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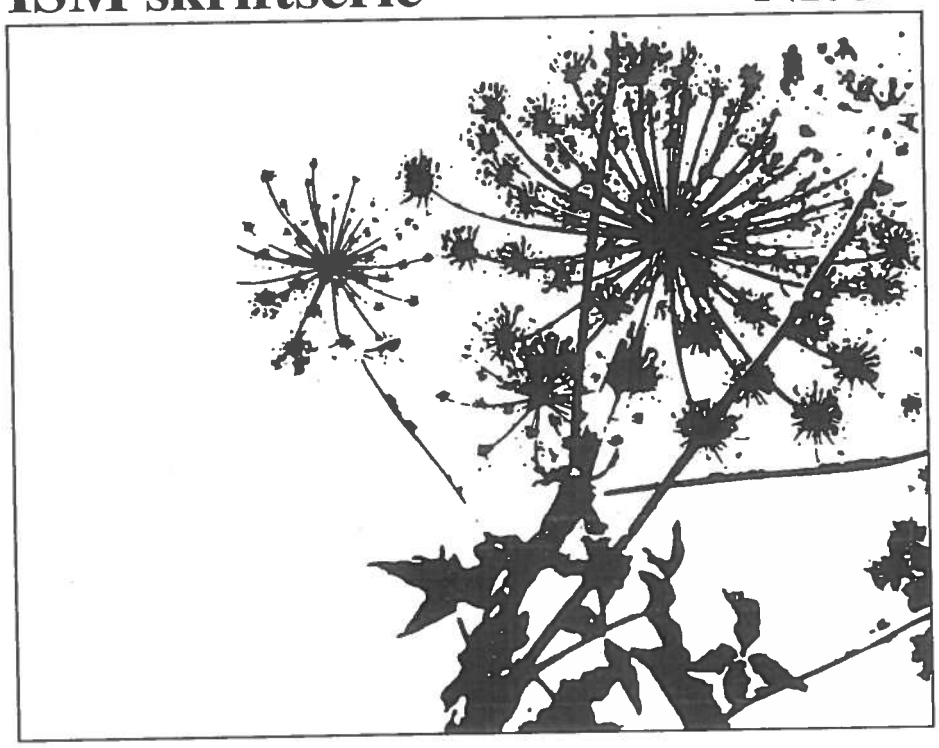

Sex steroids, bone loss and non-vertebral fractures in women and men

The Tromsø Study

By Åshild Bjørnerem

Tromsø 2007

The **Research Council** of Norway

Institute of Community Medicine
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List of papers

This thesis is based on the following papers, referred to by their Roman numerals in the text.

I. Bjørnerem Å, Straume B, Midtby M, Fønnebø V, Sundsfjord J, Svartberg J, Acharya G, Øian P, Berntsen GKR. Endogenous Sex Hormones in Relation to Age, Sex, Lifestyle Factors, and Chronic Diseases in a General Population: The Tromsø Study. J Clin Endocrinol Metab 2004; 89:6039-6047

II. Bjørnerem Å, Straume B, Øian P, Berntsen GKR. Seasonal Variation of Total and Free Estradiol, Follicle Stimulating Hormone and Dehydroepiandrosterone Sulfate in a general population. J Clin Endocrinol Metab 2006; 91:3798-802

III. Bjørnerem Å, Emaus N, Berntsen GKR, Joakimsen RM, Fønnebø V, Øian P, Seeman E, Straume B. Circulating Estradiol and Sex Hormone-Binding Globulin Predicts Bone Loss in Women and Men. The Tromsø Study. Submitted

IV. Bjørnerem Å, Ahmed LA, Joakimsen RM, Berntsen GKR, Fønnebø V, Jørgensen L, Øian P, Seeman E, Straume B. A Prospective Study of Sex Steroids, Sex Hormone-Binding Globulin and Non-Vertebral Fractures in Women and Men in the Tromsø Study. Submitted

Acknowledgements

My background is in Obstetrics and Gynaecology at the University Hospital of North Norway, and I knew little about statistics and epidemiology when I first came to the Institute of Community Medicine (ISM). I learned to appreciate epidemiology first as a student at Master of Public Health course and later as a PhD student.

I would like to thank the Tromsø population who attended and made the Tromsø Study possible. I thank Gro Berntsen for introducing me to ISM and this project, and all co-authors in the Tromsø Osteoporosis Study, Ragnar M. Joakimsen, Nina Emaus, Luai Ahmed, Lone Jørgensen and Vinjar Fønnebø for all your good contributions. I am very grateful to Bjørn Straume for spending plenty of time on teaching me so much on methods, statistics and epidemiology. A special thank to Tom Wilsgaard for clear and excellent statistical advising, to Bjarne K. Jacobsen for teaching me how to run SAS programs, and Jarle Mathiassen and Sriharan Sivasingarajah for IT-support. I want to thank the ISM for providing good working conditions and for letting me go to Melbourne in Australia on 1 year study leave. That was a great professional, social and cultural experience for me and my family. It was a pleasure to work with Ego Seeman. I am very grateful for your excellent teaching on bone fragility and how to write papers, Ego and we had a lot of fun.

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Åshild Bjørnerem, Tromsø, October 2006

Abbreviations

ANOVA /ANCOVA– analysis of variance / analysis of covariance

AFP /EFP – aluminium forearm phantom / European Forearm Phantom

BMD – bone mineral density

BMI - body mass index

BMU – basic multicellular unit

CV- coefficient of variation

CYP-19 – a gene which encodes for the aromatase enzyme

DHEA / DHEAS – dehydroepiandrosterone / dehydroepiandrosterone sulphate

DXA / SXA - dual x-ray absorptiometry / single x-ray absorptiometry

FSH – follicle stimulating hormone

FIS – Family Intervention Study

GH / IGF-I – growth hormone / insulin-like growth factor I

HR – hazard ratio

HRT – hormone replacement therapy

IHD – ischemic heart disease

IL - interleukin

M-CSF - macrophage colony-stimulating factor

OPG – osteoprotegerin

PTH - parathyroid hormone

RANK / RANKL – receptor activator of nuclear factor-kappa B / RANK ligand

SD – standard deviation

SHBG – sex hormone-binding globulin

TNF – tumor necrosis factors

TROST – The Tromsø Osteoporosis Study

1. Introduction

1.1 Background

When this thesis was planned in 2000-2001 it was well known that bone loss accelerates after menopause, and is prevented by using hormone replacement therapy (HRT) [1-6]. Case reports of young men with estrogen receptor dysfunction or aromatase deficiency showed that estrogen was important for normal growth and maturation of the male skeleton [7, 8]. However, there were few prospective studies examining the contribution of endogenous sex steroids on bone loss [9] and fracture risk in women [10-12], and no prospective study in men.

We wanted to know if levels of sex steroids were relevant as predictors of bone loss and fractures and whether this knowledge could be useful for developing future treatment by low dose of HRT. During the work on this thesis results from a large randomized controlled trial assessed the risks and benefits of estrogen plus progestin treatment in 16,608 healthy postmenopausal women [13]. The investigators concluded that the overall health risks exceeded benefits from the use of estrogen plus progestin in this group. They estimated hazard ratios with 95% confidence interval as follows: coronary heart disease 1.29 (1.02-1.63); breast cancer 1.26 (1.00-1.59); stroke 1.41 (1.07-1.85); pulmonary embolism 2.13 (1.39-3.25); colorectal cancer 0.63 (0.43-0.92); endometrial cancer 0.83 (0.47-1.47) and hip fracture 0.66 (0.45-0.98). This publication led to a reassessment of the role of HRT, which was no longer recommended in women for fracture risk reduction alone [14-16].

Left unresolved was the question whether women and men with low levels of circulating sex steroids have higher risk of bone loss or fractures. This was still relevant for our understanding of the contribution of sex steroids on bone fragility. From a clinical point of view it was interesting to know, if sex steroids are relevant predictors of bone loss or fractures, if these measurements could be useful in signalling the need for further investigation or treatment.

1.2 Epidemiology of osteoporosis

Osteoporosis is a systemic skeletal condition characterized by low bone mass and microarchitectural deterioration of bone tissue, with a consequent increase in bone fragility and susceptibility to fracture [17, 18]. Microarchitectural deterioration of bone tissue refers to the destruction of the trabecular network, with trabecular thinning and disconnection. These changes contribute substantially to reduced bone strength [19]. Since there are no satisfactory clinical tools available to assess bone quality, the diagnosis of osteoporosis depends upon the measurement of skeletal mass [18-21]. The bone mineral density (BMD) is the amount of minerals in the specific site scanned divided by the area measured. The World Health Organization definition of osteoporosis is a hip bone mineral density (BMD) 2.5 standard deviation (SD) or more below the young adult female mean [18, 22]. Osteopenia is defined as a hip BMD 1 SD or more below the young adult female mean, but less than 2.5 SD below this value. There is no consensus on a definition of osteoporosis in men, but the same absolute value for BMD as in women, are used in men [18].

In women, most of the bone loss occurs after the menopause, and the proportion of women having osteoporosis (bone fragility) increases greatly by age [23]. The prevalence of osteoporosis varies from 2 to 45% in postmenopausal women and from 0 to 36% in men [24]. The discrepancies relate mainly to differences in the specific young normal means and SD for different measurement methods, and for different patterns of age-related bone loss at various skeletal sites [25]. The accuracy of the techniques is not how closely they measure BMD, but their sensitivity and specificity to predict fractures [18]. Although the risk of fracture is similar in women and men with same BMD levels, fewer men than women develop the very low levels of BMD with greatest risk of fractures [25, 26]. Instead of degree of osteoporosis, the absolute fracture risk is recommended as the main basis for clinical treatment [18, 20, 21, 27].

1.3 Epidemiology of fractures

The occurring fractures are the clinical significance of osteoporosis [18]. Although not all fractures are due to osteoporosis, almost all type of fractures are increased in patients with low BMD [18, 28]. The BMD measure predicts fracture risk and in a meta-analysis by Marshall et al. the fracture risk approximately doubled for each SD decrease in BMD [29]. The fractures at any location were best predicted by BMD measurements from the same anatomical site, but no site was superior with respect to prediction of all types of fragility fractures [29].

Fracture of the vertebrae, proximal femur and distal forearm have long been regarded as the fragility fractures [28]. Hip-fractures, the most serious outcome of osteoporosis, cause most suffering, expenses and are associated with increased morbidity and mortality [28, 30]. As the world's population ages, the frequency of fractures will increase and this is best documented on hip fractures [28]. The incidence of hip fractures increase exponentially with age in both women and men. The age-related decrease in BMD and increase in falls, are important components of this dramatic rise in fracture risk [28]. Women have more bone loss and falls than men, and their risk of fracture is therefore about the double compared to men. Women also live longer than men, so about three-quarters of the hip fractures occurs in women [28].

There is a large variation in fracture risks world wide. The incidence of hip fractures in the Norwegian population is among the highest ever reported [31, 32], and the ten-year probability of hip fracture varied more than 15-fold between Norway and Chile [33]. The lifetime risk of a hip fracture from age 50 years varied from 1% in women from Turkey to 28.5% in women from Sweden [33], and is estimated to be about 17% for white women, and 6% for white men in the USA [28].

2 Pathogenesis of Bone Fragility

2.1 Bone composition, structure and function

Understanding the pathogenesis of bone fragility requires the study of the two properties that determine bone strength; its material composition and its structural design [34, 35]. Bone has unique properties that can meet the contradictory needs of strength and lightness, stiffness and flexibility [19]. The skeleton is well adapted for the functions of structural support, movement, protection of internal organs, a reservoir of calcium and phosphate and formation of blood cells [36].

There are two types of bone tissue: cortical and trabecular. About 80% of the skeleton is composed of cortical bone, located mainly in the shafts of long bones and surfaces of flat bones [36]. This compact bone consists of tightly packed osteons that consist of Haversian canals surrounded by concentric rings of collagen fibres arranged in lamellas, and osteocytes are located in lacunae between the rings [36]. The crystals of calcium hydroxyapatite stiffening the fibres of type 1 collagen in the extracellular matrix confer resistance to bending. Human bone is about 60% mineralized. The collagen weave confers flexibility and make bone able to absorb energy by reversible deformation during loading. Strength and lightness are also achieved by geometric structure [19]. For loading and movement, the needed stiffness and lightness are achieved by the tubular structure of long bones, where the cortical mass is placed distant from the central long axis, conferring greater resistance to bending [19, 34].

Trabecular bone is located at the ends of long bones, vertebral bodies and in the inner parts of flat bones [36]. It is lighter and less dense than cortical bone and consists of interconnecting plates and bars within which lies cavities that contain bone marrow. The trabeculae are organized to provide maximum strength similar to braces used to support a building. This structure results in a much higher surface-to-volume ratio of trabecular than cortical bone, and the potential for metabolic activity is therefore higher. The vertebral bodies

achieve lightness and flexibility by their sponge-like structure and can absorb more energy by deforming more before cracking than can long bones [34]. The skeleton is defined externally by its outer (periosteal) surface and by its components of inner surfaces: endocortical, intracortical and trabecular [19]. As the bone grows in length, bone size, bone mass and cortical thickness increase because periosteal apposition increases the diameter of the bone, and periosteal apposition is greater than the net endosteal resorption [37].

Osteoblasts, osteocytes and osteoclasts are the three types of bone cells involved in the surface phenomenon responsible for the adaptation of bone; modelling (construction) and remodelling (reconstruction) [36]. The purpose during growth is to establish the skeleton's peak bone strength and in adulthood to maintain bone strength [34, 36]. Osteoblasts are responsible for the formation and mineralization of bone, and may become bone-lining cells or osteocytes [36]. Osteocytes are mature bone cells within the bone matrix connected to one another and to osteoblasts by an extensive canalicular network. These cells are believed to play a central role in the response to mechanical stimuli, and initiating an appropriate remodelling response. Osteoclasts are large, multinucleated bone-resorbing cells, and they are formed by fusion of monocytes/macrophage precursors.

During bone remodelling, the lining cells disappear and teams of osteoclasts resorb a volume of bone, leaving a focal resorption cavity on the endosteal surface or a cutting cone within the cortex [19, 34, 36]. After a delay, osteoblasts fill the cavity with the volume of new bone that undergoes rapid primary then slower secondary mineralization. In human adults, about 20% of the trabecular bone surface is undergoing remodelling at any given time, and a cycle is believed to last between 2-8 months [36]. Provided that the same volumes of bone are removed by osteoclasts and replaced by osteoblasts within each basic multicellular unit (BMU), no net bone loss or structural damage arises [19, 34, 36]. For bone to be lost, the volume of bone resorbed must be greater than the volume of bone formed.

2.2 Peak bone mass

During growth, area BMD increases, suggesting incorrectly that bone density has risen [19, 35]. Enhancement of area BMD is mainly caused by the rise in bone size, which results in a proportional rise in the amount of mineralized bone, whereas the volumetric BMD of the whole bone remains constant or increases modestly [19, 37-39]. The bone is bigger than it was, but not denser. Area BMD provides only a two dimensional view of the three dimensional mineralized mass of bone. The length and width of the scanned bone is known but not its depth. "Area" is often dropped for the sake of brevity but at the price of understanding [35]. Since results of densitometry are a widely used endpoint in clinical practice and research, knowledge of the limitations of the method is needed for understanding of the complexity of the structural basis underlying the pathogenesis of bone fragility.

Skeletal size and volumetric BMD are similar in prepubertal girls and boys, and sex steroids are responsible for the maturation and the sexual dimorphism of the skeleton [40]. Puberty is triggered by increased secretion of gonadotropin-releasing hormone by the hypothalamus, leading to increases in gonadal secretion of sex steroids [40]. The increase in growth hormone (GH), insulin-like growth factor I (IGF-I) and estrogen act co-ordinately and support the pubertal growth spurt. The increase in estrogen is responsible for the pubertal growth spurt and epiphyseal closure in both sexes. Men with mutation in estrogen receptors or aromatase genes do not undergo rapid adolescent growth, and have elongated limb bones due to failure of the epiphyses to fuse, despite normal testosterone levels [7, 8, 41, 42].

Sex differences in bone width are established during puberty. Cortical width increases by periosteal bone formation in boys, and by less periosteal bone formation but by more endocortical apposition in girls [43-45]. Androgens, GH and IGF-I stimulate periosteal apposition in boys, whereas estrogens inhibit periosteal apposition, and it stimulates endosteal apposition in girls [19]. Thus, men build longer and wider long bones with only a slightly

thicker cortex than do women [19, 43, 44]. The cortical mass is placed further from the neutral axis of the long bone in men, conferring greater resistance to bending by the correspondingly larger muscle mass [19]. The greater bone mass in postpubertal boys than girls is likely to be due to pubertal increase in testosterone, because increase in GH and IGF-I is similar. Boys have 2 more years of prepubertal growth because of their later puberty, at age 14 rather than age 12 as in girls, and boys have a growth spurt that last for 4 yr rather than 3 yr in girls [40]. These differences largely account for the 10% greater statural height and the 25% greater peak bone mass due to the greater bone size achieved by men. Thus, growth builds a bigger but not a denser skeleton in men than in women, and the larger skeleton in men produces a stronger bone [19].

2.3 Effects of sex steroids on bone loss

After closure of the epiphyses, and cessation of growth in length, periosteal apposition continues slowly so the bone continues to increase in diameter [19, 37]. Bone mass decreases as age advances, because periosteal apposition only partly offset endosteal resorption [19]. Bone remodelling continues on the endosteal surfaces during ageing, and bones do become fragile because the cellular mechanisms responsible for constructing bone during growth (modelling) and reconstructing it during adulthood (remodelling) fail to maintain the pristine material and structural properties [19]. Bone loss starts during early adulthood in women and in men [46-50], and is probably the result of an early reduction in bone formation within each individual BMU and not due to an increase in the resorption of bone [51]. Estradiol acts to conserve bone mass and maintains balanced rates of bone formation and bone resorption, it decreases osteoclast lifespan, and may extend osteoblast lifespan [40, 52, 53]. Estrogen seems to be the dominant sex steroid regulating bone resorption, whereas both estrogen and testosterone are important in maintaining bone formation [40, 54, 55].

In premenopausal women, more than 95% of serum estradiol (the most potent estrogen), and most of the serum estrone is derived from ovarian secretion [40]. During the 2-4 years menopausal transition estradiol levels fall to 10-15% of the premenopausal levels. In postmenopausal women almost all of the circulating estrogens are derived from peripheral conversion of steroid precursors [40, 56]. In premenopausal and postmenopausal women, testosterone is derived from ovarian (25%) and adrenal secretion (25%), and from peripheral conversion (50%) [40]. The levels of testosterone decrease only moderate after menopause because it continues to be produced by the ovarian interstitium and by adrenal cortex.

At menopause, the accelerated bone loss clearly results from loss of ovarian function, because estrogen withdrawal increases the rate of remodelling, and this can be prevented by estrogen replacement [40]. First, it increases the activation frequency of BMU, so there are many more remodelling units activated on the endosteal surfaces. Second, it induces a remodelling imbalance by prolonging the resorption phase (osteoclast apoptosis is reduced) [52] and shortening the formation phase (osteoblast apoptosis is increased) [53]. The volume of the resorption cavity is increased beyond the capacity of the osteblasts to refill it, and in trabecular bone this is leading to perforation and loss of trabecular connectivity [19, 40, 57, 58]. The initial accelerated phase of bone loss is a so-called "remodelling transient", indicating the rapid fall in bone mass produced by the increase in number of BMUs, as remodelling moves from a lower to a higher rate [59]. This bone loss is a result of the normal delay in initiation of bone formation and its slower completion within the higher numbers of resorption cavities [19]. As assessed by biochemical markers, bone resorption increases by 90% at the menopause, whereas bone formation markers increase by only 45% [40, 60]. This rapid phase of postmenopausal bone loss accounts for losses of 20-30% trabecular and 5-10% of cortical bone and it subsides after 4-8 years [40]. The rate of decline in BMD slows as bone formation goes to completion in the increased numbers of BMU and steady state is restored at

the higher postmenopausal remodelling rate. After menopause the balance is more negative because estrogen deficiency increases the lifespan of osteoclasts, so more bone is resorbed, and decreases the lifespan of osteoblasts, so less bone is formed [52, 53].

In men, the peripheral conversion of steroid precursors accounts for almost all of the circulating estradiol, while more than 95% of testosterone is derived from the testis [40, 56, 61]. Except for a few older men who develop overt hypogonadism, the levels of testosterone decrease only slightly, whereas estradiol are reported to be unchanged or decreased in men with advancing age [62]. The decrease in free or bioavailable levels of testosterone and estradiol is caused by an increase in sex hormone-binding globulin (SHBG) in men [63-65].

In addition, the adrenal cortex and to a lesser extent the gonads secret large amounts of dehydroepiandrosterone (DHEA), DHEA sulphate (DHEAS) and androstenedione. The circulating DHEAS levels in men and women are 100- to 500-fold higher than testosterone and 1,000- to 10,000-fold higher than estradiol [40]. Although weakly androgenic themselves, they are an important source of substrate for the extragonadal synthesis of potent sex steroids [66, 67]. The extragonadal sites are incapable of converting cholesterol to steroid hormones, so they are dependent on circulating androgen precursors as DHEAS for production of testosterone and estradiol [66, 68]. In particular this conversion takes place in the adipose tissue, but multiple tissues including bone (osteoblasts and chondrocytes) can synthesize estradiol from testosterone, and one of the key enzymes involved is aromatase [66, 68, 69].

Although osteoporosis is often considered to be mainly a disease of women, men also lose bone with aging [1, 40]. Castrated men (sex offenders in Czechoslovakia) had a pattern of rapid bone loss similar to that of women after menopause [70]. Men do not undergo the equivalent of menopause and they exhibit a slow phase of bone loss that is similar to the slow phase in women [40]. The loss of trabecular bone in men proceeds with thinning of trabeculae rather than complete loss, as in women [71]. Bone loss is the result of a reduction in the

volume of bone formed rather than the result of an increase in the volume of bone removed in the BMUs, so trabecular connectivity is better maintained in men than in women [19]. Late in life, also intracortical remodelling increase at the surfaces within cortical bone and result in intracortical porosity, which can predispose to fractures at cortical sites such as the proximal femur [72]. Periosteal apposition, which increase the width of long bones, continues throughout life in both sexes, but men add 3-fold more bone by this process than women [35]. So more women sustain fractures than men because they start with a smaller skeleton at peak, trabecular bone loss proceeds by more architectural disruption and bone loss is offset less well by aging by periosteal apposition. Consequently, a higher proportion of elderly women than elderly men have bone size and volumetric BMD reduced to below a critical level at which the loads on the bone are greater than the bone's structural ability to tolerate them [19].

Observational studies have reported estradiol rather than testosterone as the main predictor of BMD in both sexes [63-65, 73-76], but these observations have not been consistently replicated in either sex [9, 77, 77-80]. However, because the prevailing BMD of elderly women and men is the summation of the amount of bone gained during growth and maturation and the amount lost with aging, these correlations could reflect either or both processes.

A few small longitudinal studies have shown association between estradiol and bone loss in men [81-83]. In three nested case-control studies from the Study of Osteoporotic Fractures, elderly women with lower serum levels of estradiol and higher levels of SHBG had lower cross-sectional BMD-values at the calcaneus, proximal radius, proximal femur, and lumbar spine, higher rates of bone loss from the calcaneus and proximal femur and increased risk for vertebral and hip fractures [9, 10, 84]. Although the correlations between serum estradiol levels and bone loss in late postmenopausal women were significant, they may underestimate the restraining effect on bone loss of extragonadal estrogen synthesis, which is virtually the

exclusive source of circulating estrogen levels in women after menopause [40]. This local synthesis could play a major role in sex steroid action. Moreover, because the extragonadal tissues are unable to synthesize adrenal precursors, they are dependent on the circulating levels [40]. It is likely that the process is substrate limited and the large age-related decreases in levels of circulating adrenal precursors reduce extragonadal estrogen synthesis, and thus further enhance bone loss. This raises the possibility that part of the bone loss in premenopausal women, may relate to the early decrease in levels of DHEAS [40]. A weak association between the levels of dehydroepiandrosterone sulphate (DHEAS) and BMD is reported in women, but not in men [63, 65].

The most obvious property of SHBG is its function as transport protein, and thus regulator of bioavailable sex hormones and their access to target cells [85]. Due to the high affinity for sex steroids, changes in SHBG concentration result in large changes in both free and albumin-bound steroids. Circulating sex steroids bound to SHBG have restricted access to target tissues, whereas the 1-3% fraction that is free and the 35-55% fraction that is loosely bound to albumin are readily accessible [40]. Plasma SHBG levels increase by age, thyroid hormones and estradiol treatment, and decrease by increase in body mass index (BMI), insulin and androgen treatment [78, 79, 83]. The age-related decrease in GH and IGF-I have been suggested to contribute to the increase in SHBG [86, 87], and also to bone loss [40]. SHBG is associated with BMD [73, 76, 76], bone loss [9, 88] and risk of fracture [10, 79, 89, 90]. The positive association between SHBG and markers of bone turnover add strength to the hypothesis that SHBG may participate in the pathogenesis of bone loss [78, 79, 88].

2.4 Sex steroids effects on risk of fractures

Circulating estradiol is reported both associated and not associated with occurred vertebral fractures in women [84, 91], and in men [79, 89, 91, 92]. These results from retrospective data

are difficult to interpret, as alteration in estradiol could have occurred after the fractures. Prospective studies including incident vertebral fractures, or combination of vertebral and non-vertebral fractures as end point have reported lower estradiol associated with fracture risks in women [10, 12, 90, 93]. Prospective studies including incident hip fractures have reported low estradiol associated with risk of fractures in women [10, 11] and in men [94], but the effect was dependent on weight in one study [11].

In women, higher SHBG is reported associated with increased risk of incident vertebral, hip and the combination of vertebral and non-vertebral fractures in prospective studies [10-12, 90]. In men, SHBG is associated with prevalent vertebral fractures and the combination of vertebral and non-vertebral fractures as endpoints [79, 89]. In men, the effect of SHBG on incident non-vertebral fractures has not been studied in prospective data. Most authors suggest that higher SHBG increase the risk of fractures by binding estradiol and decreasing its bioavailability [10, 12, 79].

2.5 Other causes of bone loss and fractures

The pathogenesis of bone fragility is multifactorial and no single factor as low BMD or fall alone can explain the occurrence of fractures. In addition to genetic factors, age, sex and geographic location, also prior fracture, BMI, smoking, family history, physical inactivity, use of corticosteroids and certain diseased increase the risk of fracture [18-20, 95].

Increased fracture risk caused by vitamin D and calcium deficiency is documented among the institutionalized elderly [96, 97]. Vitamin D is produced in the skin under exposure to UV radiation from sunlight, and hydroxylated in the liver and the kidneys to the active vitamin D metabolites – 25OHD and $1,25(OH)_2D$ [98], which increase intestinal calcium resorption and regulate the serum levels of calcium to promote mineralization of the skeleton [40, 99]. Nutritional intake or vitamin D supplementation is relevant when the sunlight exposure is

insufficient. Vitamin D deficiency is associated with secondary hyperparathyroidism, increased bone remodelling, bone loss and impaired bone mineralization [100].

2.6 Summary

More women than men sustain fractures because they start with a smaller skeleton at peak, trabecular bone loss proceeds by more architectural disruption and bone loss is offset less by periosteal apposition. Sex steroids have important effects on the skeleton during growth and aging. Lower levels of circulating sex steroids and higher SHBG are associated with bone loss and increased fracture risk, but this is not consistently reported. However, few prospective data are available examining the contribution of sex steroids and SHBG on fracture risk. So whether women or men with low levels of circulating sex steroids have higher risk of bone loss or fractures remains uncertain.

3. Aims of the thesis

First we wanted to make a contribution to the knowledge on whether sex steroids vary by age and by season in a large, general population living in an Artic region. Second, we wanted to increase the understanding of the pathophysiology of bone fragility, and study whether circulating sex steroids were major determinants of bone loss or fractures, and if so, whether these measurements could assist in decision making regarding fracture risk susceptibility.

The aims of the thesis were to assess in women and men aged 25-84 years:

1. The distribution of estradiol, FSH and DHEAS by age, sex and menopausal status.
2. The seasonal variation of estradiol, FSH and DHEAS.
3. Whether circulating sex steroids or SHBG predicts bone loss.
4. Whether circulating sex steroids or SHBG predicts non-vertebral fractures.

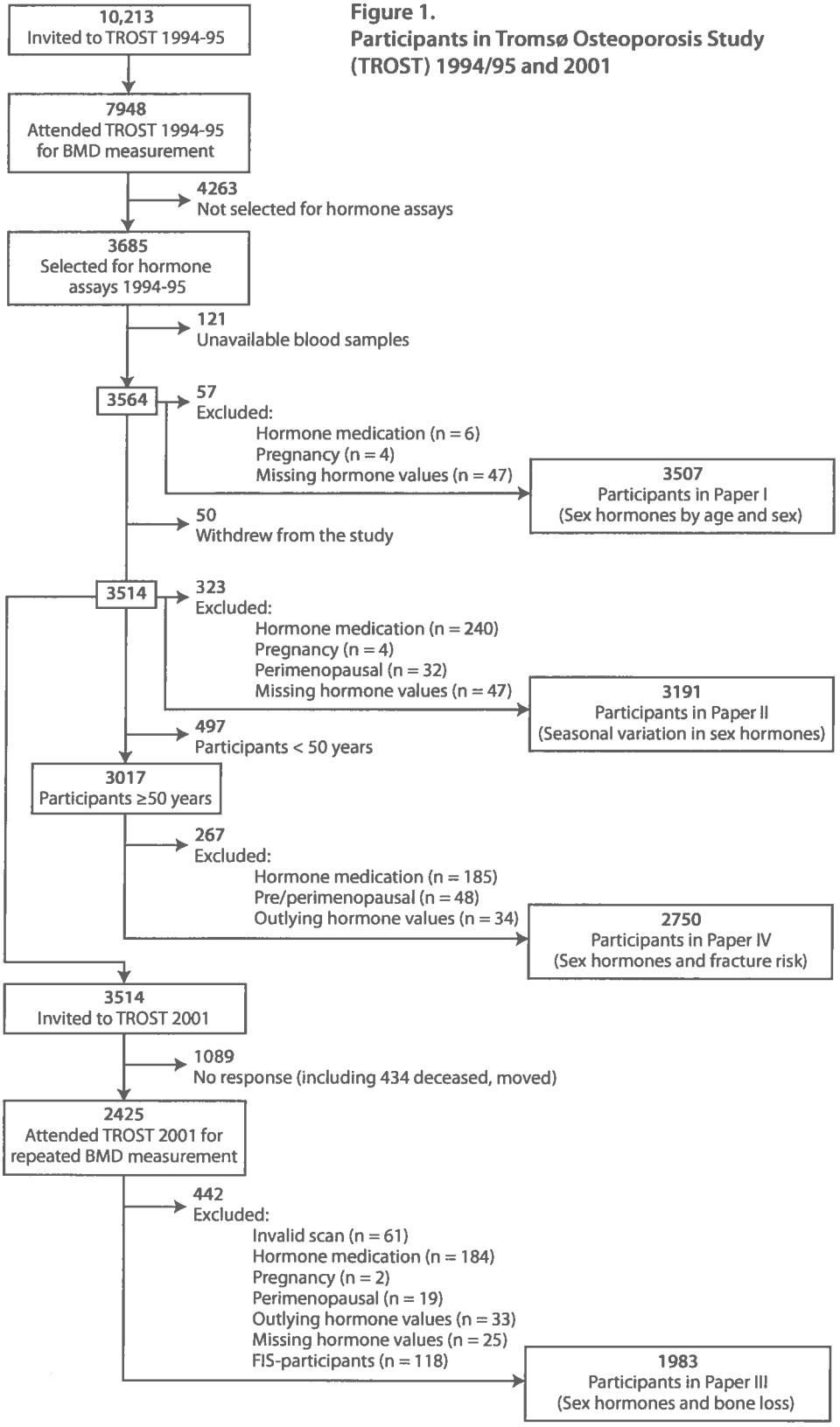


Figure 1.
Participants in Tromsø Osteoporosis Study (TROST) 1994/95 and 2001

4. Study population and methods

4.1 The Tromsø Osteoporosis Study – TROST

The Tromsø Study was initiated in 1974 with surveys repeated in 1979-80, 1986-87, 1994-95 and 2001, a single-centre, population-based prospective study of Tromsø in northern Norway, conducted by the University of Tromsø and the National Health Screening Service.

At the 1994-95 survey, all inhabitants older than 24 years were invited, and 27,159 subjects (77%) participated in the main survey (phase I). From the main survey all men aged 55-74 years, all women aged 50-74 years and 5-10% random samples of the other age groups of both sexes were invited to an extended examination, including forearm bone densitometry and blood sampling (phase II). In addition, 328 male participants of the Family Intervention Study (FIS), selected on the basis of high total cholesterol or low HDL to total cholesterol ratio, were also invited [101]. As part of this fourth Tromsø Study, TROST invited 10,213 subjects and measured bone mineral density in 7,948 subjects (response rate 78%) (Table 1), and sex hormones in a random subgroup of 3564 between September 1994 and September 1995 (Figure 1) [102, 103]. After paper I was written, 50 subjects withdrew from the study. In paper II, we thus included 3514 subjects, and in paper IV we included those 3017 subjects who were 50 years of age or older. Of the 3514 subjects, 434 deceased or moved, and the 3080 subjects still living in Tromsø, were invited to a repeated BMD measurement in 2001 and Paper III included the 2,425 (79%) who attended (Table 1, Figure 1).

Table 1. Attendence rates by age in the Tromsø Osteoporosis Study 1994-95 and 2001.

Age (years)	Invited 1994-95	Attended 1994-95	Response rates (%)	Invited 2001	Attended 2001	Response rates (%)
25-39	838	491	58.6	210	143	68.1
40-49	616	476	77.3	234	181	77.4
50-59	3739	2964	79.3	950	806	84.8
60-69	3401	2824	83.0	1185	958	80.8
70-84	1619	1193	73.7	501	337	67.3
All	10213	7948	77.8	3080	2425	78.7

4.2 Methods

4.2.1 Hormone assays

Nonfasting blood samples were taken between 0800 h and 1600 h at baseline in 1994-95, and serum was stored at -70° C for 6-7 years, until first thawed in 2001. All hormones and SHBG were measured on Immulite 2000 (Diagnostic Products Corporation, Los Angeles, CA, USA). Estradiol, testosterone and DHEAS measurements were based on competitive immunoassays, whereas follicle stimulating hormone (FSH) and SHBG measurements were based on immunometric assays. The intra- and inter-assay coefficient of variation (CV) for estradiol, DHEAS, FSH and SHBG were between 3.0-15%. The intra- and inter-assay CV for testosterone > 1nmol/l were 3.5% and 5%; while in the range 0.1 to 1.0 nmol/l, it was 12% and 20%, respectively. The lower limits of detection were 10 pmol/l for estradiol, 0.1 nmol/l for testosterone, 1.0 μmol/l for DHEAS, 0.5 IU/l for FSH, and 1.0 nmol/l for SHBG. Samples with values below limits of detection were given a value midway between zero and limit of detection. We used the method described by Vermeulen et al. to calculate free estradiol and free testosterone from total estradiol, total testosterone and SHBG levels [104]. A recent evaluation by Rinaldi et al. [105] found that method to be simple and reliable.

