a second year failed to elucidate this matter, as there was no significant change in weight in this sub-group in either the first or second year. This could be due to the small number of

subjects studied. As some evidence of a seasonal trend was again observed, at least in weight, a seasonal effect can not be discounted, which may have been significant if a larger number of subjects had been studied. However these changes may have occurred purely by chance, so it is impossible to determine which of the two possible explanations is more likely, or whether a combination of the two effects was the cause of the changes.

Influence of walking programme on fat distribution

The walking programme did not influence most of the indices of fat distribution examined in this study. However there were significant decreases in lower limb skinfold thicknesses in walkers relative to controls. The decrease of these peripheral skinfolds, with no apparent change in total fat or central skinfolds, may indicate an overall trend to a more centralised distribution of body fat.

This observation that there was a loss of lower limb skinfolds, but no significant changes in trunk skinfolds is in contrast to effects of exercise upon fat distribution reported by other workers. Leon et al. (1979) observed a significant decrease in thigh skinfold, but also decreases in abdomen, axillary, triceps and subscapular skinfolds after a 16 week walking programme. Schwartz et al. (1991) observed a decrease in subcutaneous fat at the mid-thigh site, with no change in muscle area, in young men after six months intensive endurance training. However decreases in central fat depots were also observed. In older men there were significant decreases in central adiposity but no change in thigh subcutaneous fat. Després et al. (1985) found that 20 weeks of cycle ergometer aerobic training caused a greater decrease in trunk than extremity skinfolds. The same observation was reported by Tremblay et al. (1988) after a 100 day programme of cycle ergometer training.

In all these studies, a decrease in weight and fatness was observed, with a general decrease in skinfold thickness. Himes (1988) reported that nutritional intervention generally produced greater changes in trunk than extremity skinfold thicknesses. So in the studies described, the effect of fat loss upon fat distribution could be concealing the effect of exercise on fat distribution.

Association of body composition with cardiovascular risk factors

The walking programme was not found to influence blood pressure, or blood lipid or lipoprotein levels. These findings conflict with findings in women that participation in a programme of brisk walking was associated with increase in HDL-C (Hardman et al. 1989). There were also no significant changes in means of body composition and commonly used indices of fat distribution. The suggestion of Wood et

al. (1988) that exercise induced changes in HDL-C are mediated by changes in body composition could explain the lack of any change in HDL-C here.

Changes in body composition and fat distribution in individuals were relatively small, but were significantly related to changes in other cardiovascular risk factors. Decreases in weight, total body fat, and central fat were associated with favourable changes in blood pressure and blood lipids. These associations agreed with observations in the literature.

Blood pressure has been reported to be associated with both total fatness and central adiposity (Krotkiewski 1983, Kalkhoff 1983, Blair 1984, Weinsier 1985, Després 1988). However in this study no association was observed between body composition and blood pressure, or change in body composition and change in blood pressure. This could be due in part to the exclusion of men with a blood pressure greater than 160/95 from the study. Change in systolic blood pressure was found to be associated with change in central fatness.

Triglyceride concentrations were found to be associated with both body fatness and fat distribution, and changes in body fatness were associated with changes in triglyceride concentration. These findings are in accord with those of Krotkiewski et al. (1983) and Despres et al. (1988).

Of the serum lipids studied, HDL-C was most strongly associated with body composition and fat distribution variables. The cross-sectional data showed HDL-C to be inversely associated with body weight, BMI and fat mass, and the longitudinal data showed associations with the waist-hip ratio. These findings agree with those of Garrison et al. (1986), Rhoads et al. (1976) and Despres et al. (1985). Apo A, the protein component of HDL, was also inversely associated with body weight and fat mass in baseline data.

Total cholesterol and LDL cholesterol were not associated with body fatness or fat distribution in the cross-sectional data, although Apo B (a major protein component of LDL) was correlated with % fat. In the longitudinal analysis, the changes in all three were associated with change in weight. This association of cholesterol with body weight is in accordance with the reports of Ashley & Kannel (1974), Noppa (1980) Borkan et al. (1986).

Changes in central fatness were inversely associated with changes in lp(a). There is less literature available on the association of lp(a) with body composition and fat distribution. These findings contradict the other findings where increased levels of fatness and central fat were associated with a less favourable cardiovascular risk profile.

Fat mass or % fat did not seem to be a better predictor of blood lipid levels at baseline than body weight. This could be due in part to the relatively homogeneous level of this group, who were all sedentary, and of similar age. In a group including more

physically active subjects, there may be greater variation in muscle mass, so weight would not be such a good predictor of fatness, as some individuals may be overweight but lean. So in this case body fatness may be a better predictor of blood lipid levels, as observed by Segal et al. (1987).

In the longitudinal data, changes in fat mass or % fat were not significantly correlated with changes in blood lipids, whilst changes in body weight were. This could be due to the considerably greater degree of error associated with measurements of body composition than with measurement of body weight. These errors will be of particular importance when assessing small changes in body weight.

There did not seem to be any single index of fat distribution which was a better predictor of blood lipid levels than the others studied. However, the waist hip ratio, and the fat distribution factor from factor analysis, were found to be fatness dependent. This probably accounted for the finding that the correlation of WHR with HDL decreased when adjusted for body fatness.

Conclusions

The walking programme did not influence body weight or body composition. However, significant changes were observed during the year, which could be indicative of a seasonal effect.

Involvement in the walking programme significantly decreased lower limb skinfold thicknesses. This change in calf skinfold appeared to be accompanied by an increase in the cross-sectional area of muscle and bone.

Body composition and fat distribution at baseline were significantly correlated with blood lipid levels. Lower levels of both total and central fatness were generally associated with a more favourable cardiovascular risk profile, in accordance with the literature.

Although the walking programme did not significantly influence blood lipid levels or body composition, and had little influence on central fatness, changes in body composition and fat distribution during the year were associated with changes in blood lipid levels. Again, decreases in total or central fatness were associated with improved cardiovascular risk profile.

Chapter 7

Influence of programme of brisk walking on dietary intakes of middle aged men, and associations with cardiovascular risk factors

Introduction

In the previous chapter, participation in a year long programme of brisk walking was not found to significantly influence body composition, blood pressure, or blood lipid levels. In this chapter the dietary intakes of the men involved in the brisk walking study are examined. The purpose of this investigation is threefold. Firstly, the influence of the walking programme on energy intake is examined. Secondly any changes in the composition of the diet are assessed to determine whether they may have concealed a potential change in blood lipid levels. Thirdly, the associations between dietary intakes and blood lipid levels are examined.

In the previous chapter, participation in the walking programme was not found to influence body composition. However, it would be anticipated that the total daily energy expenditure of the men examined would have increased, as walking was required to be performed in addition to any habitual exercise. These findings could be reconciled by an increase in walkers' energy intakes. Whether any adaptation in energy intake in response to the walking programme was observed is examined in this chapter.

The association of dietary intakes with incidence of CHD and levels of blood lipids was discussed in chapter 1. The nutrients reported to be most highly associated were dietary fats and lipids. Total fat intake and SFA intake have been found to be associated with TC, and PUFA intake has been inversely associated with TC (Keys et al 1980). MUFA intake was regarded as neutral in earlier studies (Keys et al 1980). However, increasing MUFA intake has been found to lower TC, but not lower HDL-C as does increasing carbohydrate intake, and hence decreasing fat intake (Mensink & Katan 1987).

Dietary cholesterol has also been found to influence TC, although this effect seems to occur in some individuals to a greater extent than others (Katan et al 1986).

Alcohol intake has been related to HDL-C and Apo A levels (Gordon et al 1981; Camargo et al 1985), although the association of alcohol intake with incidence of CHD is more controversial (Shaper et al 1987).

Men were instructed not to make any conscious changes to their diets, to attempt to control for the influence of dietary intakes upon blood lipid levels.

