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Cardiovascular risk in Danish transgender persons.

A matched historical cohort study

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Abstract

Background: Cardiovascular risk could be increased in transgender persons, but the mechanism is undetermined.

Aim: To assess the risk of cardiovascular outcomes in Danish transgender persons compared to controls.

Methods: Historical register-based cohort study in Danish transgender persons and age-matched controls. The main outcome measure was cardiovascular diagnosis (any CVD) including medicine prescriptions for CVD during 2000-2018. The transgender cohort (N=2671) included persons with ICD-10 diagnosis code of "gender identity disorder" (N=1583) and persons with legal sex change (N=1088), 1270 were assigned female at birth (AFAB) and 1401 were assigned male at birth (AMAB). Controls (N=26,710) were matched by age (n=5 controls of same and n=5 controls of other birth sex) of their respective transgender person.

Results: The median (interquartile range) age at study inclusion was 22 (18; 29) years for persons AFAB and 26 (21; 39) years for persons AMAB, The mean (SD) follow up time was 4.5 (4.2) years for persons AFAB and 5.7 (4.8) years for persons AMAB. The hazard ratio (HR) for any CVD was significantly higher in transgender persons vs controls of same and other birth sex, with highest adjusted HR in transgender persons AFAB vs control men: 2.20 (95% CI: 1.64;2.95), p<0.00. Gender affirming hormone treatment (GAHT) explained part of elevated risk of CVD in transgender persons AFAB, whereas GAHT did not contribute to the elevated risk of CVD in transgender persons AMAB.

Conclusions: The risk of cardiovascular diagnosis was increased in transgender persons. The mechanism should be further investigated.

Keywords: Transgender, Gender Identity, Gender affirming Hormonal treatment, cardiovascular, ICD-10, ATC

Introduction

The term transgender is used to describe individuals, whose gender identity differs from the assigned sex at birth. Transgender men are assigned female at birth (AFAB), but identify as men and transgender women are assigned male at birth (AMAB), but identify as women (1). Gender affirming hormone treatment (GAHT) is a cornerstone of gender affirming treatment for many transgender persons (2) and non-binary persons may also wish for GAHT. Masculinizing GAHT includes testosterone treatment (2). Feminizing GAHT includes estrogen most often combined with anti-androgen treatment (2). In Europe, cyproterone acetate is routinely prescribed to block testosterone in adult transgender women (3). The treatment goal for GAHT is testosterone and estrogen levels within reference interval for the experienced gender (2). According to Danish national guidelines, initial evaluation of transgender persons and prescription of GAHT is performed by multidisciplinary teams located at one of three national centers of gender identity (CGI) (4). The number of referrals due to gender identity related diagnoses continues to increase (1, 5). General practitioners may monitor maintenance treatment with GAHT, but this option is rarely used due to continued need for involvement of interdisciplinary collaboration between specialists. Access to genital surgery in Denmark is at present limited.

Long term cardiovascular and metabolic health in transgender study populations has been discussed and especially the possible impact of GAHT on health outcomes (3, 6). Changes in sex hormone levels during transition in transgender persons may increase risk of cardiovascular disease (7-11). Cardiovascular morbidity in transgender men could be converted from female phenotype to resemble the risk of age matched male controls and *vice versa* for transgender women, however also the birth-assigned sex could be important for long term morbidity (11-13). Testosterone treatment results in unfavorable changes in lipid levels, higher blood pressure, higher hematocrit, which could result in development of coronary artery plaques (14, 15). Estrogen treatment of transgender females may translate into increased risk of venous thromboembolism (VTE) (10, 16). Cyproterone acetate acts as a synthetic progesterone (progestin) and promotes vasoconstriction, inflammation (17), and

hyperprolactinemia (18, 19), which may add further to cardio-metabolic risk in transgender women (3). The role of testosterone treatment for development of VTE in transgender men is less clear (20). Minority stress is defined as the stress burden linked to stigma, due to events like discrimination and victimization (21). The psychological responses to these events may lead to stress-related health problems (22) and could explain the increased risk for type 1 diabetes mellitus in transgender populations (23). Feminizing GAHT was associated with insulin resistance, but data regarding risk of type 2 diabetes mellitus in transgender study cohorts were not uniform (24, 25). A recent study in patients from the out-patient clinic at the CGI in Amsterdam reported higher mortality in a Dutch study cohort (n=4568) treated with GAHT (6). In transgender women, the standard mortality ratio was 1.8, 95% confidence interval (CI) (1.6–2.0) compared to general population men and 2.8 (2.5–3.1) compared to general population women (6). In transgender men, the standard mortality rate was 1.8 (1.3–2.4) compared to general population women and 1.2 (0.9–1.6) compared to general population men (6).

Higher cardiovascular risk in transgender study populations could be due to adverse risk profile including preexisting disease. We recently reported lower socioeconomic status in a Danish study cohort of transgender persons (26). In the general population, low socioeconomic status predicts impaired health and shorter life expectancy (27, 28). Transgender persons were reluctant to attend health care facilities (29, 30) and morbidity in persons attending a CGI center could differ from transgender persons outside a CGI. According to Danish legislation, legal sex change is possible without contact to a CGI. Therefore, Danish registers make it possible to evaluate health outcomes in transgender persons with and without contact to a CGI.

In the present study, we aimed to use national registers to investigate the risk for diagnosis and treatment of cardiovascular and metabolic outcomes in Danish transgender persons. We hypothesized that cardiovascular risk could be increased in transgender persons and that use of GAHT could be a mediator for cardiovascular risk.

Material and methods

Danish health registries: The study design was a matched historical cohort study based on nationwide registers. In Denmark, all citizens are provided a unique CPR (central person register, social security number) issued at birth or upon immigration to Denmark. The Danish CPR contains 10 digits where the first six digits corresponds to birth date (DDMMYY) and the last digit indicates a person's legal sex (odd digit for males, even digit for females) (31). The CPR is a key identifier in the Danish population-based medical records and link data from Danish health and social care registers at an individual level (32, 33).The CPR ensures unambiguous linkage at personal level between national registers. We used data from the civil registration system (33) and the National Patient Register (32). The civil registration system was established in 1968 and maintains complete records of births, deaths and emigration status for all residence in Denmark (33). The National Patient Register contains data on all inpatient hospital contacts in Denmark since 1977 and outpatient contacts are included since 1995 (31). From 2002, the National Patient Register also includes contacts for private hospitals excluding private practice specialists and general practitioners. Information regarding hospital contacts was included from all persons along with dates of death if applicable. We extracted ICD-10 codes given as primary or secondary diagnoses from psychiatric and somatic parts of National Patient Register according to individual CPR. Information on all prescriptions issued by prescribers, general practitioners or specialists in Denmark from 1995 to 2018 were retrieved from the National Prescriptions Registry according to individual CPR (34). The National Prescriptions Registry contains a complete record of Anatomic Therapeutic Chemical (ATC) code, date drug dispensed and number of drug packets for all prescriptions filled at Danish pharmacies.

Transgender study population: We included Danish transgender persons according to Danish health registries. The study population consisted of a CGI-cohort and a CPR-cohort; **CGI-cohort**: The CGI-cohort included persons above the age of 15 years with international classification of diseases (ICD)-10 diagnosis code of transgender. Age cut off 15 years was chosen to exclude children

with possible difference of sex development diagnosis. The diagnosis code occurred during 2000-2018 and persons had a valid Danish address at the time of inclusion. Until 2017, the diagnosis codes F64-F649 ("transsexual, gender identity disorder") were applied (35). The diagnosis codes DZ768E-DZ768E4 ("contact regarding gender identity condition") were introduced and applied after 2017. The date of study inclusion (index date) was defined as the first date for transgender diagnosis. **CPR-cohort:** The CPR-cohort included persons with legal sex change according to their CPR after the age of 18 years during year 2000-2018. We used the civil registration system to define sex change. According to Danish law, a citizen can apply for legal sex change after the age of 18 years, which implies change of the male or female identifier of the CPR number. Until 2014, legal sex change could only be performed following gonadectomy. Since September 1st, 2014, legal sex change can be performed independent of contact to the health care system. Persons were excluded if legal sex change was reversed within 30 days. Persons in the CGI-cohort were excluded from the CPR-cohort. The index date was the date of CPR change. In persons fulfilling study criteria for both the CGI-cohort and the CPR-cohort, the index date was the first date where study criteria were fulfilled.

Exclusion criteria: Gender dysphoria is rarely diagnosed in ageing persons (36) and we excluded individuals aged ≥ 80 years at index date from the CGI-cohort and CPR-cohort (N = 19).

Assigned sex at birth was determined as the earliest recorded CPR-encoded sex. Transgender persons were divided into persons affirmed male sex at birth (AMAB) and persons assigned female sex at birth (AFAB).

Controls: For each transgender person, we randomly drew five age-matched controls of the same sex at birth and five age-matched controls of the other sex at birth from the Danish civil registration system (Figure 1). Controls were assigned the index date of their matched transgender person. Controls were born in the same year and should be alive at the index date of their respective matched transgender person. Controls could not fulfill criteria for transgender at time of extraction

and were selected without repetition. Controls who subsequently met the transgender criteria were included and entered as transgender persons on the date of sex change (N=6).

Definition of study parameters

Outcomes were defined using ICD-10 diagnosis codes and/or medical treatment (Appendix, Table 1)

Primary study outcome: Any CVD was a composite outcome defined as at least one of the following criteria: Major CVD arterial, CVD venous, hyperlipidemia, hypertension or medical treatment for CVD.

Secondary study outcomes: We defined the composite study outcomes **Any arterial outcome** defined as at least one of the following criteria: Major CVD arterial, hyperlipidemia, hypertension or medical treatment for CVD. **Any venous outcome** was defined as CVD venous or medical treatment with antithrombotic agents. **Any metabolic outcome** was defined as at least one of the following criteria (diagnosis code and/or medical treatment): Obesity, diabetes mellitus, hyperlipidemia or hypertension.

Diagnosis codes were used to define the following study outcomes: **Major CVD arterial** was defined as at least one of the criteria: Diagnosis code of transient cerebral ishemia, stroke, ischemic heart disease, cardiomyopathy, arhythmia, heart failure, intracerebal hemorrage, cerebral infarction, occlusion of vertebral and cerebral arteries, or arterosclerosis.

CVD venous was defined as diagnosis code of pulmonary embolism or venous thrombosis;

M**etabolic syndrome** was defined as diagnosis code of obesity, diabetes mellitus, gestational diabetes, hyperlipidemia, hypertension, or chronic kidney disease.

