# Cardiovascular Phenotype in Turner Syndrome—Integrating Cardiology, Genetics, and Endocrinology

Kristian H. Mortensen, Niels H. Andersen, and Claus H. Gravholt

Department of Endocrinology and Internal Medicine (K.H.M., C.H.G.), Aarhus University Hospital, 8000 Aarhus, Denmark; Department of Radiology (K.H.M.), Cambridge University Hospitals, Cambridge CB2 0QQ, United Kingdom; and Departments of Cardiology (N.H.A.) and Molecular Medicine (C.H.G.), Aarhus University Hospital, 8000 Aarhus, Denmark



Cardiovascular disease is emerging as a cardinal trait of Turner syndrome, being responsible for half of the 3-fold excess mortality. Turner syndrome has been proposed as an independent risk marker for cardiovascular disease that manifests as congenital heart disease, aortic dilation and dissection, valvular heart disease, hypertension, thromboembolism, myocardial infarction, and stroke. Risk stratification is unfortunately not straightforward because risk markers derived from the general population inadequately identify the subset of females with Turner syndrome who will suffer events. A high prevalence of endocrine disorders adds to the complexity, exacerbating cardiovascular prognosis. Mounting knowledge about the prevalence and interplay of cardiovascular and endocrine disease in Turner syndrome is paralleled by improved understanding of the genetics of the X-chromosome in both normal health and disease. At present in Turner syndrome, this is most advanced for the *SHOX* gene, which partly explains the growth deficit.

This review provides an up-to-date condensation of current state-of-the-art knowledge in Turner syndrome, the main focus being cardiovascular morbidity and mortality. The aim is to provide insight into pathogenesis of Turner syndrome with perspectives to advances in the understanding of genetics of the X-chromosome. The review also incorporates important endocrine features, in order to comprehensively explain the cardiovascular phenotype and to highlight how raised attention to endocrinology and genetics is important in the identification and modification of cardiovascular risk. **(*Endocrine Reviews* 33: 677–714, 2012)**



## I. Introduction

Turner syndrome (TS) occurs in 50 per 100,000 live-born females (1). The principally clinical diagnosis rests on the coexistence of partial or complete absence of an X-chromosome with classical traits that include reduced final height, estrogen deficiency, and infertility (2). The highly varied phenotype causes diagnostic delay as

ISSN Print 0163-769X ISSN Online 1945-7189
Printed in U.S.A.

doi: 10.1210/er.2011-1059 Received December 6, 2011. Accepted April 19, 2012.
First Published Online June 15, 2012

Abbreviations: PAR, Pseudoautosomal region; RR, relative risk; SMR, standardized mortality ratio; TS, Turner syndrome; Xq, X-chromosome.

well as nondiagnosis, with 30% of females with TS never obtaining correct diagnosis (1). Failure to diagnose TS is highly unfortunate because morbidity and mortality are a great deal higher than in the general population (1, 3, 4). Additionally, clinical assessment is poor even in diagnosed individuals (5), although multidisciplinary practice may improve this (6). Therefore, both appropriate identification of girls and women with TS and subsequent implementation of prophylactic measures to reduce morbidity and mortality leave much to be desired.

Risk assessment in TS is compromised by insufficient insight into the prevalence and causes of different syndrome-associated traits that may impact adversely on prognosis. This is especially the case for cardiovascular contributions to the excess all-cause mortality, where congenital and acquired heart diseases are felt to contribute to 8 and 41% of all-cause mortality, respectively (4). Cardiovascular risk in TS is predominantly stratified from evidence gathered on risk markers in the background population because insight into the cardiovascular phenotype in TS is limited, and previous guidelines have relied on expert consensus only (2, 7). This is unfortunate because complex patterns of not only cardiovascular but also endocrine diseases in TS render direct translation of evidence from other cohorts hazardous (8). The risk burden is so severe that TS is proposed as an independent risk factor for cardiovascular disease (9). More educated risk stratification and more appropriate clinical care can only be facilitated through a thorough delineation of the endocrine and cardiovascular phenotype in TS.

This review provides insight into the cardiovascular and endocrine phenotype in females with TS and presents an up-to-date condensation of current state-of-the-art knowledge of the cardiovascular phenotype, emphasizing the significance of not just congenital but also acquired pathologies. The aim is to provide an update on current insight into the pathogenesis of TS in relation to recent advances in the understanding of X-inactivation and its impact on female health. Moreover, the review provides an updated hypothesis on the genetic etiology of TS, highlighting our knowledge of the significance of X-chromosomal haploinsufficiency to congenital and acquired cardiovascular and endocrine traits. Finally, the review incorporates important endocrine features of TS, accounting for how genetics may explain the prevailing phenotype and that attention to endocrine factors is important in our efforts to identify and modify cardiovascular risk markers.

The full PubMed database was searched (without time restrictions) in December 2011 using the keyword "Turner syndrome" as MeSH term, as well as "Turner syndrome," "Turner's syndrome," "Turner," and "Turner's" in titles and abstracts. Articles relevant to the topic were obtained and reviewed, as well as older articles selected by the authors. Publications cited in this review were selected from those identified by the searches at the authors' discretion.

## II. Morbidity and Mortality

Increased morbidity and mortality are cardinal traits in TS (1, 3, 4, 10). The risk of premature death is increased 3-fold [standardized mortality ratio (SMR), 3.0] (1, 4), and life expectancy is reduced by at least a decade (1, 4, 11). The early-life disease burden is high, with an inverse relation between the age achieved by a girl or woman with TS and her reduction in life expectancy (11). The inferior outcomes are attributed to endocrine, nervous, cardiovascular, respiratory, digestive, and genitourinary organ disease (Fig. 1) (1, 3, 4). Of these, cardiovascular disease is increasingly recognized as the principal component in the causation of decreased life expectancy (1, 3, 4) with half of the excess morbidity attributed to cardiovascular pathology (4) (Figs. 1 and 2). In TS, cardiovascular morbidity and mortality are attributed to:

- Congenital heart disease (SMR, 20.7)
- Aortic dilation and dissection (SMR, 23.6)
- Ischemic heart disease (SMR, 2.8)
- Cerebrovascular disease (SMR, 3.9) (1, 3, 4, 11)

### A. Karyotype-phenotype correlations

All-cause mortality is raised in 45,X when compared with mosaic karyotypes (1, 4). An exacerbated prognosis is also to some extent seen in the presence of an isochromosome, and most likely also in karyotypes with Y-chromosome material. Yet, all females with TS face increased morbidity compared with the general population (3), although patterns of specific pathologies may differ between karyotypes. An increased risk of cardiac congenital anomalies has been demonstrated in 45,X (3, 12, 13), and karyotypes with an isochromosome suffer from an increased risk of diabetes and hypothyroidism (4, 14). Karyotypes other than 45,X associate with an increased risk of noncongenital cardiovascular disease (3).

The patterns of morbidity and mortality appear to be changing over time, with a trend toward a reduction in mortality (1, 4). Changes in the composition of diagnosed karyotypes are likely to contribute to this because a larger proportion of less severe phenotypes with mosaic karyotypes is now more often being correctly diagnosed (15). Especially mosaic karyotypes encompass a larger fraction (16), where cell lines of normal karyotype may associate with more favorable profiles (17). Improvement in management in the diagnosed females may also contribute to a seemingly improved prognosis. Irrespective, the prognostic improvement is small, and it represents only a trend

**Figure 1.**

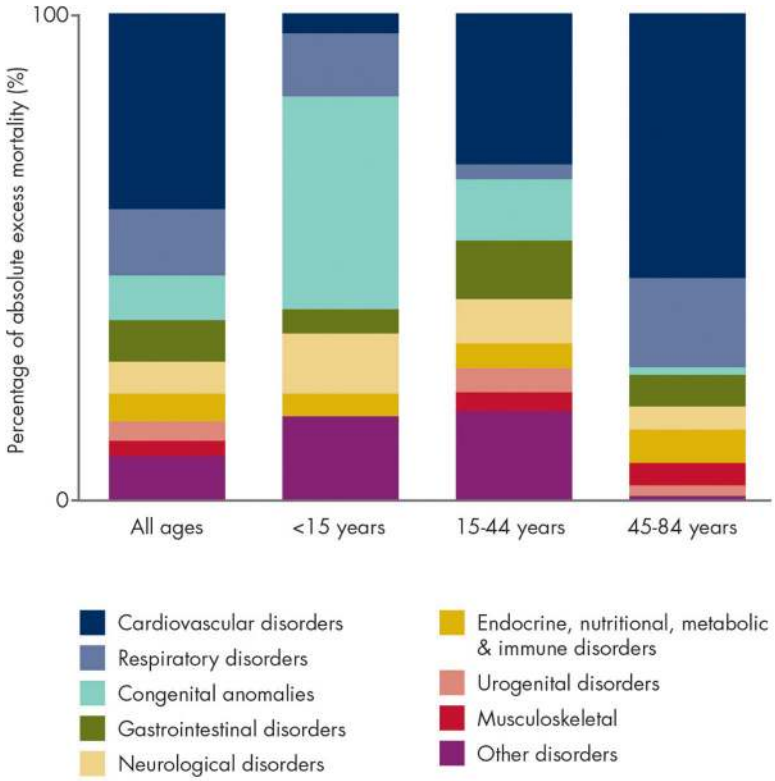


Figure 1. Differentiated mortality in TS for all ages and according to age groups (4). Categories were defined according to International Classification of Diseases 9. Numbers are adapted to express the percentage of total absolute excess risk caused by the group of disorders in question.

in a cohort with grossly increased risk of premature death (1). Important to risk prophylaxis, many females with TS are diagnosed after a delay of many years (1).

### B. Genetic background

The advent of TS is caused by *de novo* meiotic or mitotic nondisjunction that results in a spectrum of karyotypes (Table 1) (3). There can be complete absence of an X-chromosome, a structurally abnormal X-chromosome (isochromosomes or ring chromosomes), different levels of abnormal karyotypes within the body tissues (mosaicism), and karyotypes with Y-chromosome material (Table 1). Maternal risk factors for offspring with TS have been proposed to include advanced maternal age and low maternal height, although predictive values of these factors are weak for a fetus with TS (18). Also, no correlation with maternal age or even higher risk in younger mothers has also been reported (16, 19). The risk of repeated pregnancy with a fetal karyotype compatible with TS is generally not considered raised, although rare familial occurrences have been described, and a recent case series did suggest an increased risk (20, 21).

The correct number of sex chromosomes (X- and Y-chromosomes) and their correct gene expression are essential for normal health (22). In keeping with this, increased morbidity and mortality are key features of structural and numerical disorders of the sex chromosomes (1, 23, 24). Structural or numerical abnormalities of the Y-chromosome are compatible with life, whereas complete lack of X-chromosome material is incompatible with fetal survival. In fact, even the complete lack of one X-chromosome in a female may not be feasible either, and it has been hypothesized that 45,X females are indeed low-level mosaics (25, 26). Concordantly, *in utero* lethality has been proposed to be especially increased for karyotypes with 45,X monosomy, with as many as one in 100 of all recognized early pregnancies being 45,X and more than 99% of these being spontaneously aborted (27). This process of natural selection occurs predominantly in early pregnancy and leads to a prevalence of karyotypes compatible with TS of 50 per 100,000 live births (1).

### C. X-Inactivation in Turner syndrome

The 1098 known genes on the X-chromosome by far exceed the 78 genes on the male Y-chromosome (28). This large difference in gene content exists despite the sex chromosomes stemming from a common progenitor autosome (29). Full transcription of all genes on both X-chromosomes in female somatic cells, as opposed to transcription from the single X- and Y-chromosomes in somatic cells of males, is detrimental. This delivers excess transcriptional products from the two X-chromosomes in females, when compared with the somatic male counterpart (30). Therefore, somatic cells in females are subject to gene dosage compensation in early embryonic development to equalize effective dosage of X-linked genes (31). This process is termed X-inactivation, and it involves packaging of transcriptionally active chromatin into "silenced" heterochromatin (31). The X-inactivation center is essential to gene dosage compensation and is comprised of a cluster of genes situated near the centromere of the long arm of the X-chromosome (Xq). This center encodes genes that are prerequisite to X-inactivation, including more types of noncoding RNA and regulatory protein binding sites (32). Among these, the X-inactive specific transcript (*XIST*) gene is critical to X-inactivation (33) because it transcribes noncoding *XIST* RNA that coats the X-chromosome and through methylation and histone modifications represses euchromatin to

**Figure 2.**

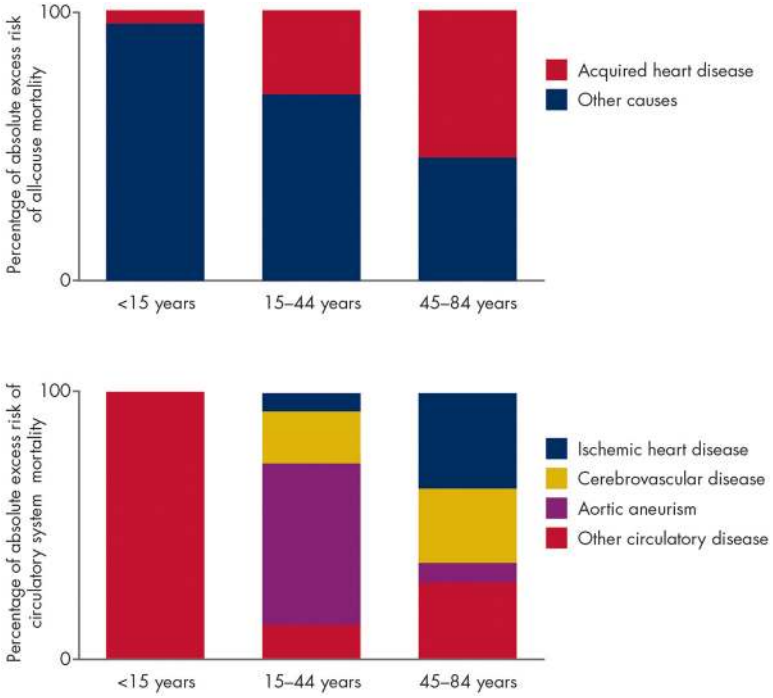


Figure 2. Acquired heart disease in TS. Acquired heart disease accounts for a large proportion of the 3-fold excess mortality, increasing in relative contribution with age in TS (*top panel*). The contributions from ischemic heart disease, cerebrovascular disease, aortic aneurysm, and other heart diseases are shown *below* (4).

form the inactive X-chromosome (34). Initially, both X-chromosomes transcribe *XIST* RNA and an antagonistic noncoding *TSIX* RNA from the X-inactivation center (35). By general random selection of the maternal or paternal X-chromosome, the balance of noncoding RNA transcribed from the X-inactivation center shifts toward *XIST* in the X-chromosome that is subsequently inactivated. The other X-chromosome maintains *TSIX* transcription during the X-inactivation maintenance phase and avoids silencing (36). The random X-inactivation process introduces in the normal female an obligatory mosaicism for the paternal and maternal X-chromosome across somatic cell lineages (37).

The inactivated X-chromosome stays inactive in the specific cell clone (37). This equalizes the effective gene dosage to that of males, with only female germ cells having the potential to reactivate an inactivated X-chromosome (37). However, approximately 25% of genes on the inactivated X-chromosome escape transcriptional silencing to varying extents, depending on the tissue, individual characteristics, and age (38–40). Escapees include two pseudoautosomal regions (PAR1 and PAR2) located at the termini of Xq and the short arm of the Xp, which are common with the Y-chromosome (Fig. 3). Several genes outside these two regions also escape silencing, whereof 25 genes are X/Y homologous and others again have lost their Y-chromosomal counterpart (paralog genes) (28). The relative expression of nonsilenced genes on the inactive X-chromosome relative to the genes of the active X-chromosome has not been clarified, but expression of escape genes from the inactivated X-chromosome may be reduced compared with the active (38, 41).

In TS, complete or partial loss of activity of the entire or even parts of the X-chromosome detrimentally impacts both prenatal and postnatal survival (1, 27). The mechanisms of gene dosage compensation are complex even in normal health, and much still needs to be resolved. In TS, the prevailing traits may result from decreased expression of paralog genes or altered expression levels of homolog genes from within or outside PAR (or a combination of these). Additional variability between females with TS (and even between cell clones in the same females due to mosaicism) may be introduced through different patterns of X-inactivation. In 45,X the one normal X will inadvertently

**TABLE 1.** Postnatal and prenatal prevalence of TS

| | | Prenatal, % (n) | Postnatal, % (n) |
|---|---|---|---|
| n | | 383 | 1063 |
| Monosomy | 45,X | 66% (254) | 40% (426) |
| Mosaic | 45,X/46,XX | 23% (90) | 24% (257) |
| Isochromosomes | 45,X/46,X,i(Xq); 46,X,i(Xq); 45,X/46,X,i(Xq)/47,X,i(Xq),i(Xq), *etc.* | 3% (11) | 10% (107) |
| Deletions | 45,X/46,X,del(X); 46,X,del(X) | 3% (13) | 6% (69) |
| Polyploidy | 45,X/46,XX/47,XXX; 45,X/47,XXX; 45,X/46,XX/47,XXX/48,XXXX | 3% (13) | 11% (117) |
| Ring chromosomes | 45,X/46,X,r(X) | <1% (2) | 3% (34) |
| Y material | 45,X/46,XY, other with Y material | | 5% (51) |

National prevalence of TS updated in 2011 from the Danish Cytogenetic Central Register (previously unpublished data), where 72–82% chose termination of the pregnancy with a trend to fewer terminations in the last half of the study period. Note is made of a decrease in the proportion of 45,X postnatal karyotypes from the previously published data in Denmark: 47% (1996) (16) to 40% now. Prenatal prevalence was gathered from a screening program of recognized pregnancies using amniocentesis and chorionic villus sampling.

**Figure 3.**

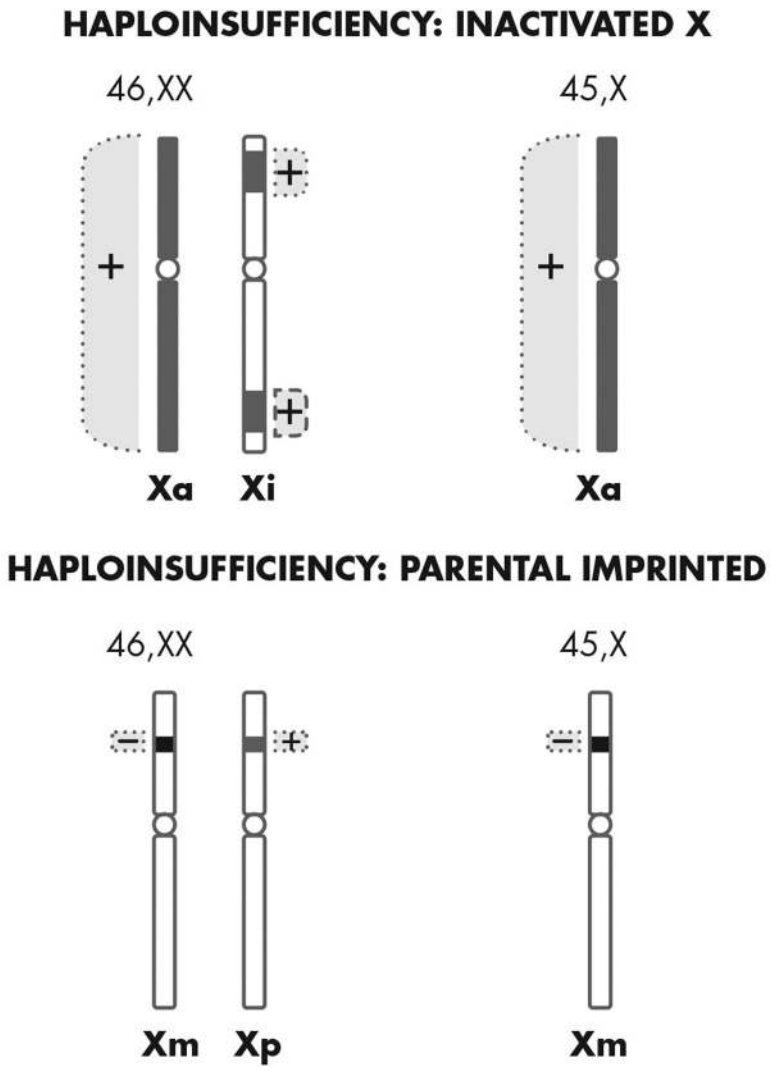


Figure 3. Models of genetic etiology in TS. Haploinsufficiency for the X-chromosome in TS may result in an abnormal phenotype through different mechanisms. In the X-inactivation hypothesis (31), haploinsufficiency for genes that normally escape X-inactivation on the otherwise inactivated X-chromosome (Xi) result in abnormal gene dosage. These escapees are mainly located in PAR1 and -2 on the long and short arms of the X-chromosomes. Thus, only genes from the activated X-chromosome (Xa) are expressed in the haploinsufficient female with TS. In the imprinting hypothesis (59), genes on the X-chromosome are expressed in a monoallelic fashion depending on paternal or maternal origin. In TS, lack of activated allele will lead to loss of expression for that gene. This is shown here for the loss of the normally expressed paternal allele (Xp) with the nonexpressed maternal allele (Xm) left in TS.

be active, whereas in structural abnormalities such as isochromosomes or ring chromosomes, inactivation of the normal X-chromosome may further aggravate the phenotype (37). The evolutionary younger Xp is subject to more escapees from inactivation than Xq (29), and structural abnormalities involving the Xp may thus carry a more adverse impact, as has been proposed for aortic dilation in TS (42).

It has become evident that a sexual dimorphism in autosomal gene regulation is present, and that more than half of all genes in a tissue are differentially expressed, although the difference in expression in the majority of these genes is only in the order of a 1.2-fold difference (30). Historically, differences between the sexes were mainly attributed to differences in the level of sex hormones, but genetic factors seem also to be involved (43). New research shows that sexual dimorphism is determined directly by the sex chromosome complement, rather than the specific sex, in addition to a modulatory role of the male-determining *Sry* gene at least in mice (44). In this study, the authors also examined differences between male $XX^{Y-}$ mice (male mice where the *Sry* gene is translocated to an autosome, thus with the same sex complement as a female) and XO mice (mice with "TS"), and found distinct differences in gene expression (44). It is not clear how X-chromosome complement causes this sexual dimorphism in gene expression, but candidate genes are genes that escape silencing (40), such as *UTX* (ubiquitously transcribed tetratricopeptide repeat gene on X-chromosome) and *JARID1C* (a histone methylase), which have previously been anticipated to play a role in the pathogenesis of TS (45, 46). Thus, if confirmed in humans, such a general mechanism of influence of sex chromosome complement on autosomal gene expression could explain features present in TS.

### D. Candidate genes

Attempts to map candidate genes that escape gene dosage compensation in TS and other disorders of the sex chromosomes are ongoing. The short stature homeobox-containing gene on chromosome X (*SHOX*) situated in PAR1 on Xp has been implicated in growth retardation and bone changes (47) and is a homeodomain transcription factor. Brain natriuretic peptide and fibroblast growth factor receptor 3 are transcriptional targets of *SHOX* (48), and thus pose a link that may be of relevance to the etiology of cardiovascular disease in TS (49). Importantly, penetrance of gene defects in TS differs from X-linked recessive disorders. This is evident in a comparison with *SHOX* haploinsufficiency in Leri-Weill dyschondrosteosis, where a *SHOX* gene mutation (a range of single gene deletions) causes only 50–75% of the height retardation encountered in TS, and congenital heart disease is not a feature of Leri-Weill dyschon-

drosteosis (50). Putatively, the missing or structurally abnormal X-chromosome delivers a more profound impact on gene expression in TS than in monogenetic disorders through mechanisms that are unaccounted for (51). Hence, a range of features is known to associate with *SHOX* haploinsufficiency in TS:

- Reduced final height
- Sensorineural hearing impairment
- High-arched palate
- Short fourth metacarpal
- Madelung deformity of the radius
- Skeletal disproportionality (51)

Altered transcriptional expression of pseudoautosomal genes in PAR1 and PAR2 other than the *SHOX* gene is also likely to carry an impact in TS, with causative loci for various cardiovascular, endocrine, skeletal, or psychological traits awaiting discovery. Most PAR genes that escape X-inactivation are to be found on Xp, where 24 genes have been identified compared with only five on Xq (28). Thus, structural abnormalities involving unbalanced translocations or deletions of Xp are expected to more severely affect the phenotype than those involving Xq (37, 52). The consequences of haploinsufficiency for X/Y gene pairs beyond PAR1/PAR2 or the absence of X heterologous genes in TS remains undefined (30). Recently, work in pluripotent embryonic stem cells from amniocytes of a TS fetus showed that the pseudoautosomal genes *ASMTL* and *PPP2R3B* were expressed in lower levels than normal cells (53). *ASMTL* may be related to methyltransferase activity (and could thus be involved in proper methylation of the genome), whereas *PPP2R3B* is involved in cell cycle control (53). Insufficient up-regulation of another pseudoautosomal gene, *CSF2RA*, during embryoid body formation was also demonstrated, consistent with data showing lower expression of this gene in embryonic stem cells with TS (54). Decreased expression of *CSF2RA* points to insufficient placentation as an explanation for the increased early fetal lethality in TS. Interestingly, the TS-induced pluripotent embryonic stem cells differentiated normally into neural-like, cardiomyocyte-like, and hepatocyte-like cells, although the cardiomyocyte-like cells displayed a spontaneous action potential that was slightly longer compared with controls, but this was not statistically significant (53). These findings may help to explain the prolonged QT interval seen in TS (55).

The potential genetic mechanisms are complex, and disease may very well be the result of deregulation within different pathways that work in concert to cause abnormality. The phenotype of TS may result from loss of proteins involved in ordinary cellular function, which are normally expressed from the inactive X-chromosome. Alternatively, loss of coding genes on an absent, inactive X-chromosome may alter gene expression from the active X-chromosomes. Loss of biallelic gene expression by either of the aforementioned mechanisms may directly impact the TS phenotype through altered expression of genes that regulate tissue genesis and maintenance, or more indirectly affect tissue phenotypes through loss of transcriptional up- or down-regulation of autosomal or even other X-linked genes. Several genes and critical regions have been proposed in TS, but the exact delineation of their roles in TS remains to be determined (54, 56–58). The same applies to the discovery of other tentative loci involved in the causation of the many cardiovascular, endocrine, skeletal, or psychological traits.