However, participation in the study did seem to make the men increasingly aware of the influence of lifestyle and diet on cardiovascular disease. Whilst every attempt was made to ensure that men did not change their diet, it is possible that the men took more interest of media reports of the effects of diet after becoming involved in the study. Dietary intakes were therefore monitored during the walking study, to determine whether changes in the composition of the diet occurred which may have influenced blood lipid levels, and even concealed a change which may otherwise have been observed. In analysis of dietary data, emphasis was placed on the nutrients discussed above.

The associations of dietary intakes with blood lipid levels were also examined. As in the previous chapter, both cross-sectional and longitudinal data were examined, and associations were compared with those reported in the literature.

Methods

Dietary intakes were assessed by means of a weighed inventory technique, as described by Marr (1971) and Bingham (1987). Weighing was performed for seven consecutive days. This period of time has been reported to be sufficient for correct ranking of subjects according to intakes of energy, protein, fat, carbohydrate and saturated fatty acids (Marr et al 1971; Nelson et al 1989). However, these authors report a period of greater than 24 days to be necessary for correct classification of polyunsaturated fatty acid and cholesterol intakes, and P:S ratio.

However it was felt that with the many commitments involved in this study, using a longer period of time may decrease the compliance of subjects, and result in incomplete recording.

Weighed inventories

The men were given a set of Soehnle or Hansen digital scales. Both had a tare facility, and a maximum capacity of 1000g, with an accuracy of 1g up to 64 g, and 2g thereafter. Scales were calibrated with known weights before use. Men were also given a folder containing blank dietary recording sheets, an instruction sheet, and an example of a day's inventory (Appendix IV). Instructions were also given verbally before measurement.

Men were instructed to record weights of all food and drink consumed, by an additive weighing technique. Weight of all leftovers was recorded. They were asked to also include method of cooking and brand names. Food was weighed after cooking, and men were asked to give recipes or relative proportions of constituents wherever possible. Plastic containers were supplied to allow a personal supply of frequently consumed foods such as milk and spreads to be kept, which were weighed at the beginning and end of each day, and whenever refilled.

The importance of recording all items of food and drink consumed was stressed. If an item was not weighed, men were asked to describe it in terms of package size, household measures, or portion sizes.

The 7 days on which recording was performed were to some extent left to the subjects. They were instructed to select a week which they felt was representative of their normal intake. However there seemed to be a tendency to avoid eating out while recording, and to avoid weeks where social occasions involving consumption of food occurred.

Wherever possible a follow-up phonecall was made to the men, 1 to 2 days after recording started, to determine whether any problems were being experienced, and to reinforce the instructions given.

Analysis

Inventories were coded in accordance with the food composition tables of Paul & Southgate (1978), and supplements (Paul et al., 1980; Tan et al, 1985; Holland et al, 1988; 1989). Weights of any foods which had not been weighed were estimated from household measures or portion sizes (Crawley, 1988).

Inventories were analysed using Microdiet (Salford University) on a Viglen personal computer. This programme contained food composition data from the food composition tables above. However, it was found that the databases of this programme were incomplete. Cholesterol content of most foods was not included, and no breakdown of fatty acids was included for a number of foods. So before analysis could be started, the cholesterol and fatty acid content of foods not included were manually added to the database. Values were obtained from a number of sources: McCance & Widdowson's "The Composition of Foods" and supplements; the database of another dietary analysis programme (Foodtabs, T. Saunders); and if not available elsewhere from "The Quick Cholesterol and Fat Counter" (Cox & Brusseau 1989). In addition values were calculated from recipes. The fatty acid breakdown of the average reported fat used for cooking by the men in the study was calculated. This was included in recipes to estimate the fatty acid compositions of fried foods and other foods such as pastries.

This updating of the database was extremely time-consuming, taking longer than the entry and analysis of all the inventories. However it was deemed necessary, as fatty acid and cholesterol intakes were of particular interest in this study.

Results

Dietary intakes at baseline

Intakes of walkers and controls at baseline are shown in Table 7.1. The average intake of British men of similar age from the OPCS dietary and nutritional survey of British adults (OPCS, 1990) is included for comparison. As the age of the men in this study fell between two age-groups of the OPCS survey, the weighted mean of these two age groups was calculated.

Dietary intakes of walkers were not significantly different from those of controls for any of the nutrients studied. Energy, protein, alcohol and fat (saturated, polyunsaturated and monounsaturated) intakes were not significantly different from average values for British men. However cholesterol intakes of walkers and controls were significantly lower than the British average.

Table 7.1

Mean dietary intakes of walkers and controls at baseline (compared with national average)

Nutrient	Average ^a	Walkers	Controls
Energy (MJ)	10.24	10.48 ± 15.63	10.70 ± 14.89
Protein (g)	86.0	91.7 ± 17.0	83.9 ± 11.9
Carbohydrate (g)	271	293 ± 62 *	295 ± 56
Alcohol (g)	25.5	21.3 ± 18.2	21.8 ± 22.9
Fat (g)	101.6	103.8 ± 23.0	106.8 ± 17.6
Saturated (g)	42.3	38.8 ± 10.9	39.2 ± 8.1
Polyunsaturated (g)	15.2	14.4 ± 5.9	16.0 ± 5.3
Monounsaturated(g)	30.8	32.3 ± 8.0	33.0 ± 6.5
Cholesterol (mg)	402	289 ± 93 *	291 ± 83 *

^a mean for British men of similar age (OPCS 1990)

* significantly different from British mean

Energy intake during walking programme

The daily energy intake of walkers and controls at baseline and at 6 and 12 months after the start of the walking programme is shown in Table 7.2. Mean energy intake of walkers decreased during the year, by nearly 0.4 MJ, whilst that of the controls remained relatively constant. However, analysis of variance revealed no significant differences between walkers and controls, no significant changes in energy intake during the year, and no significant difference in response between walkers and controls.

These changes in energy intake therefore do not explain the observation that body composition remained constant during the walking programme, whilst it would be expected that energy balance would increase. In fact the decrease in mean energy intake would if anything increase the possible energy deficit.

Seasonality of energy intake

In the previous chapter, significant changes through the year in body weight and % fat were observed: a possible seasonal effect. The changes in body weight and % fat of the men who completed weighed inventories at each time period were therefore related to the changes in energy intakes of these men (Table 7.2).

Changes in body weight through the year were no longer significant in the subgroup of subjects who completed weighed inventories. However changes in % fat through the year were still highly significant ($P < 0.001$). Changes in energy intake through the year were not significant. However a similar trend occurred as observed for % fat: lower values at 6 months than at baseline and 12 months. So it seems that these changes in fatness during the year could be partly due to changes in energy intake.

Table 7.2

Energy intake (MJ), weight (kg) and % fat of walkers and controls at baseline and at 3, 6 and 12 months after start of walking programme: mean \pm standard deviation

		Walkers n=35	Controls n=19
Energy Intake	Baseline	10.71 \pm 1.50	10.92 \pm 2.15
	6 months	10.42 \pm 1.94	10.72 \pm 1.48
	12 months	10.45 \pm 1.82	10.97 \pm 1.88
Weight	Baseline	79.4 \pm 9.2	76.3 \pm 9.0
	6 months	79.0 \pm 9.4	76.7 \pm 9.3
	12 months	79.1 \pm 9.3	76.5 \pm 9.3
% fat	Baseline	25.1 \pm 4.3	24.7 \pm 4.7
	6 months	23.5 \pm 4.3	23.5 \pm 4.7
	12 months	24.5 \pm 4.6	24.2 \pm 4.2

Composition of diet

The proportion of energy from protein, carbohydrate, fat and alcohol in walkers and controls during the walking programme is shown in Figure 7.1.