4.2.2 Forearm bone mineral density

Bone density was measured on the non-dominant forearm, at distal and ultradistal sites, with Single X-ray Absorptiometric (SXA)-devices (DTX-100 Osteometer Medi Tech, Inc., Hawthorne, California). The same study protocol was used in both baseline and follow-up studies [106]. The CV was 0.8 and 1.0% at the distal and ultradistal sites, and details of the measurement methods and the strict quality control procedures for densitometry are previously published [50, 107-110]. Briefly all scans were reviewed and reanalysed and only scans free of serious artefacts were included. The long-term performance of the densitometers

was assessed by twice daily phantom measurements with an aluminium forearm phantom, as well as by weekly measurements with the anthropomorphic European forearm phantom in the second survey.

The distal site, which contains 10-20% trabecular bone [111], includes both the radius and ulnae from the 8mm-point (point where the ulnae and radius are separated by 8 mm) and 24 mm proximally. The ultradistal site contains 50-70% trabecular bone, includes only the radius, and stretches from the 8-mm point up to the radial endplate. Only measurements from the distal site are presented, as the ultradistal measurements followed the same pattern. The mean follow-up time was 6.5 years (SD 0.4, range 5.4 to 7.4). Mean annual BMD change was calculated as the difference between the two measurements, divided by each participant's follow-up time. The annual change in BMD in %, was estimated as the difference divided by baseline BMD, and multiplied by 100.

4.2.3 Fracture registration

All non-vertebral fractures were registered from the x-ray archives of the University Hospital in Tromsø between 1 January 1994 and 12 February 2005. All fractures are registered here, as this is the only x-ray service in the city or within 250 km. The only exception would be fractures occurring while travelling with no control x-ray after returning home or fractures not referred to x-ray examination. The validation of the fracture registration is previously reported [112]. Follow-up time was assigned from baseline to the first fracture, to death (n = 506), when the participant moved (n = 177), or to the end of follow-up. The mean follow-up time was 8.4 years (range 0.01-10.4), and observed person-years were 23,034. The numbers and incidence of all first non-vertebral, hip and forearm fractures are given in Table 2 and 3.

Table 2
Numbers of non-vertebral fractures by age and sex from 1994-95 to 2005.

	Women				Men			
Age(years)	n	All	Hip	Forearm	n	All	Hip	Forearm
50-59	380	61	4	29	471	33	5	7
60-69	680	133	18	63	605	40	8	9
70-79	321	87	20	36	284	32	14	5
80-84	5	0	0	0	4	0	0	0
All	1386	281	42	128	1364	105	27	21

Table 3
Incidence of non-vertebral fractures per 10 000 person-years from 1994-95 to 2005.

	Women			Men		
Age(years)	All	Hip	Forearm	All	Hip	Forearm
50-59	183.3	12.0	87.2	78.6	11.9	16.7
60-69	235.6	31.9	111.6	77.8	15.6	17.5
70-79	354.2	81.4	146.6	143.9	62.9	22.5
80-84						
All	245.3	36.7	111.7	90.7	23.3	18.1

4.2.4 Other variables

Self-administered questionnaires were used as instruments to gain information on a broad set of variables (Appendix A-C). We included information on previous and present diseases such as cancer, asthma, diabetes, stroke and ischemic heart disease (IHD), use of any medication, current smoking status, and consumption of cigarettes, alcoholic beverages, and coffee. A physical activity score was made by adding the hours/week of moderate (m) and hard (h) physical activity, giving the hours with hard activity double weight: score = m + 2h.

As the menopausal stage could not be determined accurately in users of hormonal therapy, only nonusers were classified into pre-, peri- or postmenopausal groups. The geometric means of the sex hormones was similar whether definition of menopausal stage was based on only self-reported menopause, or based on self-reported menopause and age (Table 4). We present results with the highest possible number of observation, and based our definition of menopausal stage on self-reported menopause and age.

Table 4. Geometric means and 95% CI for the means of the sex hormones in women by definition of menopausal stage. The Tromsø Study 1994-95.

	Self-reported menopause and age		Self-reported menopause	
	Mean	95% CI	Mean	95% CI
Premenopausal women	n = 205		n = 182	
Total estradiol (pmol/liter)	179	156-206	175	151-204
Free estradiol (pmol/liter)	3.7	3.2-4.2	3.6	3.1-4.2
Total testosterone (nmol/liter)	0.5	0.4-0.5	0.5	0.4-0.6
Free testosterone (pmol/liter)	5.1	4.2-6.1	5.2	4.3-6.3
FSH (IU/liter)	6.4	5.7-7.2	6.6	5.8-7.5
DHEAS (µmol/liter)	3.3	3.1-3.6	3.3	3.1-3.6
SHBG (nmol/liter)	65.4	61.5-69.5	65.5	61.4-69.9
Postmenopausal women	n = 1334		n = 901	
Total estradiol (pmol/liter)	21	20-22	21	20-23
Free estradiol (pmol/liter)	0.4	0.4-0.5	0.4	0.4-0.5
Total testosterone (nmol/liter)	0.2	0.2-0.2	0.2	0.2-0.2
Free testosterone (pmol/liter)	2.4	2.2-2.6	2.3	2.1-2.5
FSH (IU/liter)	66.1	64.7-67.6	66.0	64.2-67.9
DHEAS (µmol/liter)	1.3	1.3-1.4	1.4	1.3-1.5
SHBG (nmol/liter)	67.9	66.2-69.6	66.6	64.6-68.7

Women who had their last menstrual period within the last three months, or within the last 3-12 months and were < 45 years of age, or had missing menstruation data and were < 45 years of age, were defined as premenopausal. Women who had stopped menstruating within the last 3-12 months and were ≥ 45 years, or with unreported last menstrual period and between 45 and 54 years, were considered perimenopausal. Women who reported that they had stopped menstruating over a year ago, or were ≥ 54 years of age were defined as postmenopausal. The women defined as postmenopausal based on age, had a mean age of 70 years (range 54-83), and 92% of them were ≥ 60 years of age. This definition of menopausal status left none of the women undefined due to missing or conflicting values. The numbers of women in each menopausal stage group are given in the respective papers. Participants were not asked about previous hysterectomy or oophorectomy. At the time of blood sampling,

menstruating women were not asked to report their date of the last menstrual period, therefore the information on menstrual phase at blood sampling is lacking.

4.2.5 Data management and statistics

The SAS Software package was used for data management and analyses. The significance level was chosen at $P < 0.05$ and P-values are two sided. In Paper I and II the sex hormones were dependent variables, and we used log-transformed hormones in all analyses due to skewed distribution. However the presented means and confidence limits were transformed back to the original units. Differences in means between groups were tested by ANOVA, and adjusted t-tests for multiple comparisons of pairs of means. In Paper I we used multiple linear regressions to model each of the hormones. The same set of covariates; age, BMI, smoking, alcohol and coffee consumption and physical activity, which were significantly associated with at least one hormone, was used systematically in all models in both sexes in paper I and II. As levels of estradiol were significantly different in users of HRT (higher levels) and contraceptive pills (lower levels) compared with non-users (Paper I), they are excluded from further analyses. Because there are major differences in levels of sex hormones between premenopausal and postmenopausal women, and between women and men, all data were analyzed separately for premenopausal and postmenopausal women and for men. The mean levels of sex hormones in perimenopasual women differed from premenopausal and postmenopausal levels (Paper I), but this group was too small for separate analyses, so we excluded them from further analyses.

In Paper II we examined the monthly means of sex hormones over the year by ANCOVA. However, these analyses tell us nothing about any cyclic seasonal pattern of variation in the hormone levels. Testing for differences in monthly means would give the same result

regardless of the order of the months that differ. It is therefore a pleasure to present results from the cosinor analyses, which directly test for pattern of variation during the year.

In Paper III data were analyses age-stratified in men, due to a significant interaction between age and SHBG. Multiple linear regression analyses were used to investigate the effect of baseline sex steroids and SHBG on BMD in cross-sectional analyses and on change in BMD in follow-up data. Baseline BMD and change in BMD were used as dependent variables, with the sex steroids and SHBG as independent variables. We included only one hormone or SHBG in each of the presented models. Log-transformation corrected for skewed distribution of the sex steroids, but results were similar in both Paper III and IV. As normality is not an assumption for independent variables, we used values of the sex steroids without transformation. We controlled for age, BMI and current smoking because these variables are known to be associated with BMD and some confounding were confirmed. To facilitate the comparison of effects of sex steroids on bone loss we calculated standardized regression coefficients that describe the change in BMD in SD, per SD change in sex steroids or SHBG.

In Paper IV we used Cox proportional hazard models to determine whether sex steroids predict fractures, and presented hazard ratios (HR) by sex specific SD change in sex steroids. The models assumption of a constant HR over time was assessed in log-log survival curves for all sex steroids and SHBG. Cox models are presented adjusted for age, BMI, height, current smoking, physical activity and BMD, because these factors are known to be associated with risk of fractures and with sex steroids, and each of them changed one or more point estimates. We rerun analyses after exclusion of fractures in fingers, toes and face and got similar results, and we also rerun analyses after exclusion of fractures with reported high-energy trauma involved, with similar results. So we have chosen to present results with all fractures included.

5.0 Main results

5.1 The distribution of estradiol, FSH and DHEAS by age, BMI and sex (Paper I)

This was a cross-sectional study of the distribution of total and free estradiol, FSH and DHEAS by age, and the relation between hormones and BMI, lifestyle factors, and chronic diseases, in 1952 women and 1555 men aged 25-84 years. By higher age, the levels of total and free estradiol were lower in postmenopausal women, and were 33 and 12% higher in men ≥ 70 years compared with men < 40 years. FSH was higher and DHEAS lower in both sexes.

With increasing BMI, total and free estradiol were higher in postmenopausal women, and free estradiol was higher in men, whereas FSH had lower levels in both groups. Smoking men had higher levels of DHEAS than non-smokers. However, men with chronic diseases had lower levels of DHEAS compared with healthy men. This was not the case in women.

5.2 The seasonal variation of estradiol, FSH and DHEAS (Paper II)

Tromsø, Norway is located at 70 degrees north and has extreme variations in the daylight exposure. The sun is below the horizon from November 28 to January 15 and does not set between May 17 and July 26. Cross-sectional data throughout a year provided a unique opportunity to test whether extreme seasonal variations in the daylight effects the levels of sex hormones in 1651 women and 1540 men aged 25-84 years and living at high latitude.

Total and free estradiol showed differences between monthly means, with peak in June in postmenopausal women, and in May in men, by analysis of covariance. By cosinor analysis, a seasonal variation in total and free estradiol was evident in women and in men, but only 0.2-0.9% of the variation in total and free estradiol was explained by season. Seasonal variations should be considered while designing studies and interpreting results of estradiol measurements to avoid bias in comparative studies.

5.3 Whether circulating sex steroids or SHBG predicts bone loss (Paper III)

We studied 1089 women and 894 men aged 25-84 years during 6.5 years follow-up to test the hypothesis that bone loss in both sexes is associated with circulating sex steroids or SHBG. Bone loss was detected in pre- and postmenopausal women and in men. Age-adjusted free estradiol and SHBG predicted bone loss in postmenopausal women and in older men (above 60 years of age). After further adjustment for BMI and smoking, only free estradiol persisted as significant independent predictors of bone loss in postmenopausal women and only SHBG in men. However, only 1-2% of the variance in bone loss was accounted for by these measurements. Therefore, measurements of sex steroids are unlikely to assist in decision making regarding fracture risk susceptibility.

5.4 Whether sex steroids or SHBG predicts non-vertebral fractures (Paper IV)

We tested the hypothesis that incident non-vertebral fractures are predicted by circulating levels of sex steroids or SHBG in a prospective study of 1386 women and 1364 men aged 50 to 84 years. During 8.4 years (range 0.01-10.4) and 23,034 person-years follow-up, 281 (20.3%) women and 105 (7.7%) men suffered non-vertebral fractures. For both sexes, fracture cases had lower BMD, higher SHBG, but sex steroids were no lower than in participants remaining fracture free. Each SD increase in SHBG increased non-vertebral fracture risk by about 20% in women (RR 1.17; 95% CI 1.03-1.33) and men (RR 1.27; 95% CI 1.03-1.55). The effect was mediated by BMD in both sexes. Fracture risk was highest in participants with the combination of SHBG in the highest tertile and BMD in the lowest tertile in women (RR 3.76; 95% CI 2.02-7.01) and in men (RR 2.98; 95% CI 1.41-6.30). However, the fracture risk prediction of the combination was only slightly better than BMD alone. No association between sex steroids and fracture risk was detected. Measurements of sex steroids or SHBG are therefore unlikely to assist in decision making regarding fracture susceptibility.

6. General discussion

6.1 Methodological considerations

6.1.1 Internal validity

The internal validity of a study refers to whether the results are representative, true or valid for the source population [113]. Selection bias, information bias and confounding may threaten the internal validity of an epidemiological study.

Selection bias

Selection bias is a systematic error in a study that stems from the procedures used to select subjects and from factors that influence study participation [114]. The Tromsø Study is a population based study well known for the high response rate, and the attendance rates were well above 75% in TROST 1994-95 and in 2001 [50, 107, 109]. The high response rate assures generalizability of results to a majority of the source population. However, the response rates in subjects below 40 years of age were 58.6% in TROST 1994-95 and 68.1% in TROST 2001 (Table 1). This lower attendance rates may treat the results in premenopausal women and in the young men.

In TROST 1994-95 the responders were compared with non-responders, and they had no higher prevalence of self-reported chronic diseases [107]. Non-responders could still be less healthy than responders. Bone loss is a powerful predictor of the general health status [115], and a tendency towards "healthy" selection bias is observed in other studies of longitudinal bone loss [116, 117]. Despite high attendance rates, there might be a "healthy" selection bias in our study, and rates of bone loss could be slightly underestimated [109]. We looked for selection bias by comparing baseline characteristics of the random subgroup with hormone measurement (n = 3507) with the total TROST population in 1994-95 (n = 7948) (Table 5). We also compared participants in the 2001 survey with both groups in the 1994-95 survey.

Table 5. Baseline characteristics in all Tromsø Osteoporosis Study (TROST) subjects in 1994-95, the subgroup with sex hormones measured in 1994-95, and those who in addition had BMD measurements repeated in 2001. The Tromsø Study 1994-95 and 2001.

	All participants with BMD in 1994-95[1] n = 7948		Participants with sex hormones in 1994-95[2] n = 3507		Participants with BMD repeated in 2001[3] n = 2425		P-values* for differences between groups		
Women	Mean	SD	Mean	SD	Mean	SD	[1] and [2]	[1] and [3]	[2] and [3]
Age (years)	58.2	10.5	59.7	11.0	59.2	10.3	< 0.001	0.01	0.006
Height (cm)	162.0	6.4	161.8	6.4	162.0	6.3	0.23	0.83	0.18
Weight (kg)	67.8	11.9	68.0	12.0	68.0	11.4	0.56	0.50	0.90
Body mass index (kg/m²)	25.9	4.4	26.0	4.5	25.9	4.2	0.21	0.51	0.56
Physical activity score	3.0	2.2	2.9	2.2	3.0	2.2	0.23	0.42	0.01
Forearm BMD[a] (g/cm²)	0.417	0.07	0.408	0.07	0.412	0.07	< 0.001	0.009	0.03
Current smoking (%)	31.9		31.0		28.3		0.39	0.004	0.03
Teetotallers (%)	24.5		27.3		25.8		0.004	0.26	0.21
Ischemic heart disease (IHD) (%)	7.1		8.6		6.4		0.01	0.31	0.004
Men									
Age (years)	59.8	10.0	60.0	10.1	59.6	9.1	0.91	0.11	0.01
Height (cm)	175.1	6.8	175.2	6.9	175.5	6.6	0.88	0.09	0.14
Weight (kg)	80.1	12.1	80.3	12.1	80.7	11.6	0.55	0.09	0.23
Body mass index (kg/m²)	26.1	3.4	26.1	3.4	26.2	3.2	0.52	0.33	0.44
Physical activity score	3.7	2.5	3.7	2.5	3.8	2.5	0.45	0.06	0.06
Forearm BMD[a] (g/cm²)	0.545	0.07	0.546	0.07	0.550	0.07	0.72	0.01	0.03
Current smoking (%)	34.6		35.5		32.9		0.46	0.21	0.07
Teetotallers (%)	13.1		12.7		11.1		0.64	0.05	0.12
Ischemic heart disease (IHD) (%)	15.7		15.6		12.3		0.92	0.002	0.003

* P-values for differences between groups within same sex by t-test and χ^2.

[a]BMD, distal forearm bone mineral density

The subgroup of women with hormone measurements was one year older, had lower BMD and higher prevalence of IHD whereas the men did not differ in any characteristics compared with the total TROST population. The women and men, who responded in 2001, were at baseline on average 4-5 months younger than the subgroup with hormone measurements. The 2001 responders had higher forearm BMD, were more physical active, were to a lesser extent current smokers and had lower prevalence of IHD. These small but significant differences, can partly explain why the non-responders to a lesser extent attended. Among the hormones we studied in Paper I, only DHEAS was associated with IHD and only in men. Among the hormones studied by Svartberg et al. [103] only free testosterone was associated with IHD after adjustment for BMI in men. As the levels of DHEAS and free testosterone were not associated with bone loss or risk of fracture (Paper III and IV), we assume the potential for "healthy" selection bias to be small.

All male participants of the Family Intervention Study (FIS), a randomized trial aimed at improvement of the cardiovascular risk profile in men aged 40-54 years who either had a high total cholesterol or a low HDL to total cholesterol ratio, were invited to TROST 1994-95 [101]. As associations between increased cardiovascular risk and BMD may exist, surplus FIS participants (n = 328) were excluded from the cross-sectional analyses of BMD as they were not viewed as representative of the general population [107]. For the same reason we excluded the 118 surplus FIS men from our analyses on longitudinal bone loss (Paper III). FIS men below 50 years of age had lower levels of free testosterone and DHEAS than other men of the same age (Table 6). However, bone loss (analysed stratified and adjusted for age) and fracture frequency did not differ significantly between FIS men and other men (Table 7 and 8). Excluding surplus FIS men did not change any of our results substantially, so we kept them included in the analyses presented in paper I, II and IV.

Table 6. Geometric means and 95% CI for the means of the sex hormones in Family Intervention Study (FIS) men compared with other men. The Tromsø Study 1994-95.

	FIS men		Other men		P-values*
	Mean	95% CI	Mean	95% CI	
Men 40-49 years	n =77		n = 49		
Total estradiol (pmol/liter)	40.7	33.9-48.8	43.1	34.3-54.2	0.70
Free estradiol (pmol/liter)	1.0	0.9-1.2	1.1	0.9-1.4	0.61
Total testosterone (nmol/liter)	12.4	11.5-13.5	14.0	12.6-15.5	0.08
Free testosterone (pmol/liter)	0.2	0.2-0.2	0.3	0.2-0.3	0.002
FSH (IU/liter)	5.6	4.9-6.3	5.1	4.3-6.0	0.43
DHEAS (µmol/liter)	3.9	3.5-4.4	5.1	4.4-5.9	0.008
SHBG (nmol/liter)	38.1	34.7-41.9	35.1	31.2-39.5	0.29
Men 50-54 years	n = 80		n = 35		
Total estradiol (pmol/liter)	47.3	40.8-54.9	39.2	31.3-49.1	0.18
Free estradiol (pmol/liter)	1.2	1.0-1.4	1.0	0.8-1.2	0.16
Total testosterone (nmol/liter)	12.1	10.8-13.6	13.0	10.9-15.5	0.53
Free testosterone (pmol/liter)	0.2	0.2-0.2	0.2	0.2-0.3	0.62
FSH (IU/liter)	7.0	6.1-8.1	6.5	5.3-8.1	0.58
DHEAS (µmol/liter)	3.6	3.2-4.1	4.3	3.6-5.2	0.11
SHBG (nmol/liter)	41.4	38.2-44.9	42.8	37.9-48.3	0.66

* P-values for differences between men within same age group tested by ANOVA.

Table 7. Means and 95% CI for the means of the bone loss in the Family Intervention Study (FIS) men compared with other men. The Tromsø Study 1994-95 and 2001.

	FIS men			Other men			P-values*
Age (years)	n	Mean	95% CI	n	Mean	95% CI	
40-49	54	-1.45	-1.94, -0.96	34	-0.85	-1.46, -0.23	0.13
50-54	64	-2.05	-2.57, -1.53	29	-1.46	-2.23, -0.69	0.22
40-54	118	-1.78	-2.14, -1.42	63	-1.13	-1.62, -0.63	0.04
40-54	118	-1.74	-2.10, -1.39	63	-1.19	-1.68, -0.70	0.08†

* Differences between men within same age group tested by ANOVA, †Age-adjusted.

Table 8. Numbers and incidence of fractures in the Family Intervention Study (FIS) men compared with other men, between 1994 and 2005. The Tromsø Study 1994-95.

	FIS men (n = 80)			Other men (n = 35)			P-values*
Age (years)	n	%	Incidence	n	%	Incidence	
50-54	6	7.5	80.3	2	5.6	61.2	0.70

* P-values for differences between men within same age group tested by χ^2.

Information bias

Information bias may occur if there are systematic measurement errors or misclassification of exposure or outcome. Misclassifications can be of two types. Differential misclassification occurs when either the misclassification of the exposure differs by the outcome status or the misclassification of the outcome differs by the exposure status. If either the misclassification of the exposure or the outcome is independent of the status of the other, the misclassification is nondifferential. Differential misclassification can either exaggerate or underestimate an association, while nondifferential misclassification tends to dilute an association [114].

The registration of non-vertebral fractures from the x-ray archives of the University Hospital in Tromsø is previously validated, and 93% of hip fractures and 97% of wrist fractures were identified [112]. The sensitivity of fractures was much better from the x-ray archives than from self-report by questionnaire. The use of radiographic records gave no over-reporting, and the minor under-reporting is probably a non-differential misclassification expected to lead to an underestimation or no change of effects.

When TROST 2001 was started one of the two densitometers underwent a major repair, and later the x-ray tube had to be replaced in both densitometers [109]. Quality control routines, in which the European Forearm Phantom (EFP) and the aluminium forearm phantom (AFP) were used, revealed that before the X-ray tube was replaced, one of the machines measured at a higher BMD level compared with the other machine [109]. The EFP measurements reflected the differences seen in the human material [110], and therefore the EFP data were used to adjust for the differences in densitometer measurements by minus 0.005 g/cm^2. When the EFP and AFP measurements were compared directly with human measurements, EFP followed the human measurements, but tended to overestimate the real differences [118]. This indicate that the correction based on the EFP probably represent an

over-adjustment [119]. The bone loss rates in our study might be slightly underestimated, but with effects we believe are either statistically or clinically significant [119].

The SXA measurements of the distal forearm is thought to be one of the most precise densitometry methods [108, 120], confirmed by the low CV% on our densitometers. Despite the high precision in SXA measurements of BMD, the method has some limitations [34, 35, 40]. Structural changes in bone can not be assessed by either SXA or by DXA, which is the most commonly used method for BMD measurement. Both methods provide data on area BMD (g/cm^2) which overestimates volumetric BMD (g/cm^3) in larger bones by failing to account for the depth of the bone [19, 35]. In smaller bone the area BMD underestimate volumetric BMD [121]. Our measurements of area BMD are not true volumetric BMD and could be confounded by size. Paradoxically, this error can improve the value of BMD as a predictor of fracture risk, since bone size is also a determinant of skeletal strength [18]. Bone loss at the forearm may not reflect perfectly the pattern of bone loss at other sites, but still peripheral BMD measurements can be used to assess fracture risk at both peripheral and central sites [29]. However, in general all absorptiometric techniques have high specificity and low sensitivity to predict fractures [21].

The circulating levels of sex steroids were measured by newer methods capable of assessing the low levels of estradiol in postmenopausal women (Paper I). However, these methods are hampered with low precision in the very low levels of sex steroids and result in some uncertainty, confirmed by the high CV% on our immunoassays (paper I-IV). Although assays with low limits of detection were used, estradiol, testosterone and DHEAS values were below the limit of detection in 23, 41 and 26%, respectively in postmenopausal women. Post-menopausal women with undetectable estradiol had a marginally higher bone loss than women with detectable estradiol levels (Paper III). Otherwise participants with undetectable levels of sex steroids did not differ from other participants in any group, and excluding the

participants with values below the limit of detection did not change any result in this study. This measurement uncertainty could result in a nondifferential misclassification of the levels of sex steroids, which could weaken true associations. However, we believe that the large sample size in this study even out some of the uncertainty related to low precision, and reduce the threat on the validity of the results.

Serum samples were frozen at -70°C for 6.5 years, and the sex hormones were measured when the samples were thawed for the first time. Levels of sex hormones have been shown to be relatively stable in frozen serum stored for 3-10 years, so the delayed analyses are not likely to represent a major problem [122, 123]. Daytime variation in sex steroids is also unlikely to have influenced our results. Although testosterone decreased by increasing sampling hour in men above 60 years of age, testosterone was no significant predictor of bone loss or fracture risk in this group. Adjustment for sampling hour did not change any results.

In premenopausal women, we could not account for the menstrual cycle phase, which was not recorded at the time of blood sampling. This may have weakened true associations in this group. Our definition of postmenopausal status was based on self-reported menopause and age. The women defined as postmenopausal based on age, had a mean age of 70 years (range 54-83), and 92% of them were 60 years of age or older. The mean levels of hormones were similar in women regardless of the menopausal definition (Table 4). The seasonality of estradiol in postmenopausal women was blunted when we used the loose definition of menopause (Paper II). Otherwise the results did not differ substantially whether women with missing response to the menopause question were included or not, so we do not believe their inclusion bias the results. Lack of information on hysterectomy before natural menopause is a limitation [124]. Nevertheless, because the rate of hysterectomy in Norway is remarkably low, it is not expected to cause a serious misclassification [125].

Confounding and interaction

Confounding might be defined as confusion, or mixing of effects, which implies that the effect of the exposure is mixed with the effect of another variable, leading to a bias [114]. A confounder must be associated with both the outcome and with the exposure, and also be imbalanced between the exposure groups to be compared. Two methods can be used to minimize bias due to confounding, stratification or regression models with inclusion of potential confounders. Sex and age are well known possible confounders. So all results are presented stratified by sex and also by menopausal status, and stratified or adjusted for age. Other possible confounders that changed one or more point estimates are thoroughly described in the method section and also in the respective papers.

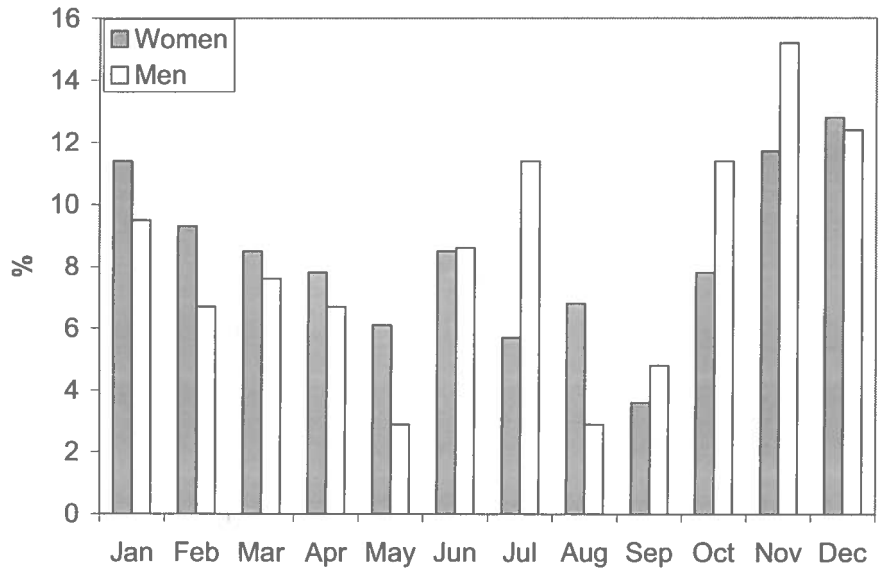
In the analyses of bone loss (Paper III), BMI was a confounder. Lower levels of free estradiol (Paper I) and higher SHBG [103] are associated with lower BMI, and lower BMI are associated with lower BMD [73, 126, 127]. Part of the effect of BMI on bone loss may be mediated through sex steroids, and part of it through other mechanisms.

Similarly in the analyses of risk of fractures (Paper IV), BMD was a confounder. Lower levels of estradiol and higher SHBG is associated with lower BMD (Paper III) and lower BMD is associated with increased risk of fractures [29]. The effect of estradiol and SHBG on risk of fractures have previously been reported to be both dependent [90, 93] and independent from BMD [10, 89]. The effect of SHBG on risk of fractures was dependent on BMD in the present study, and probably mediated through BMD. Still there might be other mechanisms whereby SHBG increase fracture risk that we do not know.

The weather condition with slippery surfaces due to ice and snow during the winter in Tromsø could be a potential confounder in the analyses of fracture risk. We estimated the monthly fracture frequency during the follow-up from 1994 to 2005, and tested for

differences in fracture frequency between summer and winter in dichotomized analyses by chi-square for equal proportions. The 6 months with mean temperature above 0°C from May to October 1994 were defined as summer months, and the 6 months with mean temperature of 0°C and below were defined as winter months from November to April (Paper II). Women had more fractures during the winter (Chi 15.0, $P < 0.001$, whereas men did not (Chi 2.8, $P = 0.10$) (Fig. 2). Unfortunately we were not able to account systematically for weather condition, because less than 30% of the radiographic reports of fractures contained information on snow and ice. Higher incidence of fractures during winter are in some reports attributed to increased numbers of falls occurring because of ice and snow [128-130]. However, residual effects of seasonal variations in fractures remain after adjustment for weather condition [131-133].

Fig. 2. The monthly non-vertebral fractures frequency (%) between 1994 and 2005.



Interaction or effect modification, a difference in effect of one factor according to the level of another factor, can have direct biological and public health relevance. In several instances we tested for interaction by inclusion of interaction terms (exposure variable multiplied by possible effect modifier). Because of a significant interaction between age and SHBG in the analyses of bone loss in men (P = 0.09), we stratified the analyses by age. Men aged 61-78 years lost more bone than men aged 25-60 years, and SHBG was associated with bone loss in older men (above 60 years), but not in younger men. In the analyses of fracture risk by combinations of tertiles of BMD and SHBG, the three-dimensional figures show that the effect of SHBG was dependent on the levels of BMD in both sexes. However, when testing the interaction term in the models, it was not significant in women (P = 0.78) and in men (P = 0.53). Interactions are not always easy to detect, and lack of significant interaction term does not necessarily exclude an interaction, because generally the power of tests for interactions may be low.

6.1.2 External validity

External validity or generalizability refers to whether the results are valid for other populations, which rely on whether the source population is representative of other populations.

The Tromsø population does not differ substantially from the Norwegian population at large with respect to age and sex distribution [134, 135]. The incidence of cardiovascular diseases, mortality, education and lifestyle is in accordance with data from other parts of Norway [136]. However, Tromsø is located at 70 degrees north and the sun is below the horizon from November 28 to January 15 and does not set between May 17 and July 26. The extreme variations in the daylight exposure at this high latitude, affects the amount and intensity of UV-exposure [137]. The inhabitants of Tromsø experience every winter

approximately six months with UV-radiation below the stated threshold need for Vitamin D production in the skin [137]. The Vitamin D metabolites regulate the serum levels of calcium and phosphorus to promote and maintain mineralization of the skeleton [40, 99]. During the Australian winter, cyclic variations in serum vitamin D lagged 1 month behind ultraviolet variation [138]. A fall in vitamin D was associated with increased parathyroid hormone (PTH), bone resorption and frequency of hip and wrist fractures that occurred 1-3 months after the trough in vitamin D levels.

Because of the location we could expect higher rates of bone loss in the Tromsø population not being representative of other populations. There are difficulties in comparing BMD change rates between populations, because of different measurement techniques used, and results presented in different age groups. However, the bone loss in Tromsø subjects aged 45-84 years was not higher compared with other cohorts [3, 50, 63, 109, 139-142]. That bone loss starts in mid-thirties in women and men [50] is in agreement with some [48, 49] but not other researchers who reported no bone loss in young adulthood [3, 81]. A recent study evaluated whether there was differences in forearm BMD between four geographic regions of Norway [143]. Mean distal forearm BMD was lower in the urban population of Tromsø (70°N), Oslo (60°N) and Bergen compared with the rural county of Nord Trøndelag, whereas there was no difference between rural part of Tromsø and Nord Trøndelag. There was no apparent north-south gradient in BMD. The hip fracture incidence in this cohort (2000) [144], in central Norway (1998) [145] and in Oslo, Norway (1997) [31] was higher than in Australia (1996) [146], Germany (1996) [147] and Japan (1994) [148]. We therefore believe that our results are applicable to other populations in Norway, but may not necessarily be representative for populations at lower latitudes.

6.2 Significance of results and strength of associations

6.2.1 Comparing our results with results from other populations

Although associations between BMD and circulating sex steroids are reported in both sexes [63-65, 73-76, 81, 82], these associations have not been confirmed in other studies [9, 77-80]. Most of the evidence are from cross-sectional studies that do not show true changes in BMD, do not allow differentiation between cause and effect, and can be hampered with bias from cohort-effects [63, 65, 73, 75, 77, 78, 80]. The few prospective studies examining the contribution of circulating sex steroids on bone loss have a relatively small sample size, which make them vulnerable to selection bias, and most of the reported associations are weak [9, 81-83, 88]. Generalizability is uncertain in some of the studies because participants are recruited by use of medical records, advertisements or membership in health insurance companies, without comparison of the participants with their target population [63, 64, 75, 81, 82]. In addition, the higher number of studies with an association may be the result of publication bias.

Whether covariates are controlled for such as BMI and SHBG may contribute to disparate results [65, 73, 75]. In agreement with Khosla et al. [81] free estradiol was associated with bone loss before, but not after we controlled for BMI in men. As previously reported, SHBG was associated with bone loss in both sexes, but the associations were weak [73, 76]. Contrary to previous reports, age-adjusted SHBG was associated with bone loss, but not after adjustment for BMI and smoking in postmenopausal women [9, 88]. The effect of SHBG can thus be mediated by BMI, known to be associated with SHBG and bone loss [64, 73, 75, 76].

The weak association between circulating sex steroids and bone loss in the present study does not exclude an important relation. If a true strong relation exists between circulating sex steroids and bone loss, many factors may obscure this association such as variability in the assay methods, fluctuations in the hormone levels due to cyclic nature of estrogen synthesis in

premenopausal women, small changes in BMD during the observation period and weak relationship between estrogen and bone remodelling. Most studies in this field have used immunoassays with a CV between 10 and 20% in at least one of the sex steroid measurements [9, 63, 65, 75, 81, 82]. Except for the rapid bone loss related to the sharp fall in estradiol in women during the menopausal transition, the change in BMD and sex steroids is slow in both sexes [9, 81, 82]. If circulating sex steroids and SHBG explain between 1-2% of an annual bone loss of 0.2-0.8%, a possible effect is obviously hard to detect [73, 81].

Few prospective data are available examining the contribution of sex steroids and SHBG on risk of incident fractures [10-12, 90, 93, 94]. Low estradiol is reported to be associated with increased risk of hip fractures in women [10, 11] and in men [94], but the effect was dependent on weight in one study [11]. That higher SHBG was associated with increased risk of incident non-vertebral fractures in both sexes in the present study, is in agreement with results from previous studies in women [10-12], whereas this is not studied in prospective data in men. The lack of association between circulating sex steroids and fractures in the present study may partly be the result of measurement uncertainty, or because many factors other than estrogen deficiency or bone fragility may contribution to risk of non-vertebral fractures, particular falls.