### E. Parental imprinting of genes on the X-chromosome

Although haploinsufficiency for X-chromosomal genes that escape silencing may cause the abnormal phenotype of TS, an alternative hypothesis suggests that parental imprinting of gene expression profiles from the X-chromosome may be involved in the causation of TS. This hypothesis is based on the fact that whereas the majority of genes are expressed from either the paternal or maternal allele, some autosomal genes are expressed in a parent of origin-dependent fashion (59). Loss of the active (imprinted) allele has been implicated in diseases such as Prader-Willi syndrome (60). In TS, this process has been associated with the X-chromosome, and some patterns of morbidity may associate with loss of either the maternal or paternal X-chromosome (Fig. 3). In the imprinting hypothesis, loss of a homolog gene on the X-chromosome will lead to null expression of this gene when it is the active (imprinted) allele. The paternal X-chromosome is absent in approximately 60–80% of 45,X monosomy, with a more equal paternal *vs.* maternal X-chromosomal deficiency in mosaic and isochromosomal karyotypes (22, 61–63). The impact of parental X-chromosome imprinting in TS is not resolved. A determinant impact has been reported on aortic valve morphology and coarctation of the aorta (63), lipid levels, visceral and abdominal fat (62), white and gray matter cerebral volume (64), and neuropsychological profile (65). Conversely, other studies have not been able to reproduce these findings (22, 66). Thus, the genetics of TS is probably more complex than previously thought and could involve processes like:

- The X-inactivation process and X-chromosome dosage compensation
- Candidate genes other than *SHOX* are probably relevant in TS
- The role of haploinsufficiency of escape genes is not yet clear

**Figure 4.**

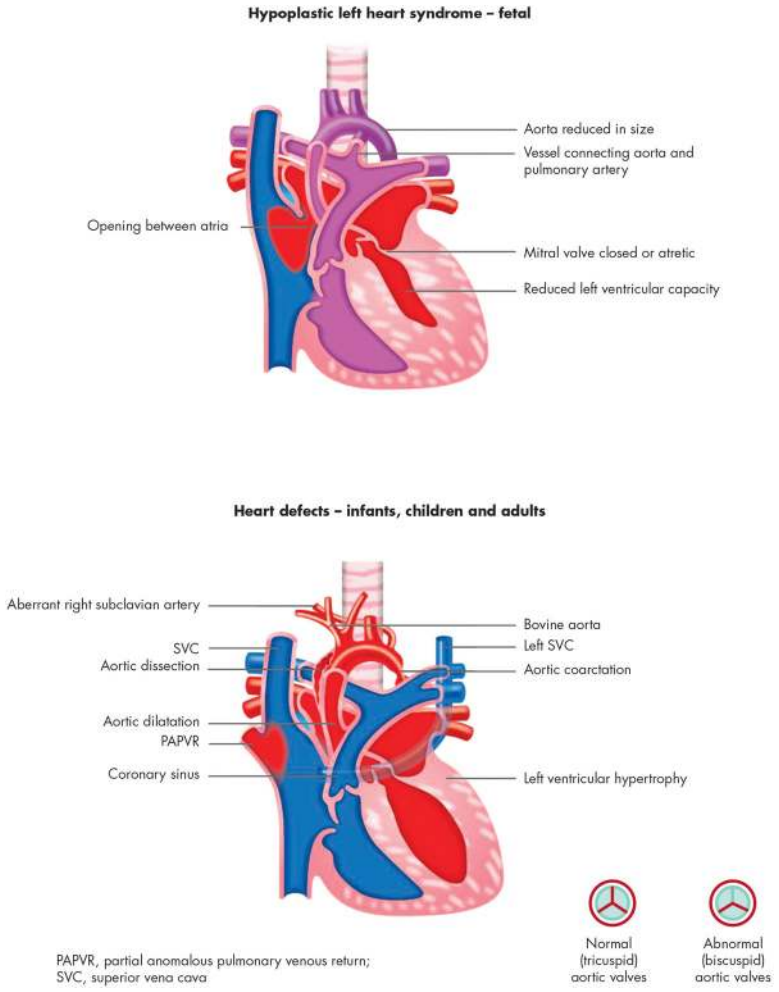


Figure 4. The *upper figure* illustrates a fetal heart with hypoplastic left heart syndrome, a condition often seen during fetal life in TS and probably contributing to the high spontaneous abortion rate. The *lower figure* illustrates the most common congenital cardiovascular malformation seen in TS. The many different malformations and acquired conditions are not necessarily present at the same time in any given individual (see text for further information). PAPVR, Partial anomalous pulmonary venous return; SVC, superior vena cava.

- Genotype-phenotype relations are not yet fully uncovered
- Parental imprinting
- Differential methylation patterns

## III. Congenital Heart Disease

The risk of congenital heart disease is considerable [relative risk (RR), 8–50] (67–69). The disease spectrum spans from trivial defects to severe and highly complex disorders, and accordingly some lesions may be asymptomatic (67, 70), whereas others are incompatible with normal life expectancy or even fetal survival (4, 71). An estimated 22–70% of all females with TS have a form of congenital heart disease (12, 67–69, 72) with congenital anomalies coexisting in other organs in 22% (12). Reports on the prevalence of congenital heart disease are highly dependent on the type of investigation used, spectrum of assessed anomalies, and karyotype distribution within the studied cohort. Excess early life mortality due to severe congenital heart disease may also cause selection bias into studies performed in adolescence and adulthood through survival with advancing age of less severe cardiac phenotypes (11).

### A. Bicuspid aortic valve

A bicuspid aortic valve is found in 15–30% of TS patients (52, 67–69, 72), compared with 1–2% of the general population (Fig. 4) (73). The prevalence would appear to be increasing due to higher diagnostic sensitivity achieved with cardiac magnetic resonance when it comes to distinguishing the tricuspid from bicuspid aortic valves (52). Congenital aortic valve anomalies of an immediately severe nature may rarely present in the infant with TS, such as aortic valve dysplasia and congenital aortic stenosis (69, 74).

Outcomes with a bicuspid aortic valve are generally favorable in childhood, with the likelihood of associated morbidity increasing with age (73). In general, for the bicuspid valve, accelerated valve calcification often promotes premature valvular stenosis. Additionally, poor leaflet coaptation can lead to aortic regurgitation, which may become aggravated in a vicious circle of increased stroke volume and aortic root dilation (73). In view of this general population evidence, it is not surprising that aortic valve dysfunction is frequent in adults with TS; aortic valve stenosis is seen in 4–16% and regurgitation in 6–45% (52, 67, 72). For comparison, trivial and clinically irrelevant aortic regurgitation and stenosis may be seen in 11 and 6% of healthy females with tricuspid aortic valves, respectively (67). As expected, the risk of valvular dysfunction in TS links closely with the presence of bicuspid aortic valves (RR, 7.9 for stenosis; and RR, 4.2 for regurgitation), and the bicuspid valve morphology is the principal cause of all moderate and severe dysfunctions (52, 67). Pediatric studies of aortic valve function in TS are limited, but stenosis is observed in 28–46% and regurgitation in 42–50% of adult females with TS and a bicuspid aortic valve (67, 75). Unfortunately, functional grading has only been the focus of a few

**Figure 5.**

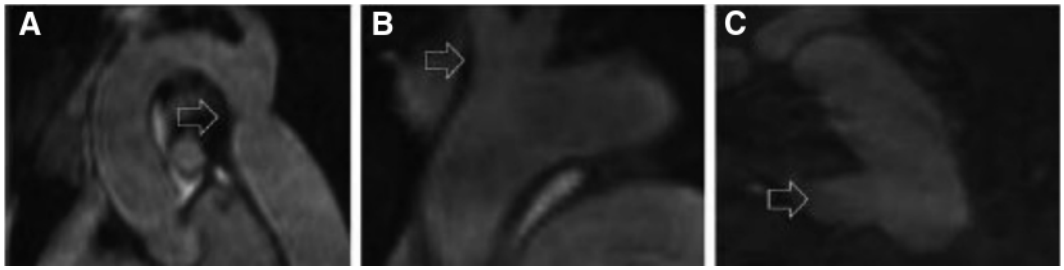


Figure 5. Congenital abnormalities of the thoracic aorta in TS. Aortic coarctation (A), common origin of the innominate and left common carotid arteries (B), and an aberrant right subclavian artery (C) as seen by cardiac magnetic resonance in TS (67). [Reproduced from K. H. Mortensen *et al.*: Abnormalities of the major intrathoracic arteries in Turner syndrome as revealed by magnetic resonance imaging. *Cardiol Young* 20:191–200, 2010 (67) with permission. © Cambridge University Press.]

studies (52, 67). Hence, insight into the general state of the aortic valve in TS is scarce, which also includes our understanding of the onset of clinically significant disease, its optimal treatment, and to a large extent also the relationship between aortic valve function and aortic dilation.

Raphe configuration of the bicuspid aortic valve may be of prognostic importance for the development of aortic valve dysfunction and ascending aortic dilation (76). The right and left aortic leaflets are mainly fused in TS (82–95%) (52, 67), which, compared with leaflet fusions that involve the noncoronary cusp, has been indicated as a positive prognostic omen in some non-TS cohorts (73). Hitherto in TS, raphe configuration has nevertheless not been demonstrated to determine aortic valve function or aortic morphology (52, 67). Important to the clinician, the annual incidence of endocarditis in the general population is increased to 0.3–2% with bicuspid aortic valves (73). Therefore, due to the frequent occurrence of not just bicuspid aortic valves but also congenital heart disease in TS, the risk of endocarditis must also be assumed to be increased in these females, although the evidence hereof is limited (77).

#### B. Coarctation of the aorta

Coarctation of the aorta is found in up to 17% of females with TS (13, 52, 67, 78), compared with 0.04% in the general population (Fig. 4) (79). The aortic obstruction often coexists with a bicuspid aortic valve (RR, 4.6) (52, 67), and coarctation of the aorta in TS is increasingly regarded as a part of an abnormal aortic phenotype (Figs. 5 and 6). This phenotype includes features such as an elongated and kinked transverse aortic arch (~50%), an aberrant right subclavian artery (~8%), a bovine aorta (~8%) (Fig. 4), and isolated cases of cervical aortic arch (13, 67, 80).

The severity of coarctation of the aorta in TS has received little attention. The prevalence of repaired coarctation of the aorta amounts to 5–12% in childhood and adulthood (13, 78, 81). In addition to this, undiagnosed coarctation of the aorta has been demonstrated by cardiac magnetic resonance in 5–8% (13, 67, 75). In reports of undiagnosed cases, the diagnosis is most often made by morphological analysis as a concentric narrowing with a posterior shelf-like morphology located typically in the descending aorta. These occult lesions have in all but one severe case been mild when assessed by cardiac magnetic resonance (neither arterial collaterals nor flow acceleration to warrant surgical intervention has been present) with very limited data from echocardiography or cardiac catheterization studies (13, 75, 80). Coarctation of the aorta may be seen in conjunction with an elongated aortic arch (13) or more adverse aortic arch phenotypes that include aortic arch hypoplasia and interrupted aortic arch (67, 80, 82).

Little is known about the optimal corrective technique of coarctation of the aorta in TS. Some surgical centers have hypothesized that aortic wall frailty increases the risk of perioperative hemorrhage and rupture of suture lines after end-to-end resections (observed in 25–38%) (83, 84). Favorable outcomes have been reported with patch angioplasty repair (83, 85). Percutaneous correction was encouraging in a very small series (86), although aortic dissection and pseudoaneurysm formation have been reported after balloon or stent repair (87–89). Most likely, the optimal surgical technique should be decided on a patient-to-patient basis and follow general experiences. Importantly, the risk of aortic dissection associated with the coarctation is not completely relieved with repair because dissections may occur even years after repair (90), which is in keeping with experiences in the correction coarctation of the aorta outside TS (91).

#### C. Associated lesions

Several mild congenital heart defects can be found in TS (Fig. 3). These include:

**Figure 6.**

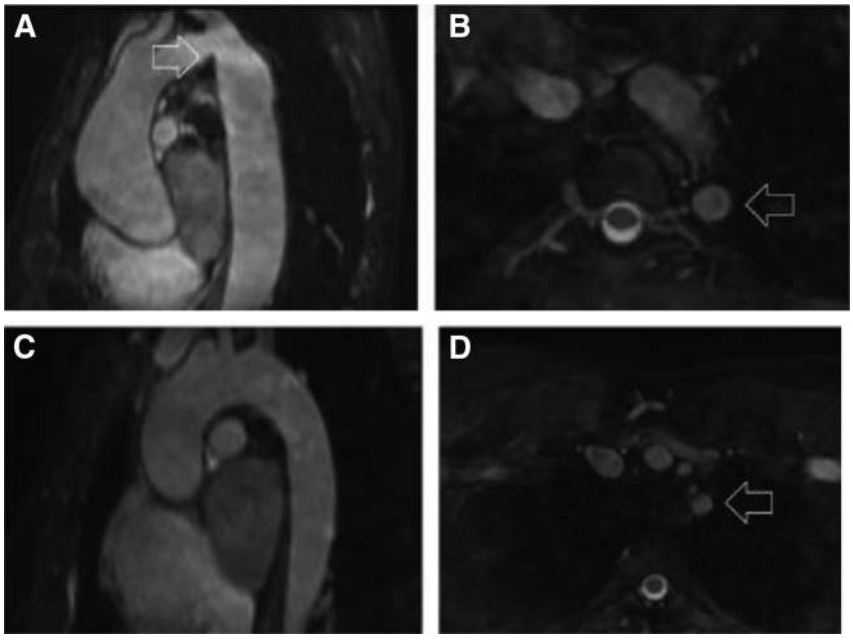


Figure 6. The transverse aortic arch phenotype in TS. The normal and elongated transverse aortic arch as seen by cardiac magnetic resonance imaging in TS (67). The abnormality is defined by the coexistence of kinking of the inferior aortic contour at the aortic isthmus (A) and a left subclavian artery origin from a position posterior to the trachea in the horizontal plane (B). The normal transverse aortic arch (C) and normal origin of the left subclavian artery (D) are illustrated for comparison; kinking is absent, and the left subclavian artery departs anterior to the posterior aspect of the trachea. [Reproduced from K. H. Mortensen *et al*.: Abnormalities of the major intrathoracic arteries in Turner syndrome as revealed by magnetic resonance imaging. *Cardiol Young* 20:191–200, 2010 (67) with permission. © Cambridge University Press.]

- Atrial septal defects (in 1–2%; RR, 19) (12, 69, 75)
- Ventricular septal defects (in 1–4%; RR, 49) (12, 69, 75)
- Partial anomalous pulmonary venous drainage (in 13–15%; RR, 20) (13, 92) (Fig. 4)
- Persistent left superior vena cava (in 8–13%) (13, 92) (Fig. 4)
- Persistent arterial duct (72)
- Interrupted inferior vena cava with azygous continuation (80)
- Pulmonary valve stenosis (72)

Prenatally, severe left-sided cardiac malformations pave the way for 45,X conspectuses comprising up to 10% of all recognized early miscarriages (27, 71, 93–95). This inferior outcome adds to a reported general inability to form an embryo proper with TS karyotypes (27, 53, 54). Spontaneous abortion occurs in 99% of all fetuses with 45,X TS, with the prevalence of TS dropping drastically from 1000 per 100,000 of all conceptions to 50 per 100,000 live births (1, 27). Fetal echocardiography performed in TS in gestational wk 11–15 has shown coarctation of the aorta (45%), hypoplastic left heart syndrome (Fig. 4) (13%), and atrioventricular septal defect (one fetus) (71). These adverse findings were confirmed by an autopsy study, which additionally revealed the presence of unicuspid aortic valves (15%) or bicuspid aortic valves (85%), whereof all but one were 45,X karyotypes (94). Moreover, midgestation fetuses have myocardial hypoplasia (96), but this must be read in a context of the general state of small for gestational age in TS (18).

At birth, a range of complex congenital heart diseases can be found. Apparently, these severe lesions associate with an even more inferior prognosis than when occurring in the general population (97, 98). They include:

- Hypoplastic left heart syndrome (97) (Fig. 4)
- Congenital aortic valve stenosis (99)
- Neonatal coarctation of the aorta (12)
- Atrioventricular septal defects

Emphasizing the complexity of congenital heart disease in TS, lesions can be seen in both monosomy 45,X (12, 52, 67) and mosaic karyotypes (100). Furthermore, characteristic exterior morphological traits may or may not be present to direct the attention toward congenital heart defects, and there is not necessarily complete concordance between external phenotype and congenital heart disease (12, 101).

#### D. Etiology of congenital heart disease

Early prenatal screening for TS relies on fetal sonography performed at gestational wk 11–14 (102). Nuchal translucency may often present in karyotypes compatible with TS, and this fetal appearance generally predicts chromosomal abnormalities and poor fetal outcome (including congenital malformations of the heart and skeleton) (103, 104). The remnant of this lymph accumulation in the lower neck region is appreciated postnatally as neck webbing in TS. Because this external feature cosegregates with congenital heart disease in TS (12, 52, 101), a causal relation between lymph accumulation and congenital heart disease has been proposed (105). Perturbations of flow and pressure are hypothesized to lead to left-sided defects by a mechanistic chain of events (106) that starts with abnormal fetal lymphangiogenesis (107, 108). The pres-

ence of severe edema in aborted TS fetuses with anomalies of the left ventricle and the aorta lends promise to this hypothesis (109). Yet, separate mechanisms have not been ruled out because postnatal neck webbing and congenital heart disease do not always coexist (12, 52). Furthermore, changes in the lymphatic vessels around the major vessels and the heart have not been demonstrated to be uniformly present in aborted fetuses with heart disease in TS (although most were examined after primary cardiac organogenesis) (109).

Insight into the genetic regulatory mechanisms behind formation and maturation of the thoracic aorta, the brachiocephalic vessels, the arterial duct, and the proximal pulmonary arteries from the five pharyngeal arch arteries may promote our understanding of congenital heart disease in TS. TGF-$\beta$2 could be of key importance in TS because knockout mice deficient of TGF-$\beta$2 suffer from obstructive lesions in the fourth pharyngeal arch artery (110) in a similar pattern to TS (94, 111). Also potentially perturbed in the fetus with TS, reduced TGF-$\beta$1 release from the endothelium in the context of low blood flow and pressure may adversely influence normal angiogenesis, which normally involves hormonal stimulation of migration and differentiation of cardiac neural crest cells into blood vessels to form vascular smooth muscle cells (112). Recent data indicate that TGF-$\beta$1 and its receptors (types I and II) activate several pathways, including SMAD-dependent and SMAD-independent pathways (113). SMADs are a family of transcriptional factors that transmit signals from receptors for TGF-$\beta$1 to the nucleus. These transmitters regulate multiple targets such as matrix proteins, collagen, fibronectin and matrix metalloproteinases, and members of the fibrinolytic system that includes plasminogen activator inhibitor-1 (110). In both syndromic (Marfan syndrome and other) and nonsyndromic (bicuspid aortic valves, degenerative aortic disease, and other) aneurysms of the aorta, SMAD2 activation and increased TGF-$\beta$1 activity have been documented in the arterial media layer, which indicates a final common pathway for thoracic aorta aneurysm syndromes (114). Molecules with potential roles in aortic wall weakening are many. A proteoglycan like biglycan was decreased in aneurysmatic aortic tissue (114), and the same biglycan was perturbed in fetuses with TS (115). Vascular endothelial growth factor has been implicated as important to early angiogenesis as well as in postnatal maintenance of aortic wall integrity in TS (57). In contrast to these potential contributors, the angiotensin type 2 receptor appears to have been ruled out in the etiology of congenital heart disease in TS (116).

A close link has been hypothesized between arteriogenesis involving neural crest cell invasion and differentiation to vascular smooth muscle cells and peripheral neurons (117). This association infers that the lifelong perturbed autonomic nervous system in TS (49, 118) could share a common origin with congenital heart disease. Moreover, normal lymph vessel development is guided by vascular endothelial growth factor C produced by blood vessels (119), and with blood vessels and peripheral nerves evolving alongside (117), a common origin of the abnormalities of these three components could explain the phenotype seen in TS with disturbances of the neural, the arterial, and the lymphatic systems. Collectively, further studies of not only angiogenesis but also genetic models and signaling molecules in TS are essential in order to identify the causative mechanisms. Unfortunately, relevant animal models of TS are not available due to different patterns of X-inactivation as well as difficulties in creating the relevant karyotypes for TS in animals (120).

## IV. Acquired Heart Disease

Acquired heart disease is a major cause of morbidity and mortality in TS (Fig. 2). An increased risk of acute aortic dissection is present from as early as the second decade of life, stroke incidence is increased from the third decade, and myocardial infarction usually appears from the fifth decade (1, 3, 4). Although acquired heart disease with age contributes increasingly to all-cause excess mortality (Fig. 2), it may often be impossible to distinguish congenital from acquired lesions. Many adverse features such as aortic dilation (67, 78), proarrhythmic traits (55, 121), and hypertension (122) appear to be interrelated, and to some extent present throughout the entire lifetime in TS.

### A. Aortic dissection

Aortic dissection occurs in 1 or 2 of 100 females with TS (123). Incidence rates are increased up to 100-fold when compared with the female background population (Fig. 4) (42, 123). This often devastating event occurs 40 yr before expected and with peak incidences in the third to fifth decades of life (Fig. 7) (123). Aortic dissection is not restricted to adulthood in TS. Incidence rates are increased even in the below 19 yr age group with 14 per 100,000 yr (123), and aortic dissection may occur as early as the first decade of life (90).

The outcome of aortic dissection is unknown in TS. The prognosis is likely to be at least equally as grave as in the general population, where 22 in 100 cases die before reaching a hospital (124). In general, prognosis depends on the location and extent of the dissection and is improved when limited to the descending aorta (Stanford B/Debakey III) compared with both isolated ascending

**Figure 7.**

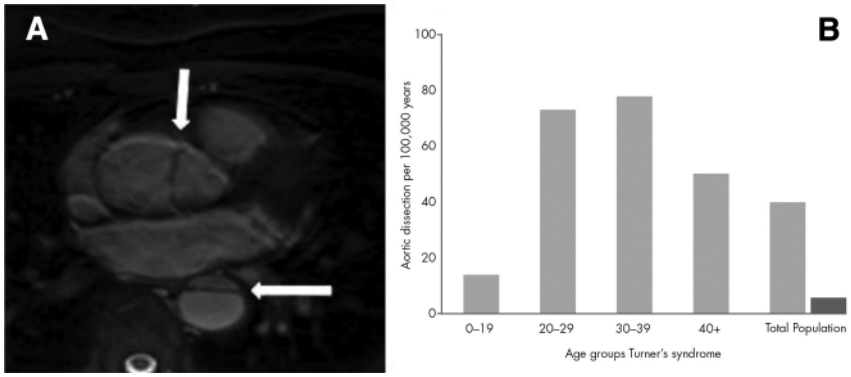


Figure 7. Aortic dissection in TS. A, Chronic Stanford type A dissection in a 50-yr-old woman with TS with 45,X/46,Xr(X) mosaic karyotype and no known risk factors for aortic dissections but moderate aortic regurgitation (81, 123). B, Aortic dissections occur at increased incidence throughout the entire life span of individuals with TS (123). [*Panel B* was reproduced from C. H. Gravholt *et al*.: Clinical and epidemiological description of aortic dissection in Turner's syndrome. *Cardiol Young* 16:430–436, 2006 (123) with permission. © Cambridge University Press.]

aortic involvement (Stanford A/Debakey II) and those propagating from the descending aorta into the ascending aorta (Stanford A/Debakey I) (125). In TS, aortic dissections predominantly include the ascending aorta (63%), with isolated descending aortic involvement occurring less frequently (37%) (123). The causative lesions are not well characterized in TS, be it intimal tear, intramural hematoma, or penetrating ulcer (126). Preponderance to Stanford type A dissection over type B is in accordance with a high prevalence of both ascending aortic dilation and bicuspid aortic valves (42, 81). Descending aortic dissection is most likely to occur in the presence of coarctation of the aorta (also after repair) (88, 90, 123).

Risk factors for aortic dissection in TS are not easily defined. Although the risk of aortic dissection and rupture is 100-fold increased (42), the event occurs on the backdrop of a syndrome prevalence of 1 in 2000 live-born girls (1), considerable diagnostic delay, a high extent of nondiagnosis (1), and aortic dissection affecting 1 or 2 of 100 females with TS over the course of an entire lifetime (123). Hence, the risk of aortic dissection may very well be severely increased, yet aortic dissections are infrequent encounters in clinical practice. Therefore, risk stratification in TS is mainly based on numerous case reports and one register-based survey (123, 127). One prospective study has assessed aortic dissection in TS (42). However, the total number of years at risk was relatively low, and it has remained difficult to conclude anything beyond a severely increased risk of dissection (42). In TS, the currently acknowledged risk markers for aortic dissection are hypertension, karyotype 45,X, aortic dilation, and left-sided obstructive lesions including bicuspid aortic valves, coarctation of the aorta, and other obstructive arch

**TABLE 2.** Risk markers for aortic dissection in TS

| General population risk factors | Evidence level | Special concerns in TS | Unresolved | Current risk factors in TS |
|---|---|---|---|---|
| Age (124) | Case reports, case series (127), one register study (123), one cohort study (42), and more cross-sectional studies of associations between aortic diameter and phenotype traits (78, 81) | Karyotype (127) | ERT? | Hypertension |
| Hypertension (125) | Expert consensus (Level C) (130) | Pregnancy (148) | Diabetes? | |
| Aortic diameter (135) | | | GH? | Aortic caliber |
| Aortic growth (130) | | | | Aortic growth |
| Bicuspid aortic valve (73) | | | | Bicuspid aortic valve |
| Aortic coarctation (347) | | | | Aortic coarctation |
| Aortic valve dysfunction (140) | | | | Aortic valve dysfunction |
| Iatrogenic trauma (348) | | | | Iatrogenic trauma |
| Aortitis (126) | | | | Age |
| | | | | Pregnancy |
| | | | | Karyotype |
| External trauma (349) | | | | External trauma |
| Drugs (126) | | | | Drugs |
| Pregnancy (126) | | | | |
| Atherosclerosis (126) | | | | Aortitis |

Risk markers for future aortic dissection in TS are currently based on evidence gathered in the general population and extrapolated onto TS as well as on limited evidence specific to TS. Hence, any conclusions on risk markers in TS are at evidence level C. ERT, Estrogen replacement therapy and estrogen deficiency; GH, GH therapy and GH deficiency.