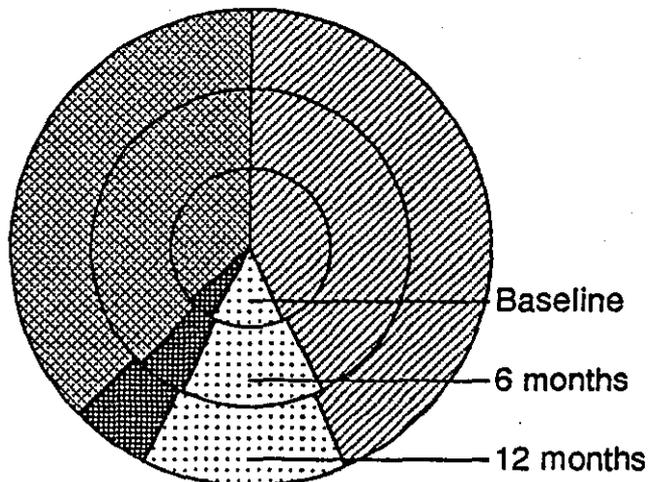
Significant differences between walkers and controls in change in protein intake during the year were observed, although there were no significant differences between the two groups. The protein intake of controls was higher at 6 and 12 months than at baseline, whilst that of walkers was lower at 12 months than at baseline and 6 months. The same findings were observed for protein as percentage of total energy intake. So the decrease in protein intake is not as a result of changes in energy intake.

There were no significant changes through the year, differences between walkers and controls, or differences in response between walkers and controls in fat, alcohol or carbohydrate intake, whether these were expressed as total intakes or percentages of energy intake.

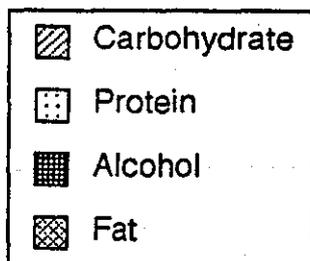
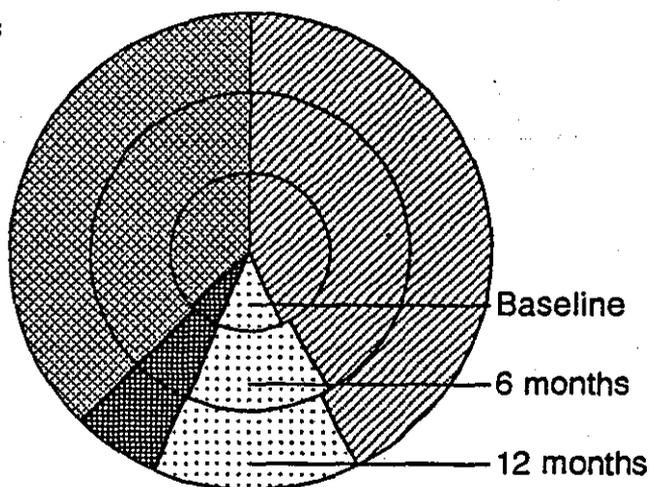
As protein intake has not generally been reported to influence blood lipid levels, it seems unlikely that these changes in the macronutrient composition of the diet will have caused a differential response in blood lipids between walkers and controls.

Figure 7.1
Proportion of energy from protein, carbohydrate and fat and alcohol

Walkers



Controls



Fatty acid and Cholesterol intakes

There were no significant changes through the year, differences between walkers and controls, or differences in response between walkers and controls in cholesterol, or saturated or monounsaturated fat intakes.

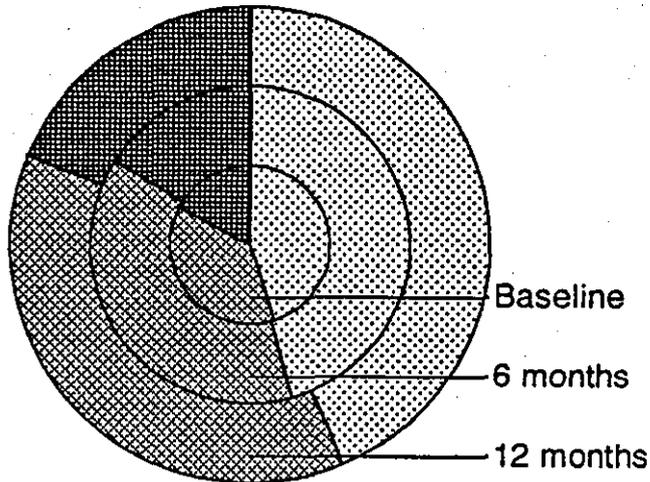
There were significant changes through the year in polyunsaturated fat intake and P:S ratio. Mean intakes were lower at 6 months than at baseline, and significantly higher at 12 months than both baseline and 6 months.

The effect of a change of this magnitude upon TC was estimated using the equations of Keys et al (1965) and Hegsted et al (1986). According to these equations, the increase in the % of energy from polyunsaturated fats would be expected to produce an decrease in TC of nearly 1 mg/dl. However, changes in TC in this study were extremely small, being generally no greater than 0.1 mg/dl.

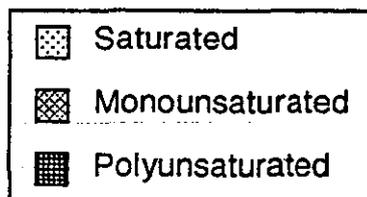
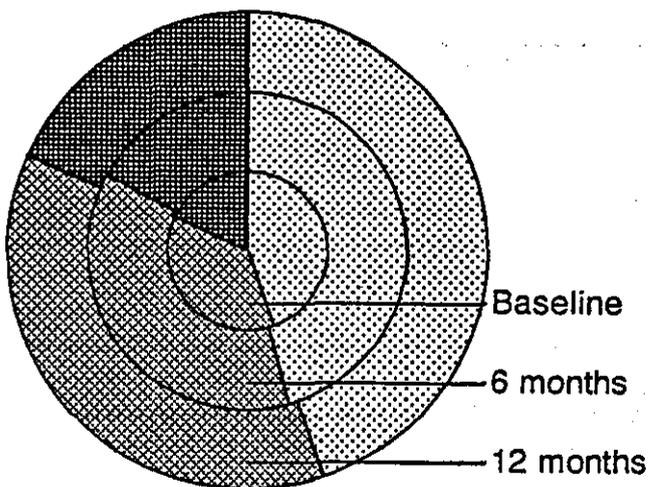
The associations of changes in PUFA intake with changes in blood lipids are examined below. This may allow determination of whether the changes in PUFA intake were associated with changes in blood lipid levels.

Figure 7.2
Fatty acid composition of diet at baseline, and at 6 and 12 months
after start of walking programme

Walkers



Controls



Association of dietary intakes with levels of cardiovascular risk factors at baseline

The baseline data from the study was used to provide a cross-sectional examination of the associations between dietary intakes and levels of other cardiovascular risk factors. Alcohol intake, fibre intake, and carbohydrate intake were significantly correlated with blood lipids levels. However no association was observed between intakes of fatty acids and blood lipids, which have often been found to be the strongest dietary predictors of blood lipid levels (McNamara 1987; Grundy 1987).

Alcohol consumption has been reported to be associated with HDL (Gordon et al 1981) and Apo A (Camargo et al 1985). However in this analysis, it was also significantly positively correlated with levels of TC and triglycerides. To determine whether these associations were dependent upon body composition or fat distribution, partial correlations were calculated. The association of alcohol intake with TC and TG was still significant after adjustment for body fatness and fat distribution ($r=0.36$ and 0.40 respectively, $P < 0.01$). Associations also persisted when non-drinkers were excluded from the analysis.

Carbohydrate intake was inversely correlated with HDL, as was fibre intake with TC, HDL and LDL.

