Gender-affirming hormone therapy: An updated literature review with an eye on the future

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Abstract

In line with increasing numbers of transgender (trans) and gender non-binary people requesting hormone treatment, the body of available research is expanding. More clinical research groups are presenting data, and the numbers of participants in these studies are rising. Many previous review papers have focused on all available data, as these were scarce, but a more recent literature review is timely. Hormonal regimens have changed over time, and older data may be less relevant for today's practice.

In recent literature, we have found that even though mental health problems are more prevalent in trans people compared to cisgender (cis) people, less psychological difficulties occur, and life satisfaction increases with gender-affirming hormone treatment (GAHT) for those who feel this is a necessity. With GAHT, body composition and contours change towards the affirmed sex. Studies in bone health are reassuring, but special attention is needed for adolescent and adult trans women, aiming at adequate dosage of hormonal supplementation and stimulating therapy compliance. Existing epidemiological data suggest that the use of (certain) estrogens in transgender women induces an increased risk of myocardial infarction and stroke, the reason that lifestyle management can be an integral part of trans health care. The observed cancer risk in trans people does not exceed the known cancer-risk differences between men and women.

Now it is time to integrate the mostly reassuring data, to leave the overly cautious approach behind, to not copy the same research questions repeatedly, and to focus on longer follow-up data with larger cohorts.

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Keywords

transgender, gender incongruence, gender-affirming hormone therapy, hormone treatment, testosterone, estrogens

Keypoints

- Gender-affirming hormonal treatment for those who request it improves mental health problems, if present.
- When clients are well informed and adequately monitored, side effects of hormone treatment are rare in the short- and mid-term.
- Virilizing and feminizing hormone treatment leads to effective body composition changes towards the affirmed sex.
- Lifestyle management can be a part of gender-affirming care aiming to interfere with a suggested increased cardiovascular risk.
- Longer-term and multicenter research needs to be a priority now.

Introduction

As early as 1923, Hirschfeld described the term “transsexual” to describe people who want to live a life according to their experienced gender, instead of their assigned gender (1). The 20th century marked the social awakening for men and women who knew that they were—as at the time often described—“trapped in the wrong body” (2). Gender incongruence is a newer term to describe the incongruence between a person’s gender identity (GI) and the assigned gender at birth (3,4). GI reflects a complex interplay of biological, psychological, environmental and cultural factors. Individuals with gender incongruence are a heterogeneous group and have been described throughout past decades with various terminologies such as transsexuals, gender queer, gender variant, gender incongruent, transgender (trans) individuals or people with gender identity disorder or gender dysphoria. Gender dysphoria refers to a profound distress or discomfort caused by the discrepancy between a person’s assigned sex at birth and gender identity (5). Not every transgender person suffers from gender dysphoria, and the need for psychological counseling and medical intervention may vary (5). For some, a social change can be sufficient. Many others will access transgender health care services, if available, to receive counseling and gender-affirming treatment. This can include hormone treatment and/or gender-affirming surgery.

Population-based estimates of gender incongruence range from 0,5% to 1,3% for birth-assigned male persons and from 0,4% to 1,2% for birth-assigned female persons (6)(7). The true prevalence is probably underestimated in this research due to the attached stigma. Luckily, over the past decades and at least in some regions, social acceptance has improved and has permitted individuals to explore their GI more freely. The number of trans people seeking gender-affirming care has increased in many specialized centers, and also other health care services are seeing a higher number of trans people (7,8). Still, sometimes there are many barriers for access to gender-affirming care or health care in general (9). Ideally, hormone treatment is started under the supervision of endocrinologists or other hormone-prescribing specialists (10). GAHT has been shown to be safe and effective in trans people in a growing body of short- and middle-term follow-up studies (11,12,21,13–20). Many previous review papers have included data from all available literature, with hormone treatments that are no longer in use, or advised against and therefore not-so-relevant anymore. The focus of this paper is to give an update on recent data available from larger cohorts, when available.

Gender-affirming hormone treatment

GAHT is feminizing or masculinizing hormone therapy through administration of exogenous hormones. For many trans and gender-nonconforming individuals, it is an essential intervention. It is important, possibly through counseling from a mental health practitioner, to identify therapy approach based on individual goals. The World Professional Association for Transgender Health (WPATH) Standards of Care, version 7 (5) suggests that adults seeking GAHT should meet the following criteria: persistent and well-documented gender incongruence, capacity to make an informed decision to consent to treatment, have reached the age of maturity and have controlled any mental health concerns. Adolescents seeking treatment may be eligible for puberty-suppressing hormones, often with gonadotrophin-releasing hormone agonists (GnRHa) as soon as pubertal changes have begun (Tanner Stage 2). It is recommended that adolescents experience the onset of puberty, as worsening of gender dysphoria can provide important information for the client, their family and the clinician (22). An intervention with puberty-suppressing hormones can help give adolescents more time to reflect on and experience their GI. The adolescent should have demonstrated long-lasting gender dysphoria and any coexisting psychological, medical or social problems should be stable enough to start treatment (5). Informed consent must be given by the adolescent and their parents or legal caretaker.

There are many variations in doses and types of hormones that are used to treat trans people. The Endocrine Society guidelines and the European Society for Sexual Medicine position statement (22)(23) provide protocols with specific information regarding the types of hormones and the suggested dosing.

Table 1 Gender-affirming hormone treatment in trans women (23)

	Mode of administration	Type	Total dose	Frequency
Estrogen	Oral	Estradiol (17beta-estradiol valerate)	2-6 mg	Once or twice daily
	Parenteral (intramuscular)	Estradiol valerate	5-30 mg	Every 1-2 weeks
		Estradiol cypionate	2-10 mg	Every week
	Transdermal	Estradiol patch	25-100 g/24 h	New patch every 3 days
		Estradiol gel	1.5 mg	Once or twice daily
Antiandrogens	Oral	Spironolactone	100 mg	Once or twice daily
		Cyproterone acetate	10-(50) mg	
GnRH agonists (gonadotropin-releasing hormone)	Intramuscular	Triptorelin	11.23 mg	3-monthly
	Intramuscular Or subcutaneous		3.75 mg	Monthly

Table 2 Gender-affirming hormone treatment in trans men (23)

	Mode of administration	Type	Total dose	Frequency
Testosterone	Intramuscular	Testosterone esters	200-250 mg	Every 2-3 weeks
		Testosterone	1,000 mg	Every 10-12

		undecanoate		weeks
	Subcutaneous	Testosterone esters	75-125 mg	Every week
	Transdermal	Androgen gel	25-100 mg	Once daily
	Oral	Testosterone undecanoate	160 mg	Once or twice daily
Progestational agents	Oral	Lynesterol	5-10 mg	Once daily
		Medroxyprogesterone	5-10 mg	Once daily
	Parenteral	Medroxyprogesterone	150 mg	Once every 3 months

Methodology

The methodology of this systematic review was based on the preferred reporting items for systematic reviews and meta analyses statement (24).

Protocol and registration

No previous protocol exists.

Eligibility criteria

Studies were selected only when participants were described as trans people or people with gender dysphoria and when there was a minimum of 10 participants. Only studies in English were considered eligible, as were cohort studies, cross-sectional studies and randomized controlled trials. Previous guidelines, review articles and meta-analyses were not included. Articles with no full text available through Ghent University were also excluded. Because the purpose of this review is to give an update of recent literature, the search strategy was limited in time. Only studies from January 1st 2015 until April 16th 2021 were included. For this paper, we focused on 5 outcomes related to GAHT: mental health and body image, body composition and contours, bone health, cardiovascular and thromboembolic safety, and cancer risk. Only studies concerning these topics were considered eligible. Sociological aspects, assessment, fertility and sexuality or other topics are therefore out of

the scope of this paper. We included studies on both adults and adolescents, and we focused especially on larger cohorts when available.