6.2.2 Biological mechanisms

Sex steroids have important effects on the skeleton in women and men, but this do not necessarily imply that the circulating levels of sex steroids play the main role in it [1, 8, 40, 42, 54, 63, 76, 88, 149, 150]. The main source of estradiol in postmenopausal women and men is peripheral conversion of circulating androgens in adipose tissue [40]. Bone possesses aromatase activity [68, 69] and local rather than circulating sex steroids regulate bone remodelling and appears to be responsible for mineralization [65, 68, 69, 73, 75, 83]. The

CYP19 genotype that encode aromatase, are reported to predict forearm bone loss independently of circulating bioavailable estradiol [83], and aromatase-inhibitors accelerate bone loss and increase fracture incidence [151].

The sex steroids produced extragonadally undoubtedly have intracrine and paracrine actions [40, 56, 66]. The term intracrinology describes the formation of sex steroids that exert their action in the same cells in which synthesis takes place, without release into the general circulation [66]. Local regulation of hormone activity in target tissues has obvious advantages. It enables fine-tuning of the intracellular concentration of active metabolites in a tissue in the presence of a wide range of circulating concentrations of hormones, thus providing mechanisms for tissue-specific responses in the absence of changes in systemic hormone production, and for the preservation of homeostasis in the face of alterations in hormonal status [40, 152]. However, this phenomenon limits the interpretation of the effects of circulating sex steroids [66]. This local estrogen production may play an important but hitherto largely unrecognised, physiological, and pathophysiological role, while the effect of circulating estradiol may be less important. Circulating estradiol levels may not be reflecting the tissue levels of estradiol, and the circulating estrogen can have little impact on the relatively high tissue levels of estrogen [68, 83, 149]. Moreover, estrogen production at the extragonadal sites is dependent on circulating androgen precursors, including testosterone [68]. In the presence of low postmenopausal estradiol levels, testosterone and DHEAS could be important for bone health in women. This was the case in our cross-sectional data, but not confirmed in our follow-up data. In men, higher serum levels of testosterone may protect against bone loss [68]. Testosterone can have a direct action on the osteoblasts through androgen receptors or an indirect action by aromatisation to estradiol [40, 54, 63].

During the last decade major progress has been made on elucidating the molecular mechanisms of estrogen action on bone cells. However, the mechanisms responsible for

increased bone remodelling at estrogen deficiency are still incompletely understood [19]. The marrow microenvironment play an essential role in bone loss by providing cytokines such as tumor necrosis factors (TNF), interleukins (IL-1 and IL-6) and macrophage colony-stimulating factor (M-CSF) [19, 40]. Systemic factors such as PTH, estrogen and 1,25 dihydroxyvitamin D3, and local factors regulate osteoclastogenesis functions trough receptors expressed in cells of the osteoblasts lineage [19]. The osteoblasts produce receptor activator of nuclear factor kappa B ligand (RANKL) and osteoprotegerin (OPG) [153]. RANKL acts via the respective receptor RANK on the osteoclasts and promotes their differentiation and activation, while OPG prevent interaction of RANKL with RANK and is and effective inhibitor of osteoclast formation. Estradiol increases OPG, and decreases M-CSF and RANK [154-157], but estradiol has not yet been shown to regulate RANKL or RANK directly [40]. An understanding of these local mechanisms will make it possible to interpret how estrogen deficiency results in bone loss in women.

Secondary hyperparathyreoidism might increase remodelling further in elderly women and men [63]. Age impairs intestinal calcium absorption and renal calcium conservation, and PTH increase to maintain serum ionic calcium by resorption of bone that contains 99% of body calcium stores [158]. Because estrogen increases intestinal calcium absorption by acting through intestinal estrogen receptors, it has been hypothesised that estrogen can mediate changes in PTH [40]. Whether estrogen deficiency is partly responsible for bone lose related to secondary hyperparathyreoidism is not clear [37].

Most authors suggest that higher SHBG increases bone loss and the risk of fractures by binding estradiol and decreasing its bioavailability [10, 12, 76, 78, 79, 88-90]. The independent effect of SHBG after adjustment for estradiol suggests other influences of SHBG on bone (Paper III). Although the fracture risk predicted by SHBG was partly dependent on BMD, the combination of SHBG and BMD predicted fracture risk slightly better than BMD

alone. This supports the notion of an independent effect of SHBG other than by affecting the BMD or bioavailability of estradiol, and this combination may identify a subgroup with high risk of fractures. However, a mathematical model may not necessarily adequately explain all the complex biological relations between hormones and other factors [65]. The interrelationships between all of the hormones and SHBG make it difficult to determine which of the parameters that have direct or indirect effect on bone [73]. SHBG is more than a transport protein. It may exert direct cellular functions by acting through specific membrane receptors [85]. There is no such evidence for bone, but an anti-estrogenic effect of SHBG is reported in breast cancer cells.

6.2.3 Assessment of fracture risk

The lifetime risk for fragility fractures after age of 50 years is about 50% in women and 20% in men [28]. The resultant high morbidity, mortality and economical costs are well recognised, and have stimulated the development of effective interventions to reduce fracture risk. Despite these advances, there are many challenges left on how to reduce this public health burden. One challenge is to better identify individuals with a high risk of fracture to target treatment more cost-effectively.

There are no clinical tools available to assess bone quality independently of bone density, so that for practical purposes the diagnosis of osteoporosis depends upon the measurement of skeletal mass, as assessed by measurements of BMD [21]. The use of BMD measurements for prognosis depends upon accuracy, which in this context is the ability of the measurement to predict fracture. BMD has high specificity but low sensitivity of fractures [21]. There are a number of clinical risk factors that provide information on fracture risk over and above that given by BMD. These include age, prior fragility fracture, a parental history of hip fractures and use of corticosteroids [21]. In general, risk factors scores show relatively poor specificity

and sensitivity in predicting fractures risk [18, 21, 159-165]. Although few individuals have many risk factors, their use in conjunction with BMD improves sensitivity of fracture prediction.

Fractures risk is often expressed as a relative risk that has different meaning in different contexts [21]. In the absence of validated population screening strategies, a case finding strategy can be developed based on the assessment of fracture probability utilising clinical risk factors, and where appropriate additional testing such as BMD. Kanis et al. recommend that risk of fracture should be expressed as a fixed-term absolute risk, i.e. probability over a 10-year interval [20, 21].

7. Implications and further research

In this context we hoped to find that measurements of sex steroids or SHBG could be relevant risk factors that could improve the sensitivity to fracture prediction. Conclusions reporting levels of estradiol or SHBG to play an important role or to be an important risk factor for bone loss [9, 73, 75, 82] might make clinicians think that such measurements could assist in decision making regarding fracture risk susceptibility. Our results do not support this notion. Careful reading of the literature does not show any convincing strong relation between circulating levels of sex steroids, bone loss and risk of fractures in any large prospective, population-based study, where possible confounders are adjusted for. Most of the reported associations are weak, and most of the results are from relatively small studies [9-12, 81-83, 88, 90, 93, 94]. So we are not convinced from existing literature that circulating levels of sex steroids and SHBG are relevant risk factors for bone loss or fracture. These findings explain why such measurements are not included among the clinical risk factors for identifying persons at risk of fracture.

Although the results in our papers on bone loss and fractures are basically negative, it is a contribution to our understanding of the role of circulating sex steroids in the pathogenesis of bone fragility. We do not doubt the important effect of estradiol and also testosterone in the cellular mechanisms in bone. This is well documented [36, 40, 54, 152, 155]. To clarify whether the small effects of circulating sex steroids on bone loss and fracture risk is a result of measurement uncertainty or because local rather than circulating sex steroids regulate bone remodelling [65, 66, 68, 69, 75, 83, 89, 149], will need future large prospective studies including better methods for hormone measurements. We also need further biological studies to elucidate what causes high SHBG levels, and by which mechanisms are sex steroids and SHBG involved in bone remodelling.

For a better understanding of the contribution of sex steroids on bone fragility we plan to study whether parity and age at menarche predict bone loss and fractures, and whether pregnancy and lactation contribute to the loss of bone that starts in young adulthood. Although the baseline sex steroids are unlikely to improve the sensitivity of fracture prediction, we want to study whether true changes in the sex steroids, particular during menopause, are related to bone loss and fracture risk. After menopause, bone loss accelerates, and we want to study the structural basis underlying this loss of bone and the hormonal factors that contribute.

The reason for studying bone structure is that fracture means structural failure [34]. Progress in understanding the pathogenensis of bone fragility is hampered by the inaccessibility of bone for investigation, but recent advances in non-invasive technology provide a quantitative measurement of the three dimensional structure of bone [166, 167]. It remains to be shown whether selection of individuals based on morphology will increase the sensitivity and specificity of fracture prediction. As there is no prospective study testing this in vivo, we hope to be able to study this in a future Tromsø Study. We would like to

investigate the distribution of bone structure traits in a general population and investigate whether bone structure traits, as cortical thickness, trabecular number or thickness predicts fractures better then BMD measured by DXA.

8. Conclusions

Our results have taught us that there still is uncertainty on whether total and free estradiol decrease by aging in men. The higher levels of FSH by aging in postmenopausal women were new knowledge. Total and free estradiol showed a seasonal variation in postmenopausal women and in men. That level of sex steroids and SHBG were weak predictors of bone loss and only SHBG predicted non-vertebral fractures in both sexes taught us that the effects of circulating sex steroids are small. The combination of low BMD and high SHBG may identify a subgroup with increased risk of fractures, but measurement of sex steroids or SHBG are unlikely to assist in clinical decision making.

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Endogenous Sex Hormones in Relation to Age, Sex, Lifestyle Factors, and Chronic Diseases in a General Population: The Tromsø Study

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The role played by endogenous hormones in many diseases makes it important to understand factors influencing their levels. This study examined the distribution of total and free estradiol, FSH, and dehydroepiandrosterone sulfate (DHEAS) by age and sex and associations of these hormones with body mass index (BMI), lifestyle factors, and chronic diseases. Plasma samples taken from 1555 men and 1952 women 25–84 yr of age in 1994–1995 Tromsø Study were analyzed in 2001.

Total estradiol increased with age among men ($P < 0.001$), with or without adjustment for BMI and lifestyle factors. FSH increased with age both in men ($P < 0.001$) as well as pre- ($P < 0.001$) and postmenopausal women ($P = 0.01$) after similar adjustment, and DHEAS decreased with age in both sexes ($P < 0.001$).

With increasing BMI, free estradiol increased in men ($P = 0.004$), total and free estradiol increased in postmenopausal women ($P < 0.001$), and FSH decreased in men ($P = 0.03$) and postmenopausal women ($P < 0.001$).

Men with chronic diseases had lower levels of DHEAS, compared with healthy men ($P < 0.001$). Smokers had higher DHEAS levels than nonsmokers. Further studies are needed to confirm these hormonal changes with age and disease. (*J Clin Endocrinol Metab* 89: 6039–6047, 2004)

HORMONES PLAY AN important role in human physiology. Sex hormones are involved in the pathophysiology of common diseases such as osteoporosis (1, 2), fractures (3, 4), breast cancer (5, 6), and cardiovascular diseases (7–9), which makes it important to understand factors that influence the levels of these hormones.

There are major differences in the levels of sex hormones with age among men and women. Hormonal changes through the menstrual cycle and the menopausal transition are well described, a greater lack of clarity is present among postmenopausal women. The menopausal transition in women is characterized by a relatively abrupt decrease in the level of total estradiol and an increase in the level of FSH (10). The effects of age and body mass index (BMI) on the levels of total estradiol and FSH are relatively small, compared with the large effect of the menopause (10). Decreased or unchanged FSH (11–13) and decreased or unchanged estradiol (12, 13) with age are reported among postmenopausal women. Most authors describe a fall in FSH whereas estradiol is unchanged with age in this group, although divergent results exist. Use of hormone replacement therapy (HRT) increases total serum estradiol and decreases the level of FSH (14). Dehydroepiandrosterone sulfate (DHEAS) levels gradually decrease with age independent of menopausal transition (15, 16).

In men, the influence of age on estradiol level is not clear. Decreased (17, 18) and unchanged levels are reported (1, 19–21). The FSH levels increase (19, 22), and the DHEAS levels gradually decrease with age in men (15, 19).

BMI is positively associated with total estradiol (12, 21) and negatively associated with FSH (12, 23) in both men and postmenopausal women. BMI is negatively associated with DHEAS among men (7, 21, 24) but not among women (7, 25).

Smokers have higher levels of DHEAS than nonsmokers of both sexes (7, 26); otherwise smoking and other lifestyle factors are reported to be either marginally or not at all associated with sex hormones (5, 21). Except for being a large source of sex hormones (27), the role of DHEAS in aging and disease is still insufficiently understood (7–9) and remains controversial.

The aim of this study was to describe the distribution of total and free estradiol, FSH, and DHEAS by age, sex, menopausal status, and use of HRT as well as the associations of total and free estradiol, FSH, and DHEAS with BMI, selected lifestyle factors, and chronic diseases in an unselected general population.

Subjects and Methods

Study population

The Tromsø study was initiated in 1974 with surveys repeated in 1979–1980, 1986–1987, and 1994–1995. The study is a single-center,

* M.M. is deceased.

Abbreviations: ANCOVA, Analysis of covariance; BMI, body mass index; CI, confidence interval; DHEAS, dehydroepiandrosterone sulphate; HRT, hormone replacement therapy; IHD, ischemic heart disease; LGS IUS, levonorgestrel-releasing intrauterine system.

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population-based prospective study in the municipality of Tromsø, Northern Norway. The fourth survey of the Tromsø population started in September 1994 and was completed in September 1995. The University of Tromsø, in cooperation with the National Health Screening Service, conducted the survey. The Regional Committee for Medical Research Ethics recommended the study. All the participants gave informed written consent.

All inhabitants over 24 yr of age were invited, and 27,159 subjects (77%) participated in the main survey (phase I). A protocol similar to that used during previous surveys in this population was followed (28, 29). From the main survey, all men aged 55–74 yr, all women aged 50–74 yr, and 5–10% random samples of the other age groups of both sexes were invited to an extended examination, including forearm bone densitometry and blood sampling (phase II). In addition, 328 male participants of the Family Intervention Study, selected on the basis of high total cholesterol or low high-density lipoprotein to total cholesterol ratio (30), were also invited. A total of 7948 individuals participated in this Tromsø Osteoporosis Study (31). Among these study participants, a subgroup of 3685 was selected for hormone assays, and blood samples from 3564 participants were available for analyses. We excluded 10 participants due to pregnancy (n = 4) or use of testosterone (n = 2), progestin (n = 1), selective estrogen receptor modulator (n = 1), or corticosteroids (n = 2). To make the analyses consistent, we also excluded 47 participants with missing values on one or more hormones, BMI, or lifestyle factors. Of the total 1952 women, all 380 women on combined HRT, estriol, or hormonal contraception are described separately but not included in the regression models. A total of 1555 men and 1572 women with hormone measurements are thus included in the main part of this study.

Questionnaires

Two extensive self-administered questionnaires were used as instruments to gain information on a broad set of variables. In the present study, we included information on previous and present diseases such as cancer; asthma; diabetes; stroke and ischemic heart disease (IHD); use of any medication; current smoking status; and consumption of cigarettes, alcoholic beverages, and coffee. An alcohol intake score was constructed by adding the number of glasses of beer, wine, and spirits, assuming an equal amount of alcohol in one unit of each type. A physical activity score was made by adding the hours per week of moderate and hard physical activity, giving the hours with hard activity double weight: score = moderate + 2 hard.

Menopause and use of hormonal therapy

Women were asked to report whether they were pregnant; the date of the last menstrual period if they still were menstruating; and if not, their age at menopause. They were also asked about the use of oral, transcutaneous, or vaginal estrogen therapy, contraceptive pills, and hormonal intrauterine device and the brand they currently used.

At blood sampling, menstruating women were not asked to report their date of the last menstrual period; therefore, the information on menstrual phase at blood sampling is lacking.

Women who reported use of hormonal therapy (n = 380) were on combined HRT (n = 215), oral or vaginal estriol (n = 111), contraceptive pills (n = 22), or levonorgestrel-releasing intrauterine system (LGS IUS) (n = 32). Only nonusers of these hormones were classified into pre-, peri-, or postmenopausal groups because the menopausal status could not be determined accurately among some of the hormone users. We have chosen to present results with the highest possible n, and therefore our definition of postmenopausal status was based on the self-reported menopause and age. Women who reported that they had stopped menstruating over a year ago (n = 900) or were 54 yr of age or older (n = 434) were defined as postmenopausal (n = 1334). Women who had stopped menstruating within the last 3–12 months and were 45 yr or older or with unreported last menstrual period and between 45 and 54 yr were considered perimenopausal (n = 33). Women who had their last menstrual period within the last 3 months (n = 176) or within the last 3–12 months and were younger than 45 yr of age (n = 6) or had missing menstruation data and were younger than 45 yr of age (n = 23) were defined as premenopausal (n = 205). This definition of menopausal status left none of the women undefined due to missing or conflicting values. Participants were not asked about previous hysterectomy or oophorectomy; accordingly, information on surgical cause of menopause was not available. Total number of postmenopausal years was calculated as current age minus age at menopause when present.

Measurements

Height and weight were measured in light clothing without shoes, and BMI was calculated as weight in kilograms divided by the square of height in meters.

Hormone assays

Nonfasting blood samples were taken between 0800 and 1600 h. Serum samples were stored at −70 C until they were first thawed for analyses of sex hormones in 2001, after a storage time of 6–7 yr. All hormones and SHBG were measured on Immulite 2000 (Diagnostic Products Corp., Los Angeles, CA). Total estradiol and DHEAS measurements were based on competitive immunoassays, whereas FSH and SHBG measurements were based on immunometric assays. The assays were run daily on randomly selected samples in batches of 100 per day.

The intra- and interassay coefficient of variation for the measurements of total estradiol, FSH, DHEAS, and SHBG were between 3.5 and 10%, depending on the level, and the levels of sensitivity were 10 pmol/liter, 0.5 IU/liter, 1.0 μmol/liter, and 1.0 nmol/liter, respectively. Samples with values below functional assay sensitivity were given a value midway between zero and assay sensitivity for the analyses: total estradiol (354 women and 61 men), FSH (eight women), and DHEAS (419 women and 85 men). Total estradiol values greater than 7340 were recoded to 7340 pmol/liter (two women), FSH greater than 170 was recoded to 170 IU/liter (one woman), and SHBG greater than 180 was recoded to 180 nmol/liter (36 women and seven men).

Free estradiol values were calculated from total estradiol and SHBG levels according to the equation:

$$FE = (TE - N \times FE)/(Ke(SHBG - TE + N \times FE)),$$

where FE is the concentration of free estradiol, TE is the concentration of total estradiol, $N = Ka \times Ca + 1$ (Ka is the association constant of albumin for estradiol = 4.21×10^4 liter/mol, and Ca is the albumin concentration set to 6.2×1^{-4} mol/liter), Ke is the association constant of SHBG for estradiol = 0.31×10^9 liter/mol (32, 33).

Statistical analysis

The SAS software package (version 8.2, SAS Institute Inc., Cary, NC), was used for both data management and analysis. For statistical analysis the values of the hormones were log transformed to correct for skewed distribution. Differences in means between groups were tested by ANOVA, analysis of covariance (ANCOVA), and adjusted t tests for multiple comparisons of pairs of means. Data are presented stratified by age. Linear trend by age was analyzed in linear regression models. Multiple linear regressions were used to model total estradiol, free estradiol, FSH, and DHEAS with the following independent variables: age, BMI, smoking, alcohol and coffee consumption, physical activity, sampling hour and season. Season and sampling hour were dropped from the final model because adjustment for them did not change any of the results. The same set of independent variables, which was significantly associated with at least one hormone, was used systematically in all hormone models among both women and men. The variables describing smoking and alcohol use had a substantial proportion of zero values. Therefore, we analyzed these variables dichotomized to check for any difference in hormone levels between smokers *vs.* nonsmokers and alcohol users *vs.* nonusers. Thereafter we checked for any dose-response effects among smokers and users of alcohol. Number of cigarettes, number of alcohol units, age at menarche, and number of menopausal years were not associated with any hormone when they were used as independent variables in multiple linear regression models. These variables are therefore not presented in the results.

The analyses were performed separately for pre- and postmenopausal women. The perimenopausal women were excluded from the multiple regression analyses due to small numbers. All P values are based on analyses of log-transformed values, and P values are two sided. However, means and confidence limits are transformed back to the original units when presented.

Results

Characteristics of the study population are shown in Table 1. The mean, median, and 95th percentile for age at menopause were 48, 49, and 55 yr, respectively, and 85% of women not using any hormone were postmenopausal by an average of 12 yr. The mean age was 38.1 (range 25–54) yr, 50.5 (range 39–57) yr, and 64.1 (range 37–83) yr among pre-, peri-, and postmenopausal women, respectively.

The geometric means and 95% confidence interval (CI) for the population were as follows: total estradiol, 49.7 (12.7–194.7) pmol/liter; free estradiol, 1.2 (0.3–4.6) pmol/liter; FSH, 7.5 (1.9–29.6) IU/liter; and DHEAS, 2.8 (0.7–11.0) μmol/liter among 1555 men, and total estradiol, 28.6 (2.5–328.5) pmol/liter; free estradiol, 0.6 (0.05–7.0) pmol/liter; FSH 48.0 (7.7–301) IU/liter; and DHEAS, 1.5 (0.3–6.8) μmol/liter among 1572 women.

Age and sex

Age- and sex-stratified distributions of total and free estradiol, FSH, and DHEAS are shown in Table 2 and Fig. 1. Among men, total ($P < 0.001$) and free estradiol levels ($P = 0.04$) were positively associated with age and were, respectively, 33 and 12% higher at age older than70 yr, compared with men younger than 40 yr. Among postmenopausal women, total ($P = 0.04$) and free estradiol levels ($P = 0.008$) were negatively associated with age and were 90% lower at

TABLE 1. Characteristics of 1555 men and 1952 women (The Tromsø study 1994–95)

Characteristics	Men			Women		
	Mean	SD	Range	Mean	SD	Range
Age (yr)	60.0	10.1	25–84	59.7	11.0	25–83
Height (cm)	175.2	6.9	140–198	161.8	6.4	138–184
Weight (kg)	80.3	12.1	44–127.5	68.0	12.0	38–143.5
BMI (kg/m^2)	26.1	3.4	16.0–39.3	26.0	4.5	16.2–52.1
No. of cigarettes/day among smokers	13.3	6.7	2–50	10.7	5.2	1–40
No. of alcohol units/2 wk among drinkers	4.8	7.3	0–104	1.9	3.4	0–34
Coffee, no. of cups/day	6.0	3.8	0–31	4.9	2.9	0–25
Physical activity score[a]	3.7	2.5	0–9	2.9	2.2	0–9
Age at menopause (yr)				48.5	4.9	20–60
Years since menopause				12.4	7.2	0–40
Current smokers (%)	35.5			31.0		
Teetotallers (%)	12.7			27.3		
Myocardial infarction (%)	9.3			3.1		
Angina pectoris (%)	10.9			7.5		
Stroke (%)	2.8			2.0		
Diabetes (%)	3.2			2.8		
Cancer (%)	3.9			6.9		
Asthma (%)	7.1			9.6		

[a] A physical activity score was made by adding the hours per week of moderate (m) and hard (h) physical activity, giving the hours with hard activity double weight: score = m + 2 h.

TABLE 2. Geometric means, 95% CI for the means and *P* values for trend of total and free estradiol, FSH, and DHEAS by age in 1555 men, 205 premenopausal women, and 1334 postmenopausal women not on hormone therapy affecting ovarian function (The Tromsø study 1994–95)

Age (yr)	n	Total estradiol (pmol/liter)		Free estradiol (pmol/liter)		FSH (IU/liter)		DHEAS (μmol/liter)	
		Mean	95% CI	Mean	95% CI	Mean	95% CI	Mean	95% CI
Men									
25–39	81	43	36–50	1.12	0.96–1.32	4.28	3.73–4.90	5.88	5.29–6.53
40–49	126	42	36–48	1.06	0.93–1.22	5.38	4.86–5.96	4.36	3.98–4.77
50–59	467	47	45–50	1.15	1.08–1.22	6.89	6.50–7.31	3.24	3.07–3.43
60–69	597	51	48–54	1.17	1.10–1.24	8.05	7.64–8.49	2.38	2.26–2.52
70–84	284	57	53–62	1.25	1.16–1.35	10.11	9.27–11.02	1.87	1.73–2.03
All	1555	50	48–51	1.16	1.13–1.21	7.50	7.25–7.77	2.75	2.66–2.85
P trend			<0.001		0.04		<0.001		<0.001
Premenopausal women									
25–39	120	185	159–215	3.94	3.42–4.55	4.99	4.49–5.55	3.85	3.54–4.19
40–49	71	188	142–249	3.63	2.77–4.77	7.57	6.14–9.32	2.65	2.29–3.06
50–59	14	108	60–193	2.36	1.32–4.23	22.32	13.30–37.44	2.84	2.08–3.86
All	205	179	156–206	3.70	3.24–4.23	6.38	5.69–7.16	3.31	3.07–3.58
P trend			0.23		0.11		<0.001		<0.001
Postmenopausal women									
25–49	7	43	12–158	0.76	0.19–2.94	46.01	20.61–102.70	1.74	0.99–3.05
50–59	379	22	20–25	0.46	0.41–0.52	65.70	62.78–68.74	1.68	1.56–1.81
60–69	630	21	20–23	0.43	0.40–0.46	65.92	63.91–67.99	1.29	1.22–1.36
70–84	318	20	18–22	0.38	0.34–0.43	67.66	65.14–70.28	1.10	1.02–1.19
All	1334	21	20–22	0.43	0.40–0.45	66.14	64.71–67.60	1.34	1.29–1.40
P trend			0.04		0.008		0.17		<0.001

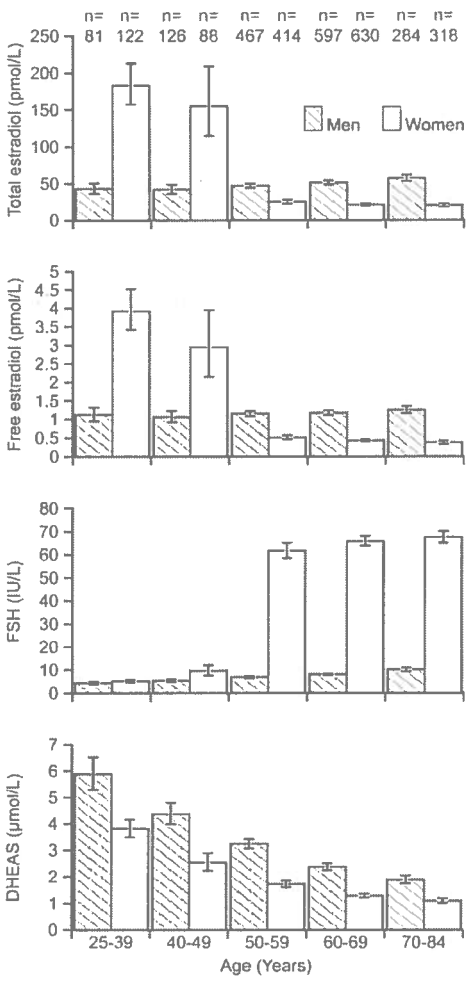

FIG. 1. Geometric means and 95% CI for the means of total and free estradiol, FSH, and DHEAS by age and sex among 1555 men and 1572 women not on hormone therapy affecting ovarian function: The Tromsø Study 1994–1995.

age older than 70 yr, compared with premenopausal women younger than 40 yr. In men and premenopausal women, FSH was positively associated with age ($P < 0.001$). Men and women older than 70 yr had, respectively, 2.4- and 13.2-fold higher levels of FSH than those younger than 40 yr. An abrupt change in the levels of estradiol and FSH was obvious among women at approximately 50 yr. DHEAS was negatively associated with age in both sexes ($P < 0.001$). The DHEAS levels at age older than 70 yr were 32 and 29% of the levels at age younger than 40 yr among men and women, respectively. The levels of DHEAS were higher among men than women ($P < 0.001$). Except for free estradiol among men ($P = 0.09$), the associations between age and all hormones studied were significant among men ($P < 0.001$) and postmenopausal women ($P = 0.02$, $P < 0.001$, $P = 0.01$, and $P < 0.001$) after adjusting for BMI and lifestyle factors. Among premenopausal women only FSH and DHEAS were associated with age after similar adjustment ($P < 0.001$).

BMI

Among men, only free estradiol was positively associated ($P = 0.004$), whereas FSH was negatively associated ($P = 0.03$) with BMI after adjustment for age and lifestyle factors (Fig. 2). Among postmenopausal women, BMI was positively associated with total and free estradiol and negatively as-

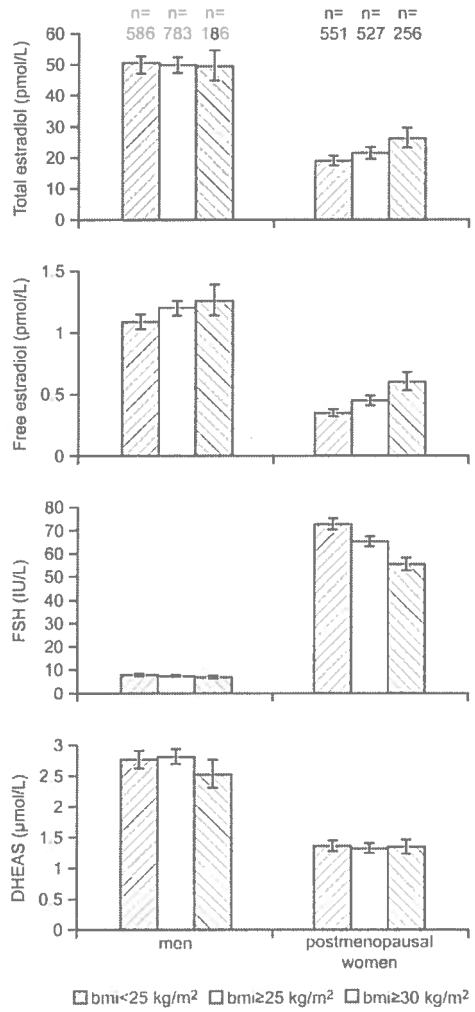

FIG. 2. Geometric means and 95% CI for the means of total and free estradiol, FSH, and DHEAS by BMI and sex adjusted for age, smoking, alcohol, coffee, and physical activity by ANCOVA among 1555 men and 1334 postmenopausal women not on hormone therapy affecting ovarian function: The Tromsø Study 1994–1995.

sociated with FSH after similar adjustment ($P < 0.001$) (Fig. 2). The levels of total and free estradiol were, respectively, 39 and 71% higher at BMI of 30 kg/m^2 or greater, compared with postmenopausal women with BMI less than 25 kg/m^2. The levels of DHEAS were not associated with BMI for either sex.

Only a small part (1–5%) of the variation in total and free estradiol levels of both sexes was explained by age, BMI, and lifestyle factors. The same independent variables explained more of the variation in FSH and DHEAS in both sexes (6–24%).

Chronic diseases

The DHEAS levels were lower among 243 men with self-reported IHD ($P < 0.001$), 43 men with stroke ($P = 0.001$), 50 men with diabetes ($P < 0.001$), 55 men with cancer ($P = 0.007$), and 110 men with asthma ($P = 0.003$), compared with 1128 healthy men after adjusting for age, BMI, and lifestyle factors (Table 3).

Menopausal status or HRT

The mean levels of all hormones studied were different among pre-, peri-, and postmenopausal groups ($P < 0.001$) (Table 4). After adjustment for age, BMI, and lifestyle factors, the differences in hormone levels by menopausal status persisted regarding total and free estradiol and FSH but not DHEAS.

Postmenopausal HRT users had higher mean levels of total and free estradiol and lower mean levels of FSH than nonusers of the same age (Table 5). The levels of DHEAS did not differ between users of HRT and nonusers. Users of contraceptive pills had a significantly lower mean level of free estradiol and FSH. The levels of estradiol did not differ between users of estriol and nonusers.

Other findings

Smoking was positively associated with DHEAS among men ($P < 0.001$) (Fig. 3) and premenopausal ($P = 0.02$) and postmenopausal women ($P = 0.03$) (data not shown). Coffee was positively associated with FSH and DHEAS ($P = 0.03$) among men and DHEAS among premenopausal women ($P = 0.01$) and postmenopausal women ($P = 0.03$). This was the case when the significance of the independent variables smoking and coffee was analyzed in separate models among women. With both variables in the model, none were significant (Fig. 3), probably due to an intercorrelation (Pearson correlation coefficient $r = 0.38$). Estradiol levels were not associated with smoking for either sex (Fig. 3).

Physical activity was negatively associated with FSH among men ($P = 0.01$). Alcohol was positively associated

TABLE 3. Geometric means, 95% CI for the means of total and free estradiol, FSH and DHEAS by chronic diseases, and *P* values for comparisons of healthy participants with participants with chronic diseases adjusted for age, BMI, smoking, alcohol, coffee, and physical activity by ANCOVA, among 1555 men and 1334 postmenopausal women not currently using hormone therapy affecting ovarian function (The Tromsø study 1994–95)

	n	Total estradiol (pmol/liter)			Free estradiol (pmol/liter)			FSH (IU/liter)			DHEAS (μmol/liter)		
		Mean	95% CI	*P*	Mean	95% CI	*P*	Mean	95% CI	*P*	Mean	95% CI	*P*
Men													
Healthy	1128	50	48–52		1.16	1.12–1.21		7.44	7.15–7.73		2.90	2.79–3.01	
Chronic disease	427	50	46–53	0.94	1.17	1.09–1.25	0.91	7.68	7.20–8.20	0.41	2.41	2.26–2.56	<0.001
IHD	243	50	46–55	0.90	1.19	1.09–1.30	0.64	7.22	6.63–7.86	0.70	2.43	2.24–2.63	<0.001
Stroke	43	50	41–62	0.89	1.23	1.00–1.52	0.57	8.76	7.20–10.67	0.06	2.22	1.85–2.66	0.001
Diabetes	50	54	44–66	0.38	1.32	1.09–1.61	0.19	7.55	6.29–9.07	0.65	2.18	1.84–2.58	<0.001
Cancer	55	40	33–48	0.03	0.90	0.75–1.09	0.01	7.89	6.63–9.38	0.36	2.41	2.05–2.82	0.007
Asthma	110	53	47–61	0.24	1.26	1.10–1.43	0.24	7.69	6.81–8.68	0.35	2.53	2.26–2.82	0.003
Postmenopausal women													
Healthy	960	21	20–22		0.42	0.40–0.45		66.63	64.97–68.33		1.35	1.29–1.41	
Chronic diseases	374	21	19–23	0.90	0.43	0.39–0.48	0.73	64.89	62.30–67.60	0.29	1.33	1.24–1.43	0.77
IHD	150	22	19–26	0.65	0.45	0.38–0.53	0.59	64.82	60.83–69.06	0.42	1.40	1.24–1.57	0.62
Stroke	35	18	13–25	0.41	0.37	0.26–0.52	0.44	64.56	56.87–73.29	0.60	1.33	1.05–1.68	0.82
Diabetes	48	20	15–26	0.64	0.41	0.31–0.55	0.86	59.15	53.03–65.96	0.03	1.33	1.09–1.63	0.83
Cancer	89	23	19–29	0.33	0.48	0.39–0.59	0.23	68.16	62.89–73.87	0.69	1.43	1.24–1.66	0.54
Asthma	132	20	17–24	0.69	0.41	0.34–0.49	0.64	64.06	59.88–68.53	0.27	1.27	1.12–1.43	0.27

IHD, Ischemic heart disease (angina and/or myocardial infarction); healthy, none of these chronic diseases. For men/postmenopausal women missing IHD (n = 3/4), stroke (n = 10/5), diabetes (n = 8/7), and cancer (n = 151/266), asthma (n = 11/13), included in the healthy group.

