What is already known on this topic?
Elevated liver function tests (LFTs) are common in adult patients Turner syndrome (TS). Potential causes and mechanisms suggested in the literature are not clear, and may include autoimmune process, venous malformations, obesity and sex hormones replacement therapy (HRT).

What this study adds?
Elevated LFTs in TS are common in children and adolescents with TS. Obesity and HRT do not increase the risk of elevated LFTs.

Abstract
Elevated liver function tests (LFTs) are common in adult patients Turner syndrome (TS). The data regarding children and adolescents are lacking.

Objective: To investigate the prevalence of abnormal LFTs in children and adolescents with TS; to analyse LFTs changes and its clinical significance during several years of observation; to evaluate the potential impact of increased BMI and sex hormones replacement therapy (HRT) on LFTs.

Methods: The analysis included 100 girls with TS (age 4-16, the mean BMI SDS 0.63 [SD 1.53]; 44 on HRT) treated with human recombinant growth hormone. A longitudinal study included 81 patients (mean follow-up period: 4.31 years, SD 0.82).

Results: Elevated LFTs were found in 34 % of patients (in 32% without HRT vs. in 36% on HRT). The relative risk of increased LFTs activity was not higher in obese vs. normal weight (OR 0.2; 95% CI 0.1-0.36, p=0.38 vs. OR 0.16; 95% CI 0.08-0.3, p=0.1). HRT did not increase the risk of abnormal LFTs activity (OR 0.8; 95% CI 0.5-1.2, p=0.37 vs. OR 0.7; 95% CI 0.4-1.1, p=0.27). During the follow-up period, no patient developed overt liver disease. There was no significant increase nor decrease of the abnormal LFTs frequency in the subsequent years of follow up.

Conclusions: Constantly elevated LFTs in TS are common in children and adolescents with TS. However the causes and clinical significance remain unclear, this study show, that obesity and HRT do not increase the risk of elevated LFTs.

Keywords: Turner syndrome, children, liver, estrogen

Introduction
Turner Syndrome (TS) affects approximately 1 per 2500 live female births and is one of the most common chromosomal aberration in females [1,2]. It is caused by a partial or complete X chromosome monosomy. Conditions often seen in TS include: short stature, ovarian dysgenesis, dysmorphic features and endocrine disturbances e.g. diabetes mellitus, thyreoiditis. According to some clinical studies, liver involvement seems to be frequent in adult TS patients with prevalence of abnormal liver functional tests (LFTs) from 20 to 80% [3-6]. The data regarding children and adolescents are lacking. The causes and clinical significance of this phenomenon are unclear, nevertheless overt liver diseases are also more common in TS patients than in general population. The hepatic histological changes reported in TS patients vary, including minimal abnormalities, steatosis, steatohepatitis, biliary involvement, nodular regenerative hyperplasia, and even cirrhosis [5-19]. Potential causes and mechanisms suggested in the literature are not clear, and may include autoimmune process, venous malformations, obesity and sex hormones replacement therapy (HRT) [5-20]. On the other hand, some basic and animal studies point to crucial a role of estrogen deficiency or estrogen receptor malfunction in the development of liver impairment [21-27].

Aims
To investigate the prevalence of abnormal LFTs in children and adolescents with TS; to analyse LFTs changes and its clinical significance during several years of observation; to evaluate the potential impact of increased BMI and sex hormones replacement therapy (HRT) on LFTs.

Methods
The analysis included 100 girls with TS (age 4-16, the mean BMI SDS 0.63 [SD 1.53]; 44 on HRT), all treated with human recombinant growth hormone. A longitudinal study included 81 patients (mean follow-up period: 4.31 years, SD 0.82). The activity of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) was measured in fresh serum samples using dry chemistry (VITROS® 5.1 FS, Ortho Clinical Diagnostics).

Statistical Analysis

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To compare the two sets of data, the Student’s t test or two-sided Mann–Whitney U test was used. For a correlation analysis, the correlation coefficient (R) and regression analysis were used. The purpose of determining the odds ratio (OR) was to use the logistic regression analysis. Statistically significant results were assumed for which the probability value was less than 0.05.

Ethics
The investigation was conducted according to the principles expressed in the Declaration of Helsinki. The participants and/or their parents signed informed consent. The study has been approved by the Jagiellonian University Bioethics Committee (decision number KBET/102/B/2012).

Results
Elevated LFTs were found in 35% of patients (in 32% without HRT vs. in 36% on HRT). Increased AST activity was present in 20% of all patients (in 18% without HRT; in 11% on HRT), and elevation of ALT in 41% of all (in 16% without HRT and in 25% on HRT). The relative risk of increased LFTs activity was not higher in obese vs. normal weight (OR 0.2; 95% CI 0.1-0.4, p=0.38 vs. OR 0.16; 95% CI 0.08-0.3, p=0.1). HRT did not increase the risk of abnormal LFTs activity (OR 0.8; 95% CI 0.5-1.2, p=0.37 vs. OR 0.7; 95% CI 0.4-1.1, p=0.27). During the follow-up period, no patient developed overt liver disease. There was no significant increase nor decrease of the abnormal LFTs frequency in the subsequent years of follow-up (Figure 1.)