Search

The following electronic databases were searched from January 1st 2015 until April 16th 2021: MEDLINE (using Pubmed interface) and Embase (using embase.com interface). MeSH terms were adapted to the relevant database. MeSH terms used were “transgender”, “gender incongruence”, “gender dysphoria” and “transsexualism”. Variant terms used were “cross-sex hormone therapy”, “hormone therapy”, “gender-affirming hormone therapy”, “gender-affirming treatment”, “sex steroid hormones”, “ testosterone”, “anti-androgen”, “estrogen”, “trans men”, and “trans women”.

Study selection

Title-abstract and full-text screening were performed by both authors. The authors determined whether the article was appropriate according to the eligibility criteria, as described above. Eventually, a final list of articles was subject to data exploration. The study selection process is described in a flow diagram (fig. 1).

Data collection process

The authors were not blinded to the author, institution and publication source of trials at any time during the study. Data extraction (characteristics of the trials, baseline characteristics of the participants, the description of intervention and outcomes) was performed using piloted extraction forms by one of the authors (LD).

Results

Study selection

The PRISMA Flow Diagram (Figure 1) summarizes the review process and selection of the studies meeting inclusion criteria. Overall, we retrieved 2,033 records from the systematic searches in 2 databases. After exclusion of duplicates and non-relevant records, a total of 91 studies remained for data extraction. Reasons for excluding references during full-text screening were other language, study design or population.

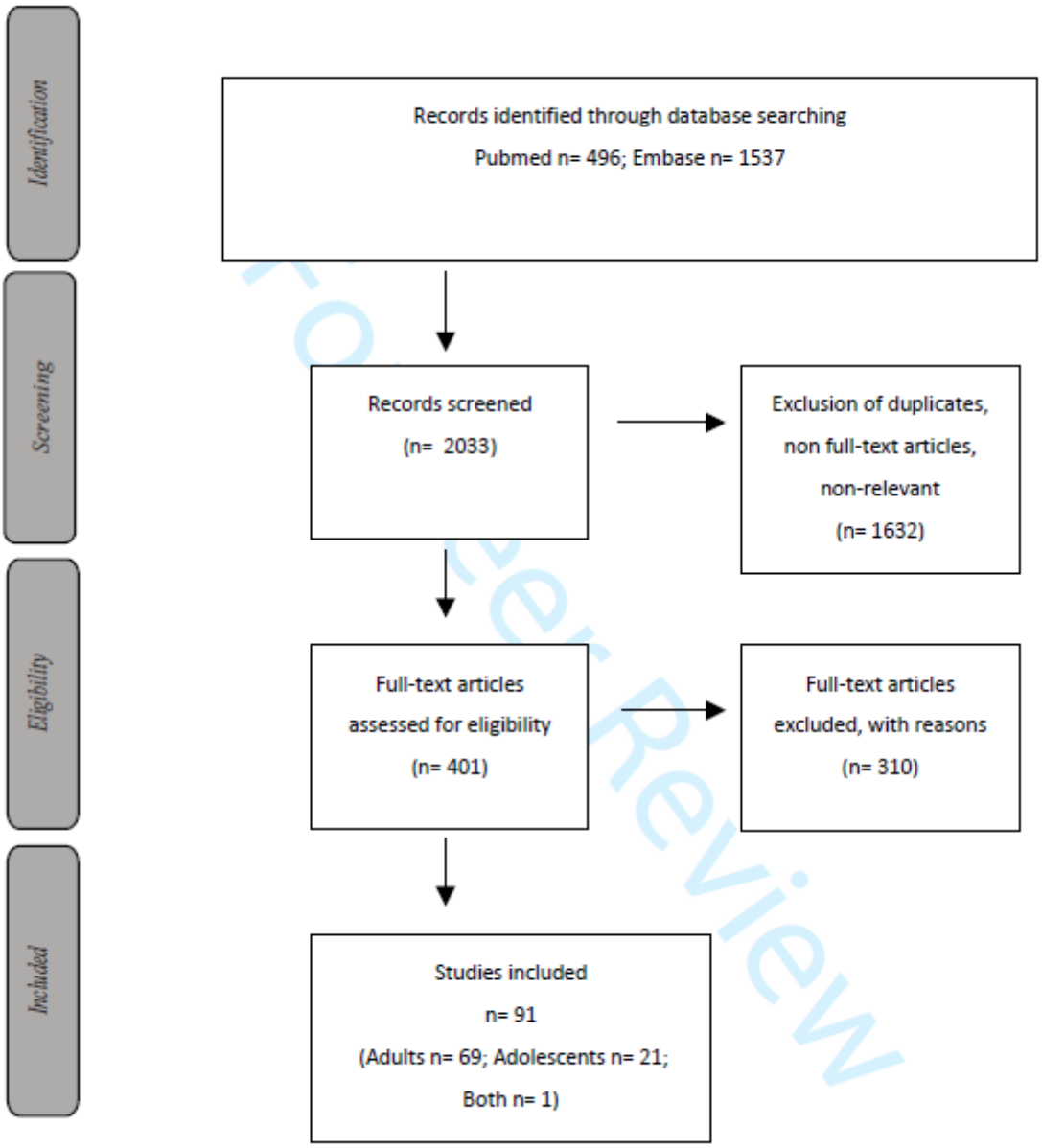

Figure 1

1. Mental Health and Body Image

Adults

Mental health diagnoses and reported substance dependence are prevalent in many adolescent and adult transgender people, underlining the need for mental health professionals in interdisciplinary gender teams.

Risk of mental health problems

An observational study from Chicago examining a population of 298 young trans women (aged 16 through 29, mean age = 23,4), of whom 72% were on GAHT (not further specified), showed an important prevalence of psychiatric problems (25). Of all participants, 41,5% had 1 or more mental health diagnoses or reported substance dependence (25). Prevalence of lifetime and current major depressive episodes was 35,4% and 14,7% respectively. Past 30-day suicidality was reported by 20,2%. Past 6-month generalized anxiety disorder and posttraumatic stress disorder occurred in 7,9% and 9,8% respectively. In a cross-sectional study from Michigan examining the rates of depression and anxiety in a cohort of 117 adult trans men (mean age = 25) of whom 82,5% were on testosterone treatment, a total of 70,3% screened positive for mild to severe depression (26). This was significantly higher compared to cis men and cis women. In the same cohort, 66,3% of participants screened positive for anxiety. A longitudinal US cohort study (27) with data from the 'Growing Up Today Study' investigated the rates of depression and anxiety in young adults (n = 7831, mean age = 25,7) from gender minority groups; use of GAHT was not specified in the gender minority group. Significant elevated levels of depressive distress and higher rates of anxiety symptoms (2- to 3-fold increased odds) were found when compared to non-gender minority respondents (27). Similar results were found in a matched retrospective cohort study from Boston, consisting of 106 trans men and 74 trans women, adults and adolescents combined, (12-29 years old, mean age = 19,6) of which 61,7% were on GAHT (not further specified) (28). They described a 2- to 3-fold increased risk of depression, anxiety disorder, suicidal ideation, suicide attempt, self-harm and having mental health treatment compared to cis controls. A large cross-sectional study from a community health center in Singapore (29) consisted of 17,521 cis men (mean age = 36,9), 9,288 cis women (mean age = 30,6), 987 trans men (mean age = 25,9), 1,002 trans women (mean age = 29,2) and 1,190 non-binary persons (428 male at birth, mean age = 28,9 and 762 female at birth, mean age = 25,4). They reported a significantly increased risk of depression, anxiety and substance abuse disorder in adult non-binary people compared to cis and trans people. They also found a higher proportion of trans women (34,6%) and non-binary people (33,7 and 36,8%) to have a depressive

disorder compared to trans men (27,5%), cis women (16,1%) and cis men (14,1%). In a chart study of 8,263 referrals to the Amsterdam gender clinic, 41 trans women and 8 trans men died by suicide, clearly more than the general population by comparison(30). A large cross-sectional study combining datasets (n = 641.860, of which n = 2,811 transgender participants) showed transgender and gender-diverse individuals were 3,03 to 6,36 times as likely to be autistic than cisgender individuals(31).