**TABLE 3.** Maximum aortic diameter in TS using cardiac magnetic resonance

| First author (Ref.) | Age (yr) | n | Assessed | Aortic diameter in comparison with controls | Aortic dilation: TS | Association[a] |
|---|---|---|---|---|---|---|
| Cleemann (78) | <24 | 41 | 9 positions | Absolute: smaller in TS except for similar in proximal descending compared to controls (n = 50); BSA-indexed: similar except for arch, isthmus, and descending where smaller in TS | 15%, total[b] | BSA, +; CoA, − |
| Hjerrild (81) | >18 | 97 | 8 positions | Absolute: comparable in TS to controls (n = 24) for all, except smaller at distal arch and isthmus | 23%, total[c] | BSA, +; BAV, +; blood pressure, +; age, + |
| Ostberg (75) | >18 | 128 | 2 positions | Absolute: mid-ascending and mid-descending in TS comparable to controls (n = 36); height-indexed: both aforementioned positions enlarged after adjustment for height and BSA | 16%, mid-ascending[b] | BAV, +; age, + |
| Matura (289) | >18 | 166 | 2 positions | Absolute: mid-ascending similar and mid-descending smaller compared to controls (n = 26); BSA-indexed, larger mid-ascending aorta in TS and similar descending aorta | 24%, mid-ascending[c] | BSA, +; BAV, +; ETA, +; karyotype, + |
| Kim (92) | <26 | 50 | 9 positions | | 2–30%, all positions, highest for aortic sinus and sinotubular junction[d] | Age, +; PAPVR, −; ETA, + |
| Dawson-Falk (80) | <35 | 40 | 1 position | | 13%: ascending aorta | |

BSA, Body surface area; ETA, elongated transverse aortic arch; BAV, bicuspid aortic valve; PAPVR, partial anomalous pulmonary venous return; CoA, coenzyme A.

[a] Associations indicate positive (+) or inverse (−) correlation between aortic diameter and the variable in question.

[b] Aortic dilation was defined as 2 SD above the mean of the control group.

[c] Aortic dilation was defined as a maximum aortic diameter exceeding 3.4 cm after height adjustment.

[d] Aortic dilation was defined as two standardized z-scores above the normal values of children and adolescents.

[e] Aortic dilation was defined as >95% CI of nomograms for BSA and aortic diameter derived from x-ray computed tomography.

lesions (Table 2). Furthermore, a risk of aortic dissection is introduced in association with diagnostic cardiac and aortic catheterizations (87, 88, 123). Aortitis as a cause of dissection has been reported in TS, but only in one case (128). With insight into the causative mechanism of aortic dissection being scarce, much is left to the attending clinician on a case-to-case basis. This is especially true because the presently acknowledged risk factors are likely to fail to identify at least 10 of 100 acute aortic syndromes (127, 129).

### *1. Aortic diameter*

Aortic diameter is the principal risk stratification tool for aortic dissection (130). Assessed by cardiac magnetic resonance, absolute aortic diameter is smaller in young girls with TS compared with healthy peers (Fig. 4) (78). Conversely, absolute aortic diameter is comparable to normal healthy peers in adulthood (allowing for smaller diameter in the arch, isthmus, and proximal descending aorta) (42, 81). Hence, aortic diameter seemingly catches up from childhood to adulthood with regard to aortic size, and this occurs despite a persistently smaller height and weight in TS. Aortic diameter in adults with TS is noted to have a larger range than healthy female peers, emphasizing the essential clinical issue with separating a normal aortic diameter from an abnormal one in TS (Table 3) (75, 78). Echocardiographic studies generally support these findings (52, 81), with one study inferring a very early enlargement of aortic diameter in children with TS (9).

Physical stature is a determinant of aortic diameter in TS (42, 81). Even when normalizing aortic diameter for body surface area, aortic dilation is evident in adults with TS in the form of larger indexed diameters at all levels (except the distal aortic arch and adjacent to the classical site of coarctation of the aorta) (42, 81). In younger females the same applies around the coarctation site, whereas other diameters are comparable to controls (78). Nevertheless, the prevalence of dilation approaches 30% when using z-scores to normalize aortic diameter in young cohorts (92). Aortic dilation is also common in adulthood, and certain factors have been found to contribute to the variation in aortic diameter in TS. Much still needs to be clarified in relation to factors determining aortic diameter, and aortic dilation may occur in the absence of such factors (67, 131). The identified determinants of aortic size are:

- Body surface area (42, 78)
- Increasing age (9, 81)

- Systemic blood pressure (81, 132)
- Karyotype (67)
- Coarctation of the aorta (67, 78)
- Elongated transverse aorta (67)
- Bicuspid aortic valve (42, 81)

Insight into aortic dilation in TS is improving with better imaging, allowing pervasive characterization of the entire thoracic aorta and expanding our knowledge of an aortic disease that extends beyond the ascending aorta. At echocardiography, acoustic windows are often restricted due to a "barrel-shaped" chest (81, 133). Magnetic resonance and x-ray computed tomography circumvent this issue and may with orthogonal postprocessing of three-dimensional imaging provide an opportunity to minimize measurement error due to angulations in two-dimensional imaging (134). In keeping with this, echocardiography in TS underestimates magnetic resonance diameters in the ascending and descending aorta (75, 80, 133). As in the general population (130), cardiac magnetic resonance is emerging as a "gold standard" for delineation of anatomical aortic variants and aortic diameter when these occur downstream from the aortic sinus. Importantly, cardiac magnetic resonance has limitations beyond cost and availability, being contraindicated in the presence of ferromagnetic objects that even with magnetic resonance-compatible devices may create significant artifact. Yet, approximately 90% of girls and women with TS will successfully complete magnetic resonance studies (75, 133).

#### *2. Risk stratification for aortic events*

An enlarged aortic diameter increases the risk of aortic dissection in TS (42). However, aortic dissection may occur with aortic diameters that are considered "normal" even after normalizing to the smaller physical stature in TS (127). Aortic dilation appears to offer good positive predictive value for future aortic dissection (42), whereas the negative predictive value is less encouraging (127). This is in accordance with evidence from the general population, where on one hand absolute aortic diameters associate with a nonlinear increment in risk (135), whereas on the other hand 20% of all aortic dissections strike at diameters below 45 mm (136). The proposal to normalize aortic diameter using body surface area arose from this shortcoming (137), but the implications to risk need further assessment; outcome studies in the general population using indexed diameters are limited, and guidelines base surgical cutoff values on nonindexed numbers (130). In keeping with this, statements on critical aortic diameter have remained conservative in TS (2) because there is no firm evidence for a specific cutoff value, where the risk associated with prophylactic aortic root replacement is superseded by the risk of acute aortic events (130).

Aortic dilation over time provides important prognostic information in populations that face a high risk of aortic dissection and rupture (130). Hence, aortic growth is a potential clinical marker that may assist in the complex risk assessment in TS. Changes in aortic diameter in TS were first prospectively studied using echocardiography. The ascending aorta grew 0.24–1.22 mm per year during 37 months in a mixed pediatric and adult cohort (age range, 3–39 yr) (133). A subsequent magnetic resonance study of a purely adult cohort showed that ascending aortic diameters increased over 29 months by 0.1–0.4 mm per year, and bicuspid aortic valves (but no other congenital anomaly or blood pressure) predicted aortic growth rates (134). For comparison, estimated aortic growth (using different imaging modalities) is 0.07 mm/yr in normal females (138), 0.1 mm/yr in thoracic aortic aneurysm populations (139), and 0.2 mm/yr in the setting of a bicuspid aortic valve (140, 141). The clinical significance of the reported aortic growth rates in TS is not known because the growth rates have not been linked with outcomes. Nevertheless, growth rates appear to be at least equivalent to other states of thoracic aortic disease, and growth rates may when used cautiously add weight to risk stratification also in TS.

Other measures than indexing to body surface area have been suggested in order to avoid over- or underestimation of thoracic aortic disease in TS (75). This includes a patient-specific ratio of ascending to descending aortic diameter (142), which is unfortunately inherently flawed in TS, where ascending aortic dilation will be underestimated in the context of frequent descending aortic dilation (75, 81). Attempts to quantify thoracic aortic disease using indices other than size have been made in TS. The aortic wall appears to be stiffened compared with healthy peers, although this is not a consistent finding, and the implications in terms of stratification for the risk of aortic dissection remain to be defined (132, 143–145).

#### *3. Pregnancy and aortic dissection*

The risk of aortic dissection and rupture during pregnancy in TS may be as high as two in 100 pregnancies (146–148), with maternal mortality approaching 86% (127). An increased occurrence of systemic arterial hypertension, if not overt eclampsia, may contribute to this risk (149, 150). However, some report gestational hypertensive disorders in TS, equally common in TS and in assisted pregnancies in general (146, 151). There are no outcome-driven criteria for absolute and relative contraindications to pregnancy (or the risk to mother and fetus, and mode of birth), although cardiovascular investigation before and

during pregnancy clearly is crucial (147). Aortic diameter exceeding 2.0 cm/m$^2$ has been proposed as an absolute contraindication for pregnancy in TS (152). Then again, aortic diameter may not accurately define and monitor the risk of dissection associated with pregnancy because aortic size was comparable between nulliparous and parous females with TS (147). While waiting for further evidence, we may have to refrain from categorical statements and limit advice to "severe" aortic dilation and complex congenital heart disease weighing against pregnancy. Therefore, counseling regarding pregnancy in TS needs to be given on a case-by-case basis, and preferably with cardiologists experienced in congenital heart disease and thoracic aortic disease taking center stage. On the note of counseling, unassisted pregnancy occurs in 2–5 per 100 females with TS (147, 153), and even females with 45,X monosomy or karyotypes with Y-material can become pregnant (146). Hence, careful information about birth control and hormonal contraceptives as well as the cardiovascular risk associated with assisted and unassisted pregnancy should be intrinsic to clinical care in potentially fertile age groups in TS (154).

#### *4. Etiology of aortic dilation*

Insight into the causative mechanism of aortic dilation, dissection, and rupture is scarce. Intima-media changes with cystic medial degeneration have been reported in 42–72% of aortic dissections in TS (123, 155). Collagen fiber composition may also be altered (123). These microscopic changes in the aortic wall composition compare well with other states of thoracic aortic aneurysm formation, where changes in vascular smooth muscle cells, elastin, collagen, and other extracellular matrix components are encountered (156). Normal tissue homeostasis within the intima-media layer results mainly from a complex interplay of contractile, secretory, and proliferative activities of vascular smooth muscle cells (156). These cells maintain the extracellular matrix in delicate interaction with extracellular matrix components, which in turn are regulated by biochemical and mechanical stimuli of the aortic wall microenvironment (156). Interactions with vascular smooth muscle cells include factors such as wall stress from the propagating aortic pulse wave (157) and blood flow perturbations in the setting of bicuspid aortic valves (158, 159). Vascular smooth muscle cells may also be regulated by cytokines secreted by immune and endothelial cells (156). In thoracic aortic aneurysms, this equilibrium of the media layer is shifted toward structural degeneration, and this process is felt secondary to a primary intrinsic wall defect leading to mechanical and elastic aortic failure (156). The culprit lesion for these adverse changes can be increased synthesis of degenerative matrix metalloproteinases (or reduced inhibitory molecules) from vascular smooth muscle cells (160). This is the case for Marfan and Loeys-Dietz syndromes (161, 162), where TGF-$\beta$1-mediated stimulation of vascular smooth muscle cell synthesis of matrix-degrading metalloproteinases is increased. These proteins then cause lyses of extracellular matrix proteins with secondary loss of structural integrity (160, 163). Other mechanisms may also be involved in increasing local protease activity, such as stimulation by immune cells within the adventitia layer (164). In theory, aortic disease in TS could result from disruption of the same or parts of these disrupted pathways of synthesis and degradation. There is a call for further studies in TS, where a myriad of factors could be causative as primary lesions or exacerbating components. Hypertension, tachycardia, or flow perturbation in relation to bicuspid aortic valves may, for instance, provide the substrate for an adverse mechanical stimulus within the aortic medial layer (49). Alternatively, immunological hyperreactivity could evoke an adverse shift in the extracellular matrix components within the aortic wall (165, 166).

Aortic wall disease may also result from a primary defect in the structural components of the extracellular matrix in TS, such as in vascular Ehlers-Danlos syndrome (167), or a primary conformational change of vascular smooth muscle cells (168). Furthermore, an ontological component has been inferred in the pathogenesis of thoracic aortic aneurysm in general (126). Aneurysm in the thoracic aorta is now considered a nonatherosclerotic process in contrast to the mainly atherosclerotic abdominal aortic aneurysms (although atherosclerosis may be superimposed in thoracic aortic disease) (130, 169). A hitherto undetermined fetal process may also be the source of cystic medial degeneration in TS, as seen in aortic dissection in this cohort (123, 127). Intriguingly, such aortic wall pathology was also observed in a 20-wk-old 45,X fetus (Fig. 8) (94). Although seen in only one case, this finding certainly infers a primary defect that could then potentially be exacerbated by the presence of other factors both ante- and postnatally. Conclusively, delineation of the precise chain of events leading to thoracic aortic aneurysm formation leading to a more complete understanding, awaits discovery both for TS and other states of syndromic and nonsyndromic aortic dilation. Certainly, relevant points of focus might include the TGF-$\beta$ system or activation of SMAD pathways in TS (112), as well as a more in-depth delineation of aortic wall changes to ascertain other potentially involved pathways.

There are concerns that recombinant human GH treatment and estrogen replacement therapy could induce unfavorable aortic wall changes. Treatment with GH may directly influence the homeostasis within the media layer

**Figure 8.**

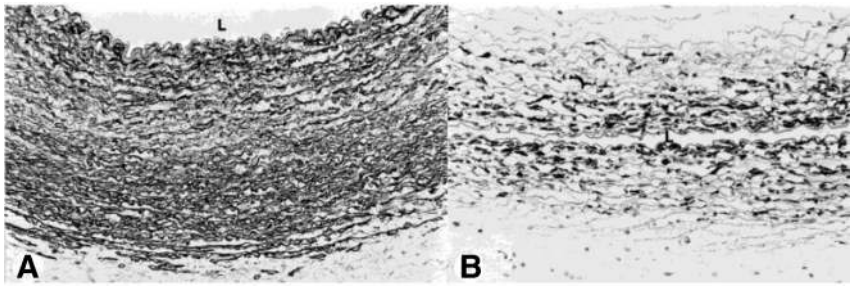


Figure 8. Changes within the tunica media are present *in utero* in TS. Aortic wall pathology may be present from an early stage, as seen in an autopsy of a 20-wk-old 45,X fetus that presented with markedly reduced numbers of smooth muscle cells and elastic fibers in a thin-walled and hypoplastic aorta (A). The appearance of the aortic wall was consistent with degeneration of the aortic media layer (A) and differed markedly from the normally developed aortic wall of a 21-wk-old normal fetus (B) (94). Illustrations are from a photomicrograph where smooth muscle cells are demonstrated with anti-α-smooth muscle actin antibody, and elastic fibers are stained with Victoria blue. [Reproduced from S. Miyabara *et al.*: Developmental analysis of cardiovascular system of 45,X fetuses with cystic hygroma. *Am J Med Genet* 68:135–141, 1997 (94) with permission. © Wiley-Liss, Inc.]

of the aortic wall, as seen in animal and human cell models (170, 171). Alternatively, such treatment may impose mechanical stress on the aortic wall as a part of a hyperkinetic syndrome comparable to that observed in acromegalic individuals (172). Hitherto, there has been no overtly adverse impact of GH treatment on aortic diameter in TS, although conclusions are limited by primarily retrospective designs without adequate controls or pretreatment comparison (144, 173). Estrogen replacement therapy appears to reduce central arterial stiffness (174), improve endothelial function (175), and decrease carotid intima-media thickness (176) in short-term studies in TS. Hence, an improved risk profile for aortic dissection has been inferred. The precise relevance of these findings to the risk of aortic events in TS is undefined. On the note of treatments that aim to decrease morbidity and mortality in TS, the extension of the experiences with prevention of dilation in Marfan syndrome (177, 178) to TS would be highly interesting. However, specific evidence is undoubtedly needed for this cohort in light of the comorbidities that renders direct comparisons with non-TS studies hazardous.

### B. Stroke

Stroke incidence in TS is increased from the second decade of life onward, overall causing increased morbidity (RR, 2.7) and mortality (SMR, 3.9) (3, 4). Only one cohort study has attempted to ascertain the etiology of stroke in TS (4). With the limitations of death certificates in identifying causes of death, an estimated 90% of events were hemorrhagic (4). Case reports have described numerous causes, which are more or less common in the general population:

- Atherosclerosis (179)
- Hemorrhage (180)
- Fibromuscular dysplasia (181)
- Moyamoya disease (182)
- Hemophilia (183)
- Congenital carotid hypoplasia (184)
- Hemangioma (185)
- Dissection of head and neck vessels (186)

Naturally, deriving causative mechanisms from solitary case sources is a dubious act due to likely publication bias. The verdict is pending, and the etiology will only be clarified with a systematic assessment of both morbidity and mortality related especially to the contribution of ischemia, intracerebral hematoma and subarachnoid hemorrhage to stroke in TS (common causes in the background population) (187).

#### *1. Cerebrovascular arteriopathy*

The burden of general population risk markers for stroke of any cause (188) is increased from a young age in TS, and among other markers includes hypertension, insulin resistance, and visceral adiposity (122, 189). These markers may influence the cardiovascular system toward early aging (190), which in turn may predispose to ischemic or hemorrhagic stroke (187). Early cardiovascular aging (191) may potentially superimpose onto any adverse *in utero* programming of the cardiovascular system (192) and be aggravated by metabolic complications (193) related to the low birth weight in TS (18). Estrogen and GH deficiencies (or equally incorrectly administered replacement therapies) could further deliver a negative impact on the vasculature (194, 195).

Carotid intima-media thickness is increased as early as the second decade of life in TS (143, 196). This surrogate risk marker quantifies the combined thickness of media and intima layer, which for the elastic carotid arteries predominantly reflects intimal pathology (principally atherosclerosis) (197). Intima-media thickness predicts the risk of stroke and coronary atherosclerosis in the general population, adding value beyond traditional risk markers (198, 199). Consequently, increased intima-media thickness in young females with TS may indicate a premature atherosclerosis. In a pediatric cohort with TS, intima-media thickness was not abnormal, but there were correlations with atherogenic factors (200), which does align with a hypothesis of early atherosclerosis. In adulthood, intima-media thickness continually associates with ath-

erosclerotic risk markers, including blood pressure, C-reactive protein, clotting factors, lipids, and age (143, 196, 201). Outcome studies are completely lacking for TS, but note can be made that increased intima-media thickness is generally considered an even more adverse prognostic omen when encountered in the young (202). Perturbations in other indices that may also reflect premature atherosclerosis have been identified in TS, such as aortic stiffness, augmentation index, and ambulatory arterial stiffness index (143, 196, 203).

Arterial dilation extends beyond the aorta and into the large arteries in TS (67, 196). Therefore, a primary and intrinsic, generalized arteriopathy may include the cerebral vasculature and potentially cause small-vessel disease and stroke (187) or promoting dissections within regional arteries (186, 204). Case reports of stroke have accounted for vascular smooth muscle cell deregulation in fibromuscular dysplasia and moyamoya disease in TS (205, 206). These arterial disorders are characterized by luminal narrowing rather than dilation (207, 208). However, their presence could be seen as indicative of disequilibrium of vascular smooth muscle cell activity and extracellular matrix components. Vascular smooth muscle cells of the head and neck arteries notably share embryonic origin with aortic vascular smooth muscle cells, stemming from pharyngeal arch arteries (I–III). The cerebral arteries are subject to the same regulatory processes during angiogenesis (112), which could support a common primary defect for arterial disease in TS. Intriguingly, other states of media layer deregulation can have a systemic component, as seen in skin biopsies of cervical artery dissections (209). Conversely, the observed association of thoracic aortic disease and conduit artery dilation in TS (67) could be a hemodynamic effect rather than marking a universal arterial disorder.

#### *2. Thromboembolism*

Atrial fibrillation could increase the risk of thromboembolic stroke in TS, although reports of atrial tachycardia are limited to date (210–212). However, P-wave dispersion is increased in TS (213) as a potential substrate for atrial fibrillation (214). Risk factors for atrial fibrillation (hypertension, type 2 diabetes, *etc.*) are overrepresented in TS, and arguably atrial arrhythmia should occur at least with the general population prevalence of one in every 200 (215). Systematic assessment is warranted, especially with atrial fibrillation often remaining subclinical until associated events unmask the arrhythmia. Other possible sources of thromboembolic stroke in TS include congenital cardiac lesions such as intracardiac shunts and thrombus (216), or calcifications and vegetations on abnormal mitral and aortic valves (217). Thromboembolism may also develop in the setting of heart failure (218), which is an anticipated feature in TS in association with the increased incidence of ischemic heart disease and type 2 diabetes, or even in normotensive and euglycemic young females with cardiomyopathy (70).

#### *3. The coagulation system*

Procoagulant disorders of the clotting system can lead to thromboembolic stroke, which is especially relevant for stroke in the young (217), and this may be a contributing factor for often premature cerebral events in TS (4). No outcome studies for thrombotic disease are available in TS. Nevertheless, the risk of thrombus formation seems increased even in the absence of functional or morphological cardiovascular substrates (Table 4). Clotting factors and times may be normal for cohorts with TS when assessed as a whole (212, 219). On the individual level, many will, however, have procoagulant levels of clotting and fibrinolytic factors (201, 219). This has been shown for C-reactive protein (40%), fibrinogen (15%), fibrin D-dimer (15%), factor VIII (25%), and von Willebrand factor (15%) (201). Proteins C and S may also be reduced (220). Inflammatory markers and clotting factors correlate with carotid intima thickness, heart rate, and blood pressure (201), which support a prognostic significance of clotting abnormalities. Of procoagulant mutations, only

**TABLE 4.** Reported events related to thrombosis in TS

| First author (Ref.) | Event | Age (yr) | Karyotype | Clotting system abnormality | Other |
|---|---|---|---|---|---|
| Jobe (350) | Deep venous thrombosis | 17 | 45,X/46,XY (q11.2) | Prothrombin G20210 mutation | Ongoing ERT |
| Donal (351) | Aortic thrombus | 56 | | | |
| Kopacek (352) | Portal vein thrombosis | 3 | 45,X | ↑ Factor VIII, ↑ von Willebrand factor | |
| | Portal vein thrombosis | 1 | 45,X | ↑ Factor VIII, ↑ von Willebrand factor | |
| | Portal vein thrombosis | 12 | 45,X | ↑ Factor VIII, ↑ von Willebrand factor | |
| Ureten (353) | Atraumatic osteonecrosis | 38 | 45,X | ↓ Protein S | Ongoing ERT |
| Pinto (354) | Portal vein thrombosis | 1 | | ↓ Protein S, ↓ Protein C, heterozygote for MTHFR mutation | |
| | Portal vein thrombosis | 2 | | ↓ Protein C; heterozygote for MTHFR mutation | |

Case reports of thrombotic and related disease in TS, occurring in the absence of reported morphological and functional disturbance to explain the lesions beyond perturbations in the clotting system. ERT, estrogen replacement therapy.

factor V Leiden G1691A gene polymorphism heterozygosity has been found more commonly in TS (13%) compared with the background population (2%) (201). An in-depth characterization of disturbances of the clotting system, their etiology, and any implications for morbidity is warranted in TS. This also applies to tendencies toward increased bleeding as a cause of hemorrhagic stroke in TS patients (221, 222), where X-linked recessive hemophilia could hypothetically manifest as frequently as in their male counterparts.

### C. Ischemic heart disease

Ischemic heart disease is more frequent in TS than in general (SMR, 3.5) (1). These events mainly raise mortality rates from the fifth decade of life in TS, occurring with incidences skewed toward older age groups when compared with other acquired heart diseases (4).

The origin of coronary artery disease in TS has not been formally assessed. A plethora of causative mechanisms conceivably contribute to causing these events as early as in the fourth decade of life. Reported causes of myocardial infarction in TS include atherosclerosis, embolism from intracardiac thrombus or coronary artery involvement in aortic dissection (223–225). However, the etiology has in many cases not been identified (226), and causative mechanisms are not easily characterized by case reports. However, looking toward atherosclerosis, in the capacity of being most common in the background population, there is ample possibility for premature atherosclerosis. Markers for atherosclerotic heart disease are frequent in TS:

- Hypertension (81)
- Insulin resistance (227) and overt diabetes (1)
- Dyslipidemia (228)
- Obesity (229)
- Estrogen deficiency (229)
- GH deficiency (229)
- Hypercoagulable clotting system (201)

Coronary artery anomalies have also been reported in TS. These include coronary arterial dilation (230), single coronary ostium (231), coronary fistulas (232), and aberrant origin from the descending thoracic aorta (233). The prevalence of these anomalies is completely unexplored. From an embryonic perspective, the proximal coronary arteries share origin with the ascending aorta and aortic valve, and adequate migration of neural crest cells and appropriate differentiation into vascular smooth muscle cells are essential to all of these structures (112). Hence, and in line with the aortic and cerebral vascular territories in TS, the delineation of connections between coronary heart disease with ontological programming of an intrinsic arterial wall defect and the interplay with comorbidities awaits further scientific progress.