Medical treatment for CVD and/or metabolic syndrome included antiobesity preparations, excluding diet products, drugs used in diabetes (insulins, blood glucose lowering drugs), antithrombotic agents, cardiac therapy (antiarrhythmics, cardiac stimulants, vasodilators), antihypertensives (antiandrenergic), diuretics, peripheral vasodilators, beta blocking agents, calcium

channel blockers, agents acting on the renin-angiotensin system, lipid modifying agents. Two or more medicine redemptions should occur within the same ATC group to be included.

Covariates

Age was calculated at the index date.

Presence of morbidity (defined as Charlson comorbidity index ≥1) was defined from hospital diagnoses within five years before the index date. The Charlson comorbidity index is based on 19 conditions (37). We calculated from the ICD-10 operationalization by Quan *et al*. *2005* (38) using the updated weights from Quan *et al*. *2011*(39).

Total personal income on the index year, excluding tax and interests, was extracted from the Income Statistics (40). We assessed income according to tertiles (high, middle, low) within age groups (≤18, 18-25, 25-30, ..., 60-65, >65 years). Data regarding baseline socioeconomic status in the cohort has been published recently (26).

GAHT following the index date was defined according to prescription and at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure). We also defined number of packages of redeemed GAHT (time-dependent cumulative continuous measure).

Feminizing treatment was defined as prescription of systemic natural estrogen (G03CA03 estradiol, G03CA04 estriol) and/or cyproterone acetate (G03HA01) and/or 5-α reductase inhibitor (G04CB01, G04CB02). Masculinizing treatment was defined as prescription of testosterone (G03BA03). In Denmark, GnRH analogue is administered without prescription for transgender persons < 18 years, and use of GnRH could not be investigated in the study cohort. Data regarding use of GAHT in the cohort has been presented recently (41).

Ethics

The study design was an open register-based cohort study and no approval was necessary from the local Ethics committee or Institutional Review Board by Danish law. The study was approved by

the Data Protection Agency and Statistics Denmark. Information regarding the study is published with OPEN ID: 939 https://open.rsyd.dk/OpenProjects/openProject.jsp?openNo=939&lang=da

Statistical analyses

Baseline characteristics in transgender persons were presented as frequencies for categorical variables. Continuous variables were summarised as medians with interquartile range. Crude analyses of baseline characteristics were performed using Wilcoxon Rank Sum test and chi-squared test.

We presented the incidence rates of cardiovascular and metabolic outcomes with 95% confidence intervals (95% CI). The cumulative incidence of any CVD was estimated by the Aalen-Johansen estimator, treating death as a competing risk. The hazard ratios (HRs) of any CVD were estimated by performing crude and adjusted Cox regression analyses and the 95% CIs were obtained by using clustered sandwich estimators. We checked that the non-proportional hazard assumption was not violated. All regression analyses included the matching factor age, and the adjusted analyses included the confounders: Charlson comorbidity index and personal income. The study population was young and N=64 had Charlson comorbidity index score =2 in the whole study population and N=14 had Charlson comorbidity index score >2. Small subgroups did not allow Charlson comorbidity index to be entered as a continuous variable and therefore, Charlson comorbidity index was entered as a dichotomous variable. According to a counterfactual approach, the HRs adjusted for potential confounders correspond to the total effect of being transgender on the risk of CVD, which can be decomposed into a controlled direct effect (CDE) and the proportion eliminated (PE) by eliminating the impact of mediation by GAHT (42). The CDE captures the influence of being transgender on the risk of CVD if the association between transgender and GAHT was hypothetically removed. In mediation analyses, we estimated CDE, by controlling for potential confounders (Charlson comorbidity index and socioeconomic status) and GAHT. GAHT was entered as a time-dependent covariate in Cox regression models – as a binary (≥2) variable for the main analyses and as a cumulative continuous variable in supplementary analyses. PE was obtained by dividing the

CDE by the total effect. The 95% CIs for CDE and PE were obtained by bootstrapping using 100 replicates.

In survival analyses, the population was followed from the index date until the date of the incident CVD diagnosis, emigration, death, or end of study period (31st Dec 2018), whatever came first. Observations with preexisting CVD conditions were excluded from the analysis. Transgender status was handled as a time-dependent variable allowing controls to switch to transgender person during follow-up.

Data management and data analyses were conducted using Stata 16 (StataCorp. 2019. *Stata Statistical Software: Release 16*. College Station, TX: StataCorp LLC) through a remote VPN access to Statistics Denmark with analysts blinded to the personal identities of the study subjects.

Sensitivity analysis

We performed a sensitivity analysis where we assessed whether findings from the regression analyses could be replicated in a population restricted to the CGI-cohort, as this would imply contact to a Danish CGI within the study period (2000-2018).

Results

A flowchart of the Danish transgender cohort and controls is shown in Figure 1. The transgender cohort (N=2671) included 1583 persons with ICD-10 diagnosis code of "gender identity disorder" and 1088 persons with legal sex change without diagnosis code, 1270 persons were assigned female at birth (AFAB) and 1401 persons were assigned male at birth (AMAB). The control group included 26,710 age-matched persons.

Baseline study characteristics are presented in Table 1. The median age at the index date was 22 (IQR: 18; 29) years in persons AFAB and 26 (IQR: 21; 39) years in persons AMAB. Transgender persons had lower income at baseline than controls. Transgender persons AFAB had lower prevalence of **any CVD,** any arterial and any metabolic outcome within five years before the index

date compared to control women. Transgender persons AFAB compared to control men had higher prevalence of morbidity. Transgender persons AMAB had lower prevalence of **any CVD**, any arterial, any venous and any metabolic outcome before the index date compared to control men. Transgender persons AMAB compared to control women had lower prevalence of morbidity (Charlson Comorbidity Index ≥ 1), **any CVD**, any arterial, and any metabolic outcome before the index date and higher prevalence of major CVD arterial.

Incidence rates of CVD and metabolic outcomes after the index date is presented in Table 2A-2B. The mean risk time (SD) was 4.5 (4.2) years for persons AFAB and 5.7 (4.8) years for persons AMAB. Transgender persons AFAB compared to control women had higher incidence rate of **any CVD** (17.38 (13.99; 21.57) vs. 11.19 (10.01; 12.50)), any arterial outcome (17.13 (13.78; 21.30) vs. 10.72 (9.57; 12.00) and diagnosis of hyperlipidemia (2.71 (1.60; 4.57) vs. 0.81 (0.55; 1.20)). Transgender persons AFAB compared to control men had higher incidence rate of **any CVD** (17.38 (13.99; 21.57) vs. 8.51 (7.50; 9.65)), any arterial (17.13 (13.78; 21.30) vs. 8.24 (7.25; 9.37)) and any metabolic outcome (21.91 (18.01; 26.65)) and diagnosis of obesity (9.43 (7.08; 12.55) vs. 1.08 (0.77; 1.52)), Table 2A.

Transgender persons AMAB compared to control men had higher incidence rate of **any CVD** (30.46 (26.35; 35.21) vs. 14.50 (13.33; 15.77)), any arterial (29.83 (25.78; 34.53) vs. 14.30 (13.14; 15.56)), any venous (11.81 (9.51; 14.66) vs. 6.38 (5.66; 7.19)), and any metabolic (33.67 (29.32; 38.66) vs. 12.76 (11.67; 13.95)) outcome, diagnosis of obesity (5.79 (4.28; 7.83) vs. 1.36 (1.06; 1.75)) and diabetes mellitus (3.04 (2.00; 4.61) vs. 1.56 (1.23; 1.98)). Transgender persons AMAB compared to control women had higher incidence rate of **any CVD** (30.46 (26.35; 35.21) vs. 16.88 (15.61; 18.26)), any arterial (29.83 (25.78; 34.53) vs. 16.40 (15.15; 17.76)), and any venous (11.81 (9.51; 14.66) vs. 5.73 (5.06; 6.49)) outcome, and hyperlipidemia (3.85 (2.66; 5.57) vs. 1.93 (1.56; 2.39)), Table 2B.

Cumulative incidence of any CVD, any arterial and any venous outcome in transgender persons vs. controls is shown in Figure 2.

Use of GAHT in transgender persons

At baseline, 18/1270 transgender persons AFAB and 32/1401 AMAB had used GAHT before the index date. In the CGI cohort, cumulative use of feminizing and masculinizing GAHT was 21.3% and 19.2% during follow up, respectively. In the CPR cohort, 5.7% used feminizing GAHT and 3.5% used masculinizing GAHT during follow up.

Crude and adjusted HR for development of any CVD are shown in Table 3A-3D. In all regression analyses, we found higher risk of developing **any CVD** in transgender persons compared to controls of same and other sex at birth. The highest HR was found in transgender persons AFAB compared to control men (adjusted HR 2.20 (95% CI: 1.64-2.95)) and in transgender persons AMAB compared to control men (adjusted HR 1.92 (95% CI: 1.59-2.33)).

Mediation analyses indicated that 33.2% (95% CI: 4.8-61.7) of the increased risk of **any CVD** in AFAB compared to control men, could be eliminated by hypothetically removing GAHT treatment (HR_{PE} 1.22 (95% CI: 1.00-1.44)) (Table 3B). The mediating role of GAHT for the risk of **any CVD** in AFAB compared to control women was uncertain when assessing GAHT as a binary variable. However, analyses including GAHT according to number of redeemed packages suggested that 26.4% (95% CI: 4.2;48.5) of the increased risk of **any CVD** in AFAB could be explained by GAHT (HR_{PE} 1.11 (95% CI: 1.04;1.19) (Appendix Table 2A). PE was statistically insignificant in models comparing persons AMAB with controls, both in analyses including GAHT as binary covariate and in analyses according to number of redeemed packages, Table 3C-D and Appendix Table 2C-D.

Sensitivity analysis

Regression analyses were repeated in the CGI-cohort alone (AFAB n=723 and AMAB n=765), which did not change significant results (appendix Tables 3A-2D).

Discussion

In the present study, we described risk of cardiovascular and metabolic outcomes in transgender persons using register-based data. The present data were unique as we had access to a national study cohort and could investigate the possible mediating effect of GAHT on study outcomes. We found higher risk of any CVD in transgender persons compared to controls of same and other birth sex with the highest risk for any CVD in transgender persons AFAB compared to control men. Furthermore, the incidence rate of arterial, venous and metabolic outcomes was higher in transgender persons compared to controls. GAHT was a statistically significant mediator of risk of any CVD in transgender persons AFAB, whereas GAHT was not a statistically significant mediator in persons AMAB.