Potential causality

In an attempt to explain such high numbers, a cross-sectional study conducted in Madrid examined protective, violence, health and well-being factors in trans (n = 180), non-binary (n = 70) and cis (n = 532) people with a mean age of 20,3 (14 to 25 years old). They found trans people to be at significantly increased risk of violence exposure and employment discrimination. Approximately 40-50% of trans and gender minority individuals suffered verbal and/or physical attacks (32). Similarly, Real et al. (33) performed a study in Porto Alegre (Brazil) and found that 34 trans women on hormone therapy (not further specified, mean age = 29) experienced significantly more discrimination and emotional and sexual abuse compared to cis men controls. Jäggi et al. (34) examined the health consequences of gender minority stigmatization in 143 trans people (mean age = 45,2) without hormone treatment in Switzerland and found a significant relation between depressive symptoms and minority stress (e.g. gender-related discrimination, rejection and victimization). Internalized transphobia, negative expectations and nondisclosure also had a positive relation with more depressive symptoms (34).

Body Image

A European multi-center study, with data collection from the European Network for the Investigation of Gender Incongruence (ENIGI), reported cross-sectional differences in satisfaction with overall appearance in trans people compared to a control sample (35). Trans men (n = 135, mean age = 23,8) scored significantly worse on all female body feature scales, and most prominently on the subscales genitalia, androgen-responsive features, body hair and body shape (35). For trans women (n = 115, mean age = 25,0), differences were found for each of the 6 male body feature subscales, and most prominently on the subscales androgen-responsive features, genitalia, androgenic hair, facial features, extremities and upper body (35). Becker et al. (36) examined body image in a cross-sectional multi-center study with and without GAHT both in trans adults (n = 120) and adolescents (n = 82). The mean age of adult trans men was 35,4, and the mean age of adult trans women was 34,9. Individuals who had undergone gender-affirming treatment (not further specified) experienced significantly improved body image (attractiveness/self-confidence and accentuation of body appearance)(36).

Effect of GAHT on mental health problems

Defreyne et al. (37) examined the effect of GAHT on anger intensity in trans men (n = 440, median age = 22,0) and trans women (n = 468, median age = 28,0) during a 3-year follow-up period. They found neither an increase in anger intensity nor a correlation between anger intensity and serum testosterone levels. The impact of GAHT on affect was also explored by Matthys et al. (38). Median total positive affect decreased in trans women (n = 451, mean age = 27,0) and trans men (n= 422, mean age = 22,0) in the first 3 months, but there was no significant difference after 36 months compared to baseline. This was in contrast with the reduction in gender dysphoria after initiation of GAHT. Experiencing social difficulties was hypothesized as an explanatory factor. A long-term follow-up study in Ulm (Germany) from Rupping et al. examined the effects of GAHT (not further specified) in 35 trans women (mean age = 52,9) and 36 trans men (mean age = 41,2) after at least 10 years since their first contact. They found that participants had significantly fewer psychological problems and interpersonal difficulties at follow-up than at the time of the initial consultation and they showed increases in life satisfaction (39). None expressed regrets about the treatment. Similar results were found in previous research. Data from 54 Italian trans people with a 2-year follow-up showed similar findings: less psychopathology, less body uneasiness and depressive symptoms with GAHT compared to those without (40). More recently, Aldridge et al. (41) showed the important mental health benefits of 18 months of GAHT (and mental health care), including decreased depression and less anxiety were confirmed in a UK cohort (n = 178).

Adolescents

Risk of mental health problems

In a cohort study conducted in California and Georgia with 588 transfeminine and 745 transmasculine children and adolescents (3-17 years old) at their initial presentation (i.e. not on hormone therapy) high rates of attention deficit (15% and 16% respectively) and depressive disorders (49% and 62% respectively) were reported, with a prevalence significantly higher than in matched reference groups (42). Lifetime suicidal ideation was 7,5% and 10,4% respectively (42). In a cross-clinic comparative study between Toronto and Amsterdam, de Vries et al. reported poorer peer relations, lower socioeconomic background and lower IQ to be predictors of behavioral and emotional problems in adolescents with gender dysphoria and without GAHT (n = 316, mean age = 15) (43). A cross-sectional study with 46 Italian adolescents (mean age = 16,0 $\pm$ 1,49), performed on the first day (i.e. no hormone therapy) of outpatient care, reported significantly higher levels of body uneasiness, as well as worse psychological functioning compared to a non-referred group of

adolescents (44). A high prevalence of suicidal ideation and attempts was also described. Up to 83,3% of transfeminine and 90% of transmasculine adolescents reported suicidal ideation in the past; for suicide attempts, this was 16,7% and 11,1% respectively (44). The overlap of gender diversity and autism spectrum disorders (ASD) has been a focus of research lately. In an Australian cohort with 859 trans young people between 14-25 years, 22,5% of participants had ever received a diagnosis of ASD by a health professional, pointing to the necessity of clinicians being aware of possible co-occurrence.

Potential causality

Risk factors for mental health problems have been described, among which were experience with physical and verbal abuse, social isolation, low self-esteem, poor peer relations and discrimination, while parent connectedness, social support, school safety and belonging, and the ability to use the chosen name were resilience-promoting factors (45).

Effect of GAHT on mental health problems

In a prospective observational study from London, Carmichael et al. (46) reported overall positive patient experience in 44 transgender adolescents (12-15 years old, median age = 13,6) with GnRHa treatment after 12 and 36 months. This is in line with previous studies. In a group of 272 Dutch trans adolescents there were increased behavioral and emotional problems in those referred to the gender clinic, but after the start of puberty blocking and GAHT, similar or better psychological functioning (i.e. fewer emotional and behavioral problems) was measured, compared to cis controls (47). Significantly improved global psychological functioning was reported by Costa et al. (48) in 201 participants referred to the Gender Identity Development Service in London, UK who started puberty suppression. Along the same lines, the US-based study by Kuper (49) showed significant improvements in body dissatisfaction, small to moderate improvements in self-reported depressive symptoms and small improvements in total anxiety symptoms for 149 adolescents (mean age = 14,9) with puberty suppression (n = 25) or with feminizing or masculinizing hormone therapy (n = 123). Similar to the findings in adults, Becker et al. (36) reported significantly improved body image in adolescents (trans boys mean age = 16,9; trans girls = 16,5) on GAHT. The above-mentioned study by Kuper (49) confirmed the reduction in body dissatisfaction.

Supplementary table 1: mental health and body image

2. Body Composition and Contours

The main purpose of GAHT is to improve the quality of life, primarily through alignment of the physical features with the gender identity. Administration of GAHT results in body composition changes. In trans men, GAHT induces virilization, such as increased muscle mass and strength and reduced fat mass. In trans women, GAHT results in feminization, such as breast development, redistribution of body fat and a decrease of lean body mass.