TABLE 4. Geometric means, 95% CI for the means of total and free estradiol, FSH, and DHEAS by menopausal status, and *P* values for comparison of pre- and perimenopausal women with postmenopausal women by ANOVA, among 1572 women not currently using hormone therapy affecting ovarian function (The Tromsø study 1994–95)

	n	Total estradiol (pmol/liter)			Free estradiol (pmol/liter)			FSH (IU/liter)			DHEAS (μmol/liter)		
		Mean	95% CI	*P*	Mean	95% CI	*P*	Mean	95% CI	*P*	Mean	95% CI	*P*
Premenopausal	205	179	156–206	<0.001	3.70	3.24–4.23	<0.001	6.38	5.69–7.16	<0.001	3.31	3.07–3.58	<0.001
Perimenopausal	33	67	38–118	<0.001	1.39	0.81–2.38	<0.001	30.39	21.23–43.52	<0.001	2.08	1.68–2.58	<0.001
Postmenopausal	1334	21	20–22		0.43	0.40–0.45		66.14	64.71–67.60		1.34	1.29–1.40	
All	1572	29	27–30		0.58	0.55–0.62		47.97	45.80–50.24		1.52	1.47–1.58	

TABLE 5. Geometric means, 95% CI for the means of total and free estradiol, FSH, and DHEAS by age and by current use of hormone therapy: HRT, oral/vaginal estriol treatment, oral contraceptive pills or LGS IUS, and *P* values for comparison of users with nonusers by ANOVA among 1952 women (The Tromsø study 1994–95)

Age (yr)	n	Total estradiol (pmol/liter)			Free estradiol (pmol/liter)			FSH (IU/liter)			DHEAS (μmol/liter)		
		Mean	95% CI	*P*	Mean	95% CI	*P*	Mean	95% CI	*P*	Mean	95% CI	*P*
Nonusers of hormone therapy													
25–49	210	171	146–199		3.48	3.00–4.04		6.68	5.91–7.55		3.22	2.98–3.49	
50–59	414	25	22–27		0.51	0.46–0.57		61.72	58.53–65.09		1.72	1.61–1.85	
60–84	948	21	19–22		0.41	0.39–0.44		66.50	64.91–68.12		1.22	1.17–1.28	
All	1572	29	27–30		0.58	0.55–0.62		47.97	45.80–50.24		1.52	1.47–1.58	
HRT													
25–49	19	150	78–288	0.67	2.74	1.47–5.12	0.43	17.09	10.06–29.01	<0.001	2.79	2.08–3.73	0.31
50–59	128	125	106–147	<0.001	2.50	2.14–2.92	<0.001	25.71	21.37–30.92	<0.001	1.54	1.38–1.72	0.12
60–84	68	116	90–149	<0.001	2.22	1.72–2.85	<0.001	22.36	16.98–29.44	<0.001	1.11	0.92–1.33	0.25
All	215	124	108–142		2.42	2.12–2.77		23.72	20.46–27.51		1.46	1.33–1.61	
Oral/vaginal estriol treatment													
25–49	1	20			0.42			86.10			3.00		
50–59	28	21	15–29	0.43	0.43	0.30–0.61	0.39	77.12	68.95–86.26	0.10	1.42	1.10–1.84	0.17
60–84	82	20	17–25	0.88	0.41	0.34–0.50	0.95	63.03	57.64–68.91	0.32	1.08	0.92–1.26	0.11
All	111	20	17–24		0.41	0.35–0.49		66.50	61.79–71.57		1.17	1.02–1.34	
Levonorgestrel intrauterine system													
25–49	25	183	110–306	0.79	3.97	2.48–6.36	0.62	7.75	5.80–10.35	0.47	3.01	2.28–3.97	0.58
50–59	7	48	18–123	0.11	1.07	0.44–2.58	0.08	64.85	51.63–81.46	0.85	2.06	1.46–2.92	0.50
All	32	136	84–221		2.98	1.90–4.68		12.33	8.39–18.13		2.77	2.19–3.50	
Oral contraceptive pills													
25–39	22	98	40–242	0.06	1.55	0.62–3.87	0.004	2.28	1.28–4.05	<0.001	3.40	2.85–4.05	0.69

with DHEAS among men ($P = 0.02$). No other lifestyle factors or BMI was significantly associated with any hormone among premenopausal women.

Total and free estradiol were significantly associated with season among men and postmenopausal women ($P < 0.001$ by partial F test). However, adjustment for season did not change any of the above reported associations.

Discussion

This study demonstrates that with increasing age, the levels of total estradiol were higher among men, total and free estradiol were lower among postmenopausal women, and the levels of FSH were higher and DHEAS lower in both sexes. With increasing BMI, free estradiol was higher in men, and levels of both total and free estradiol were higher among postmenopausal women. However, men and postmenopausal women had lower levels of FSH with increasing BMI. We found that men with chronic diseases had lower levels of DHEAS, compared with healthy men.

To our knowledge, this is the largest population-based study in which FSH, DHEAS, and estradiol of both sexes have been measured. The high response rate (77% of the eligible population) in the main survey assures generalizability of results to a majority of the source population. However, the nonresponding minority might have characteristics that differ substantially from those found in the study population. In a previous paper (34), participants of the main study who were nonresponders at the second examination (phase II) were compared with responders of phase II. They were not found to be healthier in any age or sex group, as judged by the self-reported prevalence of chronic diseases. We looked for selection bias by comparing characteristics of the subgroup that participated in the hormone studies (n = 3507) with the total phase II survey population (n = 7948). The subgroup of men did not have significantly different characteristics. Although there was a mean age difference of 1 yr (59.4 *vs.* 58.2 yr, $P < 0.001$), higher prevalence of IHD (8.6 *vs.* 7.1%, $P < 0.05$) and higher percentage of teetotalers (27.3 *vs.* 24.5%, $P < 0.01$) among the subgroup of women participating in hormone studies in comparison with the total phase II survey population, this is not expected to have influenced our results substantially.

Our study is cross-sectional; therefore, the direction of the associations cannot be determined. The cross-sectional estimates of associations between hormonal levels and age or BMI are not true measures of longitudinal changes, and individual change in hormonal levels cannot be indicated.

Self-reported age at menopause is the most straightforward way of classifying menopausal status according to the recommendation of the World Health Organization (35). Because 93% of women with missing menopause data were older than 53 yr, the classification based on age is not expected to have biased this study. Results did not differ substantially whether or not women with a missing response to the menopause question were included.

Lack of information on hysterectomy before natural menopause is a limitation of our study (36). Nevertheless, because the rate of hysterectomy in Norway is reported to be remarkably low (37), we believe that this does not introduce a serious misclassification problem.

The mean interval between filling in the questionnaire and blood sampling was 5 wk, but 78% of the women were examined within an 8-wk period. Menopausal status or use of medications may have changed during that interval. This may have weakened true associations among premenopausal women.

Assays with high sensitivity were used to measure the hormone levels. However, total estradiol and DHEAS values were below the assay sensitivity level in 18 and 21% of postmenopausal women, respectively. Therefore, it was not

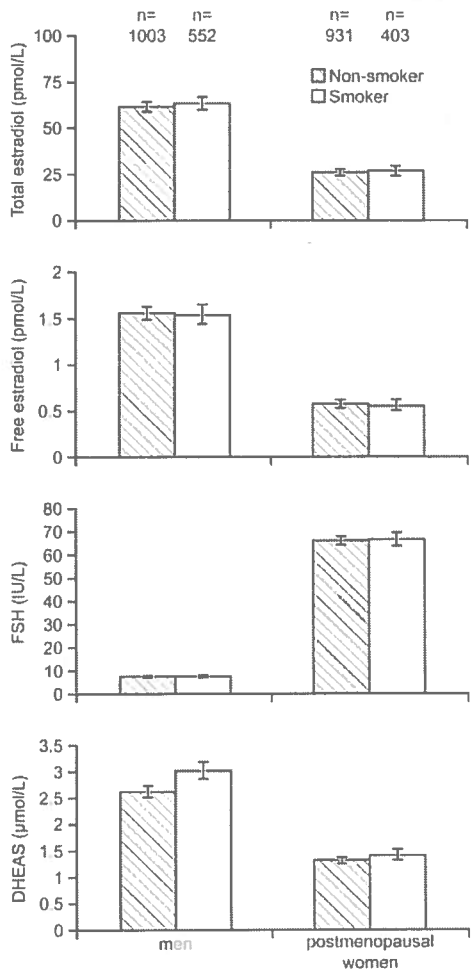

FIG. 3. Geometric means and 95% CI for the means of total and free estradiol, FSH, and DHEAS by smoking and sex adjusted for age, BMI, alcohol, coffee, and physical activity by ANCOVA among 1555 men and 1334 postmenopausal women not on hormone therapy affecting ovarian function: The Tromsø Study 1994–1995.

possible to obtain normal distribution of these hormones in this group. Blood samples taken between 0800 and 1600 h were used for the hormone measurements. Moderate diurnal variations of estradiol, FSH, and DHEAS are reported in previous mostly small studies (38, 39). In agreement with Verkasalo *et al.* (5), we did not find any systematic daytime variation in any hormone among either men or pre- and postmenopausal women. Adjustment for sampling hour did not change any result. Serum samples were frozen at −70 C for approximately 6.5 yr, and the hormone levels were measured when the samples were thawed for the first time. Levels of steroid hormones have been shown to be relatively stable in frozen plasma stored for 3–10 yr (15, 40). The delayed analyses are therefore not likely to represent a major problem. Free estradiol was calculated according to Vermeulen *et al.* (33), recently evaluated by Rinaldi *et al.* (41), and found to be a simple and reliable index of free estradiol.

No previous population-based study has reported increasing estradiol with increasing age among men. Recent large population-based studies describe levels of total estradiol unchanged (19, 21) or decreased with age (17, 18) among men. Although the reason for this discrepancy may be cohort effects and different selection criteria, our study contributes to continued uncertainty on the issue.

Among men, most of the circulating estradiol is derived from peripheral aromatization of the circulating precursor testosterone (20). The lower levels of testosterone with increasing age (19, 42) do not explain the increased levels of estradiol. A possible explanation might be increased levels of aromatase enzymes with age and the age-associated increase in fat tissue even without weight gain (20, 43, 44). Free estradiol increased to a lesser extent than total estradiol, which is explained by the well-known increase in SHBG with age (19, 42).

Whether FSH levels among postmenopausal women decrease or remain stable with increasing age remains controversial. Previous studies report a decrease in FSH (11, 12) or a stable high level of FSH (13) but no increase with age in this group. However, most of these studies are small (n = 32–60) except a population-based study by Kwekkeboom *et al.* (12), who did not account for the use of HRT that is known to lower the level of FSH (14). No previous study to our knowledge has reported increasing FSH with increasing age among postmenopausal women. Despite small changes in the absolute levels of FSH, we found that the FSH levels do not fall with age among postmenopausal women as thought previously (45). A possible explanation is decreased negative feedback on the pituitary gland due to lower levels of estradiol in this group of women. Among men, the levels of FSH at age older than 70 yr were higher than in those younger than 40 yr, but the age-related difference in the levels was less in comparison with women, as previously reported (10, 22).

The lower levels of DHEAS associated with increasing age of both sexes and higher DHEAS levels in men than in women confirm previously well-documented results (15, 26).

Our findings also confirm the well-known associations between BMI and the hormones as reported previously (12, 21). The higher levels of free estradiol but not total estradiol with increasing BMI in men is explained by the decrease of SHBG with increasing BMI (21, 42). In addition, the larger increase of free estradiol than total estradiol levels with increasing BMI among postmenopausal women is also explained by this same fall in SHBG (5, 25).

Lower DHEAS levels among men with IHD, compared with healthy men, have been reported in cross-sectional studies (9), although results from longitudinal studies are ambiguous (7, 8). The discrepancy in findings among studies may reflect the different end points used (9). DHEAS may be associated with IHD morbidity but not with cardiovascular mortality, or a more complex relationship between DHEAS and other biological processes may exist. However, among women DHEAS was not associated with IHD or other chronic diseases. This is in agreement with previous reports

(7, 46). As previously described, smokers had higher levels of DHEAS (7, 26). This intriguing finding of higher levels of DHEAS among smokers and lower levels of DHEAS among men with IHD has so far not been explained by any biological mechanism. The clinical significance of DHEAS in cardiovascular disease remains uncertain. Further prospective studies are needed for better understanding of the biological effects of DHEAS both in men and women.

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Seasonal Variation of Estradiol, Follicle Stimulating Hormone, and Dehydroepiandrosterone Sulfate in Women and Men

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Context: Seasonal variation in daylight regulates reproduction in animals living at higher latitude, but the influence of season on the sex hormones in humans remains unclear.

Objective, Design, and Participants: A cross-sectional population-based study in Tromsø, Norway (70° N) included 1651 women and 1540 men aged 25–84 yr. Circulating total estradiol (and calculated free levels), FSH, and dehydroepiandrosterone sulfate (DHEAS) were measured between September 1994 and September 1995 and provided a unique opportunity to study effects of extreme seasonal variations in the daylight on hormone levels in an arctic population.

Main Outcome Measure: Circulating total and free estradiol, FSH, and DHEAS were measured.

Results: Total and free estradiol showed differences between monthly means, with peak in June in postmenopausal women ($P < 0.001$), and in May in men ($P = 0.002$ and $P < 0.001$) by analysis of covariance. By cosinor analysis, a seasonal variation in total and free estradiol was evident in women ($P = 0.02$ and $P = 0.03$) and men ($P = 0.004$ and $P = 0.001$), but only 0.2–0.9% of the variation in total and free estradiol was explained by season. FSH and DHEAS showed no obvious seasonal variation in either sex.

Conclusions: Seasonal variations should be considered while designing studies and interpreting results of estradiol measurements to avoid bias in comparative studies. (***J Clin Endocrinol Metab* 91: 3798–3802, 2006**)

SEX HORMONES ARE involved in the pathophysiology of common diseases such as fragility fractures and cancer (1, 2), which makes it important to understand factors influencing their levels. All hormones are characterized by some rhythmic secretion and the levels can vary through the year. Seasonal variation in daylight regulates reproduction in animals living at higher latitude (3, 4), whereas the influence of season on the sex hormones in humans remains unclear (5–8).

If sex hormones vary systematically by season in some regions or for some subjects, this can cause misclassification bias in comparative studies. However, most studies involving hormone measurements do not take into account the time of measurement while designing studies or interpreting the results (9). Little information is available on seasonal variations in estradiol, FSH, and dehydroepiandrosterone sulfate (DHEAS) in both sexes. Despite having a longitudinal design, which lends them certain strength, previous studies are either small (n = 10–27) or based on selected hospital population, and the results are inconsistent (10–17). To our knowledge, seasonal variations in circulating estradiol, FSH, or DHEAS have not been investigated in a population-based study.

Tromsø, Norway, is located at 70° N and has extreme variations in the daylight exposure. The sun is below the horizon from November 28 to January 15 and does not set between May 17 and July 26. This study provided, therefore, a unique opportunity to test the hypothesis that extreme seasonal variations in the daylight affects the levels of sex hormones in women and men living at high latitude.

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Abbreviations: ANCOVA, Analysis of covariance; DHEAS, dehydroepiandrosterone sulfate; TROST, Tromsø Osteoporosis Study.

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Subjects and Methods

Subjects

The Tromsø Osteoporosis Study (TROST) is part of the Tromsø Study, a single-center, population-based, prospective study of Tromsø in northern Norway. Between September 1994 and September 1995, TROST measured bone density in 7948 subjects aged 25–84 yr (response rate 78%) (18). Among them, a random sample of 3684 were selected for hormone assays, and 3514 of them had blood samples available for analyses (19). Included in this sample were also participants in the Family Intervention Study, not viewed as representative of the general population (20). However, exclusion of these 155 surplus Family Intervention Study participants did not alter the results, so they were retained in the analyses. We excluded 323 participants due to use of hormone medication (n = 240), pregnancy (n = 4), perimenopausal status (n = 32), or missing hormone values (n = 47). All participants gave informed written consent. The regional Committee of Research Ethics and the Norwegian Data Inspectorate approved the study.

Two self-administered questionnaires were answered. We included information on current smoking status and consumption of coffee and alcohol. A physical activity score was made by adding the hours per week of moderate and hard physical activity, giving the hours with hard activity double weight: score = moderate + 2 hard. To maximize the number of observations, our definition of menopausal status was based on the self-reported menstruation data and age (19). Briefly, women who had a menstrual period within the last 3 months (n = 205) or were younger than 45 yr with missing data (n = 25) were defined as premenopausal. Women who stopped menstruating more than a year ago (n = 989) or were 54 yr of age or older with missing data (n = 432) were defined as postmenopausal. These 432 women had a mean age of 70 yr

(range 54–83), and 92% of them were 60 yr of age or older. This left 230 premenopausal and 1421 postmenopausal women and 1540 men to be included in the study. None were invited to participate in July due to summer vacation (Table 1).

Measurements

The Norwegian Meteorological Institute provided records of the monthly mean temperatures. The midmonth hours of daylight in Tromsø during the study period, were calculated as the hours between sunrise and sunset on the 15th day of each month (Fig. 1).

Height and weight were measured in light clothing without shoes, and body mass index was calculated as weight divided by the square of height (kilograms per square meter).

Nonfasting blood samples were taken between 0800 and 1600 h, and serum was stored at −70 C for 6–7 yr until first thawed in 2001. All hormones and SHBG were measured on Immulite 2000 (Diagnostic Products Corp., Los Angeles, CA). Estradiol and DHEAS measurements were based on competitive immunoassays, whereas FSH and SHBG measurements were based on immunometric assays.

The intra- and interassay coefficients of variation for estradiol and DHEAS were between 4 and 15%. The intra- and interassay coefficients of variation for FSH and SHBG were between 2 and 9%. The lower limits of detection were 10 pmol/liter for estradiol, 0.5 IU/liter for FSH, 1.0 μmol/liter for DHEAS, and 1.0 nmol/liter for SHBG. Samples with values below limits of detection were given a value midway between zero and limit of detection: estradiol (333 women, 60 men) and DHEAS (364 women, 84 men). All assays were run within a few weeks of each other using the same lot of reagents and assay kits. We used the method of Vermeulen *et al.* (21) to calculate free estradiol from total estradiol and SHBG. A recent validation by Rinaldi *et al.* (22) found that method simple and reliable.

Statistical analysis

The SAS Software package (version 8.2; SAS Institute, Cary, NC), was used for both data management and analysis. The data were analyzed separately for premenopausal and postmenopausal women, and for men. The significance level was chosen at $P < 0.05$ and P values are two sided. Because of skewed distribution, we used log-transformed hormones in all statistical analysis. However, the presented monthly means and confidence limits were transformed back to the original units.

Analysis of covariance (ANCOVA) was used to investigate the variation in monthly mean values and test for overall differences between the monthly means over the year. The months were used as a categorical explanatory variable. We adjusted for age, body mass index, current smoking (yes/no), alcohol use (yes/no), coffee drinking, and physical activity known to be associated with sex hormones (19). Because participants from the town center and the countryside met at different times of the year, we also adjusted for election districts to avoid interaction from socioeconomic status related to place of living.

The cosinor analyses were used to test seasonality in hormone values (23). We used individual values of log-transformed hormones and adjusted for all above-mentioned covariates. The essence of the method is to fit a linear regression model in which some smooth variation over time is modeled by patterns consisting of sines and cosines. This analysis provides estimates of the mean hormone value and the nonlinear parameters amplitude (distance from mean to peak of the curve) and acrophase (timing of the peak). Seasonality in hormone values was

TABLE 1. Numbers of participants by months of measurement: the Tromsø Study 1994–1995

	Premenopausal women	Postmenopausal women	Men
January	21	142	143
February	12	161	162
March	24	170	192
April	16	107	115
May	13	127	142
June	8	127	136
July			
August	18	148	151
September	78	101	88
October	10	152	169
November	14	118	137
December	16	68	105

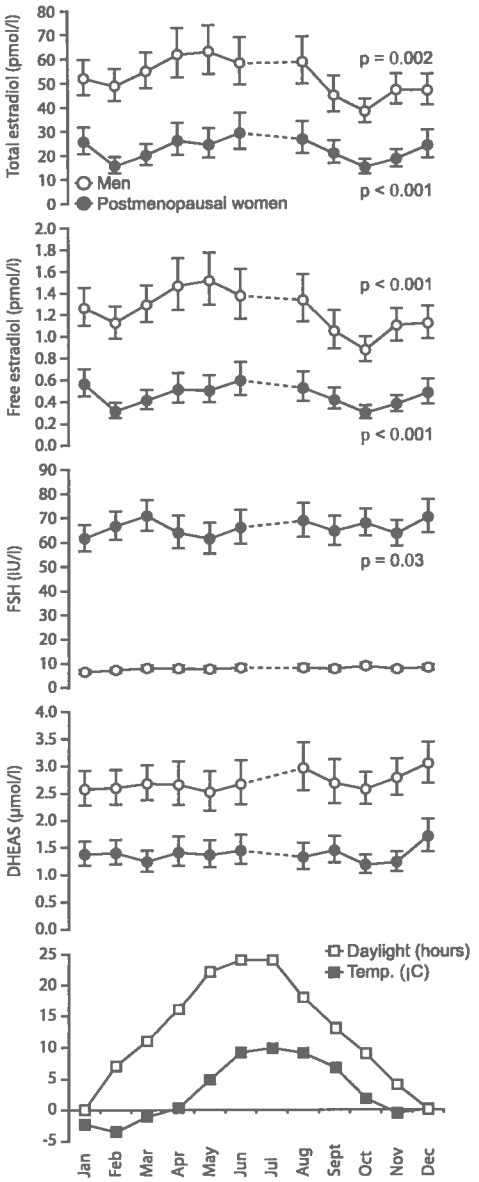

FIG. 1. Monthly geometric means with 95% confidence intervals of total and free estradiol, FSH, and DHEAS, adjusted for age, body mass index, smoking, alcohol and coffee drinking, physical activity, and election district by ANCOVAs in 1421 postmenopausal women and 1540 men. Monthly mean temperature and midmonth hours of daylight were used. The data are from the Tromsø Study 1994–1995.

evaluated by testing the null hypothesis of zero amplitude (24). The variation in hormones explained by seasonality was calculated from R^2 in cosinor analyses.

Results

In premenopausal women, total and free estradiol peaked in June (425 and 8.4 pmol/liter), the nadir month was November (76 and 1.6 pmol/liter), but there was no overall significance for differences between monthly means (P = 0.08) (data not shown). FSH and DHEAS showed no differences in monthly means in ANCOVA. In the cosinor analyses, total and free estradiol showed no seasonal variation. FSH showed a small peak in June (P = 0.04), and DHEAS showed seasonality (P = 0.004), with the peak in January (Tables 2 and 3).

In postmenopausal women, total and free estradiol showed differences between the monthly means ($P < 0.001$), with the peak in June (29.1 and 0.6 pmol/liter) and the nadir in October (14.8 and 0.3 pmol/liter), respectively, in ANCOVA (Fig. 1). FSH peaked in March (70.8 IU/liter), and the nadir occurred in May (61.3 IU/liter), but the variation by months was small (P = 0.03). In the cosinor analyses, no seasonal variation was detected when we included all postmenopausal women (Table 3). However, when we repeated the analyses after exclusion of the women 54 yr old or older with missing data on menopause (n = 432), the seasonal variation of total and free estradiol was significant (P = 0.02 and P = 0.04), and seasonality accounted for 0.2 and 0.3% of the variation in total and free estradiol (data not shown). FSH and DHEAS showed no detectable seasonality.

In men, total and free estradiol peaked in May (63.0 and 1.5 pmol/liter), and the nadir occurred October (38.0 and 0.9 pmol/liter), respectively, in ANCOVA (P = 0.002 and $P <$ 0.001) (Fig. 1). FSH and DHEAS showed no detectable differences in monthly means. In the cosinor analyses, total and free estradiol showed a highly significant seasonality, with the peak in May (P = 0.004 and P = 0.001), and seasonality accounted for 0.7 and 0.9% of the variation in total and free estradiol, respectively.

Discussion

The main finding from this study was a seasonal variation in estradiol in women and men, but only 0.2–0.9% of the variation in estradiol was explained by seasonality. Estradiol showed differences between monthly means in postmenopausal women and men. However, the cosinor analyses is a better way to test for seasonality, compared with a traditional ANOVA testing, in which any order of the months will give the same result.

Most studies on the variations in reproductive hormones by season are from northern Europe with participants younger than 50 yr (10–15). The estradiol levels have shown seasonality in studies on premenopausal women (10, 11). In this study, variability in the cyclic nature of hormonal measurements in premenopausal women and small numbers in most of the months may contribute to failure to detect significant variation in estradiol in this group. For the same reasons, we believe that the significant seasonality in DHEAS in this group may be a spurious finding. In this study, estradiol showed seasonality in postmenopausal women when we included only women who reported age at menopause. This weak association was blunted when we used a loose definition of menopause and included 432 women with missing data on menopause who were 54 yr of age or older. In a previous study on 14 elderly women, estradiol levels showed no seasonality (17). Otherwise, little is known about the seasonality of hormones among elderly women.

To our knowledge, the intriguing finding of coinciding seasonality in estradiol levels in men and postmenopausal women has not been reported previously. In agreement with results from a longitudinal study of 24 men from north Finland (13), in which the duration of daylight in summer and midwinter is comparable with Tromsø, estradiol levels peaked in May and had nadir levels in October. Others have reported no seasonality in estradiol among men (14) or women (17). The effects of seasonal variation in daylight may be blunted by the amount of artificial light in modern society. Although hormone levels peaked during the months of in-

TABLE 2. Characteristics of 230 premenopausal women, 1421 postmenopausal women, and 1540 men: the Tromsø Study 1994–1995

	Premenopausal women		Postmenopausal women		Men	
	Mean	SD	Mean	SD	Mean	SD
Age (yr)	38.6	7.1	64.1	6.4	60.1	10.0
Body mass index (kg/m²)	23.9	3.9	26.4	4.5	26.1	3.4
Coffee (no. cups/d)	5.0	3.3	4.9	2.8	6.0	3.8
Physical activity score[a]	4.2	2.6	2.7	2.1	3.7	2.5
Current smokers (%)	38.3		29.3		35.6	
Teetotallers (%)	8.3		32.7		12.8	
	Mean[b]	95% CI[c]	Mean[b]	95% CI[c]	Mean[b]	95% CI[c]
Hormones						
Total estradiol (pmol/liter)	172	23–1316	21.1	3.1–145	49.6	12.7–194
Free estradiol (pmol/liter)	3.6	0.5–25.1	0.4	0.1–3.1	1.2	0.3–4.6
FSH (IU/liter)	6.8	1.3–34.7	66.1	29.7–147	7.5	1.9–29.5
DHEAS (μmol/liter)	3.3	1.1–10.0	1.3	0.3–5.5	2.8	0.7–11.0

[a] A physical activity score was made by adding the hours per week of moderate and hard physical activity, giving the hours with hard activity double weight: score = moderate + 2 hard.
[b] Geometric mean.
[c] Confidence interval (CI) for the population (± 1.96 SD).

TABLE 3. The rhythm characteristics of log-transformed total and free estradiol, FSH, and DHEAS by cosinor analyses[a] in 230 premenopausal and 1421 postmenopausal women and 1540 men: the Tromsø Study 1994–1995

	Total estradiol (pmol/liter)	Free estradiol (pmol/liter)	FSH (IU/liter)	DHEAS (μmol/liter)
Premenopausal women				
P values[b]	0.14	0.26	0.04	0.004
Mean[c]	2.236	0.555	0.835	0.516
Amplitude[d]	0.135	0.110	0.080	0.068
Acrophase[e]	Mar 28	Mar 30	Jun 8	Jan 10
Postmenopausal women				
P values	0.09	0.21	0.60	0.89
Mean	1.325	−0.368	1.820	0.125
Amplitude	0.059	0.049	0.012	0.007
Acrophase	Mar 24	Mar 27		
Men				
P values	0.004	0.001	0.43	0.19
Mean	1.696	0.066	0.875	0.440
Amplitude	0.055	0.058	0.023	0.022
Acrophase	May 14	May 20		

[a] Adjusted for age, body mass index, smoking, alcohol and coffee drinking, physical activity, and election district.
[b] *P* value, the rhythm detection level.
[c] Mean, the annual mean level (log transformed).
[d] Amplitude is the extent of oscillation (log transformed).
[e] The acrophase is the time of peak value.

creased daylight (spring and early summer), the nadir levels were not found during the darkest season.

In animals living at higher latitudes, seasonal variation in daylight regulates reproductive function by altering the secretion of melatonin from the pineal gland (3, 4). Humans are nonseasonal breeders. Nevertheless, rhythmic seasonal variation in birth and conception rates are reported (25, 26). Although timing of human conception is mainly influenced by social factors, biological seasonal components still exist. Studies carried out at higher latitudes found higher levels of melatonin or longer duration of melatonin secretion during the dark season, compared with the light season (11–13, 27). This was not the case at lower latitudes (7, 8). In northern Finland, reproductive hormones have been shown to exhibit significant seasonal variation in men (13) and women (10, 11), with an increase in pituitary-gonadal function in late spring and early summer. However, these differences were small. Further evidence that biological factors contribute to increased fertility in the spring comes from studies of *in vitro* fertilization (28, 29). It is clear that the human pineal gland has retained the ability to respond to daylight duration, although the functional significance of this mechanism remains uncertain (7, 30). Many other factors than day length could lead to the small changes in estradiol levels, and one possible explanation is that temperature contributes. Whether seasonality in estradiol is an effect of daylight mediated by levels of melatonin could not be tested in this study because melatonin was not measured.

Epidemiological data indicate low risks for breast and prostate cancers in the arctic regions, and winter darkness and higher melatonin levels might have a protective effect against hormone-dependent tumors (6, 31). At higher latitudes, the duration of melatonin secretion has been reported to be longer in winter than summer but not at lower latitudes (7, 8). Melatonin could act as a naturally occurring antiestrogen as demonstrated on *in vivo* models of animal mammary tumors as well as *in vitro* human breast cancer cells (32–34). This melatonin hypothesis may explain the lower level of estradiol in the darkest season in the arctic regions. If estradiol levels are lower among the Norwegian population than people at lower latitudes, this may contribute in explaining the high incidence of fragility fractures in Norway (35). This hypothesis was supported by our findings of seasonality in estradiol levels in women and men in the population in Tromsø. We need comparative studies among people from different geographic locations to clarify this issue.

This study has some limitations. Although we used an assay with low limit of detection, estradiol values were below the limit in 23% of postmenopausal women. However, results did not differ substantially, whether or not women with values below the limit of detection were included. Delayed analyses of stored serum samples and daytime variation were previously discussed and results found unlikely to bias our results (19). Adjustment for sampling hour did not change any result. The characteristics of the subgroup with hormone measurements were compared with the total TROST population in a previous paper (19). The subgroup of women was 1 yr older with higher prevalence of cardiovascular diseases, whereas the men did not differ in any characteristics. The potential for selection bias is therefore assumed to be small. The cross-sectional estimates of seasonal variation of hormone levels are not true measures of longitudinal changes, and individual variations cannot be identified. A longitudinal design following up the same participants throughout the year would have been preferable. However, true seasonal variations would be expected to be evident in this study due to a large sample size, despite the cross-sectional design.

A weak but significant seasonal variation of estradiol, most of all in men, indicates that seasonality can be a source of bias in comparative studies in populations at higher latitudes. Possible seasonal variation should therefore be considered when designing studies and analyzing and interpreting results of estradiol measurements. Cases and controls should preferably have the hormones measured at

the same time of the year, and recording of blood sampling time to allow adjustment in the analyses to avoid bias. Further studies, preferably longitudinal, are needed to confirm our findings in populations at similar latitudes and test the validity of these results for other latitudes.

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Paper III

Circulating Estradiol and Sex Hormone-Binding Globulin Predict Bone Loss in Women and Men. The Tromsø Study

Abbreviated title: Sex steroids and bone loss

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Key words: bone mineral density; longitudinal studies; population-based; sex hormone-binding globulin; sex steroids

Abbreviations: BMD, bone mineral density; BMI, body mass index; CI, confidence interval; CV, coefficient of variation; DHEAS, dehydroepiandrosterone sulphate; SD, standard deviation; SHBG, sex hormone-binding globulin; TROST, Tromsø Osteoporosis Study

Abstract

INTRODUCTION: Bone loss during advancing age in women and men is partly the result of sex steroids deficiency. As the contribution of circulating sex steroids and sex hormone-binding globulin (SHBG) to bone loss remains uncertain, we sought to determine whether a single measurement of sex steroids or SHBG predicts bone loss in women and men.

METHODS: A population-based study in the city of Tromsø of 6.5 years duration (range 5.4 to 7.4) included 1089 women and 894 men aged 25-80 years. Total estradiol and testosterone, calculated free levels and SHBG were measured at baseline, and bone loss at the distal forearm was determined using bone mineral density (BMD) measurements in 1994-95 and 2001.

RESULTS: Bone loss was detected in pre- and postmenopausal women and in men. Age-adjusted free estradiol and SHBG predicted bone loss in postmenopausal women and in older men (> 60 years) ($p < 0.001 - p < 0.05$). After adjustment for body mass index (BMI) only free estradiol in postmenopausal women ($p < 0.05$) and only SHBG in older men ($p < 0.01$) persisted as significant independent predictors of bone loss. However, only 1-2% of the variance in bone loss was accounted for by these measurements.

CONCLUSION: Therefore, measurements of sex steroids are unlikely to assist in clinical decision making.

Introduction

Fragility fractures are a public health problem causing substantial morbidity, mortality and costs [1]. Reduced bone mineral density (BMD) is associated with bone fragility and is in part the result of bone loss produced by declining endogenous estradiol after menopause [2-4]. Men also lose bone as age advances and again, one underlying cause is held to be the decline in circulating estradiol as well as testosterone [5-12]. Although associations between bone loss and circulating estradiol are reported in both sexes [4, 6-13], these observations have not been consistently replicated in either sex [14-17].

One possible explanation for these inconsistencies is that other covariates such as body mass index (BMI) contribute to bone loss and these need to be taken into account [6, 11, 14]. Another factor is circulating sex hormone-binding globulin (SHBG). SHBG is associated with BMD [4, 10, 18], bone loss [14] and risk of fracture [19, 20]. However, few data are available examining the contribution of circulating SHBG on true measures of bone loss in longitudinal studies [4, 14]. We studied 1089 women and 894 men during 6.5 years to test the hypothesis that bone loss in women and men is associated with circulating sex steroids, SHBG, or both.

Materials and methods

Subjects

The Tromsø Osteoporosis Study (TROST) as part of the Tromsø Study, measured bone density in 7,948 subjects (response rate 78%) [5], and sex steroids in a random subgroup of 3564 subjects aged 25-84 years at baseline in 1994-95 (Fig. 1) [21, 22]. All 3080 still living in Tromsø, were invited to a repeated BMD measurement in 2001 and 2425 (79%) of them attended, so all participants had two BMD measurements. We excluded 442 participants due to invalid scans (n = 61), use of hormone medication (n = 184), or pregnancy (n = 2),

perimenopausal status (n = 19), outlying hormone values (n = 33), missing hormone values (n = 25), and also participants in the Family Intervention Study not representative of the general population (n = 118) [23]. In 1994-95, two self-administered questionnaires were filled in. Our definition of menopausal status were based on self-reported menstruation data, and age [21]. All participants gave informed written consent. The regional Committee of Research Ethics and the Norwegian Data Inspectorate approved the study.