### D. Hypertension

Arterial blood pressure is raised in all ages of TS patients (9, 132, 212). Hypertension affects 21–40% of children and adolescents (9, 122) and 50–58% of adults (81, 132). Characteristically for TS, hypertension affects systolic, diastolic, and pulse pressures, and there is a blunted nocturnal dipping pattern (122, 234). Morbidity (RR, 2.9) and mortality (SMR, 6.0) directly attributed to hypertension are substantial (3, 4), and there is an unfortunate degree of nondiagnosis with only 4–22% receiving treatment (9, 212).

#### *1. Essential hypertension*

Hypertension is principally described as being essential by nature in TS (212, 235), and a multifactorial etiology thus prevails. Relative sympathetic hyperactivity in TS provides a possible foundation for high output hypertension (49, 118). Increased activation of the renin-angiotensin-aldosterone system has also been indicated (235, 236) with potential secondary salt and water retention, arteriolar construction, and potentiated sympathetic drive. Conversely, lower peripheral vascular resistance has been demonstrated in young girls with TS, but this finding could relate to ongoing recombinant GH treatment (237). Abnormal levels of renin associate with abnormal captopril challenges in TS (236), but outright renovascular hypertension is rare (238). Conversely, normal renin and aldosterone have been reported for TS (49).

Oral contraceptives and postmenopausal hormone therapy have been claimed to induce hypertension (239). For TS, the verdict on this matter is unsettled (Table 5). Estrogen replacement therapy induced abnormal renin levels and captopril responses (236), and a cross-sectional study found elevated blood pressure in females with TS on oral estrogen replacement therapy (240). As opposed to these studies, renin levels were not affected by estrogen in another cohort with TS (49). Furthermore, it has been implied that estrogen counter-regulates the renin-angiotensin-aldosterone and sympathetic nervous systems in animal and human models (241). In a mixed population of patients with premature ovarian failure that included TS, transdermal 17-$\beta$-estradiol (and vaginal progesterone) reduced 24-h blood pressure and serum angiotensin II when compared with baseline and treatment with synthetic ethinyl estradiol (and norethisterone), with unchanged serum aldosterone (242). So the general appearance is that estrogen deficiency leads to a slight elevation of blood pressure, and that appropriate estrogen treatment could lower

**TABLE 5.** Cardiovascular, metabolic, and clotting factors during estrogen replacement therapy compared with no treatment in TS

| | Gravholt (234, 276) | Papagianni (309) | Ostberg (176) | Elsheik (174) | Chan. (175) | Host (312) | Lanes (219) |
|---|---|---|---|---|---|---|---|
| Participants | | | | | | | |
| n | 26 | 12 | 14 | 21 | 7 | 8 | 5 |
| Mean age (yr) | 33 | 15 | 32 | 32 | 29 | 29 | 17 |
| Intervention | | | | | | | |
| Duration | 4-month pause | 12-month treatment | | 3-month pause | 6-wk pause | 2-month pause | 6-month treatment |
| Route | Oral/transdermal | Oral | Oral | Oral | Oral | Oral | Oral |
| Status when on ERT | | | | | | | |
| Hemodynamic | ↓ 24-h diastolic AMBP, →AASI | | ↓ IMT, →FMD, →PVW, →AIX | ↓ AIX, →clinic BP | ↑ FMD, →clinic BP | | |
| Metabolic | ↓ Glucose tolerance, ↓ insulin, →glucose, ↑ FFM, →BMI, →WHR, ↑ HDL | ↑ HDL, →other lipids | ↑ HDL, ↓ glucose | ↓ Insulin, ↓ glucose, ↓ WHR, →BMI, →lipids | →Lipids | | →Lipids |
| Clotting system | | →APTT, →fibrinogen, →TRC | →Fibrinogen | | | | |
| Other | ↑ $VO_2$ max | | →IL-6, →CRP | | | ↓ Adiponectin | →PAI-I |

AASI, Ambulatory arterial stiffness index; AIX, Aortic augmentation index; AMBP, ambulatory BP; APTT, activated partial thromboplastin time; BMI, body mass index; BP, blood pressure; CRP, C-reactive protein; ERT, estrogen replacement therapy; FFM, fat free mass, FMD, forearm flow-mediated dilation; HDL, high-density lipoprotein; IMT, carotid intima-media thickness; $VO_2$ max, maximum oxygen absorption capacity; WHR, waist-to-hip ratio; TRC, platelet count; PAI-I, plasminogen activator inhibitor-I; PVW, pulse wave velocity.

blood pressure to a similar degree as antihypertensive monotherapy.

Other causes of hypertension are common in TS, such as insulin resistance [seen in some (227, 243) but not all studies (189, 244)], visceral adiposity (245, 246), and GH deficiency or resistance (8). Altered cardiovascular autoregulation due to adverse imprinting in fetal life or poor thriving early in antenatal life (247) may very well raise the risk of hypertension in TS also. Intriguingly, metabolic syndrome appears to have a component of prenatal programming in TS (248), and hypertension is a well-established component of this syndrome (249). Further study of a putative prenatal programming component is required because findings are unequivocal with birth weight and blood pressure unassociated in another study of TS (122).

### *2. Secondary hypertension*

Classical secondary hypertension is uncommon in TS. Nonetheless, hypertension does associate with vascular pathology in some females, such as coarctation of the aorta (repaired and unrepaired) (250) or, less commonly, hypoplastic transverse aortic arch (13). Still, congenital heart disease and hypertension do not always coexist (235). Renovascular (238) and endocrine hypertension (181, 251) are rare in TS, although various renal malformations occur in a third of all TS (252). Despite this, hypertension may develop secondary to impaired renal function in TS (132).

### *3. Sympathovagal dysfunction*

Sympathetic adrenergic drive is relatively increased in TS (49), which is present in utero and evident as sinus tachycardia (253). A primary developmental component is therefore likely, as is also indicated in a recent stem cell model of TS (53). However, this does not preclude a contribution for acquired factors. Certainly, changes in glucose metabolism are common in TS (189), and complications analogous to those seen in diabetic autonomous neuropathy could be of importance (254). Estrogen also contributes to normal autonomous regulation. This is appreciated by QT interval fluctuations and arrhythmic potential fluctuating over the menstrual cycle as well as by the relative tachycardia and heart rate-corrected QT-interval elongation induced by female puberty (194). Hence, estrogen deficiency in TS may play a role, although the sympathetic drive relative to parasympathetic activity was not affected by short-term estrogen replacement therapy (49). Another relevant factor in TS is the potential disturbance to the renin-angiotensin-aldosterone system (236) and its link to the overly active sympathetic nervous system.

### *4. Prognostic impact of hypertension*

Hypertension contributes to aortic dilation and dissection in TS (81, 127). In addition, elevated blood pressure marks an increased risk of ischemic heart disease and stroke (188, 255). Outcome studies in TS are lacking, but several studies have demonstrated an association between blood pressure elevation and intermediate cardiovascular markers such as carotid intima-media thickness, aortic pulse wave velocity, and aortic diameter (81, 132, 143, 196). Hypertension also causes left ventricular failure in the general population (256). Presumably, a causal rela-

**TABLE 6.** The left ventricle of the heart in TS

| First author (Ref.) | No. of participants (age) | Selection criteria | Comparative abnormalities | Putative involved mechanisms |
|---|---|---|---|---|
| Andersen (70) | 33 (>18 yr) | +, ERT; ÷, HTN, diabetes, IGTT, LVEF <50%, CHD (excl. bicuspid aortic valve) | Diastolic dysfunction (27%), systolic dysfunction (normal LVEF),[a] left atrial dilation | Hypertension, sympathovagal dysfunction, aortic arch abnormalities, aortic valve dysfunction, intrinsic cardiac defect, estrogen deficiency, GH deficiency, insulin resistance, thyroid disease |
| Sozen (355) | 31 (>18 yr) | ÷, HTN,[b] CHD, ERT, all medicines | Diastolic dysfunction, left ventricular hypertrophy, left ventricular dilation, left atrial dilation | |
| Tancredi (261) | 50 (<38 yr) | ÷, Aortic dilation[c] | Diastolic dysfunction, left ventricular hypertrophy, systolic dysfunction (normal LVEF), left ventricular dilation | |

Abnormal left ventricular function and geometry is a common finding on echocardiography in TS, although many abnormalities may only be appreciated in their true extension when taking the smaller physical stature of women with TS into account. Overall, the pathologies balance toward hypertrophy and enlarged chamber dimensions, contrasting indications of cardiac hypoplasia in fetal life. CHD, Congenital heart disease including aortic valve disease; ERT, estrogen replacement therapy; HTN, hypertension; IGTT, impaired glucose tolerance; LVEF, left ventricular ejection fraction; +, all received ERT; ÷, none had HTN, diabetes, IGTT, LVEF < 50%, and CHD.

[a] Systolic dysfunction was seen as decreased myocardial strain rate with normal ejection fraction when compared to controls (n = 33).

[b] Diagnosed hypertension was an exclusion criterion, but systolic blood pressure was higher in TS than in controls (n = 30).

[c] Hypertension was not an exclusion criterion, and diastolic blood pressure was higher in TS than in controls (n = 56).

tion may also be present in TS between the common hypertensive disorder and widespread dysfunction and abnormal geometry of the left ventricle in TS, although such association remains to be assessed in unselected cohorts in TS (Table 6).

Appropriate thresholds for antihypertensive treatment are not available in TS. However, the burden of cardiovascular morbidity and mortality certainly justifies tight blood pressure control. When a considerable burden of cardiovascular disease, including aortic dilation and risk of dissection, is present, lowering blood pressure to the lowest tolerated levels is probably paramount (130). There is no evidence to support specific treatments in TS, and the choice of therapeutic agent should follow general guidelines where reduction in blood pressure is the principal goal (257). However, some antihypertensive medicines may offer secondary benefits beyond lowering of blood pressure such as reducing progression of disease within the aortic wall in TS. Again, evidence in TS is nonexistent, and clinicians have to look to other cohorts with increased risk of aortic dilation and dissection, bearing in mind that the aortic pathology has not been disclosed in TS. The general recommendation for Marfan syndrome has been to use $\beta$-adrenergic receptor blockade as an aortic wall-stabilizing agent. Angiotensin receptor antagonists now challenge this practice by potentially superior prophylaxis for aortic dissection through TGF-$\beta$1 antagonism (177, 178, 258). Then again, combined $\alpha$- and $\beta$-adrenergic receptor blockade reduced the incidence of aortic dissection in type IV vascular Ehlers-Danlos syndrome (259). Although some lessons may be learned from Marfan syndrome, clinicians must bear in mind that causative mechanisms behind aortic dilation are unidentified in TS, and unequivocal evidence of superiority of one agent over the other is lacking, with prospective trials highly awaited. To date, only one study has showed an actual reduction in blood pressure during a generally increased attention to cardiovascular risk prophylaxis in TS (134). Unfortunately, this study allows no conclusions other than that blood pressure can be controlled through the use of antihypertensive medicines, as also indicated with short-term estrogen replacement (234).

### E. Arrhythmia

Sinus tachycardia is a lifelong phenomenon in TS (55, 260, 261). Conduction of the electrical impulse through the atria and atrioventricular node is accelerated (121), and the risk of atrial tachycardia may be increased (213), whereas bradycardia is rare (262). Furthermore, the myocardial action potential is prolonged with delayed repolarization, with pathological prolongation present in 33–36% of children and 21% of adults with TS (*i.e.*, heart rate-corrected QT interval >440 msec) (55, 121). These abnormalities of excitability and conduction are negative prognostic omens in the general population (263, 264), whereas their predictive capacity in TS remains to be determined. So far in TS, only one case of sudden, unexplained death has been related to known prolongation of the QT interval (265).

The cause of altered cardiac electrophysiology is unknown, but several aspects suggest an inherited defect:

- The 45,X karyotype has an adverse impact on the heart rate-corrected QT interval (although not consistently in all studies) (55, 121), and
- The heart rate-corrected QT interval prolongation is not associated with traditional indices of heart rate-corrected QT interval elongation (*i.e.*, age, electrolytes, left ventricular hypertrophy, medicines, coronary heart disease, and thyroid disorders) (55, 121).

It remains to be resolved whether the X-chromosomal haploinsufficiency in TS results in altered transcription of proteins involved directly in the activity of ion channels as structural or regulatory components, such as is seen in congenital long QT syndrome (266). Alternatively, altered expression of X-linked genes may function as transcriptional regulators of autosomal genes involved in ion channel activity. Interestingly, the action potential of the cardiomyocyte-like differentiated pluripotent stem cells seems prolonged in TS (53), which points toward a developmental abnormality. Further insight is necessary into the etiology, also including the role played by the relatively increased sympathetic drive seen in TS (49, 118), and likely increases automaticity, excitability, and conductivity of cardiomyocytes and cardiac conduction tissues.

## V. Endocrine and Metabolic Risk Factors

### A. Growth hormone deficiency or resistance

*SHOX* haploinsufficiency causes reduced final height in 95–99% of females with TS (267, 268). In addition to this genetic perturbation of growth, there is an unresolved imbalance in the "GH–IGF-I–IGF binding protein" axis. This disturbance may involve relative resistance to GH actions (269, 270), pituitary production failure of GH, or increased serum binding (8). Spontaneous and/or stimulated secretion of GH has been found to be diminished by some (271–273), whereas others have demonstrated normal GH secretion (274, 275). GH secretion is reduced by 50% in adulthood, with plausible influences from body composition (276). The bioactivity of circulating GH may also be reduced (277), and there are low levels of IGF-I as well as free and bioactive IGF-I (245). Moreover, proteolysis of IGF binding proteins is increased, which possibly impedes normal tissue delivery of IGF-I (278, 279). In view of this available evidence on GH signaling in TS and with normal final height being achievable with supraphysiological doses of recombinant human GH (280, 281), treatment should commence in infancy. Starting treatment early facilitates appropriate catch-up of skeletal growth, and it should be terminated upon reaching final height or when growth potential is no longer available (2).

Disorderly signaling within the GH–IGF-I axis may very well extend beyond the realm of physical stature in TS, affecting other organ systems such as the cardiovascular system. Comparatively, cardiovascular prognosis is impaired in overt GH deficiency (282, 283), and low levels of IGF-I increase the risk of stroke and myocardial infarction, even in the absence of a clearly pathological deviation of the GH–IGF-I axis (284, 285). The precise cause for this adverse association remains to be defined (195). However, GH stimulates hepatic IGF-I production through auto- and paracrine actions, which in turn regulates a myriad of cardiovascular cells, including cardiac myocytes, fibroblasts, and vascular smooth muscle cells (195). It is well established that many cardiovascular and metabolic features of TS are present in other states of GH deficiency (195). Additional study is needed to assess the role of a deviation in the GH–IGF-I axis to these common features that include insulin resistance (227) and visceral adiposity (245) as well as unfavorable changes in intima-media thickness (143), left ventricular function (70), blood pressure (212), sympathetic drive (49), exercise performance (261), and the clotting system (201). Recombinant GH treatment in adulthood is not advocated at present, and the importance of relative GH deficiency or resistance for morbidity awaits elucidation, with perturbed GH signaling continuing into adulthood in TS (286).

Increasing the activity of the GH–IGF-I axis from low normal levels benefits cardiovascular risk markers such as blood pressure, vascular resistance, and insulin sensitivity, as well as levels of the immune system (195). Conversely, hypersecretion of GH increases cardiovascular mortality as seen in acromegaly (287). In uncorrected acromegaly, an initial hyperkinetic syndrome with increased cardiac output is followed by diastolic left ventricular failure due to biventricular myocardial hypertrophy (fibrosis and cardiomyocyte hypertrophy), and ultimately dilated cardiomyopathy and heart failure ensue (288). Excess GH may also cause mitral and aortic valve calcifications, arrhythmia, and hypertension, just as glucose resistance is an issue (195). Therefore, unfavorable cardiovascular effects are a concern when supraphysiological doses of recombinant human GH are prescribed to girls and adolescents with TS over several years.

Studies of the cardiovascular impact of recombinant human GH in TS are limited by a lack of phenotype characterization before treatment (Table 7) as well a scarcity of insight into normal age-associated, cardiovascular development in children with TS. As is the case for estrogen replacement therapy, placebo-controlled studies are not likely to take place because these treatments are now recommended universally in TS (2). Somewhat reassuringly, available evidence suggests that recombinant human GH

**TABLE 7.** The impact of GH treatment on cardiovascular phenotype in TS

| | Sas (356) | van den Berg (357) | van den Berg (144) | Matura (289) | Bondy (173) | Radetti (237) |
|---|---|---|---|---|---|---|
| Participants (n) | 68 | 31 | 38 | 67 | 53 | 26 |
| GH duration | ≈7 yr | ≈9 yr | ≈9 yr | ≈4 yr | ≈5 yr | ≈5 yr |
| Design | Prospective | Retrospective (5 yr) | Retrospective (5 yr) | Retrospective[c] | Retrospective | Cross-sectional (ongoing) |
| Comparison group | Healthy peers | Healthy peers | Healthy peers | TS[d] | TS[d] | Healthy peers |
| Cardiac outcomes[a] | →LVM | →LVM, →RVM<br>↓ Biventricular volumes<br>→RV EF, →LV EF, →CI | | →LVM<br>→Biventricular volumes<br>→LV FS, →E/A ratio | | →LVM<br>→LV EF, ↑ LV FS, →CI<br>↓ Diastolic filling |
| Blood pressure | ↓ Diastolic BP<br>→Hypertension | →BP, ↑ HR | ↑ Diastolic BP, ↑ mean BP | | | ↓ SVR<br>↑ Diastolic BP, ↑ systolic BP, ↑ HR |
| Aortic outcomes[a] | | | ↑ Aortic diameter, ↓ aortic distensibility[b] | | →Aortic diameter | |

CI, Cardiac index; BP, blood pressure; EF, ejection fraction; FS, fractional shortening; HR, heart rate; LV, left ventricle; LVM, left ventricular mass; RVM, right ventricular mass; RV, right ventricle; SVR, systemic vascular resistance; E/A ratio, early/late (atrial) ventricular filling velocity.

[a] Cardiac and aortic measurements indexed for body surface area.

[b] Aortic distensibility was decreased in the mid-ascending aorta and at the level of the diaphragm.

[c] Mainly retrospective study with 65% having terminated their GH treatment.

[d] GH-treated individuals were compared to age-comparable individuals with TS who had never received GH treatment.

delivered over several years does not induce an exaggerated trophic stimulus on cardiomyocytes in TS (Table 7). Likewise, recombinant human GH does not seem to increase aortic diameter compared with nontreated females with TS (173). The impact of dose and duration of recombinant human GH is unresolved, although retrospective reports imply no adverse impact of the current doses on the aorta (144, 173, 289).

Mapping of any adverse or advantageous influences of GH treatment is essential because it is delivered to an age group where the incidence of aortic dissections is raised to 14–19 per 100,000 population years (123, 290). The cardiovascular system undergoes rapid changes with altered signaling in the GH–IGF-I axis in other states of abnormal production (288). Although evidence is limited, there are no clear indications of such influences in TS (237). Furthermore, some effects of recombinant human GH are likely to revert toward pretreatment levels even after long-term delivery. This has been noted for diastolic blood pressure in TS (291), although more convincingly for glucose metabolism (292, 293). Metabolic change may also impact the circulatory system during recombinant human GH treatment, where insulin sensitivity has mainly been reported to decrease (292–294). By contrast, lipoproteins either are unaffected (295) or undergo favorable changes (296), and visceral and total adipose tissues decrease (294, 297). The verdict is pending for metabolic influences during long-term delivery of GH treatment in childhood and adolescence. Prospective controlled studies focusing on metabolism as well as the cardiovascular system during recombinant human GH would be of great value.

### B. Estrogen deficiency

Estrogen deficiency is common in TS, where approximately 70% fail to undergo natural pubarche or menarche (298). Premature ovarian failure often ensues in adolescence, or early adulthood and is caused by apoptosis of ovarian follicles that commences in fetal life and progresses at varying rates (299, 300). The etiology of this gonadal demise is undefined. Umbilical cord blood samples have shown increased spontaneous apoptosis as well as higher levels of apoptosis mediated by TNF and CD95 pathways (301), and overexpression of Müllerian-inhibiting substance in ovarian granulosa cells of TS may also be involved (302). Due to this loss of ovarian function, lifelong deficiency of estrogen and progestin often prevails in TS, with a mere 2–5% being able to achieve unassisted pregnancy (147, 153). Hence, estrogen replacement therapy is necessary for feminization, achievement and maintenance of normal bone mineral density, and stimulation of neurocognitive development (2). Initial concerns about early introduction of estrogen as a cause of growth plate closure and reduced final height are being rejected in TS. For final height, impressive effects are shown with recombinant human GH despite early commencement of estrogen treatment (280, 303). After introduction, estrogen

replacement therapy should continue throughout adulthood, aiming for a total exposure time comparable to that in normal sex hormone synthesis (2).

Endogenous estrogen synthesis by the ovaries is crucial to cardiovascular health, as appreciated by the adverse prognostic impact of declining ovarian function in the normal menopause and premature ovarian failure (304, 305). Estrogen exerts direct cardiovascular effects on nuclear receptors of vascular smooth muscle cells, endothelial cells, and cardiomyocytes (194). Additional indirect effects are also important. Endogenous estrogen regulates hepatic synthesis of blood clotting factors and lipoproteins and promotes loss of visceral fat and increases insulin sensitivity (194). At the same time, estrogen down-regulates the immune system (194) and antagonizes the renin-angiotensin-aldosterone system (241). The precise mechanisms of action are in many instances unknown, but overall the effects of endogenous estrogen appear to be advantageous.

There may be complex and individualized influences of estrogen replacement therapy in different states of estrogen deficiency (306). The cardiovascular benefits are thought to inversely relate to the duration of estrogen deficiency before estrogen replacement therapy introduction (194). Exogenous estrogen is conceptualized as a pleiotropic hormone that may both improve and aggravate cardiovascular prognosis, depending on the atherosclerotic substrate of the individual and with timing of treatment being crucial (306). Therefore, lessens of estrogen delivery in TS should only be learned from the study of states with similar states of pronounced and early-life estrogen deficiency, rather than from cohorts with late-life deficiency after decades of normal exposure (postmenopausal women) or even normal levels (the contraceptive setting).

The effect of estrogen replacement therapy on hard cardiovascular endpoints has not been elucidated in TS. Short-term studies of intermediate risk markers suggest a beneficial impact of estrogen replacement therapy during the second to fourth decades of life in TS (Table 5). Overall, findings are congruent with known effects of estrogen on cardiovascular cells, causing acute nitrogen monoxide release and up-regulation of nitrogen monoxide synthase expression in the endothelium, which in turn relaxes vascular smooth muscle cells and has antiinflammatory effects (194). Estrogen also appears to beneficially influence the extracellular matrix synthesis and the vascular smooth muscle cell proliferation (194). For these reasons, estrogen replacement therapy in appropriate dosages could protect against atherosclerosis in TS, and may even antagonize a tentative up-regulation of the TGF-$\beta$1–vascular smooth muscle cell–matrix metalloproteinase axis in the aortic wall. However, estrogen replacement therapy may also promote a procoagulant state and contribute to an apparently increased risk of venous thrombus formation in TS. This adverse effect is seen in the contraceptive and postmenopausal setting (307, 308), and oral estrogen has been proposed to cause a procoagulant state secondary to hepatocytes that respond to high estrogen levels through increased protein synthesis (194). This may theoretically be avoided using transdermal or sc administration (194). In TS, findings of exogenous estrogen as an up-regulator of the clotting system are equivocal. Some studies report stationary levels of clotting factors (176, 219, 309), whereas others find increased fibrinogen levels during estrogen treatment (220). Although the transdermal route may offer a more physiological profile (310), route of administration did not have any influence on other cardiovascular risk markers in TS (234). Beyond administration route, the progestin component of sex hormone replacement therapy may also be of relevance to the cardiovascular effects; some progestins activate androgen and mineralocorticoid receptors (311) and potentially oppose advantageous effects of estrogens.

Metabolic changes during estrogen replacement therapy may impact cardiovascular disease. Estrogen delivery in TS has been inferred to improve metabolism with decreased visceral adipose tissue (174), increased high-density lipoprotein cholesterol (176, 234, 309), and improved levels of circulating adipokines (312). However, findings during correction of the premature ovarian failure in TS are ambiguous. Glucose tolerance may be reduced by estrogen replacement therapy during the short term, with no change in insulin sensitivity (234). Speculatively, delivery of estrogen during the longer term will improve glucose homeostasis and metabolism in TS.

### C. Androgens

Sex steroid derangement in TS includes androgen deficiency (313–315). Androgen replacement therapy in TS is controversial and far from routine, and any association between correction of androgen deficiency and cardiovascular disease is unresolved (316). Delivery of androgens in other states of female androgen deficiency benefits intermediate cardiovascular markers (317). Oxandrolone, a nonaromatizable (to estradiol) derivative of testosterone, can be used as an accelerating adjuvant to recombinant human GH in pronounced growth failure or late introduction of GH treatment in TS (2). Cardiovascular effects of oxandrolone also remain controversial, and there are concerns that delivery may worsen any adverse impact of recombinant human GH on glucose metabolism in addition to deferring feminization (318–321).

**D. Diabetes and metabolism**

Both type 1 (RR, 11.6) and type 2 (RR, 4.4) diabetes occur with increased frequency in TS and directly aggravate prognosis (1, 3, 4, 10). Glucose tolerance is impaired in 25–78% of adult TS when assessed by an oral glucose tolerance test (234, 322), with higher glucose levels, whereas insulin levels are comparable (323), higher (322), or even lower (324) in comparison with controls. The insulin response has been described as delayed compared with healthy controls (227, 322). Fasting levels of glucose are generally comparable to controls (234, 325), whereas insulin levels have been reported as increased in some cohorts (323, 325) but not in other cohorts (234, 322). Using the hyperinsulinemic euglycemic clamp, some early studies indicated increased insulin resistance in adolescents with TS (243, 325). However, these early reports are challenged by more recent studies that show no such difference when compared with controls that are meticulously matched for age and body composition (189, 227).