Adults

A retrospective cross-sectional study identified the most anticipated changes induced by GAHT(50). In adult trans women (n = 48, mean age = 28), this was breast development (35,4%) followed by gynoid fat deposition (29,2%). In trans men (n = 336, mean age = 24), the results were less focused on body composition; the most anticipated changes were cessation of menses (52,7%), followed by a deepening of the voice (32,4%) (50). To study prospective breast development during the first year of GAHT, 229 trans women (median age = 28) were described as part of the ENIGI study (51). GAHT consisted of cyproterone acetate (CPA) or spironolactone together with estrogens (oral or transdermal). Mean breast-chest difference increased to 7,9 cm (± 3,1 cm) after 1 year of hormone treatment, resulting in less than an AAA cup size (48,7%), AAA (26,4%), AA (14,2%), A (7,1%) and (3,6%) larger than an A cup. Breast development mainly occurred in the first 6 months (51). However, although the change in breast-chest difference plateaued after 9 months, sustained breast growth and development was seen during a 3-year follow-up in 69 trans women (median age = 26), leaving 58% of trans women satisfied with the result (52). Still, from a retrospective Amsterdam study of 773 trans women (median age = 50), 80% had chosen or considered breast augmentation as part of their gender-affirming treatment (53).

Klaver et al., also part of ENIGI, examined the effects of GAHT in adult trans men (n = 162, mean age = 24) and trans women (n= 179, mean age = 29) on separate body regions and whether these changes altered body shape; mean follow-up time was 380 and 377 days respectively. GAHT caused a more feminine body fat distribution and a lower waist-to-hip ratio in trans women, and a more masculine body fat distribution with a lower hip circumference in trans men. In trans women, the waist-to-hip ratio decreased (-0,03, 95% CI -0,04;-0,02) mostly due to an increase in hip circumference (+3,2 cm, 95% CI 2,3;4,0). This was based on an 18% (95% CI 13;23%) increase of body fat in the android region, an increase of 42% (95% CI 37%;46%) in the leg region and 34% (95% CI 29%;38%) in the gynoid region. (54). Similar results were found in 142 trans women (mean age = 33,7) in a 3-month follow up from Brazil (55). In trans men, a decrease in hip circumference (-1,9cm, 95% CI -3,1, -0,7) led to in an increase of the waist-to-hip ratio. This was based on a decrease of 16%

(95% CI -19%; -14%) body fat in the leg region, 14% (95% CI -16;-12%) in the gynoid region and no change in the android region (+1%, 95% CI -3%;+5%) (54). An observational Swedish cohort study (56), from Wiik et al., examining the effect of GAHT on muscle strength, size and composition with CT and MRI imaging in trans men (n = 12, mean age = 25) reported a thigh muscle volume increase of 15% and a quadriceps cross-sectional area increase of 15%. In contrast, these muscle volumes decreased during the intervention in trans women (n = 11, mean age = 27); with a decrease of total thigh muscle volume of 15% and quadriceps cross-sectional area of 4% (56). Equally, in a study with 278 trans men (mean age = 23) and 249 trans women (mean age = 28) with 1-year follow-up, grip strength increased in trans men (+6,1kg (95% CI +5,5; +6,7)) and decreased in trans women (-1,8kg (95% CI -2,6; -1,0))(57). Yun et al. reported overall similar findings in 11 Korean trans women: a general increase in fat mass focused on the gynoid region and a decrease in lean body mass and handgrip strength (58). In an Australian cross-sectional study, 43 trans men (mean age = 28, 8) were compared to 48 cis women (mean age = 28,1), and 41 trans women (mean age = 41,1) were compared to 30 cis men (mean age 32,0). All transgender participants had been > 12 months on GAHT. They found an increase of 7,8 kg (95% CI 4,0;11,5) in lean mass and higher android/gynoid fat ratio in trans men compared to cis women. Trans women experienced a decrease of lean mass of 6,9kg (95% CI -10,6;-3,1) and a lower android/gynoid fat ratio compared to cis men (59). Another ENIGI study (60) evaluated self-reported symptoms during the first year of GAHT in trans men (n = 193, median age = 23) and in trans women (n= 205, median age = 29). Weight gain (>5kg) was reported by 44,4% of trans men after 3 months of testosterone therapy and by 26,3% of trans women after 3 months of estrogen + CPA therapy.

In contrast, Gava et al. (61) performed an RCT and examined the effect of IM testosterone undecanoate, combined with dutasteride or placebo on body composition and strength in trans men (n = 14, mean age = 35,0 and 36,2 in both groups). The study reported no changes in BMI and body weight after 1 year of treatment. However, body composition did change, fat percentage decreased (-4%) and lean mass increased (+1,6%), also resulting in increased handgrip strength (61). A lack of changes with GAHT in body composition was examined in ENIGI(62); in 323 trans men (mean age = 21), a lack of change after 24 months of GAHT was observed in 20,2%. Low mean testosterone and high mean LH levels were associated with no change in this group. In 288 trans women (mean age= 25), 9,4% reported a lack of change in body composition; this was not associated with serum hormone levels. Long-term studies on the effects on insulin sensitivity are still missing, but short-term studies in smaller cohorts suggest an increase with masculinization and a decrease with feminization, also reflected in post-OGTT incretin responses (63).

Adolescents

The puberty-blocking effects of GnRHa-only in trans girls (n = 49, median age = 13,6) and trans boys (n = 67, median age = 14,2) were examined, and height, weight, fat mass and BMI were found to increase and lean body mass to decrease in the first year of treatment (12). In late-pubertal adolescents, progestins (lynestrenol and CPA) are used in some centers to attenuate endogenous hormonal effects (64). From a Belgian study of 44 trans boys and 21 trans girls treated this way for a mean duration of just less than 1 year, lean mass increased (+ 3,2 kg) as did grip strength (+ 3 kg) in trans boys, and loss of lean mass (- 2,2 kg) was shown in trans girls together with gain of fat mass (+1,5 kg) and decreased grip strength (65). When GnRHa pubertal induction with estradiol was introduced, breast development started in 3 months in 83% of participants (n = 22), and after 3 years 86% had a Tanner breast stage 4 to 5. Hip circumference increased and waist-to-hip ratio decreased (66). When GAHT was added at age 16 years, and trans boys (n = 121) and trans girls (n = 71) were examined at age 22, waist-to-hip ratio and body composition had changed toward the affirmed sex. Total body fat increased by 10%, while lean body mass decreased by 10% in trans girls, and waist-to-hip ratio decreased. In trans boys, total body fat decreased by 3%, while lean body mass increased by 3%, as did waist-to-hip ratio (67). A cross-sectional study from Colorado (68) evaluated body composition in trans boys (n = 19, mean age = 17 $\pm$ 1,4) and trans girls (n = 14, mean age = 16,3 $\pm$ 1,4). Here, trans boys were on testosterone therapy (I.M. or S.C.); all trans girls were on estradiol, with 4 trans girls also on GnRHa, 7 on spironolactone and 1 on medroxyprogesterone. Included patients were on GAHT at least 3 months, with mean treatment duration of 11,2 months in trans men and of 12,3 months in trans women. Total body fat was lower in trans boys (29% $\pm$ 7% vs 33% $\pm$ 7%), and they had higher lean mass compared to cis girls (68% $\pm$ 7% vs 64% $\pm$ 7%) (68). Trans girls had higher body fat (28% $\pm$ 6% vs 20% $\pm$ 10%) and lower lean mass than cis boys (69% $\pm$ 5% vs 77% $\pm$ 9%) (68). Insulin sensitivity was not different between trans boys and cis controls, but trans girls were more insulin resistant than cis boys.