Measurements

Bone density was measured on the non-dominant forearm, at distal and ultradistal sites, with Single X-ray Absorptiometric (SXA)-devices (DTX-100 Osteometer Medi Tech, Inc., Hawthorne, California). Only measurements from the distal site are presented in this study, as the ultradistal measurements followed the same pattern. The coefficients of variation (CV) was 0.8%, and details of the measurement methods and the strict quality control procedures for densitometry are previously published [5, 24-26]. Briefly all scans were reviewed and reanalysed and only scans free of artefacts were included. The long-term performance of the densitometers was assessed by twice daily phantom measurements with an aluminium forearm phantom, as well as by weekly measurements with the anthropomorphic European forearm phantom. The mean follow-up time was 6.5 years (standard deviations (SD) 0.4, range 5.4 to 7.4). Mean annual BMD change was calculated as the difference between the two measurements, divided by each participant's follow-up time.

Nonfasting blood samples were taken between 0800 h and 1600 h and serum was stored at -70° C for 6-7 years, until first thawed in 2001. All steroids and SHBG were measured on Immulite 2000 (Diagnostic Products Corporation, Los Angeles, CA, USA). Estradiol, testosterone and dehydroepiandrosterone sulphate (DHEAS) measurements were based on competitive immunoassays, whereas SHBG measurements were based on immunometric

assays. The lower limits of detection were 10 pmol/l for estradiol, 0.1 nmol/l for testosterone, 1.0 μmol/l for DHEAS, and 1.0 nmol/l for SHBG. The intra- and inter-assay CV for estradiol and DHEAS were between 4-15%. The intra- and inter-assay CV for testosterone > 1nmol/l were 3.5 % and 5 %; while in the range 0.1 to 1.0 nmol/l, it was 12 % and 20 %, respectively. The intra- and inter-assay CV for SHBG were 3% and 7%. Samples with values below limits of detection were given a value midway between zero and limit of detection: estradiol (220 women and 24 men), testosterone (408 women) and DHEAS (241 women and 38 men). We used the method described by Vermeulen et al. to calculate free estradiol and free testosterone from total estradiol, total testosterone and SHBG levels [27].

Statistical analysis

The data were analyzed separately for premenopausal and postmenopausal women, and age-stratified in men because of an interaction between age and SHBG ($P = 0.09$). We used a two-sided t-test and χ^2 to test for differences in baseline characteristics and sex steroids between groups within same sex.

Multiple linear regression analyses were used to investigate the effect of baseline estradiol and testosterone (total and calculated free levels), DHEAS and SHBG on BMD in cross-sectional data, and change in BMD in follow-up data. Baseline BMD and change in BMD were used as dependent variables, with the sex steroids and SHBG as continuous independent variables. We included one hormone or SHBG in each of the presented models. In additional models, the effect of SHBG independent from sex steroids was tested by inclusion of the total levels of sex steroids. The free levels of sex steroids and SHBG were not included in the same models because SHBG is component of the calculation of the free levels of sex steroids. We controlled for age (years), BMI (kg/m^2) and current smoking (no/yes), known to be associated with sex steroids and BMD [5, 6, 21, 22] in the following models:

Annual BMD change = $\beta_0 + \beta_1$sex steroids/SHBG + $\beta_2$age + β_3BMI + $\beta_4$smoking

where β_0 is the intercept, β_1 - β_4 is the regression coefficients. In order to avoid the effect of regression to the means, we considered inclusion baseline BMD and mean BMD as covariate [28]. Each subjects mean BMD was calculated by (baseline BMD + follow-up BMD) / 2. Height, physical activity, use of alcohol, calcium and vitamin D supplementation, sampling hour, baseline BMD and mean BMD were excluded as covariates, because of unchanged results. Log-transformation corrected for skewed distribution of the sex steroids, but did not improve the goodness of fit, as R^2 changed modestly in both directions. The variance in bone loss accounted for by sex steroids or SHBG was estimated from R^2, before and after inclusion of each of these variables. To facilitate the comparison of the strength of associations between sex steroid levels and change in BMD, we calculated the standardized regression coefficients, which describe the change in BMD in SD, per SD change in sex steroid or SHBG. Significance level was chosen at P-values < 0.05. The SAS Software package, v8.2 (SAS Institute Inc., Cary, NC, USA), was used for data analyses.

Results

In premenopausal women, no detectable diminution across age was observed in BMD or serum total or free estradiol in the cross sectional data (data not shown). Serum total and free testosterone diminished while SHBG increased. During a mean of 6.5 years follow-up (range 5.4-7.4) change in BMD was observed, but no association with sex steroids or SHBG was detected (Table 1-4).

In postmenopausal women, baseline BMD was lower than in premenopausal women, as were circulating estradiol and testosterone but not SHBG (Table 1). Testosterone, DHEAS

and SHBG, but not estradiol were associated with baseline BMD ($p < 0.001 - p < 0.05$) (Table 3). During follow-up, age-adjusted free estradiol and SHBG were associated with bone loss (Table 2 and 4). However, after adjustment for BMI and smoking, only free estradiol was associated ($p < 0.05$) with bone loss and accounted for 1% ($R^2 = 0.006$) of the bone loss, whereas the model accounted for 2% ($R^2 = 0.017$) by the following equation:

$\Delta BMD = 0.00526 + 0.00067$ free estradiol $+ 0.00001age + 0.00005BMI - 0.00052smoking$

Although higher free estradiol levels were associated with less bone loss, there was a considerable variability at any hormone level (Fig. 2). The 215 postmenopausal women with undetectable estradiol, had a higher bone loss of 1.04% (95% CI 0.89-1.18) than 712 women with detectable levels who lost bone 0.78% (95% CI 0.70 - 0.86) ($p = 0.003$).

Men aged 25 to 60 years had detectable diminution in BMD across this age range, but no diminution in total testosterone, total or free estradiol (data not shown). Free testosterone did diminish while SHBG increased across age. Free testosterone was associated with baseline BMD ($p < 0.05$) (Table 3). During follow-up, bone loss occurred, but no associations with sex steroids or SHBG was detected (Table 2 and 4).

Men aged 61 to 78 years had lower baseline BMD and free testosterone, and higher total estradiol and SHBG than men below 60 years of age (Table 1). Free levels of estradiol and testosterone and SHBG were associated with baseline BMD ($p < 0.001 - p < 0.05$) (Table 3). During follow up, age-adjusted free estradiol and SHBG were associated with bone loss (Fig. 2, Table 2 and 4). However, after adjustment for BMI and smoking, only SHBG was detected associated ($p < 0.01$) and accounted for 2% ($R^2 = 0.018$) of the bone loss, whereas the model accounted for 9% ($R^2 = 0.092$) by the following equation:

Δ BMD $= 0.00461 - 0.00002SHBG - 0.00012age + 0.00006BMI - 0.00099smoking.$

Discussion

The main finding from this study was that age-adjusted free estradiol and SHBG was weakly associated with bone loss in women and in men. After adjustment for BMI, free estradiol was detected associated with bone loss in postmenopausal women, whereas SHBG was associated with bone loss in older men. However, only 1-2% of the variance in bone loss was explained by these measurements.

Sex steroids have important effects on the skeleton [3, 29-31]; bone loss accelerates after menopause and is prevented by using hormone replacement therapy [4, 13, 18]. A young estrogen receptor negative man had decreased BMD despite high androgen and estrogen levels [32]. While aromatase-deficient men had decreased BMD, high androgen but undetectable estradiol levels, and estrogen treatment increased the BMD [33]. Despite these clear and reproducible associations, we were unable to confirm a strong statistical relation between circulating estradiol and bone loss in women or in men.

In men, estradiol are reported to decline with age (particular free and bioavailable levels) [7, 34], or to be stable [35, 36], while we reported rising estradiol with age [21]. Orwoll et al. reported declining free estradiol in univariate analysis, but no significant change in free estradiol by age in the multivariable analysis [37].

Although associations between BMD and circulating sex steroids are reported in both sexes [6-12], these associations have not been confirmed in other studies [14-17]. If a true association exists between sex steroids and bone loss, many factors may obscure this association such as variability in the assay method [6-14], fluctuations in hormone levels due to the cyclical nature of estrogen synthesis in premenopausal women, small changes in BMD during the observation period, weak relationship between estrogen and bone remodelling [38], secular trends in bone mass in cross sectional studies and small sample sizes in prospective studies [8, 12, 14, 39]. Whether covariates are controlled for such as BMI and SHBG, known

to be associated with both BMD and sex steroids, may contribute to the disparate results [6, 8, 10, 11]. In men above 60 years of age, free estradiol was associated with bone loss before but not after adjustment for BMI. This contrasts with the work of Khosla et al. [7, 8]. The reasons for the disparate results are not apparent but several of the above mentioned issues may contribute. Although estradiol loses significance after adjustment for BMI, this does not mean that estradiol is unimportant. Part of the effect of BMI may be mediated through sex steroids.

In addition, local rather than circulating sex steroids regulate bone remodelling. The main source of estradiol in postmenopausal women and men is peripheral conversion of circulating androgens in adipose tissue. Bone possesses aromatase activity [40, 41] and local aromatization appears to be a major source of estrogen responsible for mineralization [6, 10, 11, 39-41]. Bone loss is reported associated with the human gene encoding aromatase (CYP19), independently from circulating bioavailable estradiol [39], and aromatase-inhibitors accelerate bone loss and increase fracture incidence [42]. Circulating estradiol levels may not be reflecting the tissue levels of estradiol [39, 40]. Moreover, estrogen production at the extragonadal sites is dependent on circulating androgen precursors, including testosterone [40]. In the presence of low postmenopausal estradiol levels, testosterone and DHEAS could be important for bone health in women. In men, higher serum levels of testosterone may protect against bone loss [40].

SHBG was associated with bone loss in both sexes, but the associations were weak [4, 10]. Contrary to previous reports, only age-adjusted SHBG was associated with bone loss in postmenopausal women [14, 18]. The effect of SHBG can thus be mediated by BMI, known to be associated with both SHBG and bone loss [4, 10, 11, 43]. The previous findings of positive association between SHBG and fractures, and the positive association between SHBG and markers of bone turnover add strength to the hypothesis that rising SHBG levels

may participate in the pathogenesis of bone loss [16, 18-20, 44]. The association between SHBG and bone loss may be an indirect measure of the effects of sex steroids, because changes in SHBG result in changes in free fractions of sex steroids, and SHBG are to a lesser extent hampered with measurement errors [4]. The independent effect of SHBG after adjustment for sex steroids suggests other influences of SHBG on bone. SHBG is more than a transport protein. It may exert direct cellular functions by acting through specific membrane receptors [45]. There is no such evidence for bone, but an anti-estrogenic effect of SHBG is reported in breast cancer cells.

This study was population-based and had prolonged follow-up. There were several limitations. There was 21% attrition of subjects, but their characteristics differed little from responders. Non-responders were one year older with higher rates of cardiovascular diseases, but otherwise they did not differ in baseline BMI or levels of sex steroids. The phase of the menstrual cycle was not recorded at the time of sampling so variability in the cyclic nature of hormonal measurements in premenopausal women may have obscured associations with bone loss. Bone loss at the forearm may not reflect perfectly the pattern of bone loss at other sites. Although assays with low limits of detection were used, estradiol values were below the limit of detection in 23% of postmenopausal women. However, excluding the postmenopausal women with estradiol values below the limit of detection did not change any results. The measurement uncertainty could weaken true associations, but we believe that the large sample size in this study level out some of this measurement uncertainty, and reduce the threat on the results validity. Variations in the assays used could also explain some of the variability in the findings in the literature.

Under the assumption that there is a true association between circulating sex steroids and bone loss, most of the studies suggest that these associations are weak and so the explained variance in bone loss is small [10, 11]. Given these limitations, we suggest that

measurements of sex steroids are unlikely to assist in decision making regarding fracture risk susceptibility. Whatever the result, the explained variance in bone loss is small and will not provide additional information over that obtained from the BMD measurement. The test of this will require fracture endpoints and demonstration that the combination of BMD and circulating sex steroid measurements is a better predictor of fracture than BMD alone.

Acknowledgements

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Fig. 1. Participants of The Tromsø Osteoporosis Study (TROST) 1994-95 and 2001.

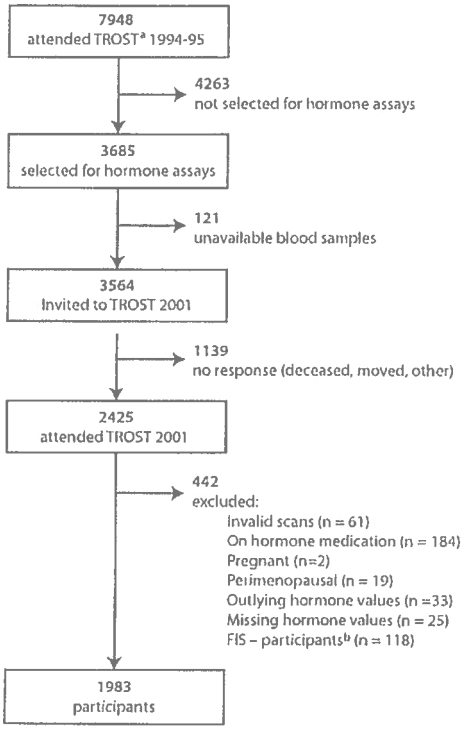

Fig. 2. Unadjusted annual change in distal forearm bone mineral density (BMD) (mg/cm²) by baseline free estradiol (pmol/l) and SHBG (nmol/l) in postmenopausal women (A) and (B), and in men > 60 years of age (C) and (D). The Tromsø Study 1994-95 and 2001.

Footnote: BMD change = $\beta_0 + \beta_1$free estradiol, and BMD change = $\beta_0 - \beta_1$SHBG, where β_0 is the intercept, β_1 is the regression coefficients, and note that the unit of BMD change is in mg/cm²; SHBG, sex hormone-binding globulin.

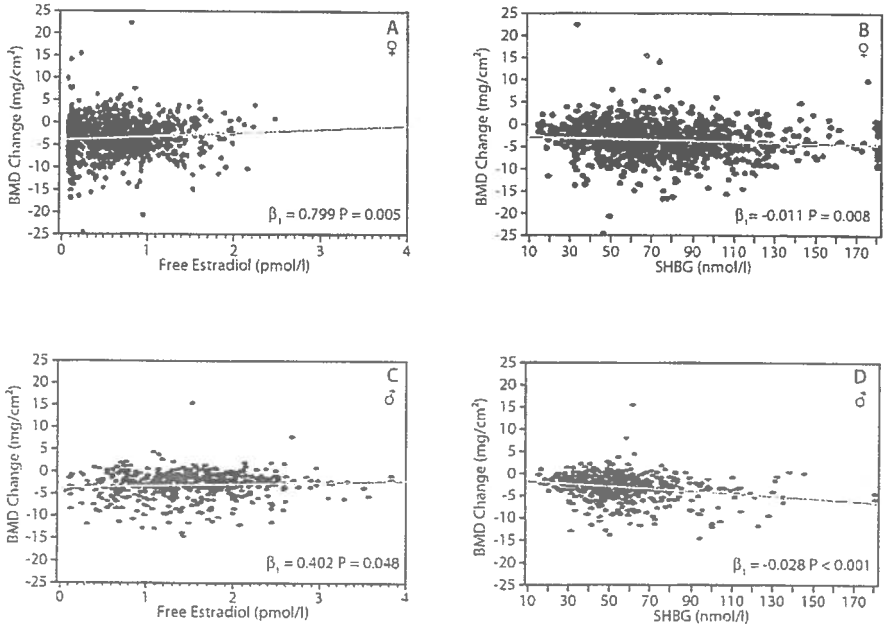

Table 1 Baseline characteristics and sex steroids in 1089 women and 894 men. The Tromsø Study 1994-95.

	Premenopausal (n = 162)		Postmenopausal (n = 927)		Men ≤ 60 yrs (n = 407)		Men >60 yrs (n = 487)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Age (years)	39.5	6.9	63.6	6.2	54.0	7.6	66.7	4.1
Height (cm)	164.6	7.0	161.4***	6.0	176.3	6.6	174.7***	6.6
Weight (kg)	65.8	11.8	68.6**	11.4	82.3	11.3	78.8***	11.5
Body mass index (kg/m²)	24.3	4.1	26.3***	4.3	26.4	3.0	25.8**	3.4
Forearm BMD[a] (g/cm²)	0.469	0.047	0.386***	0.065	0.557	0.058	0.527***	0.073
Current smokers (%)	36.4		26.0**		33.4		28.1	
Total estradiol (pmol/l)	290	315	28.7***	20.6	57.3	34.7	64.4**	29.7
Free estradiol (pmol/l)	5.7	5.5	0.6***	0.5	1.4	0.8	1.5	0.7
Total testosterone (nmol/l)	0.9	0.9	0.5***	0.6	13.6	5.3	13.0	5.0
Free testosterone (pmol/l)	11.1	12.8	5.9***	8.6	224	76	186***	60
DHEAS[b] (µmol/l)	3.7	1.9	1.7***	1.2	4.1	2.2	2.7***	1.7
SHBG[c] (nmol/l)	70.8	31.5	74.2	32.8	47.5	21.0	56.5***	23.7

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$, for differences between groups within same sex by t-test and χ^2.
[a]BMD, bone mineral density at the distal forearm.
[b]DHEAS, dehydroepiandrosterone sulphate.
[c]SHBG, sex hormone-binding globulin.

Table 2 Mean annual change in forearm bone mineral density (BMD) and 95% confidence interval (±1.96 SE) in1089 women and 894 men. The Tromsø Study 1994-95 and 2001.

	Annual change in distal forearm BMD		
Women	Mean (mg/cm²)	95% CI (mg/cm²)	Mean (%)
Premenopausal (n = 162)	-0.8	-1.2 to -0.3	-0.17
Postmenopausal (n = 927)	-3.3***	-3.5 to -3.0	-0.84
Men			
≤ 60 years (n = 407)	-1.7	-1.9 to -1.5	-0.31
> 60 years (n = 487)	-3.0***	-3.3 to -2.8	-0.59

*** P < 0.001, for differences between groups within same sex by t-test.

Table 3 Standardized regression coefficients for the forearm bone mineral density (BMD) in multiple linear regression analyses by sex steroids[a] and SHBG[b] in 1089 women and 894 men. The Tromsø Study 1994-95.

Baseline study	Distal forearm BMD (g/cm²)	
Premenopausal women (n = 162)	Age-adjusted	Adjusted for age, BMI and smoking
Total estradiol (pmol/l)	-0.07	-0.06
Free estradiol (pmol/l)	-0.03	-0.04
Total testosterone (nmol/l)	0.09	0.05
Free testosterone (pmol/l)	0.14	0.07
DHEAS[c] (μmol/l)	0.14	0.09
SHBG (nmol/l)	-0.15	-0.07
Postmenopausal women (n = 927)		
Total estradiol (pmol/l)	-0.02	-0.05
Free estradiol (pmol/l)	0.05	-0.01
Total testosterone (nmol/l)	0.09**	0.07*
Free testosterone (pmol/l)	0.14***	0.10**
DHEAS (μmol/l)	0.09**	0.10***
SHBG (nmol/l)	-0.30***	-0.25***
Men ≤ 60 years (n = 407)		
Total estradiol (pmol/l)	0.04	0.04
Free estradiol (pmol/l)	0.05	0.03
Total testosterone (nmol/l)	-0.02	0.07
Free testosterone (pmol/l)	0.07	0.11*
DHEAS (μmol/l)	-0.10	-0.07
SHBG (nmol/l)	-0.11*	-0.04
Men > 60 years (n = 487)		
Total estradiol (pmol/l)	0.05	0.05
Free estradiol (pmol/l)	0.13**	0.09*
Total testosterone (nmol/l)	-0.11*	-0.02
Free testosterone (pmol/l)	0.10*	0.12**
DHEAS (μmol/l)	0.01	0.04
SHBG (nmol/l)	-0.26***	-0.17***

* P < 0.05; ** P < 0.01; *** P < 0.001.

[a]We included one hormone or SHBG in each of the presented models.

[b]SHBG, sex hormone-binding globulin.

[c]DHEAS, dehydroepiandrosterone sulphate.

Table 4 Standardized regression coefficients for change in bone mineral density (BMD) in multiple linear regression analyses by sex steroids[a] and SHBG[b] in1089 women and 894 men. The Tromsø Study 1994-95 to 2001.

Follow-up study	Change in distal forearm BMD (g/cm²)	
Premenopausal women (n = 162)	Age-adjusted	Adjusted for age, BMI and smoking
Total estradiol (pmol/l)	-0.03	-0.02
Free estradiol (pmol/l)	-0.02	-0.02
Total testosterone (nmol/l)	-0.01	-0.01
Free testosterone (pmol/l)	0.02	-0.00
DHEAS[c] (µmol/l)	-0.09	-0.09
SHBG (nmol/l)	-0.10	-0.08
Postmenopausal women (n = 927)		
Total estradiol (pmol/l)	0.07*	0.06
Free estradiol (pmol/l)	0.09**	0.08*
Total testosterone (nmol/l)	-0.01	-0.02
Free testosterone (pmol/l)	0.04	0.02
DHEAS (µmol/l)	-0.03	-0.02
SHBG (nmol/l)	-0.09**	-0.06
Men ≤ 60 years (n = 407)		
Total estradiol (pmol/l)	-0.04	-0.04
Free estradiol (pmol/l)	-0.00	-0.00
Total testosterone (nmol/l)	-0.08	-0.08
Free testosterone (pmol/l)	-0.03	-0.03
DHEAS (µmol/l)	0.02	0.04
SHBG (nmol/l)	-0.09	-0.09
Men > 60 years (n = 487)		
Total estradiol (pmol/l)	0.03	0.04
Free estradiol (pmol/l)	0.10*	0.08
Total testosterone (nmol/l)	-0.08	-0.01
Free testosterone (pmol/l)	0.06	0.08
DHEAS (µmol/l)	0.01	0.04
SHBG (nmol/l)	-0.20***	-0.15**

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

[a]We included one hormone or SHBG in each of the presented models.

[b]SHBG, sex hormone-binding globulin.

[c]DHEAS, dehydroepiandrosterone sulphate.

Paper IV

Acta Eur J Endocr.

A Prospective Study of Sex Steroids, Sex Hormone-Binding Globulin and Non-Vertebral Fractures in Women and Men in the Tromsø Study

Abbreviated title: Sex steroids and fractures

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Key words Fractures; men; prospective study; sex steroids; sex hormone-binding globulin; women

Abbreviations ANCOVA, analysis of covariance; BMD, bone mineral density; BMI, body mass index; CI, confidence interval; CV, coefficient of variation; DHEAS, dehydroepiandrosterone sulphate; NS, non-significant; SD, standard deviation; SHBG, sex hormone-binding globulin; TROST, Tromsø Osteoporosis Study

Abstract

INTRODUCTION: As bone fragility is partly the result of sex hormone dependent bone loss, we sought to determine whether a single measurement of circulating sex steroids or sex hormone-binding globulin (SHBG), a determinant of the free hormonal fraction, predict non-vertebral fractures in women and men.

METHODS: Forearm bone mineral density (BMD), total estradiol and testosterone, calculated free levels and SHBG were measured in 1386 postmenopausal women and 1364 men aged 50 to 84 years at baseline in the Tromsø Study 1994-95. Non-vertebral fractures were documented between January 1994 and February 2005.

RESULTS: During 8.4 years (range 0.01-10.4) and 23,034 person-years follow-up, 281 (20.3%) women and 105 (7.7%) men suffered non-vertebral fractures. For both sexes, fracture cases had lower BMD, higher SHBG, but sex steroids were no lower than participants remaining fracture free. Each standard deviation (SD) increase in SHBG increased non-vertebral fracture risk by about 20% in women (HR 1.17; 95% CI 1.03-1.33) and men (HR 1.27; 95% CI 1.03-1.55). This effect was mediated by BMD in both sexes. Each SD decrease in BMD increased fracture risk by about 40% in women (HR 1.36; 95% CI 1.19-1.56) and men (HR 1.41; 95% CI 1.15-1.73). Fracture risk was highest in participants with the combination of SHBG in the highest tertile and BMD in the lowest tertile in women (HR 3.76; 95% CI 2.02-7.01) and in men (HR 2.98; 95% CI 1.41-6.30) compared with participants who had SHBG in the lowest tertile and BMD in the highest tertile. However, the fracture risk prediction of this combination was only slightly better than BMD alone in both sexes. No association between sex steroids and fracture risk was detected.

CONCLUSION: Measurements of sex steroids or SHBG are unlikely to assist in decision making regarding fracture risk susceptibility.

Introduction

Women and men who sustain fragility fractures do so partly because of low bone mineral density (BMD), which is the result of a reduced peak BMD, bone loss or both [1, 2]. Bone loss occurs before menopause but is accelerated after menopause and is the result of sex hormone deficiency [3-6]. Consequently, a low level of circulating sex steroids may be a sensitive and specific predictor of fractures and so may be useful in signalling the need for further investigation or treatment.

Some investigators have reported lower sex steroids and higher sex hormone-binding globulin (SHBG) associated with bone loss [5-14], and increased fracture risk but this is not consistently reported [15-25], perhaps because many factors influence fracture risk, particularly falls [26, 27]. However, few prospective data are available examining the contribution of sex steroids and SHBG on fracture risk [15, 18, 20, 22, 24]. In the companion paper we reported that estradiol and SHBG predicted bone loss in postmenopausal women and elderly men, but the associations were weak in both [28].

If sex steroids or SHBG (a determinant of the free hormonal fraction) predicts fractures, the combination of BMD and sex steroids/SHBG measurements may improve prediction. We therefore examined the independent and combined contribution of sex steroids/SHBG and BMD to fracture risk in both sexes. We tested the hypothesis that incident non-vertebral fractures in women and men are predicted by circulating levels of sex steroids or SHBG in a prospective study of 1386 women and 1364 men during 23,034 person-years follow-up.

Materials and methods

Subjects

The Tromsø Osteoporosis Study (TROST), as part of the Tromsø Study, involved measurement of bone density in 6981 subjects aged 50 to 84 years (response rate 80%) [29],

and sex steroid levels in a random subgroup of 3017 subjects at baseline in 1994-95 [30, 31]. We excluded 267 participants due to hormone medication (n = 185), or pre- or perimenopausal status (n = 48), or outlying hormone values not believed to be true measurements (n = 34), and 2750 participants are thus included in this study. The participants with invalid scans (n = 45) were excluded from the analyses including BMD. All participants gave informed written consent. The regional Committee of Research Ethics and the Norwegian Data Inspectorate approved the study.

Two self-administered questionnaires were filled in. A physical activity score was made by adding the hours/week of moderate and hard physical activity, giving the hours with hard activity double weight: score = moderate + 2hard. Our definition of postmenopausal status were based on self-reported menopause (n = 955), and age $\geq$ 54 years if data was missing (n = 431) [30]. Exclusion of the women with missing data did not change the results.

All non-vertebral fractures were registered from the x-ray archives of the University Hospital in Tromsø between 1 January 1994 and 12 February 2005. All fractures are registered here, as this is the only x-ray service in the city or within 250 km. The only exception would be fractures occurring while traveling with no control x-ray after returning home. The validation of the fracture registration is previously reported [32]. Follow-up time was assigned from baseline to the first fracture, to death (n = 506), when the participant moved (n = 177), or to the end of follow-up. The mean follow-up time was 8.4 years (range 0.01-10.4), and person-years were 23,034.

Measurements

Height and weight were measured in light clothing without shoes, and body mass index (BMI) was calculated as weight divided by the square of height (kg/m^2). Bone density was measured on the non-dominant distal forearm, with Single X-ray Absorptiometric (SXA)-devices

(DTX-100 Osteometer Medi Tech, Inc., Hawthorne, California). The coefficients of variation (CV) was 0.8%, and details of the measurement methods and the strict quality control procedures for densitometry are previously published [29, 33].

Nonfasting blood samples were taken between 0800 h and 1600 h and serum was stored at -70° C for 6-7 years, until first thawed in 2001. All steroids and SHBG were measured on Immulite 2000 (Diagnostic Products Corporation, Los Angeles, CA, USA). Estradiol, testosterone and dehydroepiandrosterone sulphate (DHEAS) measurements were based on competitive immunoassays, whereas SHBG measurements were based on immunometric assays. The lower limits of detection were 10 pmol/l for estradiol, 0.1 nmol/l for testosterone, 1.0 μmol/l for DHEAS, and 1.0 nmol/l for SHBG. The intra- and inter-assay CV for estradiol and DHEAS were between 4-15%. The intra- and inter-assay CV for testosterone > 1nmol/l were 3.5 % and 5 %; while in the range 0.1 to 1.0 nmol/l, it was 12 % and 20 %, respectively. The intra- and inter-assay CV for SHBG were 3% and 7%. Samples with values below limits of detection were given a value midway between zero and limit of detection. We used the method described by Vermeulen et al. to calculate free estradiol and free testosterone from total estradiol, total testosterone and SHBG levels [30, 34].

Statistical analysis

Women and men were analyzed separately. We used a two-sided t test, χ^2 test and analyses of covariance (ANCOVA) to test for differences in means in baseline characteristics and sex steroids by fracture status. Cox proportional hazard models were used to determine whether baseline total and free estradiol, total and free testosterone, dehydroepiandrosterone sulphate (DHEAS) and SHBG predict fractures. We included one hormone or SHBG in each of the presented models. In additional models, the effect of SHBG independent from sex steroids was tested by inclusion of total levels of sex steroids. The free levels of sex steroids and

SHBG were not included in the same models because SHBG is component of the calculation of free levels of sex steroids. Log-transformation corrected for skewed distribution of the sex steroids, but did not change any results.

We controlled for age (years), BMI (kg/m^2), height (cm), current smoking (yes/no), physical activity score and BMD (g/cm^2), known to be associated with sex steroids and fracture risk [30, 31, 35, 36]. A history of previous fractures, chronic diseases, alcohol consumption and supplementation of calcium were excluded as covariates, because of unchanged results. Exclusion of participants with high-energy trauma fractures (n = 55) did not change the results, so all fractures were included. We tested interaction terms between age, BMI, BMD, sex steroids and SHBG in the models, and none was significant. The proportionality assumptions of the models were verified. The SAS Software package, v9 (SAS Institute Inc., Cary, NC, USA) was used, and significance level was chosen at $P < 0.05$.

Results

During a mean follow up of 23,034 person-years, 281 (20.3%) of 1386 postmenopausal women and 105 (7.7%) of 1364 men suffered incident non-vertebral fractures (Table 1). Women with fractures were older, taller and had lower BMI than women without fractures. For both sexes, fracture cases had lower BMD, higher SHBG, but circulating levels of sex steroids were no lower (Table 1 and 2).

In women, no significant association between sex steroids and risk of non-vertebral fractures was detected (Table 3). Women with undetectable levels of estradiol (total estradiol < 10 pmol/l, n = 327) had no higher risk of fractures than women with detectable levels, HR 0.99 (95% CI 0.75-1.31). Each SD increase in SHBG increased the risk of non-vertebral fractures by about 20% (HR 1.17; 95% CI 1.03-1.33, P = 0.02) after adjustment for age, BMI,

height, smoking and physical activity (Table 3). After further adjustment for BMD, the increased risk was blunted and no longer statistically significant (HR 1.09; 95% CI 0.95-1.24, P = 0.21). The age-adjusted risk of wrist, not hip fractures increased by each SD increase in SHBG (HR 1.25; 95% CI 1.06-1.47), but not after adjustment for BMI and other covariates (HR 1.18; 95% CI 0.98-1.42).

In men, there was a marginally increased risk of fractures by higher levels of testosterone, but no other association between sex steroids and fractures was detected (Table 3). Each SD increase in total testosterone increased the risk of non-vertebral fractures by about 20% after adjustment for age, BMI, height, smoking and physical activity (HR 1.21; 95% CI 1.00-1.47, P = 0.053), and after further adjustment for BMD (P = 0.056). Testosterone and SHBG correlated (r = 0.55), and testosterone (HR 1.14; 95% CI 0.91-1.43) and SHBG (HR 1.15; 95% CI 0.89-1.48) were not associated with fracture risk after adjustment for each other. Each SD increase in SHBG increased the risk of non-vertebral fractures by about 20% after adjustment for age, BMI, height, smoking and physical activity (HR 1.27; 95% CI 1.03-1.55, P = 0.02), and after further adjustment for BMD (HR 1.22; 95% CI 1.00-1.50, P = 0.056) (Table 3). The age-adjusted risk of hip, not wrist fractures increased for each SD increase in SHBG (HR 1.37; 95% CI 1.00-1.88), but not after adjustment for BMI and other covariates (HR 1.41; 95% CI 0.98-2.02).

In both sexes, baseline BMD was associated with risk of non-vertebral fractures before and after adjusting for age, BMI, height, smoking, physical activity and SHBG. Each SD decrease in BMD increased the risk for fracture by 40% in women (HR 1.36; 95% CI 1.19-1.56) and men (HR 1.41; 95% CI 1.15-1.73) (Table 3). Fracture risk was increased in participants with SHBG in the highest and BMD in the lowest tertile (Fig. 1). About 16% of the cohort had the combination of the two traits, with the highest HR 3.76 (95% CI 2.02-7.01)

in women, and HR 2.98 (95% CI 1.41-6.30) in men, compared with participants who had SHBG in the lowest tertile and BMD in the upper tertile (Fig. 2).

In women, the combination of low BMD and high SHBG measurement had HR 1.52 (95% CI 1.14-2.02), while low BMD alone had HR 1.38 (95% CI 1.08-1.77), compared with all other women. Similarly in men, the combination of the two traits had HR 2.32 (95% CI 1.46-3.68), while low BMD alone had HR 2.14 (95% CI 1.42-3.21), compared with all other men. There was no significant interaction between BMD and SHBG in women (P = 0.78) or in men (P = 0.53).

Discussion

The main finding from this population-based prospective study was that a single measurement of circulating sex steroids did not predict incident non-vertebral fractures in either sex. A higher SHBG and lower BMD predicted risk of fractures in both sexes, but the effect of SHBG was small and partly dependent of the association between BMD and fractures.

An association between circulating estradiol, bone loss or risk of vertebral and non-vertebral fractures is reported in some but not all studies in women or men [5-25, 38-41]. Circulating estradiol is reported both associated and not associated with prevalent vertebral fractures in women [16, 19], and in men [17, 19, 21, 25]. These results from retrospective data are difficult to interpret, as alteration in estradiol could have occurred after the fractures. Prospective studies including incident vertebral fractures, or combination of vertebral and non-vertebral fractures as end point have reported lower estradiol associated with fracture risks in women [15, 20, 22, 23]. Prospective studies including incident hip fractures have reported low estradiol associated with risk of fractures in women [15, 18] and in men [24], but the effect was dependent on weight in one study [18].

The lack of association between circulating sex steroids and risk of non-vertebral fractures in the present study may be the result of several factors. First, non-vertebral fractures are traumatic and the contribution of bone fragility to the fracture event may be relatively less than in vertebral fractures, as these generally involve minimal trauma. Second, peak bone mass is an important determinant of bone strength in old age and may make a more important contribution to bone fragility than estrogen deficiency [9, 42]. Third, bone loss occurs before menopause and is independent of sex steroids. Fourth, a single measurement of sex steroids has a large variance and so may not adequately reflect long term exposure to estrogen deficiency.

