Linear age-course effects on the associations between body mass index, triglycerides, and female breast and male liver cancer risk: An internal replication study of 800,000 individuals

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What´s new?

We systematically investigated interactions between attained age and metabolic factors in relation to cancer risk. For female breast cancer and liver cancer among men, elevated BMI and triglycerides were associated with decreased risks in younger ages and increased risks in older ages with linearly changing patterns of associations across age. The findings for liver cancer are novel, and the linearly changing associations across age for breast cancer suggest that other age-related factors than menopause *per se* modify the associations between metabolic factors and breast cancer risk.

Abbreviations:

40-y: the 40-year programme

BMI: body mass index

HR: hazard ratio

ICD: International Classification of Diseases

Me-Can: Metabolic syndrome and Cancer project

MPP: Malmö Preventive Project

NCS: Norwegian Counties Study

PH: proportional hazards

SD: standard deviation

VIP: Västerbotten Intervention Project

VHM&PP: Vorarlberg Health Monitoring and Prevention Programme

Z score: standardized exposures with a mean value of zero and a standard deviation of one

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Abstract

Apart from the consistently observed differential association between obesity and breast cancer risk by menopausal status, the associations between obesity and other metabolic imbalances with risks of cancers have not been systematically investigated across the age-course. We created two random 50-50% cohorts from six European cohorts comprising 813,927 individuals. In the "discovery cohort", we used Cox regression with attained age as time-scale and tested interactions between body mass index (BMI), blood pressure, plasma glucose, triglycerides and cholesterol, and attained age in relation to cancer risk. Results with a P-value below 0.05 were additionally tested in the "replication cohort" where a replicated result was considered evidence of a linear interaction with attained age. These findings were investigated by flexible parametric survival models for any age-plateaus in their shape of associations with cancer risk across age. Consistent with other studies, BMI was negatively related to breast cancer risk (n cases=11,723) among younger (premenopausal) women. However, the association remained negative for several years after menopause and, although gradually weakening over age, the association became positive only at 62 years of age. This linear and positive age-interaction was also found for triglycerides and breast cancer, and for BMI and triglycerides in relation to liver cancer among men (n cases=444). These findings are unlikely to be due to chance owing to the replication. The linear age-interactions in breast cancer may suggest an influence by other age-related factors than menopause; however, further investigation of age-related effect modifiers in both breast and liver cancer is needed.

Introduction

Obesity is an established risk factor for a range of different cancers, including cancer of the oesophagus, colorectum, liver, kidney, multiple myeloma, and endometrium.[1] The shape of the associations between obesity and cancers have been investigated in detail across the exposure range allowing for non-linear associations,[2, 3] but potential differential associations across the age-course have not been specifically investigated. Menopause occurs at around 50 years of age, and female hormone-related cancers, especially breast cancer, have commonly been investigated separately by menopausal status. Obesity has been consistently related to a decreased breast cancer risk before menopause and an increased risk after menopause.[2-4] In our previous study of breast cancer, we found a similar pattern of association also for plasma triglycerides and risk at pre- and postmenopausal ages.[5]

Most cohort studies of metabolic factors and cancer risk have used Cox proportional hazards regression, which assumes a constant multiplicative effect of the risk factor over time, i.e. proportional hazards (PH). Violation of PH when using age as time-scale, which is commonly used in studies of cancer risk, indicates that the hazard ratio (HR) for the violated factor varies by attained age. However, the reporting of PH assumption tests and of age-dependent results in case of violation of the main exposure have been lacking in survival studies overall,[6, 7] and to our knowledge, no previous study has systematically assessed whether risk for cancer at different sites due to metabolic factors vary across attained age.

The aim of this study was to systematically investigate whether the associations between metabolic factors (body mass index [BMI], blood pressure, and plasma glucose, triglycerides and total cholesterol) and risks for a range of cancer forms vary across attained age in a large European cohort.