Table 7.3

Correlation of dietary intakes with cardiovascular risk factors at baseline

	S BP	D BP	TC	HDL	LDL	TG	ApoA	ApoB	lp(a)
energy	.03	-.04	-.10	-.01	-.16	.13	.09	-.07	.14
protein	-.07	.15	-.21	-.07	-.18	-.09	-.06	-.11	.03
alcohol	.20	-.04	.37**	.29*	.20	.32**	.43**	.22	.22
carbohydrate	-.07	-.11	-.24	-.30*	-.23	.12	-.23	-.12	.07
fibre	-.13	.01	-.25*	-.25*	-.25*	.11	-.20	-.16	.09
fat	.02	.04	-.09	.16	-.11	-.11	.18	-.12	.06
SFA	-.06	-.02	-.13	.10	-.16	-.06	.10	-.17	.13
MUFA	.03	-.00	-.06	.14	-.06	-.13	.11	-.10	.11
PUFA	.18	.16	-.24	-.03	-.24	-.07	-.02	-.21	-.15
cholesterol	-.03	.01	-.05	.19	-.05	-.14	.18	-.07	.15
P:S ratio	.14	.09	-.11	-.13	-.10	.02	-.14	-.05	-.16

* $P < 0.05$ ** $P < 0.01$

Association of changes in diet with changes in cardiovascular risk factors

The longitudinal data were also examined. Correlations of changes in dietary intakes with changes in cardiovascular risk factors during the study are shown in Table 7.4.

Very few significant correlations were observed. Changes in alcohol intake were significantly positively correlated with changes in total cholesterol and Apo A. These associations again persisted when non-drinkers were excluded.

Changes in intakes of dietary fats were not correlated with changes in blood lipids, as observed above. Although significant changes in protein and PUFA intakes were observed above, the level of these changes were not correlated with cardiovascular risk factors.

Table 7.4

Correlation of changes in dietary intakes with changes in cardiovascular risk factors

	S BP	DBP	TC	HDL	LDL	TG	ApoA	ApoB	lp(a)
energy	-.15	-.09	-.00	.23	-.08	.00	.18	-.16	.04
protein	-.20	.08	.01	.21	-.06	.02	.08	-.07	.13
alcohol	.04	-.13	.25*	.15	.19	.11	.36**	.09	-.00
carbohydrate	-.21	-.09	-.06	.16	-.11	-.04	.10	-.20	-.06
fibre	.01	-.05	-.00	.18	-.04	-.08	.06	-.14	-.09
fat	-.08	-.05	-.08	.17	-.13	-.02	.06	-.15	.09
SFA	-.15	-.01	-.15	.14	-.19	-.02	.03	-.24	-.01
MUFA	-.05	.04	-.19	.09	-.21	-.10	-.02	-.21	.06
PUFA	.01	-.14	-.17	.05	-.15	-.15	-.07	-.18	.07
cholesterol	-.11	-.10	.02	.20	-.01	-.09	.02	-.02	.06
P:S ratio	.12	-.09	-.17	-.04	-.08	-.23	-.05	-.09	.09

Discussion

Compliance

The recording of weighed inventories was the most unpopular aspect of the study for most of the men involved. This was reflected by the numbers of men completing the weighed inventory: 64 at baseline; 54 at 6 months and 60 at 12 months after the start of the walking programme.

In some cases failure to complete inventories was due to extensive travelling during the measurement period. In other cases, there were difficulties in selecting a representative week. For example, one subject was a school-teacher, and reported large differences in diet between term-time and holidays. The baseline measurement was made during a holiday period, so it was decided that other measurements should be made during holidays too, to allow consistent results. However there was no holiday period at the time of the 6 month measurement, so this measurement was missed. Other men were unwilling to record food intakes at times of heavy work commitments, and so also missed one or more measurements.

These difficulties are possibly an unavoidable consequence of measuring a sample of men in full employment, who seemed to have great work commitments. Bramwell (1961); Marr (1965) and Bramwell & Marr (1970) studied adult men, and found 10% refused to record intakes, whilst 7% produced doubtful records. In this study 91% of inventories were completed, and there was no evidence for the reliability of those that were completed to be doubted. The overall compliance in this study thus seems to be slightly better than the above results.

No significant differences were found between intakes of men who completed all measurements, and those who did not complete measurement. This suggests that those who did complete measurement were representative of the group as a whole.

Dietary intakes at baseline

The men in this study were found to have a significantly lower cholesterol intake than the average for British men (OPCS 1991). There were also non-significant differences in total and saturated fat intakes: although fat intake tended to be higher in this study than in a random sample, saturated fat intake was lower.

These findings suggest that the diet of the men in this study may be "healthier" than that of the general population. This could be to some extent due to self-selection: the fact that they volunteered for this study indicates that these men have some interest in health or fitness. Another factor may be the exclusion of men with high TC levels, who might have had higher intakes of dietary cholesterol and saturated fat.

Influence of the walking programme on energy intake

Energy intake was not found to differ significantly between walkers and controls, or through the year. The response of energy intake did not differ significantly between walkers and controls. However there was a trend to a decrease in mean energy intake during the year, in walkers but not controls. These findings are hard to reconcile with the anticipated increase in energy expenditure of walkers during the year, whilst mean body fatness remained constant.

It is possible that walkers reduced their energy expenditure during the rest of the day, despite being instructed to complete the brisk walking in addition to the habitual amount of walking and other physical activity, or over-reported the amount of walking completed. However, as discussed in the previous chapter, changes in fitness were observed which would tend to indicate that some change in the amount of physical activity performed had occurred, and that this change was sufficient to induce physiological adaptation.

It is also possible that body composition or energy intake measurements were not sensitive enough to detect changes. According to Marr et al. (1971) and Nelson et al. (1989), 7 days of recording should be sufficient to correctly classify energy intake. However, it is possible that the recording period chosen was not representative, or that a trend occurred towards consistent under-reporting (as reported by Livingstone et al. 1990). However these explanations could not explain why the energy intake of walkers decreased, whilst that of controls remained constant, as these factors should affect both groups to an equal extent. It is possible that walkers felt that they were expected to show improvements, and modified their diet during the recording period to conform with these expectations. Another possibility is that the large commitment required by the walkers in completing the exercise programme allowed them less time, or made them less well-motivated, to complete inventories reliably.

Seasonality of energy intake

Although there were not significant changes in energy expenditure, there was a trend to lower mean energy expenditure at 6 months than at baseline and 12 months, in both walkers and controls. This trend might explain the lower body fatness at 6 months, described in the previous chapter. Seasonal variations in energy intake have also been reported by Marr (1965), with higher intakes in the winter. To determine whether this effect was seen in this study, energy intakes measured during summer months (June to August) were compared to those during winter months (December to February). Mean energy intake was lower in summer than the winter, being 10.8 and 11.5 MJ in summer and winter respectively. However, this difference was not

significant. The lack of significance could be due to the small proportion of measurements made during summer and winter, because of the timescale of the study (Table 6.1).

Composition of the diet during the walking programme

There were no significant differences between walkers and controls, differences through the year, or differences in response of walkers and controls, in carbohydrate, fat or alcohol intakes. There was however a different response in protein intake between walkers and controls. However change in protein intake alone would probably not be expected to influence blood lipid levels.

There was a significant increase in intake of polyunsaturated fat over the year, in both walkers and controls. Changes in PUFA intakes have been reported to influence blood lipid levels (Keys et al 1965; Hegsted et al 1986), and it was estimated that the observed change in PUFA intake might be expected to cause a decrease in TC of nearly 1 mg/dl. However changes in blood lipids observed in this study were small.

There are two possible explanations for these findings. Firstly, the intraindividual variation in PUFA intake is large, and Nelson et al (1989) have reported that intakes should be assessed for 15 - 30 days to achieve a valid measure of habitual diet. So the changes observed could be due simply to the 7 day weighed inventories used in this study failing to characterise habitual intakes, so these might not be real changes.

The second possibility is that involvement in the study has increased subjects' awareness of the association of diet with cardiovascular disease, resulting in either an actual modification in habitual intakes, or a temporary modification while diet was being recorded, and subjects were more than usually conscious of their diet.