Supplementary table 2: Body composition and contours

3. Bone Health

Hormonal treatment in trans persons can affect bone health. The effect on periosteal and endocortical growth in bone geometry is partly regulated by sex steroids. As GAHT changes the hormonal exposure, it plays a key role in bone turnover and preserving bone mineral density (BMD) (69)(70).

Adults

Bone mineral density in trans men

The ENIGI study investigated the first-year effect of GAHT in trans men (n = 199, median age = 24). Total hip (TH) BMD increased by 1,04% (95% CI 0,64-1,44); the increase in lumbar spine (LS) was larger in trans men aged >50 years compared with trans men <50 years, and no increase was observed at the femoral neck (FN) (71). When changes in BMD were investigated in 543 trans men (median age = 25) during the first 10 years of GAHT, no changes in LS BMD were noted, but the LS Z-score increased by + 0,34 (72). No changes in bone geometry were found in a group of 473 trans men (baseline, or after 5, 15 or 25 years of GAHT), and in hip structure analysis a trabecular bone score decreased(73). In the ENIGI cohort of 23 trans men (mean age = 27) treated 1 year with IM testosterone undecanoate, together with a gain in muscle mass (+10,4%) and loss of fat mass (-9,7%), increased levels of procollagen-1 N-terminal peptide (P1NP) and carboxy-terminal telopeptide of type 1 collagen (1CTP) and a small but significant increase in trabecular BMD at the distal radius and total hip were seen (74). The increase of trabecular BMD might have been an indirect effect of testosterone therapy due to an increase in muscle mass, which imposed a higher strain on the bone and thereafter stimulated higher bone modeling and remodeling (74). Secondly, testosterone may also have acted on BMD indirectly through estrogen aromatization (74).

In trans men, bone turnover markers P1NP, alkaline phosphatase (ALP) and sclerostin increased by 33%, 16% and 15% respectively after 1 year of therapy, but opposite effects were found on bone turnover in trans men aged ≥50 years compared to younger trans men. In trans men aged ≥50 years, decreases of P1NP, 1CTP and sclerostin of 19%, 32% and 10% respectively were described. The older group of trans men benefited the most from GAHT as they were assumed to be estrogen-deficient as a result of their postmenopausal state at baseline (70). The increase of estrogen concentration after the aromatization of testosterone resulted in decreased bone resorption. In contrast, Gava et al. (61) reported no differences in bone metabolism and BMD after 1 year of treatment with intramuscular testosterone undecanoate. A nationwide cohort study in the Netherlands investigated 1,036 trans men, aged 40 +/- 14 years, using GAHT for median 9 years. Fracture risk was not increased (75).

Bone mineral density in trans women

In a prospective observational study from Ghent, Belgium examining BMD in adult trans women (n = 49, median age = 33) before GAHT, after 1 year and after 2 years, an increase at FN, radius, LS and total body, but not at TH was found (76). From the same ENIGI cohort with 231 trans women (median age = 28), investigations into the first-year effect of GAHT showed an increase of BMD in LS (+3,67%; 95% CI 3,20-4,13), TH (+0,97%; 95% CI 0,62-1,31) and FN (+1,86%; 95% CI 1,41-2,31) (71). In

a cohort of 50 trans women treated with CPA or leuprolide acetate and 1 or 2 mg oral estradiol for 5 years, LS and total body BMD increased after 3 years (77). In comparison with cis women, trans women (n = 31, median age = 29) on GAHT with estrogens (12,9%), CPA or spironolactone (16,1%) or a combination of both (70,9%) had lower Z-scores at LS (0,26 ± 1,42 vs. 0,50 ±1,19) and at FN (-0,41 ± 0,95 vs. 0,29 ± 1,04) (78). When changes in BMD were investigated in 543 trans women (median age = 25) during the first 10 years of GAHT, no change in LS BMD was noted, but the LS Z-score increased by + 0,34 (72). Of note, prior to the start of GAHT, 21,9% of trans women had low BMD for age, confirming earlier studies by Van Caenegem (79). No changes in bone geometry were found in a group of 535 trans women (at baseline, or after 5, 15 or 25 years of GAHT), and in hip structure analysis, a trabecular bone score increased (73). During the first year of GAHT (n = 121 trans women, median age = 30), ALP, 1CTP and sclerostin decreased by 19%, 11% and 8% respectively (70). The prevalence of low bone mass was described in a group of 57 trans women, aged 45,3 ±11,3, at least 2 years after gender-confirming surgery. Low bone mass (Z-score ≤2) was present in 40% compared to the sex assigned at birth, with lower serum estradiol levels and low compliance to estrogen treatment in comparison to those with higher bone mass. Intermediate-to-high fracture risk was found in 14% of the sample. A high percentage of hypovitaminosis D was seen (93%)(80). From a nationwide cohort study in the Netherlands with 1,089 trans women aged < 50(mean 38 ± 9) and 934 women aged ≥ 50(mean 60 ± 8) using GAHT for a median of 8 and 19 years respectively, the fracture risk was higher in older trans women, compared to age-matched reference cis men (OR 1,90). In young trans women, fracture risk tended to be increased compared with age-matched reference cis women (OR 1,49) (75). An ENIGI study by Vlot et al. (70) found decreases in ALP, CTx and sclerostin by 19% (95% CI, –21 to –16), 11% (95% CI, –18 to –4) and 8% (95% CI, –13 to –4) respectively after 1 year of HT.

Adolescents

Bone mineral density

The effect on bone density of GnRHa as monotherapy in 44 12-15 year olds with persistent and severe gender dysphoria was studied at baseline and yearly thereafter (up to 36 months)(46). There was no change from baseline in LS BMD at 12 months, nor in TH BMD at 34 and 36 months, but at 24 months LS bone mineral content and BMD were higher than at baseline. When 51 trans girls and 70 trans boys were treated with GnRHa treatment alone, bone mineral apparent density (BMAD) stabilized and/or showed a small decrease, but Z-scores decreased in all groups as did bone markers (81). During 3 years of combined GnRHa and GAHT, a significant increase in BMAD was found. Z-scores remained below 0 in trans girls, but normalized in trans boys (81). CPA alone during 10,6 (5 to

31) months in 21 trans girls also limited normal bone expansion and pubertal bone mass accrual, mostly at the LS (65). Klink et al. (82) performed a longitudinal observational study to determine the effects of GnRHa and GAHT on BMD in trans girls (n = 15, median age = 14,9 at baseline) and trans boys (n= 19, median age = 15,0). In trans girls, Z-scores decreased (-0,8 to -1,4) during GnRHa treatment, but not significantly. During GAHT, absolute LS BMD increased, but the Z-score at 22 years old was still lower than at the start of the treatment (82). In trans boys, both LS and FN BMD and the respective Z-scores showed a trend decrease (+0,2 to -0,3) during GnRHa monotherapy, and increased between the start of GAHT and at 22 years of age. But also here, LS BMD Z-scores were lower compared to the start of GnRHa treatment (not significant) (82). Of note, in trans women, the LS BMD Z-score was below the population mean at the start of the treatment, similar to the findings in 62 trans boys reported by Stoffers et al.(83). Less participation in sports activities with a negative impact on mechanical loading and lower 25(OH)-vitamin D status were suggested as causal factors. A retrospective cohort study from van der Loos et al. (69) investigated the changes in bone geometry among trans adolescents (106 trans girls and 216 trans boys) using GnRHa and GAHT when started in different stages of puberty. After ≥ 2 years of GAHT, they found a similar subperiosteal width and endocortical diameter to the experienced gender when GnRHa was started during early puberty. Trans participants starting in mid- and late puberty remained within the reference curve of the sex assigned at birth (69).