Methods

Population

We used data from the Metabolic syndrome and Cancer project (Me-Can) 2.0, which pools six cohorts with recruitment periods as follows: in Norway, Oslo I (1972-73), the Norwegian Counties Study (NCS, 1974-88) and the 40-year programme (40-y, 1985-99); in Sweden, the Västerbotten Intervention Project (VIP, 1985-ongoing) and the Malmö Preventive Project (MPP, 1974-92); and in Austria, the Vorarlberg Health Monitoring and Prevention Programme (VHM&PP, 1985-2005). The project is a continuation of Me-Can 1.0, which has been extensively described previously with regards to the purpose, population, and measurements of included cohorts.[8] In contrast to Me-Can 1.0 on which Me-Can publications in 2009-2015 were based,[8] Me-Can 2.0 does not include Cohort of Norway (CONOR), but it includes the full Oslo I, NCS, and 40-y cohorts. Me-Can 2.0 also includes additional individuals and health observations in the VIP between the years 2006 and 2014 and in the VHM&PP between the years 2003 and 2005. Questionnaire information on tobacco use and number of years smoking were also added to the Me-Can database, as well as other variables that, however, are not available in all cohorts. Measurement methods of

metabolic factors have been previously described in detail for all cohorts,[8] which in the VIP is valid for measurements performed before Sept 1, 2009, after which, however, the measurement methods for blood pressure and plasma lipids changed. Before that date, blood pressure was measured in supine position, and was thereafter measured in sitting position. Plasma cholesterol and triglycerides were measured on a Reflotron bench-top analyzer at the examining health care centre before Sept 1, 2009, and were thereafter measured at the Clinical Chemistry department at the nearest hospital using standard enzymatic methods. For research purposes, formula have been developed to transform levels between new and old measurement methods of blood pressure and plasma lipids, which we used in the present study. These formula were based on individuals with measurements using both old and new measurements methods (n=838 for triglycerides, 1,197 for cholesterol, and 648 for blood pressure). Triglyceride and cholesterol levels measured on Sept 1, 2009 onwards were converted to old measurement levels using the formulas: 0.177 + (0.932 × triglyceride level) and 0.170 + (0.939 × cholesterol level). Formula for blood pressure were age- and sex-specific (**Supplementary Table 1**). The study was approved by ethical committees in Norway, Sweden, and Austria.

Follow-up

Individuals in the cohorts were linked to their respective national cancer register, or regional cancer register for the VHM&PP, and national cause of death register for information on cancer diagnoses and death causes. The Norwegian and Swedish cohorts were additionally

linked to their respective national total population register for information on emigration. Follow-up included the year 2012 in Norway and 2014 in Sweden and Austria, which was an extension of seven (Austria), eight (Sweden), or eleven (Norway) years of follow-up for cancer diagnoses compared to in Me-Can 1.0. We used the 7th revision of the International Classification of Diseases (ICD-7) to categorise cancers. All malignant cancers, except basalioma, were included, as well as blood cancers (ICD-7 200-209) of uncertain or unknown behavior. In situ cancers were excluded except urothelial carcinoma in situ, which were included because they frequently progress to higher stage and muscle-invasive tumours.[9, 10]

Selection criteria

Amongst 843,522 individuals with 1,557,855 health observations in Me-Can 2.0, we selected 813,927 individuals with one observation each (**Figure 1**). The largest number of exclusions were performed due to a prevalent cancer (26,158 observations), non-fasting status during the first three years in the VHM&PP (77,031 observations), missing data on fasting status (29,842 observations), and the selection of one observation amongst individuals with more than one observation, in which case we selected the first health observation with complete data on metabolic factors and with a fasting state if that was available (609,143 observations). The relatively large number of observations with fasting status missing were primarily a result of not requesting information on fasting status. Out of the 302 individuals excluded due to an older age than 100 years at the end of follow-up, 300 originated from the

VHM&PP in which missing emigration data might have caused a minor loss to follow-up and thus immortalised individuals in the dataset.

Statistical analysis

We investigated metabolic factors (BMI, blood pressure, glucose, cholesterol, and triglycerides) transformed to standardized exposures (Z score) with a mean value of zero and a standard deviation (SD) of one using the formula: (metabolic level-mean)/SD within subgroups of sex, cohort and fasting time (<4h, 4-7 h, and ≥8 h), except blood pressure that was standardized by sex and cohort only. Triglycerides and glucose had skewed distributions so their values were log-transformed before standardisation. Blood pressure was investigated as mid-blood pressure, i.e. the average of systolic plus diastolic blood pressure.[11] In all analyses, we excluded individuals with values more extreme than ±3 SDs of the main exposure (maximum 1.3% of individuals).