Associations of dietary intakes with cardiovascular risk factors

There were generally fewer significant correlations between cardiovascular risk factors and dietary intakes than were observed with body composition or fat distribution. The nutrients which were related to blood lipids were alcohol (in both cross-sectional and longitudinal data) and carbohydrate and fibre (in cross-sectional data only).

Alcohol intake was positively correlated with TC, HDL, Apo A and triglyceride levels. Changes in alcohol intake were also positively correlated with changes in TC and Apo A. The association of alcohol with HDL and ApoA has been reported by Gordon et al (1981) and Camargo et al (1985). This association would provide a protective role for alcohol. If HDL levels are increased, without decreases

in other lipoprotein cholesterol subfractions, then this will cause an increase in TC, so the association of alcohol intake with TC is explained by the association with HDL. However the positive association with triglyceride levels would indicate an unfavourable role for alcohol.

The negative correlation of fibre intake with blood lipid levels agrees with the findings that increasing fibre intake can cause a lowering of blood lipid levels (Morris et al 1977; Judd & Truswell 1985).

Although changes in protein and PUFA intake were observed, levels of these changes were not associated with changes in cardiovascular risk factors. So it does not seem that the observed changes in these nutrients have influenced cardiovascular risk factor levels.

The methodology used in this study might have contributed to the lack of observed associations of dietary fats with blood lipid levels. The database used was based on The Fourth Edition of McCance and Widdowson's "The Composition of Foods" (Paul and Southgate, 1978). In this edition, the fatty acid composition of fried foods or foods containing fats, such as pastries, was not given. The fatty acid breakdown was therefore calculated from the average fat used in cooking by the men in this sample. This may have resulted in decreasing the inter-individual variation in saturated, monounsaturated, and polyunsaturated fatty acid intakes. This might in turn have resulted in correlations between fats and blood lipids being lower than if separate codes had been used for foods containing animal fats and vegetable fats. This shortcoming may be overcome in future, as the recently released Fifth Edition of "The Composition of Foods" contains separate codes for foods prepared with animal and vegetable fats.

Conclusions

There was very little change in dietary intakes during the brisk walking study. There was no change in energy intake in the walkers to account for the lack of any change in body composition, whilst energy expenditure had presumably increased.

The only nutrient in which the change in the walking group was different to that for the controls was protein. Intake of polyunsaturated fats increased in both groups over the year. Neither of these nutrients were found to be correlated with cardiovascular risk factors. The only nutrients which were observed to be associated with other cardiovascular risk factors were alcohol, fibre and carbohydrate.

Chapter 8

Discussion

The techniques for measurement of body composition, and its associations with risk factors for, and incidence of, cardiovascular disease were discussed in chapter 1. However in many of the studies of cardiovascular disease discussed, overweight was assessed from weight for height, or body mass index. As overweight is not necessarily associated with over-fatness or obesity, the use of weight for height or BMI as an indicator of body composition may confound the relationship between fatness and disease. For example, increased cardiovascular risk factors have been observed in obese, but not overweight lean, men (Segal et al. 1987). This highlights the need for measurement of body composition in such studies. These studies tend to require extremely large numbers of subjects, so some of the traditional methods for the measurement of body composition may be inappropriate, because they are too time-consuming or expensive.

Techniques which may be suitable for large-scale or field studies were therefore assessed in chapters 3, 4 and 5. Body composition techniques were then applied in a study of the influence of a programme of brisk walking on cardiovascular risk factors in chapter 6. Blood cholesterol and lipoproteins were assessed, to quantify any change in cardiovascular risk. As discussed in chapter 1, it has been suggested that the exercise induced change in lipoproteins may be mediated by changes in body composition. Blood pressure is also related to body composition (Kannel et al. 1967). So the relationships between changes in body composition and changes in other risk factors were examined.

Fat distribution has been reported to be an independent risk factor for cardiovascular disease (Lapidus 1984; Larsson 1984), as well as being associated with other risk factors such as blood pressure (Blair et al. 1983) and blood lipid levels (Albrink & Meigs 1964). For this reason, the relationship between changes in fat distribution and changes in other risk factors were also examined in chapter 6.

Dietary intakes were also monitored during the walking programme (chapter 7). As reviewed in chapter 1, a number of dietary components have been found to be related to incidence of cardiovascular disease, and other risk factors, especially blood lipid levels. So any changes in dietary composition were assessed, to determine whether these would confound the observed effects of exercise. Changes in total energy intake were also assessed to provide information on the effect of the walking programme on energy balance.

"Field" methods for measurement of human body composition

Near infra-red interactance

The application of near infra-red interactance for measurement of body composition is relatively recent, first being described by Conway et al. (1984). Since then it has not been extensively evaluated.

The measurement of body composition by near infra-red interactance was examined in chapters 3 and 4. Four groups of subjects were studied: 63 middle-aged men, 13 female athletes, 27 young men, and 27 young women.

Near infra-red interactance was found to underestimate fatness in all groups studied. The size of this underestimation was often sufficient to misclassify the fatness of the groups studied. The standard error of estimation of near infra-red interactance was found to be greater than that for skinfold thickness.

In the middle-aged men and female athletes, a significant trend towards increasing underestimation at increasing levels of fatness was observed. This trend was not apparent in the young men and women.

In all groups of subjects studied, the raw interactance data was found to be as well correlated with fatness as the near infra-red interactance estimate of fatness. Percentage fat by near infra-red interactance was calculated from equations containing variables such as weight and height, as well as interactance data, despite the fact that the inclusion of these variables would be expected to improve the estimation. This, in conjunction with the underestimation of fatness observed with these equations, suggests that these equations may be inappropriate in the groups studied.

The manufacturers state that equations were generated individually for each instrument, by regression with hydrostatic weighing data. However they do not give any information on the hydrostatic weighing methodology used, or the number or characteristics of subjects in the validation sample. Perhaps the estimation of body composition by near infra-red interactance could be improved by the use of better validated prediction equations.

Although interactance, and estimates of percentage fat by interactance, were found to be correlated with fatness assessed by other techniques, higher correlations were observed with biceps skinfold. So it seems that in the samples studied, biceps skinfold was a better predictor of fatness than interactance.

In chapter 4, interactance was also measured at a number of other sites, to determine whether any combination of sites would provide a better estimate of body composition relative to skinfold thickness than measurement at biceps site alone. However measurements at other sites were less well correlated with fatness than measurements at biceps site. No combination of several sites was found to improve estimation of body composition.

To further assess the relative merits of skinfold thickness and near infra-red interactance, the association between these techniques and subcutaneous adipose tissue thickness (SCATT) was assessed in chapter 5. The purpose of the study was to determine which method was the best predictor of subcutaneous adipose tissue thickness, and whether interactance was dependent also upon muscle thickness.

Optical density measurements were found to be correlated with SCATT, but not with muscle thickness. The correlation of optical densities with SCATT was considerably greater at the biceps site than the other sites measured. This supports the choice of the biceps site for measurement, and explains why no combination of several sites was found to predict body fatness better than measurement at biceps site alone. However skinfold thicknesses were found to be a better predictor of SCATT than NIRI.

Optical density measurements of Futrex are made at two wavelengths: one with a higher absorbance for fat, and the other with a higher absorbance for water. Theoretically, the relative size of the optical densities at two wavelengths should be able to give more detailed information of the composition of the measurement site. However the ratio of the two optical densities was found to be less well correlated with SCATT than a single optical density measurement. Adding the second optical density measurement was not found to improve the correlation with SCATT above that seen with the first.

The two wavelengths at which optical density is measured by Futrex are relatively close together in the near infra-red interactance spectrum: 940 and 950 nm. Conway et al. (1984) used a scanning spectrophotometer, with which they were able to obtain measurements at a much wider range of wavelengths. They found measurements at 916 and 1026 nm to best characterise body fatness. Similarly Conway and Norris (1987) used measurements at 867 and 914.5 nm.