The marginally increased risk of fractures by higher testosterone was not present after adjustment for SHBG and may be due to confounding effect of SHBG. Both higher testosterone and higher SHBG are associated with lower weight, known to be associated with increased fracture risk [35].

In the present study, higher SHBG was associated with increased risk of incident non-vertebral fractures, but the risk explained by SHBG was small in both sexes and partly dependent on BMD. In women, higher SHBG is reported associated with increased risk of incident vertebral, hip and the combination of vertebral and non-vertebral fractures in prospective studies [15, 18, 20, 22]. In men, SHBG is associated with prevalent vertebral fractures and the combination of vertebral and non-vertebral fractures as endpoint [17, 21]. In men, the effect of SHBG on incident non-vertebral fractures has not been studied in prospective data. Most authors suggest that higher SHBG increase the risk of fractures by binding estradiol and decreasing its bioavailability [15, 20, 21]. The independent effect of SHBG on risk of fractures adjusted for estradiol in the present study is against this explanation. Some investigators report that subjects with both high SHBG and low estradiol had the highest risk of incident vertebral and hip fractures [15, 22]. Center et al. reported

SHBG, but not estradiol to be associated with prevalent fractures (vertebral and non-vertebral) in men, and the effect was independent of BMD [17].

An increased risk of fracture in participants with the combination of SHBG in the highest and BMD in the lowest tertile, was more than either trait alone. Although the fracture risk prediction by the combination of SHBG and BMD was only slightly better than BMD alone, the combination may identify a subgroup with high risk of fractures. This supports the notion of an independent effect of SHBG other than by affecting the BMD or bioavailability of estradiol. SHBG is more than a transport protein, and may act through specific membrane receptors [43]. An anti-estrogenic effect of SHBG is reported in breast cancer cells, but there is no evidence on direct effect of SHBG on bone.

This study has limitations and the true strengths of the relationships between sex steroids/SHBG and fracture risk may have been underestimated. Although assays with low limits of detection were used, estradiol values were below the limits of detection in 24% of postmenopausal women. However, excluding the women with values below the limit of detection did not change the results. The measurement uncertainty could weaken true associations, and the more precisely measured SHBG could therefore appear to be more strongly associated with the outcome. However, we believe that the large sample size in this study even out some of the measurement uncertainty related to low precision, and reduce the threat on the validity of the results. Variations in the assays used could also explain some of the variability in the findings in the literature.

Adjustment for daytime or seasonal fluctuations in estradiol did not change the results. Responders to study invitations are often more healthy than non-responders. However, previous comparison between responders and non-responders in TROST, and between the subgroup with hormone measurements and total TROST population, gave no indication of

differences between the groups [29, 30]. The potential for selection bias is therefore assumed to be small.

The lack of association between sex steroids and fractures and the weak association between SHBG and fractures make these traits unlikely to assist in decision making regarding fracture risk susceptibility.

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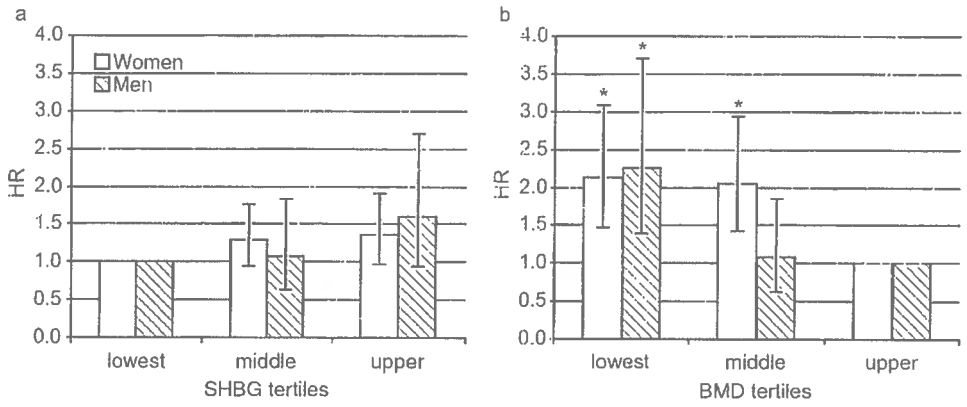
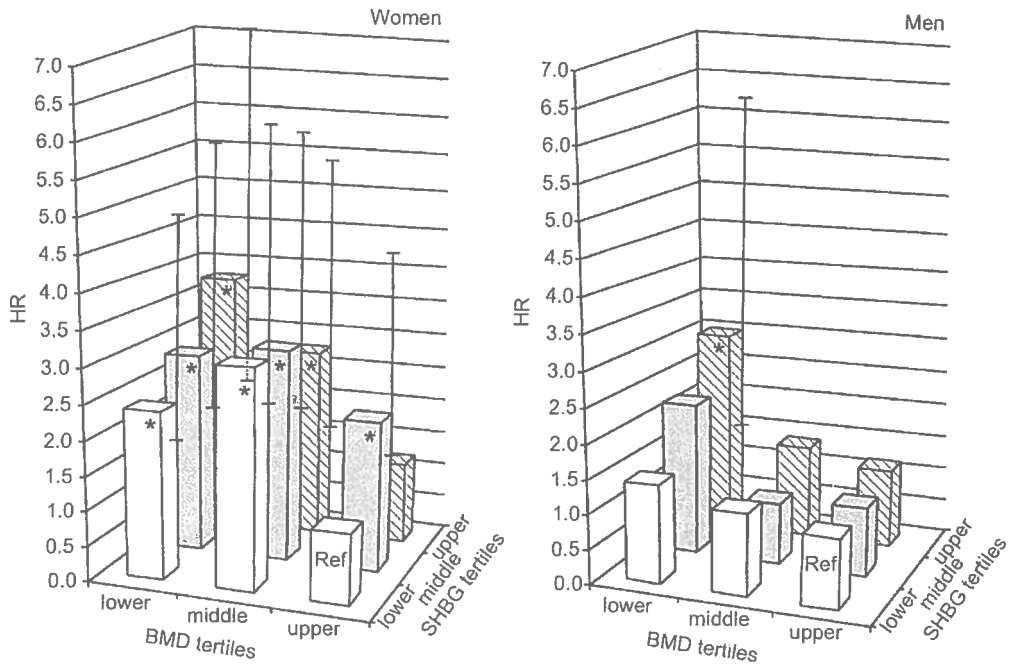
Fig. 1a and 1b.
The hazard ratios (HR) with 95% CI of non-vertebral fractures by tertiles of SHBG and BMD in women and men, adjusted for age, BMI, height, smoking and physical activity. The Tromsø Study 1994-95.



Footnote: The SHBG tertiles in women: < 57, 57-84 and > 84 nmol/l, in men: < 43, 43-58 and > 58 nmol/l. The BMD tertiles in women: < 0.362, 0.362 – 0.423, > 0.423 g/cm², in men: <0.509, 0.509-0.565 and > 0.565 g/cm². * P < 0.05.

Fig. 2.
The hazard ratios (HR) with 95% CI of non-vertebral fractures by combinations of tertiles of SHBG and BMD in women and men, adjusted for age, BMI, height, smoking and physical activity. The Tromsø Study 1994-95.



Footnote: The tertiles values are given in Fig. 1, CI including 1 is not presented. * $P < 0.05$.

Table 1
Baseline characteristics of participants by fracture status. The Tromsø Study 1994-95.

Women	No fractures n = 1105		Non-vertebral fractures n = 281	
	Mean	SD	Mean	SD
Age (years)	64.0	6.2	65.6***	6.2
Height (cm)	160.8	6.0	162.0**	6.2
Weight (kg)	68.7	12.4	67.8	11.5
Body mass index (kg/m²)	26.6	4.6	25.9*	4.3
Physical activity score[a]	2.7	2.1	2.6	2.1
Forearm BMD[b] (g/cm²)	0.387	0.066	0.360***	0.063
Current smokers (%)	30.0		28.8	
			n	%
All non-vertebral fractures			281	20.3
Hip fractures			42	3.0
Wrist fractures			128	9.2
Men	n = 1259		n = 105	
Age (years)	62.9	6.6	63.9	7.0
Height (cm)	174.7	6.9	175.8	6.2
Weight (kg)	79.8	12.2	80.6	11.3
Body mass index (kg/m²)	26.1	3.5	26.1	3.2
Physical activity score[a]	3.7	2.5	3.3	2.3
Forearm BMD[b] (g/cm²)	0.536	0.069	0.511***	0.070
Current smokers (%)	34.4		37.1	
			n	%
All non-vertebral fractures			105	7.7
Hip fractures			27	2.0
Wrist fractures			21	1.5

*P < 0.05; ** P < 0.01; *** P < 0.001, for between groups within same sex by t-test.
[a]A physical activity score was made by adding hours/week of moderate and hard physical activity; score = moderate + 2hard
[b]BMD, bone mineral density at the distal forearm.

Table 2
Geometric means with 95% CI and P-values for differences in means of sex steroids and SHBG[a] by fracture status by ANCOVA[b] in 1386 women and 1364 men. The Tromsø Study 1994-95.

Women	No fractures		Non-vertebral fractures		
	mean	95% CI	mean	95% CI	P-values
Total estradiol (pmol/l)	19.9	18.9-21.1	20.0	18.0-22.3	0.94
Free estradiol (pmol/l)	0.4	0.4-0.4	0.4	0.4-0.4	0.74
Total testosterone (nmol/l)	0.2	0.2-0.2	0.2	0.2-0.3	0.30
Free testosterone (pmol/l)	2.3	2.1-2.5	2.4	2.1-2.9	0.57
DHEAS[c] (μmol/l)	1.3	1.3-1.4	1.3	1.2-1.5	0.75
SHBG (nmol/l)	67.0	65.4-68.6	70.7	67.4-74.1	0.046
Men					
Total estradiol (pmol/l)	50.7	48.8-52.6	53.1	46.6-60.5	0.50
Free estradiol (pmol/l)	1.2	1.1-1.2	1.2	1.1-1.4	0.77
Total testosterone (nmol/l)	11.6	11.2-12.0	12.5	11.2-14.1	0.18
Free testosterone (pmol/l)	175	169-181	182	162-205	0.52
DHEAS (μmol/l)	2.5	2.4-2.6	2.4	2.1-2.7	0.28
SHBG (nmol/l)	49.5	48.5-50.5	53.6	49.9-57.4	0.03

Missing hormone values in women/men: total estradiol 1/5, free estradiol 1/19, total testosterone 2/7, free testosterone 2/20, DHEAS 1/7 and SHBG 0/15.
[a]SHBG, sex hormone-binding globulin.
[b]ANCOVA, analyses of covariance, adjusted for age (years), body mass index (kg/cm^2), height (cm), current smoking (no/yes) and physical activity score.
[c]DHEAS, dehydroepiandrosterone sulphate.

Table 3
The hazard ratios (HR) with 95% CI of non-vertebral fractures by sex-specific standard deviations (SD) change in sex steroids[a], SHBG[b] and BMD[c] in 1386 women and 1364 men. The Tromsø Study 1994-95.

	Per unit of change (SD)	Age-adjusted HR	HR adjusted for age, BMI[d], height, smoking and physical activity	HR adjusted for age, BMI, height, smoking, physical activity and BMD
Women				
Total estradiol	÷ 21 pmol/l	0.98 (0.87-1.10)	0.99 (0.88-1.11)	0.96 (0.85-1.08)
Free estradiol	÷ 0.5 pmol/l	0.93 (0.82-1.05)	0.95 (0.84-1.07)	0.93 (0.82-1.06)
Total testosterone	÷ 0.7 nmol/l	1.06 (0.96-1.18)	1.09 (0.98-1.21)	1.11 (1.00-1.24)
Free testosterone	÷ 9 pmol/l	1.00 (0.89-1.12)	1.04 (0.92-1.16)	1.07 (0.95-1.20)
DHEAS[e]	÷ 1.2 µmol/l	1.02 (0.90-1.16)	1.03 (0.91-1.17)	1.05 (0.93-1.20)
SHBG	÷ 33 nmol/l	1.20 (1.08-1.34) ***	1.17 (1.03-1.33) *	1.09 (0.95-1.24)
BMD	- 66 mg/cm²	1.39 (1.22-1.58) ***	1.39 (1.21-1.59) ***	1.36 (1.19-1.56)[c] ***
Men				
Total estradiol	÷ 31 pmol/l	1.07 (0.89-1.29)	1.06 (0.88-1.28)	1.05 (0.86-1.27)
Free estradiol	÷ 0.7 pmol/l	1.03 (0.85-1.25)	1.02 (0.84-1.24)	1.02 (0.83-1.25)
Total testosterone	÷ 5.2 nmol/l	1.19 (0.99-1.42)	1.21 (1.00-1.47)	1.20 (1.00-1.46)
Free testosterone	÷ 69 pmol/l	1.09 (0.90-1.33)	1.08 (0.89-1.32)	1.11 (0.91-1.35)
DHEAS	÷ 1.9 µmol/l	0.84 (0.67-1.06)	0.84 (0.67-1.06)	0.84 (0.66-1.07)
SHBG	÷ 25 nmol/l	1.21 (1.01-1.46) *	1.27 (1.03-1.55) *	1.22 (1.00-1.50)
BMD	- 69 mg/cm²	1.41 (1.16-1.71) ***	1.46 (1.19-1.79) ***	1.41 (1.15-1.73)[f] ***

*P < 0.05; *** P ≤ 0.001
[a]We included one hormone or SHBG in each of the presented models.
[b]SHBG, sex hormone-binding globulin.
[c]BMD, bone mineral density at the distal forearm.
[d]BMI, body mass index.
[e]DHEAS, dehydroepiandrosterone sulphate.
[f]Models of the BMD effects are adjusted for SHBG, instead of BMD.

Appendix A

First questionnaire Tromsø Study 1994-95, Norwegian and English

Innbydelse til

HELSEUNDERSØKELSEN

Fødselsdato Personnr. Kommune Kretsnr.

Velkommen til helseundersøkelsen i Tromsø!

Helseundersøkelsen kommer nå til Tromsø. Tid og sted for frammøte finner du nedenfor. Du finner også en orientering om undersøkelsen i den vedlagte brosjyren.

Vi ber deg fylle ut spørreskjemaet på baksiden og ta det med til undersøkelsen.

Undersøkelsen blir mest verdifull om frammøtet blir så fullstendig som mulig. Vi håper derfor at du har mulighet til å komme. Møt selv om du kjenner deg frisk, om du er under legebehandling, eller om du har fått målt kolesterol og blodtrykk i den senere tid.

Vennlig hilsen

Kommunehelsetjenesten
Fagområdet medisin, Universitetet i Tromsø
Statens helseundersøkelser

N

Hvordan er helsen din nå? *Sett bare ett kryss.*

Dårlig	12	1
Ikke helt god		2
God		3
Svært god		4

Har du, eller har du hatt:

	JA	NEI	Alder første gang
Hjerteinfarkt 13			år
Angina pectoris (hjertekrampe) 16			år
Hjerneslag/hjerneblødning 19			år
Astma 22			år
Diabetes (sukkersyke) 25			år

Bruker du medisin mot høyt blodtrykk?

Nå	28	1
Før, men ikke nå		2
Aldri brukt		3

Har du i løpet av det siste året vært plaget med smerter og/eller stivhet i muskler og ledd som har vart i minst 3 måneder sammenhengende? 29 JA NEI

Har du de siste to ukene følt deg:

	Nei	Litt	En god del	Svært mye
Nervøs og urolig? 30				
Plaget av angst? 31				
Trygg og rolig? 32				
Irritabel? 33				
Glad og optimistisk? 34				
Nedfor/deprimert? 35				
Ensom? 36				
	1	2	3	4

Røykte noen av de voksne hjemme da du vokste opp? 37 JA NEI

Bor du, eller har du bodd, sammen med noen dagligrøykere etter at du fylte 20 år? 38 JA NEI

Hvis "JA", hvor mange år tilsammen? 39 Antall år

Hvor lenge er du vanligvis daglig tilstede i røykfylt rom? 41 Antall timer

Sett 0 hvis du ikke oppholder deg i røykfylt rom.

Røyker du selv: JA NEI

- Sigaretter daglig? 43
- Sigarer/sigarillos daglig? 44
- Pipe daglig? 45

Hvis du har røykt daglig tidligere, hvor lenge er det siden du sluttet? 46 Antall år

Hvis du røyker daglig nå eller har røykt tidligere:

- Hvor mange sigaretter røyker eller røykte du vanligvis daglig? 48 Antall sigaretter
- Hvor gammel var du da du begynte å røyke daglig? 52 Alder år
- Hvor mange år tilsammen har du røykt daglig? 54 Antall år

MOSJON

Hvordan har din fysiske aktivitet i fritiden vært det siste året? *Tenk deg et ukentlig gjennomsnitt for året.*

Arbeidsvei regnes som fritid.

	Timer pr. uke: Ingen	Under 1	1-2	3 og mer
Lett aktivitet *(ikke svett/andpusten)* 56				
Hard fysisk aktivitet *(svett/andpusten)* 57				
	1	2	3	4

KAFFE

Hvor mange kopper kaffe drikker du daglig?

Sett 0 hvis du ikke drikker kaffe daglig.

		Antall kopper
Kokekaffe	58	
Annen kaffe	60	

ALKOHOL

Er du total avholdsmann/-kvinne? 62 JA NEI

Hvor mange ganger i måneden drikker du vanligvis alkohol? *Regn ikke med lettøl.*

Sett 0 hvis mindre enn 1 gang i mnd. 63 Antall ganger

Hvor mange glass øl, vin eller brennevin drikker du vanligvis i løpet av to uker? 65

Regn ikke med lettøl.

Sett 0 hvis du ikke drikker alkohol.

Øl	Vin	Brennevin
glass	glass	glass

FETT

Hva slags margarin eller smør bruker du vanligvis på brødet? *Sett ett kryss.*

Bruker ikke smør/margarin	71	1
Meierismør		2
Hard margarin		3
Bløt (soft) margarin		4
Smør/margarin blanding		5
Lettmargarin		6

UTDANNING/ARBEID

Hvilken utdanning er den høyeste du har fullført?

Grunnskole, 7-10 år, framhaldsskole, folkehøgskole	72	1
Realskole, middelskole, yrkesskole, 1-2-årig videregående skole		2
Artium, øk.gymnas, allmennfaglig retning i videregående skole		3
Høgskole/universitet, mindre enn 4 år		4
Høgskole/universitet, 4 år eller mer		5

Hva slags arbeidssituasjon har du nå?

Lønnet arbeid	73	
Heltids husarbeid	74	
Utdanning, militærtjeneste	75	
Arbeidsledig, permittert	76	

Hvor mange timer lønnet arbeid har du i uka? 77 Antall timer

Mottar du nå noen av følgende ytelser?

Syketrygd (sykmeldt)	79	
Attføring	80	
Uførepensjon	81	
Alderspensjon	82	
Sosialstøtte	83	
Arbeidsløshetstrygd	84	

SYKDOM I FAMILIEN

Har en eller flere av foreldre eller søsken hatt hjerteinfarkt (sår på hjertet) eller angina pectoris (hjertekrampe)? 85

JA	NEI	VET IKKE

EGEN HELSE

Hvordan er helsen din nå? *Sett bare ett kryss.*

Dårlig	12	1
Ikke helt god		2
God		3
Svært god		4

Har du, eller har du hatt:

	JA	NEI	Alder første gang
Hjerteinfarkt 13			år
Angina pectoris (hjertekrampe) 18			år
Hjerneslag/hjerneblødning 19			år
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Diabetes (sukkersyke) 25			år

Bruker du medisin mot høyt blodtrykk?

Nå	28	1
Før, men ikke nå		2
Aldri brukt		3

Har du i løpet av det siste året vært plaget med smerter og/eller stivhet i muskler og ledd som har vart i minst 3 måneder sammenhengende? 29 — JA / NEI

Har du de siste to ukene følt deg:

	Nei	Litt	En god del	Svært mye
Nervøs og urolig? 30				
Plaget av angst? 31				
Trygg og rolig? 32				
Irritabel? 33				
Glad og optimistisk? 34				
Nedtrykt/deprimert? 35				
Ensom? 36				
	1	2	3	4

RØYKING

Røykte noen av de voksne hjemme da du vokste opp? 37 — JA / NEI

Bor du, eller har du bodd, sammen med noen dagligrøykere etter at du fylte 20 år? 38 — JA / NEI

Hvis "JA", hvor mange år tilsammen? 39 — Antall år

Hvor lenge er du vanligvis daglig tilstede i røykfylt rom? 41 — Antall timer

Sett 0 hvis du ikke oppholder deg i røykfylt rom.

Røyker du selv: — JA / NEI

Sigaretter daglig?	43	
Sigarer/sigarillos daglig?	44	
Pipe daglig?	45	

Hvis du har røykt daglig tidligere, hvor lenge er det siden du sluttet? 46 — Antall år

Hvis du røyker daglig nå eller har røykt tidligere:

Hvor mange sigaretter røyker eller røykte du vanligvis daglig?	48	Antall sigaretter
Hvor gammel var du da du begynte å røyke daglig?	52	Alder år
Hvor mange år tilsammen har du røykt daglig?	54	Antall år

MOSJON

Hvordan har din fysiske aktivitet i fritiden vært det siste året? *Tenk deg et ukentlig gjennomsnitt for året.*
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Hard fysisk aktivitet *(svett/andpusten)* 57				
	1	2	3	4

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Sett 0 hvis du ikke drikker kaffe daglig.

Kokekaffe	58	Antall kopper
Annen kaffe	60	Antall kopper

ALKOHOL

Er du total avholdsmann/-kvinne? 62 — JA / NEI

Hvor mange ganger i måneden drikker du vanligvis alkohol? *Regn ikke med lettøl.*
Sett 0 hvis mindre enn 1 gang i mnd. 63 — Antall ganger

Hvor mange glass øl, vin eller brennevin drikker du vanligvis i løpet av to uker? 65
Regn ikke med lettøl.
Sett 0 hvis du ikke drikker alkohol.

Øl	Vin	Brennevin
glass	glass	glass

FETT

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Bruker ikke smør/margarin	71	1
Meierismør		2
Hard margarin		3
Blot (soft) margarin		4
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Lettmargarin		6

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Realskole, middelskole, yrkesskole, 1-2-årig videregående skole		2
Artium, øk.gymnas, allmennfaglig retning i videregående skole		3
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Høgskole/universitet, 4 år eller mer		5

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Mottar du nå noen av følgende ytelser?

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Uførepensjon	81	
Alderspensjon	82	
Sosialstøtte	83	
Arbeidsløshetstrygd	84	

Har en eller flere av foreldre eller søsken hatt hjerteinfarkt (sår på hjertet) eller angina pectoris (hjertekrampe)? 85 — JA / NEI / VET IKKE

English translation of invitation with the first questionnaire used in the health survey in Tromsø 1994/95

Translation based on translations by Kevin McCafferty and Anne Clancy

HEALTH SURVEY
INVITATION

"This is your chance"

Date of birth Social security No.

Municipality Electoral ward No.

Welcome to the Tromsø Health Survey!

The Health Survey is coming to Tromsø. This leaflet will tell you when and where. You will also find information about the survey in the enclosed brochure.

We would like you to fill in the form overleaf and take it with you to the examination.

The more people take part in the survey, the more valuable its results will be. We hope, therefore, that you will be able to come. Come along even if you feel healthy, if you are currently receiving medical treatment, or if you have had your cholesterol and blood pressure levels taken recently.

Yours sincerely,
Municipal Health Authorities
Faculty of Medicine - University of Tromsø
National Health Screening Service

"This is a real opportunity — Take it!"

Your own health

What is your current state of health?
Tick one box only.

Poor	☐
Not so good	☐
Good	☐
Very good	☐

Do you have, or have you ever had:

	YES	NO	Age first time
Myocardial infarction	☐	☐	_____ years
Angina pectoris	☐	☐	_____ years
Stroke/ brain haemorrhage	☐	☐	_____ years
Asthma	☐	☐	_____ years
Diabetes	☐	☐	_____ years

Do you take medicine for high blood pressure?

At the moment	☐
Used to, but not any longer	☐
Never have	☐

Have you during the last year suffered from pains and/or stiffness in muscles and joints that have lasted continuously for at least 3 months?

YES ☐ NO ☐

Have you in the last two weeks felt:

	No	A little	A lot	Very much
Nervous or worried?	☐	☐	☐	☐
Anxious?	☐	☐	☐	☐
Secure and calm?	☐	☐	☐	☐
Irritable?	☐	☐	☐	☐
Happy and optimistic?	☐	☐	☐	☐
Down/depressed?	☐	☐	☐	☐
Lonely?	☐	☐	☐	☐

Smoking

Did any of the adults at home smoke while you were growing up? YES ☐ NO ☐

Do you now, or have you previously, lived with daily smokers after your 20th birthday?

YES ☐ NO ☐

If "YES", for how many years in all? ____Years

How many hours a day do you normally spend in smoke-filled rooms? _____Hours
Put 0 if you do not spend time in smoke-filled rooms.

Do you yourself smoke:	YES	NO
Cigarettes daily?	☐	☐
Cigars/cigarillos daily?	☐	☐
Pipe daily ?	☐	☐

If you previously smoked daily, how long is it since you stopped? _____ Years

If you smoke daily at the moment, or have smoked before:

How many cigarettes do you smoke/did you smoke per day? _____ Cigarettes

How old were you when you began smoking daily? Age _____ Years

How many years in all have you smoked daily? _____ Years

Exercise

How has your physical activity in leisure time been during this last year? *Think of your weekly average for the year. Time spent going to work counts as leisure time.*

	Hours pr. week None	Less than 1	1-2	3 or more
Light activity (*not sweating or out of breath*)	☐	☐	☐	☐
Hard activity (*sweating/ out of breath*)	☐	☐	☐	☐

Coffee

How many cups of coffee do you drink daily? *Put 0 if you do not drink coffee daily.* Cups

Boiled coffee ☐☐☐
(i.e., grind boiled and allowed to draw)
Other coffee ☐☐☐

Alcohol

Are you a teetotaler? YES ☐ NO ☐

How many times a month do you normally drink alcohol? *Do not count low-alcohol beer. Put 0 if less than once a month.* ____ Times

How many glasses of beer, wine or spirits do you normally drink in a fortnight? *Do not count low-alcohol beer. Put 0 if less than once a month.*

Beer Glasses	Wine Glasses	Spirits Glasses
☐☐	☐☐	☐☐

Fat

What kind of margarine or butter do you normally use on bread? *Tick one box only.*

Don't use butter/margarine	☐
Creamery butter	☐
Hard margarine	☐
Soft margarine	☐
Butter/margarine blend	☐
Light margarine	☐

Education/work

What is the highest level of education you have completed?

7-10 years primary/secondary school, modern secondary school, folk high school	☐
Technical school, middle school, vocational.. school, 1-2 years' senior high school	☐
A-levels/High school diploma, (3-4 years)	☐
College/university, less than 4 years	☐
College/university, 4 or more years	☐

What is your current work situation?

Paid work	☐
Full-time housework	☐
Education, military service	☐
Unemployed, redundant	☐

How many hours of paid work do you have pr. week? _____ Hours

Do you receive any of the following benefits?

Sickness benefit (sick leave)	☐
Rehabilitation benefit	☐
Disability pension	☐
Old-age pension	☐
Social welfare benefits	☐
Unemployment benefit	☐

Illness in the family

Have one or more of your parents or siblings had a heart attack or had angina (heart cramp)?

YES	NO	DON'T KNOW
☐	☐	☐

Appendix B

Second questionnaire for subjects aged <70 years, Tromsø Study 1994-95, Norwegian and English

Helseundersøkelsen i Tromsø

Hovedformålet med Tromsøundersøkelsene er å skaffe ny kunnskap om hjerte-karsykdommer for å kunne forebygge dem. I tillegg skal undersøkelsen øke kunnskapen om kreftsykdommer og andre alminnelige plager som f.eks. allergier, smerter i muskulatur og nervøse lidelser. Vi ber deg derfor svare på noen spørsmål om forhold som kan ha betydning for risikoen for disse og andre sykdommer.

Skjemaet er en del av Helseundersøkelsen som er godkjent av Datatilsynet og av Regional komite for medisinsk forskningsetikk. Svarene brukes bare til forskning og behandles strengt fortrolig. Opplysningene kan senere bli sammenholdt med informasjon fra andre offentlige helseregistre etter de regler som Datatilsynet og Regional komite for medisinsk forskningsetikk gir.

Hvis du er i tvil om hva du skal svare, sett kryss i den ruten som du synes passer best.

Det utfylte skjema sendes i vedlagte svarkonvolutt. Portoen er betalt.

På forhånd takk for hjelpen!

Med vennlig hilsen

Fagområdet medisin
Universitetet i Tromsø **Statens helseundersøkelser**

Hvis du ikke ønsker å besvare spørreskjemaet, sett kryss i ruten under og returner skjemaet. Da slipper du purring.

Jeg ønsker ikke å besvare spørreskjemaet ..... 17 ☐

Dag Mnd År

Dato for utfylling av skjema: ..... 18 ...../...../.....

BAKGRUNN

I hvilken kommune bodde du da du fylte 1 år?

..... 24 - 28

Hvis du ikke bodde i Norge, oppgi land i stedet for kommune.

Hvordan var de økonomiske forhold i familien under din oppvekst?
- Meget gode ..... 29 ☐
- Gode ☐
- Vanskelige ☐
- Meget vanskelige ☐

Hvor mange av de første 3 årene av ditt liv
- bodde du i by? ..... 30 _____ år
- hadde dere katt eller hund i hjemmet? ..... 31 _____ år

Hvor mange av de første 15 årene av ditt liv
- bodde du i by? ..... 32 _____ år
- hadde dere katt eller hund i hjemmet? ..... 34 _____ år

BOLIG

Hvem bor du sammen med?

Sett ett kryss for hvert spørsmål og angi antall.	Ja	Nei	Antall
Ektefelle/samboer 36	☐	☐	
Andre personer over 18 år 37	☐	☐	_____
Personer under 18 år 40	☐	☐	_____

Hvor mange av barna har plass i barnehage? ..... 43 _____

Hvilken type bolig bor du i?
- Enebolig/villa ..... 45 ☐ 1
- Gårdsbruk ☐ 2
- Blokk/terrasseleilighet ☐ 3
- Rekkehus/2-4 mannsbolig ☐ 4
- Annen bolig ☐ 5

Hvor stor er din boenhet? ..... 46 _____ m²

I omtrent hvilket år ble boligen bygget? ..... 48 _____

	Ja	Nei
Er boligen isolert etter 1970? 53	☐	☐
Bor du i underetasje/kjeller? 54	☐	☐
Hvis "Ja", er gulvbelegget lagt på betong? 55	☐	☐

Hvordan er boligen hovedsakelig oppvarmet?
- Elektrisk oppvarming ..... 56 ☐
- Vedfyring ☐
- Sentralvarmeanlegg oppvarmet med:
  - Parafin ☐
  - Elektrisitet ☐

	Ja	Nei
Er det heldekkende tepper i stua? 60	☐	☐
Er det katt i boligen? 61	☐	☐
Er det hund i boligen? 62	☐	☐

ARBEID

Hvis du er i lønnet eller ulønnet arbeid, hvordan vil du beskrive ditt arbeid?
- For det meste stillesittende arbeid? ..... 63 ☐ 1
  (f.eks. skrivebordsarbeid, montering)
- Arbeid som krever at du går mye? ☐ 2
  (f.eks. ekspeditørarb., lett industriarb., undervisning)
- Arbeid hvor du går og løfter mye? ☐ 3
  (f.eks. postbud, pleier, bygningsarbeid)
- Tungt kroppsarbeid? ☐ 4
  (f.eks. skogsarb., tungt jordbruksarb., tungt bygn.arb.)

Kan du selv bestemme hvordan arbeidet ditt skal legges opp?
- Nei, ikke i det hele tatt ..... 64 ☐ 1
- I liten grad ☐ 2
- Ja, i stor grad ☐ 3
- Ja, det bestemmer jeg selv ☐ 4

	Ja	Nei
Har du skiftarbeid, nattarbeid eller går vakter? 65	☐	☐

Har du noen av følgende yrker (heltid eller deltid)?

Sett ett kryss for hvert spørsmål.	Ja	Nei
Sjåfør 66	☐	☐
Bonde/gårdbruker	☐	☐
Fisker	☐	☐

EGNE SYKDOMMER

Har du noen gang hatt:
Sett ett kryss for hvert spørsmål. Oppgi alderen ved hendelsen.
*Hvis det har skjedd flere ganger, hvor gammel var du **siste** gang?*

	Ja	Nei	Alder
Lårhalsbrudd 69	☐	☐	
Brudd ved håndledd/underarm 72	☐	☐	
Nakkesleng (whiplash) 75	☐	☐	
Skade som førte til sykehusinnleggelse 78	☐	☐	
Sår på magesekken 81	☐	☐	
Sår på tolvfingertarmen 84	☐	☐	
Magesår-operasjon 87	☐	☐	
Operasjon på halsen 90	☐	☐	

Har du eller har du hatt:
Sett ett kryss for hvert spørsmål.

	Ja	Nei
Kreftsykdom 93	☐	☐
Epilepsi (fallesyke)	☐	☐
Migrene	☐	☐
Kronisk bronkitt	☐	☐
Psoriasis	☐	☐
Benskjørhet (osteoporose) 98	☐	☐
Fibromyalgi/fibrositt/kronisk smertesyndrom	☐	☐
Psykiske plager som du har søkt hjelp for	☐	☐
Stoffskiftesykdom (skjoldbruskkjertel)	☐	☐
Sykdom i leveren	☐	☐
Nyresten 103	☐	☐
Blindtarmsoperasjon	☐	☐
Allergi og overfølsomhet		
Atopisk eksem (f.eks. barneeksem)	☐	☐
Håndeksem	☐	☐
Høysnue	☐	☐
Matvareallergi 108	☐	☐
Annen overfølsomhet (ikke allergi)	☐	☐

Hvor mange ganger har du hatt forkjølelse, influensa, "ræksjuka" og lignende siste halvår? 110 _____ ganger

	Ja	Nei
Har du hatt dette siste 14 dager? 112	☐	☐

SYKDOMMER I FAMILIEN

Kryss av for de slektningene som har eller har hatt noen av sykdommene:
Kryss av for "Ingen" hvis ingen av slektningene har hatt sykdommen.