The cause for diabetes in TS is unexplained. Insulin release after an iv glucose tolerance test has been shown to be reduced, especially at the early time-points (first-phase insulin response) (189). This infers that impaired β-cell function, rather than decreased insulin sensitivity, could be the foundation of impaired glucose tolerance in TS (244). Reduced gastric inhibitory polypeptide in young girls with TS who had impaired glucose tolerance, compared with age-matched girls with TS with normal glucose tolerance, suggests that an imbalance in incretin–β-cell cross talk might contribute to disturbances in insulin secretion (326). Moreover, insulin sensitivity was normal in young females with TS compared with controls closely matched for age and body mass index, whereas discrete perturbations in glucose handling were present (189). In the same study, blood glucose levels were increased during oral and iv glucose tolerance test, with a poor compensatory increase in insulin and a reduced insulin-to-glucose ratio (189).

Haploinsufficiency of genes on Xp increases the risk of type 2 diabetes to 18–23%, and haploinsufficiency of Xp combined with trisomy for Xq genes (karyotypes with isochromosome Xq) further increases this risk (327). This was shown in a large cohort of TS females with a mean age of 35 yr and 25% of these TS females had type 2 diabetes (327), where the distribution was:

- del Xq: type 2 diabetes rate of 9% (compares with the general population)
- 45,X: type 2 diabetes rate of 18%
- del Xp: type 2 diabetes rate of 23%
- Isochromosome Xq: type 2 diabetes rate of 43%

The discovery of specific causative genes on the X-chromosome is likely to follow. Gene array data suggest that overexpression of Xq transcription factors is involved in altered pancreatic islet and β-cell function, as well as in proinflammatory action in TS, which was mirrored by increased levels of circulating C-reactive protein, IGF-II, and anti-glutamic acid decarboxylase 65 antibodies (327). Clinical studies have documented raised C-reactive protein (326, 328) as well as glutamic acid decarboxylase 65 antibodies (166), especially linked to isochromosome Xq (166). Such data infer a link between autoimmunity and diabetes in TS. Collectively, oral and iv glucose tolerance tests and clamp studies suggest that a number of factors might interact and in combination explain the perturbed β-cell function.

With premature ovarian failure affecting most adult females with TS, it is essential to evaluate the impact of estrogen replacement on glucose metabolism. Glucose tolerance assessed by oral glucose tolerance test was impaired in 78% after 6 months of estrogen replacement therapy compared with 50% before such treatment (234). However, free fat mass and physical fitness increased during exogenous estrogen treatment, lending promise of further improvements in glucose metabolism with extended treatment duration. A meta-analysis evaluated the effect of estrogen replacement therapy in the postmenopausal setting and found that muscle strength was increased during active treatment (329), as has also been indicated in TS (234, 330). Route of administration of estrogen replacement therapy (oral or transdermal) does not seem to influence glucose metabolism in TS (234, 240, 310). Incorporating data from conditions other than TS, which also include young women with surgically induced menopause, it seems that 17-β-estradiol in the normal range is associated with beneficial effects on insulin sensitivity, whereas both a hypo- and hyperestrogenic milieu is associated with insulin resistance (331, 332). Indeed, 17-β-estradiol and its α and β receptors have profound beneficial effects on energy homeostasis, skeletal muscle, adipose tissue, liver, pancreas (β-cells), and the cardiovascular system (333).

Increased liver enzymes are common in TS (334), and cirrhosis is 5-fold more common (3). Deficiency of sex hormones, especially 17-β-estradiol, seems to play an essential role in the hepatic abnormalities because treatment with estrogen replacement therapy reduces or even normalizes the liver enzymes (276, 335). The liver primarily expresses estrogen receptor α, and to some extent estrogen receptor β also, and recent evidence shows that stimulation of estrogen receptor α protects against inflammation and hypercholesterolemia, and that it is essential for normal glucose homeostasis (333). The histopathology of liver biopsies in females with TS who had persistently el-

evated liver enzymes showed a range of diseases, which included cirrhosis associated with obliterative portal venopathy, multiple focal nodular hyperplasia, and nodular regenerative hyperplasia (336). Less severe changes were also present, such as portal fibrosis, inflammatory infiltrates, and nonalcoholic fatty liver disease. The overall conclusion was that the principal causative mechanisms were congenital vascular disorders and nonalcoholic fatty liver disease (336). 17-$\beta$-Estradiol directly mediates transcriptional regulation of vascular endothelial growth factor through estrogen receptor $\alpha$ and $\beta$, activating receptors 1 and 2 for this growth factor on hepatic sinusoidal endothelial cells (337). This leads to paracrine release of growth and survival factors protecting hepatocytes from toxins and promoting hepatocyte proliferation. In this way, a lack of estrogen could lead to accelerated hepatocyte apoptosis and leakage of liver enzymes and vice versa (337). In a recent study, an abnormality of hepatic lipid storage was suggested because females with TS with high cholesterol and high body mass index had higher $\gamma$-glutamyl transferase, and magnetic resonance imaging confirmed intra-hepatocellular lipid deposition in TS (338). With the available evidence in mind, it can be concluded that estrogen replacement has a prominent role in maintaining normal liver metabolism in TS.

### E. Autoimmunity

Autoimmunity is increased in TS (165, 166), resulting not only in type 1 diabetes but also thyroiditis (RR, 16.6) (3). Thyroid autoimmune disease principally manifests as hypothyroidism, which increases in prevalence with age to affect 23–34% of adolescents and adults with TS (166, 339). No links have been made between hypothyroidism and cardiovascular morbidity in TS. However, inadequate diagnosis and suboptimal substitution may aggravate the prevailing metabolic and cardiovascular phenotype (340).

Haploinsuffiency of X-chromosome material has been proposed as a possible primary cause for autoimmunity (335), although lack of estrogen and up-regulation of proinflammatory cytokines (245) as well as accelerated apoptosis of T-cell subsets may also be involved (301). Estrogen has numerous effects on the immune system encompassing modulation of T cells (341) and inhibition of inflammation (342), although studies in humans and animal models are inconclusive. From periovulatory to pregnancy levels, 17-$\beta$-estradiol has been shown to have a stimulatory effect on IL-4, IL-10, and interferon $\alpha$, but it inhibits TNF from CD4+ T cells, indicating a shift toward down-regulation of T-cell autoimmunity (342). At the same levels, 17-$\beta$-estradiol has been shown to stimulate antibody secretion by CD5+ B cells while suppressing bone marrow B-cell lineage precursors (342). The exact role of estrogen in autoimmunity in TS is unknown. Estrogen could induce an increased B-cell autoimmunity, but at the same time decrease T-cell autoimmunity (342). Other causes for the breakdown of self-tolerance such as fetal microchimerism, skewing of X-chromosome inactivation, gene duplication, or up-regulation of proinflammatory cytokines are being investigated. Androgen predominantly exerts an inhibitory effect on the immune system (342) and could therefore also be implicated through the relative androgen insufficient state in TS (314).

## VI. Future Perspectives

*SHOX* haploinsufficiency is currently the only genetic mechanism that explains part of the genotype and phenotype of TS (343). Here, we have presented a host of other putative mechanisms, which may or may not play a role in cardiovascular disease among TS patients. For many years, the pathogenesis of TS has eluded researchers, and the discovery of *SHOX* gene and its role in TS has naturally led researchers to believe that another gene or several genes play similar roles to partially or completely explain the spectrum of phenotypes. Such genes could be situated in one of the two PAR of the X-chromosome, although genes outside of this area may also play a role in the pathogenesis of TS. Interestingly, pseudoautosomal genes such as *ASMTL* and *PPP2R3B* in experimental models were expressed at lower levels than in normal cells (53). Because *ASMTL* is a methyltransferase, it can be speculated that improper methylation of the genome plays a role in TS. At present, the involvement of *CSF2RA* points toward insufficient placentation as the most plausible explanation for the high fetal lethality in TS (53, 344). However, these findings need to be confirmed in additional studies before any firm conclusions can be drawn. Likewise, the process and consequences of X-chromosome inactivation (or the lack hereof) also need to be studied in further detail. Another perspective comes from recent scientific advances indicating that sexual dimorphism between males and females plays a role in transcriptional regulation of autosomal genes and that this leads to differential expression between males and females of more than half of the coding genome (44). This must be studied in further detail, and the exact role of sexual dimorphism and the possible changes inflicted by the lack of one X-chromosome in TS need to be explored. Suffice it to say, the genetic mechanisms behind TS are far from understood, and the interaction between different genetic mechanisms may even complicate matters further. TS may actually be the result of deregulation within many different pathways that work

in concert to cause deviation from normality. Thus, linking genetics with phenotypic traits should prove extremely interesting in future studies, and we may very well have to revise our understanding of the genetics behind TS in the coming years. At present, we can probably divide the phenotypic traits of TS into (Fig. 9): 1) congenital cardiovascular malformations of unresolved pathogeneses; 2) decreased intrauterine viability, where haploinsufficiency for X-linked pseudoautosomal genes operating in the placenta has been suggested to be involved (*CSF2RA* and possibly other genes); 3) ovarian dysgenesis, where many genes have been implicated, but none proven; 4) predilection to autoimmune disease of unknown geneses; and 5) altered brain development, especially social-cognitive development, which is altered in many cases, often in a more "male-like" direction.

Today, we can conclude that females with TS suffer from a condition with a high risk of congenital and acquired heart diseases, diabetes, metabolic disorders, and autoimmune disease (Fig. 9), which necessitates multidisciplinary care with involvement of many specialties (2). In many centers (6, 155), guidelines for multidisciplinary practice have been implemented, but clinical care has remained suboptimal elsewhere (5). The prevalence of associated diseases has been well detailed, but we lack a clear understanding of the pathophysiological mechanisms, including details on the genetic and molecular events that take place and lead to the diverse phenotypes. Life expectancy is decreased (11), and conditions related to the heart and great vessels are involved in about half of all excess deaths (4). Mounting evidence supports the notion that the specific X-chromosomal deficit and the tissue affected

**Figure 9.**

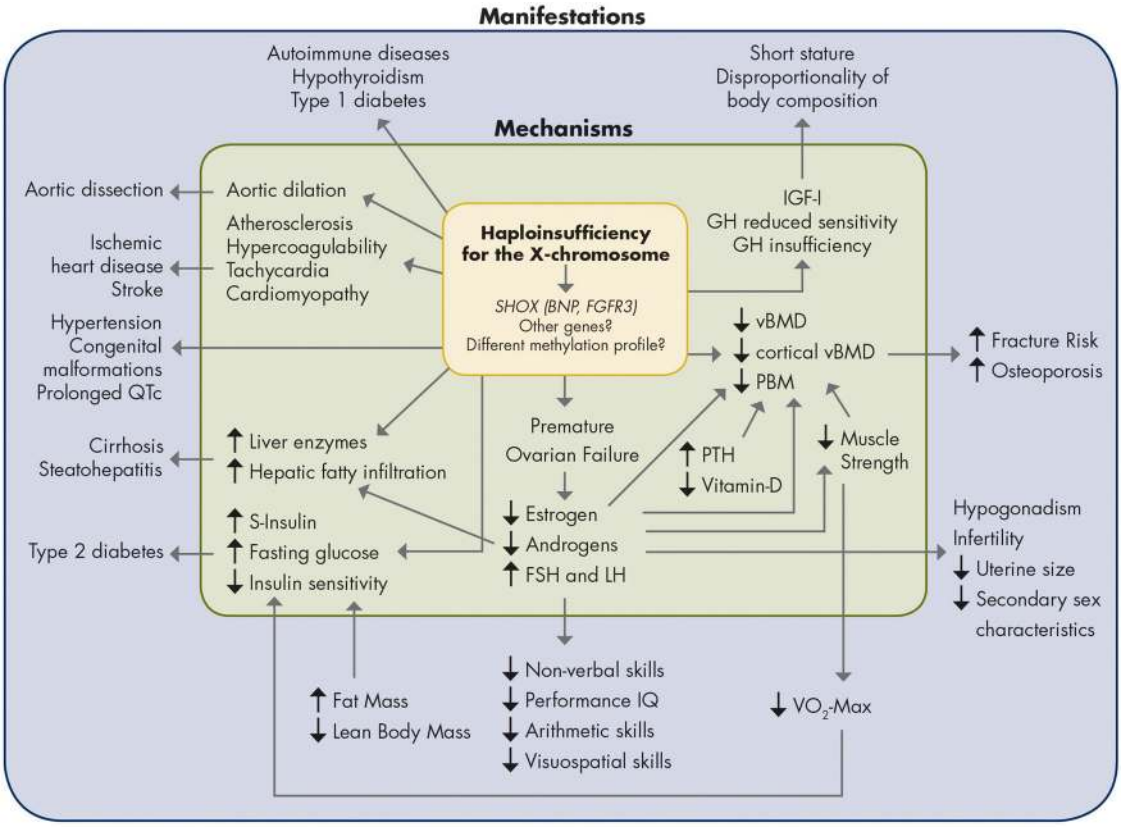


Figure 9. The serious effect of haploinsufficiency of genes on the X-chromosome (and possibly other genetic mechanisms working in concert) is at the center of the current understanding of pathogenesis in TS. This leads to a range of effects for the cardiovascular system (congenital and acquired diseases), the endocrine system (especially with premature ovarian failure and thus female hypogonadism), body composition, as well as the brain and other organs. Hypogonadism has pervasive effects, affecting: 1) different hormone levels; 2) cardiovascular features; 3) metabolic features; and 4) features related to sex hormones, such as infertility. In addition, mounting evidence suggests that hypogonadism in TS leads either directly or indirectly to a reduced quality of life. Haploinsufficiency of genes on the X-chromosome has been implicated in the presence of an increased risk of congenital malformations, although no specific genes have been identified so far. *Arrows* indicate possible consequences—not all interactions have been shown in scientific studies. $VO_2$max, Maximum capacity to transport and utilize oxygen during incremental exercise; BNP, brain natriuretic peptide; FGFR3, fibroblast growth factor receptor 3; BMD, bone mineral density; PBM, peak bone mineral mass; IQ, intelligence quotient.

by this karyotype will determine the pathology encountered by the affected girl or woman. Although TS is not primarily considered a syndrome of premature aging (345), a number of characteristics point to some form of accelerated process—for instance, premature occurrence of aortic dissection, stroke, diabetes, autoimmune disease, and sensorineural deafness similar to presbycusis (346). So with these aspects in mind, a more thorough understanding of the total task that females with TS pose to the clinician is expected to evolve, facilitating better clinical care and more targeted development of new medications and translation from basic research to future treatments.

## VII. Conclusion

Females with TS suffer a multifaceted syndrome that manifests within the realm of several organ systems, whereof many especially cardiovascular components are only just being unraveled (Figs. 4 and 9). There is ample opportunity for progress because our understanding of the causation behind the plethora of associated cardiovascular traits remains patchy. An in-depth description of the genetic mechanisms leading to the advent of this X-chromosomal disorder should facilitate fundamental changes in diagnosis, treatment, and monitoring. Treatment of TS comes at a high cost for society, with an estimated 78,000 females affected in the United States and another 125,500 females affected in the European Union. If we can pinpoint new genetic mechanisms leading to the abundance of clinical manifestations and traits related to TS, we will have provided the important framework for development of new and targeted therapy.

## Acknowledgments

Address all correspondence and requests for reprints to: Dr. Claus H. Gravholt, M.D., Ph.D., Department of Endocrinology and Internal Medicine (MEA), Aarhus University Hospital, Norrebrogade 44, DK-8000 Aarhus C, Denmark. E-mail: ch.gravholt@dadlnet.dk.

This work was supported by the Danish Medical Research Council (Grant 22-01-0395), the Danish Agency for Science Technology and Innovation (Grant 271-07-0333), and an unrestricted grant from Novo Nordisk (all to C.H.G.). Further supporting grants were from the Danish Heart Foundation, Korning Fonden, Hede Nielsens Fond, Eva og Henry Fraenkels Minde Fond, and Snedkermester Sophus Jacobsen and Hustru Astrid Jacobsens Fond (all to K.H.M.).

Disclosure Summary: The authors have nothing to disclose.

## References

1. **Stochholm K, Juul S, Juel K, Naeraa RW, Gravholt CH** 2006 Prevalence, incidence, diagnostic delay, and mortality in Turner syndrome. J Clin Endocrinol Metab 91:3897–3902
2. **Bondy CA** 2007 Care of girls and women with Turner syndrome: a guideline of the Turner Syndrome Study Group. J Clin Endocrinol Metab 92:10–25
3. **Gravholt CH, Juul S, Naeraa RW, Hansen J** 1998 Morbidity in Turner syndrome. J Clin Epidemiol 51:147–158
4. **Schoemaker MJ, Swerdlow AJ, Higgins CD, Wright AF, Jacobs PA** 2008 Mortality in women with Turner syndrome in Great Britain: a national cohort study. J Clin Endocrinol Metab 93:4735–4742
5. **Bondy C, Bakalov VK, Lange ED, Ceniceros I** 2006 Deficient medical care for adults with the Turner syndrome. Ann Intern Med 145:866–867
6. **Freriks K, Timmermans J, Beerendonk CC, Verhaak CM, Netea-Maier RT, Otten BJ, Braat DD, Smeets DF, Kunst DH, Hermus AR, Timmers HJ** 2011 Standardized multidisciplinary evaluation yields significant previously undiagnosed morbidity in adult women with Turner syndrome. J Clin Endocrinol Metab 96:E1517–E1526
7. **Saenger P, Wikland KA, Conway GS, Davenport M, Gravholt CH, Hintz R, Hovatta O, Hultcrantz M, Landin-Wilhelmsen K, Lin A, Lippe B, Pasquino AM, Ranke MB, Rosenfeld R, Silberbach M** 2001 Recommendations for the diagnosis and management of Turner syndrome. J Clin Endocrinol Metab 86:3061–3069
8. **Gravholt CH** 2004 Epidemiological, endocrine and metabolic features in Turner syndrome. Eur J Endocrinol 151: 657–687
9. **Lopez L, Arheart KL, Colan SD, Stein NS, Lopez-Mitnik G, Lin AE, Reller MD, Ventura R, Silberbach M** 2008 Turner syndrome is an independent risk factor for aortic dilation in the young. Pediatrics 121:e1622–e1627
10. **Swerdlow AJ, Schoemaker MJ, Higgins CD, Wright AF, Jacobs PA** 2005 Mortality and cancer incidence in women with extra X chromosomes: a cohort study in Britain. Hum Genet 118:255–260
11. **Price WH, Clayton JF, Collyer S, De Mey R, Wilson J** 1986 Mortality ratios, life expectancy, and causes of death in patients with Turner's syndrome. J Epidemiol Community Health 40:97–102
12. **Mazzanti L, Cacciari E** 1998 Congenital heart disease in patients with Turner's syndrome. Italian Study Group for Turner Syndrome (ISGTS). J Pediatr 133:688–692
13. **Ho VB, Bakalov VK, Cooley M, Van PL, Hood MN, Burklow TR, Bondy CA** 2004 Major vascular anomalies in Turner syndrome: prevalence and magnetic resonance angiographic features. Circulation 110:1694–1700
14. **Elsheikh M, Wass JA, Conway GS** 2001 Autoimmune thyroid syndrome in women with Turner's syndrome—the association with karyotype. Clin Endocrinol (Oxf) 55: 223–226
15. **Hanson L, Bryman I, Barrenäs ML, Janson PO, Wahlström J, Albertsson-Wikland K, Hanson C** 2001 Genetic analysis of mosaicism in 53 women with Turner syndrome. Hereditas 134:153–159
16. **Gravholt CH, Juul S, Naeraa RW, Hansen J** 1996 Prenatal and postnatal prevalence of Turner's syndrome: a registry study. BMJ 312:16–21
17. **El-Mansoury M, Barrenäs ML, Bryman I, Hanson C, Larsson C, Wilhelmsen L, Landin-Wilhelmsen K** 2007

Chromosomal mosaicism mitigates stigmata and cardiovascular risk factors in Turner syndrome. Clin Endocrinol (Oxf) 66:744–751
18. **Hagman A, Wennerholm UB, Källén K, Barrenäs ML, Landin-Wilhelmsen K, Hanson C, Bryman I** 2010 Women who gave birth to girls with Turner syndrome: maternal and neonatal characteristics. Hum Reprod 25:1553–1560
19. **Warburton D, Kline J, Stein Z, Susser M** 1980 Monosomy X: a chromosomal anomaly associated with young maternal age. Lancet 1:167–169
20. **Leichtman DA, Schmickel RD, Gelehrter TD, Judd WJ, Woodbury MC, Meilinger KL** 1978 Familial Turner syndrome. Ann Intern Med 89:473–476
21. **Larizza D, Danesino C, Maraschio P, Caramagna C, Klersy C, Calcaterra V** 2011 Familial occurrence of Turner syndrome: casual event or increased risk? J Pediatr Endocrinol Metab 24:223–225
22. **Sagi L, Zuckerman-Levin N, Gawlik A, Ghizzoni L, Buyukgebiz A, Rakover Y, Bistritzer T, Admoni O, Vottero A, Baruch O, Fares F, Malecka-Tendera E, Hochberg Z** 2007 Clinical significance of the parental origin of the X chromosome in Turner syndrome. J Clin Endocrinol Metab 92:846–852
23. **Stochholm K, Juul S, Gravholt CH** 2010 Mortality and incidence in women with 47,XXX and variants. Am J Med Genet A 152A:367–372
24. **Stochholm K, Juul S, Gravholt CH** 2010 Diagnosis and mortality in 47,XYY persons: a registry study. Orphanet J Rare Dis 5:15
25. **Held KR, Kerber S, Kaminsky E, Singh S, Goetz P, Seemanova E, Goedde HW** 1992 Mosaicism in 45,X Turner syndrome: does survival in early pregnancy depend on the presence of two sex chromosomes? Hum Genet 88:288–294
26. **Fernández R, Méndez J, Pásaro E** 1996 Turner syndrome: a study of chromosomal mosaicism. Hum Genet 98:29–35
27. **Hassold TJ** 1986 Chromosome abnormalities in human reproductive wastage. Trends Genet 2:105–110
28. **Ross MT, Grafham DV, Coffey AJ, Scherer S, McLay K, Muzny D, Platzer M, Howell GR, Burrows C, Bird CP, Frankish A, Lovell FL, Howe KL, Ashurst JL, Fulton RS, Sudbrak R, Wen G, Jones MC, Hurles ME, Andrews TD, Scott CE, Searle S, Ramser J, Whittaker A, Deadman R, Carter NP, Hunt SE, Chen R, Cree A, Gunaratne P, Havlak P, Hodgson A, Metzker ML, Richards S, Scott G, Steffen D, Sodergren E, Wheeler DA, Worley KC, Ainscough R, Ambrose KD, Ansari-Lari MA, *et al.*** 2005 The DNA sequence of the human X chromosome. Nature 434:325–337
29. **Lahn BT, Page DC** 1999 Four evolutionary strata on the human X chromosome. Science 286:964–967
30. **Yang C, Chapman AG, Kelsey AD, Minks J, Cotton AM, Brown CJ** 2011 X-Chromosome inactivation: molecular mechanisms from the human perspective. Hum Genet 130:175–185
31. **Lyon MF** 1961 Gene action in the X-chromosome of the mouse (*Mus musculus L.*). Nature 190:372–373
32. **Brown CJ, Lafreniere RG, Powers VE, Sebastio G, Ballabio A, Pettigrew AL, Ledbetter DH, Levy E, Craig IW, Willard HF** 1991 Localization of the X inactivation centre on the human X chromosome in Xq13. Nature 349:82–84
33. **Brown CJ, Ballabio A, Rupert JL, Lafreniere RG, Grompe M, Tonlorenzi R, Willard HF** 1991 A gene from the region of the human X inactivation centre is expressed exclusively from the inactive X chromosome. Nature 349:38–44
34. **Jeppesen P, Turner BM** 1993 The inactive X chromosome in female mammals is distinguished by a lack of histone H4 acetylation, a cytogenetic marker for gene expression. Cell 74:281–289
35. **Lee JT, Davidow LS, Warshawsky D** 1999 Tsix, a gene antisense to Xist at the X-inactivation centre. Nat Genet 21:400–404
36. **Heard E, Clerc P, Avner P** 1997 X-chromosome inactivation in mammals. Annu Rev Genet 31:571–610
37. **Lyon MF** 2002 X-chromosome inactivation and human genetic disease. Acta Paediatr Suppl 91:107–112
38. **Anderson CL, Brown CJ** 1999 Polymorphic X-chromosome inactivation of the human TIMP1 gene. Am J Hum Genet 65:699–708
39. **Wareham KA, Lyon MF, Glenister PH, Williams ED** 1987 Age related reactivation of an X-linked gene. Nature 327:725–727
40. **Carrel L, Willard HF** 2005 X-inactivation profile reveals extensive variability in X-linked gene expression in females. Nature 434:400–404
41. **Disteche CM, Filippova GN, Tsuchiya KD** 2002 Escape from X inactivation. Cytogenet Genome Res 99:36–43
42. **Matura LA, Ho VB, Rosing DR, Bondy CA** 2007 Aortic dilatation and dissection in Turner syndrome. Circulation 116:1663–1670
43. **Ober C, Loisel DA, Gilad Y** 2008 Sex-specific genetic architecture of human disease. Nat Rev Genet 9:911–922
44. **Wijchers PJ, Yandim C, Panousopoulou E, Ahmad M, Harker N, Saveliev A, Burgoyne PS, Festenstein R** 2010 Sexual dimorphism in mammalian autosomal gene regulation is determined not only by Sry but by sex chromosome complement as well. Dev Cell 19:477–484
45. **Xu J, Deng X, Disteche CM** 2008 Sex-specific expression of the X-linked histone demethylase gene Jarid1c in brain. PLoS One 3:e2553
46. **Xu J, Deng X, Watkins R, Disteche CM** 2008 Sex-specific differences in expression of histone demethylases Utx and Uty in mouse brain and neurons. J Neurosci 28:4521–4527
47. **Rao E, Weiss B, Fukami M, Rump A, Niesler B, Mertz A, Muroya K, Binder G, Kirsch S, Winkelmann M, Nordsiek G, Heinrich U, Breuning MH, Ranke MB, Rosenthal A, Ogata T, Rappold GA** 1997 Pseudoautosomal deletions encompassing a novel homeobox gene cause growth failure in idiopathic short stature and Turner syndrome. Nat Genet 16:54–63
48. **Marchini A, Häcker B, Marttila T, Hesse V, Emons J, Weiss B, Karperien M, Rappold G** 2007 BNP is a transcriptional target of the short stature homeobox gene SHOX. Hum Mol Genet 16:3081–3087
49. **Gravholt CH, Hansen KW, Erlandsen M, Ebbehøj E, Christiansen JS** 2006 Nocturnal hypertension and impaired sympathovagal tone in Turner syndrome. J Hypertens 24:353–360
50. **Kosho T, Muroya K, Nagai T, Fujimoto M, Yokoya S, Sakamoto H, Hirano T, Terasaki H, Ohashi H, Nishimura G, Sato S, Matsuo N, Ogata T** 1999 Skeletal features and growth patterns in 14 patients with haploinsufficiency of