CVD in transgender persons: The present study cohort was young, which resulted in limited number of major CVD outcomes. Therefore a relatively broad definition of CVD was applied in the present study as the primary study outcome. The definition of any CVD included ICD-10 diagnosis codes of CVD and ATC codes for medical treatment of CVD. This method allowed us to detect milder cardiovascular outcomes, which would be treated at the general practitioner without registering of hospital diagnoses. In a similar way, we reported incidence of arterial and vascular study outcomes defined by ICD-10 diagnosis codes and medical treatment. Short follow up duration and limited number of cardiovascular events in transgender study cohorts is a limitation in all studies due to expansion of transgender care especially within the last decade (6, 9, 43). Higher cardiovascular risk in transgender persons is supported by recent studies in selected study cohorts (6, 8, 9). Our finding of HR for CVD ranging from 1.63 to 2.20 in transgender persons corresponded to recent prospective and cross sectional studies in transgender study cohorts (6, 7, 10, 43). Most recently, de Blok *et al* (6) showed higher mortality in transgender men (AFAB) and transgender women (AMAB) compared to controls, which included approximately two-fold higher risk for death due to CVD. The study resembled the present study regarding mean age of inclusion (23 years for transgender men and 30 years for transgender women) (6). Median follow up duration was longer (five years for transgender men and 11 years for transgender women) and only

transgender persons with contact to the Gender Identity Clinic of Amsterdam were included (6). Persons attending a CGI undergo annual examinations according to national and international guidelines (1, 44). Systematic screening for medical disease could result in surveillance bias in the transgender cohort and over-diagnosis of study outcomes compared to the control group. Higher incidence rate of CVD independent of use of GAHT should be interpreted in this context and part of the higher incidence rate of study outcomes in the present study could be result of systematic evaluation and screening. Danish national guidelines define gender-affirming treatment as highly specialized treatment, which should be initiated by gender teams at a CGI. In the present study, persons in the CPR-cohort included 42% of the transgender study cohort, which implied no contact to a Danish CGI during 2000-2018. Therefore, we could investigate study outcomes in transgender persons with and without contact to a Danish CGI. Omitting persons from the CPR-cohort did not change significant results. Only a small number of transgender persons in the CPR-cohort were treated with GAHT (41). However, GAHT can be self-prescribed and purchased on the internet, which would not be detected in the current study setting. The rate of self-prescribed use of GAHT could be considerable (45). Self-medication could lead to higher morbidity in transgender persons without regular health care contacts and we plan to evaluate long-term morbidity in the CGI-cohort and CPR-cohort in future studies.

GAHT as a mediator for CVD in transgender persons: Interestingly, our data supported that use of GAHT mediated any CVD in transgender persons AFAB in contrast to transgender persons AMAB. Use of GAHT was entered as a mediator in regression models as binary variable (exposure yes/no) and as continuous variable (number of redeemed packages), which did not change significant results. The cumulative incidence of prescription of GAHT was 21% in the CGI-cohort, which allowed us to investigate CVD risk in users and non-users of GAHT. Furthermore, study outcomes were investigated in transgender persons independent of attendance to a GCI, which implied information regarding CVD risk in persons, who could be lost to follow up in other studies.

National settings regarding transgender care would affect study outcomes. Use of GAHT mediated any CVD in transgender persons AFAB, which could imply a negative impact of masculinizing GAHT on cardiovascular risk. However, several confounders could not be accounted for, given the current register based design. GAHT was defined by ATC code and we could not distinguish between transdermal and intramuscular application. In the present study, the IR per 1,000 person-years of any arterial outcome was significantly higher in transgender persons AFAB compared to control women and control men, whereas the IR per 1,000 person-years of any venous outcome was comparable in transgender persons AFAB and controls. Our results could support higher risk for arterial vascular disease in transgender persons AFAB. However, the number of incident ICD-10 codes for arterial and venous diagnoses was limited and our data did not allow for a sensitivity analysis regarding the individual components of our composite endpoint of any CVD. The mechanism for possible higher cardiovascular risk in transgender study cohorts awaits clarification (10, 46). Unfavorable lipid profile during GAHT was associated with higher risk of CVD events according to the Framingham 30-year CVD risk estimate in transgender men but not in transgender women (47), and the importance of lipid profile for prospective CVD risk should be investigated. Testosterone treatment has been considered safe regarding cardiovascular outcomes (1), but the present study support that more data are needed regarding vascular risk during masculinizing hormonal treatment (11, 15).

As recently reported, adult feminizing treatment in Denmark most often include prescription of cyproterone acetate, whereas spironolactone is less often used and GnRH agonist is not generally available for adult transgender persons (3, 4). Cardiovascular adverse effects of cyproterone acetate on lipid profile and coagulation system are well described (3). In recent years, the initial dosage of cyproterone acetate has been reduced from 25-50 mg/day to 12.5 mg with further down titration within the first year (3) and studies regarding the lowest effective dosage of cyproterone acetate are ongoing (48, 49). The present data could be reassuring regarding use of feminizing treatment for transgender care as GAHT did not mediate any CVD in regression analyses. However, genital

surgery is not widely available in Denmark and many transgender women will continue use of cyproterone acetate for several years. The number of incident ICD-10 diagnoses codes for arterial and venous CVD was limited in transgender persons AMAB in the present study, but we found a trend for higher IR per 1,000 person-years of CVD venous compared to control men (p=0.09). We plan to investigate the risk of long-term adverse effects of cyproterone acetate during longer follow up of the present study cohort.

Baseline cardiovascular and metabolic diagnoses were less frequent in transgender persons compared to controls and the presence of morbidity at baseline was 3%. Recent studies reported that wish for hormone management was one of the primary aims for seeking health care programs in transgender study cohorts (50, 51). GAHT may not be prescribed in persons with severe physical disease and contraindications (1). A possible explanation for our findings could be that persons with contraindications for GAHT are less likely to seek transgender care (26). A recent register based study found that cancer was diagnosed at later stages in transgender persons and worse survival for many cancer types was partly explained through existing barriers to access the healthcare system (30). Existing medical diseases could be underdiagnosed in transgender persons due to reluctance to seek general healthcare. This could be an alternative explanation of our finding of lower prevalence of preexisting cardiovascular and metabolic diagnoses in our transgender study cohort.
Socioeconomic status was lower in transgender persons compared to controls (26). However, socioeconomic status was not a significant confounder for any CVD in the present study. There is no individual cost for health care consultations in Denmark and all expenses are covered by the public health care system. The cost for GAHT is partly reimbursed by the general medicine support system in line with other prescription drugs. Socioeconomic status could be an important confounder of cardiovascular risk in countries with unequal access to the national health care system.

Strengths and limitations apply to this study. **S**tudy strengths are nationwide data and the possibility to investigate possible mediators of CVD. Inclusion of persons with legal sex change allowed us to investigate study outcomes in persons without contact to CGI. Some limitations apply to the present study. We obtained transgender diagnosis by available ICD-10 codes and CPR changes. As a result, transgender persons without hospital contacts and/or persons with no wish for CPR change would not be included in our population. The present study results may not apply for non-binary persons. Persons who terminated transgender care at a CGI before year 2000 would be left outside the dataset. In Denmark, referral to national centres of CGI has increased dramatically during the last decade (5) and follow up duration was relatively limited in this study. Repeated data analysis will be necessary to obtain longer follow up duration. In Denmark, change of CPR is rarely performed, except for transgender persons. Protection of persecuted persons could also allow CPR change, but these cases will not usually involve change of the sex-identifier of the CPR number. We divided persons according to assigned gender at birth, which could lead to misclassification of sex in foreigners if they immigrated after the date of the legal sex change. Furthermore, misclassification might occur if an individual received a transgender diagnosis or had legal sex change performed before year 2000. Similarly, the CPR-cohort was defined by conditioning on the future, i.e. that the individual did not enter the CGI-cohort during the study period. Use of GAHT was defined according to redemption databases, which imply methodological challenges (52). Register based data did not allow us to investigate the influence of BMI, family history of CVD, minority stress and lifestyle factors (diet, smoking, exercise) on study outcomes (10, 46).

Conclusion

Cardiovascular and metabolic outcomes were more prevalent in transgender persons compared to controls. GAHT exposure could contribute to the elevated cardiovascular risk in transgender person assigned female at birth. Future studies will be able to bring further knowledge regarding mechanisms for higher cardiovascular risk in transgender persons.

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Legends:

Figure 1: Flowchart of Danish transgender cohort and controls

Flowchart of included study cohort

AFAB: Assigned female at birth, AMAB: assigned male at birth

CGI: Center of gender identity, CPR: Social security number

Figure 2: Cumulative incidence of any CVD, any arterial and any venous outcome in transgender persons vs. controls

Cumulative incidence of any CVD in transgender persons vs. controls

AFAB: Assigned female at birth, AMAB: assigned male at birth

Table 1: Baseline characteristics of Danish transgender cohort and controls and CVD diagnoses before the index date.

Baseline characteristics including morbidity and cardiovascular diagnoses five years before the index date and medication history one year before index date.

Morbidity defined as Charlson comorbidity index ≥1

P1: AFAB vs. control women, P2: AFAB vs. control men

P3: AMAB vs. control women, P4: AMAB vs. control men

AFAB: Assigned female at birth, AMAB: Assigned male at birth, ATC: Anatomical Therapeutic Chemical Classification System, CVD: cardiovascular disease, IQR: Interquartile range

Table 2A. Incidence rates of cardiovascular and metabolic outcomes in Danish transgender cohort (AFAB) and controls

Presented per 1000 PY (person years), preexcisting diseases excluded for diseases in question

P1: AFAB vs control women, P2: AFAB vs. control men

AFAB: Assigned female at birth, ATC: Anatomical Therapeutic Chemical Classification System,

CVD: Cardiovascular disease, IR: incidence rate

Table 2B. Incidence rates of cardiovascular and metabolic outcomes in Danish transgender cohort (AMAB) and controls

Presented per 1000 PY (person years), preexcisting diseases excluded for diseases in question

P1: AMAB vs control men, P2: AMAB vs. control women

AMAB: Assigned male at birth, ATC: Anatomical Therapeutic Chemical Classification System, CVD: Cardiovascular disease, IR: incidence rate

Table 3A. Crude and adjusted Hazard ratios in Danish transgender cohort, AFAB (N=1238) vs. control women (N=6091) and development of any CVD outcome

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).

GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).

Subjects with preexisting CVD excluded from the analysis.

N>445 exposed to GAHT in the analysis

AFAB: Assigned female at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio

$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Table 3B. Crude and adjusted Hazard ratios in Danish transgender cohort, AFAB (N=1238) vs. control men (N=6139) and development of any CVD outcome

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).

GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).

Subjects with preexisting CVD excluded from the analysis.

N<353 exposed to GAHT in the analysis

AFAB: Assigned female at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio

$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Table 3C. Crude and adjusted Hazard ratios in Danish transgender cohort, AMAB (N=1325) vs. control men (N=6463) and development of any CVD outcome

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/⩾1)), socioeconomic status, and use of GAHT (yes/no).

GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).

Subjects with preexisting CVD excluded from the analysis.