It is therefore possible that the accuracy of the measurement could be improved by using more widely separated wavelengths, although this may cause equipment to be more expensive and less portable. However a measure of SCATT at a single site, however accurately it is obtained, will not result in an accurate measure of body fatness, because of differences in fat distribution.

Bioelectrical impedance

Reports of the accuracy of estimation of body composition from bioelectrical impedance have varied widely. To attempt to explain the discrepancy of these reports, various equations for estimation of body composition by impedance were compared in chapter 4.

Different prediction equations were found to produce significantly different estimates of body composition. The standard error of estimation from the different equations was found to be remarkably similar. However some equations were found to increasingly overestimate fatness at increasing levels of fatness, whilst this was not the case for other equations. So it seems that the use of different equations might contribute to the discrepancy between reports. Other causes may be differences between different instruments, as reported by Deurenberg et al. (1989), or differences resulting from electrode placement (Elsen et al. 1987).

The impedance of the body can also be influenced by a number of other factors. In chapter 4, strenuous exercise preceding measurement was found to significantly decrease impedance. Ambient and skin temperature have also been found to influence impedance (Caton et al. 1988), as has consumption of liquids (Khaled et al. 1988; Deurenberg et al. 1988). The findings of the latter author that beef tea influenced impedance whilst normal tea did not, brings up important questions as to the influence of electrolyte intake on impedance measurement. If electrolyte intake influences impedance, then systematic differences may be found between individuals or groups consuming different diets. In this case the time-scale of the effect of electrolytes on impedance needs to be quantified, to allow this effect to be controlled for.

All of the factors above, and the degree to which they were controlled for, may contribute to the different findings by different authors discussed in chapter 1. They may also influence the appropriateness of the technique for field studies, where the controlling of all these factors may be difficult or impossible.

Skinfold thicknesses

Skinfold thicknesses were assessed by comparison with hydrostatic weighing in chapter 3, and the standard error of estimation of the technique found to be acceptable (4.6 and 3.1 % fat in middle-aged men and female athletes respectively). In the middle-aged men a trend was found to increasing underestimation at increasing levels of fatness, although this trend was relatively weak.

The association between skinfold thickness and SCATT was examined in chapter 5. Skinfold thicknesses were generally found to be well correlated with SCATT from ultrasound. Significantly different skinfold compressibilities at different sites were observed. At the anterior thigh site in women, a negative compressibility was observed, indicating that the skinfold thickness was more than twice the SCATT by ultrasound. This was largely due extremely large skinfold measurements on two subjects, whilst SCATT (ultrasound) was near the mean for the group. In 4 other women in this study, no reliable measurement of thigh skinfold

could be made, due to the tautness of the tissue. These findings suggests that in some women, the amount of connective tissue present at this site may prevent accurate skinfold thickness measurement, so if this site was used for assessment of body composition or fat distribution, unreliable estimates may be obtained in some women.

Comparison of suitability of techniques for field studies

The estimation of body composition from skinfold thicknesses is cheap and acceptably accurate for most purposes. However, the measurement of skinfold thicknesses requires considerable observer training, and inter-observer differences have been reported. The technique was also slightly more time consuming than the other techniques measured, and less socially acceptable.

Near infra-red interactance was consistently found to provide less accurate estimates of both body fatness and SCATT than skinfold thickness. However measurement is extremely fast, socially acceptable, would require minimal observer training, and is not likely to be subject to inter-observer differences.

The accuracy of bioelectrical impedance was not directly assessed here. Reports of its accuracy are inconsistent, possibly because of the use of different instruments and prediction equations, which were found to perform differently in chapter 4. The method is fast, socially acceptable, and reasonably cheap. However the major limitation of its use for field studies is the controlling of the conditions discussed above, which may be especially difficult in field conditions. The use of different methodologies may also mean that results may often not be comparable.

Overall, it would seem that measurement of skinfold thickness is likely to be the most appropriate technique for field studies, so long as observers are well trained. However if the accuracy of near infra-red interactance could be improved, and suitable prediction equations developed, near infra-red interactance might be an ideal tool for such studies.

Influence of a year-long programme of brisk walking

Influence of brisk walking programme on body composition, fat distribution and energy intake

Participation in the walking programme was not found to influence body weight or fatness, or energy intake. As men were instructed to complete the brisk walking of the program in addition to any other walking or physical activity to which they were accustomed, it would have been expected that their total daily energy expenditure should have increased. This would be expected to produce an adaptation in either energy stores or energy intake.

It is possible that the total daily energy expenditure did not change. This could occur if the walkers had not been complying with the walking programme, or if they decreased the amount of physical activity performed during the rest of the day.

The different changes in fitness of the walking group and control group, described in chapter 6, suggest that the walkers had been complying, and had increased the amount of physical activity performed sufficiently to induce physiological change. The men were all employed in sedentary occupations, and were not engaged in any programme of physical activity prior to commencing the walking programme. It therefore seems unlikely that the men could have decreased physical activity during the rest of the day sufficiently to counteract the increased energy expenditure of brisk walking 30 minutes per day.

Other possibilities are that the assessment of either body composition or energy intake was not sensitive enough to detect changes. Body composition was assessed by two techniques: hydrostatic weighing and skinfold thickness, and body weight was also monitored during the walking programme. All these methods separately confirmed that no change in body composition occurred. Hydrostatic weighing has been reported to be subject to bias in assessment of body composition during exercise programmes, in that athletes have been reported to have denser lean tissue than contemporaries of the same age (MacDougall et al. 1983). So involvement in an exercise program could cause an increase in lean tissue density. However this would result in an underestimation of body fatness, rather than an overestimation which might conceal a decrease in body fatness. However, because the density of muscle is less than that of the mineral content of the body, muscular development has been associated with a decrease in the density of the fat-free mass (Womersley et al., 1976). This factor may cause an overestimation of fatness after an exercise programme, if muscularity increased.

Similarly, changes in body weight may be subject to bias during an exercise program if muscularity changes, as it is impossible to determine whether a change in weight is due to a change in muscularity or fatness. So a decrease in fat mass may be obscured by an increase in muscle mass.

There is no evidence that estimation of body composition from skinfold thickness is subject to a consistent bias as a result of an exercise program. However, in chapter 6, changes in fat distribution as a result of the walking programme were observed: skinfold thicknesses of the lower limb decreased in the walking group. So it is possible that the decrease in fatness of the lower limb produced a decrease in total fatness, which was not detected by measurements of skinfold thicknesses on the upper body, where no decrease in fatness occurred.

So it is possible that changes in fat distribution may have concealed changes in fatness according to skinfold thicknesses, and changes in muscularity concealed changes in fatness according to hydrostatic weighing and body weight. The changes in cross-sectional muscle and bone area of the lower limb were estimated in chapter 6. This did not change differently in walkers and controls at the thigh site, whilst a 2% increase occurred at the calf site. This change in muscularity does not seem sufficient to greatly influence estimates of body composition. Similarly, changes in skinfold thicknesses of the lower limb were small, and unlikely to contribute to greatly to changes in total fatness. So whilst the possibility of decreases in fatness being underestimated after involvement in the walking programme exists, it seems likely that this underestimation is small.

A 7-day weighed inventory has been reported to be sufficient to correctly rank individuals on the basis of habitual energy intake (Marr et al. 1971; Nelson et al. 1989). However Livingstone et al. (1990) reported considerable underestimation of energy intake using the weighed inventory technique in studies of energy balance. Such an underestimation would be expected to affect walking and control groups, and baseline and 12-month measurements, to an equal extent. It would therefore not be expected to be responsible for concealing a change in energy intake in the walking group.