We investigated linear interactions between metabolic factors and attained age, and cancer risk for cancer forms with at least 400 incident cases separately among men or women. Individuals in the study were followed from one year after baseline in order to reduce the risk for reverse causation, until the date of first cancer diagnosis, emigration, death, or end of follow-up, whichever occurred first. For cancers of the liver, pancreas, and kidney (renal cell), which are both subject to longer latency periods before clinical diagnosis and for which an existing tumour might affect metabolic levels, we additionally explored the impact of excluding the first six years of follow-up. We used Cox regression with attained age

as time-scale, and included a time-dependent product term of attained age and a metabolic factor as interaction variable in the analysis to assess the linear exposure-age interaction.[12] We adjusted for continuous baseline age and seven categories of smoking (never-smoker, ex-smoker, smokers in tertiles of pack-years, smokers with pack-years missing [7.0% of smokers], and missing smoking status [0.3%]). Our recent study in the VIP and the VHM&PP of the relationship between the five metabolic factors investigated in the present study showed that BMI was overall modestly correlated with the other metabolic factors and, out of the metabolic factors investigated, was the strongest determinant for future change of another metabolic factor.[13] Therefore, in the present study, analyses of metabolic exposures other than BMI were additionally adjusted for BMI continuous Z score. The model was stratified by cohort and birth cohort (before 1923, 1923-30, 1931-38, 1939-46, 1947-54, 1955 and later), and additionally by quintiles of baseline age or by smoking categories if any of these factors interacted linearly with attained age (P-value<0.05 in **Supplementary Table 2**).

To avoid false positive interactions, metabolic exposure-age interactions were evaluated using an internal replication approach. We split the population into two random 50-50% datasets, one "discovery cohort" and one "replication cohort", and tested one exposure-age interaction at a time in the discovery cohort. Results with P-values below 0.05 were additionally tested in the replication cohort, out of which results with a P-value below 0.05 were considered evidence of an interaction. For confirmed findings in the replication cohort, we further investigated the findings in the full study population. First, we calculated

the linear age-interaction estimates and derived HRs at specific ages and the age at which the HRs crossed 1.0. Second, we plotted HRs over attained age in the full study population by using cubic splines of the studied time-interaction variable in flexible parametric survival models.[14] This model is similar to the Cox model but the baseline hazard function is estimated parametrically using cubic splines. We used the same adjustments as in the Cox models, but additionally adjusted for cohort and birth cohort as the flexible parametric survival model software does not allow for stratification.

In relation to the findings for BMI and triglycerides and breast cancer among women and liver cancer among men, we performed a number of posthoc and sensitivity analyses by use of flexible parametric survival models. We calculated age-dependent HRs in relation to levels of blood pressure, glucose and cholesterol in order to see whether the patterns of associations for these factors would be similar to those of BMI and triglycerides. We also tested flexible parametric survival models in relation to breast cancer using various degrees of freedom for the cubic splines of the age-dependent associations for BMI and triglycerides. The results showed largely similar patterns but, naturally, with larger flexibility of the spline with increasing degrees of freedom (**Supplementary Figure 1**). The Akaike Information Critera and Bayesian Information Criteria were used to assess the best fit of the tested models,[14] and the lowest values were found for the model with one degree of freedom for both breast and liver cancer (**Supplementary Table 3**). This indicates that the age-dependent associations can be expressed as a linear function of log time, however, the

other models had very similar fitted values. In all models, four degrees of freedom were used to model the spline of the baseline hazard.

Posthoc and sensitivity analyses of age-dependent HRs for BMI and triglycerides were also performed for a) liver cancer of men and women combined, b) breast cancer in subgroups of age at measurement: <40 years, 40-49 years and ≥50 years, c) breast cancer with adjustments for age at first delivery (no child, <20 years, 20-24 years, 25-29 years, 30-34 years, and ≥35 years) and number of deliveries (0, 1, 2, 3, and ≥4) based on nation-wide registry data in Sweden and Norway (the multi-generation register and the national tax registry), and d) breast and liver cancer with adjustment for plasma gamma glutamyl-transferase as a moderate and objective marker for (heavy) alcohol consumption,[15-17] but also for non-alcoholic fatty liver disease. In the latter analysis, we adjusted for quartiles of gamma glutamyl-transferase, in the MPP using separate cut-points by fasting time, and in the VHM&PP using separate cut-points by measurements performed before or after Jan 1 2003 when gamma glutamyl-transferase measurements had been changed.