Bone turnover markers

Vlot et al. (84) investigated the effects of GnRHa and GAHT on bone turnover markers and bone mineral apparent density (BMAD) in adolescents. In this study from Amsterdam, there were 34 trans boys (median age = 15,1) and 22 trans girls (median age = 13,5) with a follow-up duration of 24 months. They found decreases of P1NP and 1CTP during GnRHa treatment, indicating decreased bone turnover with GnRHa. In both younger (<14 years old) trans girls, and older and younger (< and > 14 years old) trans boys, BMAD Z-scores of the lumbar spine (LS) decreased during GnRHa (84). Two years after initiation of GAHT, the bone turnover markers decreased further in all groups except for trans boys >14 years old, but the BMAD increased and Z-scores returned to normal, especially at the LS (84).

Supplementary table 3: Bone Health

4. Cardiovascular and thromboembolic safety

Adults

Acute myocardial infarction

The Behavioral Risk Factor Surveillance System cross-sectional data reported a >2-fold and 4-fold increase in the rate of myocardial infarction in trans men (n = 1267, mean age = 51,4) compared to cis men and cis women respectively, even after adjusting for other cardiovascular risk factors (85). Trans women (n = 1788, mean age = 53,1) had a >2-fold increase in the rate of myocardial infarction compared to cis women, but there was no difference compared to cis men (85). An important limitation of this Washington DC-based study is the lack of detailed information on the GAHT used.

VTE and stroke

Compared to cis controls, a US cohort of 2,842 transfeminine individuals with a mean follow-up of 4,0 years showed a significantly higher incidence of ischemic stroke and VTE. No conclusions could be reached for 2,118 transmasculine participants (86). A retrospective cohort study from the Netherlands also found a higher risk for strokes and VTEs in 2,517 adult trans women (median age = 30, mean and median follow-up durations = 9,07 and 5,95 years) compared to both reference cis women and cis men, and a higher risk of myocardial infarctions in adult trans women and adult trans men (n = 1358, median age = 23, mean and median follow-up durations 8,10 and 4,10 years) when compared to reference cis women (87). In a retrospective U.S. cohort, Pyra et al. (88) reported a thromboembolism incidence of 0,8% in trans women on estrogen with or without an antiandrogen (n=2509, median age = 30), with no association between thromboembolism and hormone therapy as assessed by blood concentrations. However, recent progestin prescriptions were associated with increased odds of thromboembolism (aOR 2,95; 95% CI 1,02-8,57) (88). Only 0,2% of trans men on testosterone (n = 1893, median age = 26) developed thromboembolism in this study (88). A retrospective study investigated the effect of estrogen (oral, transdermal or intramuscular) therapy on the postoperative risk of developing venous thromboembolism (VTE) events in adult trans women (n = 662), trans men (n = 232) and non-binary persons (n = 25) (89). Seventy-one % of trans women were taking spironolactone and 19% were on CPA in combination with estrogen therapy. Kozato et al. reported no increased risk of VTE when estrogen treatment was continued throughout surgery compared to suspended estrogen treatment (89). No difference was detected in VTE rates

among different routes of estradiol administration. In a German cohort of 155 trans women, 1,9% developed VTE (21). Lim et al. (90) demonstrated hypercoagulable parameters on both thromboelastography and thrombin generation in trans women (n = 26, median age = 32,8) on estrogen treatment (with or without CPA/spironolactone) compared to cis men with a shift towards cis female parameters. The study was not designed to determine whether these changes influence clinical thrombotic risks. The effect of GAHT on coagulation parameters was examined in a Dutch study with 98 trans women and 100 trans men, before and after 12 months. Trans women had more procoagulant profiles with increases in Factor IX, XI and a decrease in protein C. The route of administration (oral vs. transdermal) and age influenced the changes in coagulation parameters. Reassuringly, changes in trans men were not procoagulant overall, and differences between transdermal versus intramuscular testosterone were small. The effect of GAHT on blood pressure was negligible in all studies (91)(88)(92).

Risk factors trans men

A prospective cohort study, part of ENIGI, examined the effects of 1 year of hormone treatment on serum lipid levels (91). Unfavorable changes in lipid profiles were observed in trans men (n = 188, mean age = 26,4) with an increase of total cholesterol (4,1%; 95% CI 1,5-6,6), LDL-cholesterol (13%; 95% CI 9,2-16,8) and triglycerides (36,9%; 95% CI 29,8-44,1). HDL-cholesterol decreased by 10,8% (95% CI -14,0 to -7,6). Similar results were reported in Liu et al. (Taiwan) and Allen et al. (Dallas, Texas) (93)(94) with an increase in LDL-cholesterol and a decrease in HDL-cholesterol in trans men during the first year of testosterone therapy. Liu et al. (93) examined the effects of 1 year of testosterone therapy in trans men (n = 65, mean age = 27,9) and found an increase in LDL-cholesterol from 124,3 mg/dl to 131,3 mg/dl and a decrease in HDL-cholesterol from 57,9 mg/dl to 52,8 mg/dl. Allen et al. found an increase in LDL-cholesterol and a relative decrease in HDL-cholesterol in trans men (n = 91, mean age = 27,8) and an increase of HDL-cholesterol in 126 trans women. In a study comparing testosterone undecanoate and testosterone enanthate injections in 50 trans men aged 21-42 years, similar lipid changes were found—an increase of total cholesterol, triglycerides and LDL-cholesterol, and a slight reduction in HDL-cholesterol (95). Kirisawa et al. (96) found no difference on triglycerides and total cholesterol in Japanese trans men (n = 85, mean age = 27,1) on testosterone enanthate (125mg-250mg) after 24 months. When cardiovascular risk was calculated according to the Framingham 30-year cardiovascular disease risk estimate, changes in lipid profile over time determined the risk increase in trans men; this was not the case in trans women (97).