N=381 exposed to GAHT in the analysis

AMAB: Assigned male at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio

$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion

676 eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the
677 direction of HR_{CDE} and HE_{PE} was the same.

678 **Table 3D. Crude and adjusted Hazard ratios in Danish transgender cohort, AMAB (N=1325)**
679 **vs. control women (N=6404) and development of any CVD outcome**

680 Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models
681 corrected for morbidity (Charlson comorbidity index (0/⩾1)), socioeconomic status, and use of
682 GAHT (yes/no).
683 GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-
684 dependent binary measure).
685 Subjects with preexisting CVD excluded from the analysis.
686 N=594 exposed to GAHT in the analysis
687 AMAB: Assigned male at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone
688 treatment, HR: Hazard ratio
689 $HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard
690 ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion
691 eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the
692 direction of HR_{CDE} and HE_{PE} was the same.
693

Table 1: Baseline characteristics of Danish transgender cohort and controls and CVD diagnoses before the index date.

	Transgender AFAB N= 1270	**Control Women N= 6350**	**Control Men N= 6350**	**P1**	**P2**	**Transgender AMAB N=1401**	**Control Men N=7005**	**Control Women N=7005**	**P3**	**P4**
Baseline										
Age, median (IQR)	22(18;29)	22(18;29)	22(19;29)			26(21;39)	26(21;39)	26(21;39)		
Morbidity (yes)	43(3.4%)	190(3.0%)	153(2.4%)	0.46	**0.04**	39(2.8%)	196(2.8%)	281(4.0%)	0.98	**0.03**
Income low tertile	864(68.0%)	2201(34.7%)	2152(33.9%)	0.00	0.00	962(68.7%)	2174(31.0%)	2391(34.1%)	**0.00**	**0.00**
Income middle tertile	277(21.8%)	2286(36.0%)	1893(29.8%)	0.00	0.00	287(20.5%)	2110(30.1%)	2609(37.2%)	**0.00**	**0.00**
Any CVD outcome	32(2.5%)	259(4.1%)	211(3.3%)	**0.01**	0.14	76(5.4%)	542(7.7%)	601(8.6%)	**0.01**	**0.00**
Any arterial outcome	30(2.4%)	250(3.9%)	207(3.3%)	**0.01**	0.09	75(5.4%)	539(7.7%)	583(8.3%)	**0.01**	**0.00**
Any venous outcome	15(1.2%)	59(0.9%)	54(0.9%)	0.40	0.26	20(1.4%)	163(2.3%)	123(1.8%)	**0.04**	0.39
Any metabolic outcome	52(4.1%)	477(7.5%)	216(3.4%)	**0.00**	0.22	71(5.1%)	548(7.8%)	794(11.3%)	**0.00**	**0.00**
Medical history ICD-10										
Major CVD Arterial	16(1.3%)	58(0.9%)	60(0.9%)	0.25	0.30	34(2.4%)	143(2.0%)	100(1.4%)	0.36	**0.01**
CVD Venous	<5	16(0.3%)	6(0.1%)	.	.	<5	18(0.3%)	28(0.4%)	.	.
Obesity	23(1.8%)	199(3.1%)	29(0.5%)	**0.01**	**0.00**	10(0.7%)	33(0.5%)	231(3.3%)	0.25	**0.00**
Diabetes mellitus	14(1.1%)	97(1.5%)	48(0.8%)	0.25	0.21	17(1.2%)	75(1.1%)	92(1.3%)	0.64	0.76
Hyperlipidemia	5(0.4%)	14(0.2%)	13(0.2%)	0.26	0.21	9(0.6%)	47(0.7%)	30(0.4%)	0.91	0.28
Hypertension	11(0.9%)	24(0.4%)	37(0.6%)	**0.02**	0.24	20(1.4%)	102(1.5%)	86(1.2%)	0.94	0.54
Chronic kidney disease	.(.%)	6(0.1%)	9(0.1%)	.	.	<5	12(0.2%)	<5	.	.
Medication history ATC										
A08: Antiobesity	.(.%)	18(0.3%)	<5	.	.	<5	8(0.1%)	36(0.5%)	.	.
A10: Diabetes	10(0.8%)	84(1.3%)	61(1.0%)	0.12	0.56	13(0.9%)	108(1.5%)	101(1.4%)	0.08	0.13
B01: Antithrombotic	13(1.0%)	49(0.8%)	50(0.8%)	0.36	0.40	19(1.4%)	157(2.2%)	104(1.5%)	**0.04**	0.72
C01-C09: Cardiovascular	19(1.5%)	171(2.7%)	130(2.0%)	**0.01**	0.20	40(2.9%)	414(5.9%)	448(6.4%)	**0.00**	**0.00**
C10: Antilipid	8(0.6%)	54(0.9%)	71(1.1%)	0.43	0.12	22(1.6%)	212(3.0%)	159(2.3%)	**0.01**	0.10

Baseline characteristics including morbidity and cardiovascular diagnoses five years before the index date and medication history one year before index date. Transgender cohort divided according to assigned gender at birth.
Morbidity defined as Charlson comorbidity index ≥1

P1: AFAB vs. control women, P2: AFAB vs. control men
P3: AMAB vs. control men, P4: AMAB vs. control women
AFAB: Assigned female at birth, ATC: Anatomical Therapeutic Chemical Classification System, CVD: cardiovascular disease, IQR: Interquartile range

Table 2A. Incidence rates of cardiovascular and metabolic outcomes in Danish transgender cohort (AFAB) and controls

	Mean risk time (year)	Incident Transgender AFAB N=1270		Control women N=6350		Control men N=6350			
		N(%)	IR	N(%)	IR	N(%)	IR	P1	P2
Any CVD outcome	4.5	82(6.6%)	17.38(13.99;21.57)	311(5.1%)	11.19(10.01;12.50)	241(3.9%)	8.51(7.50;9.65)	**0.03**	**0.00**
Any arterial outcome	4.5	81(6.5%)	17.13(13.78;21.30)	299(4.9%)	10.72(9.57;12.00)	234(3.8%)	8.24(7.25;9.37)	**0.02**	**0.00**
Any venous outcome	4.7	21(1.7%)	4.15(2.71;6.36)	108(1.7%)	3.58(2.97;4.33)	92(1.5%)	3.07(2.50;3.76)	0.91	0.57
Any metabolic outcome	4.5	100(8.2%)	21.91(18.01;26.65)	535(9.1%)	20.55(18.88;22.36)	226(3.7%)	7.94(6.97;9.04)	0.32	**0.00**
ICD-10, CVD									
Major CVD Arterial	4.7	19(1.5%)	3.56(2.25;5.66)	96(1.5%)	3.18(2.61;3.89)	96(1.5%)	3.20(2.62;3.90)	0.98	0.98
CVD venous	4.8	5(0.4%)	0.96(0.40;2.30)	38(0.6%)	1.24(0.90;1.70)	23(0.4%)	0.75(0.50;1.13)	0.37	0.87
ICD-10, metabolic syndrome									
Obesity	4.7	47(3.8%)	9.43(7.08;12.55)	307(5.0%)	10.63(9.50;11.89)	33(0.5%)	1.08(0.77;1.52)	0.07	**0.00**
Diabetes mellitus	4.8	11(0.9%)	1.96(1.05;3.64)	67(1.1%)	2.21(1.74;2.81)	33(0.5%)	1.08(0.77;1.52)	0.53	0.13
Hyperlipidemia	4.8	14(1.1%)	2.71(1.60;4.57)	25(0.4%)	0.81(0.55;1.20)	39(0.6%)	1.27(0.93;1.74)	**0.01**	0.06
Hypertension	4.7	18(1.4%)	3.33(2.07;5.36)	73(1.2%)	2.40(1.91;3.02)	65(1.0%)	2.14(1.68;2.74)	0.41	0.21
Chronic kidney disease	4.8	<5	.	<5	.	12(0.2%)	0.39(0.22;0.69)	.	.
ATC, medical treatment									
A08: Antiobesity	4.8	<5	.	8(0.1%)	0.26(0.13;0.52)	<5	.	.	.
A10: Diabetes	4.8	15(1.2%)	2.92(1.76;4.85)	79(1.3%)	2.61(2.09;3.25)	34(0.5%)	1.12(0.80;1.57)	0.82	**0.01**
B01: Antithrombotic	4.7	20(1.6%)	3.95(2.55;6.11)	90(1.4%)	2.97(2.42;3.66)	82(1.3%)	2.73(2.20;3.39)	0.67	0.42
C01-C09: Cardiovascular	4.6	54(4.3%)	10.95(8.38;14.29)	187(3.0%)	6.50(5.63;7.50)	170(2.7%)	5.84(5.03;6.79)	**0.02**	**0.01**
C10: Antilipid	4.7	29(2.3%)	5.70(3.96;8.21)	81(1.3%)	2.68(2.15;3.33)	109(1.7%)	3.66(3.04;4.42)	**0.01**	0.17

Presented per 1000 PY (person years), preexcisting diseases excluded for diseases in question

P1: AFAB vs control women, P2: AFAB vs. control men

AFAB: Assigned female at birth, ATC: Anatomical Therapeutic Chemical Classification System, CVD: Cardiovascular disease, IR: incidence rate

Table 2B. Incidence rates of cardiovascular and metabolic outcomes in Danish transgender cohort (AMAB) and controls