SHOX: implications for the development of Turner syndrome. J Clin Endocrinol Metab 84:4613–4621
51. **Binder G** 2011 Short stature due to SHOX deficiency: genotype, phenotype, and therapy. Horm Res Paediatr 75: 81–89
52. **Sachdev V, Matura LA, Sidenko S, Ho VB, Arai AE, Rosing DR, Bondy CA** 2008 Aortic valve disease in Turner syndrome. J Am Coll Cardiol 51:1904–1909
53. **Li W, Wang X, Fan W, Zhao P, Chan YC, Chen S, Zhang S, Guo X, Zhang Y, Li Y, Cai J, Qin D, Li X, Yang J, Peng T, Zychlinski D, Hoffmann D, Zhang R, Deng K, Ng KM, Menten B, Zhong M, Wu J, Li Z, Chen Y, Schambach A, Tse HF, Pei D, Esteban MA** 2012 Modeling abnormal early development with induced pluripotent stem cells from aneuploid syndromes. Hum Mol Genet 21:32–45
54. **Urbach A, Benvenisty N** 2009 Studying early lethality of 45,XO (Turner's syndrome) embryos using human embryonic stem cells. PLoS One 4:e4175
55. **Bondy CA, Ceniceros I, Van PL, Bakalov VK, Rosing DR** 2006 Prolonged rate-corrected QT interval and other electrocardiogram abnormalities in girls with Turner syndrome. Pediatrics 118:e1220–e1225
56. **Boucher CA, Sargent CA, Ogata T, Affara NA** 2001 Breakpoint analysis of Turner patients with partial Xp deletions: implications for the lymphoedema gene location. J Med Genet 38:591–598
57. **Brandenburg H, Steegers EA, Gittenberger-de Groot AC** 2005 Potential involvement of vascular endothelial growth factor in pathophysiology of Turner syndrome. Med Hypotheses 65:300–304
58. **Adachi M, Tachibana K, Asakura Y, Muroya K, Ogata T** 2000 Del(X)(p21.1) in a mother and two daughters: genotype-phenotype correlation of Turner features. Hum Genet 106:306–310
59. **Cattanach BM, Kirk M** 1985 Differential activity of maternally and paternally derived chromosome regions in mice. Nature 315:496–498
60. **Nicholls RD, Knoll JH, Butler MG, Karam S, Lalande M** 1989 Genetic imprinting suggested by maternal heterodisomy in nondeletion Prader-Willi syndrome. Nature 342: 281–285
61. **Ko JM, Kim JM, Kim GH, Lee BH, Yoo HW** 2010 Influence of parental origin of the X chromosome on physical phenotypes and GH responsiveness of patients with Turner syndrome. Clin Endocrinol (Oxf) 73:66–71
62. **Van PL, Bakalov VK, Zinn AR, Bondy CA** 2006 Maternal X chromosome, visceral adiposity, and lipid profile. JAMA 295:1373–1374
63. **Chu CE, Donaldson MD, Kelnar CJ, Smail PJ, Greene SA, Paterson WF, Connor JM** 1994 Possible role of imprinting in the Turner phenotype. J Med Genet 31:840–842
64. **Cutter WJ, Daly EM, Robertson DM, Chitnis XA, van Amelsvoort TA, Simmons A, Ng VW, Williams BS, Shaw P, Conway GS, Skuse DH, Collier DA, Craig M, Murphy DG** 2006 Influence of X chromosome and hormones on human brain development: a magnetic resonance imaging and proton magnetic resonance spectroscopy study of Turner syndrome. Biol Psychiatry 59:273–283
65. **Skuse DH, James RS, Bishop DV, Coppin B, Dalton P, Aamodt-Leeper G, Bacarese-Hamilton M, Creswell C, McGurk R, Jacobs PA** 1997 Evidence from Turner's syndrome of an imprinted X-linked locus affecting cognitive function. Nature 387:705–708
66. **Bondy CA, Matura LA, Wooten N, Troendle J, Zinn AR, Bakalov VK** 2007 The physical phenotype of girls and women with Turner syndrome is not X-imprinted. Hum Genet 121:469–474
67. **Mortensen KH, Hjerrild BE, Andersen NH, Sørensen KE, Hørlyck A, Pedersen EM, Lundorf E, Christiansen JS, Gravholt CH** 2010 Abnormalities of the major intrathoracic arteries in Turner syndrome as revealed by magnetic resonance imaging. Cardiol Young 20:191–200
68. **Prandstraller D, Mazzanti L, Picchio FM, Magnani C, Bergamaschi R, Perri A, Tsingos E, Cacciari E** 1999 Turner's syndrome: cardiologic profile according to the different chromosomal patterns and long-term clinical follow-up of 136 nonpreselected patients. Pediatr Cardiol 20:108–112
69. **Völkl TM, Degenhardt K, Koch A, Simm D, Dörr HG, Singer H** 2005 Cardiovascular anomalies in children and young adults with Ullrich-Turner syndrome—the Erlangen experience. Clin Cardiol 28:88–92
70. **Andersen NH, Hjerrild BE, Sørensen K, Pedersen EM, Stochholm K, Gormsen LC, Hørlyck A, Christiansen JS, Gravholt CH** 2006 Subclinical left ventricular dysfunction in normotensive women with Turner's syndrome. Heart 92:1516–1517
71. **Surerus E, Huggon IC, Allan LD** 2003 Turner's syndrome in fetal life. Ultrasound Obstet Gynecol 22:264–267
72. **Gøtzsche CO, Krag-Olsen B, Nielsen J, Sørensen KE, Kristensen BO** 1994 Prevalence of cardiovascular malformations and association with karyotypes in Turner's syndrome. Arch Dis Child 71:433–436
73. **Siu SC, Silversides CK** 2010 Bicuspid aortic valve disease. J Am Coll Cardiol 55:2789–2800
74. **Kutay V, Yakut C** 2005 Absence of right superior vena cava and aortic annular hypoplasia in a patient with Turner's syndrome. J Card Surg 20:375–376
75. **Ostberg JE, Brookes JA, McCarthy C, Halcox J, Conway GS** 2004 A comparison of echocardiography and magnetic resonance imaging in cardiovascular screening of adults with Turner syndrome. J Clin Endocrinol Metab 89:5966–5971
76. **Schaefer BM, Lewin MB, Stout KK, Gill E, Prueitt A, Byers PH, Otto CM** 2008 The bicuspid aortic valve: an integrated phenotypic classification of leaflet morphology and aortic root shape. Heart 94:1634–1638
77. **Załuska R, Kazłowski J, Gumkowska A, Pikto-Pietkiewicz W** 2008 Congenital disorders of the cardiovascular system and their complications in a 21-year-old woman with Turner syndrome–a case report. Kardiol Pol 66:63–66
78. **Cleemann L, Mortensen KH, Holm K, Smedegaard H, Skouby SO, Wieslander SB, Leffers AM, Leth-Espensen P, Pedersen EM, Gravholt CH** 2010 Aortic dimensions in girls and young women with Turner syndrome: a magnetic resonance imaging study. Pediatr Cardiol 31:497–504
79. **Hoffman JI, Kaplan S** 2002 The incidence of congenital heart disease. J Am Coll Cardiol 39:1890–1900
80. **Dawson-Falk KL, Wright AM, Bakker B, Pitlick PT, Wilson DM, Rosenfeld RG** 1992 Cardiovascular evaluation in Turner syndrome: utility of MR imaging. Australas Radiol 36:204–209
81. **Hjerrild BE, Mortensen KH, Sørensen KE, Pedersen EM,**

**Andersen NH, Lundorf E, Hansen KW, Hørlyck A, Hager A, Christiansen JS, Gravholt CH** 2010 Thoracic aortopathy in Turner syndrome and the influence of bicuspid aortic valves and blood pressure: a CMR study. J Cardiovasc Magn Reson 12:12

82. **Mutlu M, Dilber E, Aslan Y, Okten A, Oztürk O** 2010 A Turner syndrome case associated with anal atresia, interrupted aortic arch and multicystic dysplastic kidney. Turk J Pediatr 52:215–217
83. **Ravelo HR, Stephenson LW, Friedman S, Chatten J, Rashkind WJ, Vidas M, Edmunds Jr LH** 1980 Coarctation resection in children with Turner's syndrome: a note of caution. J Thorac Cardiovasc Surg 80:427–430
84. **Beder SD, Driscoll DJ, McNamara DG, Cooley DA** 1982 Coarctation repair in children with Turner's syndrome. Tex Heart Inst J 9:49–52
85. **Brandt 3rd B, Heintz SE, Rose EF, Ehrenhaft JL, Clark EB** 1984 Repair of coarctation of the aorta in children with Turner syndrome. Pediatr Cardiol 5:175–177
86. **Zanjani KS, Thanopoulos BD, Peirone A, Alday L, Giannakoulas G** 2010 Usefulness of stenting in aortic coarctation in patients with the Turner syndrome. Am J Cardiol 106:1327–1331
87. **Fejzic Z, van Oort A** 2005 Fatal dissection of the descending aorta after implantation of a stent in a 19-year-old female with Turner's syndrome. Cardiol Young 15:529–531
88. **Badmanaban B, Mole D, Sarsam MA** 2003 Descending aortic dissection post coarctation repair in a patient with Turner's syndrome. J Card Surg 18:153–154
89. **Wu IH, Wu MH, Chen SJ, Wang SS, Chang CI** 2012 Successful deployment of an iliac limb graft to repair acute aortic rupture after balloon aortoplasty of recoarctation in a child with Turner syndrome. Heart Vessels 27:227–230
90. **Sybert VP** 1998 Cardiovascular malformations and complications in Turner syndrome. Pediatrics 101:E11
91. **Oliver JM, Gallego P, Gonzalez A, Aroca A, Bret M, Mesa JM** 2004 Risk factors for aortic complications in adults with coarctation of the aorta. J Am Coll Cardiol 44:1641–1647
92. **Kim HK, Gottliebson W, Hor K, Backeljauw P, Gutmark-Little I, Salisbury SR, Racadio JM, Helton-Skally K, Fleck R** 2011 Cardiovascular anomalies in Turner syndrome: spectrum, prevalence, and cardiac MRI findings in a pediatric and young adult population. AJR Am J Roentgenol 196:454–460
93. **Eiben B, Bartels I, Bähr-Porsch S, Borgmann S, Gatz G, Gellert G, Goebel R, Hammans W, Hentemann M, Osmers R, Rauskolb R, Hansmann I** 1990 Cytogenetic analysis of 750 spontaneous abortions with the direct-preparation method of chorionic villi and its implications for studying genetic causes of pregnancy wastage. Am J Hum Genet 47:656–663
94. **Miyabara S, Nakayama M, Suzumori K, Yonemitsu N, Sugihara H** 1997 Developmental analysis of cardiovascular system of 45,X fetuses with cystic hygroma. Am J Med Genet 68:135–141
95. **Haak MC, Bartelings MM, Gittenberger-De Groot AC, Van Vugt JM** 2002 Cardiac malformations in first-trimester fetuses with increased nuchal translucency: ultrasound diagnosis and postmortem morphology. Ultrasound Obstet Gynecol 20:14–21
96. **Barr Jr M, Oman-Ganes L** 2002 Turner syndrome morphology and morphometrics: cardiac hypoplasia as a cause of midgestation death. Teratology 66:65–72
97. **Reis PM, Punch MR, Bove EL, van de Ven CJ** 1999 Outcome of infants with hypoplastic left heart and Turner syndromes. Obstet Gynecol 93:532–535
98. **Patel A, Hickey E, Mavroudis C, Jacobs JP, Jacobs ML, Backer CL, Gevitz M, Mavroudis CD** 2010 Impact of noncardiac congenital and genetic abnormalities on outcomes in hypoplastic left heart syndrome. Ann Thorac Surg 89:1805–1813; discussion 1813–1814
99. **van Egmond H, Orye E, Praet M, Coppens M, Devloo-Blancquaert A** 1988 Hypoplastic left heart syndrome and 45X karyotype. Br Heart J 60:69–71
100. **Bidot-López P, Matisoff D, Talner NS, Hsia YE** 1978 Hypoplastic left heart in a patient with 45,X/46,XX/47,XXX mosaicism. Am J Med Genet 2:341–343
101. **Loscalzo ML, Van PL, Ho VB, Bakalov VK, Rosing DR, Malone CA, Dietz HC, Bondy CA** 2005 Association between fetal lymphedema and congenital cardiovascular defects in Turner syndrome. Pediatrics 115:732–735
102. **Nicolaides KH** 2011 Screening for fetal aneuploidies at 11 to 13 weeks. Prenat Diagn 31:7–15
103. **Allan LD** 2006 The mystery of nuchal translucency. Cardiol Young 16:11–17
104. **Hyett J, Perdu M, Sharland G, Snijders R, Nicolaides KH** 1999 Using fetal nuchal translucency to screen for major congenital cardiac defects at 10–14 weeks of gestation: population based cohort study. BMJ 318:81–85
105. **Clark EB** 1984 Neck web and congenital heart defects: a pathogenic association in 45 X-O Turner syndrome? Teratology 29:355–361
106. **Hu N, Christensen DA, Agrawal AK, Beaumont C, Clark EB, Hawkins JA** 2009 Dependence of aortic arch morphogenesis on intracardiac blood flow in the left atrial ligated chick embryo. Anat Rec (Hoboken) 292:652–660
107. **Berdahl LD, Wenstrom KD, Hanson JW** 1995 Web neck anomaly and its association with congenital heart disease. Am J Med Genet 56:304–307
108. **van der Putte SC** 1977 Lymphatic malformation in human fetuses. A study of fetuses with Turner's syndrome or status Bonnevie-Ullrich. Virchows Arch A Pathol Anat Histol 376:233–246
109. **Lacro RV, Jones KL, Benirschke K** 1988 Coarctation of the aorta in Turner syndrome: a pathologic study of fetuses with nuchal cystic hygromas, hydrops fetalis, and female genitalia. Pediatrics 81:445–451
110. **Goumans MJ, Liu Z, ten Dijke P** 2009 TGF-$\beta$ signaling in vascular biology and dysfunction. Cell Res 19:116–127
111. **Bondy CA** 2008 Congenital cardiovascular disease in Turner syndrome. Congenit Heart Dis 3:2–15
112. **Gittenberger-de Groot AC, Azhar M, Molin DG** 2006 Transforming growth factor $\beta$-SMAD2 signaling and aortic arch development. Trends Cardiovasc Med 16:1–6
113. **Derynck R, Zhang YE** 2003 Smad-dependent and Smad-independent pathways in TGF-$\beta$ family signalling. Nature 425:577–584
114. **Gomez D, Al Haj Zen A, Borges LF, Philippe M, Gutierrez PS, Jondeau G, Michel JB, Vranckx R** 2009 Syndromic and

non-syndromic aneurysms of the human ascending aorta share activation of the Smad2 pathway. J Pathol 218:131–142

115. **von Kaisenberg CS, Prols F, Nicolaides KH, Maass N, Meinhold-Heerlein I, Brand-Saberi B** 2003 Glycosaminoglycans and proteoglycans in the skin of aneuploid fetuses with increased nuchal translucency. Hum Reprod 18: 2544–2561
116. **Struwe E, Krammer K, Dötsch J, Metzler M, Dörr HG, Cesnjevar R, Rascher W, Koch A** 2006 No evidence for angiotensin type 2 receptor gene polymorphism in intron 1 in patients with coarctation of the aorta and Ullrich-Turner syndrome. Pediatr Cardiol 27:636–639
117. **Carmeliet P, Tessier-Lavigne M** 2005 Common mechanisms of nerve and blood vessel wiring. Nature 436:193–200
118. **Zuckerman-Levin N, Zinder O, Greenberg A, Levin M, Jacob G, Hochberg Z** 2006 Physiological and catecholamine response to sympathetic stimulation in Turner syndrome. Clin Endocrinol (Oxf) 64:410–415
119. **Saharinen P, Tammela T, Karkkainen MJ, Alitalo K** 2004 Lymphatic vasculature: development, molecular regulation and role in tumor metastasis and inflammation. Trends Immunol 25:387–395
120. **Fan G, Tran J** 2011 X chromosome inactivation in human and mouse pluripotent stem cells. Hum Genet 130:217–222
121. **Bondy CA, Van PL, Bakalov VK, Sachdev V, Malone CA, Ho VB, Rosing DR** 2006 Prolongation of the cardiac QTc interval in Turner syndrome. Medicine (Baltimore) 85: 75–81
122. **Nathwani NC, Unwin R, Brook CG, Hindmarsh PC** 2000 Blood pressure and Turner syndrome. Clin Endocrinol (Oxf) 52:363–370
123. **Gravholt CH, Landin-Wilhelmsen K, Stochholm K, Hjerrild BE, Ledet T, Djurhuus CB, Sylvén L, Baandrup U, Kristensen BØ, Christiansen JS** 2006 Clinical and epidemiological description of aortic dissection in Turner's syndrome. Cardiol Young 16:430–436
124. **Olsson C, Thelin S, Ståhle E, Ekbom A, Granath F** 2006 Thoracic aortic aneurysm and dissection: increasing prevalence and improved outcomes reported in a nationwide population-based study of more than 14,000 cases from 1987 to 2002. Circulation 114:2611–2618
125. **Hagan PG, Nienaber CA, Isselbacher EM, Bruckman D, Karavite DJ, Russman PL, Evangelista A, Fattori R, Suzuki T, Oh JK, Moore AG, Malouf JF, Pape LA, Gaca C, Sechtem U, Lenferink S, Deutsch HJ, Diedrichs H, Marcos y Robles J, Llovet A, Gilon D, Das SK, Armstrong WF, Deeb GM, Eagle KA** 2000 The International Registry of Acute Aortic Dissection (IRAD): new insights into an old disease. JAMA 283:897–903
126. **LeMaire SA, Russell L** 2011 Epidemiology of thoracic aortic dissection. Nat Rev Cardiol 8:103–113
127. **Carlson M, Silberbach M** 2007 Dissection of the aorta in Turner syndrome: two cases and review of 85 cases in the literature. J Med Genet 44:745–749
128. **Apostolopoulos T, Kyriakidis M, Toutouzas P** 1992 Endarteritis of the aortic arch in Turner's syndrome with cystic degeneration of the aorta. Int J Cardiol 35:417–419
129. **Lin AE, Lippe B, Rosenfeld RG** 1998 Further delineation of aortic dilation, dissection, and rupture in patients with Turner syndrome. Pediatrics 102:e12
130. **Hiratzka LF, Bakris GL, Beckman JA, Bersin RM, Carr VF, Casey Jr DE, Eagle KA, Hermann LK, Isselbacher EM, Kazerooni EA, Kouchoukos NT, Lytle BW, Milewicz DM, Reich DL, Sen S, Shinn JA, Svensson LG, Williams DM** 2010 2010 ACCF/AHA/AATS/ACR/ASA/SCA/SCAI/SIR/STS/SVM guidelines for the diagnosis and management of patients with thoracic aortic disease: a report of the American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines, American Association for Thoracic Surgery, American College of Radiology, American Stroke Association, Society of Cardiovascular Anesthesiologists, Society for Cardiovascular Angiography and Interventions, Society of Interventional Radiology, Society of Thoracic Surgeons, and Society for Vascular Medicine. Circulation 121:e266–e369
131. **Prandstraller D, Mazzanti L, Giardini A, Lovato L, Tamburrino F, Scarano E, Cicognani A, Fattori R, Picchio FM** 2009 Correlations of phenotype and genotype in relation to morphologic remodelling of the aortic root in patients with Turner's syndrome. Cardiol Young 19:264–271
132. **Elsheikh M, Casadei B, Conway GS, Wass JA** 2001 Hypertension is a major risk factor for aortic root dilatation in women with Turner's syndrome. Clin Endocrinol (Oxf) 54:69–73
133. **Lanzarini L, Larizza D, Prete G, Calcaterra V, Meloni G, Sammarchi L, Klersy C** 2007 Aortic dimensions in Turner's syndrome: two-dimensional echocardiography versus magnetic resonance imaging. J Cardiovasc Med (Hagerstown) 8:428–437
134. **Mortensen KH, Hjerrild BE, Stochholm K, Andersen NH, Sørensen KE, Lundorf E, Hørlyck A, Pedersen EM, Christiansen JS, Gravholt CH** 2011 Dilation of the ascending aorta in Turner syndrome—a prospective cardiovascular magnetic resonance study. J Cardiovasc Magn Reson 13:24
135. **Davies RR, Goldstein LJ, Coady MA, Tittle SL, Rizzo JA, Kopf GS, Elefteriades JA** 2002 Yearly rupture or dissection rates for thoracic aortic aneurysms: simple prediction based on size. Ann Thorac Surg 73:17–27; discussion 27–28
136. **Parish LM, Gorman 3rd JH, Kahn S, Plappert T, St John-Sutton MG, Bavaria JE, Gorman RC** 2009 Aortic size in acute type A dissection: implications for preventive ascending aortic replacement. Eur J Cardiothorac Surg 35:941–945; discussion 945–946
137. **Davies RR, Gallo A, Coady MA, Tellides G, Botta DM, Burke B, Coe MP, Kopf GS, Elefteriades JA** 2006 Novel measurement of relative aortic size predicts rupture of thoracic aortic aneurysms. Ann Thorac Surg 81:169–177
138. **Burman ED, Keegan J, Kilner PJ** 2008 Aortic root measurement by cardiovascular magnetic resonance: specification of planes and lines of measurement and corresponding normal values. Circ Cardiovasc Imaging 1:104–113
139. **Coady MA, Rizzo JA, Hammond GL, Kopf GS, Elefteriades JA** 1999 Surgical intervention criteria for thoracic aortic aneurysms: a study of growth rates and complications. Ann Thorac Surg 67:1922–1926; discussion 1953–1958
140. **Davies RR, Kaple RK, Mandapati D, Gallo A, Botta Jr**