We also investigated the shape of associations between absolute BMI level and Z score of triglycerides and breast cancer risk in subgroups of attained age: <50 years, 50-59 years, 60-69 years, and ≥70 years, by use of cubic splines in Cox models with the same strata and adjustments as in the previously described Cox models, with additional adjustment for cohort in analyses of BMI. We compared the fit of a linear model with the fit of the cubic

spline model by use of likelihood-ratio tests, in which the linear model was nested in a model that additionally included cubic splines.

All statistical analyses were performed in STATA MP/2 version 15 (StataCorp LP, College Station, Texas).

Results

The mean baseline age among the 813,927 participants in the study was 43 years (SD=9), and 65% were 35 to 44 years old (**Table 1**). Mean BMI was 25 kg/m^2 (SD=4), and 11% were obese (BMI ≥30 kg/m^2). The proportion of smokers was 29% among women and 34% among men. Follow-up time was on average 20 years (SD=8), and exclusion of the first follow-up year in survival analysis resulted in 7,515,453 person-years among women and 7,611,787 person-years among men. Participant characteristics were almost identical in the discovery and the replication cohort (**Supplementary Table 4**).

Among women, P-values for age-dependent metabolic variables were below 0.05 on seven occasions in the discovery cohort, which were replicated for BMI and triglycerides in relation to breast cancer risk (**Supplementary Table 5**). The corresponding finding among men was four in the discovery cohort, which was replicated for BMI and triglycerides in relation to liver cancer risk. In analyses of liver, pancreas and renal cell cancer excluding the first six years of follow-up, we found no significant age-interactions for any of the metabolic factors. For liver cancer among men, the P-values for age-interaction with BMI and triglycerides had increased to ≥0.05 in the discovery cohort, which may, however, have

been caused by limited statistical power in these analyses (n cases = 390 in the full cohort), as well as in the analyses of pancreas and renal cell cancer (n cases within the first follow-up years are shown in **Supplementary Table 6**).

Table 2 shows HRs at specific ages for BMI and triglycerides in relation to female breast and male liver cancer derived from linear interaction models with age (**Supplementary Table 7**). Higher levels of BMI and triglycerides were risk factors only at older ages and were negatively related to risk at younger ages. The age at which HRs crossed 1.0 was for breast cancer 62 years for BMI and 66 years for triglycerides, and for liver cancer among men 53 years for BMI and 72 years for triglycerides. HRs over attained age for BMI and triglycerides are shown in **Figure 2** in relation to female breast cancer, and in **Figure 3** in relation to liver cancer among men. For liver cancer, the age-interaction with BMI and triglycerides became less clear with the inclusion of women (n cases=209) (**Supplementary Figure 2**). HRs across age for mid-blood pressure, glucose, and cholesterol in relation to breast cancer among women and liver cancer among men are shown in **Supplementary Figures 3** and **4**.

The effect of interaction with attained age for BMI, triglycerides and breast cancer risk showed minor differences between subgroups of baseline age (**Supplementary Figure 5**), and minor influence by additional adjustment for parity and age at first birth (**Supplementary Figure 6**), or plasma gamma glutamyl-transferase level (**Supplementary Figure 7**). For liver cancer, adjustment for gamma glutamyl-transferase did not change the

interactions between BMI, triglycerides and attained age, but HRs were attenuated (**Supplementary Figure 8**).

For female breast cancer, we further plotted the shapes of associations across the exposure scales of BMI in absolute levels and Z score of triglycerides in relation to risk in subgroups of attained age (**Figure 4**). The associations were approximately linear within each age-group (P-values comparing the Cox spline model with a linear model were ≥0.22 and ≥0.12, for BMI and triglycerides, respectively) and showed gradually changing patterns of associations with increasing age.

Discussion

We systematically investigated the interaction between metabolic factors and attained age in relation to cancer risk in a large European study and found that BMI and triglycerides interacted positively with age in relation to breast cancer among women and liver cancer among men. Higher levels of BMI and triglycerides were related to a decreased risk for breast cancer before 62 and 66 years of age, but with an increased risk after that age. For male liver cancer, the age cut-points were at 53 and 72 years, respectively. However, rather than drastic changes of associations around these ages, and inherent to the linear tests performed to detect interactions, HRs for these metabolic factors changed linearly across the age-course.