	Mean risk time (year)	Incident Transgender AMAB N=1401		Control men N=7005		Control women N=7005		P1	P2
		N(%)	IR	N(%)	IR	N(%)	IR		
Any CVD outcome	5.7	185(14.0%)	30.46(26.35;35.21)	546(8.4%)	14.50(13.33;15.77)	625(9.8%)	16.88(15.61;18.26)	**0.00**	**0.00**
Any arterial outcome	5.7	182(13.7%)	29.83(25.78;34.53)	539(8.3%)	14.30(13.14;15.56)	610(9.5%)	16.40(15.15;17.76)	**0.00**	**0.00**
Any venous outcome	6.1	82(5.9%)	11.81(9.51;14.66)	268(3.9%)	6.38(5.66;7.19)	246(3.6%)	5.73(5.06;6.49)	**0.01**	**0.00**
Any metabolic outcome	5.6	203(15.3%)	33.67(29.32;38.66)	484(7.5%)	12.76(11.67;13.95)	870(14.0%)	24.84(23.24;26.54)	**0.00**	0.23
ICD-10, CVD									
Major CVD Arterial	6.1	47(3.4%)	6.39(4.77;8.56)	258(3.8%)	6.10(5.40;6.89)	214(3.1%)	4.91(4.29;5.61)	0.57	0.51
CVD venous	6.3	17(1.2%)	2.31(1.44;3.72)	53(0.8%)	1.20(0.92;1.57)	72(1.0%)	1.62(1.28;2.04)	0.09	0.54
ICD-10, metabolic syndrome									
Obesity	6.2	43(3.1%)	5.79(4.28;7.83)	60(0.9%)	1.36(1.06;1.75)	423(6.2%)	10.08(9.16;11.09)	**0.00**	**0.00**
Diabetes mellitus	6.2	22(1.6%)	3.04(2.00;4.61)	68(1.0%)	1.56(1.23;1.98)	125(1.8%)	2.84(2.38;3.38)	**0.04**	0.57
Hyperlipidemia	6.2	28(2.0%)	3.85(2.66;5.57)	99(1.4%)	2.26(1.86;2.76)	86(1.2%)	1.93(1.56;2.39)	0.10	**0.02**
Hypertension	6.1	50(3.6%)	7.01(5.31;9.25)	207(3.0%)	4.84(4.23;5.55)	203(2.9%)	4.67(4.07;5.36)	0.22	0.18
Chronic kidney disease	6.3	<5	.	21(0.3%)	0.47(0.31;0.73)	15(0.2%)	0.31(0.18;0.52)	.	.
ATC, medical treatment									
A08: Antiobesity	6.3	10(0.7%)	1.36(0.73;2.53)	5(0.1%)	0.11(0.05;0.27)	30(0.4%)	0.68(0.47;0.97)	**0.00**	0.16
A10: Diabetes	6.2	34(2.4%)	4.73(3.38;6.62)	83(1.2%)	1.91(1.54;2.37)	128(1.9%)	2.90(2.44;3.45)	**0.00**	0.14
B01: Antithrombotic	6.1	76(5.5%)	10.87(8.68;13.61)	250(3.7%)	5.94(5.24;6.72)	219(3.2%)	5.08(4.45;5.79)	**0.01**	**0.00**
C01-C09: Cardiovascular	5.8	154(11.3%)	24.18(20.65;28.32)	385(5.8%)	9.74(8.81;10.76)	451(6.9%)	11.58(10.56;12.70)	**0.00**	**0.00**
C10: Antilipid	6.1	78(5.7%)	11.25(9.01;14.04)	299(4.4%)	7.22(6.45;8.09)	259(3.8%)	6.08(5.39;6.87)	**0.04**	**0.01**

Presented per 1000 PY (person years), preexcisting diseases excluded for diseases in question

P1: AMAB vs control men, P2: AMAB vs. control women

AMAB: Assigned male at birth, ATC: Anatomical Therapeutic Chemical Classification System, CVD: Cardiovascular disease, IR: incidence rate

Table 3A. Crude and adjusted Hazard ratios in Danish transgender cohort, AFAB (N=1238) vs. control women (N=6091) and development of any CVD outcome

					Mediation analysis		
	Crude HR (95% CI)	Adjusted HR (95% CI)	Adjusted HR (95% CI)	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (% (95% CI))
Transgender (yes/no)	**1.63 (1.27;2.08) 0.00**	**1.62 (1.27;2.07) 0.00**	**1.63 (1.26;2.11) 0.00**	**1.63 (1.26;2.11) 0.00**	**1.57 (1.21;2.03) 0.00**	1.04 (0.96;1.12)	9.9 (0.0;30.2)
Confounders							
Morbidity		1.22 (0.67;2.22) 0.51		1.22 (0.67;2.22) 0.51	1.21 (0.67;2.21) 0.53		
Income							
Low tertile			1.00 (0.76;1.31) 0.99	0.99 (0.76;1.30) 0.97	1.00 (0.76;1.32) 0.98		
Middle tertile			1.02 (0.78;1.35) 0.86	1.02 (0.78;1.35) 0.87	1.03 (0.78;1.36) 0.82		
Mediator							
≥2 GAHT prescriptions (yes/no)					1.21 (0.87;1.69) 0.26		

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).
Subjects with preexisting CVD excluded from the analysis.
N>445 exposed to GAHT in the analysis
AFAB: Assigned female at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Table 3B. Crude and adjusted Hazard ratios in Danish transgender cohort, AFAB (N=1238) vs. control men (N=6139) and development of any CVD outcome

					Mediation analysis		
	Crude HR (95% CI)	Adjusted HR (95% CI)	Adjusted HR (95% CI)	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (%(95% CI))
Transgender (yes/no)	**2.11 (1.61;2.76) 0.00**	**2.11 (1.61;2.76) 0.00**	**2.20 (1.64;2.95) 0.00**	**2.20 (1.64;2.95) 0.00**	**1.80 (1.27;2.55) 0.00**	**1.22 (1.00;1.44)**	**33.2(4.8;61.7)**
Confounders		0.90 (0.43;1.88)		0.93 (0.45;1.92)	0.88 (0.41;1.86)		
Morbidity		0.79		0.84	0.73		
Income			0.95 (0.70;1.28)	0.95 (0.70;1.28)	0.98 (0.72;1.32)		
Low tertile			0.73	0.74	0.88		
Middle tertile			1.21 (0.91;1.60) 0.19	1.21 (0.91;1.60) 0.19	1.22 (0.92;1.61) 0.17		
Mediator							
≥2 GAHT prescriptions (yes/no)					**1.86 (1.14;3.05) 0.01**		

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).
Subjects with preexisting CVD excluded from the analysis.
N<353 exposed to GAHT in the analysis
AFAB: Assigned female at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Table 3C. Crude and adjusted Hazard ratios in Danish transgender cohort, AMAB (N=1325) vs. control men (N=6463) and development of any CVD outcome

					Mediation analysis		
	Crude HR (95% CI)	Adjusted HR (95% CI)	Adjusted HR (95% CI)	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (%(95% CI))
Transgender (yes/no)	**2.00 (1.68;2.39) 0.00**	**2.00 (1.68;2.39) 0.00**	**1.93 (1.59;2.33) 0.00**	**1.92 (1.59;2.33) 0.00**	**1.93 (1.57;2.38) 0.00**	1.00 (0.90;1.10)	.
Confounders		1.05 (0.64;1.73)		1.06 (0.64;1.74)	1.06 (0.64;1.74)		
Morbidity		0.84		0.83	0.83		
Income			1.16 (0.96;1.41)	1.16 (0.96;1.41)	1.16 (0.96;1.41)		
Low tertile			0.12	0.12	0.13		
Middle tertile			1.11 (0.91;1.35) 0.29	1.11 (0.91;1.35) 0.29	1.11 (0.91;1.35) 0.29		
Mediator							
≥2 GAHT prescriptions (yes/no)					0.99 (0.71;1.37) 0.94		

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).
Subjects with preexisting CVD excluded from the analysis.
N=381 exposed to GAHT in the analysis
AMAB: Assigned male at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Table 3D. Crude and adjusted Hazard ratios in Danish transgender cohort, AMAB (N=1325) vs. control women (N=6404) and development of any CVD outcome

					Mediation analysis		
	Crude HR (95% CI)	Adjusted HR (95% CI)	Adjusted HR (95% CI)	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (%(95% CI))
Transgender (yes/no)	**1.78 (1.51;2.11) 0.00**	**1.79 (1.52;2.12) 0.00**	**1.73 (1.46;2.05) 0.00**	**1.74 (1.46;2.06) 0.00**	**1.80 (1.51;2.14) 0.00**	0.97 (0.92;1.02)	.
Confounders		1.32 (0.93;1.86)		1.32 (0.93;1.86)	1.33 (0.94;1.88)		
Morbidity		0.12		0.12	0.10		
Income			1.17 (0.96;1.42)	1.17 (0.96;1.43)	1.17 (0.96;1.43)		
Low tertile			0.12	0.11	0.12		
Middle tertile			1.04 (0.85;1.29) 0.69	1.05 (0.85;1.30) 0.63	1.05 (0.86;1.30) 0.62		
Mediator							
≥2 GAHT prescriptions (yes/no)					0.85 (0.67;1.08) 0.19		

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).

GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).

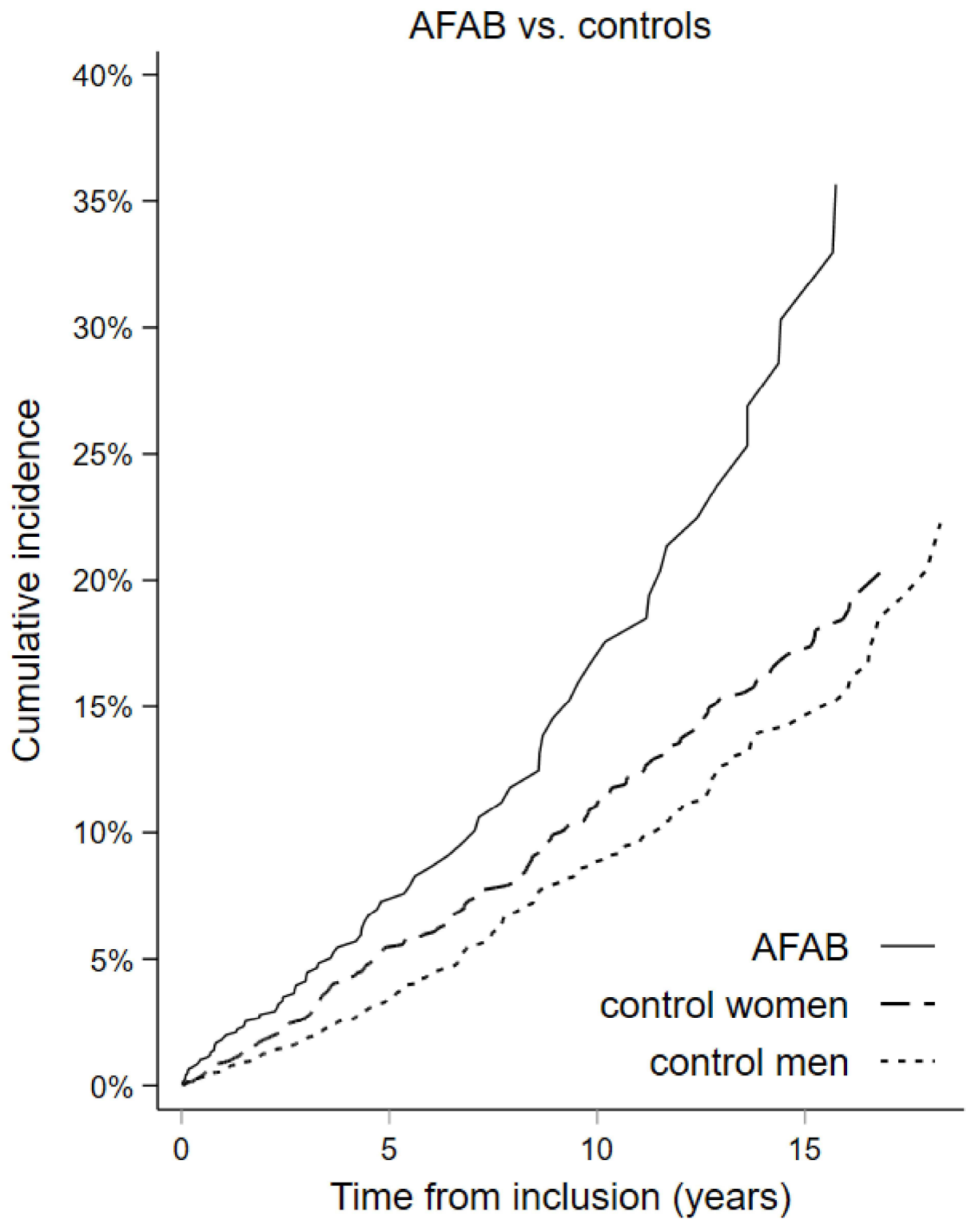
Subjects with preexisting CVD excluded from the analysis.