One factor which may affect the walking and control groups differently is the involvement in the walking programme. All the subjects were men in full employment, and many found time to be a restraint on the amount of walking performed. It is possible that men in the walking group completed the inventories less reliably than men in the control group, because of the amount of extra time and commitment involved in completing the walking programme.

The lack of any change in body composition or energy intake of the walking group could thus be ascribed to a number of factors. It is impossible to determine whether this lack of change could be due to an adaptation in energy balance, or to the factors described above. However, of the factors above, a decrease in physical activity of the walkers during the rest of the day, and failure of weighed inventories to detect changes in habitual energy intake seem most likely.

Seasonal variation in body fatness and energy intake

Changes in body weight and body fatness during the year were observed, in both walking and control groups. Body weight and fatness were significantly lower at 3 month and 6 month testing than at baseline and 12 months. Changes in energy intake during the year were not significant, although mean energy intakes were lower at 6 months than at baseline and 12 months. The greatest changes in fatness were

observed at 3 month testing, so it is possible that if energy intake had been assessed at 3 months, significant changes might have been observed. It therefore seems likely that the changes in fatness during the year were due to a changes in energy intake.

This variation during the year could be due to a seasonal effect. Such effects have been observed in developing countries, due to large variations in physical activity and in the availability of food. These influences are likely to be of less importance in men employed in sedentary occupations, in affluent societies. Nevertheless, Marr (1966) and Hartog et al. (1965) observed lower energy intakes in summer than winter, in affluent societies. The lowest levels of fatness in this study were observed in the summer, as were lowest energy intakes.

It is also possible that this effect is due to the influence of involvement in the study. Men were not given any feedback until the end of the study, to prevent any changes in lifestyle being induced as a consequence. However, they seemed to show an increased interest in the effect of diet and lifestyle after joining the study. It is possible that this increased interest caused temporary changes in lifestyle, thus causing some of the changes in fatness and energy intake observed above.

Influence of brisk walking programme on cardiovascular risk factors

The brisk walking programme was not found to have any significant influence on levels of cardiovascular risk factors. This could be due to the existence of a threshold level of exercise below which changes are not induced (as reported by Paffenbarger et al. 1978, and Haskell et al. 1986) which was not met in this study. Alternatively, the findings of Wood et al. (1987) that exercise induced changes in blood lipid levels are mediated by changes in body composition suggest that the lack of change in blood lipid levels could be due to the lack of change in body composition.

Changes in the composition of the diet during the walking programme

The composition of the diet was monitored during the walking programme, to determine whether any changes occurred which might influence blood lipid levels. There were very few changes in nutrient intakes during the year. Intake of polyunsaturated fatty acids and P:S ratio increased in both walking and control groups: this may be a response to increased awareness of the influence of diet on cardiovascular disease. However, since this change was observed in both groups, this finding is unlikely to have produced differential changes in blood lipids in the two groups, and so will not have confounded the influence of the walking programme on blood lipids.

The change in protein intake was significantly different between the two groups. However this change in protein intake occurred without significant changes in intakes of fats or other nutrients. This change on its own is also unlikely to have confused the results.

Association of body composition, fat distribution and dietary intakes with cardiovascular risk factors

Cross-sectional comparison

The correlations of body composition, fat distribution and dietary intakes with levels of other cardiovascular risk factors at baseline were examined. A number of significant associations were observed, despite the fact that the sample size was smaller than that usually considered necessary for cross-sectional comparisons. Perhaps this is because the homogeneity of this group resulted in decreased inter-individual variability, which has been reported to be a confounding factor in cross-sectional studies.

A number of significant associations were observed between body composition or fat distribution and blood lipid levels. Results were in agreement with the literature, as discussed in chapter 6. Indices of both total and central fatness were found to be associated with a less favourable risk factor profile.

Fewer significant associations were observed between blood lipids and dietary intakes. Interestingly, the nutrients associated with blood lipid levels were carbohydrate, alcohol and fibre, rather than fats, which have been reported to be strongly associated with blood lipid levels (Keys et al. 1980). However, some larger cross-sectional studies have observed no significant associations between fatty acid intakes and blood lipid levels, maybe because of large variation between individuals in intrinsic blood lipid levels (Stallones et al., 1983). Because of the small size of the group studied here, the lack of a significant association can not be considered important, and may be partly due to the methodology used, as discussed in chapter 7.

Longitudinal comparison

Changes in body composition, fat distribution, and dietary intakes during the brisk walking programme were related to changes in other cardiovascular risk factors to allow a cross-sectional comparison. These changes were all relatively small, and whilst involvement in the walking programme may have contributed to the changes, they were not significantly different in the walking group and the control group.

Although the walking programme did not significantly alter blood lipid and lipoprotein levels, small changes in body composition and fat distribution during the year were significantly correlated with changes in levels of blood lipids and

lipoproteins. This finding would tend to support the hypothesis of Wood et al. (1988) and Tran and Weltman (1985) that the exercise induced changes in blood lipids and lipoproteins are mediated by changes in body composition. It is also possible that changes in fat distribution play an additional part, as changes in fat distribution were also associated with changes in blood lipid levels.

There was very little association between changes in dietary intakes during the year with blood lipid and lipoprotein levels. However changes in dietary intakes were small: men had been instructed not to alter their diets. The difficulty of characterising habitual intakes of some nutrients with a 7 day weighed inventory may also have decreased associations. However the lack of association provides confirmation that changes in diet during the year did not affect the response of blood lipids.

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Appendix I

Statement of Informed consent

I have read and understood the description of the study given, and agree to take part. I realise that I am able to withdraw from the study at any stage, without having to give my reasons for withdrawal.

Signed..... Date.....

Appendix II

Leaflet for recruitment of subjects for brisk walking study

Loughborough University of Technology BRITISH HEART FOUNDATION PROJECT

An opportunity to participate in a major study of the effects of regular exercise on risk factors for coronary heart disease

We are looking for men aged between 42 and 59 from the Loughborough area to take part in a study funded by the British Heart Foundation. The purpose of the study is to examine the influence of regular, brisk walking on fitness, body fatness, and the transport of cholesterol in the blood. Subjects will follow a prescribed programme of brisk walking for one year, fitting the walking into their daily routine. A series of tests to evaluate changes will be conducted at baseline and after 3, 6 and 12 months, involving several visits to the laboratory on each occasion.

If you (i) are a non-smoker or an occasional smoker, (ii) currently take no regular exercise and (iii) are aged 42 to 59, contact Dr Adrienne Hardman (Loughborough 223265), Professor Peter Jones (Loughborough 223003) or Dr Nick Norgan (Loughborough 223009) as soon as possible for more information.



Appendix III

Description of brisk walking study given to potential volunteers

BRITISH HEART FOUNDATION PROJECT - 1989

Purpose of the Study:

The purpose of the study is to examine the influence of regular brisk walking on i) physical fitness, ii) the amount and distribution of body fat, iii) the concentration in blood of cholesterol and other markers of coronary heart disease risk and iv) psychological well-being.

Criteria for acceptance into the Study

1. Neither currently on a programme of regular exercise nor employed in a strenuous job.
2. Not at high risk of coronary heart disease (CHD), i.e. non-smoker or occasional smoker, free of known cardiovascular disease, blood pressure below 160/95 mm Hg, plasma cholesterol less than 6.7 mmol/l.
3. Willing to accept random allocation to either the Walking Group or to the Control Group, with a 2:1 chance of being in the Walking Group. A programme of walking and monitoring will be conducted for the Control Group after the present study is complete.

Explanation of the Procedures:

- A) Four exercise tests will be undertaken, three of these in the laboratory - after a practise session to become familiar with the apparatus.
- i) Test one involves walking uphill on a motorised treadmill at a fixed speed for 16 minutes. The gradient will be increased at the end of each four minute period. Heart rate will be recorded throughout from three chest electrodes and expired air will be collected at intervals using a mouthpiece and respiratory valve.