Apart from breast cancer, we had no expectations of age-dependent associations with specific cancer forms. This hypothesis-free approach together with the

many tests performed (n=185) required correction for multiple testing. Without such correction, about nine false positive findings using 5% alpha-level would have been expected in our study. Bonferroni correction or its derivates are often used to correct for multiple testing,[18] but due to their one-off corrections of P-values, a very low P-value is required to reach significance. Additionally, these methods do not consider varying statistical power of each test, such as per cancer site in our study. Therefore, we used replication, which is common practice in genome-wide association studies discovering new gene loci for diseases or traits. Relative to other standard correction methods,[18] our replication approach enabled us to increase the chances of positive findings especially for rarer cancers, whilst also reducing the risk of false positives. However, we cannot exclude the possibility of false positive or negative findings, especially for rarer cancer forms with lower statistical power.

Our results for BMI and breast cancer correspond with the large number of observational studies suggesting a negative association between BMI and breast cancer risk before menopause but a positive association after menopause.[2-4] The division at menopause in studies of obesity is motivated by a plausible biological explanation for the association after menopause, at the same time, the biological support is much weaker for the negative association before menopause.[3, 19, 20] Two recent Mendelian randomization studies evaluated the potential causal association between BMI and breast cancer risk, and showed negative associations with breast cancer risk regardless of menopausal status.[21, 22] The authors speculated that the timing of obesity measurement might explain the divergent findings compared to observational studies. This, however, found no support in our study

(**Supplementary Figure 5**), though our youngest age-group might have been too old to capture young adulthood. However, results in our study provide an alternative explanation for the seemingly differential associations by study design. Higher BMI levels, especially obese levels (BMI ≥30 kg/m^2), decreased the risk for breast cancer an additional number of years after menopausal age, and the association changed continuously across age. This suggests that other age-related factors than menopause *per se* may cause the age-dependent association between obesity and breast cancer. The reasons for this is unclear, and one potential explanation may be that older women are more frequently diagnosed with estrogen and progesterone-receptor-positive breast tumours, which has shown a complex association with obesity.[23-25]

Our findings for triglycerides and breast cancer risk were similar to those for BMI, and likewise, results from observational studies of triglycerides reflected those of BMI with a graded association by menopause or menopausal age.[5, 26, 27] The largest study to date on triglycerides and breast cancer risk showed a suggestive negative association in a population of women who, at diagnosis, were primarily in their early postmenopausal ages.[28] These findings are supported in a recent Mendelian randomization study,[29] with a mean age of 66.5 years at breast cancer diagnosis (unpublished data in reference no. 29), i.e. the age at which the HR for triglycerides crossed 1.0 in our study. Thus, the present study and emerging evidence in recent studies suggest a similar age-dependent association between triglycerides and breast cancer risk to that of obesity and breast cancer, which may not depend on menopause.

The causes for our findings on liver cancer are unclear and may have several explanations. The male to female incidence ratio of liver cancer ranges from 2:1 to 4:1 across populations,[30] which has been hypothesized to be caused by a higher prevalence of strong risk factors among men including hepatitis B and C virus infection, alcohol intake, smoking, and also potentially by sex hormones.[31] Moreover, obesity has been related to an increased liver cancer risk among men in particular, which may suggest an influence by sex-specific effect modifiers.[32] Our results additionally suggest that such effect modifiers are age-related. Estrogens may be involved in liver cancer,[31] and whilst circulating estrogen levels increase with age among men,[33] it decreases among women after menopause. Potentially, the higher estrogen levels among older men act in synergy with obesity and elevated triglycerides in relation to risk. Alternatively, our results might reflect higher risks with a longer exposure to metabolic imbalances (correlated with higher age), as has been found for obesity duration in relation to several cancer forms among women.[34] Alternatively, obesity and triglycerides may act in synergy with effect modifiers that increase in strength with a longer exposure duration, e.g. the aforementioned main risk factors for liver cancer. Finally, our results might also reflect differential associations between BMI, triglycerides, and different liver cancer tumours typically occurring in younger versus older age.[35]