**DM, Elefteriades JA, Coady MA** 2007 Natural history of ascending aortic aneurysms in the setting of an unreplaced bicuspid aortic valve. Ann Thorac Surg 83:1338–1344
141. **Tzemos N, Therrien J, Yip J, Thanassoulis G, Tremblay S, Jamorski MT, Webb GD, Siu SC** 2008 Outcomes in adults with bicuspid aortic valves. JAMA 300:1317–1325
142. **Aronberg DJ, Glazer HS, Madsen K, Sagel SS** 1984 Normal thoracic aortic diameters by computed tomography. J Comput Assist Tomogr 8:247–250
143. **Baguet JP, Douchin S, Pierre H, Rossignol AM, Bost M, Mallion JM** 2005 Structural and functional abnormalities of large arteries in the Turner syndrome. Heart 91:1442–1446
144. **van den Berg J, Bannink EM, Wielopolski PA, Pattynama PM, de Muinck Keizer-Schrama SM, Helbing WA** 2006 Aortic distensibility and dimensions and the effects of growth hormone treatment in the Turner syndrome. Am J Cardiol 97:1644–1649
145. **Sharma J, Friedman D, Dave-Sharma S, Harbison M** 2009 Aortic distensibility and dilation in Turner's syndrome. Cardiol Young 19:568–572
146. **Bryman I, Sylvén L, Berntorp K, Innala E, Bergström I, Hanson C, Oxholm M, Landin-Wilhelmsen K** 2011 Pregnancy rate and outcome in Swedish women with Turner syndrome. Fertil Steril 95:2507–2510
147. **Hadnott TN, Gould HN, Gharib AM, Bondy CA** 2011 Outcomes of spontaneous and assisted pregnancies in Turner syndrome: the U.S. National Institutes of Health experience. Fertil Steril 95:2251–2256
148. **Karnis MF, Zimon AE, Lalwani SI, Timmreck LS, Klipstein S, Reindollar RH** 2003 Risk of death in pregnancy achieved through oocyte donation in patients with Turner syndrome: a national survey. Fertil Steril 80:498–501
149. **Chevalier N, Letur H, Lelannou D, Ohl J, Cornet D, Chalas-Boissonnas C, Frydman R, Catteau-Jonard S, Greck-Chassain T, Papaxanthos-Roche A, Dulucq MC, Couet ML, Cédrin-Durnerin I, Pouly JL, Fénichel P** 2011 Materno-fetal cardiovascular complications in Turner syndrome after oocyte donation: insufficient prepregnancy screening and pregnancy follow-up are associated with poor outcome. J Clin Endocrinol Metab 96:E260–E267
150. **Bodri D, Vernaeve V, Figueras F, Vidal R, Guillén JJ, Coll O** 2006 Oocyte donation in patients with Turner's syndrome: a successful technique but with an accompanying high risk of hypertensive disorders during pregnancy. Hum Reprod 21:829–832
151. **Foudila T, Söderström-Anttila V, Hovatta O** 1999 Turner's syndrome and pregnancies after oocyte donation. Hum Reprod 14:532–535
152. **Bondy C, Rosing D, Reindollar R** 2009 Cardiovascular risks of pregnancy in women with Turner syndrome. Fertil Steril 91:e31–e32
153. **Hovatta O** 1999 Pregnancies in women with Turner's syndrome. Ann Med 31:106–110
154. **Mortensen KH, Rohde MD, Uldbjerg N, Gravholt CH** 2010 Repeated spontaneous pregnancies in 45,X Turner syndrome. Obstet Gynecol 115:446–449
155. **Elsheikh M, Dunger DB, Conway GS, Wass JA** 2002 Turner's syndrome in adulthood. Endocr Rev 23:120–140
156. **El-Hamamsy I, Yacoub MH** 2009 Cellular and molecular mechanisms of thoracic aortic aneurysms. Nat Rev Cardiol 6:771–786
157. **Humphrey JD** 2008 Mechanisms of arterial remodeling in hypertension: coupled roles of wall shear and intramural stress. Hypertension 52:195–200
158. **Della Corte A, Quarto C, Bancone C, Castaldo C, Di Meglio F, Nurzynska D, De Santo LS, De Feo M, Scardone M, Montagnani S, Cotrufo M** 2008 Spatiotemporal patterns of smooth muscle cell changes in ascending aortic dilatation with bicuspid and tricuspid aortic valve stenosis: focus on cell-matrix signaling. J Thorac Cardiovasc Surg 135:8–18, 18.e1–2
159. **Cotrufo M, Della Corte A** 2009 The association of bicuspid aortic valve disease with asymmetric dilatation of the tubular ascending aorta: identification of a definite syndrome. J Cardiovasc Med (Hagerstown) 10:291–297
160. **Pratt B, Curci J** 2010 Arterial elastic fiber structure. Function and potential roles in acute aortic dissection. J Cardiovasc Surg (Torino) 51:647–656
161. **Neptune ER, Frischmeyer PA, Arking DE, Myers L, Bunton TE, Gayraud B, Ramirez F, Sakai LY, Dietz HC** 2003 Dysregulation of TGF-$\beta$ activation contributes to pathogenesis in Marfan syndrome. Nat Genet 33:407–411
162. **Loeys BL, Schwarze U, Holm T, Callewaert BL, Thomas GH, Pannu H, De Backer JF, Oswald GL, Symoens S, Manouvrier S, Roberts AE, Faravelli F, Greco MA, Pyeritz RE, Milewicz DM, Coucke PJ, Cameron DE, Braverman AC, Byers PH, De Paepe AM, Dietz HC** 2006 Aneurysm syndromes caused by mutations in the TGF-$\beta$ receptor. N Engl J Med 355:788–798
163. **Azhar M, Schultz Jel J, Grupp I, Dorn 2nd GW, Meneton P, Molin DG, Gittenberger-de Groot AC, Doetschman T** 2003 Transforming growth factor $\beta$ in cardiovascular development and function. Cytokine Growth Factor Rev 14:391–407
164. **He R, Guo DC, Sun W, Papke CL, Duraisamy S, Estrera AL, Safi HJ, Ahn C, Buja LM, Arnett FC, Zhang J, Geng YJ, Milewicz DM** 2008 Characterization of the inflammatory cells in ascending thoracic aortic aneurysms in patients with Marfan syndrome, familial thoracic aortic aneurysms, and sporadic aneurysms. J Thorac Cardiovasc Surg 136:922–929, 929.e1
165. **Jørgensen KT, Rostgaard K, Bache I, Biggar RJ, Nielsen NM, Tommerup N, Frisch M** 2010 Autoimmune diseases in women with Turner's syndrome. Arthritis Rheum 62:658–666
166. **Mortensen KH, Cleemann L, Hjerrild BE, Nexo E, Locht H, Jeppesen EM, Gravholt CH** 2009 Increased prevalence of autoimmunity in Turner syndrome—influence of age. Clin Exp Immunol 156:205–210
167. **Germain DP** 2007 Ehlers-Danlos syndrome type IV. Orphanet J Rare Dis 2:32
168. **Milewicz DM, Guo DC, Tran-Fadulu V, Lafont AL, Papke CL, Inamoto S, Kwartler CS, Pannu H** 2008 Genetic basis of thoracic aortic aneurysms and dissections: focus on smooth muscle cell contractile dysfunction. Annu Rev Genomics Hum Genet 9:283–302
169. **Haimovici H, Maier N** 1964 Fate of aortic homografts in canine atherosclerosis. 3. Study of fresh abdominal and thoracic aortic implants into thoracic aorta: role of tissue susceptibility in atherogenesis. Arch Surg 89:961–969

170. **Castillo C, Cruzado M, Ariznavarreta C, Gil-Loyzaga P, Lahera V, Cachofeiro V, Tresguerres JA** 2005 Effect of recombinant human growth hormone administration on body composition and vascular function and structure in old male Wistar rats. Biogerontology 6:303–312
171. **Erikstrup C, Pedersen LM, Heickendorff L, Ledet T, Rasmussen LM** 2001 Production of hyaluronan and chondroitin sulphate proteoglycans from human arterial smooth muscle—the effect of glucose, insulin, IGF-I or growth hormone. Eur J Endocrinol 145:193–198
172. **Lombardi G, Galdiero M, Auriemma RS, Pivonello R, Colao A** 2006 Acromegaly and the cardiovascular system. Neuroendocrinology 83:211–217
173. **Bondy CA, Van PL, Bakalov VK, Ho VB** 2006 Growth hormone treatment and aortic dimensions in Turner syndrome. J Clin Endocrinol Metab 91:1785–1788
174. **Elsheikh M, Bird R, Casadei B, Conway GS, Wass JA** 2000 The effect of hormone replacement therapy on cardiovascular hemodynamics in women with Turner's syndrome. J Clin Endocrinol Metab 85:614–618
175. **Chan NN, Vallance P, Colhoun HM, MacAllister RJ, Hingorani AD, Conway GS** 2002 The effects of hormone replacement therapy on endothelial function in women with Turner's syndrome. Clin Endocrinol (Oxf) 56:615–620
176. **Ostberg JE, Storry C, Donald AE, Attar MJ, Halcox JP, Conway GS** 2007 A dose-response study of hormone replacement in young hypogonadal women: effects on intima media thickness and metabolism. Clin Endocrinol (Oxf) 66:557–564
177. **Brooke BS, Habashi JP, Judge DP, Patel N, Loeys B, Dietz 3rd HC** 2008 Angiotensin II blockade and aortic-root dilation in Marfan's syndrome. N Engl J Med 358:2787–2795
178. **Ahimastos AA, Aggarwal A, D'Orsa KM, Formosa MF, White AJ, Savarirayan R, Dart AM, Kingwell BA** 2007 Effect of perindopril on large artery stiffness and aortic root diameter in patients with Marfan syndrome: a randomized controlled trial. JAMA 298:1539–1547
179. **Irioka T, Mizusawa H** 2011 Ischemic stroke in a young adult with Turner syndrome. Neurol Sci 32:317–319
180. **Finsterer J, Zartl M, Samec P** 2000 Spontaneous cerebral haemorrhage without hypertension in non-mosaic 45X Turner's syndrome. J Clin Neurosci 7:341–343
181. **Knisely AS, Sweeney K, Ambler MW** 1988 Pheochromocytoma and sudden death as a result of cerebral infarction in Turner's syndrome: report of a case. J Forensic Sci 33:1497–1502
182. **Spengos K, Kosmaidou-Aravidou Z, Tsivgoulis G, Vassilopoulou S, Grigori-Kostaraki P, Zis V** 2006 Moyamoya syndrome in a Caucasian woman with Turner's syndrome. Eur J Neurol 13:e7–e8
183. **Gandhi NN, Hartman ME, Williford WL, Peters MA, Cheifetz IM, Turner DA** 2011 Successful use of extracorporeal membrane oxygenation for pH1N1-induced refractory hypoxemia in a child with hypoplastic left heart syndrome. Pediatr Crit Care Med 12:e398–e401
184. **Komori H, Matsuishi T, Abe T, Nagata Y, Ohtaki E, Kojima K, Yukizane S** 1993 Turner syndrome and occlusion of the internal carotid artery. J Child Neurol 8:412–415
185. **Rosenblum B, Rothman AS, Lanzieri C, Song S** 1986 A cavernous sinus cavernous hemangioma. Case report. J Neurosurg 65:716–718
186. **Muscat P, Lidov M, Nahar T, Tuhrim S, Weinberger J** 2001 Vertebral artery dissection in Turner's syndrome: diagnosis by magnetic resonance imaging. J Neuroimaging 11:50–54
187. **Donnan GA, Fisher M, Macleod M, Davis SM** 2008 Stroke. Lancet 371:1612–1623
188. **O'Donnell MJ, Xavier D, Liu L, Zhang H, Chin SL, Rao-Melacini P, Rangarajan S, Islam S, Pais P, McQueen MJ, Mondo C, Damasceno A, Lopez-Jaramillo P, Hankey GJ, Dans AL, Yusoff K, Truelsen T, Diener HC, Sacco RL, Ryglewicz D, Czlonkowska A, Weimar C, Wang X, Yusuf S** 2010 Risk factors for ischaemic and intracerebral haemorrhagic stroke in 22 countries (the INTERSTROKE study): a case-control study. Lancet 376:112–123
189. **Hjerrild BE, Holst JJ, Juhl CB, Christiansen JS, Schmitz O, Gravholt CH** 2011 Delayed $\beta$-cell response and glucose intolerance in young women with Turner syndrome. BMC Endocr Disord 11:6
190. **Zieske AW, Malcom GT, Strong JP** 2002 Natural history and risk factors of atherosclerosis in children and youth: the PDAY study. Pediatr Pathol Mol Med 21:213–237
191. **Nilsson PM, Lurbe E, Laurent S** 2008 The early life origins of vascular ageing and cardiovascular risk: the EVA syndrome. J Hypertens 26:1049–1057
192. **Barker DJ, Winter PD, Osmond C, Margetts B, Simmonds SJ** 1989 Weight in infancy and death from ischaemic heart disease. Lancet 2:577–580
193. **Whincup PH, Kaye SJ, Owen CG, Huxley R, Cook DG, Anazawa S, Barrett-Connor E, Bhargava SK, Birgisdottir BE, Carlsson S, de Rooij SR, Dyck RF, Eriksson JG, Falkner B, Fall C, Forsén T, Grill V, Gudnason V, Hulman S, Hyppönen E, Jeffreys M, Lawlor DA, Leon DA, Minami J, Mishra G, Osmond C, Power C, Rich-Edwards JW, Roseboom TJ, Sachdev HS, Syddall H, Thorsdottir I, Vanhala M, Wadsworth M, Yarbrough DE** 2008 Birth weight and risk of type 2 diabetes: a systematic review. JAMA 300:2886–2897
194. **Turgeon JL, Carr MC, Maki PM, Mendelsohn ME, Wise PM** 2006 Complex actions of sex steroids in adipose tissue, the cardiovascular system, and brain: insights from basic science and clinical studies. Endocr Rev 27:575–605
195. **Colao A** 2008 The GH-IGF-I axis and the cardiovascular system: clinical implications. Clin Endocrinol (Oxf) 69:347–358
196. **Ostberg JE, Donald AE, Halcox JP, Storry C, McCarthy C, Conway GS** 2005 Vasculopathy in Turner syndrome: arterial dilatation and intimal thickening without endothelial dysfunction. J Clin Endocrinol Metab 90:5161–5166
197. **Van Bortel LM** 2005 What does intima-media thickness tell us? J Hypertens 23:37–39
198. **Mancia G, Laurent S, Agabiti-Rosei E, Ambrosioni E, Burnier M, Caulfield MJ, Cifkova R, Clément D, Coca A, Dominiczak A, Erdine S, Fagard R, Farsang C, Grassi G, Haller H, Heagerty A, Kjeldsen SE, Kiowski W, Mallion JM, Manolis A, Narkiewicz K, Nilsson P, Olsen MH, Rahn KH, Redon J, Rodicio J, Ruilope L, Schmieder RE, Struijker-Boudier HA, van Zwieten PA, Viigimaa M, Zanchetti A** 2009 Reappraisal of European guidelines on hyperten-

sion management: a European Society of Hypertension Task Force document. J Hypertens 27:2121–2158
199. **Lorenz MW, Markus HS, Bots ML, Rosvall M, Sitzer M** 2007 Prediction of clinical cardiovascular events with carotid intima-media thickness: a systematic review and meta-analysis. Circulation 115:459–467
200. **Pirgon Ö, Atabek ME, Oran B, Güçlü R** 2008 Atherogenic lipid profile and systolic blood pressure are associated with carotid artery intima-media thickness in children with Turner syndrome. J Clin Res Pediatr Endocrinol 1:62–71
201. **Gravholt CH, Mortensen KH, Andersen NH, Ibsen L, Ingerslev J, Hjerrild BE** 2012 Coagulation and fibrinolytic disturbances are related to carotid intima thickness and arterial blood pressure in Turner syndrome. Clin Endocrinol (Oxf) 76:649–656
202. **Lorenz MW, von Kegler S, Steinmetz H, Markus HS, Sitzer M** 2006 Carotid intima-media thickening indicates a higher vascular risk across a wide age range: prospective data from the Carotid Atherosclerosis Progression Study (CAPS). Stroke 37:87–92
203. **Mortensen KH, Hansen KW, Erlandsen M, Christiansen JS, Gravholt CH** 2009 Ambulatory arterial stiffness index in Turner syndrome: the impact of sex hormone replacement therapy. Horm Res 72:184–189
204. **Fuentes K, Silveira DC, Papamitsakis NI** 2007 Spontaneous carotid artery dissection in a patient with Turner syndrome. Cerebrovasc Dis 24:543–544
205. **Jagannath AD, Rastogi U, Spooner AE, Lin AE, Agnihotri AK** 2010 Aortic dissection and moyamoya disease in Turner syndrome. Am J Med Genet A 152A:2085–2089
206. **Lancman M, Mesropian H, Serra P, Granillo R** 1991 Turner's syndrome, fibromuscular dysplasia, and stroke. Stroke 22:269–271
207. **Olin JW, Sealove BA** 2011 Diagnosis, management, and future developments of fibromuscular dysplasia. J Vasc Surg 53:826–836.e1
208. **Scott RM, Smith ER** 2009 Moyamoya disease and moyamoya syndrome. N Engl J Med 360:1226–1237
209. **Grond-Ginsbach C, Pjontek R, Aksay SS, Hyhlik-Dürr A, Böckler D, Gross-Weissmann ML** 2010 Spontaneous arterial dissection: phenotype and molecular pathogenesis. Cell Mol Life Sci 67:1799–1815
210. **Klein LW, Levin JL, Weintraub WS, Agarwal JB, Helfant RH** 1984 Pseudocoarctation of the aortic arch in a patient with Turner's syndrome. Clin Cardiol 7:621–623
211. **De Filippo P, Colombo S, Brambilla R, Borghi A, Cantù F** 2008 Ablation of atrial tachycardia from a giant left superior vena cava using integration with computed tomographic imaging. Circ Arrhythm Electrophysiol 1:143–144
212. **Landin-Wilhelmsen K, Bryman I, Wilhelmsen L** 2001 Cardiac malformations and hypertension, but not metabolic risk factors, are common in Turner syndrome. J Clin Endocrinol Metab 86:4166–4170
213. **Sozen AB, Cefle K, Kudat H, Ozturk S, Oflaz H, Pamukcu B, Akkaya V, Isguven P, Palanduz S, Ozcan M, Goren T, Guven O** 2008 Atrial and ventricular arryhthmogenic potential in Turner syndrome. Pacing Clin Electrophysiol 31:1140–1145
214. **Dilaveris PE, Gialafos JE** 2001 P-wave dispersion: a novel predictor of paroxysmal atrial fibrillation. Ann Noninvasive Electrocardiol 6:159–165
215. **Camm AJ, Kirchhof P, Lip GY, Schotten U, Savelieva I, Ernst S, Van Gelder IC, Al-Attar N, Hindricks G, Prendergast B, Heidbuchel H, Alfieri O, Angelini A, Atar D, Colonna P, De Caterina R, De Sutter J, Goette A, Gorenek B, Heldal M, Hohloser SH, Kolh P, Le Heuzey JY, Ponikowski P, Rutten FH, Vahanian A, Auricchio A, Bax J, Ceconi C, Dean V, Filippatos G, Funck-Brentano C, Hobbs R, Kearney P, McDonagh T, Popescu BA, Reiner Z, Sechtem U, Sirnes PA, Tendera M, Vardas PE, Widimsky P, Vardas PE, Agladze V, Aliot E, Balabanski T, Blomstrom-Lundqvist C, Capucci A, Crijns H, Dahlöf B, Folliguet T, Glikson M, Goethals M, Gulba DC, Ho SY, Klautz RJ, Kose S, McMurray J, Perrone Filardi P, Raatikainen P, Salvador MJ, Schalij MJ, Shpektor A, Sousa J, Stepinska J, Uuetoa H, Zamorano JL, Zupan I** 2010 Guidelines for the management of atrial fibrillation: the Task Force for the Management of Atrial Fibrillation of the European Society of Cardiology (ESC). Europace 12:1360–1420
216. **Bakiler AR, Cangar S, Aksit S, Unver A, Yaprak I, Caglayan S** 1993 Disappearing intracardiac thrombi in both atria after mumps in a patient with Turner syndrome. Chest 103:1611–1612
217. **Ferro JM, Massaro AR, Mas JL** 2010 Aetiological diagnosis of ischaemic stroke in young adults. Lancet Neurol 9:1085–1096
218. **Witt BJ, Gami AS, Ballman KV, Brown Jr RD, Meverden RA, Jacobsen SJ, Roger VL** 2007 The incidence of ischemic stroke in chronic heart failure: a meta-analysis. J Card Fail 13:489–496
219. **Lanes R, Gunczler P, Palacios A, Villaroel O** 1997 Serum lipids, lipoprotein lp (a), and plasminogen activator inhibitor-1 in patients with Turner's syndrome before and during growth hormone and estrogen therapy. Fertil Steril 68:473–477
220. **Calcaterra V, Gamba G, Montani N, de Silvestri A, Terulla V, Lanati G, Larizza D** 2011 Thrombophilic screening in Turner syndrome. J Endocrinol Invest 34:676–679
221. **Kelsey G, Monagle P, Barnes C** 2006 Delayed diagnosis of congenital factor IX deficiency (Christmas disease) in a girl with Turner's syndrome. Clin Lab Haematol 28:355–356
222. **Weinspach S, Siepermann M, Schaper J, Sarikaya-Seiwert S, Rieder H, Gerigk M, Höhn T, Laws HJ** 2009 Intracranial hemorrhage in a female leading to the diagnosis of severe hemophilia A and Turner syndrome. Klin Padiatr 221:167–171
223. **Aligeti VR, Horn HR** 2007 Turner's syndrome and coronary artery disease. Am J Cardiol 99:741–742
224. **Gerlis LM, Gatzoulis M, Ho SY, Sheppard MN** 2004 Sudden unexpected cardiac death in a patient with Turner's syndrome. Cardiol Young 14:192–196
225. **Garvey P, Elovitz M, Landsberger EJ** 1998 Aortic dissection and myocardial infarction in a pregnant patient with Turner syndrome. Obstet Gynecol 91:864
226. **Kalin MF, Zumoff B** 1990 Sex hormones and coronary disease: a review of the clinical studies. Steroids 55:330–352
227. **Salgin B, Amin R, Yuen K, Williams RM, Murgatroyd P, Dunger DB** 2006 Insulin resistance is an intrinsic defect

independent of fat mass in women with Turner's syndrome. Horm Res 65:69–75
228. **Kozlowska-Wojciechowska M, Jez W, Zdrojewski T, Chwojnicki K** 2006 Are young women with Turner syndrome at greater risk of coronary artery disease? Eur J Cardiovasc Prev Rehabil 13:467–469
229. **Hjerrild BE, Mortensen KH, Gravholt CH** 2008 Turner syndrome and clinical treatment. Br Med Bull 86:77–93
230. **Polkampally PR, Matta JR, McAreavey D, Bakalov V, Bondy CA, Gharib AM** 2011 Aneurysmal dilatation of medium caliber arteries in Turner syndrome. Congenit Heart Dis 6:382–383
231. **Oohara K, Yamazaki T, Sakaguchi K, Nakayama M, Kobayashi A** 1995 Acute aortic dissection, aortic insufficiency, and a single coronary artery in a patient with Turner's syndrome. J Cardiovasc Surg (Torino) 36:273–275
232. **Forgosh LB, Conetta DA** 1992 Cardiac assessment in Turner's syndrome: a case for the inclusion of coronary angiography. J Am Soc Echocardiogr 5:281–284
233. **Luckraz H, Mohammed A, Youhana A** 2006 A prominent collateral coronary artery. Ann Thorac Surg 81:1518
234. **Gravholt CH, Naeraa RW, Nyholm B, Gerdes LU, Christiansen E, Schmitz O, Christiansen JS** 1998 Glucose metabolism, lipid metabolism, and cardiovascular risk factors in adult Turner's syndrome. The impact of sex hormone replacement. Diabetes Care 21:1062–1070
235. **Nathwani NC, Unwin R, Brook CG, Hindmarsh PC** 2000 The influence of renal and cardiovascular abnormalities on blood pressure in Turner syndrome. Clin Endocrinol (Oxf) 52:371–377
236. **Virdis R, Cantu MC, Ghizzoni L, Ammenti A, Nori G, Volta C, Cravidi C, Vanelli M, Balestrazzi P, Bernasconi S, Giovannelli G** 1986 Blood pressure behaviour and control in Turner syndrome. Clin Exp Hypertens A 8:787–791
237. **Radetti G, Crepaz R, Milanesi O, Paganini C, Cesaro A, Rigon F, Pitscheider W** 2001 Cardiac performance in Turner's syndrome patients on growth hormone therapy. Horm Res 55:240–244
238. **Jelić S, Marisavljević D** 1997 [Turner's syndrome with mosaic karyotype and renovascular hypertension]. Srp Arh Celok Lek 125:48–50
239. **Boldo A, White WB** 2011 Blood pressure effects of the oral contraceptive and postmenopausal hormone therapies. Endocrinol Metab Clin North Am 40:419–432, ix
240. **Giordano R, Forno D, Lanfranco F, Manieri C, Ghizzoni L, Ghigo E** 2011 Metabolic and cardiovascular outcomes in a group of adult patients with Turner's syndrome under hormonal replacement therapy. Eur J Endocrinol 164: 819–826
241. **Komukai K, Mochizuki S, Yoshimura M** 2010 Gender and the renin-angiotensin-aldosterone system. Fundam Clin Pharmacol 24:687–698
242. **Langrish JP, Mills NL, Bath LE, Warner P, Webb DJ, Kelnar CJ, Critchley HO, Newby DE, Wallace WH** 2009 Cardiovascular effects of physiological and standard sex steroid replacement regimens in premature ovarian failure. Hypertension 53:805–811
243. **Caprio S, Boulware S, Diamond M, Sherwin RS, Carpenter TO, Rubin K, Amiel S, Press M, Tamborlane WV** 1991 Insulin resistance: an early metabolic defect of Turner's syndrome. J Clin Endocrinol Metab 72:832–836
244. **Bakalov VK, Cooley MM, Quon MJ, Luo ML, Yanovski JA, Nelson LM, Sullivan G, Bondy CA** 2004 Impaired insulin secretion in the Turner metabolic syndrome. J Clin Endocrinol Metab 89:3516–3520
245. **Gravholt CH, Hjerrild BE, Mosekilde L, Hansen TK, Rasmussen LM, Frystyk J, Flyvbjerg A, Christiansen JS** 2006 Body composition is distinctly altered in Turner syndrome: relations to glucose metabolism, circulating adipokines, and endothelial adhesion molecules. Eur J Endocrinol 155: 583–592
246. **Ostberg JE, Attar MJ, Mohamed-Ali V, Conway GS** 2005 Adipokine dysregulation in Turner syndrome: comparison of circulating interleukin-6 and leptin concentrations with measures of adiposity and C-reactive protein. J Clin Endocrinol Metab 90:2948–2953
247. **Eriksson JG, Forsén TJ, Kajantie E, Osmond C, Barker DJ** 2007 Childhood growth and hypertension in later life. Hypertension 49:1415–1421
248. **Baldin AD, Siviero-Miachon AA, Fabbri T, de Lemos-Marini SH, Spinola-Castro AM, Baptista MT, D'Souza-Li LF, Morcillo AM, Maciel-Guerra AT, Guerra-Junior G** 2012 Turner syndrome and metabolic derangements: another example of fetal programming. Early Hum Dev 88: 99–102
249. **Cornier MA, Dabelea D, Hernandez TL, Lindstrom RC, Steig AJ, Stob NR, Van Pelt RE, Wang H, Eckel RH** 2008 The metabolic syndrome. Endocr Rev 29:777–822
250. **Tanous D, Benson LN, Horlick EM** 2009 Coarctation of the aorta: evaluation and management. Curr Opin Cardiol 24:509–515
251. **Okuno S, Nishi N, Sakaki T** 2005 Putaminal hemorrhage in a case of Turner's syndrome with hyperaldosteronemia. No Shinkei Geka 33:171–176
252. **Lippe B, Geffner ME, Dietrich RB, Boechat MI, Kangarloo H** 1988 Renal malformations in patients with Turner syndrome: imaging in 141 patients. Pediatrics 82:852–856
253. **Liao AW, Snijders R, Geerts L, Spencer K, Nicolaides KH** 2000 Fetal heart rate in chromosomally abnormal fetuses. Ultrasound Obstet Gynecol 16:610–613
254. **Singh JP, Larson MG, O'Donnell CJ, Wilson PF, Tsuji H, Lloyd-Jones DM, Levy D** 2000 Association of hyperglycemia with reduced heart rate variability (The Framingham Heart Study). Am J Cardiol 86:309–312
255. **Yusuf S, Hawken S, Ounpuu S, Dans T, Avezum A, Lanas F, McQueen M, Budaj A, Pais P, Varigos J, Lisheng L** 2004 Effect of potentially modifiable risk factors associated with myocardial infarction in 52 countries (the INTERHEART study): case-control study. Lancet 364:937–952
256. **Kannel WB** 2000 Vital epidemiologic clues in heart failure. J Clin Epidemiol 53:229–235
257. **Mancia G, De Backer G, Dominiczak A, Cifkova R, Fagard R, Germano G, Grassi G, Heagerty AM, Kjeldsen SE, Laurent S, Narkiewicz K, Ruilope L, Rynkiewicz A, Schmieder RE, Boudier HA, Zanchetti A, Vahanian A, Camm J, De Caterina R, Dean V, Dickstein K, Filippatos G, Funck-Brentano C, Hellemans I, Kristensen SD, McGregor K, Sechtem U, Silber S, Tendera M, Widimsky P, Zamorano JL, Erdine S, Kiowski W, Agabiti-Rosei E, Ambrosioni E, Lindholm LH, Viigimaa M, Adamopoulos S, Agabiti-Rosei E, Ambrosioni E, Bertomeu V, Clement D, Erdine S, Farsang C, Gaita D, Lip G, Mallion JM, Manolis AJ,**