	Mor	Far	Bror	Søster	Barn	Ingen
Hjerneslag eller hjerneblødning 113	☐	☐	☐	☐	☐	☐
Hjerteinfarkt før 60 års alder 119	☐	☐	☐	☐	☐	☐
Kreftsykdom 125	☐	☐	☐	☐	☐	☐
Astma 131	☐	☐	☐	☐	☐	☐
Mage/tolvfingertarm-sår 137	☐	☐	☐	☐	☐	☐
Benskjørhet (osteoporose) 143	☐	☐	☐	☐	☐	☐
Psykiske plager 149	☐	☐	☐	☐	☐	☐
Allergi 155	☐	☐	☐	☐	☐	☐
Diabetes (sukkersyke) 161	☐	☐	☐	☐	☐	☐
– alder da de fikk diabetes 167	___	___	___	___	___	

SYMPTOMER

	Ja	Nei
Hoster du omtrent daglig i perioder av året? 177	☐	☐
Hvis "Ja":		
Er hosten vanligvis ledsaget av oppspytt? 178	☐	☐
Har du hatt slik hoste så lenge som i en 3 måneders periode i begge de to siste år? 179	☐	☐
Har du hatt episoder med piping i brystet? 180	☐	☐
Hvis "Ja", har dette oppstått:		
Sett ett kryss for hvert spørsmål.		
Om natten 181	☐	☐
Ved luftveisinfeksjoner	☐	☐
Ved fysiske anstrengelser	☐	☐
Ved sterk kulde	☐	☐
Har du merket anfall med plutselig endring i pulsen eller hjerterytmen siste år? 185	☐	☐

Hvor ofte er du plaget av søvnløshet?
- Aldri, eller noen få ganger i året 186 ☐ 1
- 1-2 ganger i måneden ☐ 2
- Omtrent en gang i uken ☐ 3
- Mer enn en gang i uken ☐ 4

Hvis du er plaget av søvnløshet i perioder, når på året er du mest plaget?
- Ingen spesiell tid 187 ☐ 1
- Særlig i mørketiden ☐ 2
- Særlig i midnattsoltiden ☐ 3
- Særlig vår og høst ☐ 4

	Ja	Nei
Har du det siste året vært plaget av søvnløshet slik at det har gått ut over arbeidsevnen? 188	☐	☐

Hvor ofte er du plaget av hodepine?
- Sjelden eller aldri 189 ☐ 1
- En eller flere ganger i måneden ☐ 2
- En eller flere ganger i uken ☐ 3
- Daglig ☐ 4

Hender det at tanken på å få alvorlig sykdom bekymrer deg?
- Ikke i det hele tatt 190 ☐ 1
- Bare i liten grad ☐ 2
- En del ☐ 3
- Ganske mye ☐ 4

BRUK AV HELSEVESENET

Hvor mange ganger har du siste året, på grunn av egen helse eller sykdom, vært:
*Sett **0** hvis du **ikke** har hatt slik kontakt.*

	Antall ganger siste år
Hos vanlig lege/legevakt 191	
Hos psykolog eller psykiater	
Hos annen legespesialist utenfor sykehus	
På poliklinikk 197	
Innlagt i sykehus	
Hos bedriftslege	
Hos fysioterapeut 203	
Hos kiropraktor	
Hos akupunktør	
Hos tannlege 209	
Hos naturmedisiner (homøopat, soneterapeut o.l.)	
Hos håndspålegger, synsk eller "leser"	

LEGEMIDLER OG KOSTTILSKUDD

Har du det siste året periodevis brukt noen av de følgende midler daglig eller nesten daglig?
Angi hvor mange måneder du brukte dem.
*Sett 0 hvis du **ikke** har brukt midlene.*

Legemidler
- Smertestillende (215) _____ mnd.
- Sovemedisin _____ mnd.
- Beroligende midler _____ mnd.
- Medisin mot depresjon (221) _____ mnd.
- Allergimedisin _____ mnd.
- Astmamedisin _____ mnd.

Kosttilskudd
- Jerntabletter (227) _____ mnd.
- Kalktabletter eller benmel _____ mnd.
- Vitamin D-tilskudd _____ mnd.
- Andre vitamintilskudd (233) _____ mnd.
- Tran eller fiskeoljekapsler _____ mnd.

Har du de siste 14 dager brukt følgende legemidler eller kosttilskudd?
*Sett **ett** kryss for **hvert** spørsmål.*

	Ja	Nei
Legemidler		
Smertestillende medisin (237)	☐	☐
Febersenkende medisin	☐	☐
Migrenemedisin	☐	☐
Eksemsalve	☐	☐
Hjertemedisin (ikke blodtrykksmedisin)	☐	☐
Kolesterolsenkende medisin (242)	☐	☐
Sovemedisin	☐	☐
Beroligende medisin	☐	☐
Medisin mot depresjon	☐	☐
Annen nervemedisin	☐	☐
Syrenøytrallserende midler (247)	☐	☐
Magesårsmedisin	☐	☐
Insulin	☐	☐
Tabletter mot diabetes (sukkersyke)	☐	☐
Tabletter mot lavt stoffskifte (thyroxin)	☐	☐
Kortisontabletter (252)	☐	☐
Annen medisin	☐	☐
Kosttilskudd		
Jerntabletter	☐	☐
Kalktabletter eller benmel	☐	☐
Vitamin D-tilskudd	☐	☐
Andre vitamintilskudd (267)	☐	☐
Tran eller fiskeoljekapsler	☐	☐

VENNER

Hvor mange gode venner har du som du kan snakke fortrolig med og gi deg hjelp når du trenger det? (259) ____ gode venner
Tell ikke med de du bor sammen med, men ta med andre slektninger!

Hvor mange av disse gode vennene har du kontakt med minst en gang i måneden? (261) ____

	Ja	Nei
Føler du at du har nok gode venner? (263)	☐	☐

Hvor ofte tar du vanligvis del i foreningsvirksomhet som f.eks. syklubb, idrettslag, politiske lag, religiøse eller andre foreninger?
- Aldri, eller noen få ganger i året (264) ☐ 1
- 1-2 ganger i måneden ☐ 2
- Omtrent en gang i uken ☐ 3
- Mer enn en gang i uken ☐ 4

KOSTVANER

Hvis du bruker smør eller margarin på brødet, hvor mange skiver rekker en liten porsjonspakning vanligvis til? Vi tenker på slik porsjonspakning som du får på fly, på kafé o.l. (10-12 gram).

Den rekker til omtrent (265) _____ skiver

Hva slags fett blir vanligvis brukt til **matlaging** (ikke på brødet) i din husholdning?
- Meierismør (266) ☐
- Hard margarin ☐
- Bløt (Soft) margarin ☐
- Smør/margarin blanding ☐
- Oljer (270) ☐

Hva slags type brød (kjøpt eller hjemmebakt) spiser du vanligvis?
Sett ett eller to kryss!

	Loff (271)	Fint brød	Kneip-brød	Grov-brød	Knekke-brød (275)
Brødtypen ligner mest på:	☐	☐	☐	☐	☐

Hvor mye (i **antall** glass, kopper, poteter eller brødskiver) spiser eller drikker du vanligvis **daglig** av følgende matvarer?
*Kryss av for **alle** matvarene.*

	0	Færre enn 1	1-2	3-4	5-6	Mer enn 6
Helmelk (søt eller sur) (glass) (276)	☐	☐	☐	☐	☐	☐
Lettmelk (søt eller sur) (glass)	☐	☐	☐	☐	☐	☐
Skummet melk (søt eller sur) (glass)	☐	☐	☐	☐	☐	☐
Te (kopper)	☐	☐	☐	☐	☐	☐
Appelsinjuice (glass)	☐	☐	☐	☐	☐	☐
Poteter (281)	☐	☐	☐	☐	☐	☐
Brødskiver totalt (inkl. knekkebrød)	☐	☐	☐	☐	☐	☐
Brødskiver med						
– fiskepålegg (f.eks. makrell i tomat)	☐	☐	☐	☐	☐	☐
– magert kjøttpålegg (f.eks. skinke)	☐	☐	☐	☐	☐	☐
– fetere kjøttpålegg (f.eks. salami)	☐	☐	☐	☐	☐	☐
– gulost (285)	☐	☐	☐	☐	☐	☐
– brunost	☐	☐	☐	☐	☐	☐
– kaviar	☐	☐	☐	☐	☐	☐
– syltetøy og annet søtt pålegg	☐	☐	☐	☐	☐	☐
	1	2	3	4	5	6

Hvor mange **ganger i uka** spiser du vanligvis følgende matvarer?
*Kryss av for **alle** matvarene.*

	Aldri	Færre enn 1	1	2-3	4-5	Omtrent daglig
Yoghurt (289)	☐	☐	☐	☐	☐	☐
Kokt eller stekt egg	☐	☐	☐	☐	☐	☐
Frokostblanding/havregryn o.l.	☐	☐	☐	☐	☐	☐
Middag med						
– rent kjøtt	☐	☐	☐	☐	☐	☐
– pølser/kjøttpudding/-kaker	☐	☐	☐	☐	☐	☐
– feit fisk (f.eks. laks/ørr) (295)	☐	☐	☐	☐	☐	☐
– mager fisk (f.eks. torsk)	☐	☐	☐	☐	☐	☐
– fiskeboller/-pudding/-kaker	☐	☐	☐	☐	☐	☐
– grønnsaker	☐	☐	☐	☐	☐	☐
Majones, remulade o.l.	☐	☐	☐	☐	☐	☐
Gulrøtter (300)	☐	☐	☐	☐	☐	☐
Blomkål/kål/brokkoli	☐	☐	☐	☐	☐	☐
Epler/pærer	☐	☐	☐	☐	☐	☐
Appelsiner, mandariner o.l.	☐	☐	☐	☐	☐	☐
Sukkerholdige leskedrikker	☐	☐	☐	☐	☐	☐
Sukkerfrie («Light») leskedrikker	☐	☐	☐	☐	☐	☐
Sjokolade	☐	☐	☐	☐	☐	☐
Vafler, kaker o.l. (307)	☐	☐	☐	☐	☐	☐
	1	2	3	4	5	6

ALKOHOL

Hvor ofte pleier du å drikke	øl?	vin?	brennevin?
Aldri, eller noen få ganger i året	☐ 308	☐	☐ 1
1-2 ganger i måneden	☐	☐	☐ 2
Omtrent 1 gang i uken	☐	☐	☐ 3
2-3 ganger i uken	☐	☐	☐ 4
Omtrent hver dag	☐	☐	☐ 5 310

Omtrent hvor ofte har du i løpet av siste år drukket alkohol tilsvarende minst 5 halvflasker øl, en helflaske vin eller 1/4 flaske brennevin?

- Ikke siste år ..... 311 ☐ 1
- Noen få ganger ☐ 2
- 1 - 2 ganger per måned ☐ 3
- 1 - 2 ganger i uken ☐ 4
- 3 eller flere ganger i uken ☐ 5

I omtrent hvor mange år har ditt alkoholforbruk vært slik du har svart i spørsmålene over? ..... 312 _____ år

SLANKING

Omtrent hvor mange ganger har du bevisst prøvd å slanke deg? Sett **0** hvis ingen forsøk.

- før 20 år ..... 314 _____ ganger
- senere ..... 316 _____ ganger

Hvis du har slanket deg, omtrent hvor mange kilo har du på det meste gått ned i vekt?

- før 20 år ..... 318 _____ kg
- senere ..... 320 _____ kg

Hvilken vekt ville du være tilfreds med (din "trivselsvekt")? ..... 322 _____ kg

UFRIVILLIG URINLEKKASJE

Hvor ofte har du ufrivillig urinlekkasje?

- Aldri ..... 325 ☐ 1
- Ikke mer enn en gang i måneden ☐ 2
- To eller flere ganger i måneden ☐ 3
- Ukentlig eller oftere ☐ 4

Dine kommentarer:

BESVARES BARE AV KVINNER

MENSTRUASJON

Hvor gammel var du da du fikk menstruasjon første gang? ..... 326 _____ år

Hvis du ikke lenger har menstruasjon, hvor gammel var du da den sluttet? ..... 328 _____ år

Når du ser bort fra svangerskap og barselsperiode, har du noen gang vært blødningsfri i minst 6 måneder? ..... 330 Ja ☐ Nei ☐

Hvis "Ja", hvor mange ganger? ..... 331 _____ ganger

Hvis du fremdeles har menstruasjon eller er gravid:

Hvilken dato startet din siste menstruasjon? ..... 333 dag/ mnd/ år ___/___/___

Bruker du vanligvis smertestillende legemidler for å dempe menstruasjonsplager? ..... 339 Ja ☐ Nei ☐

SVANGERSKAP

Hvor mange barn har du født? ..... 340 _____ barn

Er du gravid nå? ..... 342 Ja ☐ Nei ☐ Usikker ☐

Har du i forbindelse med svangerskap hatt for høyt blodtrykk og/eller eggehvite (protein) i urinen? ..... 343 Ja ☐ Nei ☐

Hvis "Ja", i hvilket svangerskap?

	Svangerskap Første	Senere
For høyt blodtrykk 344	☐	☐
Eggehvite i urinen 346	☐	☐

Hvis du har født, fyll ut for hvert barn barnets fødselsår og omtrent antall måneder du ammet barnet.

Barn:	Fødselsår:	Antall måneder med amming:
1	348 _____	_____
2	_____	_____
3	356 _____	_____
4	_____	_____
5	364 _____	_____
6	_____	_____

PREVENSJON OG ØSTROGEN

Bruker du eller har du brukt:	Nå	Før	Aldri
P-pille (også minipille) 372	☐	☐	☐
Hormonspiral	☐	☐	☐
Østrogen (tabletter eller plaster) 374	☐	☐	☐
Østrogen (krem eller stikkpiller)	☐ 1	☐ 2	☐ 3

Hvis du bruker p-pille, hormonspiral eller østrogen; hvilket merke bruker du nå?

376 .....

Hvis du bruker eller har brukt p-pille:

Alder da du begynte med P-piller? ..... 380 _____ år

Hvor mange år har du tilsammen brukt P-piller? ..... 382 _____ år

Dersom du har født, hvor mange år brukte du P-piller før første fødsel? ..... 384 _____ år

Hvis du har sluttet å bruke P-piller:

Alder da du sluttet? ..... 386 _____ år

Takk for hjelpen! Husk å postlegge skjemaet idag!

Helsundersøkelsene i Tromsø

English translation of the second questionnaire used in the health survey in Tromsø 1994/95 for subjects younger than 70 years.

Based on translations by K. McCafferty and A. Clancy

TROMSØ HEALTH SURVEY

The main aim of the Tromsø survey is to improve our knowledge of heart and circulatory conditions in order to aid prevention. The survey is also intended to improve our knowledge of cancer and other general conditions, such as allergies, muscle pains and nervous conditions. We would therefore like you to answer some questions about factors that may be relevant for your risk of getting these and other illnesses.

This form is part of the Health Survey, which has been approved by the Norwegian Data Inspectorate and the Regional Board of Research Ethics. The answers will only be used for research purposes and will be treated in strict confidence. The information you give us may later be stored along with information from other public health registers in accordance with the rules laid down by the Data Inspectorate and the Regional Board of Research Ethics.

If you are unsure about what to answer, tick the box that you feel fits best.

The completed form should be sent to us in the enclosed pre-paid envelope.

Thank you in advance for helping us.

Yours sincerely,

Faculty of Medicine
University of Tromsø

National Health
Screening Service

If you do not wish to answer the questionnaire, tick the box below and return the form. Then you will not receive reminders.

I do not wish to answer the questionnaire. ☐

Date for filling in this form: Day/Month/Year

CHILDHOOD/YOUTH

What Norwegian municipality did you live in at the age of 1 year? _____

If you did not live in Norway, give country of residence instead of municipality.

How was your family's economic situation while you were growing up?

- Very good ☐
- Good ☐
- Difficult ☐
- Very difficult ☐

For how much of the first three years of your life

- did you live in a town/city? ____ Years
- did your family have a cat or dog in the home? ____ Years

For how much of the first 15 years of your life

- did you live in a town/city? ____ Years
- did your family have a cat or dog in the home? ____ Years

HOME

Who do you live with?

Tick once for each item and give the number of persons.

	YES	NO	Number
Spouse/partner	☐	☐	_____
Other persons over 18 years	☐	☐	_____
Persons under 18 years	☐	☐	_____

How many of the children go to day care/kindergarten/ nursery school? ____

What type of home do you live in?

- Villa/ detached house ☐
- Farm ☐
- Flat /Apartment ☐
- Terraced /semi-detached house ☐
- Other ☐

How big is your home? _____ m2

Approximately what year was your home built? _____

	YES	NO
Has your home been insulated after 1970?	☐	☐
Do you live on the bottom floor/cellar level?	☐	☐
If "YES", is the floor laid on concrete?	☐	☐

What is the main source of heat in your home?

- Electric heating ☐
- Wood-burning stove ☐
- Central heating system using:
  - Paraffin ☐
  - Electricity ☐

	YES	NO
Do you have fitted carpets in the living-room?	☐	☐
Is there a cat in your home?	☐	☐
Is there a dog in your home?	☐	☐

WORK

If you are in paid or unpaid work, which statement describes your work best?

- I am mainly seated while working (*e.g., at a desk/assembly work*) ☐
- My work requires a lot of walking (*e.g., shop assistant, light industrial work, teaching*) ☐
- My work entails a lot of walking and lifting (*e.g., postman/woman, nurse, building work*) ☐
- I do heavy physical work (*e.g., forestry, heavy agricultural/construction work*) ☐

Do you have any influence on how your work is organised?

- No, not at all ☐
- To a small extent ☐
- Yes, to a large extent ☐
- Yes, I decide myself ☐

	YES	NO
Are you on call; do you work shifts or nights?	☐	☐

Do you do any of the following jobs (full- or part-time)?

Tick one box only for each item.	YES	NO
Driver	☐	☐
Farmer	☐	☐
Fisherman	☐	☐

YOUR OWN ILLNESSES

Have you ever had:

Tick one box only for each item. Give your age at the time. If you have had the condition several times, how old were you last time?

	YES	NO	AGE
Hip fracture	☐	☐	____
Wrist/forearm fracture	☐	☐	____
Whiplash	☐	☐	____
Injury requiring hospital admission	☐	☐	____
Stomach ulcer	☐	☐	____
Duodenal ulcer	☐	☐	____
An operation for stomach/ duodenal ulcer	☐	☐	____
Throat/ neck operation	☐	☐	____

Have you you ever had, or do you still have:

Tick one box only for each item.	YES	NO
Cancer	☐	☐
Epilepsy	☐	☐
Migraine	☐	☐
Chronic bronchitis	☐	☐
Psoriasis	☐	☐
Osteoporosis	☐	☐
Fibromyalgia/fibrositis/ chronic pain syndrome	☐	☐
Psychological problems for which you have sought help	☐	☐
Thyroid disease	☐	☐
Liver disease	☐	☐
Kidney stone	☐	☐
Appendectomy	☐	☐
Allergy and hypersensitivity:		
Atopic eczema (e.g., childhood eczema)	☐	☐
Hand eczema	☐	☐
Hay fever	☐	☐
Food allergy	☐	☐
Other hypersensitivity (not allergy)	☐	☐

How many times have you had a cold, influenza (flue), vomiting/diarrhoea, or similar in the last six months?

_____ times

Have you had any of these in the last two weeks?

YES	NO
☐	☐

ILLNESS IN THE FAMILY

Tick the appropriate box for relatives that have, or have ever had the following illnesses: *Tick "None" if none of your relatives have had the condition.*

	Mother	Father	Brother	Sister	Child	None
Stroke or brain haemorrhage	☐	☐	☐	☐	☐	☐
Myocardial infarction before age 60	☐	☐	☐	☐	☐	☐
Cancer	☐	☐	☐	☐	☐	☐
Asthma	☐	☐	☐	☐	☐	☐
Stomach/ duodenal ulcer	☐	☐	☐	☐	☐	☐
Osteoporosis	☐	☐	☐	☐	☐	☐
Psychological problems	☐	☐	☐	☐	☐	☐
Allergy	☐	☐	☐	☐	☐	☐
Diabetes	☐	☐	☐	☐	☐	☐
-age when they got diabetes	___	___	___	___	___	___

SYMPTOMS

	YES	NO
Do you cough approximately every day of the year?	☐	☐
If "Yes": Is your cough productive ?	☐	☐
Have you had this kind of cough for as long as 3 months in each of the last two years?	☐	☐
Have you had periods of wheezing in your chest?	☐	☐
If "Yes", has this occurred: *Tick one box only for each item.*		
At night	☐	☐
In connection with respiratory infections	☐	☐
In connection with physical exertion	☐	☐
In connection with very cold weather	☐	☐
Have you noticed sudden changes in your pulse or heart rhythm in the last year?	☐	☐

How often do you suffer from sleeplessness?

- Never, or just a few times a year ☐
- 1-2 times a month ☐
- Approximately once a week ☐
- More than once a week ☐

If you suffer from periods of sleeplessness, what times of the year does it affect you most?

- No particular time of year ☐
- Especially during the dark winter months ☐
- Especially during the midnight sun period ☐
- Especially in spring and autumn ☐

Have you in the last twelve months suffered from sleeplessness to the extent that it has affected your ability to work? YES ☐ NO ☐

How often do you suffer from headaches?

- Seldom/Never ☐
- Once a month or more ☐
- Once a week or more ☐
- Every day ☐

Does the thought of getting a serious illness ever worry you?

- Not at all ☐
- Only a little ☐
- Some ☐
- Very much ☐

USE OF HEALTH SERVICES

How many visits have you made during the past year due to your own health or illness? *Tick 0 if you have not had such contact*

	Number of times the past year
To a general practitioner (GP)/ Emergency GP	____
Psychologist or psychiatrist	____
Other medical specialist (not at a hospital)	____
Hospital out-patient clinic	____
Hospital admission	____
Medical officer at work	____
Physiotherapist	____
Chiropractor	____
Acupuncturist	____
Dentist	____
Alternative medical practitioner (homoeopath, foot zone therapist, etc.)	____
Healer, Faith healer, clairvoyant	____

MEDICATION AND DIETARY SUPPLEMENTS

Have you for any length of time in the past year used any of the following medicines every day or almost daily? Indicate how many months you used them for. *Write 0 for items you have not used.*

Medication:

- Painkillers ____ mths
- Sleeping pills ____ mths
- Tranquilizers ____ mths
- Antidepressants ____ mths
- Allergy drugs ____ mths
- Asthma drugs ____ mths

Dietary supplements

- Iron tablets ____ mths
- Calcium tablets or bonemeal ____ mths
- Vitamin D supplement ____ mths
- Other vitamin supplements ____ mths
- Cod liver oil or fish oil capsules ____ mths

Have you in the last 14 days used the following medicines or dietary supplements? *Tick one box only for each item.*

Medicines	YES	NO
Painkillers	☐	☐
Antipyretic drugs (to reduce fever)	☐	☐
Migraine drugs	☐	☐
Eczema cream/ointment	☐	☐
Heart medicine (not blood pressure)	☐	☐
Lipid lowering drugs	☐	☐
Sleeping pills	☐	☐
Tranquilizers	☐	☐
Antidepressants	☐	☐
Other drugs for nervous conditions	☐	☐
Antacids	☐	☐
Gastric ulcer drugs	☐	☐
Insulin	☐	☐
Diabetes tablets	☐	☐
Thyroxin tablets (for metabolic disorder)	☐	☐
Cortisone tablets	☐	☐
Other medicine(s)	☐	☐

Dietary supplements	YES	NO
Iron tablets	☐	☐
Calcium tablets or bonemeal	☐	☐
Vitamin D supplement	☐	☐
Other vitamin supplements	☐	☐
Cod liver oil or fish oil capsules	☐	☐

FRIENDS

How many good friends do you have whom you can talk confidentially with and who give you help when you need it? _____ good friends

Do not count people you live with, but do include other relatives!

How many of these good friends do you have contact with at least once a month? _____

Do you feel you have enough good friends? YES ☐ NO ☐

How often do you normally take part in organised gatherings, e.g., sewing circles, sports clubs, political meetings, religious or other associations?

- Never, or just a few times a year ☐
- 1-2 times a month ☐
- Approximately once a week ☐
- More than once a week ☐

DIET

If you use butter or margarine on your bread, how many slices does a small catering portion normally cover? By this, we mean the portion packs served on planes, in cafés, etc. (i.e., 10-12g)

A catering portion is enough for about ____ slices.

What kind of fat is normally used in **cooking** (not on the bread) in your home?

- Creamery butter ☐
- Hard margarine ☐
- Soft margarine ☐
- Butter/margarine blend ☐
- Oils ☐

What kind of bread (bought or home-made) do you usually eat? *Tick one or two boxes!*

The bread I eat is most similar to

- White bread ☐
- Light textured brown bread ☐
- Ordinary brown bread ☐
- Coarse brown bread ☐
- Crisp bread ☐

How much (in **number** of glasses, cups, potatoes or slices) do you usually eat or drink **daily** of the following foodstuffs? *Tick one box for **each** foodstuff.*

	0	Less than 1	1-2	3-4	5-6	More than 6
Full cream milk (fresh or soured) (glasses)	☐	☐	☐	☐	☐	☐
Semi-skimmed milk (low-fat) (fresh or soured) (glasses)	☐	☐	☐	☐	☐	☐
Skimmed milk (fresh or soured) (glasses)	☐	☐	☐	☐	☐	☐
Tea (cups)	☐	☐	☐	☐	☐	☐
Orange juice (glasses)	☐	☐	☐	☐	☐	☐
Potatoes	☐	☐	☐	☐	☐	☐
Slices of bread in total (incl. crispbread)	☐	☐	☐	☐	☐	☐
Slices of bread with fish (e.g., mackerel in tomato sauce	☐	☐	☐	☐	☐	☐
- lean meat (e.g., ham)	☐	☐	☐	☐	☐	☐
- fat meat (e.g., salami)	☐	☐	☐	☐	☐	☐
- cheese (e.g. Gouda/ Norvegia)	☐	☐	☐	☐	☐	☐
- brown cheese	☐	☐	☐	☐	☐	☐
- smoked cod caviar	☐	☐	☐	☐	☐	☐
- jam and other sweet spreads	☐	☐	☐	☐	☐	☐

How many **times per week** do you normally eat the following foodstuffs? *Tick a box for **all** foodstuffs listed.*

	Never	Less than 1	1	2-3	4-5	Roughly every day
Yoghurt	☐	☐	☐	☐	☐	☐
Boiled or fried egg	☐	☐	☐	☐	☐	☐
Breakfast cereal/ oat meal, etc.	☐	☐	☐	☐	☐	☐
For dinner - meat	☐	☐	☐	☐	☐	☐
- sausage/meatloaf/ meatballs	☐	☐	☐	☐	☐	☐
- fat fish (e.g., salmon/ redfish)	☐	☐	☐	☐	☐	☐
- lean fish (e.g., cod)						
- fishballs/fishpudding/ fishcakes	☐	☐	☐	☐	☐	☐
- vegetables	☐	☐	☐	☐	☐	☐
Mayonnaise, remoulade	☐	☐	☐	☐	☐	☐
Carrots	☐	☐	☐	☐	☐	☐
Cauliflower/cabbage/ broccoli	☐	☐	☐	☐	☐	☐
Apples/pears	☐	☐	☐	☐	☐	☐
Oranges, mandarines	☐	☐	☐	☐	☐	☐
Sweetened soft drinks	☐	☐	☐	☐	☐	☐
Sugarfree ("Light") soft drinks	☐	☐	☐	☐	☐	☐
Chocolate	☐	☐	☐	☐	☐	☐
Waffles, cakes, etc.	☐	☐	☐	☐	☐	☐

ALCOHOL

How often do you usually drink	beer?	wine?	spirits?
Never, or just a few times a year	☐	☐	☐
1-2 times a month	☐	☐	☐
Roughly once a week	☐	☐	☐
2-3 times a week	☐	☐	☐
Roughly every day	☐	☐	☐

Approximately how often in the last year have you drunk alcohol that equals at least 5 small bottles of beer, a bottle of wine, or 1/4 bottle of spirits?

- Not in the last year ☐
- Just a few times ☐
- 1-2 times a month ☐
- 1-2 times a week ☐
- 3 or more times a week ☐

For approximately how many years has your alcohol comsumption been as you described above? _____ years

WEIGHT REDUCTION

About how many times have you deliberately tried to lose weight? *Write 0 if you never have.*

- before age 20 _____ times
- after age 20 _____ times

If you have lost weight, about how many kilos have you ever lost at the most?

- before age 20 _____ times _____ kg
- after age 20 _____ times _____ kg

What weight would you be satisfied with (your "ideal weight")? _____ kg

URINARY INCONTINENCE

How often do you suffer from urinary incontinence?

Never	☐
Not more than once a month	☐
Two or more times a month	☐
Once a week or more	☐

Your comments:

Thank you for helping us! Remember to post the form today!

Tromsø Health Survey

TO BE ANSWERED BY WOMEN ONLY

MENSTRUATION

How old were you when you had your first menstruation? _____ years

If you no longer menstruate, how old were you when you stopped having menstruation? _____ years

Apart from pregnancy and after giving birth, have you ever stopped having menstruation for 6 months or more?

YES ☐ NO ☐

If "Yes", how many times? _____ times

If you still menstruate or are pregnant:

What date did your last menstruation begin?

day/month/year ___ / ___ / ___

Do you normally use painkillers to relieve period pains?

YES ☐ NO ☐

PREGNANCY

How many children have you given birth to? _____ children

Are you pregnant at the moment? YES ☐ NO ☐ Don't know ☐

During pregnancy, have you had high blood pressure and/or proteinuria? YES ☐ NO ☐

If "Yes", during which pregnancy?

	Pregnancy	
	First	Later
High blood pressure	☐	☐
Proteinuria	☐	☐

If you have given birth, fill out for each child the year of birth and approximately how many months you breastfed the child.

Child:	Year of birth:	Number of months breastfed:
1	_____	_____ months
2	_____	_____ months
3	_____	_____ months
4	_____	_____ months
5	_____	_____ months
6	_____	_____ months

CONTRACEPTION AND OESTROGEN

Do you, or have you ever, used:	Now	Used to	Never:
Contraceptive pills (incl.minipill)	☐	☐	☐
A hormonal intrauterine device	☐	☐	☐
Oestrogen (tablets or patches)	☐	☐	☐
Oestrogen (cream or suppositories)	☐	☐	☐

If you use contraceptive pills, hormonal intrauterine device, or oestrogen, what brand do you currently use?

_____

If you use, or have ever used, contraceptive pills:

Age when you began taking the pill? _____years

How many years in total have you taken the pill? _____years

If you have given birth, how many years did you take the pill before your first child? _____years

If you have stopped taking the pill:

Age when you stopped? _____years

Appendix C

Second questionnaire for subjects aged > 70 years, Tromsø Study 1994-95, Norwegian and English

Helseundersøkelsen i Tromsø

for dem som er 70 år og eldre.

Hovedformålet med Tromsøundersøkelsene er å skaffe ny kunnskap om hjerte-karsykdommer for å kunne forebygge dem. De skal også øke kunnskapen om kreftsykdommer og alminnelige plager som f.eks. allergier, smerter i muskulatur og nervøse lidelser. Endelig skal de gi kunnskap om hvorledes den eldste delen av befolkningen har det. Vi ber deg derfor svare på spørsmålene nedenfor.

Skjemaet er en del av Helseundersøkelsen som er godkjent av Datatilsynet og av Regional komite for medisinsk forskningsetikk. Svarene brukes bare til forskning og behandles strengt fortrolig. Opplysningene kan senere bli sammenholdt med informasjon fra andre offentlige helseregistre etter de regler som Datatilsynet og Regional komite for medisinsk forskningsetikk gir.

Hvis du er i tvil om hva du skal svare, sett kryss i den ruten som du synes passer best.

Det utfylte skjema sendes i vedlagte svarkonvolutt. Portoen er betalt.

På forhånd takk for hjelpen!

Med vennlig hilsen

Fagområdet medisin
Universitetet i Tromsø — Statens helseundersøkelser

Hvis du ikke ønsker å besvare spørreskjemaet, sett kryss i ruten under og returner skjemaet. Da slipper du purring.

Jeg ønsker ikke å besvare spørreskjemaet ..... 17 ☐

Dag Mnd År

Dato for utfylling av skjema: ..... 18 ...../...../.....

OPPVEKST

I hvilken kommune bodde du da du fylte 1 år?

..... 24 28

Hvis du ikke bodde i Norge, oppgi land i stedet for kommune.

Hvordan var de økonomiske forhold i familien under din oppvekst?

- Meget gode ..... 29 ☐ 1
- Gode ..... ☐ 2
- Vanskelige ..... ☐ 3
- Meget vanskelige ..... ☐ 4

Hvor gamle ble dine foreldre?

- Mor ble ..... 30 _____ år
- Far ble ..... 32 _____ år

BOLIG

Hvem bor du sammen med?
Sett ett kryss for hvert spørsmål og angi antall.

		Ja	Nei	Antall
Ektefelle/samboer	34	☐	☐	
Andre personer over 18 år	35	☐	☐	_____
Personer under 18 år	36	☐	☐	_____

Hvilken type bolig bor du i?

- Enebolig/villa ..... 41 ☐ 1
- Gårdsbruk ..... ☐ 2
- Blokk/terrasseleilighet ..... ☐ 3
- Rekkehus/2-4 mannsbolig ..... ☐ 4
- Annen bolig ..... ☐ 5

Hvor lenge har du bodd i boligen du bor i nå? ..... 42 _____ år

		Ja	Nei
Er boligen tilpasset til dine behov?	44	☐	☐
Hvis "Nei", er det problemer med:			
Plassen i boligen	45	☐	☐
Ujevn, for høy eller for lav temperatur	46	☐	☐
Trapper	47	☐	☐
Toalett	48	☐	☐
Bad/dusj	49	☐	☐
Vedlikehold	50	☐	☐
Annet (spesifiser)	51	☐	☐
Ønsker du å flytte til en eldrebolig?	52	☐	☐

TIDLIGERE ARBEID OG ØKONOMI

Hvordan vil du beskrive det arbeidet du hadde de siste 5-10 årene før du ble pensjonist?

- For det meste stillesittende arbeid? ..... 53 ☐ 1
  (f.eks. skrivebordsarbeid, montering)
- Arbeid som krever at du går mye? ..... ☐ 2
  (f.eks. ekspeditørarbeid, husmor, undervisning)
- Arbeid hvor du går og løfter mye? ..... ☐ 3
  (f.eks. postbud, pleier, bygningsarbeid)
- Tungt kroppsarbeid? ..... ☐ 4
  (f.eks. skogsarb., tungt jordbruksarb., tungt bygn.arb.)

Har du hatt noen av følgende yrker (heltid eller deltid)?

		Ja	Nei
Sett ett kryss for hvert spørsmål.			
Sjåfør	54	☐	☐
Bonde/gårdbruker	55	☐	☐
Fisker	56	☐	☐

Hvor gammel var du da du ble pensjonert? ..... 57 _____ år

Hva slags pensjon har du?

- Minstepensjon ..... 59 ☐
- Tilleggspensjon ..... 60 ☐

Hvordan er din økonomi nå?

- Meget god ..... 61 ☐ 1
- God ..... ☐ 2
- Vanskelig ..... ☐ 3
- Meget vanskelig ..... ☐ 4

HELSE OG SYKDOM

Er helsen din blitt forandret det siste året?
- Ja, dårligere ..... 62 ☐ 1
- Nei, uforandret ..... ☐ 2
- Ja, bedre ..... ☐ 3

Hvordan synes du at helsen din er nå i forhold til andre på samme alder?
- Mye dårligere ..... 63 ☐ 1
- Litt dårligere ..... ☐ 2
- Omtrent lik ..... ☐ 3
- Litt bedre ..... ☐ 4
- Mye bedre ..... ☐ 5

EGNE SYKDOMMER

Har du noen gang hatt:
Sett ett kryss for hvert spørsmål. Oppgi alderen ved hendelsen.
Hvis det har skjedd flere ganger, hvor gammel var du siste gang?

		Ja	Nei	Alder
Lårhalsbrudd	64	☐	☐	_____
Brudd ved håndledd/underarm	67	☐	☐	_____
Nakkesleng (whiplash)	70	☐	☐	_____
Skade som førte til sykehusinnleggelse	73	☐	☐	_____
Sår på magesekken	76	☐	☐	_____
Sår på tolvfingertarmen	79	☐	☐	_____
Magesår-operasjon	82	☐	☐	_____
Operasjon på halsen	85	☐	☐	_____

Har du eller har du hatt:
Sett ett kryss for hvert spørsmål.