Weaknesses of our study include the use of a general obesity measure rather than abdominal obesity that is more specific in relation to metabolic imbalances, as well as the assessment of total cholesterol rather than its components. We also lacked complete data on potentially important confounders such as socioeconomic status, occupational

exposures, inflammatory markers, physical activity, and dietary intake including alcohol. However, analyses including adjustment for gamma glutamyl-transferase as an objective marker for alcohol intake[15-17] did not change the patterns of associations for breast and liver cancer, but the overall lowered HRs for liver cancer presumably reflect alcohol as a confounder and/or liver damage and more specifically non-alcoholic fatty liver disease as one pathway to liver cancer.[36, 37] Moreover, data on medications would have been desirable in our study, especially during follow-up given the relatively young population and long follow-up. Strengths of our study include the large population that provided robust risk estimates and facilitated the replication approach. Replicated age-exposure interactions using the Cox regression model were in addition modelled using non-linear models in order to detect possible effect of plateaus in the association. Moreover, we used high-quality registers to track and determine cancers,[38-40] and we adjusted in detail for smoking, an important risk factor for several cancer forms.

In conclusion, elevated BMI and triglycerides were related to decreased risks of breast cancer among younger women, but to increased risks among older women. The continuously changing associations across age rather than drastic changes around the age of menopause questions the notion that menopausal factors underlie the consistently observed differential associations between BMI and breast cancer risk before versus after menopause. Similar to breast cancer, BMI and triglycerides interacted positively with attained age in relation to liver cancer among men. These are unlikely to be chance findings

given the replication in the study, but the underlying causes are unknown and require further investigation in other studies.

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Conflict of interest

The authors declare that they have no conflict of interest.

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Table 1. Baseline characteristics of the study population

Characteristics	**Women (n=407,474)**		**Men (n=406,453)**	
Cohort, n (%)				
Oslo			17,856	4
NCS	43,074	11	44,658	11
40y	209,776	51	192,971	47
VHM&PP	94,032	23	80,963	20
VIP	50,965	13	49,868	12
MPP	9,627	2	20,137	5
Age, years				
Mean (SD)	42.9	9.6	43.0	8.8
Categories, n (%)				
<35	49,291	12	44,908	11
35-44	265,819	65	261,919	64
45-54	46,951	12	58,281	14
≥55	45,413	11	41,345	10
Smoking status, n (%)				
Never-smoker	195,655	48	156,273	38
Ex-smoker	94,343	23	111,365	27
Smoker	116,191	29	137,338	34
Fasting time, hours, n (%)				
<4	208,623	51	205,260	51
4-7	38,806	10	43,050	11
≥8	160,045	39	158,143	39
Body mass index, kg/m^2				
Mean (SD)	24.6	4.3	25.6	3.4
Categories, n (%)				
<25	254,545	63	188,970	47
25-29	107,942	27	176,160	44
≥30	42,992	11	39,581	10
Blood pressure, mm Hg				
Mean (SD) systolic blood pressure	126	18	133	16
Mean (SD) diastolic blood pressure	77	11	82	10
Mean (SD) mid-blood pressure[b]	102	13	107	12
Categories, n (%)				
<140 systolic and <90 diastolic blood pressure	314,363	77	255,396	63
140-159 systolic or 90-99 diastolic blood pressure	66,194	16	113,227	28
≥160 systolic or ≥100 diastolic blood pressure	26,665	7	37,564	9
Glucose, mmol/l[c]				

Mean (SD)	5.0	1.1	5.2	1.3
Categories, n (%)				
<6.1 in serum/plasma, <5.6 in whole blood	142,967	92	134,917	89
6.1-6.9 in serum/plasma, 5.6-6.0 in whole blood	8,548	6	10,875	7
≥7.0 in serum/plasma, ≥6.1 in whole blood	4,437	3	6,182	4
Cholesterol, mmol/l[d]				
Mean (SD)	5.5	1.2	5.6	1.2
Categories, n (%)				
<5.2	70,263	44	61,436	39
5.2-6.1	50,377	32	52,725	33
≥6.2	39,224	25	43,818	28
Triglycerides, mmol/l[d]				
Mean (SD)	1.2	0.8	1.6	1.2
Categories, n (%)				
<1.7	132,497	83	106,252	68
1.7-2.2	16,002	10	24,166	16
≥2.3	10,618	7	25,522	16
Years of follow-up				
Mean (SD)	19.4	7.3	19.7	8.1
Categories, n (%)				
<5	16,249	4	18,520	5
5-14	86,551	21	89,708	22
15-24	229,918	56	214,449	53
≥25	74,756	18	83,776	21

Oslo, Oslo Study I; NCS, Norwegian Counties Study; 40y, Age 40 Programme; VHM&PP, Vorarlberg Health Monitoring and Prevention Programme; VIP, Västerbotten Intervention Project; MPP, Malmö Prevention Project; SD, standard deviation.