- ii) The test described above will be repeated but using treadmill gradients selected to represent 50%, 60%, 70% and 80% of each man's estimated maximal capacity. Five very small (25 μ l) samples of blood will be obtained by pricking the thumb, before exercise and at the end of each 4-minute period of walking.
 - iii) On another occasion, subjects will walk for 20 min on the motorised treadmill at a gradient selected to be equivalent to 70% of their own estimated maximum capacity. Heart rate and expired air will be collected as described above and thumbprick blood samples taken before and on three occasions during the test. Subjects will also exhale maximally into an instrument which measures lung function before and after exercise.
 - iv) Finally, each subject will walk one mile outside, at a brisk pace. During the walk heart rate will be recorded from two chest electrodes using short range telemetry.
- B) The amount of fat under the skin will be determined using calipers at 8 different sites and a number of body girths will be measured. Total body fat will be determined from body density derived by underwater weighing. This is done in a tank of warm water (36° C) and involves breathing out maximally through a snorkel whilst submerged. The volume of air left in the lungs after breathing out as far as possible, is determined by taking three deep breaths from a bag of 100% oxygen fitted to the snorkel.
- C) Two, 20 ml samples of blood will be taken from a vein in the arm for the analysis of plasma cholesterol and other biochemical markers for CHD. This needs to be done in the morning after a 12 hour fast. Haemoglobin concentration and the proportion of red cells in the blood will also be measured in three blood samples.
- D) Resting blood pressure will be measured.
- E) All food and drink consumed over a period of one week will be weighed and recorded so that average daily nutrient and energy intake can be determined.

- F) Several questionnaires will be completed. These can all be done at home. They ask questions about i) your current physical activity level, ii) present and past smoking habits and, iii) how you feel about yourself (self-esteem).

Walking:

Those allocated to the Walking Group will follow a brisk walking programme. This will be individually prescribed on a fortnightly basis and will increase to an average of about three miles a day by the end of six months and thereafter. The first session will be on the University site so that each man's self-selected brisk pace can be determined.

The Control Group will be asked to maintain their habitual lifestyle until this trial is complete. After one year the subjects in this group will embark on the brisk walking programme described above.

Repeated measurements:

The measurements described above, with the exception of the underwater weighing, will be repeated after three months of regular walking and again after six months. After one year all measurements, including the underwater weighing, will be repeated.

Risks and discomforts:

Although all exercise testing will be at a submaximal level, the possibility exists that, very occasionally, certain changes may occur during the test. They include abnormal blood pressure, fainting, disorder of the heart beat and, in very rare instances, heart attack. The preliminary screening undertaken before the study is designed to minimise such risks. Sampling of venous blood may cause minor bruising or haematoma (a small accumulation of blood under the skin). All experiments will be conducted by trained and experienced staff and closely monitored. You should feel free to ask for more information or explanation at any time.

Benefits:

This trial is being conducted because it is not known whether regular, brisk walking can influence fitness and/or health in men. There can therefore be no guarantee that you personally will benefit from taking part. We do, however, have every expectation that you will enjoy contributing to a worthwhile study. The very least you will gain is a greater understanding of the body's responses to exercise.

If you need more information please contact David Stensel (4326), Katherine Brooke-Wavell (3005) or any other member of the BHF Research Group.

Dr Adrienne Hardman (3265)

Dr Peter Jones (3003)

Dr Nick Norgan (3009)

Appendix IV

Instructions and forms for recording of weighed inventories

Instruction sheet

INSTRUCTIONS FOR THE USE OF THE FOOD DIARY

It is important that you weigh and record everything that you eat and drink during the week of the survey, following the instructions below.

- Please (i) start a separate page for each day
(ii) start a separate line for each item.

Column 1

Record meal, and time and place of eating.

Column 2

Describe each item as accurately as possible, stating where relevant:

- (i) type and brand
- (ii) whether food is fresh, dried, canned, frozen, salted, smoked, etc.
- (iii) whether food is cooked, if so give method of cooking e.g. fried, baked etc.

Column 3

Record the weight of each item, after cooking:

- (i) Place scales on level surface
- (ii) Place plate or container on top of scales
- (iii) Press zero button. (This will turn on scales)
- (iv) Once zero appears, add first item of food
- (v) Record weight displayed
- (vi) Press zero button before weighing next item
- (vii) If the scale is not used for twenty seconds, a red flashing light will appear, indicating that it is about to switch off. If the zero button is pressed, the last weight will be recalled, if not it will switch itself off after one minute.
- (viii) Four decimal points appearing along the bottom of the display indicate that the batteries are becoming low, so please replace them with those provided.

Wherever possible, record weights in grams. If this is not possible, record weights in household measures (e.g. sugar or jam in teaspoons, stating whether level, rounded, or heaped).

Column 4

Record the weight of any leftovers, such as food remaining on plate, weight of container in which food has been weighed, apple cores etc.

Columns 5 and 6

Please leave blank.

If a food consists of several ingredients, please list each on a separate line i. e. instead of writing 'one cheese sandwich', record separately the weights of the bread, margarine, cheese etc. For made-up dishes, please supply recipes, or proportions of ingredients, wherever possible.

Please remember to record all drinks, as well as food, giving weights where possible, or volumes if these are known. Record separately weights of added milk and sugar.

Items which are used several times each day (e. g. butter, sugar, milk) may be kept in a container which need only be weighed at the beginning and end of each day, and on refilling. Containers are provided for this purpose:

- (i) Fill the container with the food
- (ii) At the beginning of the day, weigh the container with the food, describing the food in column 2 and recording the weight in column 3, as previously.
- (iii) Do not record the food on subsequent occasions on which it is used from the container.
- (iv) At the end of the day, record the total weight of container and food remaining in the container in the 'leftovers' column (column 4).

Please ensure that you are the only person to use food from the container.

An example sheet is given overleaf. If you have any queries or difficulties, please contact me on Loughborough 223005.

Katherine Brooke-Wavell

Example Recording Sheet

Name..... Date.....

Number..... Day of week.....

1. Meal, time and place	2. Precise description of food and drink, and method of cooking	3. Weight (g)	4. Container /leftovers (g)	5.	6. Leave Blank
<i>Breakfast</i>	<i>Cornflakes (Kelloggs)</i>	28			
<i>8.30 am</i>	<i>Milk (Sainsbury's virtually fat-free)</i>	48			
<i>home</i>	<i>Bread (Mother's Pride, large white sliced, toasted)</i>	76			
	<i>Robinsons lemon marmalade</i>	12			
	<i>Coffee, instant</i>	2			
	<i>Milk (whole, pasteurised)</i>	10			
<i>Lunch</i>	<i>Cheese (cheddar)</i>	55			
<i>1 pm</i>	<i>Bread (white, crusty)</i>	76			
<i>Pub</i>	<i>Butter</i>	4			
<i>(the Ship)</i>	<i>Chutney 2 teaspoons</i>				
	<i>Kronenburg lager 1 pint</i>				
<i>Snack</i>	<i>Coffee (instant)</i>	2			
<i>3.30 pm</i>	<i>Coffee-mate</i>	6			
<i>Office</i>	<i>Mars Bar</i>	35			
	<i>Apple</i>	76	8-core		
<i>Dinner</i>	<i>Beefburger (Bejam, frozen, grilled)</i>	102			
<i>6.30 pm</i>	<i>Potatoes, old, roast</i>	322	74-		
<i>Home</i>	<i>Peas (Bird Eye frozen, boiled)</i>	50	leftover		
	<i>Yoghurt (Ski, strawberry)</i>	162	10-carton		
<i>Snack 8.30</i>	<i>Milk chocolate digestive biscuits</i>	56			
<i>During day</i>	<i>Flora margarine (in container)</i>	215	195		
	<i>Sugar (in container)</i>	106	94		