		Ja	Nei
Kreftsykdom	88	☐	☐
Epilepsi (fallesyke)		☐	☐
Migrene		☐	☐
Parkinsons sykdom		☐	☐
Kronisk bronkitt		☐	☐
Psoriasis	93	☐	☐
Benskjørhet (osteoporose)		☐	☐
Fibromyalgi/fibrositt/kronisk smertesyndrom		☐	☐
Psykiske plager som du har søkt hjelp for		☐	☐
Stoffskiftesykdom (skjoldbruskkjertel)		☐	☐
Sykdom i leveren	96	☐	☐
Gjentatt, ufrivillig urinlekkasje		☐	☐
Grønn stær		☐	☐
Grå stær		☐	☐
Slitasjegikt (artrose)		☐	☐
Leddgikt	103	☐	☐
Nyrestein		☐	☐
Blindtarmsoperasjon		☐	☐
Allergi og overfølsomhet			
Atopisk eksem (f.eks. barneeksem)		☐	☐
Håndeksem		☐	☐
Høysnue	108	☐	☐
Matvareallergi		☐	☐
Annen overfølsomhet (ikke allergi)		☐	☐

Hvor mange ganger har du hatt forkjølelse, influensa, "ræksjuka" og lignende siste halvår? 111 ____ ganger

		Ja	Nei
Har du hatt dette de siste 14 dager?	113	☐	☐

SYKDOM I FAMILIEN

Kryss av for de slektningene som har eller har hatt noen av sykdommene:
Kryss av for "ingen" hvis ingen av slektningene har hatt sykdommen.

		Mor	Far	Bror	Søster	Barn	Ingen
Hjerneslag eller hjerneblødning	114	☐	☐	☐	☐	☐	☐
Hjerteinfarkt før 60 års alder	120	☐	☐	☐	☐	☐	☐
Kreftsykdom	126	☐	☐	☐	☐	☐	☐
Høyt blodtrykk	132	☐	☐	☐	☐	☐	☐
Astma	138	☐	☐	☐	☐	☐	☐
Benskjørhet (osteoporose)	144	☐	☐	☐	☐	☐	☐
Slitasjegikt (artrose)	150	☐	☐	☐	☐	☐	☐
Psykiske plager	156	☐	☐	☐	☐	☐	☐
Alderdomssløvhet	162	☐	☐	☐	☐	☐	☐
Diabetes (sukkersyke)	168	☐	☐	☐	☐	☐	☐
– alder da de fikk diabetes	174	___	___	___	___	___	

SYMPTOMER

		Ja	Nei
Hoster du omtrent daglig i perioder av året?	184	☐	☐
Hvis "Ja":			
Er hosten vanligvis ledsaget av oppspytt?	185	☐	☐
Har du hatt slik hoste så lenge som i en 3 måneders periode i begge de to siste år?	186	☐	☐
Har du hatt episoder med piping i brystet?	187	☐	☐
Hvis "Ja", har dette oppstått:			
Sett ett kryss for hvert spørsmål.			
Om natten	188	☐	☐
Ved luftveisinfeksjoner		☐	☐
Ved fysiske anstrengelser		☐	☐
Ved sterk kulde	191	☐	☐
Har du merket anfall med plutselig endring i pulsen eller hjerterytmen siste år?	192	☐	☐
Har du gått ned i vekt siste året?	193	☐	☐
Hvis "Ja":			
Hvor mange kilo?	194	_____ kg	

Hvor ofte er du plaget av søvnløshet?
- Aldri, eller noen få ganger i året ..... 196 ☐ 1
- 1-2 ganger i måneden ..... ☐ 2
- Omtrent en gang i uken ..... ☐ 3
- Mer enn en gang i uken ..... ☐ 4

Hvis du er plaget av søvnløshet i perioder, når på året er du mest plaget?
- Ingen spesiell tid ..... 197 ☐ 1
- Særlig i mørketiden ..... ☐ 2
- Særlig i midnattsoltiden ..... ☐ 3
- Særlig vår og høst ..... ☐ 4

		Ja	Nei
Pleier du å ta en lur på dagen?	198	☐	☐
Føler du at du vanligvis får nok søvn?		☐	☐

Er du plaget av:		Nei	Litt	I stor grad
Svimmelhet	200	☐	☐	☐
Dårlig hukommelse		☐	☐	☐
Kraftløshet		☐	☐	☐
Forstoppelse	203	☐	☐	☐

Hender det at tanken på å få alvorlig sykdom bekymrer deg?

- Ikke i det hele tatt ..... 204 ☐
- Bare i liten grad ☐
- En del ☐
- Ganske mye ☐

LEGEMLIGE FUNKSJONER

Klarer du selv disse gjøremålene i det daglige uten hjelp fra andre?	Ja	Med noe hjelp	Nei
Gå innendørs i samme etasje 205	☐	☐	☐
Gå i trapper	☐	☐	☐
Gå utendørs	☐	☐	☐
Gå ca. 500 meter	☐	☐	☐
Gå på toalettet	☐	☐	☐
Vaske deg på kroppen 210	☐	☐	☐
Bade eller dusje	☐	☐	☐
Kle på og av deg	☐	☐	☐
Legge deg og stå opp	☐	☐	☐
Spise selv	☐	☐	☐
Lage varm mat 215	☐	☐	☐
Gjøre lett husarbeid (f.eks. oppvask)	☐	☐	☐
Gjøre tyngre husarbeid (f.eks. gulvvask)	☐	☐	☐
Gjøre innkjøp	☐	☐	☐
Ta bussen	☐	☐	☐

	Ja	Vanskelig	Nei
Kan du høre vanlig tale (evt. med høreapparat)? 220	☐	☐	☐
Kan du lese (evt. med briller)? 221	☐	☐	☐

Er du avhengig av noen av disse hjelpemidlene?

	Ja	Nei
Stokk 222	☐	☐
Krykke	☐	☐
Gåstol (rullator)	☐	☐
Rullestol	☐	☐
Høreapparat	☐	☐
Trygghetsalarm 227	☐	☐

BRUK AV HELSEVESENET

Hvor mange ganger har du siste året, på grunn av egen helse eller sykdom, vært:
Sett 0 hvis du ikke har hatt slik kontakt.

	Antall ganger siste år
Hos vanlig lege/legevakt 228	
Hos psykolog eller psykiater	
Hos annen legespesialist utenfor sykehus	
På poliklinikk 234	
Innlagt i sykehus	
Hos fysioterapeut	
Hos kiropraktor 240	
Hos akupunktør	
Hos tannlege	
Hos fotterapeut 246	
Hos naturmedisiner (homøopat, soneterapeut o.l.)	
Hos håndspåleggger, synsk eller "leser"	

Har du hjemmehjelp?	Ja	Nei
Privat 252	☐	☐
Kommunal	☐	☐
Har du hjemmesykepleie?	☐	☐

Er du fornøyd med helse- og hjemmetjenesten i kommunen?	Ja	Nei	Vet ikke
Prinsippet med fast lege 255	☐	☐	☐
Hjemmesykepleien	☐	☐	☐
Hjemmehjelpen	☐	☐	☐

Er du trygg på at du kan få hjelp av helse- og hjemmetjenesten hvis du trenger det?

- Trygg ..... 258 ☐ 1
- Ikke trygg ☐ 2
- Svært utrygg ☐ 3
- Vet ikke ☐ 4

LEGEMIDLER OG KOSTTILSKUDD

Har du det siste året periodevis brukt noen av de følgende midler daglig eller nesten daglig? Angi hvor mange måneder du brukte dem.
Sett 0 hvis du ikke har brukt midlene.

Legemidler

Smertestillende 259	____ mnd.
Sovemedisin	____ mnd.
Beroligende midler	____ mnd.
Medisin mot depresjon 265	____ mnd.
Allergimedisin	____ mnd.
Astmamedisin	____ mnd.
Hjertemedisin (ikke blodtrykksmedisin) 271	____ mnd.
Insulin	____ mnd.
Tabletter mot diabetes (sukkersyke)	____ mnd.
Tabletter mot lavt stoffskifte (thyroxin) 277	____ mnd.
Kortisontabletter	____ mnd.
Midler mot forstoppelse	____ mnd.

Kosttilskudd

Jerntabletter 283	____ mnd.
Vitamin D-tilskudd	____ mnd.
Andre vitamintilskudd	____ mnd.
Kalktabletter eller benmel 289	____ mnd.
Tran eller fiskeoljekapsler	____ mnd.

FAMILIE OG VENNER

Har du nær familie som kan gi deg hjelp og støtte når du trenger det? 293	Ja ☐	Nei ☐
Hvis "Ja": Hvem kan gi deg hjelp?		
Ektefelle/samboer 294	☐	
Barn	☐	
Andre	☐	

Hvor mange gode venner har du som du kan snakke fortrolig med og gi deg hjelp når du trenger det? ..... 297 ____ gode venner
Tell ikke med dem du bor sammen med, men ta med andre slektninger!

	Ja	Nei
Føler du at du har nok gode venner? 299	☐	☐

Føler du at du hører med i et fellesskap (gruppe av mennesker) som stoler på hverandre og føler forpliktelse overfor hverandre (f.eks. i politisk parti, religiøs gruppe, slekt, naboskap, arbeidsplass eller organisasjon)?

- Sterk tilhørighet ..... 300 ☐ 1
- Noe tilhørighet ☐ 2
- Usikkert ☐ 3
- Liten eller ingen tilhørighet ☐ 4

Hvor ofte tar du vanligvis del i foreningsvirksomhet som f.eks. syklubb, idrettslag, politiske lag, religiøse eller andre foreninger?

- Aldri, eller noen få ganger i året ..... 301 ☐ 1
- 1-2 ganger i måneden ..... ☐ 2
- Omtrent en gang i uken ..... ☐ 3
- Mer enn en gang i uken ..... ☐ 4

KOSTVANER

Antall

Hvor mange måltider spiser du vanligvis daglig (middag og brødmåltid)? ..... 302 _____

Hvor mange ganger i uken spiser du varm middag? ..... 304 _____

Hva slags type brød (kjøpt eller hjemmebakt) spiser du vanligvis?
Sett ett eller to kryss.

	Loff	Fint brød	Knekp-brød	Grov-brød	Knekke-brød
Brødtypen ligner mest på:	☐ 306	☐	☐	☐	☐ 310

Hva slags fett blir til vanligvis brukt til *matlaging* (ikke på brødet) i din husholdning?

- Meierismør ..... 311 ☐
- Hard margarin ..... ☐
- Bløt (Soft) margarin ..... ☐
- Smør/margarin blanding ..... ☐
- Oljer ..... 315 ☐

Hvor mye (i antall glass, poteter eller brødskiver) spiser/drikker du vanligvis daglig av følgende matvarer?
Kryss av for alle matvarene.

	Ingen	Mindre enn 1	1-2	3 og mer
Melk alle sorter (glass) 316	☐	☐	☐	☐
Appelsinjuice (glass)	☐	☐	☐	☐
Poteter	☐	☐	☐	☐
Brødskiver totalt (inkl. knekkebrød) ...	☐	☐	☐	☐
Brødskiver med				
– fiskepålegg (f.eks. makrell i tomat)	☐	☐	☐	☐
– gulost	☐	☐	☐	☐
– kaviar 322	☐ 1	☐ 2	☐ 3	☐ 4

Hvor mange ganger i uka spiser du vanligvis følgende matvarer?
Kryss av for alle matvarene.

	Aldri	Sjeldnere enn 1	1	2 og mer
Yoghurt 323	☐	☐	☐	☐
Kokt eller stekt egg	☐	☐	☐	☐
Frokostblanding/havregryn o.l.	☐	☐	☐	☐
Middag med				
– rent kjøtt	☐	☐	☐	☐
– fett fisk (f.eks. laks/uer)	☐	☐	☐	☐
– mager fisk (f.eks. torsk) 328	☐	☐	☐	☐
– grønnsaker (rå eller kokte)	☐	☐	☐	☐
Gulrøtter (rå eller kokte)	☐	☐	☐	☐
Blomkål/kål/brokkoli	☐	☐	☐	☐
Epler/pærer	☐	☐	☐	☐
Appelsiner, mandariner o.l. 333	☐ 1	☐ 2	☐ 3	☐ 4

TRIVSEL

Hvordan trives du med å bli gammel - alt i alt?

- Godt ..... 334 ☐ 1
- Ganske bra ..... ☐ 2
- Opp og ned ..... ☐ 3
- Dårlig ..... ☐ 4

Hvordan ser du på livet fremover?

- Lyst ..... 335 ☐ 1
- Ikke så verst ..... ☐ 2
- Noksa bekymret ..... ☐ 3
- Mørkt ..... ☐ 4

BESVARES BARE AV KVINNER

MENSTRUASJON

Hvor gammel var du da du fikk menstruasjon første gang? ..... 336 _____ år

Hvor gammel var du da menstruasjonen sluttet? ..... 338 _____ år

SVANGERSKAP

Hvor mange barn har du født? ..... 340 _____ barn

Hvis du har født, fyll ut for hvert barn barnets fødselsår og omtrent antall måneder du ammet barnet.
Hvis du har født mer enn 6 barn, noter fødselsår og antall måneder med amming for dem nederst på siden.

Barn:	Fødselsår:	Antall måneder med amming:
1	342 _____	_____
2	346 _____	_____
3	_____	_____
4	_____	_____
5	358 _____	_____
6	_____	_____

Har du i forbindelse med svangerskap hatt for høyt blodtrykk og/eller eggehvite (protein) i urinen? ..... 366 Ja ☐ Nei ☐

Hvis "Ja", i hvilket svangerskap?

	Svangerskap Første	Senere
For høyt blodtrykk 367	☐	☐
Eggehvite i urinen 369	☐	☐

ØSTROGEN-MEDISIN

Bruker du, eller har du brukt, østrogen-medisin?

	Nå	Før	Aldri
Tabletter eller plaster 371	☐	☐	☐
Krem eller stikkpiller 372	☐	☐	☐

Hvis du bruker østrogen, hvilket merke bruker du nå?

..... 373

Dine kommentarer:

Takk for hjelpen! Husk å postlegge skjemaet idag!

English translation of the second questionnaire used in the health survey in Tromsø 1994/95 for subjects 70 years or older.

Based on translations by Kevin McCafferty and Anne Clancy.

TROMSØ HEALTH SURVEY for the over 70s

The main aim of the Tromsø survey is to improve our knowledge of heart and circulatory conditions in order to aid prevention. The survey is also intended to improve our knowledge of cancer and other general conditions, such as allergies, muscle pains and nervous conditions. The ultimate aim is to gain an overview of the general health of the elderly population. We would therefore like you to answer the questions below.

This form is part of the Health Survey, which has been approved by the Norwegian Data Inspectorate and the Regional Board of Research Ethics. The answers will only be used for research purposes and will be treated in strict confidence. The information you give us may later be stored along with information from other public health registers in accordance with the rules laid down by the Data Inspectorate and the Regional Board of Research Ethics.

If you are unsure about what to answer, tick the box that you feel fits best.

The completed form should be sent to us in the enclosed pre-paid envelope.

Thank you in advance for helping us.

Yours sincerely,

Faculty of Medicine
University of Tromsø

National Health
Screening Service

If you do not wish to answer the questionnaire, tick the box below and return the form. Then you will not receive reminders.

I do not wish to answer the questionnaire. ☐

Date for filling in this form: Day/Month/Year

CHILDHOOD/YOUTH

What Norwegian municipality did you live in at the age of 1 year?

If you did not live in Norway, give country instead of municipality.

How was your family's financial situation while you were growing up?

- Very good ☐
- Good ☐
- Difficult ☐
- Very difficult ☐

How old were your parents when they died?

- Mother _____ years
- Father _____ years

HOME

Who do you live with?
Tick one box for each item and give the number of persons.

	YES	NO	Number
Spouse/partner	☐	☐	_____
Other persons over 18 years		☐	☐ _____
Persons under 18 years		☐	☐ _____

What type of home do you live in?

- Villa/detached house ☐
- Farm ☐
- Apartment/flat in block/terrace ☐
- Terraced/semi-detached house ☐
- Other ☐

How long have you lived in your present home? ____years

Is your home adapted to your needs? YES ☐ NO ☐
If "No", do you have problems with:

- Space ☐
- Variable temperature/too cold/too warm ☐
- Stairs ☐
- Toilet ☐
- Bath/shower ☐
- Maintenance ☐
- Other (please specify) ☐

Would you like to move into a retirement home?
YES ☐ NO ☐

PREVIOUS WORK AND FINANCIAL SITUATION

Which statement best describes the type of work you did for the last 5-10 years before you retired?

- I was mainly seated while working (*e.g., desk/assembly work*) ☐
- My work required a lot of walking (*e.g., shop assistant, housewife, teaching*) ☐
- My work required a lot of walking and lifting (*e.g., postman, nurse, construction work*) ☐
- I did heavy physical work (*e.g., forestry, heavy agricultural work, heavy construction work*) ☐

Did you do any of the following jobs (full- or part-time)?

Tick one box only for each item.	YES	NO
Driver	☐	☐
Farmer	☐	☐
Fisherman	☐	☐

How old were you when you retired? _____ years

What kind of pension do you have?

- Basic state pension ☐
- Additional pension ☐

How is your current financial situation?

- Very good ☐
- Good ☐
- Difficult ☐
- Very difficult ☐

HEALTH AND ILLNESS

Has your state of health changed in the last year?

- Yes, it has got worse ☐
- No, unchanged ☐
- Yes, it has got better ☐

How do you feel your health is now compared to others of your age?

- Much worse ☐
- A little worse ☐
- About the same ☐
- A little better ☐
- Much better ☐

YOUR OWN ILLNESSES

Have you ever had:

Tick one box only for each item. Give your age at the time. If you have had the condition several times, how old were you last time?

	YES	NO	AGE
Hip fracture	☐	☐	_____
Wrist / forearm fracture	☐	☐	_____
Whiplash	☐	☐	_____
Injury requiring hospital admission	☐	☐	_____
Stomach ulcer	☐	☐	_____
Duodenal ulcer	☐	☐	_____
Stomach/duodenal ulcer operation	☐	☐	_____
Throat/neck surgery	☐	☐	_____

Have you ever had, or do you still have:

Tick one box only for each item.	YES	NO
Cancer	☐	☐
Epilepsy	☐	☐
Migraine	☐	☐
Chronic bronchitis	☐	☐
Psoriasis	☐	☐
Osteoporosis	☐	☐
Fibromyalgia/fibrositis/ chronic pain syndrom	☐	☐
Psychological problems for which you have sought help	☐	☐
Thyroid disease	☐	☐
Liver disease	☐	☐
Thyroid disease	☐	☐
Liver disease	☐	☐
Recurrent urinary incontinence	☐	☐
Glaucoma	☐	☐
Cataract	☐	☐
Arthrosis (osteoarthritis)	☐	☐
Rheumatoid arthritis	☐	☐
Kidney stone	☐	☐
Appendectomy	☐	☐
Allergy and hypersensitivity		
Atopic eczema (e.g., childhood eczema)	☐	☐
Hand eczema	☐	☐
Hay fever	☐	☐
Food allergy	☐	☐
Other hypersensitivity (not allergy)	☐	☐

How many times have you had a cold, influenza (flue), diarrhea/vomiting, or similar in the last six months?

_____ times

Have you had any of these in the last two weeks?

YES ☐ NO ☐

ILLNESS IN THE FAMILY

Tick off relatives who have, or have ever had, any of the following conditions:

Tick "None" for conditions which none of your relatives have had.

	Mother	Father	Brother	Sister	Child	None
Stroke or brain haemorrhage	☐	☐	☐	☐	☐	☐
Myocardial infarction before age 60	☐	☐	☐	☐	☐	☐
Cancer	☐	☐	☐	☐	☐	☐
Hypertension	☐	☐	☐	☐	☐	☐
Asthma	☐	☐	☐	☐	☐	☐
Osteoporosis	☐	☐	☐	☐	☐	☐
Arthrosis (osteoarthritis)	☐	☐	☐	☐	☐	☐
Psychological problems	☐	☐	☐	☐	☐	☐
Dementia	☐	☐	☐	☐	☐	☐
Diabetes	☐	☐	☐	☐	☐	☐
-age when they got diabetes	___	___	___	___	___	___

SYMPTOMS

Do you cough daily for periods of the year?	YES	NO
	☐	☐
If "Yes":		
Is your cough productive?	☐	☐
Have you had this kind of cough for as long as 3 months in each of the last two years?	☐	☐
Have you had periods of wheezing in your chest?	☐	☐
If "Yes", has this occurred:		
Tick one box only for each item.		
At night	☐	☐
In connection with respiratory infections	☐	☐
In connection with physical exertion	☐	☐
In connection with very cold weather	☐	☐
Have you noticed sudden changes in your pulse or heart rhythm in the last year?	☐	☐
Have you lost weight in the last year?	☐	☐
If "Yes":		
How many kilograms?	_____ kg	

How often do you suffer from sleeplessness?

- Never, or just a few times a year ☐
- 1-2 times a month ☐
- Approximately once a week ☐
- More than once a week ☐

If you suffer from periods of sleeplessness, what times of the year does it affect you most?

- No particular time of year ☐
- Especially during the 'dark winter months' ☐
- Especially during the midnight sun period ☐
- Especially in spring and autumn ☐

Do you usually take a nap during the day? YES ☐ NO ☐

Do you feel that you normally get enough sleep? YES ☐ NO ☐

	No	A little	A lot
Do you suffer from:	☐	☐	☐
Dizziness	☐	☐	☐
Poor memory	☐	☐	☐
Lack of energy	☐	☐	☐
Constipation	☐	☐	☐

Does the thought of getting a serious illness ever worry you?

- Not at all ☐
- Only a little ☐
- Some ☐
- Very much ☐

BODILY FUNCTIONS

Can you manage the following everyday activities on your own without help from others?

	Yes	With some help	No
Walking indoors on one level	☐	☐	☐
Walking up/down stairs	☐	☐	☐
Walking outdoors	☐	☐	☐
Walking approx. 500 metres	☐	☐	☐
Going to the toilet	☐	☐	☐
Washing yourself	☐	☐	☐
Taking a bath/shower	☐	☐	☐
Dressing and undressing	☐	☐	☐
Getting in and out of bed	☐	☐	☐
Eating meals	☐	☐	☐
Cooking ☐	☐	☐	
Doing light housework (e.g., washing up)	☐	☐	☐
Doing heavier housework (e.g., cleaning floors)	☐	☐	☐
Going shopping	☐	☐	☐
Taking the bus	☐	☐	☐

	Yes	With difficulty	No
Can you hear normal speech (if necessary with a hearing aid)?	☐	☐	☐
Can you read (if necessary with glasses)?	☐	☐	☐

Are you dependent on any of the following aids?

	Yes	No
Walking stick	☐	☐
Crutches	☐	☐
Walking frame/Zimmer frame	☐	☐
Wheelchair	☐	☐
Hearing aid	☐	☐
Safety alarm device	☐	☐

USE OF HEALTH SERVICES

How many visits have you made during the past year due to your own health or illness:

*Tick **0** if you have **not** had such contact*

	Number of times the past year
To a general practitioner (GP)/ emergency GP	____
Psychologist or psychiatrist	____
Other medical specialist (not at a hospital)	____
Hospital out-patient clinic	____
Hospital admission	____
Physiotherapist	____
Chiropractor	____
Acupuncturist	____
Dentist	____
Chiropodist	____
Alternative medical practitioner (homoeopath, foot zone therapist, etc.)	____
Healer, Faith healer, clairvoyant	____

	Yes	No
Do you have domestic help?		
Private	☐	☐
Municipal	☐	☐
Do you receive services from the district nurse?	☐	☐

Are you pleased with the health care and home assistance services your municipality supplies?

	Yes	No	Don't know
Assigned family GP	☐	☐	☐
District nurse	☐	☐	☐
Home assistance	☐	☐	☐

Do you feel confident that you can receive the health care and home assistance you require if you need it?

- Confident ☐
- Not confident ☐
- Very unsure ☐
- Don't know ☐

MEDICATION AND DIETARY SUPPLEMENTS

Have you for any length of time in the past year used any of the following medicines every day or almost daily?
Indicate how many months you used them for.
*Write **0** for items you have **not** used.*

Medication:

Painkillers	_____ mths
Sleeping pills	_____ mths
Tranquillizers	_____ mths
Antidepressants	_____ mths
Allergy drugs	_____ mths
Asthma drugs	_____ mths
Heart medicine (not blood pressure)	_____ mths
Insulin	_____ mths
Diabetes tablets	_____ mths
Thyroxin tablets (for metabolic disorder)	_____ mths
Cortisone tablets	_____ mths
Remedies for constipation	_____ mths

Dietary supplements:

Iron tablets	_____ mths
Vitamin D supplement	_____ mths
Other vitamin supplements	_____ mths
Calcium tablets or bonemeal	_____ mths
Cod liver oil or fish oil capsules	_____ mths

FAMILY AND FRIENDS

Do you have close relatives who can give you help and support when you need it? Yes ☐ No ☐

If "Yes", who can give you help?

- Spouse/partner ☐
- Children ☐
- Others ☐

How many good friends do you have whom you can talk confidentially with and who give you help when you need it?

_____good friends

Do not count people you live with, but do include other relatives!

Do you feel you have enough good friends? Yes ☐ No ☐

Do you feel that you belong to a community or group of people who can depend on each other and who feel committed to each other (e.g., a political party, religious group, relatives, neighbours, work place, or organisation)?

- Strong sense of belonging ☐
- Some sense of belonging ☐
- Not sure ☐
- Little or no sense of belonging ☐

How often do you normally take part in organised gatherings, e.g., sewing circles, sports clubs, political meetings, religious or other associations?

- Never, or just a few times a year ☐
- 1-2 times a month ☐
- Approximately once a week ☐
- More than once a week ☐

DIET

How many meals a day do you normally eat (dinner and smaller meals)? _____ Number

How many times a week do you eat a hot dinner? _____ Number

What kind of bread (bought or home-made) do you usually eat? *Tick one or two boxes!*

The bread I eat is most similar to

- White bread ☐
- Light textured brown bread ☐
- Ordinary brown bread ☐
- Coarse brown bread ☐
- Crisp bread ☐

What kind of fat is normally used in **cooking** (not on the bread) in your home?

- Creamery butter ☐
- Hard margarine ☐
- Soft margarine ☐
- Butter/margarine blend ☐
- Oils ☐

How much (in **number** of glasses, cups, potatoes or slices) do you usually eat or drink **daily** of the following foodstuffs? *Tick one box for **each** foodstuff.*

	0	Less than 1	1-2	3-4	5-6	6-
Milk of all types (glasses)	☐	☐	☐	☐	☐	☐
Orange juice (glasses)	☐	☐	☐	☐	☐	☐
Potatoes	☐	☐	☐	☐	☐	☐
Slices of bread in total (incl. crispbread)	☐	☐	☐	☐	☐	☐
Slices of bread with fish (e.g., mackerel in tomato sauce)	☐	☐	☐	☐	☐	☐
- cheese (e.g., Norwegia)	☐	☐	☐	☐	☐	☐
- smoked cod caviar	☐	☐	☐	☐	☐	☐

How many **times per week** do you normally eat the following foodstuffs? *Tick a box for **all** foodstuffs listed.*

	Never	Less than 1	1	2-3	4-5	Roughly every day
Yoghurt	☐	☐	☐	☐	☐	☐
Boiled or fried egg	☐	☐	☐	☐	☐	☐
Breakfast cereal/ oat meal, etc.	☐	☐	☐	☐	☐	☐
For dinner - meat	☐	☐	☐	☐	☐	☐
- fat fish (e.g., salmon/ redfish)	☐	☐	☐	☐	☐	☐
- lean fish (e.g., cod)						
- vegetables (raw or cooked)	☐	☐	☐	☐	☐	☐
Carrots (raw or cooked)	☐	☐	☐	☐	☐	☐
Cauliflower/cabbage/broccoli	☐	☐	☐	☐	☐	☐
Apples/pears	☐	☐	☐	☐	☐	☐
Oranges, mandarines, etc.	☐	☐	☐	☐	☐	☐

WELL BEING

How content do you generally feel with growing old?

- Good ☐
- Quite good ☐
- Up and down ☐
- Bad ☐

What is your view of the future?

- Bright ☐
- Not too bad ☐
- Quite worried ☐
- Dark ☐

TO BE ANSWERED BY WOMEN ONLY

MENSTRUATION

How old were you when you had your first menstruation? _____ years

How old were you when you stopped having menstruations? _____ years

PREGNANCY

How many children have you given birth to? _____children

If you have given birth, fill out for each child the year of birth and approximately how many months you breastfed the child. If you have given birth to more than 6 children, note their birthyear and number of months you breastfed at the space provided below for comments.

Child:	Year of birth:	Number of months breastfed:
1	_____	_____ months
2	_____	_____ months
3	_____	_____ months
4	_____	_____ months
5	_____	_____ months
6	_____	_____ months

During pregnancy, have you had high blood pressure and/or proteinuria? Yes ☐ No ☐

If "Yes", during which pregnancy?

	Pregnancy First	Later
High blood pressure	☐	☐
Proteinuria	☐	☐

OESTROGEN

Do you, or have you ever used oestrogen:

	Now	Used to	Never
Tablets or patches	☐	☐	☐
Cream or suppositories	☐	☐	☐

If you use oestrogen, what brand do you currently use?

_____

Your comments:

Thank you for helping us! Remember to post the form today! **Tromsø Health Survey**

ISM SKRIFTSERIE - FØR UTGITT:

1. Bidrag til belysning av medisinske og sosiale forhold i Finnmark fylke, med særlig vekt på forholdene blant finskættede i Sør-Varanger kommune.
Av Anders Forsdahl, 1976. (nytt opplag 1990)

2. Sunnhetstilstanden, hygieniske og sosiale forhold i Sør-Varanger kommune 1869-1975 belyst ved medisinal-beretningene.
Av Anders Forsdahl, 1977.

3. Hjerte-karundersøkelsen i Finnmark - et eksempel på en populasjonsundersøkelse rettet mot cardiovasculære sykdommer. Beskrivelse og analyse av etterundersøkelsesgruppen.
Av Jan-Ivar Kvamme og Trond Haider, 1979.

4. D. The Tromsø Heart Study: Population studies of coronary risk factors with special emphasis on high density lipoprotein and the family occurrence of myocardial infarction.
Av Olav Helge Førde og Dag Steinar Thelle, 1979.

5. D. Reformer i distriktshelsetjenesten III: Hypertensjon i distriktshelsetjenesten.
Av Jan-Ivar Kvamme, 1980.

6. Til professor Knut Westlund på hans 60-års dag, 1983.

7.* Blodtrykksovervåkning og blodtrykksmåling.
Av Jan-Ivar Kvamme, Bernt Nesje og Anders Forsdahl, 1983.

8.* Merkesteiner i norsk medisin reist av allmennpraktikere - og enkelte utdrag av medisinalberetninger av kulturhistorisk verdi.
Av Anders Forsdahl, 1984.

9. "Balsfjordsystemet." EDB-basert journal, arkiv og statistikksystem for primærhelsetjenesten.
Av Toralf Hasvold, 1984.

10. D. Tvunget psykisk helsevern i Norge. Rettsikkerheten ved slikt helsevern med særlig vurdering av kontrollkommisjonsordningen.
Av Georg Høyer, 1986.

11. D. The use of self-administered questionnaires about food habits. Relationships with risk factors for coronary heart disease and associations between coffee drinking and mortality and cancer incidence.
Av Bjarne Koster Jacobsen, 1988.

12.* Helse og ulikhet. Vi trenger et handlingsprogram for Finnmark.
Av Anders Forsdahl, Atle Svendal, Aslak Syse og Dag Thelle, 1989.

13. D. Health education and self-care in dentistry - surveys and interventions.
Av Anne Johanne Søgaard, 1989.

14. Helsekontroller i praksis. Erfaringer fra prosjektet helsekontroller i Troms 1983-1985.
Av Harald Siem og Arild Johansen, 1989.

15. Til Anders Forsdahls 60-års dag, 1990.

16. D. Diagnosis of cancer in general practice. A study of delay problems and warning signals of cancer, with implications for public cancer information and for cancer diagnostic strategies in general practice.
Av Knut Holtedahl, 1991.

17. D. The Tromsø Survey. The family intervention study. Feasibility of using a family approach to intervention on coronary heart disease. The effect of lifestyle intervention of coronary risk factors.
Av Synnøve Fønnebø Knutsen, 1991.

18. Helhetsforståelse og kommunikasjon. Filosofi for klinikere.
Av Åge Wifstad, 1991.

19. D. Factors affecting self-evaluated general health status - and the use of professional health care services.
Av Knut Fylkesnes, 1991.

20. D. Serum gamma-glutamyltransferase: Population determinants and diagnostic characteristics in relation to intervention on risk drinkers.
Av Odd Nilssen, 1992.

21. D. The Healthy Faith. Pregnancy outcome, risk of disease, cancer morbidity and mortality in Norwegian Seventh-Day-Adventists.
Av Vinjar Fønnebø, 1992.

22. D. Aspects of breast and cervical cancer screening.
Av Inger Torhild Gram, 1992.

23. D. Population studies on dyspepsia and peptic ulcer disease: Occurrence, aetiology, and diagnosis. From The Tromsø Heart Study and The Sørreisa Gastrointestinal Disorder Studie.
Av Roar Johnsen, 1992.

24. D. Diagnosis of pneumonia in adults in general practice.
Av Hasse Melbye, 1992.

25. D. Relationship between hemodynamics and blood lipids in population surveys, and effects of n-3 fatty acids.
Av Kaare Bønaa, 1992.

26. D. Risk factors for, and 13-year mortality from cardiovascular disease by socioeconomic status. A study of 44690 men and 17540 women, ages 40-49. **Av Hanne Thürmer, 1993.**

27. Utdrag av medisinalberetninger fra Sulitjelma 1891-1990. **Av Anders Forsdahl, 1993.**

28. Helse, livsstil og levekår i Finnmark. Resultater fra Hjerte-karundersøkelsen i 1987-88. Finnmark III. **Av Knut Westlund og Anne Johanne Søgaard, 1993.**

29. D. Patterns and predictors of drug use. A pharmacoepidemiologic study, linking the analgesic drug prescriptions to a population health survey in Tromsø, Norway. **Av Anne Elise Eggen, 1994.**

30. D. ECG in health and disease. ECG findings in relation to CHD risk factors, constitutional variables and 16-year mortality in 2990 asymptomatic Oslo men aged 40-49 years in 1972. **Av Per G. Lund-Larsen, 1994.**

31. D. Arrhythmia, electrocardiographic signs, and physical activity in relation to coronary heart risk factors and disease. The Tromsø Study. **Av Maja-Lisa Løchen, 1995.**

32. D. The Military service: mental distress and changes in health behaviours among Norwegian army conscript. **Av Edvin Schei, 1995.**

33. D. The Harstad injury prevention study: Hospital-based injury recording and community-based intervention. **Av Børge Ytterstad, 1995.**

34.* D. Vilkår for begrepsdannelse og praksis i psykiatri. En filosofisk undersøkelse. **Av Åge Wifstad, 1996.** (utgitt Tano Aschehoug forlag 1997)

35. Dialog og refleksjon. Festskrift til professor Tom Andersen på hans 60-års dag, 1996.

36. D. Factors affecting doctors´ decision making. **Av Ivar Sønbø Kristiansen, 1996.**

37. D. The Sørreisa gastrointestinal disorder study. Dyspepsia, peptic ulcer and endoscopic findings in a population. **Av Bjørn Bernersen, 1996.**

38. D. Headache and neck or shoulder pain. An analysis of musculoskeletal problems in three comprehensive population studies in Northern Norway. **Av Toralf Hasvold, 1996.**

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