[a]The number (%) of participants with missing values were for: smoking status, 2,762 (0.3%); body mass index, 3,737 (0.5%); blood pressure, 518 (0.1%); glucose, 293,652 (36%); cholesterol, 866 (1%); and triglycerides 3,900 (5%).

[b]Mid-blood pressure defined as (systolic+diastolic blood pressure)/2.

[c]Includes participants who fasted >8 hours and who had glucose measured in serum or plasma, i.e. not participants in the MPP who had glucose measured in whole blood.

[d]Includes participants who fasted >8 hours.

Table 2. Estimates of hazard ratios at specific ages, per standard deviation increase in BMI and triglycerides in relation to breast cancer in women and liver cancer in men, derived from results from Cox proportional hazards models in Supplementary Table 6.

Age, years	Breast cancer (N cases=11,723)		Liver cancer (N cases=444)	
	HR for BMI[a]	HR for triglycerides[a]	HR for BMI[a]	HR for triglycerides[a]
30	0.79	0.86	0.64	0.53
40	0.85	0.90	0.78	0.62
50	0.92	0.94	0.95	0.72
60	0.99	0.98	1.15	0.83
70	1.06	1.02	1.39	0.96
80	1.14	1.06	1.69	1.12
90	1.22	1.10	2.04	1.30
Age where HR=1[b]	62	66	53	72

HR, hazard ratio; BMI, body mass index.

[a]Hazard ratio derived from time-interaction variable of the metabolic factor calculated in Cox models with age as time-scale, stratified for cohort and birth cohort (six categories), and adjusted for baseline age (continuous), smoking (seven categories), and continuous Z score for BMI (except for BMI).

[b]Time were $\exp(\log(HR)+\log(HR_{time\text{-}interaction}*time))=1$.

Figure 1. Flowchart of exclusions and selections of individuals and health observations in the study. ↪ denotes exclusions. Abbreviations: VIP, Västerbotten Intervention Project; MPP, Malmö Preventive Project; BMI, body mass index; VHM&PP, Vorarlberg Health Monitoring and Prevention Programme.

Figure 2. Age-dependent hazard ratio with 95% confidence interval for breast cancer among women (N cases=11,723) per standard deviation increase in A) body mass index and B) triglycerides. Calculations are based on flexible parametric survival models with age as time scale and cubic splines for the age interaction, adjusted for cohort, birth cohort (six categories), baseline age (continuous), smoking (seven categories), and continuous Z score for BMI (except for BMI), with four degrees of freedom in the baseline hazard and three degrees of freedom in the time dependent effect of the metabolic factors.

Figure 3. Age-dependent hazard ratio with 95% confidence interval for liver cancer among men (N cases=444) per standard deviation increase in A) body mass index and B) triglycerides. Calculations are based on flexible parametric survival models with age as time scale and cubic splines for the age interaction, adjusted for cohort, birth cohort (six categories), baseline age (continuous), smoking (seven categories), and continuous Z score for BMI (except for BMI), with in A) three degrees of freedom for the baseline hazard and two for the time dependent effect of body mass index and in B) two degrees of freedom for the baseline hazard and one for the time dependent effect of triglycerides.

Figure 4. Hazard ratio of breast cancer among women for levels of A) body mass index (BMI) and B) triglycerides (in Z score) by attained age with N cases: 2,657 for <50 years, 4,591 50-60 years, 3,024 60-70 years, and 1,668>70 years. Models were derived from Cox models using cubic spline, with knots placed at 5, 35, 65 and 95 centiles. Participants with a BMI higher than 40, and with a triglyceride Z score >3 and <-3 were excluded from the respective analysis. Grid lines on the x axis represent centiles 10 and 90.