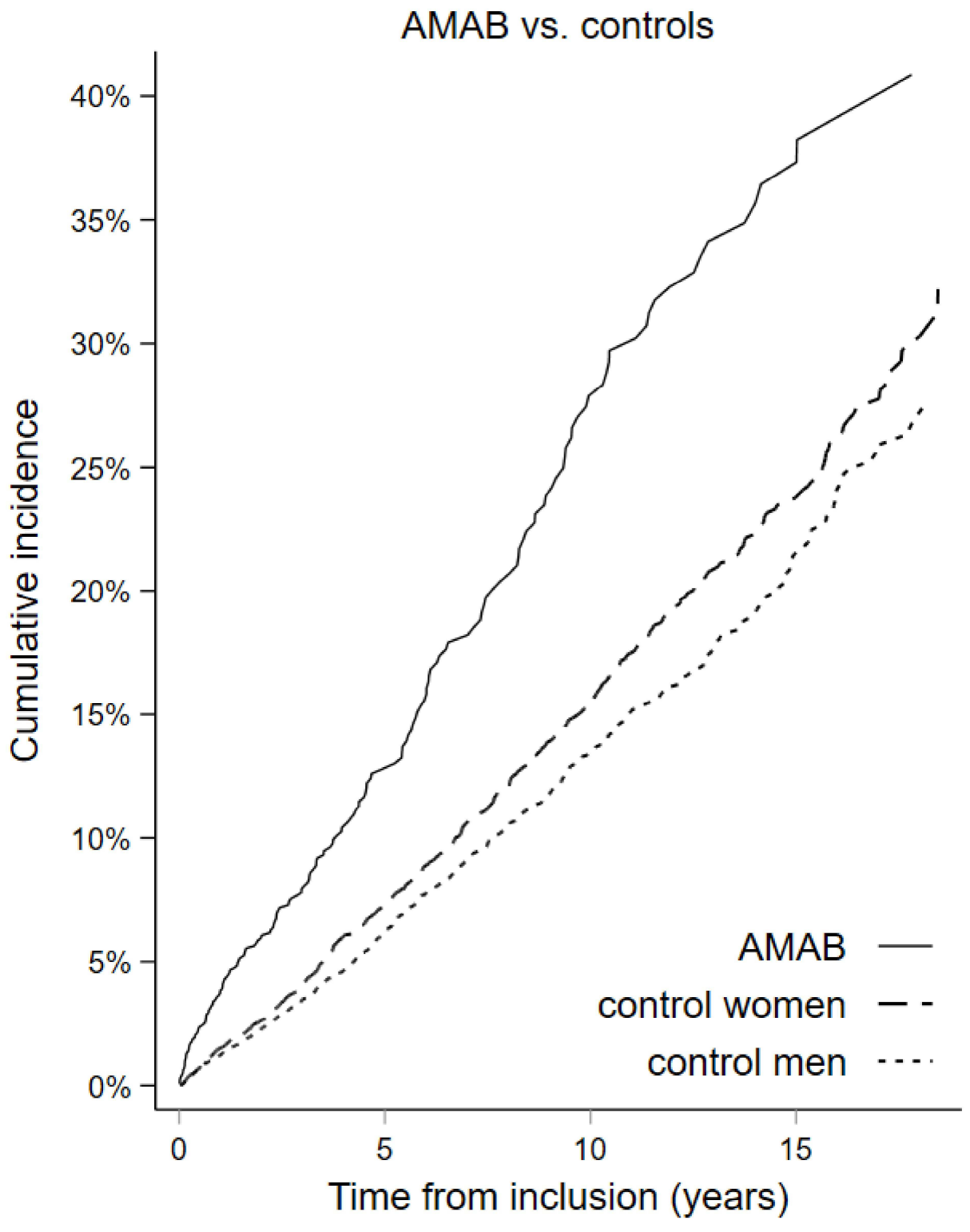
N=594 exposed to GAHT in the analysis

AMAB: Assigned male at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio

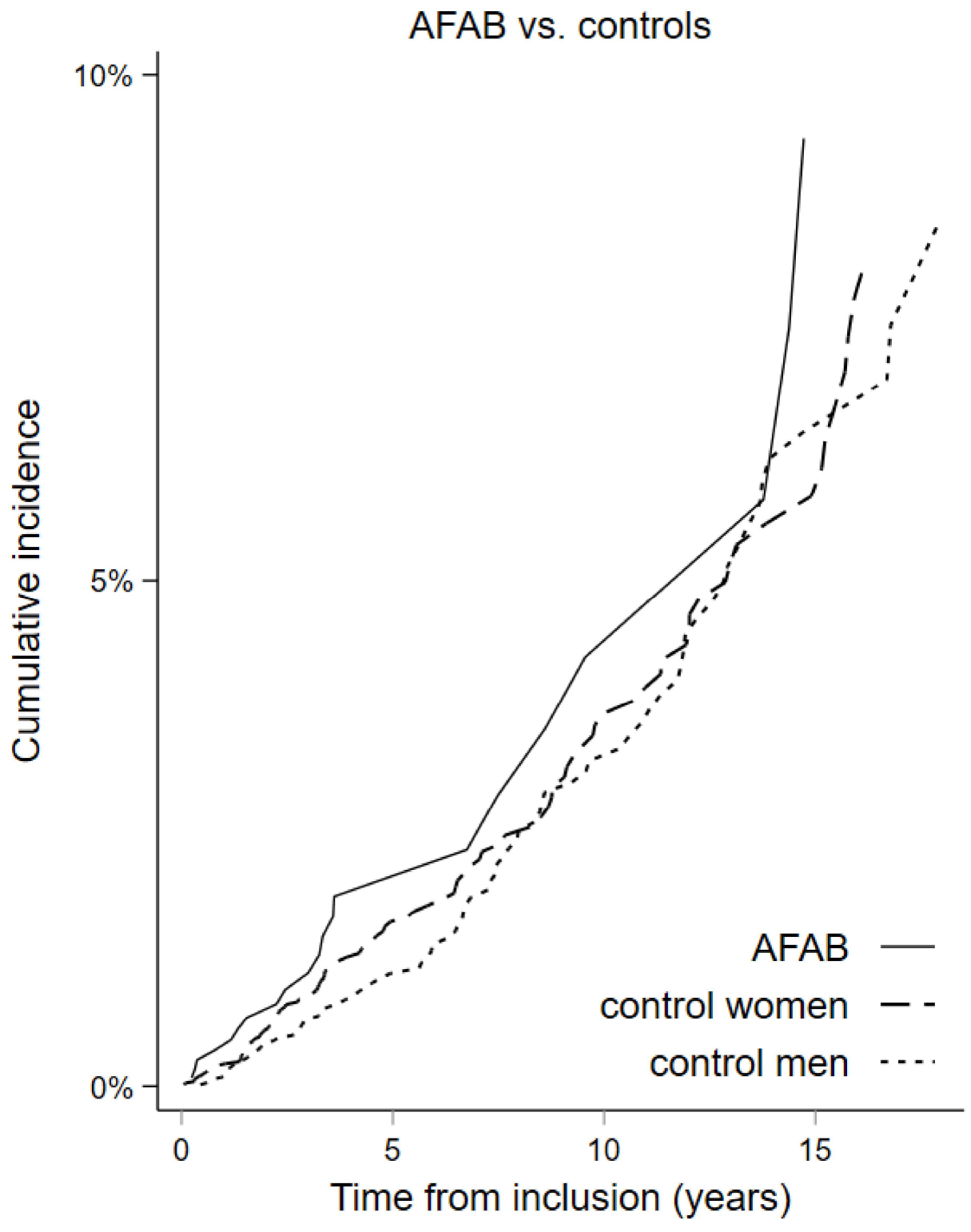
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

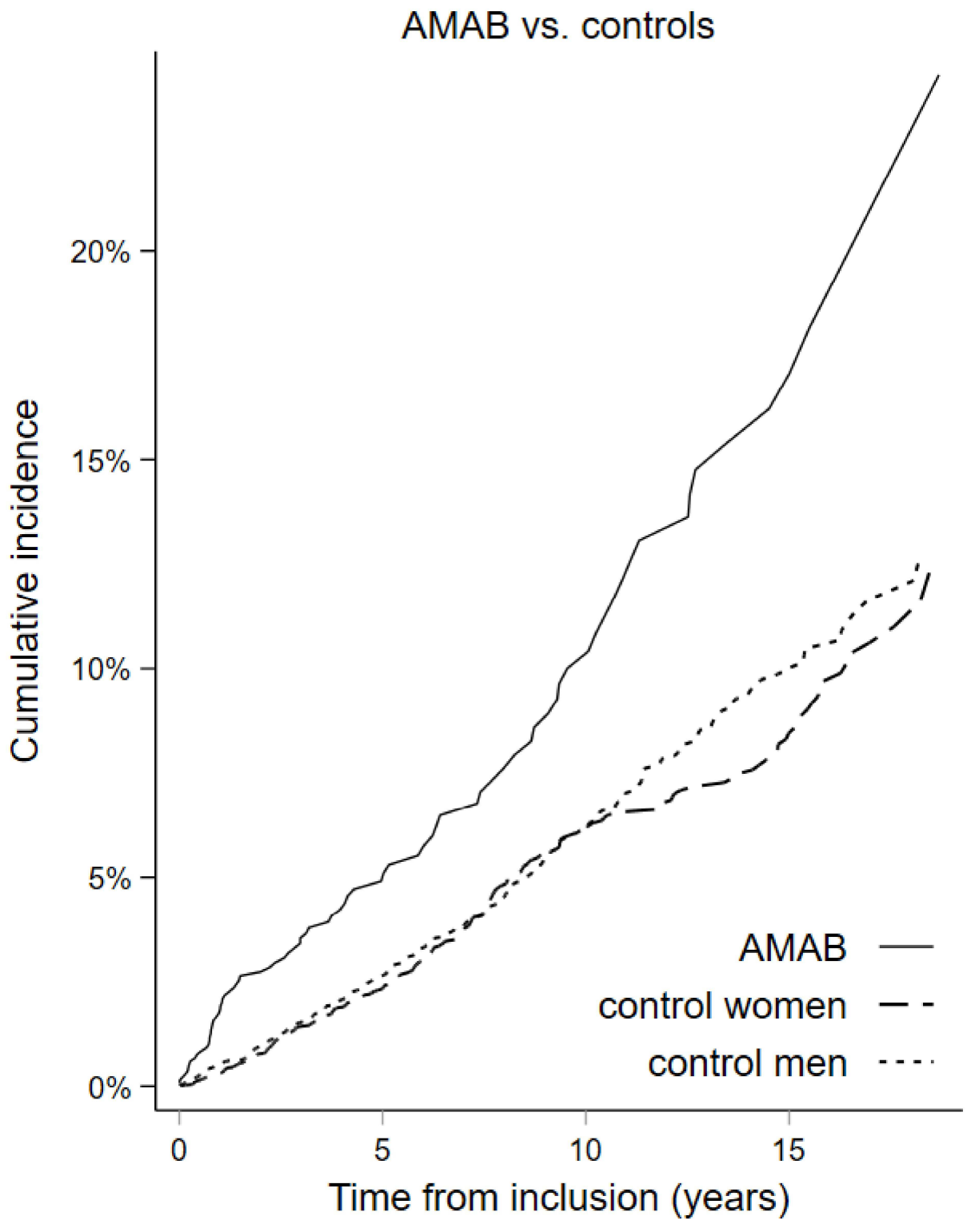
Cumulative incidence function of any cardiovascular disease