**Nilsson PM, O'Brien E, Ponikowski P, Redon J, Ruschitzka F, Tamargo J, van Zwieten P, Waeber B, Williams B** 2007 2007 Guidelines for the management of arterial hypertension: The Task Force for the Management of Arterial Hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). J Hypertens 25:1105–1187

258. **Habashi JP, Judge DP, Holm TM, Cohn RD, Loeys BL, Cooper TK, Myers L, Klein EC, Liu G, Calvi C, Podowski M, Neptune ER, Halushka MK, Bedja D, Gabrielson K, Rifkin DB, Carta L, Ramirez F, Huso DL, Dietz HC** 2006 Losartan, an AT1 antagonist, prevents aortic aneurysm in a mouse model of Marfan syndrome. Science 312:117–121
259. **Ong KT, Perdu J, De Backer J, Bozec E, Collignon P, Emmerich J, Fauret AL, Fiessinger JN, Germain DP, Georgesco G, Hulot JS, De Paepe A, Plauchu H, Jeunemaitre X, Laurent S, Boutouyrie P** 2010 Effect of celiprolol on prevention of cardiovascular events in vascular Ehlers-Danlos syndrome: a prospective randomised, open, blinded-endpoints trial. Lancet 376:1476–1484
260. **Berg KA, Clark EB, Astemborski JA, Boughman JA** 1988 Prenatal detection of cardiovascular malformations by echocardiography: an indication for cytogenetic evaluation. Am J Obstet Gynecol 159:477–481
261. **Tancredi G, Versacci P, Pasquino AM, Vittucci AC, Pucarelli I, Cappa M, Di Mambro C, Marino B** 2011 Cardiopulmonary response to exercise and cardiac assessment in patients with Turner syndrome. Am J Cardiol 107: 1076–1082
262. **Golzio PG, Franco E, Chiribiri A** 2006 Atrio-ventricular synchronization by single VDD lead inserted through persistent left superior vena cava in patient with Turner's syndrome. Pacing Clin Electrophysiol 29:1181–1182
263. **Kannel WB, Kannel C, Paffenbarger Jr RS, Cupples LA** 1987 Heart rate and cardiovascular mortality: the Framingham Study. Am Heart J 113:1489–1494
264. **Roden DM** 2008 Clinical practice. Long-QT syndrome. N Engl J Med 358:169–176
265. **Dalla Pozza R, Bechtold S, Kääb S, Buckl M, Urschel S, Netz H, Schwarz HP** 2006 QTc interval prolongation in children with Ulrich-Turner syndrome. Eur J Pediatr 165: 831–837
266. **Bokil NJ, Baisden JM, Radford DJ, Summers KM** 2010 Molecular genetics of long QT syndrome. Mol Genet Metab 101:1–8
267. **Blaschke RJ, Rappold GA** 2001 SHOX in short stature syndromes. Horm Res 55(Suppl 1):21–23
268. **Davenport ML** 2010 Approach to the patient with Turner syndrome. J Clin Endocrinol Metab 95:1487–1495
269. **Westwood M, Tajbakhsh SH, Siddals KW, Whatmore AJ, Clayton PE** 2011 Reduced pericellular sensitivity to IGF-I in fibroblasts from girls with Turner syndrome: a mechanism to impair clinical responses to GH. Pediatr Res 70: 25–30
270. **Whatmore AJ, Patel L, Clayton PE** 2009 A pilot study to evaluate gene expression profiles in peripheral blood mononuclear cells (PBMCs) from children with GH deficiency and Turner syndrome in response to GH treatment. Clin Endocrinol (Oxf) 70:429–434
271. **Lin TH, Kirkland JL, Kirkland RT** 1988 Growth hormone assessment and short-term treatment with growth hormone in Turner syndrome. J Pediatr 112:919–922
272. **Massarano AA, Brook CG, Hindmarsh PC, Pringle PJ, Teale JD, Stanhope R, Preece MA** 1989 Growth hormone secretion in Turner's syndrome and influence of oxandrolone and ethinyl oestradiol. Arch Dis Child 64:587–592
273. **Zadik Z, Landau H, Chen M, Altman Y, Lieberman E** 1992 Assessment of growth hormone (GH) axis in Turner's syndrome using 24-hour integrated concentrations of GH, insulin-like growth factor-I, plasma GH-binding activity, GH binding to IM9 cells, and GH response to pharmacological stimulation. J Clin Endocrinol Metab 75:412–416
274. **Wit JM, Massarano AA, Kamp GA, Hindmarsh PC, van Es A, Brook CG, Preece MA, Matthews DR** 1992 Growth hormone secretion in patients with Turner's syndrome as determined by time series analysis. Acta Endocrinol (Copenh) 127:7–12
275. **Lanes R, Brito S, Suniaga M, Moncada G, Borges M** 1990 Growth hormone secretion in pubertal age patients with Turner's syndrome. J Clin Endocrinol Metab 71:770–772
276. **Gravholt CH, Naeraa RW, Fisker S, Christiansen JS** 1997 Body composition and physical fitness are major determinants of the growth hormone-insulin-like growth factor axis aberrations in adult Turner's syndrome, with important modulations by treatment with 17 $\beta$-estradiol. J Clin Endocrinol Metab 82:2570–2577
277. **Foster CM, Borondy M, Markovs ME, Hopwood NJ, Kletter GB, Beitins IZ** 1994 Growth hormone bioactivity in girls with Turner's syndrome: correlation with IGF-I. Pediatr Res 35:218–222
278. **Gravholt CH, Frystyk J, Flyvbjerg A, Orskov H, Christiansen JS** 2001 Reduced free IGF-I and increased IGFBP-3 proteolysis in Turner syndrome: modulation by female sex steroids. Am J Physiol Endocrinol Metab 280:E308–E314
279. **Gravholt CH, Chen JW, Oxvig C, Overgaard MT, Christiansen JS, Frystyk J, Flyvbjerg A** 2006 The GH-IGF-IGFBP axis is changed in Turner syndrome: partial normalization by HRT. Growth Horm IGF Res 16:332–339
280. **Ross JL, Quigley CA, Cao D, Feuillan P, Kowal K, Chipman JJ, Cutler Jr GB** 2011 Growth hormone plus childhood low-dose estrogen in Turner's syndrome. N Engl J Med 364:1230–1242
281. **Stephure DK** 2005 Impact of growth hormone supplementation on adult height in Turner syndrome: results of the Canadian randomized controlled trial. J Clin Endocrinol Metab 90:3360–3366
282. **Stochholm K, Laursen T, Green A, Laurberg P, Andersen M, Kristensen LØ, Feldt-Rasmussen U, Christiansen JS, Frydenberg M, Gravholt CH** 2008 Morbidity and GH deficiency: a nationwide study. Eur J Endocrinol 158:447–457
283. **Sherlock M, Ayuk J, Tomlinson JW, Toogood AA, Aragon-Alonso A, Sheppard MC, Bates AS, Stewart PM** 2010 Mortality in patients with pituitary disease. Endocr Rev 31:301–342
284. **Johnsen SP, Hundborg HH, Sørensen HT, Orskov H, Tjønneland A, Overvad K, Jørgensen JO** 2005 Insulin-like growth factor (IGF) I, -II, and IGF binding protein-3 and

risk of ischemic stroke. J Clin Endocrinol Metab 90:5937–5941

285. **Juul A, Scheike T, Davidsen M, Gyllenborg J, Jørgensen T** 2002 Low serum IGF-I is associated with increased risk of ischemic heart disease: a population-based case-control study. Circulation 106:939–944
286. **Gravholt CH, Veldhuis JD, Christiansen JS** 1998 Increased disorderliness and decreased mass and daily rate of endogenous growth hormone secretion in adult Turner syndrome: the impact of body composition, maximal oxygen uptake and treatment with sex hormones. Growth Horm IGF Res 8:289–298
287. **Bengtsson BA, Edén S, Ernest I, Odén A, Sjögren B** 1988 Epidemiology and long-term survival in acromegaly. A study of 166 cases diagnosed between 1955 and 1984. Acta Med Scand 223:327–335
288. **Colao A, Vitale G, Pivonello R, Ciccarelli A, di SC, Lombardi G** 2004 The heart: an end-organ of GH action. Eur J Endocrinol 151(Suppl 1):S93–S101
289. **Matura LA, Sachdev V, Bakalov VK, Rosing DR, Bondy CA** 2007 Growth hormone treatment and left ventricular dimensions in Turner syndrome. J Pediatr 150:587–591
290. **Bolar K, Hoffman AR, Maneatis T, Lippe B** 2008 Long-term safety of recombinant human growth hormone in Turner syndrome. J Clin Endocrinol Metab 93:344–351
291. **Bannink EM, van der Palen RL, Mulder PG, de Muinck Keizer-Schrama SM** 2009 Long-term follow-up of GH-treated girls with Turner syndrome: BMI, blood pressure, body proportions. Horm Res 71:336–342
292. **Sas TC, de Muinck Keizer-Schrama SM, Stijnen T, Aanstoot HJ, Drop SL** 2000 Carbohydrate metabolism during long-term growth hormone (GH) treatment and after discontinuation of GH treatment in girls with Turner syndrome participating in a randomized dose-response study. Dutch Advisory Group on Growth Hormone. J Clin Endocrinol Metab 85:769–775
293. **Menke LA, Sas TC, Stijnen T, Zandwijken GR, de Muinck Keizer-Schrama SM, Otten BJ, Wit JM** 2011 Effect of oxandrolone on glucose metabolism in growth hormone-treated girls with Turner syndrome. Horm Res Paediatr 75:115–122
294. **Gravholt CH, Naeraa RW, Brixen K, Kastrup KW, Mosekilde L, Jørgensen JO, Christiansen JS** 2002 Short-term growth hormone treatment in girls with Turner syndrome decreases fat mass and insulin sensitivity: a randomized, double-blind, placebo-controlled, crossover study. Pediatrics 110:889–896
295. **Querfeld U, Döpper S, Gradehand A, Kiencke P, Wahn F, Zeisel HJ** 1999 Long-term treatment with growth hormone has no persisting effect on lipoprotein (a) in patients with Turner's syndrome. J Clin Endocrinol Metab 84:967–970
296. **van Teunenbroek A, de Muinck Keizer-Schrama SM, Aanstoot HJ, Stijnen T, Hoogerbrugge N, Drop SL** 1999 Carbohydrate and lipid metabolism during various growth hormone dosing regimens in girls with Turner syndrome. Dutch Working Group on Growth Hormone. Metabolism 48:7–14
297. **Wooten N, Bakalov VK, Hill S, Bondy CA** 2008 Reduced abdominal adiposity and improved glucose tolerance in growth hormone-treated girls with Turner syndrome. J Clin Endocrinol Metab 93:2109–2114
298. **Pasquino AM, Passeri F, Pucarelli I, Segni M, Municchi G** 1997 Spontaneous pubertal development in Turner's syndrome. Italian Study Group for Turner's Syndrome. J Clin Endocrinol Metab 82:1810–1813
299. **Borgström B, Birgit B, Hreinsson J, Julius H, Rasmussen C, Carsten R, Sheikhi M, Maryam S, Fried G, Gabriel F, Keros V, Victoria K, Fridström M, Margareta F, Hovatta O, Outi H** 2009 Fertility preservation in girls with Turner syndrome: prognostic signs of the presence of ovarian follicles. J Clin Endocrinol Metab 94:74–80
300. **Modi DN, Sane S, Bhartiya D** 2003 Accelerated germ cell apoptosis in sex chromosome aneuploid fetal human gonads. Mol Hum Reprod 9:219–225
301. **Gupta S, Chiplunkar S, Gupta A, Gollapudi S** 2003 Increased spontaneous, tumor necrosis factor receptor- and CD95 (Fas)-mediated apoptosis in cord blood T-cell subsets from Turner's syndrome. Genes Immun 4:239–243
302. **Modi D, Sane S, Bhartiya D** 2009 Over-expression of Mullerian inhibiting substance mRNA in the Turner syndrome ovary. Sex Dev 3:245–252
303. **van Pareren YK, de Muinck Keizer-Schrama SM, Stijnen T, Sas TC, Jansen M, Otten BJ, Hoorweg-Nijman JJ, Vulsma T, Stokvis-Brantsma WH, Rouwé CW, Reeser HM, Gerver WJ, Gosen JJ, Rongen-Westerlaken C, Drop SL** 2003 Final height in girls with Turner syndrome after long-term growth hormone treatment in three dosages and low dose estrogens. J Clin Endocrinol Metab 88:1119–1125
304. **Løkkegaard E, Jovanovic Z, Heitmann BL, Keiding N, Ottesen B, Pedersen AT** 2006 The association between early menopause and risk of ischaemic heart disease: influence of hormone therapy. Maturitas 53:226–233
305. **Maclaran K, Horner E, Panay N** 2010 Premature ovarian failure: long-term sequelae. Menopause Int 16:38–41
306. **Mendelsohn ME, Karas RH** 2005 Molecular and cellular basis of cardiovascular gender differences. Science 308:1583–1587
307. **Canonico M, Plu-Bureau G, Lowe GD, Scarabin PY** 2008 Hormone replacement therapy and risk of venous thromboembolism in postmenopausal women: systematic review and meta-analysis. BMJ 336:1227–1231
308. **Helmerhorst FM, Bloemenkamp KW, Rosendaal FR, Vandenbroucke JP** 1997 Oral contraceptives and thrombotic disease: risk of venous thromboembolism. Thromb Haemost 78:327–333
309. **Papagianni V, Deligeoroglou E, Makrakis E, Botsis D, Creatsas G** 2011 Response to hormonal treatment of young females with primary or very premature ovarian failure. Gynecol Endocrinol 27:291–299
310. **Taboada M, Santen R, Lima J, Hossain J, Singh R, Klein KO, Mauras N** 2011 Pharmacokinetics and pharmacodynamics of oral and transdermal 17$\beta$ estradiol in girls with Turner syndrome. J Clin Endocrinol Metab 96:3502–3510
311. **Schumacher M, Guennoun R, Ghoumari A, Massaad C, Robert F, El-Etr M, Akwa Y, Rajkowski K, Baulieu EE** 2007 Novel perspectives for progesterone in hormone replacement therapy, with special reference to the nervous system. Endocr Rev 28:387–439
312. **Høst C, Bojesen A, Frystyk J, Flyvbjerg A, Christiansen JS, Gravholt CH** 2010 Effect of sex hormone treatment on

circulating adiponectin and subforms in Turner and Klinefelter syndrome. Eur J Clin Invest 40:211–219
313. **Apter D, Lenko HL, Perheentupa J, Söderholm A, Vihko R** 1982 Subnormal pubertal increases of serum androgens in Turner's syndrome. Horm Res 16:164–173
314. **Gravholt CH, Svenstrup B, Bennett P, Sandahl Christiansen J** 1999 Reduced androgen levels in adult Turner syndrome: influence of female sex steroids and growth hormone status. Clin Endocrinol (Oxf) 50:791–800
315. **Martin DD, Schweizer R, Schwarze CP, Elmlinger MW, Ranke MB, Binder G** 2004 The early dehydroepiandrosterone sulfate rise of adrenarche and the delay of pubarche indicate primary ovarian failure in Turner syndrome. J Clin Endocrinol Metab 89:1164–1168
316. **Zuckerman-Levin N, Frolova-Bishara T, Militianu D, Levin M, Aharon-Peretz J, Hochberg Z** 2009 Androgen replacement therapy in Turner syndrome: a pilot study. J Clin Endocrinol Metab 94:4820–4827
317. **Davis SR, Burger HG** 2003 The role of androgen therapy. Best Pract Res Clin Endocrinol Metab 17:165–175
318. **Wilson DM, Frane JW, Sherman B, Johanson AJ, Hintz RL, Rosenfeld RG** 1988 Carbohydrate and lipid metabolism in Turner syndrome: effect of therapy with growth hormone, oxandrolone, and a combination of both. J Pediatr 112:210–217
319. **Haeusler G, Schmitt K, Blümel P, Plöchl E, Waldhör T, Frisch H** 1996 Insulin, insulin-like growth factor-binding protein-1, and sex hormone-binding globulin in patients with Turner's syndrome: course over age in untreated patients and effect of therapy with growth hormone alone and in combination with oxandrolone. J Clin Endocrinol Metab 81:536–541
320. **Menke LA, Sas TC, de Muinck Keizer-Schrama SM, Zandwijken GR, de Ridder MA, Odink RJ, Jansen M, Delemarre-van de Waal HA, Stokvis-Brantsma WH, Waelkens JJ, Westerlaken C, Reeser HM, van Trotsenburg AS, Gevers EF, van Buuren S, Dejonckere PH, Hokken-Koelega AC, Otten BJ, Wit JM** 2010 Efficacy and safety of oxandrolone in growth hormone-treated girls with Turner syndrome. J Clin Endocrinol Metab 95:1151–1160
321. **Gault EJ, Perry RJ, Cole TJ, Casey S, Paterson WF, Hindmarsh PC, Betts P, Dunger DB, Donaldson MD** 2011 Effect of oxandrolone and timing of pubertal induction on final height in Turner's syndrome: randomised, double blind, placebo controlled trial. BMJ 342:d1980
322. **Holl RW, Kunze D, Etzrodt H, Teller W, Heinze E** 1994 Turner syndrome: final height, glucose tolerance, bone density and psychosocial status in 25 adult patients. Eur J Pediatr 153:11–16
323. **Polychronakos C, Letarte J, Collu R, Ducharme JR** 1980 Carbohydrate intolerance in children and adolescents with Turner syndrome. J Pediatr 96:1009–1014
324. **Cicognani A, Mazzanti L, Tassinari D, Pellacani A, Forabosco A, Landi L, Pifferi C, Cacciari E** 1988 Differences in carbohydrate tolerance in Turner syndrome depending on age and karyotype. Eur J Pediatr 148:64–68
325. **Stoppoloni G, Prisco F, Alfano C, Iafusco D, Marrazzo G, Paolisso G** 1990 Characteristics of insulin resistance in Turner syndrome. Diabete Metab 16:267–271
326. **Heinze E, Schlickenrieder J, Holl RW, Ebert R** 1991 Reduced secretion of gastric inhibitory polypeptide in Turner patients with impaired glucose tolerance. Eur J Pediatr 150:339–342
327. **Bakalov VK, Cheng C, Zhou J, Bondy CA** 2009 X-Chromosome gene dosage and the risk of diabetes in Turner syndrome. J Clin Endocrinol Metab 94:3289–3296
328. **Gravholt CH, Leth-Larsen R, Lauridsen AL, Thiel S, Hansen TK, Holmskov U, Naeraa RW, Christiansen JS** 2004 The effects of GH and hormone replacement therapy on serum concentrations of mannan-binding lectin, surfactant protein D and vitamin D binding protein in Turner syndrome. Eur J Endocrinol 150:355–362
329. **Greising SM, Baltgalvis KA, Lowe DA, Warren GL** 2009 Hormone therapy and skeletal muscle strength: a meta-analysis. J Gerontol A Biol Sci Med Sci 64:1071–1081
330. **Gravholt CH, Nyholm B, Saltin B, Schmitz O, Christiansen JS** 2001 Muscle fiber composition and capillary density in Turner syndrome: evidence of increased muscle fiber size related to insulin resistance. Diabetes Care 24:1668–1673
331. **Livingstone C, Collison M** 2002 Sex steroids and insulin resistance. Clin Sci (Lond) 102:151–166
332. **Godsland IF** 2005 Oestrogens and insulin secretion. Diabetologia 48:2213–2220
333. **Barros RP, Gustafsson JÅ** 2011 Estrogen receptors and the metabolic network. Cell Metab 14:289–299
334. **El-Mansoury M, Berntorp K, Bryman I, Hanson C, Innala E, Karlsson A, Landin-Wilhelmsen K** 2008 Elevated liver enzymes in Turner syndrome during a 5-year follow-up study. Clin Endocrinol (Oxf) 68:485–490
335. **Elsheikh M, Hodgson HJ, Wass JA, Conway GS** 2001 Hormone replacement therapy may improve hepatic function in women with Turner's syndrome. Clin Endocrinol (Oxf) 55:227–231
336. **Roulot D, Degott C, Chazouillères O, Oberti F, Calès P, Carbonell N, Benferhat S, Bresson-Hadni S, Valla D** 2004 Vascular involvement of the liver in Turner's syndrome. Hepatology 39:239–247
337. **Gravholt CH, Poulsen HE, Ott P, Christiansen JS, Vilstrup H** 2007 Quantitative liver functions in Turner syndrome with and without hormone replacement therapy. Eur J Endocrinol 156:679–686
338. **Koulouri O, Ostberg J, Conway GS** 2008 Liver dysfunction in Turner's syndrome: prevalence, natural history and effect of exogenous oestrogen. Clin Endocrinol (Oxf) 69: 306–310
339. **El-Mansoury M, Bryman I, Berntorp K, Hanson C, Wilhelmsen L, Landin-Wilhelmsen K** 2005 Hypothyroidism is common in Turner syndrome: results of a five-year follow-up. J Clin Endocrinol Metab 90:2131–2135
340. **Klein I, Danzi S** 2007 Thyroid disease and the heart. Circulation 116:1725–1735
341. **Pernis AB** 2007 Estrogen and CD4+ T cells. Curr Opin Rheumatol 19:414–420
342. **Straub RH** 2007 The complex role of estrogens in inflammation. Endocr Rev 28:521–574
343. **Blaschke RJ, Rappold G** 2006 The pseudoautosomal regions, SHOX and disease. Curr Opin Genet Dev 16:233–239
344. **Joseph M, Cantú ES, Pai GS, Willi SM, Papenhausen PR, Weiss L** 1996 Xp pseudoautosomal gene haploinsufficiency and linear growth deficiency in three girls with chro-

mosome Xp22;Yq11 translocation. J Med Genet 33:906–911
345. **Kveiborg M, Gravholt CH, Kassem M** 2001 Evidence of a normal mean telomere fragment length in patients with Ullrich-Turner syndrome. Eur J Hum Genet 9:877–879
346. **Güngör N, Böke B, Belgin E, Tunçbilek E** 2000 High frequency hearing loss in Ullrich-Turner syndrome. Eur J Pediatr 159:740–744
347. **Vriend JW, Mulder BJ** 2005 Late complications in patients after repair of aortic coarctation: implications for management. Int J Cardiol 101:399–406
348. **Januzzi JL, Sabatine MS, Eagle KA, Evangelista A, Bruckman D, Fattori R, Oh JK, Moore AG, Sechtem U, Llovet A, Gilon D, Pape L, O'Gara PT, Mehta R, Cooper JV, Hagan PG, Armstrong WF, Deeb GM, Suzuki T, Nienaber CA, Isselbacher EM** 2002 Iatrogenic aortic dissection. Am J Cardiol 89:623–626
349. **Richens D, Kotidis K, Neale M, Oakley C, Fails A** 2003 Rupture of the aorta following road traffic accidents in the United Kingdom 1992–1999. The results of the co-operative crash injury study. Eur J Cardiothorac Surg 23:143–148
350. **Jobe S, Donohoue P, Di Paola J** 2004 Deep venous thrombosis and Turner syndrome. J Pediatr Hematol Oncol 26:272
351. **Donal E, Coisne D, Corbi P** 2000 Images in cardiology: a case report of aortic arch mobile thrombi. Heart 84:614
352. **Kopacek Zilz C, Keller Brenner J, Elnecave RH** 2006 Portal vein thrombosis and high factor VIII in Turner syndrome. Horm Res 66:89–93
353. **Ureten K, Ozturk MA, Bostanci A, Ceneli O, Ozbek M, Haznedaroglu IC** 2010 Atraumatic osteonecrosis after estrogen replacement therapy associated with low protein S level in a patient with Turner syndrome. Clin Appl Thromb Hemost 16:599–601
354. **Pinto RB, Silveira TR, Bandinelli E, Röhsig L** 2004 Portal vein thrombosis in children and adolescents: the low prevalence of hereditary thrombophilic disorders. J Pediatr Surg 39:1356–1361
355. **Sozen AB, Cefle K, Kudat H, Ozturk S, Oflaz H, Akkaya V, Palanduz S, Demirel S, Ozcan M, Goren T, Guven O** 2009 Left ventricular thickness is increased in nonhypertensive Turner's syndrome. Echocardiography 26:943–949
356. **Sas TC, Cromme-Dijkhuis AH, de Muinck Keizer-Schrama SM, Stijnen T, van Teunenbroek A, Drop SL** 1999 The effects of long-term growth hormone treatment on cardiac left ventricular dimensions and blood pressure in girls with Turner's syndrome. Dutch Working Group on Growth Hormone. J Pediatr 135:470–476
357. **van den Berg J, Bannink EM, Wielopolski PA, Hop WC, van Osch-Gevers L, Pattynama PM, de Muinck Keizer-Schrama SM, Helbing WA** 2008 Cardiac status after childhood growth hormone treatment of Turner syndrome. J Clin Endocrinol Metab 93:2553–2558