Discussion
The reported elevated LFTs activity in TS patients ranges from 20 to 80%, with the highest values in the older patients [3-6, 27-28]. The percentage of liver dysfunction in TS seems to increase with the age. In the recent large study (842 pediatric patients) only 3.4% of 698 examined presented abnormal LFTs activity, but only in only in 5 patients younger than 10 years of age [39]. Research carried out in older age groups indicates a much more frequent occurrence of abnormal LFTs values. In the study of El-Mansoury et al. 36% of 218 adult TS patients presented with abnormal levels of one or more liver enzymes at the beginning, and subsequent 23% developed abnormal LFTs during a 5-year follow-up [27]. In our study, we found a similar number of young TS patients with elevated LFTs at the beginning of the study (34%), but we did not observe any progression during the follow-up period (Figure 1). Although generally liver diseases in patients with TS are more common than in the general population, so far no direct correlation has been found between their development and the occurrence of abnormal LFTs in the preceding period. In most of the published studies LFTs did not progress to overt liver diseases. Also, little is known about the factors predisposing to abnormal LFTs. The literature suggests the possible participation of obesity and HRT by analogy to the results of research conducted in various groups of patients [4]. Because of their short stature and abnormal body proportions, women with TS are more likely to be overweight and obese [4,17-19,30]. In our present study, no relationship was found between obesity and LFTs. The relative risk of the development of LFTs was comparable in patients with obesity and normal BMI-SDS. This finding remains in accordance with few earlier studies in this field, which confirmed obesity as a frequent finding in TS patients, but without correlation to liver impairment [5,27,31].

Another potential factor widely considered in older publications as a cause of hepatotoxicity is estrogen replacement therapy [32,33]. Estrogen receptors are expressed in the liver and estrogens probably play one of the most important role in hepatic lipid homeostasis [34,35]. Despite many studies performed in this field, the causative role of estrogens is not well established. Some reports suggested that estrogen replacement therapy in TS patients can cause deterioration of liver function and in some patients discontinuation of therapy was followed by a decrease in enzyme levels [36]. On the contrary, some more recent valuable studies point to a potential role of estrogen replacement as a favourable factor improving liver function [21]. Although some studies reported alterations in LFTs in TS patients treated with estrogens, these alterations did not improve with the discontinuation of replacement therapy [13, 20]. More recent studies found also elevated LFTs in young patients before HRT, and some, showed beneficial impact of estrogens introduction [4,28,31]. In our study, sex hormones replacement therapy did not increase the risk of elevated ALT and AST activity. As we examined a group of pediatric TS patients, it can be difficult to compare our results with studies based on results of adult TS patients, however the more recent observations studies conducted in post-menopausal women without HRT revealed an increased risk of liver steatosis in comparison to pre-menopausal ones [21, 37, 38]. For this reason, the importance of estrogen in liver function has become the subject of many experimental studies. And it turned out that that many basic and animal studies revealed a crucial role of estrogens and estrogen receptor deficiency in the pathogenesis of liver dysfunction. Estrogens can mediate their biologic effects in the liver through a number of mechanisms. The classic mechanism involves its binding to the steroid nuclear hormone receptors, α or β. Both have the classic features of steroid hormone receptors [39]. Estrogens can also alter cell signaling via estrogen receptor α or β localized in the cell membrane. In addition to membrane localized α and β receptors, estrogens can signal through another cell surface receptor, the G protein coupled estrogen receptor (GPER, also called Gpr30) which is expressed in multiple tissues including liver [40]. It has been shown recently that the loss of receptor α in the liver is associated with hepatic steatosis and inflammation, and its gene expression is lower in patients with NASH [41]. Zhu et al. found that estrogen treatment may reverse aspects of pathway-selective insulin resistance by promoting insulin action on glucose metabolism but limiting hepatic lipid and diacylglycerol deposition [22,23]. Estrogen treatment reduces liver fat storage on several levels, mainly by blocking insulin signaling to liver acetyl-CoA carboxylase and reducing hepatic apoB100 and phospholipid transfer protein. This protective effect of estrogen treatment requires intact hepatic estrogen signaling through estrogen receptor α. By contrast, hepatic estrogen singling may not be required for the effects of estrogen treatment on body weight and adiposity [22,23]. Moreover, Kao et al. found that estrogen receptor α could be an important mediator of liver regeneration [42]. What is more, it has been shown that estrogen receptor β agonist might provide therapeutic benefits in liver steato-hepatitis by directly modulating the bile acid receptors in the liver, which have important functions in the liver, and, indirectly, by inhibiting adiposity [43]. The mechanisms by which estrogen signaling protects against hepatic steatosis also include reductions in de novo lipogenesis, as reported by Gao et al. [25]. These mechanisms may be helpful for understanding mechanisms of liver impairment in TS patients and the favourable action of estrogen replacement.

Conclusion
Constantly elevated LFTs in TS are common in children and adolescents with TS. However the causes and clinical significance remain unclear, this study show, that obesity and HRT do not increase the risk of elevated LFTs.

Authors contribution
M.W. Conception or design of the work, data collection, data analysis and interpretation, drafting the article, final approval of the
version to be Publisher

A.R. Conception or design of the work, data collection, data analysis and interpretation, drafting the article, D.J. data collection, data analysis and interpretation, drafting the article, critical revision of the article J.S. critical revision of the article, final approval of the version to be Publisher

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Figure 1. Percentage of abnormal results of LFTs during subsequent years of observation

![Figure 1](https://example.com/figure1.png)