Risk factors trans women

In trans women (n = 242, mean age = 32,3), van Velzen et al. (91) reported favorable effects with decreased levels of total cholesterol (-9,7%; 95% CI -11,3 to -8,1), LDL-cholesterol (-6,0%; 95% CI -8,6 to 3,6), HDL- cholesterol (-9,3%; 95% CI -11,4 to -7,3) and triglycerides (-10,2%; 95% CI -14,5 to -5,9). Liu et al. showed a trend decrease of LDL-cholesterol from 104,2mg/dl to 100,4mg/dl and a trend increase of HDL-cholesterol from 63,9mg/dl to 65,0mg/dl in trans women (n = 45, mean age = 26,0) during the first year of GAHT. Similarly, Allen et al. found no significant changes in LDL-cholesterol, HDL-cholesterol or triglycerides (n = 126, mean age = 31,1) (94). In a retrospective study, trans women (mean age = 34,4) on either spironolactone (n = 82) or CPA (n = 31) were compared with regard to the effects on HDL-cholesterol (98). After 12 months, HDL-cholesterol had increased by 0,10 mmol/l in the spironolactone group and decreased by 0,07 mmol/l in the CPA group (98). Gava et al. examined the effect of CPA versus GnRHa in combination with transdermal estradiol in trans women (n = 40, mean age CPA group = 32,9, mean age GnRHa group = 29,4) on biochemical parameters (14). They found a decrease in total cholesterol in the CPA group and an increase in the GnRHa group, a decrease of HDL-cholesterol in the CPA group and an increase in the GnRHa group, and no significant change in triglycerides and LDL-cholesterol. Similar findings were reported when comparing spironolactone (n = 82) to CPA (n = 31), with increases of HDL-cholesterol with spironolactone, and decreases with CPA (98). A worse cardiovascular risk profile was also described with CPA inducing lower HDL-cholesterol compared to spironolactone or GnRHa as anti-androgen in a group of 126 trans women (99). Insulin sensitivity is yet another cardiometabolic risk factor, but the literature on this topic is limited. The effects of GAHT on insulin sensitivity have been described above. Of note, so far most of the cardiovascular data described above pertain to still relatively young groups and do not make use of contemporary noninvasive risk stratification, for example by using coronary calcium score calculation. Only 1 cohort of 20 trans men with 12 months of GAHT had an evaluation with flow-mediated dilation and intima-media thickness (IMT) by carotid ultrasound, showing a higher mean-maximum common IMT(100). The number of cardiovascular events is still relatively small, but caution is needed as trans persons will age. Aging trans men and women should be monitored in a similar way as cis people, and management should focus on BMI, smoking, sedentary lifestyle, hypercholesterolemia, type 2 diabetes and hypertension.

Hematocrit

Over 54 weeks of follow-up in adult trans men treated with intramuscular testosterone undecanoate after 1 year, an increase in hematocrit from 40,7% (± 1,9%) to 44,8% (± 2,6%) and an increase in hemoglobin from 13,8 g/dl (± 0,7 g/dl) to 14,8 g/dl (± 1,0 g/dl) was reported (61). Defreyne et al. (101) reported analogous results with increased serum hematocrit levels (+4,9%, 95% CI 3,82-5,25) during the first year of testosterone treatment (n = 192, mean age = 41,1), with the most

pronounced increase in the first 3 months (+2,7%, 95% CI 1,94-3,29). Trans men receiving shorter-acting testosterone esters had a greater increase (Δ 0,8%) in serum hematocrit levels compared to trans men receiving 3 monthly injections of testosterone undecanoate, which might be explained by pharmacokinetic profiles (101). A long-term follow-up cohort study from Madsen et al. (102) reported erythrocytosis in 11% (hematocrit >0,50 L/l), 3,7% (>0,52 l/l) and 0,5% (> 0,54l/l) of trans men (n = 1037, mean age at start of GAHT = 22,5) on testosterone therapy. The largest increase was seen in the first year of testosterone therapy, but the probability of developing erythrocytosis still increased in the following 20 years (10% after 1 year, 38% after 10 years)(102). In this study, in contrast to Defreyne et al. (101), long-acting undecanoate showed higher risk for hematocrit levels >0,50 l/l (OR 2,9, 95% CI 1,7-5,0) compared to short-acting ester injections. Erythrocytosis was 5,6% in a German cohort with 233 trans men (21). In contrast, in trans women (n = 239, mean age = 45,0) on estrogen in combination with CPA, a decrease of serum hematocrit levels by 4,1% (95% CI 3,50-4,37) was seen after 3 months, after which only small decreases were observed during follow-up (101). Along the same lines, Allen et al. (94) reported increases of hematocrit levels in trans men (n = 91, mean age = 27,8) during testosterone treatment and decreases in trans women (n = 126, mean age = 31,1) during estrogen treatment with or without an anti-androgen, with no changes after 6 months of therapy when compared to measurements after 5 years of GAHT. A similar pattern was shown in 183 trans women and 119 trans men with follow-up longer than 6 months (103).

Adolescents

A retrospective cohort compared transgender adolescents (71 trans girls, 121 trans boys) at the age of 22, who started GnRHa treatment at a mean age of 15, to cis peers. For trans girls, changes in BMI, systolic and diastolic blood pressure, glucose, HOMA-IR and lipid values were similar or more favorable compared to peers. The same was true for trans boys, showing reassuring data regarding cardiovascular risk. Body weight management was at the center of attention, as obesity prevalence was 9,9% in trans girls and 6,6% in boys, compared to 2,2 and 3,0% in cis girls and boys (104). No significant changes in blood pressure were seen in 28 trans adolescents treated with oral estrogen for 1 year or more (66), and this was confirmed in a retrospective study with 19 trans girls treated with GnRHa, of whom 15 continued with estrogen treatment (105).

Jarin et al. (92) found a significant decrease in mean HDL-cholesterol level from 50,2 to 45,0 mg/dl in trans male adolescents (n = 72, mean age = 16). In trans girls (n = 44), no significant changes were found. Trans male adolescents (n = 45) who were solely treated with androgenic progestins (lynestrenol) showed a significantly more unfavorable lipid profile compared to lynestrenol in

combination with testosterone esters. Jarin et al. (92) examined the effects of 6 months of GAHT in trans boys (n = 72, mean age = 16) on testosterone (25-100mg weekly, subcutaneous) and in trans girls (n = 44, mean age = 18) on estrogen therapy (orally 1-8mg per day, intramuscularly 20 - 80mg per month or transdermally 0,025 - 0,200mg weekly) with or without anti-androgens. They reported significant increases in hemoglobin (from 13,5 g/dl to 15,0 g/dl) and hematocrit (from 39,4% to 44,5%) in trans boys, but no changes in trans girls.

Supplementary table 4: Cardiovascular and thromboembolic safety

5. Cancer risk

The observed cancer risk in trans people does not exceed the known cancer-risk differences between men and women, and practitioners must be aware that trans people may develop problems in relevant anatomy. Some have advocated following cancer-screening guidelines according to the organs present and relevant risk behavior (110).

GAHT may affect the risk of hormone-sensitive cancer types, and a gender difference is described for several other cancer types, suggesting a role of sex hormones. Epidemiological data for trans people are still limited, which is why approaches are often drawn from cis screening guidelines. Also, to single out the effect of GAHT is impossible as age is an important determinant, lifestyle interventions are part of good clinical practice, and social and mental health changes occur and relate to changed health-seeking behavior. Cancer screening rates have been described and compared with rates among cis people in 120 trans participants (age = 21-74, GAHT not specified) of whom 86 were eligible for cervical, 30 for breast and 38 for colorectal screening. Lower screening rates for cervical (56% vs. 72%, 95% CI 0,25 to 0,62), breast (33% vs. 65%, 95% CI 0,12 to 0,59) and colorectal cancer (55% vs. 70%, 95% CI 0,26 to 0,99) in trans compared to cis people were found. The authors suggested that patients and healthcare practitioners being unaware of the relevant screening guidelines could be an explanation (106), and others have discussed the barriers towards accessing the healthcare system leading to later cancer diagnosis and worse survival for many cancer types (107).

A retrospective case series of 81 trans men (median age = 31) receiving testosterone therapy with a median duration of 4 years and undergoing hysterectomy found no cases of endometrial hyperplasia or malignancy (108). Current literature lacks consensus on the need for endometrial surveillance or risk-reducing surgery, but in this study there does not appear to be an excess of endometrial pathology (108). Jacoby et al. (109) conducted a retrospective study to determine the incidence of

precancerous and malignant lesions at the time of gender-affirming surgery. In biopsies of trans men who underwent bilateral mastectomies (n = 193), 8,8% contained a high-risk lesion and 1 patient harbored lobular carcinoma in situ (109). This patient was not on hormone therapy. Of the 8 trans men who underwent vaginectomy, 1 patient had a high-grade vulvar intraepithelial neoplasia associated with high-risk human papilloma virus (109).