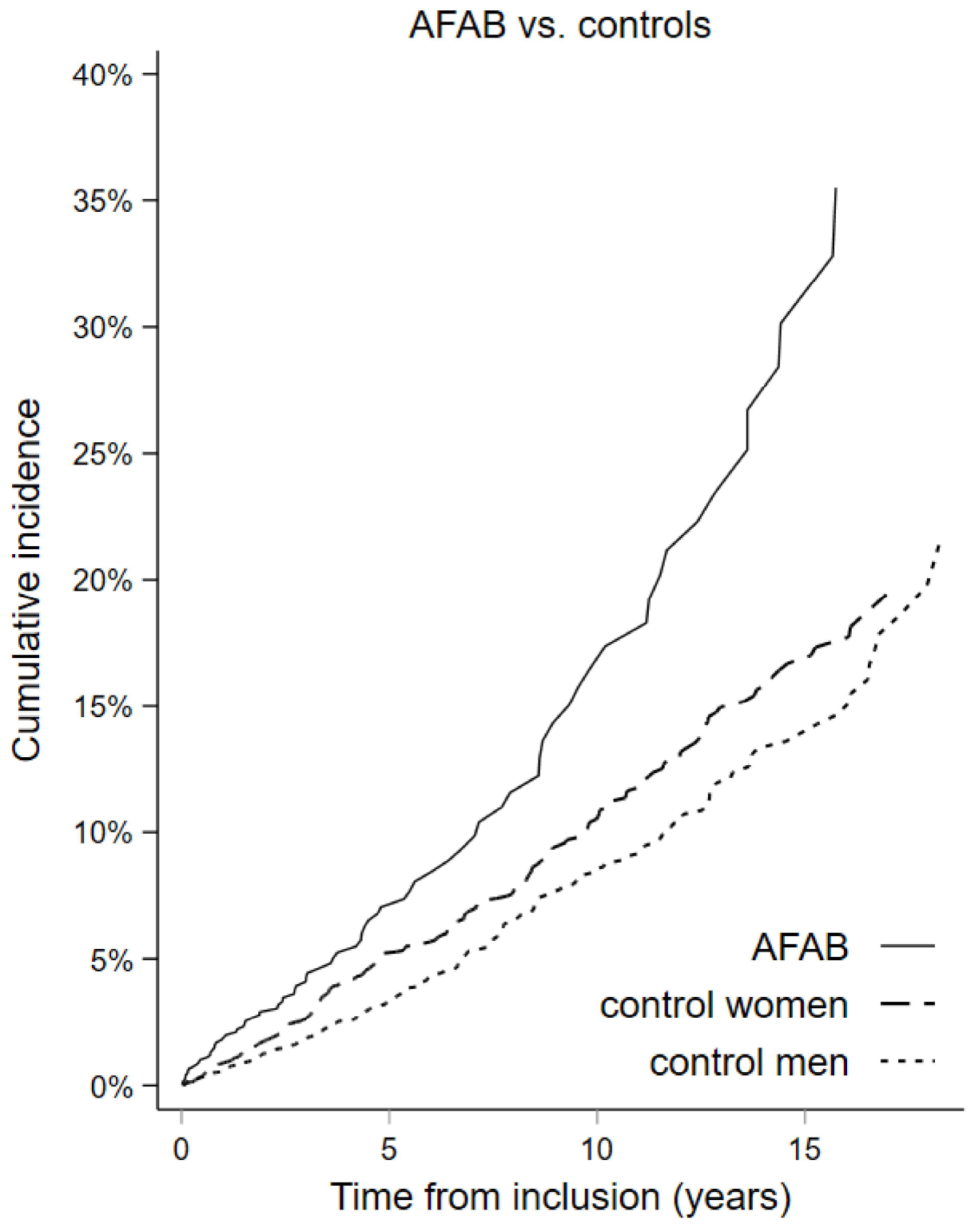


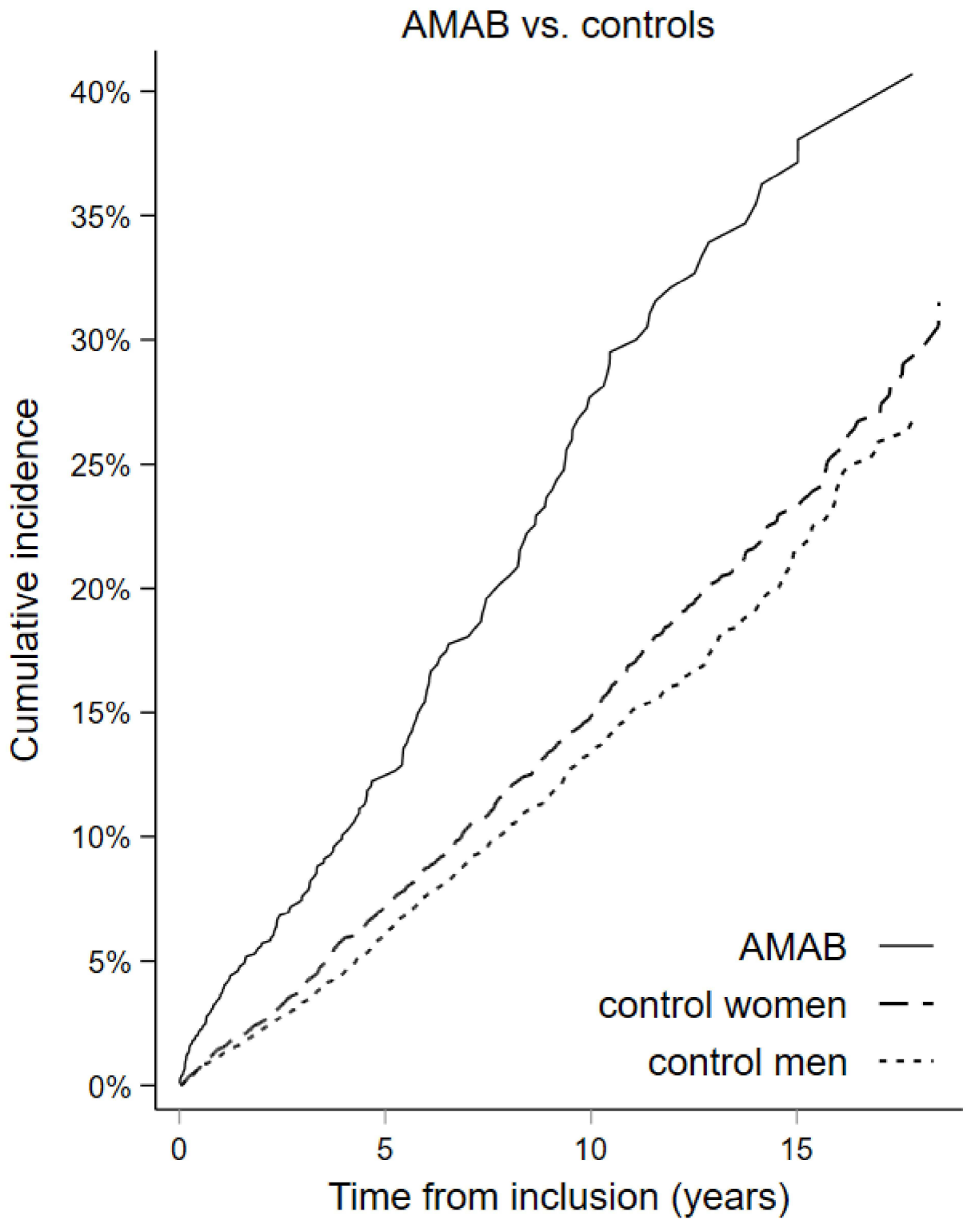
Cumulative incidence function of any venous cardiovascular disease





Cumulative incidence function of any arterial cardiovascular disease





Appendix table 1, Overview of study outcomes

Primary outcome	
Any CVD outcome	Major CVD arterial (defined below) OR CVD venous (defined below) OR hyperlipidemia: E780-E785 OR Hypertension: I10-I15 OR Medical treatment for CVD (≥2 redemptions) B01: Antithrombotic agents C01: Cardiac therapy (antiarrhythmics, cardiac stimulants, vasodilators) C02: Antihypertensives (antiandrenergic) C03: Diuretics C04: Peripheral vasodilators C07: Beta blocking agents C08: Calcium channel blockers C09: Agents acting on the renin-angiotensin system C10: Lipid modifying agents
Secondary outcomes	
Any arterial outcome	Hyperlipidemia: E780-E785 OR Major CVD arterial (defined below) OR Hypertension OR Medical treatment for CVD (≥2 redemptions) B01: Antithrombotic agents C01: Cardiac therapy (antiarrhythmics, cardiac stimulants, vasodilators) C02: Antihypertensives (antiandrenergic) C03: Diuretics C04: Peripheral vasodilators C07: Beta blocking agents C08: Calcium channel blockers C09: Agents acting on the renin-angiotensin system C10: Lipid modifying agents
Any venous outcome	CVD venous OR Medical treatment B01: Antithrombotic agents (≥2 redemptions)
Any metabolic outcome	Obesity: DE66 OR