In a retrospective Dutch study (110) of 2,260 trans women, with a median duration of 13 years of GAHT in different variations, incidence rates for breast cancer were lower compared to cis women (incidence ratio 0,3). However, during GAHT the risk increased towards cis female levels. From a retrospective Dutch study of breast biopsies available from medical files of 2,616 trans women, 6 lesions appeared to be breast cancer, with a benign versus malignant breast biopsy ratio of 88:12, comparable to the ratio in cis women (90:10). This finding led to the advice to follow breast care guideline as developed for cis women. Trans women hold a lower risk of prostate cancer compared to cis controls (standardized incidence ratio 0,2); 6 prostate cancers were diagnosed in 2,281 trans women with a median follow-up time of 14 years. Of the 94 trans women who underwent vaginoplasty with concomitant orchiectomy, 1 patient (1,1%) was found to have a mixed germ cell tumor (109). Another 40-year-old trans woman was found to have a well-differentiated papillary peritoneal mesothelioma (109). Recently, a possible association between the use of CPA and the occurrence of meningioma has come to our attention (111) (112). The risk of meningioma associated with the use of high-dose CPA was examined in a French cohort study of cis women (n = 253,777) and trans women (n = 10,876) (113). There were 2 groups of cis women; the control group received a cumulative CPA dose of < 3gr, and the exposed group received a cumulative CPA dose of > 3gr. The incidence of meningioma in the 2 groups was 4,5 and 23,8 per 100,000 person years respectively (RR 5,2; 95% CI 3,2 – 8,6). In the cohort of trans women participants, the incidence of meningioma was 20,7 (n = 3) per 100,000 person years in the exposed group versus 0 in the control group. These 3 participants were taking high daily doses of 100-150mg CPA per day for an exposure period of 3-4,5 years (113). Of note, an appreciable reduction in risk was observed after discontinuation of treatment. Even if hyperprolactinemia occurs frequently in feminizing treatment with an adjunct anti-androgen CPA (114) but not with spironolactone (115)(99), the occurrence of prolactinoma is rare. From the Amsterdam cohort described by Nota et al.(111) with 2,555 trans women (median age at start of GAHT = 31), we learned prolactinomas are diagnosed more often in trans women compared to control Dutch cis women (4,0; 2,1-7,9) and cis men (26,5; 12,9-48,6).

Supplementary table 5: Cancer risk

Discussion

In line with the increase in transgender people requesting gender-affirming care, more research initiatives have taken off. Where we previously had to rely for our clinical practice on guidance from single-center studies with small numbers and short follow-up durations, nowadays we have more data from larger cohorts and longer follow-up durations. We must acknowledge that most studies with a high number of participants are based on mostly Northern European and some US based cohorts, with many variations in hormonal protocols. Whether these data are applicable for other regions remains unclear. These European and US studies often happen within the context of well-organized gender teams, in which we know these are not globally accessible or even active. Nevertheless, the summary of currently available evidence is reassuring regarding the effectiveness and safety of GAHT. Integrating this knowledge, an overly cautious approach seems unnecessary (116)(66)(117), and many centers are currently extending the time between outpatient visits, also as a way to deal with many clients.

The ideal age to start GAHT in adolescence is heavily debated. Pubertal suppression may be started when Tanner stage 2 to 3 is reached. These early pubertal changes often lead to the adolescents experiencing increased distress. GnRHa is considered to be a safe intervention and has been shown to improve global psychological functioning (46)(47)(48)(49). More difficult to determine is an ideal age to start sex hormones to induce puberty. Currently, the recommended age to start is 16, but this is based on (local) jurisdiction and not on the individual cognitive capacity of the adolescent to give an informed decision. With puberty suppression, we can also extend the diagnostic phase, which gives the adolescent more time to reflect. Therefore, the age of 16 seems reasonable to start puberty induction, but the need for an individual assessment of the ability to make an informed decision must not be overlooked and may influence decisions.

GAHT reduces mental health problems in transgender people and helps obtain the desired physical features (37,40,59,60,41,46,51,52,55–58), but the role of the mental health professional in the gender team is clear from the relatively high prevalence of mental health problems. The main somatic concerns are bone health, cardiovascular events and cancer risk. In trans men, BMD was found to increase during GAHT; in trans women, it was less conclusive (71,72,74,76–78). The available literature on cardiovascular endpoints is still emerging and a focus of future research; however, in trans women there is a significantly increased risk of VTE (86–88). Blood pressure is not significantly changed by GAHT (88,91,92). In general, lipid profile is favorably changed in trans women in contrast to trans men. But in trans women treated with CPA, a negative effect is seen on HDL levels (91,93,94,98). Trans men have an increased risk of elevated hematocrit levels, but this is manageable (61,94,101,102). The discrepancy between the effects of GAHT on cardiovascular risk

factors and the available data on cardiovascular endpoints remains to be elucidated against the backdrop of changing hormonal interventions over time. There is no evidence of elevated cancer risk of breast, endometrium or prostate (108)(109)(110). But there is a small but higher risk of prolactinoma occurrence in trans women and a small increased risk of meningioma in trans women on CPA (111)(113). The general advice is to screen according to the organs present and relevant risk behavior, until further data become available (110).

As GAHT is relatively new and research is still limited, many research questions remain. Longer-term prospective follow-up studies are needed, and currently a >10-year analysis of data seems a logical next step as shorter-term data are reassuring. The research agenda should be determined in participation with the transgender and gender-diverse group, and a more integrated biopsychosocial approach accounting for influences of minority stress is advisable. Maybe it is also time for randomized controlled trials to examine treatment proposals e.g., estrogen with or without progestogens as feminizing treatment, its potential added value physically and psychologically, or effectiveness and negative effects of intramuscular or sublingual estrogens. From the presented research on breast development, it has become clear that the current hormonal strategies are often insufficient and better interventions are needed. Also, because of financial considerations, endocrine studies have included hormone-naïve participants only; many questions remain for those who present at a gender clinic and are hormone non-naïve. Most studies presented in this review are of binary trans persons or those who chose feminizing or masculinizing treatments. Very little is known about gender non-binary people's goals for hormonal interventions and what to advise. When confronted with terminology such as micro dosing—nowadays requested in outpatient clinics—we cannot rely on data, but have to be creative based on our knowledge of the effects and side effects of GAHT in binary trans people. Underlying mechanisms of already made observations also need to be elucidated. Finally, the body of literature on GAHT in adolescents is quite limited and is also restricted to a few centers working with young trans people. Likewise, the data on GAHT in elderly trans people remain nearly inexistent. A wealth of research questions is still out there, as so far we have only scratched the surface.

Limitations

For this review we relied on published data from a limited group of clinical research teams that are active in this field. Per definition, this is biased literature, with participants having access to well-organized research centers. In the papers, feminizing and masculinizing treatments are described, assuming participants are trans men or trans women. Categorization of gender identity can only rely

on self-identification, and we assume some of the people categorized as trans are in fact non-binary individuals seeking GAHT. Also, many variations in GAHT exist, making generalization difficult.

Conclusions

The body of available data on GAHT in trans people is steadily increasing, and short-to-midterm outcomes are quite reassuring in relation to effectiveness and safety. Integration of these findings in clinical care is timely. Still, there are many trans people who are confronted with mental health difficulties, and appropriate counseling should be easily available. Therefore, lowering barriers to health care that are still present is very meaningful.

Conflict of interest statement:

The authors report no conflicts of interest in this work.

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