	Diabetes mellitus, gestational diabetes: E10- E11, E13, E14, 024 OR Hyperlipidemia: E780-E785 OR Hypertension: I10-I15 OR Medical treatment with (≥2 redemptions) A08: Antiobesity preparations, excluding diet products A10: Drugs used in diabetes (insulins, blood glucose lowering drugs) C02: Antihypertensives (antiandrenergic) C03: Diuretics C04: Peripheral vasodilators C07: Beta blocking agents C08: Calcium channel blockers C09: Agents acting on the renin-angiotensin system C10: Lipid modifying agents
Major CVD arterial	Transient cerebral ishemia, stroke: G45-G46 OR Ischemic heart disease: I20-I25 OR Cardiomyopathy: I42 OR Arrhythmia: I47-I49 OR Heart failure: I50 OR Intracerebal hemorrage: I61 OR Cerebral infarction, occlusion of vertebral and cerebral arteries: I63-I66 OR Arterosclerosis: I70
CVD venous	Pulmonary embolism: I26 OR Venous thrombosis: I80-82
Metabolic syndrome	Obesity: DE66 OR Diabetes mellitus, gestational diabetes: E10- E11, E13, E14, 024 OR Hyperlipidemia: E780-E785 OR Hypertension: I10-I15 OR Cronic kidney disease: N18

Appendix Table 2A. Adjusted Hazard ratios in Danish transgender cohort, AFAB (n=1238) vs. control women (N=6091) and development of any CVD outcome – continues cumulative GAHT

		Mediation analysis		
	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (% (95% CI))
Transgender (yes/no)	**1.63 (1.26;2.11) 0.00**	**1.47 (1.12;1.92) 0.01**	**1.11 (1.04;1.19)**	**26.4 (4.2;48.5)**
Mediator				
GAHT (number of packages)		**1.01 (1.01;1.02) 0.00**		

Hazard ratios estimated using Cox regression presented for corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT.
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent continues cumulative measure).
Subjects with preexisting CVD excluded from the analysis.
AFAB: Assigned female at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
ratio for the proportion eliminated by the impact of GAHT (=HRTE / HRCDE). PE: Proportion eliminated by the impact of GAHT (=HRTE - HRCDE)/(HRTE - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Appendix Table 2B. Adjusted Hazard ratios in Danish transgender cohort, AFAB (n=1238) vs. control men (N=6139) and development of any CVD outcome – continues cumulative GAHT

		Mediation analysis		
	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (% (95% CI))
Transgender (yes/no)	**2.20 (1.64;2.95) 0.00**	**1.98 (1.45;2.69) 0.00**	**1.11 (1.03;1.19)**	**18.6 (0.0;32.3)**
Mediator GAHT (number of packages)		**1.01 (1.01;1.02) 0.00**		

Hazard ratios estimated using Cox regression presented for corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT.
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent continues cumulative measure).
Subjects with preexisting CVD excluded from the analysis.
AFAB: Assigned female at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Appendix Table 2C. Adjusted Hazard ratios in Danish transgender cohort, AMAB (n=1325) vs. control men (N=6463) and development of any CVD outcome – continued cumulative GAHT

		Mediation analysis		
	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (% (95% CI))
Transgender (yes/no)	**1.92 (1.59;2.33) 0.000**	**1.93 (1.59;2.34) 0.000**	1.00 (0.95;1.04)	.
Mediator GAHT (number of packages)		1.00 (0.99;1.01) 0.85		

Hazard ratios estimated using Cox regression presented for corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT.
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent continues cumulative measure).
Subjects with preexisting CVD excluded from the analysis.
AMAB: Assigned male at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Appendix Table 2D. Adjusted Hazard ratios in Danish transgender cohort, AMAB (n=1325) vs. control women (N=6404) and development of any CVD outcome – continued cumulative GAHT

		Mediation analysis		
	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (% (95% CI))
Transgender (yes/no)	**1.74 (1.46;2.06) 0.000**	**1.75 (1.47;2.09) 0.000**	0.99 (0.96;1.02)	.
Mediator GAHT (number of packages)		1.00 (0.99;1.01) 0.52		

Hazard ratios estimated using Cox regression presented for corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT.
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent continues cumulative measure).
Subjects with preexisting CVD excluded from the analysis.
AMAB: Assigned male at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Appendix, table 3A. Crude and adjusted Hazard ratios in Danish CGI-cohort, AFAB (n=722) vs. control women (N=3627) and development of any CVD outcome

					Mediation analysis		
	Crude HR (95% CI)	Adjusted HR (95% CI)	Adjusted HR (95% CI)	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (% (95% CI))
Transgender (yes/no)	**1.59 (1.15;2.20) 0.01**	**1.59 (1.15;2.21) 0.01**	**1.58 (1.13;2.21) 0.01**	**1.59 (1.14;2.22) 0.01**	**1.52 (1.09;2.13) 0.01**	1.04 (0.94;1.15)	11.3 (0.0;43.0)
Confounders							
Morbidity		0.70 (0.28;1.79) 0.460		0.70 (0.28;1.79) 0.46	0.70 (0.28;1.78) 0.45		
Income							
Low tertile			1.00 (0.71;1.41) 0.99	1.00 (0.71;1.42) 0.98	1.01 (0.72;1.43) 0.95		
Middle tertile			0.96 (0.67;1.37) 0.81	0.96 (0.67;1.38) 0.82	0.97 (0.68;1.40) 0.87		
Mediator							
≥2 GAHT prescriptions (yes/no)					1.23 (0.81;1.86) 0.33		

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).
Subjects with preexisting CVD excluded from the analysis.
N=368 exposed to GAHT in the analysis
AFAB: Assigned female at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio

$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Appendix, table 3B. Crude and adjusted Hazard ratios in Danish CGI-cohort, AFAB (n=722) vs. control men (N=3648) and development of any CVD outcome

					Mediation analysis		
	Crude HR (95% CI)	Adjusted HR (95% CI)	Adjusted HR (95% CI)	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (% (95% CI))
Transgender (yes/no)	**2.15 (1.49;3.09) 0.00**	**2.16 (1.49;3.11) 0.00**	**2.23 (1.52;3.27) 0.00**	**2.23 (1.52;3.28) 0.00**	**1.82 (1.16;2.86) 0.01**	**1.22 (0.96;1.49)**	**33.2 (0.0;73.3)**
Confounders							
Morbidity		0.64 (0.24;1.70) 0.37		0.66 (0.25;1.75) 0.40	0.65 (0.24;1.76) 0.40		
Income							
Low tertile			0.89 (0.60;1.33) 0.57	0.90 (0.60;1.33) 0.59	0.91 (0.61;1.36) 0.65		
Middle tertile			1.08 (0.74;1.57) 0.70	1.07 (0.74;1.57) 0.71	1.08 (0.74;1.57) 0.67		
Mediator							
≥2 GAHT prescriptions (yes/no)					1.92 (1.01;3.63) 0.05		

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).
Subjects with preexisting CVD excluded from the analysis.
N=307 to GAHT in the analysis
AFAB: Assigned female at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Appendix Table 3C. Crude and adjusted Hazard ratios in Danish CGI-cohort, AMAB (n=759) vs. control men (N=3777) and development of any CVD outcome

					Mediation analysis		
	Crude HR (95% CI)	Adjusted HR (95% CI)	Adjusted HR (95% CI)	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (% (95% CI))
Transgender (yes/no)	**1.91 (1.55;2.35) 0.00**	**1.90 (1.54;2.34) 0.00**	**1.86 (1.49;2.31) 0.00**	**1.85 (1.49;2.30) 0.00**	**1.80 (1.42;2.28) 0.00**	1.03 (0.88;1.17)	5.8 (0.0;36.7)
Confounders							
Morbidity		1.17 (0.67;2.03) 0.59		1.17 (0.67;2.03) 0.58	1.17 (0.67;2.03) 0.58		
Income							
Low tertile			1.13 (0.90;1.42) 0.30	1.13 (0.90;1.42) 0.30	1.13 (0.90;1.42) 0.30		
Middle tertile			1.08 (0.86;1.36) 0.51	1.08 (0.86;1.36) 0.50	1.08 (0.86;1.36) 0.51		
Mediator							
≥2 GAHT prescriptions (yes/no)					1.09 (0.73;1.61) 0.69		

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).
Subjects with preexisting CVD excluded from the analysis.
N=314 exposed to GAHT in the analysis
AMAB: Assigned male at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.

Appendix table 3D. Crude and adjusted Hazard ratios in Danish CGI-cohort, AMAB (n=759) vs. control women (N=3762) and development of any CVD outcome

					Mediation analysis		
	Crude HR (95% CI)	Adjusted HR (95% CI)	Adjusted HR (95% CI)	Adjusted all potential confounders HR_{Total} (95% CI)	Adjusted all potential confounders and GAHT HR_{CDE} (95% CI)	HR_{PE} (95% CI)	PE (% (95% CI))
Transgender (yes/no)	**1.76 (1.44;2.15) 0.000**	**1.76 (1.44;2.15) 0.000**	**1.72 (1.41;2.11) 0.000**	**1.73 (1.41;2.12) 0.000**	**1.79 (1.46;2.20) 0.000**	0.96 (0.91;1.02)	.
Confounders							
Morbidity		1.36 (0.92;2.02) 0.125		1.38 (0.93;2.04) 0.110	1.38 (0.93;2.06) 0.107		
Income							
Low tertile			1.22 (0.95;1.56) 0.117	1.23 (0.96;1.57) 0.106	1.22 (0.95;1.57) 0.111		
Middle tertile			1.13 (0.87;1.46) 0.366	1.14 (0.88;1.49) 0.312	1.15 (0.88;1.49) 0.305		
Mediator							
≥2 GAHT prescriptions (yes/no)					0.85 (0.64;1.12) 0.240		

Hazard ratios estimated using Cox regression presented for crude model (adjusted for age) and models corrected for morbidity (Charlson comorbidity index (0/≥1)), socioeconomic status, and use of GAHT (yes/no).
GAHT defined as at least two redemptions of feminizing and/or masculinizing treatment (time-dependent binary measure).
Subjects with preexisting CVD excluded from the analysis.
N=469 exposed to GAHT in the analysis
AMAB: Assigned male at birth, CVD: cardiovascular disease, GAHT: Gender affirming hormone treatment, HR: Hazard ratio
$HR_{Total:}$ Hazard ratio for total effect, HR_{CDE}: Hazard ratio for controlled direct effect, HR_{PE}: Hazard ratio for the proportion eliminated by the impact of GAHT (=HR_{Total} / HR_{CDE}). PE: Proportion eliminated by the impact of GAHT (=HR_{Total} – HR_{CDE})/(HR_{Total} - 1)*100, only presented if the direction of HR_{CDE} and HE_{PE} was the same.